Effects of low-dose alteplase and intensive blood pressure control on brain (micro-) circulation: imaging analyses from the ENCHANTED trial

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A thesis submitted for the degree of

Doctor of Philosophy

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Surname/Family Name	:	**Zhou**
Given Name/s	:	**Zien**
Abbreviation for degree	:	**PhD**
Faculty	:	**Medicine**
School	:	**The George Institute for Global Health**
Thesis Title	:	**Effects of low-dose alteplase and intensive blood pressure control on brain (micro-)circulation: imaging analyses from the ENCHANTED trial**

Background and aims: Worldwide, the burden of acute ischaemic stroke (AIS) is high in terms of disability-adjusted life-years lost. Prior research shows that imaging features of brain frailty (atrophy and severe leucoaraiosis) and acute ischaemia (visible ischaemic lesions, hypoattenuation, large ischaemic lesion, swelling, and hyper-attenuated arteries) on non-contrast computerised tomography (CT) scans are associated with symptomatic intracerebral haemorrhage (sICH) and mortality in thrombolysis-treated AIS patients. This thesis aimed to elucidate other CT and magnetic resonance imaging (MRI) factors related to abnormalities in the brain (micro-) circulation (including fluid-attenuated inversion recovery hyperintense arteries [FLAIR-HAs], a single penetrating artery occlusion presented as a lacunar infarct, and vascular obstruction at different sites) that determine prognosis in thrombolysis-treated AIS patients, and that may modify the effects of treatment with either intravenous alteplase by dose or in the control of blood pressure (BP) according to intensity.

Methods: Secondary analyses of datasets from the international randomised trial - Enhanced Control of Hypertension and Thrombolysis Stroke Study (ENCHANTED) - were conducted after central classification of brain scan images.

Results: FLAIR-HAs on MRI are frequently present in AIS due to presumed cardioembolism. They indicate a favourable prognosis in such thrombolysis-treated AIS patients, despite also being associated with an increased risk of intracerebral haemorrhage within the region of infarction.

A meta-analysis of the ENCHANTED data with similar studies shows that FLAIR-HAs are not associated with the functional outcome overall but are associated with recovery, specifically in patients with endovascular therapy for AIS. FLAIR-HAs are also associated with early recanalisation or haemorrhagic complications, and early neurologic deterioration.

My studies on lacunar AIS show no differences in the treatment effect of low- versus standard-dose alteplase, and early intensive versus guideline-recommended BP lowering, on functional recovery and sICH across lacunar and non-lacunar cases of AIS.

Furthermore, functional recovery by alteplase dose or BP lowering intensity is not modified by the degree or site of vascular obstruction in cerebral vessels noted on CT or MRI angiography.

Conclusions: My thesis has established that several CT/MRI brain imaging features of brain (micro-) circulation are important prognostic markers of functional recovery and sICH in thrombolysis-treated AIS patients. However, these imaging markers do not appear to modify the treatment effects of randomised alteplase dose and degree of BP-lowering among participants of the ENCHANTED trial.

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The work in this thesis is the result of original research by myself, and has not been submitted for a higher degree at any other university or institution. Any contribution made to the research by others with whom I have worked at UNSW or elsewhere is acknowledged in the thesis.

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1. Zhou Z, Yoshimura S, Delcourt C, et al. Thrombolysis outcomes in acute ischaemic stroke by fluid-attenuated inversion recovery hyperintense arteries. Stroke 2020;51(7):2240-3.
2. Zhou Z, Malavera A, Yoshimura S, et al. Clinical prognosis of FLAIR hyperintense arteries in ischaemic stroke patients: a systematic review and meta-analysis. J Neurol Neurosurg Psychiatry 2020;91(5):475-82.
3. Zhou Z, Delcourt C, Xia C, et al. Low- versus standard-dose alteplase in acute lacunar ischaemic stroke: the ENCHANTED trial. Neurology 2021;96(11):e1512-6.
4. Zhou Z, Xia C, Carcel C, et al. Intensive versus guideline-recommended blood pressure reduction in acute lacunar stroke with intravenous thrombolysis therapy: the ENCHANTED trial. Eur J Neurol 2021;28(3):783-93.
5. Zhou Z, Xia C, Mair G, et al. Thrombolysis outcomes according to arterial characteristics of acute ischaemic stroke by alteplase dose and blood pressure target. Int J Stroke. 2021:17474930211025436 (Online ahead of print).

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I declare that:

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Zien Zhou		05/06/2021

Postgraduate Coordinator's Declaration (to be filled in where publications are used in lieu of Chapters)

I declare that:

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- the minimum requirements for the format of the thesis have been met.

PGC's Name	**PGC's Signature**	**Date (dd/mm/yy)**
Maree Hackett		08/02/2022

Details of publication #1:
Full title: Thrombolysis outcomes in acute ischaemic stroke by fluid-attenuated inversion recovery hyperintense arteries
Authors: Zien Zhou, Sohei Yoshimura, Candice Delcourt, Richard I. Lindley, Shoujiang You, Alejandra Malavera, Takako Torii-Yoshimura, Cheryl Carcel, Xia Wang, Xiaoying Chen, Mark W. Parsons, Andrew M. Demchuk, Joanna M. Wardlaw, Grant Mair, Thompson G. Robinson, John Chalmers, Jianrong Xu, Craig S. Anderson
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The Candidate's Contribution to the Work

Dr. Zhou contributed to the conception, rationale, and design of this study, participated in the image assessment, cross-checked all imaging data and performed data analyses, drafted the initial manuscript, revised the manuscript, drafted the responses to the reviewers' comments, and prepared the final draft of the manuscript for publication.

Location of the work in the thesis and/or how the work is incorporated in the thesis:

Chapter 2: This chapter presented post-hoc analyses from the ENCHANTED trial with respect to the predictors of fluid-attenuated inversion recovery sequence hyperintense arteries (FLAIR-HAs) on MRI and the associations of FLAIR-HAs with thrombolysis outcomes in AIS patients. The findings of this study provide a prompt for clinicians to recognise the presence of FLAIR-HAs where MRI is used in the management of AIS patients.

Primary Supervisor's Declaration

I declare that:

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- All of the co-authors of the publication have reviewed the above information and have agreed to its veracity by signing a 'Co-Author Authorisation' form.

Supervisor's name	*Supervisor's signature*	*Date (dd/mm/yy)*
Craig S. Anderson		*01/09/2021*

Details of publication #2:
Full title: Clinical prognosis of FLAIR hyperintense arteries in ischaemic stroke patients: a systematic review and meta-analysis
Authors: Zien Zhou, Alejandra Malavera, Sohei Yoshimura, Candice Delcourt, Grant Mair, Rustam Al-Shahi Salman, Andrew M. Demchuk, Joanna M. Wardlaw, Richard I. Lindley, Craig S. Anderson
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The Candidate's Contribution to the Work

Dr. Zhou contributed to the conception, rationale, and design of this study, developed and registered the study protocol, conducted the literature search, screening, and data extraction, performed data analyses, wrote the first draft of the manuscript, drafted the responses to the reviewers' comments, and prepared the final draft of the manuscript for publication.

Location of the work in the thesis and/or how the work is incorporated in the thesis:

Chapter 3: This chapter is a systematic review and study-level meta-analysis on associations of FLAIR-HAs with clinical prognosis in AIS patients. The study presented a comprehensive analysis to address this issue through the stratification by studies with different treatments administered to the patients and different methodologies of FLAIR-HAs assessment.

Primary Supervisor's Declaration

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Supervisor's name	*Supervisor's signature*	*Date (dd/mm/yy)*
Craig S. Anderson		*01/09/2021*

Details of publication #3:

Full title: Low-dose vs standard-dose alteplase in acute lacunar ischaemic stroke: the ENCHANTED trial

Authors: Zien Zhou, Candice Delcourt, Chao Xia, Sohei Yoshimura, Cheryl Carcel, Takako Torii-Yoshimura, Shoujiang You, Alejandra Malavera, Xiaoying Chen, Maree L. Hackett, Mark Woodward, John Chalmers, Jianrong Xu, Thompson G. Robinson, Mark W. Parsons, Andrew M. Demchuk, Richard I. Lindley, Grant Mair, Joanna M. Wardlaw, Craig S. Anderson

Journal or book name: Neurology

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The Candidate's Contribution to the Work

Dr. Zhou contributed to the conception, rationale, and design of this study, participated in the image assessment, cross-checked all imaging data and performed data analyses, wrote the first draft of the manuscript, drafted the responses to the reviewers' comments, and prepared the final draft of the manuscript for publication.

Location of the work in the thesis and/or how the work is incorporated in the thesis:

Chapter 4: This chapter is a secondary analysis from the ENCHANTED alteplase dose arm that explored the efficacy and safety of low- versus standard-dose intravenous alteplase for lacunar versus nonlacunar AIS according to prespecified definitions based on clinical and adjudicated imaging findings. The results provided Class II evidence that low-dose alteplase had no additional benefit or safety than standard-dose alteplase for lacunar AIS patients.

Primary Supervisor's Declaration

I declare that:

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- All of the co-authors of the publication have reviewed the above information and have agreed to its veracity by signing a 'Co-Author Authorisation' form.

Supervisor's name	*Supervisor's signature*	*Date (dd/mm/yy)*
Craig S. Anderson		*01/09/2021*

Details of publication #4:
Full title: Intensive versus guideline-recommended blood pressure reduction in acute lacunar stroke with intravenous thrombolysis therapy: The ENCHANTED trial
Authors: Zien Zhou, Chao Xia, Cheryl Carcel, Sohei Yoshimura, Xia Wang, Candice Delcourt, Alejandra Malavera, Xiaoying Chen, Grant Mair, Mark Woodward, John Chalmers, Andrew M. Demchuk, Richard I. Lindley, Thompson G. Robinson, Mark W. Parsons, Joanna M. Wardlaw, Craig S. Anderson
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The Candidate's Contribution to the Work

Dr. Zhou contributed to the conception, rationale, and design of this study, participated in the image assessment, cross-checked all imaging data and performed data analyses, wrote the first draft of the manuscript, drafted the responses to the reviewers' comments, and prepared the final draft of the manuscript for publication.

Location of the work in the thesis and/or how the work is incorporated in the thesis:

Chapter 5: This chapter is a secondary analysis from the ENCHANTED blood pressure (BP) arm that explored differential effects of early intensive versus guideline-recommended BP lowering between lacunar and non-lacunar AIS according to prespecified definitions based on clinical and adjudicated imaging findings. The results showed no differences in the treatment effect of early intensive versus guideline-recommended BP lowering across lacunar and non-lacunar AIS.

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Craig S. Anderson		*01/09/2021*

Details of publication #5:
Full title: Thrombolysis outcomes according to arterial characteristics of acute ischaemic stroke by alteplase dose and blood pressure target
Authors: Zien Zhou, Chao Xia, Grant Mair, Candice Delcourt, Sohei Yoshimura, Xiaosheng Liu, Zengai Chen, Alejandra Malavera, Cheryl Carcel, Xiaoying Chen, Xia Wang, Rustam Al-Shahi Salman, Thompson G. Robinson, Richard I. Lindley, John Chalmers, Joanna M. Wardlaw, Mark W. Parsons, Andrew M. Demchuk, Craig S. Anderson
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The Candidate's Contribution to the Work

Dr. Zhou contributed to the conception, rationale, and design of this study, set up an ENCHANTED angiography imaging reading from, participated in the image assessment, cross-checked all imaging data and performed analyses, wrote the first draft of the manuscript, drafted the responses to the reviewers' comments, and prepared the final draft of the manuscript for publication.

Location of the work in the thesis and/or how the work is incorporated in the thesis:

Chapter 6: This chapter presented a study with the aim to explore the influence of low-dose intravenous alteplase and intensive BP lowering on outcomes of AIS according to status/site of vascular obstruction in the ENCHANTED participants. The main findings from Chapter 6 give light to future research in exploring whether AIS patients with medium vessel occlusion would benefit from low-dose alteplase and whether caution should be advised for intensive BP lowering in thrombolysis-treated patients with large vessel occlusion.

Primary Supervisor's Declaration

I declare that:

- the information above is accurate
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- All of the co-authors of the publication have reviewed the above information and have agreed to its veracity by signing a 'Co-Author Authorisation' form.

Supervisor's name	*Supervisor's signature*	*Date (dd/mm/yy)*
Craig S. Anderson		*01/09/2021*

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"Five years ago, I left home. I dedicate this thesis to my beloved father, who passed away in the autumn of 2020, and everyone who has supported me in this PhD adventure."

Abstract

Background and aims: Worldwide, the burden of acute ischaemic stroke (AIS) is high in terms of disability-adjusted life-years lost. Prior research has shown that imaging features of brain frailty (atrophy and severe leucoaraiosis) and acute ischaemia (visible ischaemic lesions, hypoattenuation, large ischaemic lesion, swelling, and hyper-attenuated arteries) on non-contrast computerised tomography (CT) are associated with symptomatic intracerebral haemorrhage (sICH) and mortality in thrombolysis-treated AIS patients. This thesis aimed to elucidate other brain (micro-) circulation imaging markers (including fluid-attenuated inversion recovery hyperintense arteries [FLAIR-HAs], a single penetrating artery occlusion presented as a lacunar infarct, and vascular obstruction at different sites) of prognosis in thrombolysis-treated AIS patients and determine whether any of them modify the effects of a randomised dose of intravenous alteplase and intensity of blood pressure (BP) control that was evaluated in a large international clinical trial.

Methods: This work pertains to secondary analyses of datasets from an international randomised trial - the Enhanced Control of Hypertension and Thrombolysis Stroke Study (ENCHANTED) – involving over 3000 participants, where their brain images were centrally analyses with other clinical and outcome data.

Results: FLAIR-HAs on magnetic resonance imaging (MRI) are common in AIS presumed to be due to cardioembolism. They indicate a favourable prognosis in thrombolysis-treated AIS patients, despite also being associated with an increased risk of haemorrhage within the area of infarction.

Meta-analysis of the ENCHANTED data with other studies shows that FLAIR-HAs are not associated with the functional outcome overall, but are associated with outcomes specifically in patients who receive endovascular therapy for AIS. FLAIR-HAs are also associated with early recanalisation or haemorrhagic complications and early neurologic deterioration.

My studies of lacunar AIS show no differences in the treatment effect of low- versus standard-dose alteplase, and early intensive versus guideline-recommended BP lowering, on functional recovery and sICH between lacunar and non-lacunar AIS.

Furthermore, functional recovery by alteplase dose or BP lowering intensity is also not modified by the location and degree of vascular obstruction within the cerebral circulation noted on CT or MRI angiography.

Conclusions: The data outlined in this thesis has established that several CT/MRI imaging markers of brain (micro-) circulation abnormality have prognostic significance for functional recovery and sICH in thrombolysis-treated AIS patients. However, no imaging markers appear to modify the treatment effects of alteplase dose and BP-lowering in the ENCHANTED trial.

Ethical Clearance

All participating hospital sites received approval from their relevant ethics committees (or Institutional Review Board) before initiating the ENCHANTED trial. Written informed consent was obtained from each patient or his/her legal surrogate in situations where a participant could not provide this directly due to neurological impairment.

Publications, Conference Presentations, and Awards

Publications arising from this thesis

1. **Zhou Z**, Yoshimura S, Delcourt C, Lindley RI, You S, Malavera A, Torii-Yoshimura T, Carcel C, Wang X, Chen X, Parsons MW, Demchuk AM, Wardlaw JM, Mair G, Robinson TG, Chalmers J, Xu J, Anderson CS. Thrombolysis outcomes in acute ischaemic stroke by fluid-attenuated inversion recovery hyperintense arteries. *Stroke* 2020;51(7):2240-3.

2. **Zhou Z**, Malavera A, Yoshimura S, Delcourt C, Mair G, Al-Shahi Salman R, Demchuk AM, Wardlaw JM, Lindley RI, Anderson CS. Clinical prognosis of FLAIR hyperintense arteries in ischaemic stroke patients: a systematic review and meta-analysis. *J Neurol Neurosurg Psychiatry* 2020;91(5):475-82.

3. **Zhou Z**, Delcourt C, Xia C, Yoshimura S, Carcel C, Torii-Yoshimura T, You S, Malavera A, Chen X, Hackett ML, Woodward M, Chalmers J, Xu J, Robinson TG, Parsons MW, Demchuk AM, Lindley RI, Mair G, Wardlaw JM, Anderson CS. Low- versus standard-dose alteplase in acute lacunar ischaemic stroke: the ENCHANTED trial. *Neurology* 2021;96(11):e1512-6.

4. **Zhou Z**, Xia C, Carcel C, Yoshimura S, Wang X, Delcourt C, Malavera A, Chen X, Mair G, Woodward M, Chalmers J, Demchuk AM, Lindley RI, Robinson TG, Parsons MW, Wardlaw JM, Anderson CS. Intensive versus guideline-recommended blood pressure reduction in acute lacunar stroke with intravenous thrombolysis therapy: the ENCHANTED trial. *Eur J Neurol* 2021;28(3):783-93.

5. **Zhou Z**, Xia C, Mair G, Delcourt C, Yoshimura S, Liu X, Chen Z, Malavera A, Carcel C, Chen X, Wang X, Al-Shahi Salman R, Robinson TG, Lindley RI, Chalmers J, Wardlaw JM, Parsons MW, Demchuk AM, Anderson CS. Thrombolysis outcomes according to arterial characteristics of acute ischaemic stroke by alteplase dose and blood pressure target. *Int J Stroke*. 2021:17474930211025436 (Online ahead of print).

Other publications during the candidature

1. Li C, **Zhou Z**, Neuen BL, Yu J, Huang Y, Young T, Li J, Li L, Perkovic V, Jardine MJ, Keay L, Markoulli M, Rosenthal N, Capuano G, Yavin Y, Neal B, Arnott C. Sodium-glucose co-transporter-2 inhibition and ocular outcomes in patients with type 2 diabetes: a systematic review and meta-analysis. *Diabetes Obes Metab* 2021;23(1):252-7.

2. Xia C, Wang X, Lindley RI, Delcourt C, Chen X, **Zhou Z**, Guo R, Carcel C, Malavera A, Calic Z, Mair G, Wardlaw JM, Robinson TG, Anderson CS. Early decompressive hemicraniectomy in thrombolyzed acute ischemic stroke patients from the international ENCHANTED trial. Sci Rep. 2021;11(1):16495.

3. Yu J, **Zhou Z**, Mahaffey KW, Matthews DR, Neuen BL, Heerspink HJL, Jardine MJ, Li J, Perkovic V, Neal B, Arnott C. An exploration of the heterogeneity in effects of SGLT2 inhibition on cardiovascular and all-cause mortality in the EMPA-REG OUTCOME, CANVAS Program, DECLARE-TIMI 58, and CREDENCE trials. *Int J Cardiol* 2021;324:165-72.

4. **Zhou Z**, Jardine MJ, Li Q, Neuen BL, Cannon CP, de Zeeuw D, Edwards R, Levin A, Mahaffey KW, Perkovic V, Neal B, Lindley RI; CREDENCE Trial Investigators. Effect of SGLT2 inhibitors on stroke and atrial fibrillation in diabetic kidney disease: results from the CREDENCE trial and meta-analysis. *Stroke* 2021;52(5):1545-56.

5. Jardine M, **Zhou Z**, Lambers Heerspink HJ, Hockham C, Li Q, Agarwal R, Bakris GL, Cannon CP, Charytan DM, Greene T, Levin A, Li JW, Neuen BL, Neal B, Oh R, Oshima M, Pollock C, Wheeler DC, de Zeeuw D, Zhang H, Zinman B, Mahaffey KW, Perkovic V. Kidney, cardiovascular, and safety outcomes of canagliflozin according to baseline albuminuria: a CREDENCE secondary analysis. *Clin J Am Soc Nephrol* 2021;16(3):384-95.

6. Jardine MJ, **Zhou Z**, Mahaffey KW, Oshima M, Agarwal R, Bakris G, Bajaj HS, Bull S, Cannon CP, Charytan DM, de Zeeuw D, Di Tanna GL, Greene T, Heerspink HJL, Levin A, Neal B, Pollock C, Qiu R, Sun T, Wheeler DC, Zhang H, Zinman B, Rosenthal N, Perkovic V; CREDENCE Study Investigators. Renal, cardiovascular, and safety outcomes of canagliflozin by baseline kidney function: a secondary analysis of the CREDENCE randomised trial. *J Am Soc Nephrol* 2020;31(5):1128-39.

7. Delcourt C, Wang X, **Zhou Z**, Wardlaw JM, Mair G, Robinson TG, Chen X, Yoshimura S, Torii-Yoshimura T, Carcel C, Calic Z, Tan WY, Malavera A, Anderson CS, Lindley RI. Brain imaging abnormalities and outcome after acute ischaemic stroke: the ENCHANTED trial. J Neurol Neurosurg Psychiatry 2020;91(12):1290-6.

8. Rådholm K, **Zhou Z**, Clemens K, Neal B, Woodward M. Effects of sodium-glucose co-transporter-2 inhibitors in type 2 diabetes in women versus men. *Diabetes Obes Metab* 2020;22(2):263-6.

9. Chen X, Delcourt C, Sun L, **Zhou Z**, Yoshimura S, You S, Malavera A, Torii-Yoshimura T, Carcel C, Arima H, Hackett ML, Robinson T, Song L, Wang X, Lindley RI, Chalmers J, Anderson CS; ENCHANTED Investigators. Brain imaging signs and health-related quality of life after acute ischaemic stroke: analysis of ENCHANTED alteplase dose arm. *Cerebrovasc Dis* 2020;49(4):427-36.

10. Xia C, Wang X, Lindley RI, Delcourt C, **Zhou Z**, Chen X, Carcel C, Malavera A, Calic Z, Anderson CS; ENCHANTED Investigators. Combined utility of blood glucose and white blood cell in predicting outcome after acute ischaemic stroke: The ENCHANTED trial. *Clin Neurol Neurosurg* 2020;198:106254.

11. **Zhou Z**, Lindley RI, Rådholm K, Jenkins B, Watson J, Perkovic V, Mahaffey KW, de Zeeuw D, Fulcher G, Shaw W, Oh R, Desai M, Matthews DR, Neal B. Canagliflozin and stroke in type 2 diabetes mellitus. *Stroke* 2019;50(2):396-404.

12. Morotti A, Boulouis G, Dowlatshahi D, Li Q, Barras CD, Delcourt C, Yu Z, Zheng J, **Zhou Z**, Aviv RI, Shoamanesh A, Sporns PB, Rosand J, Greenberg SM, Al-Shahi Salman R, Qureshi AI, Demchuk AM, Anderson CS, Goldstein JN, Charidimou A; International NCCT ICH Study Group. Standards for detecting, interpreting, and reporting noncontrast computed tomographic markers of intracerebral haemorrhage expansion. *Ann Neurol* 2019;86(4):480-92.

13. **Zhou Z**, Jardine M, Perkovic V, Matthews DR, Mahaffey KW, de Zeeuw D, Fulcher G, Desai M, Oh R, Simpson R, Watts NB, Neal B. Canagliflozin and fracture risk in individuals with type 2 diabetes: results from the CANVAS Program. *Diabetologia* 2019;62(10):1854-67.

14. Xu Y, Shanthosh J, **Zhou Z**, Somerville E, Anderson CS, Glozier N, Hackett ML. Prevalence of driving and traffic accidents among people with seizures: a systematic review. *Neuroepidemiology* 2019;53(1-2):1-12.

15. Xu Y, **Zhou Z**, Shanthosh J, Hackett ML, Anderson CS, Glozier N, Somerville E. Who is driving and who is prone to have traffic accidents? A systematic review and meta-analysis among people with seizures. *Epilepsy Behav* 2019;94:252-7.

16. Badve MS, **Zhou Z**, van de Beek D, Anderson CS, Hackett ML. Frequency of post-stroke pneumonia: systematic review and meta-analysis of observational studies. *Int J Stroke* 2019;14(2):125-36.

17. Ge X, Zhao H, **Zhou Z**, Li X, Sun B, Wu H, Wan J, Xu J, Villablanca JP, Liu X. Association of Fractional Flow on 3D-TOF-MRA with cerebral perfusion in patients with MCA stenosis. *Am J Neuroradiol* 2019;40(7):1124-31.
18. **Zhou Z**, Yu J, Carcel C, Delcourt C, Shan J, Lindley RI, Neal B, Anderson CS, Hackett ML. Resuming anticoagulants after anticoagulation-associated intracranial haemorrhage: systematic review and meta-analysis. *BMJ Open* 2018;8(5):e019672.
19. Badve MS, **Zhou Z**, Anderson CS, Hackett ML. Effectiveness and safety of antibiotics for preventing pneumonia and improving outcome after acute stroke: systematic review and meta-analysis. *J Stroke Cerebrovasc Dis* 2018;27(11):3137-47.
20. Zhang F, **Zhou Z**, Tang D, Zheng D, Cheng J, Lin L, Xu J, Zhao X, Wu H. Diffusion-weighted MRI in solitary pulmonary lesions: associations between apparent diffusion coefficient and multiple histopathological parameters. *Sci Rep* 2018;8(1):11248.
21. Ladwig S, **Zhou Z**, Xu Y, Wang X, Chow CK, Werheid K, Hackett ML. Comparison of treatment rates of depression after stroke versus myocardial infarction: a systematic review and meta-analysis of observational data. *Psychosom Med* 2018;80(8):754-63.

Conference presentations

1. **Zien Zhou**, Candice Delcourt, Chao Xia, Sohei Yoshimura, Cheryl Carcel, Takako Torii-Yoshimura, Shoujiang You, Alejandra Malavera, Xiaoying Chen, Maree L. Hackett, Mark Woodward, John Chalmers, Jianrong Xu, Thompson G. Robinson, Mark W. Parsons, Andrew M. Demchuk, Richard I. Lindley, Grant Mair, Joanna M. Wardlaw, Craig S. Anderson. Low- versus standard-dose alteplase in acute lacunar ischaemic stroke: the ENCHANTED trial. 2021 International Stroke Conference, USA, 17-19 March 2021, moderated poster (virtual online).
2. **Zien Zhou**, Alejandra Malavera, Sohei Yoshimura, Candice Delcourt, Grant Mair, Rustam Al-Shahi Salman, Andrew M Demchuk, Joanna M Wardlaw, Richard I Lindley, Craig S Anderson. Clinical prognosis of FLAIR hyperintense arteries in ischaemic stroke patients: a systematic review and meta-analysis. 2020 European Stroke Organisation and the World Stroke Organization Jointing Conference, Vienna, Austria, 7-9 Nov 2020, E-poster (virtual online).
3. **Zien Zhou**, Meg J. Jardine, Qiang Li, Brendon L. Neuen, Christopher P. Cannon, Dick de Zeeuw, Robert Edwards, Adeera Levin, Kenneth W. Mahaffey, Vlado Perkovic, Bruce

Neal, Richard I. Lindley. Canagliflozin and stroke in diabetic kidney disease: results from the CREDENCE trial. the American College of Cardiology (ACC) 69th annual scientific session and expo, Chicago, USA, 29-30 Mar 2020, E-poster (virtual online).

4. **Zien Zhou**, Meg J. Jardine, Hiddo Heerspink, Brendon L. Neuen, Qiang Li, Clare Arnott, Kenneth W. Mahaffey, Vlado Perkovic, Bruce Neal. Effects of SGLT2 inhibitors on stroke in type 2 diabetes according to baseline kidney function. the American College of Cardiology (ACC) 69th annual scientific session and expo, Chicago, USA, 29-30 Mar 2020, E-poster (virtual online).
5. **Zhou Z**, Yoshimura S, Delcourt C, Lindley RI, You S, Malavera A, Torii-Yoshimura T, Carcel C, Wang X, Chen X, Parsons MW, Demchuk AM, Wardlaw JM, Mair G, Robinson TG, Chalmers J, Xu J, Anderson CS. Thrombolysis outcomes in acute ischaemic stroke by fluid-attenuated inversion recovery hyperintense arteries. Stroke Society of Australasia scientific meeting 2019, Canberra, Australia, 10-13 September 2019, oral presentation.
6. **Zien Zhou**, Sohei Yoshimura, Candice Delcourt, Richard I Lindley, Shoujiang You, Alejandra Malavera, Takako Torii-Yoshimura, Cheryl Carcel, Xia Wang, Xiaoying Chen, Mark W Parsons, Andrew M Demchuk, Joanna M Wardlaw, Grant Mair, Thompson G Robinson, John Chalmers, Jianrong Xu, Craig S Anderson. Thrombolysis outcomes in acute ischaemic stroke by fluid-attenuated inversion recovery hyperintense arteries. 5th European Stroke Organisation Conference, Milan, Italy, 22-24 May 2019, E-poster.

Scholarships and awards

1. UNSW Sydney Scientia PhD Scholarship (March 2018 to March 2022).
2. Awarded Paul Dudley White International Scholar Award by the American Heart Association in March 2021 for the highest ranked abstract from Australia at the International Stroke Conference 2021.

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List of Abbreviations

ACA: anterior cerebral artery

ACEI: angiotensin converting enzyme inhibitor

ADC: apparent diffusion coefficient

AF: atrial fibrillation

AI: artificial intelligence

AIC: acute ischaemic change

AIS: acute ischaemic stroke

AOL: arterial occlusive lesion score

AOR: adjusted odds ratio

ASPECTS: Alberta Stroke Program Early CT score

BA: basilar artery

BP: blood pressure

CAA: cerebral amyloid angiopathy

CAD: coronary artery disease

CBF: cerebral blood flow

CBV: cerebral blood volume

CE-MRA: contrast-enhanced magnetic resonance angiography

CI: confidence interval

CT: computed tomography

CTA: computerised tomographic angiography

DBP: diastolic blood pressure

DICOM: Digital Imaging and Communications in Medicine

DM: diabetes mellitus

DWI: diffusion-weighted imaging

ECASS: European-Australian Cooperative Acute Stroke Study

ENCHANTED: Enhanced Control of Hypertension and Thrombolysis Stroke Study

END: early neurological deterioration

EVT: endovascular therapy

FLAIR: fluid-attenuated inversion recovery sequence

FLAIR-HAs: FLAIR hyperintense arteries

FU: follow-up

GCS: Glasgow coma scale

HI: haemorrhagic infarcts

ICA: internal carotid artery

ICAO: internal carotid artery occlusion (or obstruction)

ICH: intracerebral haemorrhage

IQR: interquartile range

IST-3: the third International Stroke Trial

LVO: large vessel occlusion (or obstruction)

MCA: middle cerebral artery

MRA: magnetic resonance angiography

MRI: magnetic resonance imaging

mRS: modified Rankin scale

MTT: mean transit time

MVO: medium vessel occlusion (or obstruction)

NIHSS: National Institutes of Health Stroke Scale

NINDS: National Institutes of Neurological Diseases and Stroke

NNCT: non-contrasted computed tomography

NVO: no vessel occlusion (or obstruction)

OCSP: the Oxfordshire Community Stroke Project

OR: odds ratio

PCA: posterior cerebral artery

PH: parenchymal haematomas

PWI: perfusion-weighted imaging

RR: risk ratio

SE/GRE: spin echo/gradient recalled echo sequence

rtPA: recombinant human tissue plasminogen activator

SBP: systolic blood pressure

SD:	standard deviation
sICH:	symptomatic intracerebral haemorrhage
SITS-MOST:	Safe Implementation of Thrombolysis in Stroke Monitoring Study
TGI:	The George Institute for Global Health
TIA:	transient ischaemic attack
TICI:	thrombolysis in cerebral infarction
TIMI:	thrombolysis in myocardial infarction grade.
T_{max}:	time to maximum
TOAST:	The Trial of ORG 10172 in Acute Stroke Treatment classification
TTP:	time to peak
WAKE-UP:	The Efficacy and Safety of MRI-based Thrombolysis in Wake-Up Stroke trial

Chapter 1: Introduction

Stroke is a leading cause of death and disability. It induces a heavy disease burden globally. The Global Burden of Disease Study 2016 showed that there were 5.5 (95% uncertainty interval [UI] 5.3 to 5.7) million deaths and 116.4 (95% UI 111.4 to 121.4) million years lived with disability due to stroke in 2016, worldwide.[1] In China, the most populous country, stroke was the first cause of years of life lost in 2017, although there was a decrease in disability-adjusted life-years from 1990 to 2017.[2] Furthermore, there are enormous direct and indirect economic losses due to the treatment and rehabilitation in stroke patients: the total costs of post-stroke care for inpatient and outpatient were highest in the USA ($4850 per patient-month) and lowest in Australia ($752 per patient-month) according to a recent systematic review of related economic data published from January 2000 to August 2016.[3] Thus, strategies to prevent stroke by modifying conventional risk factors (such as high blood pressure [BP], diabetes mellitus, hyperlipaemia, atrial fibrillation, smoking) and effective treatments to reduce poor prognosis after stroke are urgently needed, albeit challenging.

There are two main pathological subtypes (ischaemic and haemorrhagic) of stroke, and a majority of stroke patients (80%) have an ischaemic stroke, caused by the obstruction/occlusion of carotid or a cerebral artery (named as "culprit artery"), resulting in a reduction of downstream cerebral blood flow and dysfunction of corresponding cerebral tissue. Growth or rupture of atherosclerotic plaque and cardiogenic embolus usually account for forming the "culprit artery." Clinical symptoms after ischaemic stroke onset include face dropping on one side, limb paresis, vision disturbance, dizziness/vertigo, difficulty in speaking and understanding speech, and disability of walking. Modern treatment for acute ischaemic stroke (AIS) is based on the principle that a better recovery of cerebral function and a more favourable prognosis depends on earlier recanalisation of the "culprit artery" and quicker reperfusion of the ischaemic brain area. The first approved treatment for AIS is intravenous thrombolysis using recombinant tissue plasminogen activator (rt-PA, alteplase), converting plasminogen to plasmin followed by breaking down fibrin, an essential protein for the formation of thrombus.[4] In 1996, based on the National Institute of Neurological Disorders and Stroke (NINDS) trial,[5] alteplase was licensed in North America for intravenous use in AIS patients within 3 hours after stroke onset. Since then, there have been rapid developments in the strategies of AIS administration. Some treatments have been applied in clinical practice based on large-scale randomised clinical trials

or related meta-analysis with confirmed results, such as extending the time window of intravenous thrombolysis to 4.5 hours,[6-8] dual antiplatelet therapy (clopidogrel plus aspirin) in minor AIS or transient ischaemic attack,[9-11] endovascular treatment for large vessel occlusion,[12-18] and multi-model imaging-guided intravenous thrombolysis in wake-up stroke patients or endovascular treatment.[18-23] Meanwhile, the efficacy and safety of other promising interventions are still in debate due to limited evidence from high-quality clinical trials or conflicting results in observational studies, such as new drugs of intravenous thrombolysis, low-dose intravenous alteplase use, intensive BP management in candidates for intravenous thrombolysis or endovascular treatment, and the necessity of intravenous thrombolysis before endovascular treatment.

The Enhanced Control of Hypertension and Thrombolysis Stroke Study (ENCHANTED) was the first large-scale, international, multicentre, 2×2 quasi-factorial, prospective, randomised, open-label, blinded-endpoint trial that assessed the effectiveness of low-dose (0.6 mg/kg; 15% as bolus, 85% as infusion during 1 hour) versus standard-dose (0.9 mg/kg; 10% as bolus, 90% as infusion during 1 hour) intravenous alteplase (alteplase arm), and more intensive BP lowering (target systolic BP [SBP] 130-140 mmHg within 1 hour after randomisation) versus guideline-recommended BP control (SBP <180 mmHg) (BP arm) in adult patients with AIS.[24-28] The main result papers of the ENCHANTED trial are outlined in Appendices A and B. In August 2016, the ENCHANTED Imaging Project was set out to explore associations of imaging signs with thrombolysis outcomes in AIS patients and identify any imaging signs that would modify the randomised treatment effects of alteplase dose and BP target. After receiving standardised training whereby each reader completed the ACCESS (Acute Cerebral CT Evaluation of Stroke Study) training module of recognised acute and pre-existing AIS imaging features (http://www.ed.ac.uk/edinburgh-imaging/access), a research team with a background in stroke but not necessarily imaging interpretation expertise (non-expert: one radiologist [myself], eight-stroke neurologists and two-stroke neurology trainees) assessed the presence, extent, and severity of, and swelling from, acute ischaemic changes (including arterial territory, border zone, small subcortical and brainstem/cerebellar infarcts), coexisting old vascular lesions and their subtypes, white matter lesions, and brain volume loss, on all collected images from the ENCHANTED trial, using an electronic scoring system modified from the third International Stroke Trial (IST-3).[29] Since September 2019, the team also assessed the presence, location, extent (IST-3 angiography score and modified Thrombolysis in Cerebral Infarction [TICI] score),[30] clot burden score,[31] visual residual flow,[32] the collateral score of vascular

obstruction[33] on collected baseline computed tomographic angiography (CTA) or magnetic resonance angiography (MRA), and recanalisation (revised Arterial Occlusive Lesion [rAOL] score)[32] after alteplase treatment in participants with both baseline and follow-up angiography. Copies of the ENCHANTED image interpretation forms are list in Appendices C and D.

This thesis is based on clinical and adjudicated imaging data from both arms of the ENCHANTED trial. This Chapter provides an overview of prior studies on which my research is based. Firstly, I perform a literature review on the association of alteplase dose and thrombolysis outcomes, followed by an overview of the current status of BP management in AIS candidates for intravenous thrombolysis and an introduction to AIS-related imaging signs. Finally, I present an outline of gaps in current research, the objectives of my studies, and a description of thesis structure.

1.1 Alteplase dose and thrombolysis outcomes in AIS

1.1.1 Optimal alteplase dose from early pilot studies

In the early 1990s, before carrying out the NINDS trial, two pilot studies investigated the optimal dose of alteplase for its urgent use in AIS with an open-label, dose-escalation design. The first study included 74 adult AIS patients (18 to 80 years) within 1.5 hours after symptom onset and grouped according to different doses of alteplase received: 0.35 mg/kg (n=6), 0.60 mg/kg (n=12), 0.85 mg/kg (n=30), 0.95 mg/kg (n=25) and 1.08 mg/kg (n=1).[34] The results showed there were numerically more patients with major neurological improvement at 24 hours post-treatment in high-dose groups (0.85 mg/kg, 55%; 0.95 mg/kg, 50%) than in low-dose groups (0.35 mg/kg, 33%; 0.6 mg/kg, 33%) (statistical test not reported). No intracerebral parenchymal haematoma or other major haemorrhagic complications occurred in dose groups of ≤0.85 mg/kg, but they were observed in the remaining groups with the higher dose. The second study investigated the efficacy and safety of alteplase in 20 adult AIS patients who had longer time intervals (1.5 to 3 hours) from symptom onset to treatment.[35] Participants were grouped as 0.6 mg/kg (n=8), 0.85 mg/kg (n=6) and 0.95 (n=6) mg/kg. No intracerebral haemorrhage (ICH) occurred in the 0.6 mg/kg group, and each of the other two groups had one case of ICH (17%). The number (percentage) of significant neurological improvements at 24 hours in the three groups were 2 (25%), 1 (17%), and 0 (0%), respectively (statistical test not reported).

These two early pilot studies suggested that intravenous thrombolysis by alteplase with a dose of ≤0.95 mg/kg was relatively safe and effective for early neurological improvement. Subsequently, the NINDS trial, the first randomised, double-blind study to assess the effectiveness of alteplase versus placebo in AIS within 3 hours of onset, adopted a dose of 0.9 mg/kg. It confirmed the efficacy of alteplase on 90-day improved functional outcome post-randomisation, despite an increase of symptomatic ICH (sICH).[5] Unfortunately, there had been no randomised trial to compare 0.9 mg/kg versus a lower dose of alteplase directly in the 1990s because of no approval for funding.

1.1.2 Japan approved the use of low-dose alteplase (0.6 mg/kg) in AIS

From 2002 to 2010, a series of cohort studies in Japanese AIS patients were conducted to investigate the association of low-dose (0.6 mg/kg) intravenous alteplase with 90-day functional outcome and the incidence of early symptomatic ICH (sICH): Japan Alteplase Clinical Trial (J-ACT),[36] Stroke Acute Management with Urgent Risk-factor Assessment and Improvement Study (SAMURAI),[37] Japan Alteplase Clinical Trial II (J-ACT II),[38] and the Japan post-Marketing Alteplase Registration Study (J-MARS).[39] Based on comparable proportions of functional recovery and sICH between the J-ACT and earlier studies conducted in the USA and Europe where 0.9 mg/kg alteplase was adopted, Japan approved the intravenous use of 0.6 mg/kg alteplase (the only dose) in AIS within 3 hours after stroke onset in October 2005. Subsequent SAMURAI, J-ACT II, and J-MARS, with larger samples or in patients with large vessel occlusion (LVO) identified by image, further suggested the safety and efficacy of its use in routine clinical practice for Japanese AIS patients. Table 1.1 summarises these four cohort studies and contemporary or earlier randomised placebo-controlled trials whereby 0.9 mg/kg alteplase was adopted. A direct comparison of 0.6 mg/kg versus 0.9 mg/kg intravenous alteplase or 0.6 mg/kg versus placebo through a randomised control trial (RCT) was never undertaken in Japan due to concern over ethics and because RCTs conducted in the 1990s had identified the superiority of 20 million international units (MIU) of duteplase (equal to 0.6 mg/kg alteplase) versus placebo with respect to clinical improvement after AIS.[40-42]

1.1.3 Controversy on low-dose alteplase (0.6 mg/kg) for AIS in other Asian countries

Some pathophysiological mechanisms support the use of low-dose intravenous alteplase in Asian patients with AIS: these include race difference in the levels of plasma fibrinogen, plasminogen activator inhibitor-I, and coagulation factor XIII activity (lower in Japanese than

in Caucasians);[46-48] different composition of AIS causes between Asians and Caucasians (intracranial atherosclerosis being the primary cause of AIS in Asian patients);[49,50] and genetic differences.[51-53] Furthermore, the use of 0.6 mg/kg alteplase can save half of the cost compared with 0.9 mg/kg, ideal for patients with limited income in developing Asian countries.

After approving 0.6 mg/kg intravenous alteplase use for AIS in Japan, researchers from other Asian countries investigated its efficacy and safety for patients in their own countries, with controversial results. An observational study conducted in Singapore (where multi-ethnic Asians) did not favour low-dose intravenous alteplase for AIS.[54] The results showed standard-dose intravenous alteplase (0.9 mg/kg) was associated with a lower rate of sICH (1.2%) and higher rate of 90-day functional independence (59%) compared with the low-dose (0.5 mg/kg) group (14.5% and 35%, respectively). As assumed by the authors, a delay in recanalisation by low-dose alteplase might cause an increased risk of sICH and disability after thrombolysis.[55] According to a search strategy made by our team,[56] I identified 15 observational studies from Asian countries (except Japan) after searching in Medline and Embase from inception to the end of December 2020. Seven studies favoured low-dose intravenous alteplase (1 from Vietnam, 1 from Pakistan, and 5 in the Chinese population),[57-63] three studies favoured standard-dose alteplase (1 from Singapore and 2 in the Chinese population),[54,64,65] and five studies showed neutral results (1 from South Korea and 4 in the Chinese population).[66-70] Table 1.2 summarises these studies. Potential reasons for the controversial results include various definitions of low-dose alteplase across studies, differences in the expertise of intravenous thrombolysis across hospitals, small samples in most of the studies, and bias caused by being prone to assign AIS patients with a high risk of ICH to receive low-dose alteplase.

1.1.4 Alteplase arm of the ENCHANTED trial

The alteplase arm of the ENCHANTED trial firstly investigated the association of low-dose versus standard-dose intravenous alteplase with thrombolysis outcomes in AIS patients with an international, multicentre, prospective, randomised, open-label, blinded-endpoint design.[24-26] From March 2012 to August 2015, 3310 AIS patients (63% Asians) recruited at 111 clinical centres from 13 countries and randomised to receive low-dose intravenous alteplase (N=1654; 0.6 mg/kg, 15% as bolus, 85% as infusion during 1 hour) or standard-dose intravenous alteplase (N=1643; 0.9 mg/kg, 10% as bolus, 90% as infusion during 1 hour) within 4.5 hours after symptom onset. The study aimed to identify the noninferiority between the two-dose groups with respect to death or disability (primary outcome, defined as modified Rankin scale [mRS]

scores 2-6) and ordinal mRS scores shift (secondary outcome) at 90-day post-randomisation and to determine whether low-dose intravenous alteplase was superior to standard-dose with respect to sICH (safety outcome, defined by SITS-MOST criteria). Other outcomes included major disability (mRS scores 3-6) at 90-day post-randomisation, mortality at 7 and 90 days, and early neurologic deterioration (END, increase of ≥4 points in the NIHSS score) within 72 hours post-randomisation. The results, published in 2016, did not show the noninferiority of low-dose versus standard-dose intravenous alteplase for the primary outcome (odds ratio [OR], 1.09; 95% confidence interval [CI], 0.95-1.25) (upper limit of 95% CI > noninferiority margin of 1.14). The noninferiority between the two-dose groups for 90-day ordinal mRS scores shift was confirmed (OR 1.00, 95% CI 0.89-1.13). Low-dose alteplase significantly reduced the risk of sICH (OR 0.48 0.27-0.86).

Several pre-specified or post-hoc subgroup analyses from ENCHANTED by baseline characteristics were performed to identify what kind of AIS patients would benefit from low-dose intravenous alteplase. Clinical factors (age, ethnicity, severity, prior stroke, and diabetes mellitus, prior treatment of lipid-lowering agents, and kidney function) did not modify the randomised treatment effects of low-dose versus standard-dose alteplase on 90-day functional outcomes,[71-75] except for prior antiplatelet treatment that low-dose alteplase improved the 90-day functional outcome in thrombolysis-treated AIS patients with prior antiplatelet therapy (mRS score shift: no prior antiplatelet therapy OR 1.07, 95% CI 0.93-1.23, with prior antiplatelet therapy OR 0.76, 95% CI 0.59-0.98, $P_{interaction}$ =0.02).[76] An observational study based on the ENCHANTED data showed a combination of favourable clinical characteristics, including younger age, lower SBP, mild neurological impairment, and no atrial fibrillation, diabetes mellitus, or premorbid symptoms predicted a net advantage from low-dose alteplase compared with standard-dose. Corresponding logistic regression models were also validated externally using real-world registry data.[77] However, direct application of these models in clinical practice is infeasible since cut-off values of these factors to predict good prognosis by low-dose alteplase were not provided. In addition, the models did not involve critical factors from neuroimaging data, such as recanalisation, collateral circulation, infarct size, and location.

1.1.5 Meta-analysis of low-dose versus standard-dose alteplase in AIS

There were seven systematic reviews and meta-analyses on low-dose versus standard-dose alteplase in AIS published from 2013 to 2019, including one involving individual participant

Table 1.1. Studies by which Japan approved the use of low-dose (0.6 mg/kg) intravenous alteplase in AIS and contemporary or earlier studies in the USA and Europe

Source	Location and conducting period	Study design	Included criteria	rt-PA dose	Number of patients	Functional outcome at 3 months, n (%)	Safety outcome, n (%)
Studies in Japan							
J-ACT, 2006 (36)	Japan, 04/2002-09/2003	Prospective cohort	As in the NINDS trial	0.6 mg/kg	103	mRS scores 0-1: 38 (36.9)	sICH within 36 hrs: 6 (5.8) Death within 3 months: 10 (9.7)
SAMURAI, 2009 (37)	Japan, 10/2005-07/2008	Retrospective cohort	As in the NINDS trial	0.6 mg/kg	600	mRS scores 0-1: 199 (33.2)	sICH within 36 hrs: 23 (3.8) Any ICH within 36 hrs: 119 (19.8) Death within 3 months: 43 (7.2)
J-ACT II, 2010 (38)	Japan, 03/2007-07/2008	Prospective cohort	AIS patients within 3 hours of onset & MRA identified MCA occlusion	0.6 mg/kg	58	mRS scores 0-1: 27 (46.6)	sICH within 36 hrs: 0 (0.0) Death within 3 months: 1 (1.7)
J-MARS, 2010 (39)	Japan, 10/2005-10/2007	Prospective cohort	AIS patients within 3 hours of onset who received 0.6 mg/kg intravenous alteplase in Japan	0.6 mg/kg	7492	mRS scores 0-1: 1637/4944 (33.1)	sICH within 36 hrs: 259 (3.5) sICH at 3 months: 329 (4.4) Death within 3 months: 985 (13.1)
Contemporary or earlier studies in the USA and Europe							
NINDS Part 2, 1995 (5)	USA, 01/1991-10/1994	Randomised control trial	AIS patients with known time of onset, a deficit measurable on the NIHSS and no intracranial haemorrhage on CT scan	0.9 mg/kg	168	mRS scores 0-1: 39 (23.2)	sICH: 12 (7.1); Any ICH: 21 (12.5) [within 36 hrs]
				Placebo	165	mRS scores 0-1: 26 (15.8)	sICH: 2 (1.2); Any ICH: 8 (4.8) [within 36 hrs]
ECASS II, 1998 (43)	Europe, Australia, and New Zealand 10/1996-01/1998	Randomised control trial	AIS patients (18-80 years) who could be treated within 6 hrs of symptom onset, no or only minor early signs of infarction on the initial CT scan	0.9 mg/kg	409	mRS scores 0-1: 165 (40.3)	PH2: 33 (8.1); PH1+PH2: 48 (11.8) Any ICH: 197 (48.4) [within 7 days]
				Placebo	391	mRS scores 0-1: 143 (36.6)	PH2: 3 (0.8); PH1+PH2: 12 (3.1) Any ICH: 155 (40.2) [within 7 days]

Source	Location and conducting period	Study design	Included criteria	rt-PA dose	Number of participants	Functional outcome at 3 months, n (%)	Safety outcome, n (%)
Contemporary or earlier studies in the USA and Europe							
SITS-MOST, 2007 (44)	Europe, 12/2002-04/2006	Prospective cohort	AIS patients (18-80 years) within 3 hrs of stroke onset	0.9 mg/kg	6483	mRS scores 0-2: 3362/6136 (54.8)	sICH within 24 hrs: 107/6444 (1.7) sICH at 7 days: 468/6438 (7.3) Death within 3 months: 701/6218 (11.3)
ECASS III, 2008 (7)	Europe, 07/2003-11/2007	Randomised control trial	AIS patients (18-80 years), onset of symptoms 3 to 4.5 hrs before initiation of study-drug	0.9 mg/kg	418	mRS scores 0-1: 219 (52.4)	sICH: 10 (2.4) Any ICH: 113 (27.0) Death within 3 months: 28 (6.7)
				Placebo	403	mRS scores 0-1: 182 (45.2)	sICH: 1 (0.2) Any ICH: 71 (17.6) Death within 3 months: 31 (7.7)
IST-3, 2012 (45)	12 countries, 05/2000-07/2011	Randomised control trial	AIS patients; the time of stroke onset was known; treatment could be started within 6 hrs of onset; no intracranial haemorrhage and structural brain lesions on CT or MRI	0.9 mg/kg	1515	OHS 0-2 at 6 months: 554 (36.6)	sICH within 7 days: 104 (6.9) Death within 7 days: 163 (11) Death at 6 months: 408 (26.9)
				Placebo	1520	OHS 0-2 at 6 months: 534 (35.1)	sICH at 7 days: 16 (1.1) Death within 7 days: 107 (7.0) Death at 6 months: 407 (26.8)

AIS denotes acute ischaemic stroke; CT, computerised tomography; ECASS II, The Second European Cooperative Acute Stroke Study; ECASS III, The Third European Cooperative Acute Stroke Study; ICH, intracerebral haemorrhage; IST-3, The Third International Stroke Trial; J-ACT, Japan Alteplase Clinical Trial; J-ACT II, Japan Alteplase Clinical Trial II; J-MARS, The Japan post-Marketing Alteplase Registration Study; NIHSS, National Institute of Health stroke scale; NINDS, The National Institute of Neurological Disorders and Stroke rt-PA Stroke Study; MCA, middle cerebral artery; MRA, magnetic resonance angiography; MRI, magnetic resonance imaging; mRS, modified Rankin scale; OHS, Oxford Handicap Score; PH, parenchymal haemorrhage; rt-PA: recombinant tissue-type plasminogen activator; SAMURAI, Stroke Acute Management with Urgent Risk-factor Assessment and Improvement Study; sICH, symptomatic intracerebral haemorrhage; SITS-MOST, The Safe Implementation of Thrombolysis in Stroke-Monitoring Study.

Table 1.2. Studies from Asian countries (except Japan) on thrombolysis outcomes by low-dose intravenous alteplase in AIS

Source	Location and conducting period	Study design	Included criteria	rt-PA dose	Number of patients	Functional outcome at 3 months, n (%)	Safety outcome, n (%)
Favoured low-dose intravenous alteplase							
Chao et al, 2010 (57)	Taiwan China, 12/2004-07/2008	Prospective cohort	AIS patients who were admitted to Taiwan hospitals and used alteplase to treat	0.7 mg/kg	116	mRS scores 0-2: 60/102 (58.8)	sICH* within 3 months: 6 (5.2) Death within 3 months: 8 (6.9)
				0.9 mg/kg	125	mRS scores 0-2: 57/117 (48.7)	sICH* within 3 months: 13 (10.4) Death within 3 months: 16 (12.8)
Nguyen et al, 2010 (58)	Vietnam, 05/2006-05/2009	Prospective cohort	As in the NINDS trial	0.6 mg/kg	48	mRS scores 0-1: 27 (56.3)	sICH within 24 hrs: 1 (2.1) Death within 3 months: 1 (2.1)
				0.9 mg/kg	73	mRS scores 0-1: 25 (34.2)	sICH within 24 hrs: 4 (5.5) Death within 3 months: 9 (12.3)
Chao et al, 2014 (59)[†]	Taiwan China, 12/2004-11/2011	Prospective cohort	As in the SITS-MOST study, including an upper age limit of 80 years	0.6 mg/kg	181	mRS scores 0-1: 56/146 (38.4)	sICH* within 3 months: 10 (5.5) Death within 3 months: 14 (7.7)
				0.7 mg/kg	199	mRS scores 0-1: 44/156 (28.2)	sICH* within 3 months: 10 (5.0) Death within 3 months: 19 (9.6)
				0.8 mg/kg	202	mRS scores 0-1: 46/171 (26.9)	sICH* within 3 months: 12 (5.9) Death within 3 months: 18 (8.9)
				0.9 mg/kg	422	mRS scores 0-1: 124/367 (33.8)	sICH* within 3 months: 31 (7.3) Death within 3 months: 35 (8.3)

AIS denotes acute ischaemic stroke; ICH, intracerebral haemorrhage; mRS, modified Rankin scale; NIHSS, National Institute of Health stroke scale; NINDS, The National Institute of Neurological Disorders and Stroke rt-PA Stroke Study; NR, not reported; sICH, symptomatic intracerebral haemorrhage; SITS-MOST, The Safe Implementation of Thrombolysis in Stroke-Monitoring Study.

Source	Location and conducting period	Study design	Included criteria	rt-PA dose	Number of patients	Functional outcome at 3 months, n (%)	Safety outcome, n (%)
Zheng et al, 2016 (60)[†]	Mainland China, 06/2012-02/2014	Retrospective cohort	AIS patients admitted within 4.5 hrs after onset; without intracranial haemorrhage; NIHSS scores 4-22.	0.6-0.7[‡] mg/kg	90	mRS scores 0-1: 65 (72.2)	ICH: 0 (0.0) Death within 3 months: 2 (2.2)
				0.6-0.7[§] mg/kg	90	mRS scores 0-1: 49 (54.4)	ICH: 0 (0.0) Death within 3 months: 3 (3.3)
				0.9 mg/kg	40	mRS scores 0-1: 22 (55.0)	ICH: 0 (0.0) Death within 3 months: 0 (0.0)
Ong et al, 2017 (61)	Taiwan China, 01/2007-12/2015	Retrospective cohort	AIS patients (aged ≤80 years) who received intravenous alteplase ≤0.9 mg/kg	0.6 mg/kg	71	mRS scores 0-2 at 6 months: 32/63 (50.8)	sICH at discharge: 1 (1.4)
				0.7 mg/kg	59	mRS scores 0-2 at 6 months: 32/57 (56.1)	sICH at discharge: 5 (8.5)
				0.8 mg/kg	88	mRS scores 0-2 at 6 months: 55/85 (64.7)	sICH at discharge: 8 (9.1)
				0.9 mg/kg	56	mRS scores 0-2 at 6 months: 21/55 (38.2)	sICH at discharge: 12 (21.4)
Zhao et al, 2018 (62)	Mainland China, 07/2012-12/2016	Retrospective cohort	AIS patients (18-80 years) within 4.5 hrs of onset; no ICH; NIHSS scores <25; normal activated partial thromboplastin time, prothrombin time, and fibrinogen.	0.6-0.89 mg/kg	1115	mRS scores 0-1: 402 (36.1)	sICH within 24 hrs: 25 (2.2) Death within 3 months: 61 (5.5)
				0.9 mg/kg	372	mRS scores 0-1: 140 (37.6)	sICH within 24 hrs: 22 (5.9) Death within 3 months: 27 (7.3)
Zaman Babar et al, 2019 (63)	Pakistan, 01/2007-10/2016	Retrospective cohort	AIS patients who received intravenous altepalse	0.6 mg/kg	79	mRS scores 0-2: 40 (50.6)	sICH*: 3 (3.8) Death within 3 months: 1 (1.3)

Source	Location and conducting period	Study design	Included criteria	rt-PA dose	Number of patients	Functional outcome at 3 months, n (%)	Safety outcome, n (%)
Favoured standard-dose intravenous alteplase							
Sharma et al, 2010 (54)	Singapore, 01/2000-05/2008	Retrospective cohort	As in the NINDS trial	0.5 mg/kg	48	mRS scores 0-1: 17 (35.4)	sICH within 24 hrs: 7 (14.5) Death within 3 months: 5 (10.4)
				0.9 mg/kg	82	mRS scores 0-1: 48 (58.5)	sICH within 24 hrs: 1 (1.2) Death within 3 months: 11 (13.4)
Liao et al, 2014 (64)	Mainland China, 05/2007-04/2012	Prospective cohort	Patients presenting with AIS who were given intravenous alteplase within 4.5 hours after symptom onset (as in the SITS-MOST study)	0.5-0.7 mg/kg	75	mRS scores 0-1: 31/74 (41.9)	sICH* within 36 hrs: 0 (0.0) Death within 3 months: 4/74 (5.4)
				0.7-0.85 mg/kg	131	mRS scores 0-1: 61/127 (48.0)	sICH* within 36 hrs: 12 (9.2) Death within 3 months: 11/127 (8.7)
				0.9 mg/kg	678	mRS scores 0-1: 358/665 (53.8)	sICH* within 36 hrs: 33 (4.9) Death within 3 months: 49/666 (7.4)
Liu et al, 2019 (65)	Mainland China, 05/2007-04/2012	Prospective cohort	AIS patients (70-80 years) with alteplase treatment within 4.5 hrs after onset; with diabetes or serum glucose >9.0 mmol/L; NIHSS score >20; or with cardioembolism	0.5-0.7 mg/kg	60	mRS scores 0-1: 17 (28.3)	sICH* within 36 hrs: 3 (5.0) Death within 3 months: 11 (18.3)
				0.85-0.95 mg/kg	494	mRS scores 0-1: 209 (42.3)	sICH* within 36 hrs: 30 (6.1) Death within 3 months: 66 (13.4)
Comparable between low-dose and standard-dose alteplase							
Zhou et al, 2010 (66)	Mainland China, 1998-2008	Retrospective cohort	As in the NINDS trial (AIS patients who were given alteplase within 4.5 hours)	0.6-0.7 mg/kg	23	mRS scores 0-1: 8 (34.8)	sICH* within 24 hrs: 1 (4.3) Death within 3 months: 4 (17.4)
				0.8 mg/kg	31	mRS scores 0-1: 12 (38.7)	sICH* within 24 hrs: 3 (9.7) Death within 3 months: 5 (16.1)
				0.9 mg/kg	51	mRS scores 0-1: 26 (51.0)	sICH* within 24 hrs: 5 (9.8) Death within 3 months: 6 (11.8)

Source	Location and conducting period	Study design	Included criteria	rt-PA dose	Number of patients	Functional outcome at 3 months, n (%)	Safety outcome, n (%)
Chen et al, 2012 (67)	Taiwan China, 08/2006-12/2010	Retrospective cohort	As in the SITS-MOST study, but without an upper age limit	0.7 mg/kg	105	mRS scores 0-1: 43 (41.0)	sICH* within 36 hrs: 5 (4.8) Death in hospital: 8 (7.6)
				0.9 mg/kg	156	mRS scores 0-1: 60 (38.4)	sICH* within 36 hrs: 4 (2.6) Death in hospital: 9 (5.8)
Pan et al, 2013 (68)	Mainland China, NR	Retrospective cohort	As in the NINDS trial	<0.75 mg/kg	31	mRS scores 0-1: 16 (51.5)	sICH* within 24 hrs: 3 (9.1) Death within 3 months: 1 (3.0)
				0.75-0.9 mg/kg	33	mRS scores 0-1: 20 (61.2)	sICH* within 24 hrs: 5 (16.1) Death within 3 months: 1 (3.2)
				0.9 mg/kg	19	mRS scores 0-1: 11 (57.1)	sICH* within 24 hrs: 2 (10.5) Death within 3 months: 1 (5.3)
Kim et al, 2015 (69)	South Korea, 11/2009-03/2013	Prospective cohort	AIS patients (within 4.5 hrs after onset) identified by neuroimage and received intravenous alteplase	0.6 mg/kg	450	mRS scores 0-1: 146 (32.4)	sICH: 38 (8.4) Death within 3 months: 57 (12.7)
				0.9 mg/kg	1076	mRS scores 0-1: 380 (35.3)	sICH: 69 (6.4) Death within 3 months: 151 (14.0)
Yang et al, 2016 (70)	Mainland China, 01/2013-01/2016	Retrospective cohort	AIS patients (with mild symptom defined as NIHSS score ≤5 at admission) who received alteplase within 4.5 hrs after symptom onset	0.6 mg/kg	46	mRS scores 0-1: 34 (73.9)	sICH within 24 hrs: 2 (4.3) Death in hospital: 2 (4.3)
				0.9 mg/kg	62	mRS scores 0-1: 44 (71.0)	sICH within 24 hrs: 3 (4.8) Death in hospital: 1 (1.6)

*NINDS criteria.
†Favoured low-dose (0.6 mg/kg) alteplase in elderly patients (71–80 years).
‡With no consideration of the results of blood clotting and hepatic and kidney function.
§After receiving the normal results of blood clotting and hepatic and kidney function.

data from Asian stroke registers.[78-84] All these studies concluded a comparable efficacy of low-dose versus standard-dose alteplase for 90-day functional outcomes. Two studies confirmed a reduced risk of sICH by low-dose alteplase after data pooling.[80,82]

1.2 BP lowering and thrombolysis outcomes in AIS

1.2.1 High BP levels after stroke predict poor prognosis

Elevated BP is common in acute stroke patients, with around 70% of the patients having SBP >140 mmHg at presentation in hospital, as reported in one cross-sectional survey based on a nationally registered dataset from the USA (N=563,704, 31% with SBP <140 mmHg, 56% with SBP 140-184 mmHg, 13% with SBP 185-219 mmHg, 0.1% with SBP ≥220 mm Hg).[85] Several factors or mechanisms contribute to the evaluation of BP after stroke onsets, such as pain, pre-existing hypertension, infection or inflammatory reaction, mental stress caused by hospitalisation, activation of the renin-angiotensin-aldosterone system and sympathetic neuroendocrine system, compromised sensitivity of cardiac baroreceptor, and raised intracranial pressure caused by the growth of brain parenchymal haematoma or oedema.[86]

Although evaluated BP levels would decrease spontaneously in part of the patients within seven days post-stroke onset, around one-third of stroke patients have persistent hypertension in the first week, which is associated with poor clinical prognosis. A systematic review, including 32 cohort studies and 10,892 stroke patients, found high BP in AIS or primary ICH was associated with subsequent death (mean arterial BP OR [95% CI] 1.61 [1.12-2.31] for ICH), death or dependency (SBP OR [95% CI] 2.69 [1.13-6.40] for ICH, weighted mean difference [95% CI] +11.73 mmHg [1.30-22.16] for AIS), and death or neurologic deterioration (SBP OR [95% CI] 5.57 [1.42-21.86] for ICH).[87] A recent observational study in AIS with larger samples (N=309,611) showed a J-shaped/U-shaped relationship between SBP and outcomes with comparable results when other BP parameters were analysed: both higher and lower SBP (versus central reference value) increased the risk of in-hospital death (adjusted OR [95% CI] 1.24 [1.19-1.30] for 200 versus 150 mmHg, 1.16 [1.13-1.20] for 120 versus 150 mmHg), not discharged home (1.15 [1.12-1.18] for 200 versus 150 mmHg, 1.11 [1.09-1.13] for 120 versus 150 mmHg), and inability to walk independently at discharge (1.09 [1.06-1.11] for 200 versus 150 mmHg, 1.16 [1.13-1.18] for 120 versus 150 mmHg).[88]

1.2.2 Patients with ICH benefit from early intensive BP lowering

Early BP lowering showed some benefit in acute ICH, as confirmed in the second Intensive Blood Pressure Reduction in Acute Cerebral Haemorrhage Trial 2 (INTERACT2).[89] INTERACT2 was an international, multicentre, prospective, randomised, open-label, blinded end-point trial that investigated the effects of rapid lowering of elevated BP to improve the outcome in patients with ICH. The trial included 2,839 participants who had spontaneous ICH and elevated SBP (150-220 mm Hg) within 6 hours after ICH onset and randomly assigned them to receive intensive BP-lowering treatment (with a target SBP <140 mmHg within 1 hour) or contemporary guideline-recommended treatment (with a target SBP <180 mmHg) through the use of BP-lowering agents according to local physician's choosing.[90-91] The results indicated intensive BP lowering versus the control improved 90-day functional outcomes defined by ascending ordinal mRS shift (OR [95% CI] 0.87 [0.77-1.00], P=0.04), although there was no significant reduction in the rate of death or severe disability (mRS 3-6 [primary outcome], 0.87 [0.75-1.01], P = 0.06). The rates of mortality and nonfatal serious adverse events were comparable between the two randomised groups.[89]

Intensive BP lowering reduced haematoma growth without compromising peri-haematoma cerebral blood flow and aggravating peri-haematoma oedema, as identified from INTERACT2, INTERACT1 (pilot-phase study of INTERACT2), and other imaging studies,[92-94] might account for the favourable clinical prognosis from early intensive BP lowering in patients with primary ICH. Based on the results of INTERACT2, a revision of guidelines for the management of spontaneous ICH by the American Heart Association (AHA)/American Stroke Association (ASA) was made in 2015: "acute lowering of SBP to 140 mm Hg is recommended for ICH patients presenting with SBP between 150 to 220 mmHg and no contraindication to acute BP treatment or suggested for those presenting with SBP >220 mm Hg and having the condition of continuous intravenous infusion and frequent BP monitoring."[95]

1.2.3 Current BP management in thrombolysis-treated AIS

Contrary to the benefit of early intensive BP lowering in ICH, the optimal BP management in AIS patients who are candidates for reperfusion treatment (intravenous thrombolysis or mechanical thrombectomy [MT]) remains uncertain. Current guideline recommendation (BP <185/110 and <180/105 mmHg, before and for 24 hours after alteplase, respectively)[96,97] is based on expert opinion, given an apparent association of elevated BP and increased risk of

symptomatic ICH in thrombolysis-treated AIS.[88,98] Results from observational studies on the association of decreased BP levels with thrombolysis outcomes in AIS are inconsistent. Post-hoc analyses from the IST-3 trial defined a potential benefit of BP reduction or receiving BP-lowering treatment over 24 hours in AIS patients, with a significant reduction of 6-month poor outcome regardless of intravenous use of alteplase (a large BP reduction, OR [95% CI] 0.93 [0.89-0.97]; receiving BP-lowering treatment: 0.78 [0.65-0.93]).[99] However, two recent studies observed adverse effects of intraprocedural BP lowering on the growth of infarct volume and functional outcomes in patients who received MT.[100,101] A meta-analysis of 13 RCTs with 12,703 participants reported a neutral effect of BP lowering on the prevention of 90-day death or dependency in the early period of AIS (pooled relative risk by random-effects model [95% OR] 1.04 [0.96-1.13]) as well as the neutral effect on recurrent vascular events, disability or death, all-cause mortality, recurrent stroke, and serious adverse events.[102] *In summary, there had been insufficient evidence to form management recommendations on early BP lowering target in AIS.*

1.2.4 BP arm of the ENCHANTED trial

The BP arm of the ENCHANTED trial investigated the effectiveness of more intensive versus contemporaneous guideline-recommended management of BP in thrombolysis-treated AIS patients with an international, multicentre, prospective, randomised, open-label, blinded-endpoint design.[24,27,28] From March 2012 to April 2018, 2196 thrombolysis-eligible AIS patients within 6 hours of symptom onset and with a baseline SBP ≥150 mmHg were randomly assigned to intensive BP lowering group (target SBP 130-140 mmHg within 1 hour, N=1081) or guideline-recommended BP control group (target SBP <180 mmHg, N=1115) over 72 hours. The study aimed to test the hypotheses that following the use of intravenous thrombolysis, a strategy of early intensive BP lowering is superior to guideline-recommended BP control for improving 90-day functional recovery (ascending ordinal mRS shift [primary outcome], disability or death [mRS scores 2-6], major disability or death [mRS scores 3-6]) and reducing the risk of ICH (safety outcome) in AIS patients. Other outcomes included death or END (≥4 points increase in NIHSS) within 24 hours and 72 hours post-randomisation and sICH defined by the SITS-MOST criteria and several criteria from other studies.

The results, published in 2019, showed significantly less intracranial haemorrhage in the early intensive BP lowering group (n=160, 14.8%) compared with guideline-recommended BP control group (n=209, 18.7%) (OR [95% CI] 0.75 [0.60-0.94]). There was no difference in the

functional recovery at 90 days post-randomisation between the two randomised groups (mRS ordinal shift, OR [95% CI] 1.01 [0.87-1.17]). The rate of any serious adverse events that occurred during the trial was also comparable between the two groups (19.4% versus 22.0%, OR [95% CI] 0.86 [0.70-1.05]).

1.2.5 Paradox of BP lowering in AIS

Currently, there is still no defined optimal early BP-lowering target to recommend for AIS patients. The association of early BP lowering with thrombolysis outcomes in AIS may be modified by whether or not there is successful recanalisation (or reperfusion) after intravenous thrombolysis or MT.[103] Before recanalisation, cerebral blood perfusion in the infarct lesion mainly comes from posterior blood flow through collateral circulation in leptomeningeal, with no or a small amount of anterior cerebral blood flow. At this stage, BP-lowering might induce adverse influence on establishing collateral circulation, followed by less cerebral blood perfusion in the vulnerable and evolving, ischaemic penumbral region to accelerate infarct core growth. All these will lead to unfavourable functional recovery in both the short and long term. Thus, increased BP level before recanalisation may benefit patients with AIS, as identified in several studies based on imaging findings.[100,101,104] Once successful recanalisation is achieved, evaluated BP levels will promote injury of the fragile blood-brain barrier and increase ICH risk in the ischaemic territory. Thus, BP-lowering after recanalisation is beneficial for AIS, as confirmed in recent findings in AIS patients who received endovascular treatment with successful recanalisation.[105-107]

In the ENCHANTED BP arm, there is a possibility that participants received BP lowering before alteplase initiating fibrinolysis of the thrombus and achieving recanalisation, which might offset any benefit of reduced ICH after reperfusion and lead to a neutral outcome of 90-day functional recovery by intensive BP lowering versus guideline-recommended BP control. Smaller than envisaged SBP difference between groups (mean ± standard 144.3 ± 10.2 mmHg in the intensive BP lowering group, 149.8 ± 12.0 mmHg in the guideline-recommended group, deviation Δ6 mmHg over 24 hours) and inclusion of good prognosis patients with mild-to-moderate severity (baseline NIHSS median score 7) are another two explanations to the neutral results of 90-day functional recovery.

1.3 Imaging of AIS

1.3.1 Significance of brain imaging in AIS

Brain CT and MRI are essential for AIS diagnosis. Multi-model CT and MRI scanning, including non-contrast CT (NCCT) and MRI, CT angiography (CTA) and MRI angiography (MRA), perfusion imaging, and magnetic susceptibility imaging, can help to exclude ICH and provide important information related to AIS, including acute ischaemic change in parenchymal, infarct location, the site of vascular obstruction or occlusion, clot length and permeability, the volume of infarct core and penumbra, collateral circulation status, and coexisting old vascular lesions, small vessel diseases (white matter hyperintensity, lacune, perivascular space enlargement, microbleed), and brain volume loss.[108-110]

It is hard to say which imaging modality is better for the diagnosis of AIS. NCCT remains the primary imaging modality used to evaluate patients with suspected AIS, given its 24 hours availability in the emergency department, fast scanning time, and high sensitivity and specificity to identify primary ICH or haemorrhagic transformation after AIS.[111] However, it is challenging to detect acute ischaemic change on NCCT within 3 hours after AIS onset. In contrast, MRI has a high sensitivity in detecting early infarct lesions, and there is signal change on diffusion-weighted imaging (DWI) within minutes after AIS onset, even the infarct lesion is small or located in the posterior fossa.[112-114] Nevertheless, MRI has apparent shortcomings, such as not being available widely, no use at night in most hospitals, long scanning time, and ineligibility to patients with a pacemaker, defibrillator, cardiac stent, artificial heart valve, and claustrophobia.

Besides the necessity in detecting AIS, there is increasing recognition of the importance of brain images in predicting clinical prognosis and guiding the management of AIS patients.[115] For patients with wake-up stroke who is unclear of the stroke onset, the WAKE-UP (Efficacy and Safety of MRI-based Thrombolysis in Wake-Up Stroke) trial showed intravenous alteplase use led to significantly better 90-day functional recovery and numerically more intracranial haemorrhages compared to no use in those who had DWI-FLAIR mismatch (having signal change on DWI but no on FLAIR sequence).[20] The result supports using the DWI-FLAIR mismatch sign to indicate whether a patient with wake-up stroke is within the time window of intravenous thrombolysis (<4.5 hours after stroke onset). Based on perfusion imaging-guided thrombolysis, the EXTEND (Extending the Time for Thrombolysis in Emergency Neurological

Deficits) trial showed a possibility of intravenous alteplase use between 4.5 and 9 hours after stroke onset in patients with a sign of perfusion lesion-ischaemic core mismatch, given significantly more participants with favourable 90-day functional outcome in the alteplase group than the placebo group.[21] The DEFUSE 3 (Endovascular Therapy Following Imaging Evaluation for Ischaemic Stroke) trial[18] and the DAWN (DWI or CTP Assessment with Clinical Mismatch in the Triage of Wake-Up and Late Presenting Strokes Undergoing Neurointervention with Trevo) trial[19] confirmed imaging signs from CTP and MRI could be used to select AIS patients (with intracranial ICA or proximal MCA occlusion and presenting at 6 to 24 hours after stroke onset) who would benefit from thrombectomy. Accordingly, CTP or DWI sequence with or without MR perfusion is recommended in these clinical populations in an updated guideline from the AHA/ASA.[116]

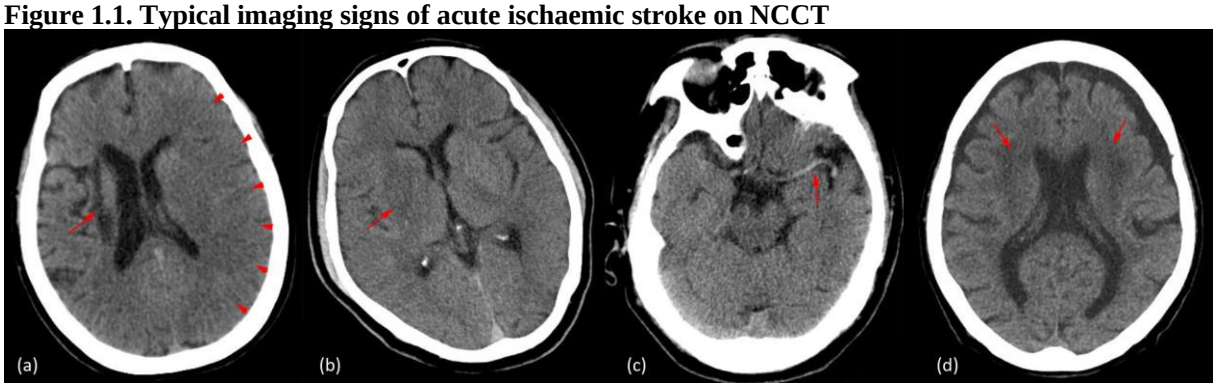
1.3.2 Non-contrast computed tomography (NCCT)

Brain NCCT can provide a series of two-dimensional, cross-sectional, anatomical images of the brain based on X-rays having different attenuation levels when passing through different brain tissues, as measured by different CT values and corresponding normalised grayscale images. Unfortunately, there is no consensus on the optimal acquisition parameters of brain NCCT for a suspected AIS patient, and brain NCCT acquisition protocols are reported inconsistently across the publications. Nonetheless, image acquisition should follow the principle of optimised dose reduction and image quality.[117-121] Before obtaining the brain NCCT scan, all items that may result in artefacts around the head need to be removed. In order to suppress motion artefacts, the patient's head could be immobilised with wedge sponges on either side of the head or to use a helical scan model with faster scanning speed compared with axial mode.

Acute ischaemic changes on brain NCCT include signs of swelling (cortical sulcal effacement, obscuration of sylvian fissure, and loss of insular ribbon), brain parenchymal CT density change (loss of grey-white matter cortex differentiation, loss of basal ganglia outline, and focal hypoattenuation), and hyper-attenuated artery sign.[110] *Cortical sulcal effacement* is defined as loss of precise delineation of the grey-white interface in the margins of the cortical sulci, and the sulci are obscured when comparing with contralateral counterpart (Figure 1.1-a). *Obscuration of sylvian fissure and loss of insular ribbon* is presented as decreased precision in the delineation of the grey-white interface at the lateral margin of the insula. A mild decrease of x-ray attenuation in the infarct lesion will lead to the *loss of grey-white matter differentiation*

in the cortex or basal ganglia, and an apparent decrease of X-ray attenuation will further cause *focal hypoattenuation* with a clear borderline (Figure 1.1-b). It is important to discriminate focal hypoattenuation led by ischaemia with the *coexisting old vascular lesion(s)*, which usually has lower CT density values with a sharper outline than acute infarct lesion (Figure 1.1-a). *Hyper-attenuated artery sign* consists of hyper-density of an arterial structure; this is highly specific to an acute thrombus.[122] An absolute density of >43 HU on brain NCCT with thin slices (slice thickness ≤3 mm) can determine a hyperdense artery sign (Figure 1.1-c). In addition, *brain atrophy* and *white matter lesions* can be semi-quantitatively measured on NCCT (Figure 1.1-d).[123,124]

Figure 1.1. Typical imaging signs of acute ischaemic stroke on NCCT



(a) cortical sulcal effacement (arrow head) and coexisting old vascular lesion (arrow); (b) focal hypoattenuation (arrow); (c) hyper-attenuated artery sign (arrow); (d) white matter lesions (arrow) and brain atrophy.

The reader reliability and accuracy of detecting acute ischaemic changes are not high. An early systematic review including 15 studies showed that the prevalence of all early infarction signs on NCCT was 61(± 21)%, and the interobserver agreement ranged from 0.14 to 0.78. Corresponding mean sensitivity and specificity were 66% (range, 20% to 87%) and 87% (56% to 100%), respectively.[110] The ENCHANTED imaging data showed inter-reader reliability was fair to moderate for most acute ischaemic changes (κ=0.36 for assessing swelling, 0.40 for identifying brain parenchymal CT density change due to ischaemia, and 0.44 for identifying hyperdense arteries), atrophy and white matter lesions were more consistently assessed (κ=0.49 for both). Accuracy was moderate to substantial for acute ischaemic changes (κ=0.49 to 0.64) but only fair to moderate for old vascular lesions (κ=0.35 to 0.54).[125]

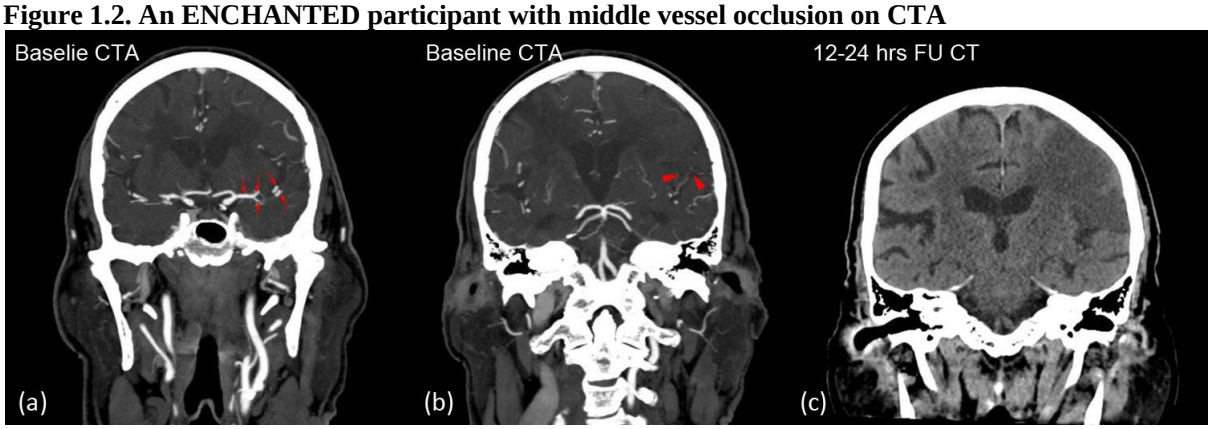
With respect to the association of acute ischaemic changes on brain NCCT with thrombolysis outcomes, data from the NINDS rt-PA stroke trial found there was no difference in the effect of alteplase versus placebo across AIS patients (N=616) with or without early infarct signs on NCCT obtained with 3 hours post-stroke.[126] Based on the results, the authors concluded that

early ischaemic changes were not associated with an increased risk of adverse outcomes after intravenous alteplase use. In contrast, secondary analysis from the IST-3 trial involving more thrombolysis-treated AIS patients (N=3017) showed a reduction in 6-month functional independence was predicted by the presence of focal hypoattenuation (OR [95% CI] 0.66 [0.55-0.81]), large infarct lesion (0.51 [0.38-0.68]), swelling (0.59 [0.46-0.75]), hyper-attenuated artery (0.59 [0.47-0.75]), atrophy (0.74 [0.59-0.94]), and white matter lesions (0.72 [0.59-0.87]). Old infarct lesion (1.72 [1.18-2.51]), focal hypoattenuation (1.54 [1.04-2.27]), and hyper-attenuated artery (1.54 [1.03-2.29]) predicted sICH.[29]

1.3.3 Computed tomography angiography (CTA)

CTA is usually used to assess intra- or extracranial vessel occlusion and its site for AIS patients. After intravenous injection of an iodinated radiocontrast agent, we can observe intra- or extracranial vessels when performing CT scanning immediately. Figure 2.1 shows a typical case of ENCHANTED participants with middle vessel occlusion confirmed on CTA. Advantages of CTA include minimal invasiveness, short scanning time, excellent vascular anatomic rendering, and a low risk of complications. All these make CTA an ideal tool in detecting vessel occlusion and assessing clot length, clot permeability or residual flow, leptomeningeal collateral circulation, and intra- or extracranial arterial stenosis in AIS patients for whom treatment decision-making is urgent.[127,128]

Figure 1.2. An ENCHANTED participant with middle vessel occlusion on CTA



(a) left M1 and proximal M2 segments of the middle cerebral artery are shown on baseline CTA (arrow); (b) fewer sylvian branches (arrowhead) compared to contralateral side; (c) infarct at left temporal lobe is seen on follow-up CT.

However, it needs to be cautious about contrast agent-related complications, such as contrast agent extravasation into subcutaneous soft tissue when intravenous injection, iodine allergy reaction, and contrast-induced nephropathy.[129-131] CTA can be acquired either in a single

arterial phase (performing CT scanning at the time point of contrast agent passing through cerebral arteries) or in multiple phases (arterial, late arterial, and early venous phases). Multiphase CTA is superior to the single-phase CTA in detecting distal intracranial arterial occlusion, identifying tandem lesion and carotid terminus occlusion, and assessing clot length and the status of leptomeningeal collateral circulation, despite an increase in acquisition time and radiation dose.[33,111,132-134]

Cerebral artery obstruction on CTA can be scaled from normal artery to complete occlusion according to modified Thrombolysis in Cerebral Infarction (TICI) scale[135] or IST-3 Angiography Score.[30] Both intravenous thrombolysis and endovascular thrombectomy are approved for the treatment of LVO, given that their benefit to functional recovery is confirmed in large-scale randomised clinical trials and related meta-analyses. However, evidence of treatment for AIS patients without vascular obstruction or with obstruction at internal carotid artery (ICA) or medium cerebral artery is limited, with most data derived from observational studies with relatively small samples.[136-142] One multi-centre prospective cohort study including 575 AIS patients showed that more distal thrombus location, greater clot permeability, and longer time to recanalisation assessment were associated with recanalisation of arterial occlusion after intravenous alteplase use.[32] Other clot characteristics (length, clot burden score, the presence of migration) are also associated with early recanalisation and functional recovery after intravenous thrombolysis or endovascular thrombectomy, as reported in several recent studies.[143-146] In addition, the status of leptomeningeal collateral circulation is associated with infarct growth, and it could predict clinical outcomes.[33,147-149]

1.3.4 Computed tomography perfusion (CTP)

Brain CTP can evaluate infarct core and ischaemic penumbra based on repeated CT scanning after intravenous injection of an iodinated radiocontrast agent. In the infarct core, neural cells are thought to be damaged irreversibly. In contrast, neural cells in the ischaemic penumbra are salvageable once reperfusion occurs. After setting regions of interest (ROIs) of arterial input function (usually in the A2 segment of the anterior cerebral artery) and venous outflow function (usually in the superior sagittal sinus or torcular herophili), CTP parameters can be acquired automatically using specific commercial or CT vendor-provided software. Key CTP parameters include cerebral blood volume (CBV), cerebral blood flow (CBF), mean transit time (MTT), time to peak (TTP), and time to maximum (T_{max}). CBV (unit, mL/100 g) is the volume of blood passing through a given volume of brain parenchyma.[111] CBF (unit, mL/100 g/min) is defined

as the rate of blood passing through a given volume of brain parenchyma per unit time.[111] MTT (unit, seconds) is defined as the time required for blood passing through a given volume of brain parenchyma. TTP (unit, seconds) measures the time taken from contrast injection until the maximum peak of contrast enhancement.[150] T_{max} (unit, seconds) is defined as the time between AIF and the tissue contrast agent concentration, calculated by AIF deconvolution.[151] Formula CBF=CBV/MTT can reflect the relationship among CBV, CBF, and MTT.

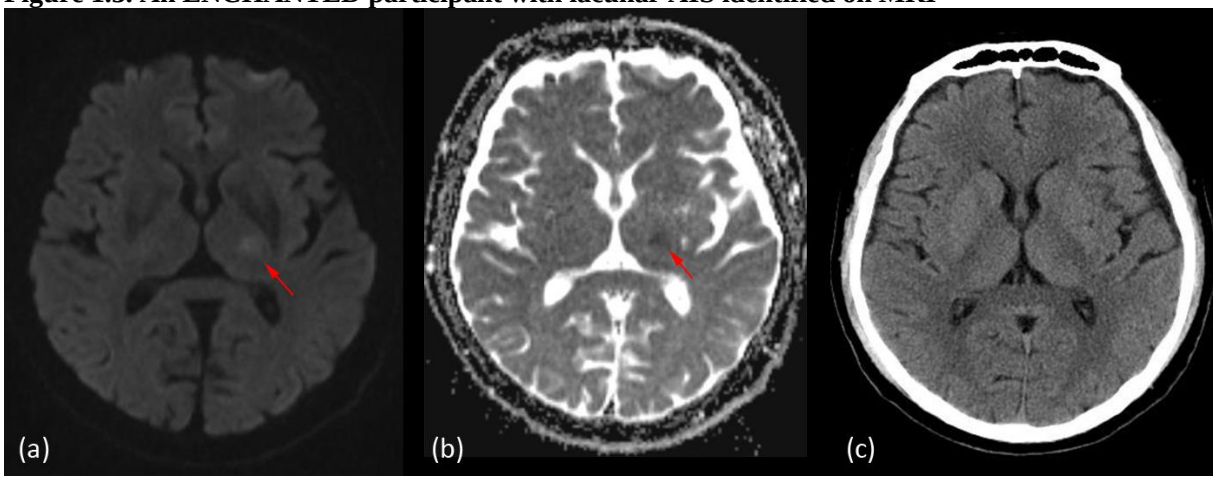
Although prior studies recommended cut-off values of CBV decrease (<2-2.5 ml/100 g),[152,153] CBF decrease (the reduction of relative CBF >30-45% of contralateral normal counterpart),[152,154,155] and MTT increase (>8.3 seconds),[156] cut-off values for infarct core and ischaemic penumbra have not been unified. CT machines from different vendors and CTP parameters calculated from different postprocessing techniques may partly account for it. It is widely accepted that relative CBF <30% to the contralateral normal counterpart is the threshold of infarct core, as it correlates better with the infarct core on DWI.[157,158] Data from the DEFUSE trial suggested a T_{max} of >5.5 seconds as the threshold of ischaemic penumbra on CTP.[159] The DEFUSE 3 trial adopted T_{max} >6 seconds as the threshold of the ischaemic penumbra. It indicated the eligibility of endovascular thrombectomy for AIS patients who were last known to be well 6 to 16 hours ago if having an infarct core <70 mL, infarct core-penumbra mismatch volume >15mL, and a mismatch ratio of ≥1.8 on CTP.[18] The same T_{max} threshold to define ischaemic penumbra was used in the EXTEND trial, which found a benefit to functional recovery in AIS patients who were given intravenous alteplase between 4.5 to 9 hours after stroke onset or on awakening with stroke symptoms if having an infarct core <70 mL, infarct core-penumbra mismatch volume >10 mL, and a mismatch ratio of ≥1.2 on CTP.[21]

1.3.5 Magnetic resonance imaging (MRI)

Although MRI is not the first tool for diagnosing AIS in most hospitals, diffusion-weighted MR imaging (DWI) is much more sensitive and specific than CT scanning in detecting acute infarct lesions, with a signal change immediately after stroke onset.[160] The swelling of neural cells and less free-moving intercellular fluid in the ischaemic area will lead to diffusion restriction, which can be quantitatively reflected by a decreased apparent diffusion coefficient (ADC) and mean diffusivity (MD) from DWI.[161] Accordingly, the infarct lesion is shown as hyperintensity on DWI and hypo-intensity on the ADC map. Figure 1.3 shows the DWI and ADC map acquired before randomisation from a participant of the ENCHANTED trial who had lacunar AIS. DWI is superior to CTP in estimating the infarct core and infarct volume. Purushotham et al. showed

that an ADC ≤620 × 10^{-6} mm^2/s is the optimal threshold for identifying infarct core with a sensitivity of 69% and a specificity of 78%.[162] Although DWI has the advantage of detecting small infarct lesions, including those in the posterior circulation and acute lacunar infarct (< 20 mm in the territory of penetrating arteries), an AIS patient may have normal MRI due to the reversibility of DWI lesion, which is seen in one-fourth of AIS patients according to a recent systematic review.[163] An earlier observational study also reported that nearly one-third of patients with non-disabling stroke did not have a relevant infarct lesion on DWI.[164] Potential explanations include reducing CBF causing symptoms but not severe enough to cause diffusion restriction, spontaneous recanalisation before MRI scanning, and missed DWI lesion when scanning.

Figure 1.3. An ENCHANTED participant with lacunar AIS identified on MRI

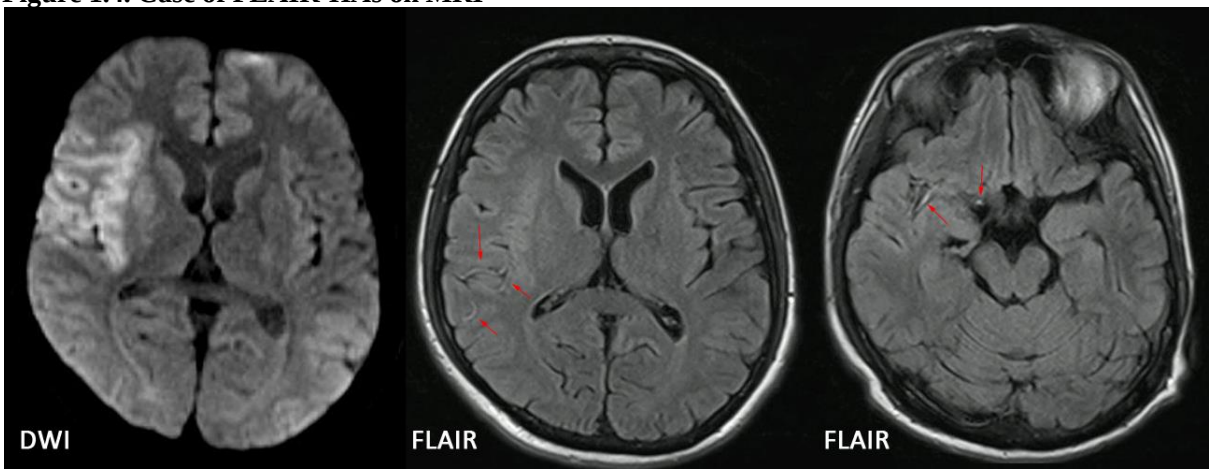


(a) A small infarct lesion at left internal capsule shown as hyperintensity on DWI (arrow); (b) the infarct lesion is shown as hypo-intensity on corresponding ADC map; (c) there is no ischaemic change or infarct lesion on NCCT.

FLAIR hyperintensity arteries (FLAIR-HAs) are a common sign of acute infarct on MRI, with a prevalence of more than 45% in AIS patients.[165,166] They are defined as focal, tubular, or serpentine hyperintensities relative to the grey matter in the subarachnoid space, corresponding to the typical arterial course on FLAIR sequence (a case from ENCHANTED is shown in figure 1.4).[167] FLAIR-HAs can correspond to fresh thrombus or slow retrograde (or anterograde) flow in the leptomeningeal collateral circulation from haemodynamic impairment after intracranial arterial occlusion.[168,169] Thus, FLAIR-HAs may reflect good collaterals[170] and the presence of a large ischaemic penumbra when located distal to the clot or beyond the ischaemic core.[171,172] However, the association of FLAIR-HAs with clinical prognosis has not been clearly defined. Potential reasons include retrospective study design, relatively small samples, varying methodologies of FLAIR-HAs assessment, different treatments given to patients, and heterogeneity of endpoints across prior studies. FLAIR sequence is valuable for identifying the

onset time in those with wake-up stroke or being last known well. Thomalla et al. showed that having an infarct lesion on DWI but not on FLAIR (i.e., DWI-FLAIR mismatch, Figure 1.5) can identify AIS patients who are within 3 hours after stroke onset, with moderate sensitivity (48%) but high specificity (93%).[173] The WAKE-UP trial and the following meta-analysis found a benefit of intravenous alteplase use in patients with unknown time onset who have DWI-FLAIR mismatch on MRI.[20,23] Although it is not confirmed in the main results of the recent THAWS (Thrombolysis for Acute Wake-Up and Unclear-Onset Strokes With Alteplase at 0.6 mg/kg) trial, a subset of THAWS participants with DWI-ASPECTS (Alberta Stroke Program Early CT Score) 5 to 8 and DWI-FLAIR mismatch get better 90-day functional outcome after low-dose intravenous alteplase use versus placebo.[174,175]

Figure 1.4. Case of FLAIR-HAs on MRI



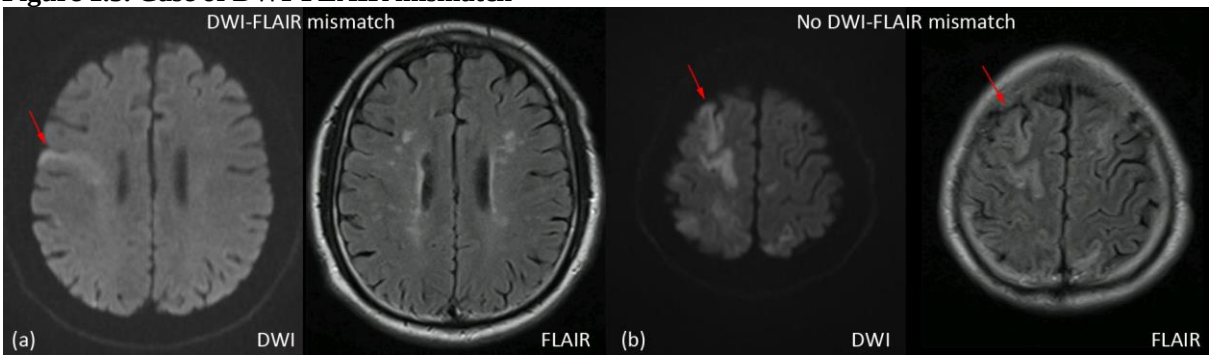
An ENCHANTED participant with an ischaemic lesion in right MCA territory on baseline DWI. FLAIR-HAs were present at right proximal (M1 segment), middle (M2 segment/sylvian fissure), and distal (M3-6 segment) MCA on baseline FLAIR sequence (arrows).

MRI is also sensitive to detect chronic microbleeds, which are shown as small spots (≤10 mm) of a signal void with associated blooming on susceptibility-weighted imaging (SWI) or gradient-echo T2*-weighted sequences (Figure 1.6).[176] Persistent hypertension and advanced cerebral amyloid angiopathy (CAA) are two main causes of chronic microbleeds. Hypertensive vasculopathy is associated with microbleeds in the deep brain area (such as basal ganglia, thalamus, brainstem, and cerebellum), and advanced CAA is associated with microbleeds in the brain lobe. It is still uncertain whether AIS patients who have microbleeds on MRI are predisposed to sICH after intravenous thrombolysis.[176-179]

1.3.6 Magnetic resonance angiography (MRA) and perfusion-weighted MRI (PWI)

Intra- and extra-cerebral vascular characteristics can be evaluated noninvasively on MRA either using the 3D-time of flight (TOF) or contrast-enhanced MRA (CE-MRA) technique (Figure 1.7). 3D-TOF MRA is helpful to identify acute proximal LVO but not reliable to identify the occlusion of distal vessel branches. Furthermore, it is possible of vascular pseudo-occlusion or overestimation of vascular stenosis on reconstructed maximum intensity projection images of 3D-TOF MRA, caused by blood turbulence when blood flows from normal lumen to a stenosis segment.[180,181] CE-MRA is acquired after intravenous injection of gadolinium-based contrast agent. Hyperintensity vascular lumen and hypo-intensity vascular wall and surrounding tissue can be discriminated when gadolinium-based contrast agent reduces the T1 relaxation time of intravascular blood. CE-MRA is more reliable than 3D-TOF MRA in grading vascular stenosis and occlusion. Dynamic CE-MRA can show the pathological features of the vessel wall due to atherosclerosis, such as intraplaque haemorrhage and lipid component, which are significantly associated with an increased risk of AIS.[182-184]

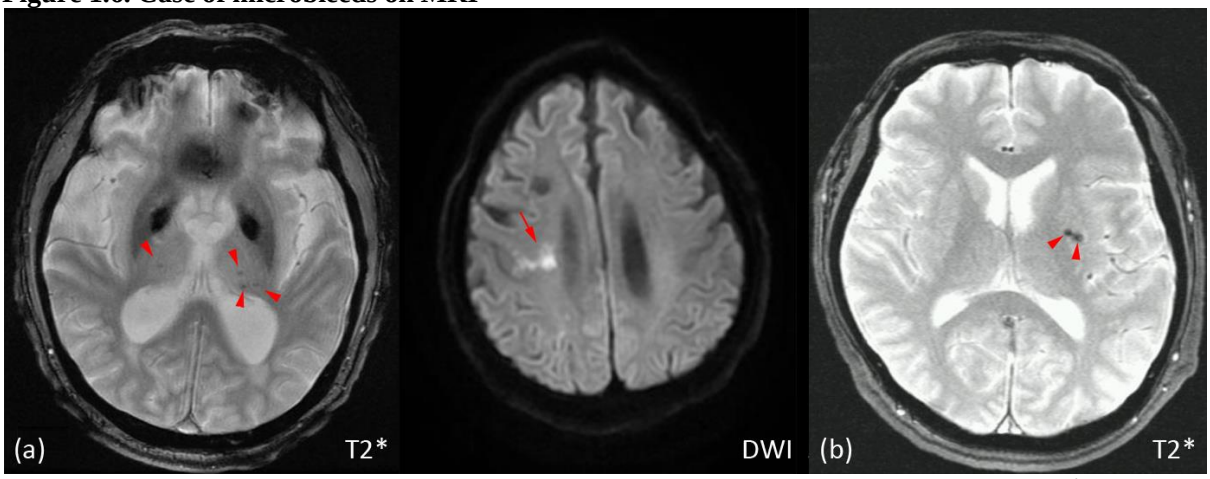
Figure 1.5. Case of DWI-FLAIR mismatch



(a) An ENCHANTED participant with DWI-FLAIR mismatch; (b) no DWI-FLAIR mismatch

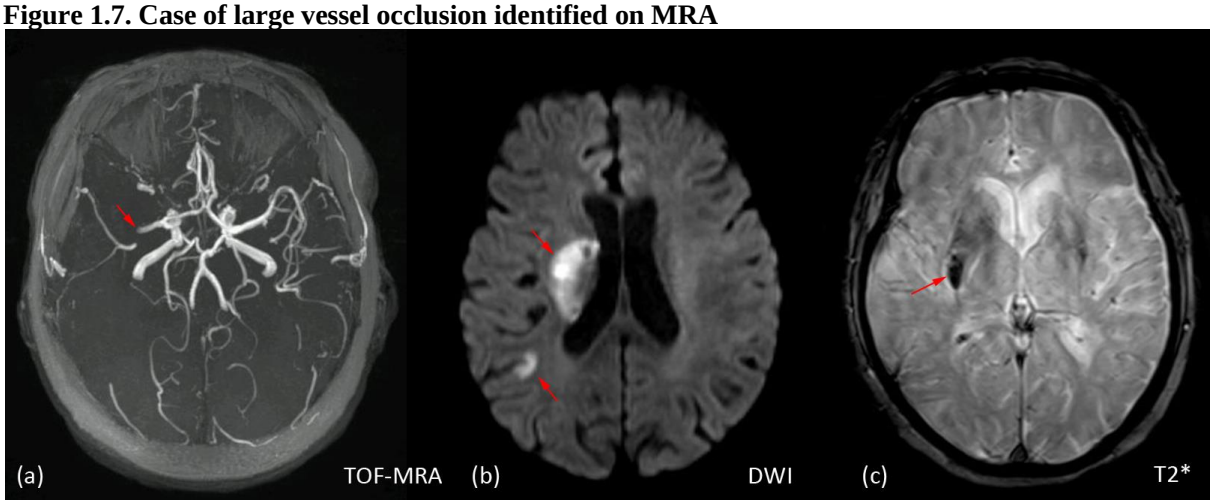
PWI is achieved through dynamic susceptibility-weighted contrast-enhanced MRI (DSC) sequence. Like CTP, PWI can display hypoperfusion area due to LVO and discriminate infarct core and ischaemic penumbra. After intravenous bolus injection of the gadolinium-based contrast agent, cerebral capillaries will have a nonlinear signal loss on T2* images, which can be tracked by DSC sequence to generate a haemodynamic time-to-signal intensity curve and a set of perfusion parameters (CBV, CBF, TTP, Tmax, and MTT).[112,185] The combination of PWI with DWI is valuable for identifying infarct core-ischaemic penumbra mismatch. A mismatch of >20% is one of the criteria for endovascular thrombectomy in LVO patients beyond the time window.[186]

Figure 1.6. Case of microbleeds on MRI



(a) An ENCHANTED participant with microbleeds at bilateral thalamus (arrowhead) on T2* image. An infarct lesion at right centrum semiovale (arrow) is shown on DWI; (b) another ENCHANTED participant with microbleeds at left basal ganglia on T2* image.

Figure 1.7. Case of large vessel occlusion identified on MRA



(a) An ENCHANTED participant with right MCA occlusion identified on MRA; (b) Infarct lesions at right basal ganglia and posterior border zone were detected on DWI; (c) bleeding was seen at right basal ganglia on T2* image.

1.3.7 Artificial intelligence (AI) in AIS imaging

AI technology for imaging analysis is developing quickly with some clinical applications in infarct lesion detection, core and penumbra volume estimation, LVO identification, ASPECTS grading, and prognosis prediction in AIS patients. AI can solve a specific task in a way that mimics human behaviour through computer algorithms. Machine learning (defined as algorithms enabling computers to learn from examples without explicit programming) and deep learn (defined as a set of inspired neural networks that do not require predetermined inputs) are two methods to achieve AI imaging analysis.[187] Several commercial AI software for AIS imaging diagnosis and analysis have been licensed in the USA and Europe, such as RapidAI

(USA) and Brainomix (UK). Nagel et al. reported that e-ASPECTS from the Brainomix was non-inferior to neuroradiologists in scoring ASPECTS on NCCT in AIS patients.[188] Subsequent study using the ENCHANTED data showed that e-ASPECTS was significantly associated with baseline neurological severity and independently predicted 90-day functional recovery and adverse outcomes.[189] Nevertheless, limitations of current AI techniques for AIS include limited use, few standard datasets with large samples for computer learning or neural networks setting, and no shared codes for external algorithm replication. More training data through multisite collaboration and open-source datasets and codes might help in the future.

1.4 Gaps in current research

Extensive subgroup analyses from the ENCHANTED trial have explored whether there are differential treatment effects of intravenous low-dose versus standard-dose alteplase or intensive BP lowering versus guideline-recommended BP lowering on functional outcomes according to conventionally clinical factors.[26,28,71-76] In addition, imaging data from the ENCHANTED showed that features of brain frailty (atrophy and severe leucoaraiosis) and acute ischaemia (visible ischaemic lesions, hypoattenuation, large ischaemic lesion, swelling, and hyper-attenuated arteries) on NCCT, are associated with symptomatic intracerebral haemorrhage (sICH) and mortality in thrombolysis-treated AIS patients (Tables 1.3 to 1.5). However, these features do not appear to modify the effects of different doses of intravenous alteplase (Figures 1.8 and 1.9).[190] More insights into other imaging features of brain (micro-) circulation are required, which might be helpful to form management strategies in thrombolysis-treated AIS patients. Some gaps in current literature that I had endeavoured to address are listed below:

1. Although FLAIR-HAs on MRI are recognised for nearly two decades, *predictors to the presence of FLAIR-HAs and their prognostic significance in AIS patients has not been well defined* despite reflecting the presence of a large ischaemic penumbra and good collateral circulation from slow retrograde flow distal to the occluded vessel (‘clot’) or ischaemic lesion.[170-172]
2. Patients with lacunar AIS, who are thought to have a single penetrating artery occlusion, are eligible to receive intravenous thrombolysis in routine clinical practice, given comparable favourable outcomes to other common AIS pathologic subtypes. Nonetheless, *clinical concern persists over whether the modest risk of thrombolysis-*

Table 1.3. Logistic regression analysis of associations between individual imaging signs and sICH or functional outcomes in the ENCHANTED trial, adjusted for age, NIHSS score, and time to randomisation

		sICH (IST3)		Death within 7 days		Death within 90 days		mRS 0-2	
		Adjusted OR (95% CI)	**P value**	**Adjusted OR (95% CI)**	**P value**	**Adjusted OR (95% CI)**	**P value**	**Adjusted OR (95% CI)**	**P value**
Early ischaemic signs	Visible ischaemic lesion	1.3 (0.82-2.08)	0.2666	1.52 (1.06-2.18)	**0.0243**	1.27 (0.97-1.68)	0.0819	0.7 (0.59-0.84)	**0.0001**
	Hypoattenuation	1.17 (0.72-1.9)	0.5233	1.51 (1.04-2.18)	**0.0287**	1.27 (0.96-1.68)	0.0968	0.74 (0.62-0.9)	**0.0018**
	Severe hypoattenuation	0.9 (0.28-2.91)	0.8642	1.24 (0.52-2.96)	0.6203	0.87 (0.43-1.74)	0.6908	1.16 (0.76-1·78)	0.4825
	Large lesion	0.83 (0.25-2.7)	0.5460	2.67 (1.52-4.71)	**0.0007**	1.94 (1.19-3.17)	**0.0078**	0.52 (0.33-0·81)	**0.0035**
	Very large lesion	0 (0-0)	0.9876	12.08 (0.65-223.57)	0.0942	6.88 (0.39-120.4)	0.1864	0 (0-0)	0.9701
	Swelling	1.24 (0.78-1.99)	0.3597	1.49 (1.03-2.14)	**0.0323**	1.27 (0.96-1·67)	0.0886	0.7 (0.58-0.84)	**0.0001**
	Hyperattenuated arteries	1.71 (1.01-2.89)	**0.0446**	2.17 (1.48-3.18)	**0.0001**	1.87 (1.39-2.52)	**0.0000**	0.63 (0.5-0.79)	**0.0001**
	Atrophy	0.88 (0.51-1.52)	0.5420	0.99 (0.64-1.53)	0.9629	1.52 (1.08-2.15)	**0.0178**	0.83 (0.68-1.01)	0.0625
	Severe atrophy	1.59 (0.88-2.86)	0.1246	0.52 (0.29-0.96)	**0.0352**	1.29 (0.9-1.84)	0.1594	0.83 (0.64-1.06)	0.1327
Pre-existing signs	Leukoaraiosis	0.84 (0.5-1.4)	0.4964	1.07 (0.71-1.61)	0.7613	1.17 (0.87-1.58)	0.2881	0.71 (0.59-0.86)	**0.0005**
	Severe leukoaraiosis	0.78 (0.36-1.69)	0.533	1.2 (0.68-2.11)	0.5266	1.74 (1.2-.2.54)	**0.0037**	0.68 (0.51-0.89)	**0.0055**
	Old infarct	0.95 (0.59-1.54)	0.838	1.24 (0.85-1.81)	0.2646	1.2 (0.91-1.58)	0.2072	0.7 (0.59-0.84)	**0.0001**

CI denotes confidence interval; ENCHANTED Enhanced Control of Hypertension and Thrombolysis Stroke Study; IST-3, the third International Stroke Trial; mRS denotes modified Rankin scale; OR, odds ratio; sICH symptomatic intracerebral haemorrhage. Data are adjusted OR (95% CI), p values. Variable results are shown as yes versus no.

Table 1.4. Full multivariable logistic regression models for symptomatic intracerebral haemorrhage and functional outcome at 3 months in the ENCHANTED trial

	sICH OR (95%CI)*	P value	mRS 0-2 OR (95%CI)*	P value
Age (years)	1.01 (0.99-1.03)	0.4557	0.98 (0.97-0.99)	**<0.0001**
NIHSS score	1.03 (0.99-1.07)	0.1038	0.86(0.85-0.88)	**<0.0001**
Time to randomisation (hours)	0.97 (0.77-1.21)	0.7652	0.99 (0.92-1.06)	0.7068
Treatment group (standard vs. low)	1.63 (1.00-2.64)	**0.0487**	1.08 (0.90-1.29)	0.3983
Antiplatelet (Yes vs. No)	1.35 (0.80-2.29)	0.2606	0.95 (0.77-1.17)	0.6415
Lesion size (large or very large vs. others)	0.65 (0.19-2.23)	0.4898	0.56 (0.34-0.9)	**0.0178**
Swelling	0.87 (0.22-3.48)	0.8492	0.56 (0.32-0.97)	**0.0379**
Hyperattenuated arteries	1.81 (1.03-3.17)	**0.0380**	0.70 (0.55-0.9)	**0.0043**
Old infarct	0.93 (0.56-1.56)	0.7969	0.78 (0.64-0.94)	**0.0090**
Hypoattenuation (mild vs. none)	1.44 (0.35-5.9)	0.6091	1.31 (0.74-2.31)	0.3472
Hypoattenuation (severe vs. none)	0.91 (0.13-6.45)	0.9276	1.94 (0.96-3.89)	0.0631
Atrophy (mild vs. none)	0.95 (0.51-1.76)	0.8707	0.81 (0.64-1.01)	0.0599
Atrophy (severe vs. none)	1.73 (0.78-3.86)	0.1802	0.74 (0.54-1.03)	0.0708
Leukoaraiosis (mild vs. none)	0.76 (0.39-1.48)	0.4260	0.80 (0.63-1.01)	0.0657
Leukoaraiosis (severe vs. none)	0.67 (0.29-1.54)	0.3409	0.64 (0.47-0.87)	**0.0043**

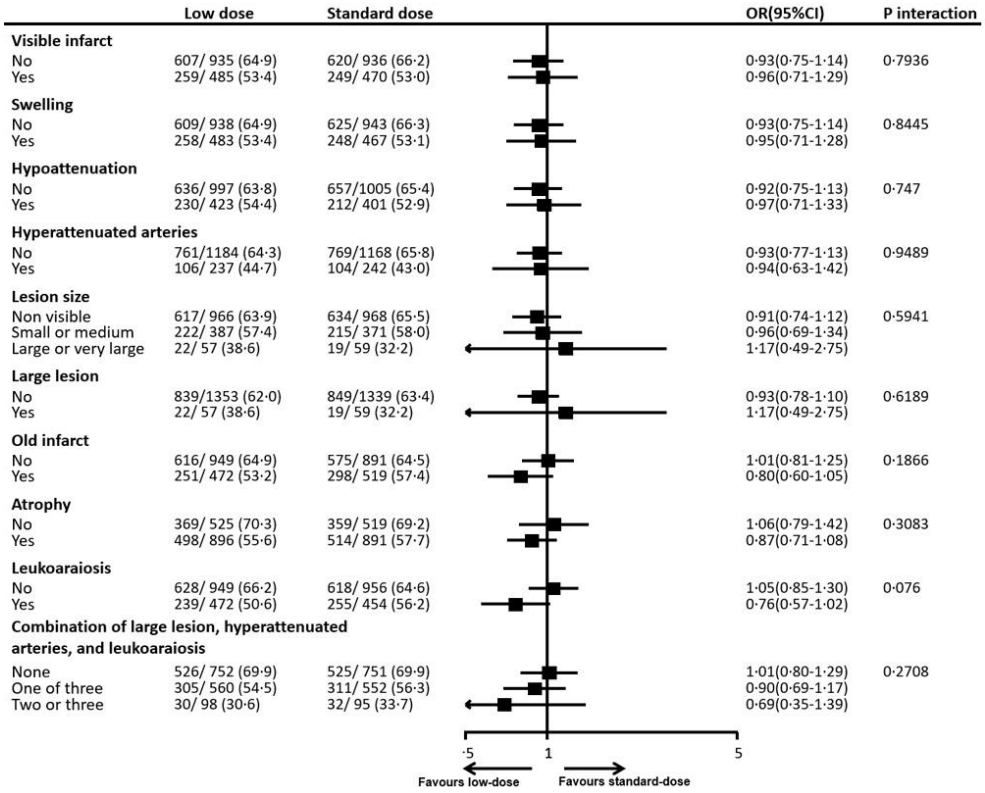
mRS denotes modified Rankin Score; NIHSS, National Institutes of Health Stroke Scale; OR, odds ratio; sICH, symptomatic intracerebral haemorrhage according to IST3 definition.
Data are adjusted for age, NIHSS and time to randomisation.

Table 1.5. Multivariable logistic regression models selected by stepwise logistic regression for sICH (IST3 definition) and functional outcome at 90 days in the ENCHANTED trial

	sICH OR (95%CI)	P value	mRS 0-2 OR (95%CI)	P value
Age (years)	1.01(0.99-1.03)	0.1947	0.98(0.97-0.99)	**<0.0001**
NIHSS score	1.03(0.99-1.07)	0.1130	0.86(0.85-0.88)	**<0.0001**
Time to randomisation (hours)	0.95(0.76-1.2)	0.6755	0.99(0.92-1.06)	0.7505
Treatment group (standard vs. low-dose)	1.60(0.99-2.59)	0.0545	1.08(0.91-1.29)	0.3887
Lesion size (large or very large vs. others)			0.57(0.35-0.92)	**0.0207**
Swelling			0.79(0.65-0.97)	**0.0256**
Hyperattenuated arteries	1.86(1.09-3.17)	**0.0227**	0.68(0.54-0.87)	**0.0018**
Old infarct			0.76(0.63-0.91)	**0.0038**
Hypoattenuation (mild vs. none)				
Hypoattenuation (severe vs. none)				
Leukoaraiosis (mild vs. none)			0.78(0.62-0.99)	**0.0369**
Leukaraiosis (severe vs. none)			0.61(0.46-0.82)	**0.0011**

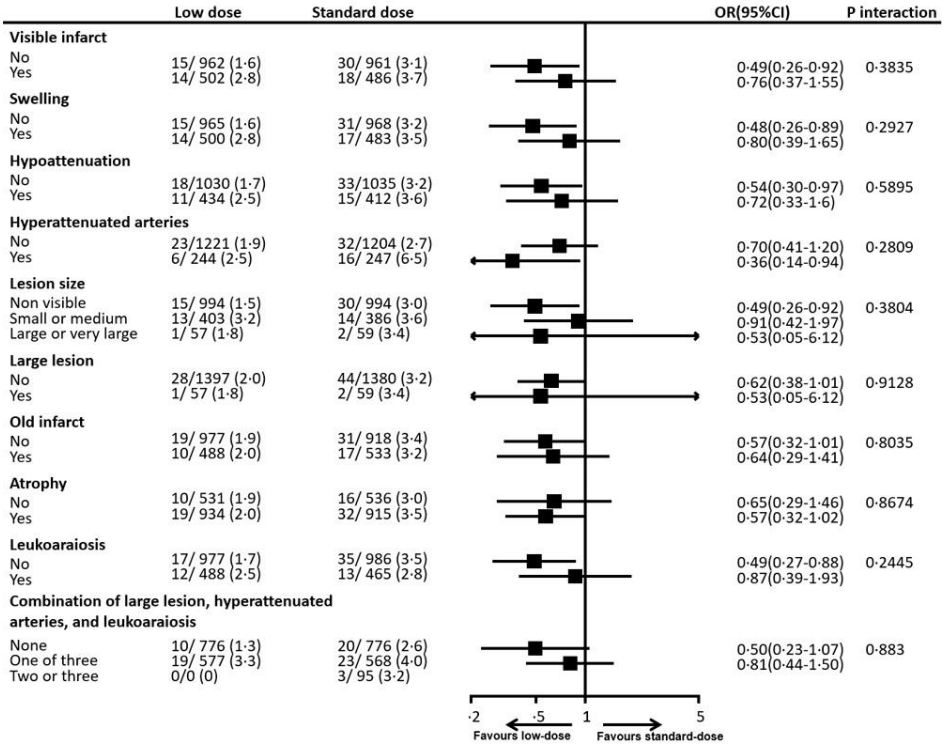
mRS denotes modified Rankin Score; NIHSS, National Institutes of Health Stroke Scale; OR, odds ratio; sICH symptomatic intracerebral haemorrhage according to IST3 definition
Data are adjusted for age, NIHSS and time to randomisation.

Figure 1.8. Effects of low-dose alteplase as compared with standard-dose alteplase on the functional outcome (mRS 3-6), according to imaging characteristics in the ENCHANTED trial



mRS denotes modified Rankin scale. Data are odds ratio (95% CI), P values for interaction.

Figure 1.9. Effects of low-dose alteplase as compared with standard-dose alteplase on the safety outcome (sICH), according to imaging characteristics in the ENCHANTED trial



mRS denotes modified Rankin scale; sICH, symptomatic intracerebral haemorrhage. Data are odds ratio (95% CI), P values for interaction.

related ICH could offset the modest benefits of intravenous thrombolysis for lacunar AIS, where the natural course is generally more benign than other AIS subtypes.

3. In the ENCHANTED trial, there is no heterogeneity of randomised alteplase dose and BP-lowering target on the primary outcome across different AIS subtypes defined by the TOAST (Trial of Org 10172 in Acute Stroke Treatment) classification system ($P_{interaction}$=0.88 in the alteplase dose arm and $P_{interaction}$=0.90 in the BP arm). *However, this approach has moderate diagnostic sensitivity and specificity. It may mix patients with non-lacunar AIS with the target population of lacunar AIS.*
4. Evidence of thrombolysis outcomes in AIS patients without vascular obstruction or with obstruction at the internal carotid artery or medium cerebral artery is limited, with most data derived from observational studies. Furthermore, *evidence in relation to randomised alteplase dose and BP target and key clinical outcomes, according to vascular obstruction status/site, is lacking*.

1.5 Objectives of the thesis

The scope of this thesis includes responding to all issues listed above. I have assessed some other imaging changes (or signs) of AIS, and investigated their associations with thrombolysis outcomes using patient data from the ENCHANTED trial. The specific aims of this thesis are:

1. To determine factors associated with FLAIR-HAs on MRI and their prognostic significance in thrombolysis-treated patients with AIS from the ENCHANTED trial alteplase-dose arm (Chapter 2).
2. Extending Chapter 2 through a systematic review and meta-analysis of the association of FLAIR-HAs with early vascular recanalisation or reperfusion, ICH, cerebral infarct growth, and clinical outcomes in AIS (Chapter 3).
3. To determine any differential efficacy and safety of low- versus standard-dose intravenous alteplase in the ENCHANTED participants with lacunar (versus non-lacunar) AIS identified by the combination of clinical and adjudicated imaging findings (Chapter 4).
4. To determine any differential efficacy and safety of intensive versus guideline-recommended BP lowering between lacunar and non-lacunar AIS in the BP arm of the ENCHANTED trial (Chapter 5).

5. To explore the influence of low-dose intravenous alteplase and intensive BP lowering on the outcomes of AIS according to the status/location of the vascular obstruction in the ENCHANTED participants (Chapter 6).

1.6 Structure of the thesis

This thesis contains a series of published manuscripts. All references are cited within each Chapter and outlined in the 'References' section.

The studies contained within this thesis explored associations of brain (micro-) circulation features on images with thrombolysis outcomes in AIS patients by alteplase dose or BP lowering target. Chapters 2 and 3 investigate clinical prognosis in AIS by FLAIR-HAs in the ENCHANTED trial and from a study-level meta-analysis, respectively. In Chapters 4 and 5, I summarise studies on the effects of randomised alteplase dose and BP-lowering on thrombolysis outcomes in lacunar AIS, representing a single penetrating artery occlusion. In Chapters 6, I present findings on the prognostic significance according to arterial characteristics of AIS by alteplase dose and BP-lowering target. In Chapter 7, I summarise and discuss the main findings of this body of research, the implications for clinical practice, and highlight areas for future research. Finally, the thesis appendices are list in Chapter 8.

References

1. GBD 2016 Stroke Collaborators. Global, regional, and national burden of stroke, 1990-2016: a systematic analysis for the Global Burden of Disease Study 2016. Lancet Neurol 2019;18(5):439-58.
2. Zhou M, Wang H, Zeng X, et al. Mortality, morbidity, and risk factors in China and its provinces, 1990-2017: a systematic analysis for the Global Burden of Disease Study 2017. Lancet 2019;394(10204):1145-58.
3. Rajsic S, Gothe H, Borba HH, et al. Economic burden of stroke: a systematic review on post-stroke care. Eur J Health Econ 2019;20(1):107-34.
4. Li XK, Lijnen HR, Nelles L, et al. Biochemical and biologic properties of rt-PA del (K296-G302), a recombinant human tissue-type plasminogen activator deletion mutant resistant to plasminogen activator inhibitor-1. Blood 1992;79(2):417-29.
5. National Institute of Neurological Disorders and Stroke rt-PA Stroke Study Group. Tissue plasminogen activator for acute ischaemic stroke. N Engl J Med. 1995;333(24):1581-7.
6. Hacke W, Donnan G, Fieschi C, et al. Association of outcome with early stroke treatment: pooled analysis of ATLANTIS, ECASS, and NINDS rt-PA stroke trials. Lancet 2004;363(9411):768-74.
7. ECASS Investigators. Thrombolysis with alteplase 3 to 4.5 hours after acute ischaemic stroke. N Engl J Med 2008;359(13):1317-29.
8. Lees KR, Bluhmki E, von Kummer R, et al. Time to treatment with intravenous alteplase and outcome in stroke: an updated pooled analysis of ECASS, ATLANTIS, NINDS, and EPITHET trials. Lancet 2010;375(9727):1695-703.
9. CHANCE Investigators. Clopidogrel with aspirin in acute minor stroke or transient ischaemic attack. N Engl J Med 2013;369(1):11-9.
10. Johnston SC, Easton JD, Farrant M, et al. Clopidogrel and Aspirin in Acute Ischaemic Stroke and High-Risk TIA. N Engl J Med 2018;379(3):215-25.
11. Pan Y, Elm JJ, Li H, et al. Outcomes Associated With Clopidogrel-Aspirin Use in Minor Stroke or Transient Ischaemic Attack: A Pooled Analysis of Clopidogrel in High-Risk Patients With Acute Non-Disabling Cerebrovascular Events (CHANCE) and Platelet-Oriented Inhibition in New TIA and Minor Ischaemic Stroke (POINT) Trials. JAMA Neurol 2019;76(12):1466-73.
12. MR CLEAN Investigators. A randomised trial of intraarterial treatment for acute ischaemic stroke. N Engl J Med 2015;372(1):11-20.
13. ESCAPE Trial Investigators. Randomised assessment of rapid endovascular treatment of ischaemic stroke. N Engl J Med 2015;372(11):1019-30.
14. EXTEND-IA Investigators. Endovascular therapy for ischaemic stroke with perfusion-imaging selection. N Engl J Med 2015;372(11):1009-18.
15. SWIFT PRIME Investigators. Stent-retriever thrombectomy after intravenous t-PA vs. t-PA alone in stroke. N Engl J Med 2015;372(24):2285-95.
16. REVASCAT Trial Investigators. Thrombectomy within 8 hours after symptom onset in ischaemic stroke. N Engl J Med 2015;372(24):2296-306.
17. HERMES collaborators. Endovascular thrombectomy after large-vessel ischaemic stroke: a meta-analysis of individual patient data from five randomised trials. Lancet 2016;387(10029):1723-31.

18. DEFUSE 3 Investigators. Thrombectomy for Stroke at 6 to 16 Hours with Selection by Perfusion Imaging. N Engl J Med 2018;378(8):708-18.
19. DAWN Trial Investigators. Thrombectomy 6 to 24 Hours after Stroke with a Mismatch between Deficit and Infarct. N Engl J Med 2018;378(1):11-21.
20. WAKE-UP Investigators. MRI-Guided Thrombolysis for Stroke with Unknown Time of Onset. N Engl J Med 2018;379(7):611-22.
21. EXTEND Investigators. Thrombolysis Guided by Perfusion Imaging up to 9 Hours after Onset of Stroke. N Engl J Med 2019;380(19):1795-803.
22. EXTEND, ECASS-4, and EPITHET Investigators. Extending thrombolysis to 4·5-9 h and wake-up stroke using perfusion imaging: a systematic review and meta-analysis of individual patient data. Lancet 2019;394(10193):139-47.
23. Thomalla G, Boutitie F, Ma H, et al. Intravenous alteplase for stroke with unknown time of onset guided by advanced imaging: systematic review and meta-analysis of individual patient data. Lancet 2020;396(10262):1574-84.
24. Huang Y, Sharma VK, Robinson T, et al. Rationale, design, and progress of the ENhanced Control of Hypertension ANd Thrombolysis strokE stuDy (ENCHANTED) trial: An international multicentre 2×2 quasi-factorial randomised controlled trial of low- vs. standard-dose rt-PA and early intensive vs. guideline-recommended blood pressure lowering in patients with acute ischaemic stroke eligible for thrombolysis treatment. Int J Stroke 2015;10(5):778-88.
25. ENCHANTED Investigators. Statistical analysis plan for evaluating low- vs. standard-dose alteplase in the ENhanced Control of Hypertension and Thrombolysis strokE stuDy (ENCHANTED). Int J Stroke 2015;10(8):1313-5.
26. ENCHANTED Investigators and Coordinators. Low-Dose versus Standard-Dose Intravenous Alteplase in Acute Ischaemic Stroke. N Engl J Med 2016;374(24):2313-23.
27. Anderson CS, Woodward M, Arima H, et al. Statistical analysis plan for evaluating different intensities of blood pressure control in the ENhanced Control of Hypertension And Thrombolysis strokE stuDy. Int J Stroke 2019;14(5):555-8.
28. ENCHANTED Investigators and Coordinators. Intensive blood pressure reduction with intravenous thrombolysis therapy for acute ischaemic stroke (ENCHANTED): an international, randomised, open-label, blinded-endpoint, phase 3 trial. Lancet 2019;393(10174):877-88.
29. IST-3 collaborative group. Association between brain imaging signs, early and late outcomes, and response to intravenous alteplase after acute ischaemic stroke in the third International Stroke Trial (IST-3): secondary analysis of a randomised controlled trial. Lancet Neurol 2015;14(5):485-96.
30. Mair G, von Kummer R, Adami A, et al. Arterial Obstruction on Computed Tomographic or Magnetic Resonance Angiography and Response to Intravenous Thrombolytics in Ischaemic Stroke. Stroke 2017;48(2):353-60.
31. Tan IY, Demchuk AM, Hopyan J, et al. CT angiography clot burden score and collateral score: correlation with clinical and radiologic outcomes in acute middle cerebral artery infarct. AJNR Am J Neuroradiol 2009;30(3):525-31.
32. INTERRSeCT Study Investigators. Association of Clinical, Imaging, and Thrombus Characteristics With Recanalisation of Visible Intracranial Occlusion in Patients With Acute Ischaemic Stroke. JAMA. 2018;320(10):1017-26.

33. Menon BK, d'Esterre CD, Qazi EM, et al. Multiphase CT Angiography: A New Tool for the Imaging Triage of Patients with Acute Ischaemic Stroke. Radiology 2015;275(2):510-20.
34. Brott TG, Haley EC Jr, Levy DE, et al. Urgent therapy for stroke. Part I. Pilot study of tissue plasminogen activator administered within 90 minutes. Stroke 1992;23(5):632-40.
35. Haley EC Jr, Levy DE, Brott TG, et al. Urgent therapy for stroke. Part II. Pilot study of tissue plasminogen activator administered 91-180 minutes from onset. Stroke 1992;23(5):641-5.
36. Japan Alteplase Clinical Trial (J-ACT) Group. Alteplase at 0.6 mg/kg for acute ischaemic stroke within 3 hours of onset: Japan Alteplase Clinical Trial (J-ACT). Stroke 2006;37(7):1810-5.
37. Stroke Acute Management with Urgent Risk-factor Assessment and Improvement Study Investigators. Routine use of intravenous low-dose recombinant tissue plasminogen activator in Japanese patients: general outcomes and prognostic factors from the SAMURAI register. Stroke 2009;40(11):3591-5.
38. Japan Alteplase Clinical Trial II Group. Effects of 0.6 mg/kg intravenous alteplase on vascular and clinical outcomes in middle cerebral artery occlusion: Japan Alteplase Clinical Trial II (J-ACT II). Stroke 2010;41(3):461-5.
39. J-MARS Investigators. Thrombolysis with 0.6 mg/kg intravenous alteplase for acute ischaemic stroke in routine clinical practice: the Japan post-Marketing Alteplase Registration Study (J-MARS). Stroke 2010;41(9):1984-9.
40. Mori E, Yoneda Y, Tabuchi M, et al. Intravenous recombinant tissue plasminogen activator in acute carotid artery territory stroke. Neurology 1992;42(5):976-82.
41. Sharma VK, Kawnayn G, Sarkar N. Acute ischaemic stroke: comparison of low-dose and standard-dose regimes of tissue plasminogen activator. Expert Rev Neurother 2013;13(8):895-902.
42. Ramaiah SS, Yan B. Low-dose tissue plasminogen activator and standard-dose tissue plasminogen activator in acute ischaemic stroke in Asian populations: a review. Cerebrovasc Dis 2013;36(3):161-6.
43. Hacke W, Kaste M, Fieschi C, et al. Randomised double-blind placebo-controlled trial of thrombolytic therapy with intravenous alteplase in acute ischaemic stroke (ECASS II). Second European-Australasian Acute Stroke Study Investigators. Lancet 1998;352(9136):1245-51.
44. SITS-MOST investigators. Thrombolysis with alteplase for acute ischaemic stroke in the Safe Implementation of Thrombolysis in Stroke-Monitoring Study (SITS-MOST): an observational study. Lancet 2007;369(9558):275-82.
45. IST-3 collaborative group. The benefits and harms of intravenous thrombolysis with recombinant tissue plasminogen activator within 6 h of acute ischaemic stroke (the third international stroke trial [IST-3]): a randomised controlled trial. Lancet 2012;379(9834):2352-63.
46. Iso H, Folsom AR, Sato S, et al. Plasma fibrinogen and its correlates in Japanese and US population samples. Arterioscler Thromb 1993;13(6):783-90.
47. Iso H, Folsom AR, Koike KA, et al. Antigens of tissue plasminogen activator and plasminogen activator inhibitor 1: correlates in nonsmoking Japanese and Caucasian men and women. Thromb Haemost 1993;70(3):475-80.

48. Kangsadalampai S, Board PG. The Val34Leu polymorphism in the A subunit of coagulation factor XIII contributes to the large normal range in activity and demonstrates that the activation peptide plays a role in catalytic activity. Blood 1998;92(8):2766-70.
49. Leng X, Hurford R, Feng X, et al. Intracranial arterial stenosis in Caucasian versus Chinese patients with TIA and minor stroke: two contemporaneous cohorts and a systematic review. J Neurol Neurosurg Psychiatry. 2021 Mar 30:jnnp-2020-325630. doi: 10.1136/jnnp-2020-325630. Online ahead of print.
50. Hurford R, Wolters FJ, Li L, et al. Prevalence, predictors, and prognosis of symptomatic intracranial stenosis in patients with transient ischaemic attack or minor stroke: a population-based cohort study. Lancet Neurol 2020;19(5):413-21.
51. Ikram MA, Seshadri S, Bis JC, et al. Genomewide association studies of stroke. N Engl J Med 2009;360(17):1718-28.
52. Söderholm M, Pedersen A, Lorentzen E, et al. Genome-wide association meta-analysis of functional outcome after ischaemic stroke. Neurology 2019;92(12):e1271-83.
53. Matsushita T, Umeno J, Hirakawa Y, et al. Association study of the polymorphisms on chromosome 12p13 with atherothrombotic stroke in the Japanese population. J Hum Genet 2010;55(7):473-6.
54. Sharma VK, Tsivgoulis G, Tan JH, et al. Feasibility and safety of intravenous thrombolysis in multiethnic Asian stroke patients in Singapore. J Stroke Cerebrovasc Dis 2010;19(6):424-30.
55. Ng KW, Sharma VK. Low-dose versus standard-dose tissue plasminogen activator for intravenous thrombolysis in Asian acute ischaemic stroke patients. Stroke 2010;41(8):e545-6.
56. Wang X, You S, Sato S, et al. Current status of intravenous tissue plasminogen activator dosage for acute ischaemic stroke: an updated systematic review. Stroke Vasc Neurol 2018;3(1):28-33.
57. Chao AC, Hsu HY, Chung CP, et al. Outcomes of thrombolytic therapy for acute ischaemic stroke in Chinese patients: the Taiwan Thrombolytic Therapy for Acute Ischaemic Stroke (TTT-AIS) study. Stroke 2010;41(5):885-90.
58. Nguyen TH, Truong AL, Ngo MB, et al. Patients with thrombolysed stroke in Vietnam have an excellent outcome: results from the Vietnam Thrombolysis Registry. Eur J Neurol 2010;17(9):1188-92.
59. Chao AC, Liu CK, Chen CH, et al. Different doses of recombinant tissue-type plasminogen activator for acute stroke in Chinese patients. Stroke 2014;45(8):2359-65.
60. Zheng M, Lei H, Cui Y, et al. Clinical efficacy and safety of hypernormal shortened door to needle time (DNT) plus individualized low-dose alteplase therapy in treating acute ischaemic stroke. Pak J Med Sci 2016;32(4):811-6.
61. Ong CT, Wong YS, Wu CS, et al. Outcome of stroke patients receiving different doses of recombinant tissue plasminogen activator. Drug Des Devel Ther 2017;11:1559-66.
62. Zhao G, Huang T, Zheng M, et al. Comparative Analysis on Low- and Standard-Dose Regimes of Alteplase Thrombolytic Therapy for Acute Ischaemic Stroke: Efficacy and Safety. Eur Neurol 2018;79(1-2):68-73.
63. Zaman Babar MU, Khan AZ, Hakim H, et al. Utilization and outcomes with low dose tissue plasminogen activator as intravenous thrombolytic therapy for ischaemic stroke at Aga Khan University Hospital, Karachi: a retrospective analysis. J Pak Med Assoc 2019;69(11):1705-10

64. Liao X, Wang Y, Pan Y, et al. Standard-dose intravenous tissue-type plasminogen activator for stroke is better than low doses. Stroke 2014;45(8):2354-8.
65. Liu M, Pan Y, Zhou L, et al. Low-dose rt-PA may not decrease the incidence of symptomatic intracranial haemorrhage in patients with high risk of symptomatic intracranial haemorrhage. Neurol Res 2019;41(5):473-79.
66. Zhou XY, Wang SS, Collins ML, et al. Efficacy and safety of different doses of intravenous tissue plasminogen activator in Chinese patients with ischaemic stroke. J Clin Neurosci 2010;17(8):988-92.
67. Chen CH, Hsieh CY, Lai TB, et al. Optimal dose for stroke thrombolysis in Asians: low dose may have similar safety and efficacy as standard dose. J Thromb Haemost 2012;10(7):1270-5.
68. Pan SM, Liu JF, Liu M, et al. Efficacy and safety of a modified intravenous recombinant tissue plasminogen activator regimen in Chinese patients with acute ischaemic stroke. J Stroke Cerebrovasc Dis 2013;22(5):690-3.
69. Kim BJ, Han MK, Park TH, et al. Low-Versus Standard-Dose Alteplase for Ischaemic Strokes Within 4.5 Hours: A Comparative Effectiveness and Safety Study. Stroke 2015;46(9):2541-8.
70. Yang J, Yu F, Liu H, et al. A Retrospective Study of thrombolysis with 0.6 mg/kg Recombinant Tissue Plasminogen Activator (rt-PA) in Mild Stroke. Sci Rep 2016;6:31344.
71. Wang X, Robinson TG, Lee TH, et al. Low-Dose vs Standard-Dose Alteplase for Patients With Acute Ischaemic Stroke: Secondary Analysis of the ENCHANTED Randomised Clinical Trial. JAMA Neurol 2017;74(11):1328-35.
72. Song L, Wang X, Robinson T, et al. Characteristics, management and response to alteplase in China versus non-China participants of the ENCHANTED trial. Stroke Vasc Neurol 2017;2(2):53-8.
73. Chen G, Wang X, Robinson TG, et al. Comparative effects of low-dose versus standard-dose alteplase in ischaemic patients with prior stroke and/or diabetes mellitus: The ENCHANTED trial. J Neurol Sci 2018;387:1-5.
74. Minhas JS, Wang X, Arima H, et al. Lipid-Lowering Pretreatment and Outcome Following Intravenous Thrombolysis for Acute Ischaemic Stroke: A Post Hoc Analysis of the Enhanced Control of Hypertension and Thrombolysis Stroke Study Trial. Cerebrovasc Dis 2018;45(5-6):213-20.
75. Carr SJ, Wang X, Olavarria VV, et al. Influence of Renal Impairment on Outcome for Thrombolysis-Treated Acute Ischaemic Stroke: ENCHANTED (Enhanced Control of Hypertension and Thrombolysis Stroke Study) Post Hoc Analysis. Stroke 2017;48(9):2605-9.
76. Robinson TG, Wang X, Arima H, et al. Low- Versus Standard-Dose Alteplase in Patients on Prior Antiplatelet Therapy: The ENCHANTED Trial (Enhanced Control of Hypertension and Thrombolysis Stroke Study). Stroke 2017;48(7):1877-83.
77. Wang X, Lee KJ, Moullaali TJ, et al. Who will benefit more from low-dose alteplase in acute ischaemic stroke? Int J Stroke 2020;15(1):39-45.
78. Wardlaw JM, Koumellis P, Liu M. Thrombolysis (different doses, routes of administration and agents) for acute ischaemic stroke. Cochrane Database Syst Rev 2013;2013(5):CD000514.
79. Liu MD, Ning WD, Wang RC, et al. Low-Dose Versus Standard-Dose Tissue Plasminogen Activator in Acute Ischaemic Stroke in Asian Populations: A Meta-Analysis. Medicine (Baltimore) 2015;94(52):e2412.

80. Tan G, Wang H, Chen S, et al. Efficacy and safety of low dose alteplase for intravenous thrombolysis in Asian stroke patients: a meta-analysis. Sci Rep 2017;7(1):16076.
81. Liu H, Zheng H, Cao Y, et al. Low- versus Standard-Dose Intravenous Tissue-Type Plasminogen Activator for Acute Ischaemic Stroke: An Updated Meta-Analysis. J Stroke Cerebrovasc Dis 2018;27(4):988-97.
82. Cheng JW, Zhang XJ, Cheng LS, et al. Low-Dose Tissue Plasminogen Activator in Acute Ischaemic Stroke: A Systematic Review and Meta-Analysis. J Stroke Cerebrovasc Dis 2018;27(2):381-90.
83. Wang X, You S, Sato S, et al. Current status of intravenous tissue plasminogen activator dosage for acute ischaemic stroke: an updated systematic review. Stroke Vasc Neurol 2018;3(1):28-33.
84. Wang X, Li J, Moullaali TJ, et al. Low-dose versus standard-dose alteplase in acute ischaemic stroke in Asian stroke registries: an individual patient data pooling study. Int J Stroke 2019;14(7):670-7.
85. Qureshi AI, Ezzeddine MA, Nasar A, et al. Prevalence of elevated blood pressure in 563,704 adult patients with stroke presenting to the ED in the United States. Am J Emerg Med 2007;25(1):32-8.
86. Appleton JP, Sprigg N, Bath PM. Blood pressure management in acute stroke. Stroke Vasc Neurol 2016;1(2):72-82.
87. Willmot M, Leonardi-Bee J, Bath PM. High blood pressure in acute stroke and subsequent outcome: a systematic review. Hypertension 2004;43(1):18-24.
88. Bangalore S, Schwamm L, Smith EE, et al. Blood pressure and in-hospital outcomes in patients presenting with ischaemic stroke. Eur Heart J 2017;38(37):2827-35.
89. Anderson CS, et al. Rapid blood-pressure lowering in patients with acute intracerebral haemorrhage. N Engl J Med 2013;368(25):2355-65.
90. Delcourt C, Huang Y, Wang J, et al. The second (main) phase of an open, randomised, multicentre study to investigate the effectiveness of an intensive blood pressure reduction in acute cerebral haemorrhage trial (INTERACT2). Int J Stroke 2010;5(2):110-6.
91. Anderson C, Heeley E, Heritier S, et al. Statistical analysis plan for the second INTEnsive blood pressure Reduction in Acute Cerebral haemorrhage Trial (INTERACT2): a large-scale investigation to solve longstanding controversy over the most appropriate management of elevated blood pressure in the hyperacute phase of intracerebral haemorrhage. Int J Stroke 2013;8(5):327-8.
92. Carcel C, Wang X, Sato S, et al. Degree and Timing of Intensive Blood Pressure Lowering on Haematoma Growth in Intracerebral Haemorrhage: Intensive Blood Pressure Reduction in Acute Cerebral Haemorrhage Trial-2 results. Stroke 2016; 47(6):1651-3.
93. Anderson CS, Huang Y, Arima H, et al. Effects of early intensive blood pressure-lowering treatment on the growth of haematoma and perihaematomal oedema in acute intracerebral haemorrhage: the Intensive Blood Pressure Reduction in Acute Cerebral Haemorrhage Trial (INTERACT). Stroke 2010;41(2):307-12.
94. Butcher KS, Jeerakathil T, Hill M, et al. The Intracerebral Haemorrhage Acutely Decreasing Arterial Pressure Trial. Stroke 2013;44(3):620-6.
95. Hemphill JC 3rd, Greenberg SM, Anderson CS, et al. Guidelines for the Management of Spontaneous Intracerebral Haemorrhage: A Guideline for Healthcare Professionals From the American Heart Association/American Stroke Association. Stroke 2015;46(7):2032-60.

96. Powers WJ, Rabinstein AA, Ackerson T, et al. 2018 Guidelines for the Early Management of Patients With Acute Ischaemic Stroke: A Guideline for Healthcare Professionals From the American Heart Association/American Stroke Association. Stroke 2018;49(3):e46-e110.
97. Sandset EC, Anderson CS, Bath PM, et al. European Stroke Organisation (ESO) guidelines on blood pressure management in acute ischaemic stroke and intracerebral haemorrhage. Eur Stroke J 2021;0(0):1-42.
98. Ahmed N, Wahlgren N, Brainin M, et al. Relationship of blood pressure, antihypertensive therapy, and outcome in ischaemic stroke treated with intravenous thrombolysis: retrospective analysis from Safe Implementation of Thrombolysis in Stroke-International Stroke Thrombolysis Register (SITS-ISTR). Stroke 2009;40(7): 2442-9.
99. Berge E, Cohen G, Lindley RI, et al. Effects of blood pressure and blood pressure-lowering treatment during the first 24 hours among patients in the Third International Stroke Trial of thrombolytic treatment for acute ischaemic stroke. Stroke 2015; 46(12): 3362-9.
100. Treurniet KM, Berkhemer OA, Immink RV, et al. A decrease in blood pressure is associated with unfavourable outcome in patients undergoing thrombectomy under general anesthesia. J Neurointerv Surg 2018;10(2):107-11.
101. Petersen NH, Ortega-Gutierrez S, Wang A, et al. Decreases in blood pressure during thrombectomy are associated with larger infarct volumes and worse functional outcome. Stroke 2019;50(7):1797-804.
102. Lee M, Ovbiagele B, Hong KS, et al. Effect of Blood Pressure Lowering in Early Ischaemic Stroke: Meta-Analysis. Stroke 2015;46(7):1883-9.
103. Bulwa Z, Gomez CR, Morales-Vidal S, et al. Management of Blood Pressure After Acute Ischaemic Stroke. Curr Neurol Neurosci Rep. 2019;19(6):29.
104. Hong L, Cheng X, Lin L, et al. The blood pressure paradox in acute ischaemic stroke. Ann Neurol 2019;85(3):331-339.
105. Anadani M, Orabi MY, Alawieh A, et al. Blood Pressure and Outcome After Mechanical Thrombectomy With Successful Revascularization. Stroke 2019;50(9):2448-54.
106. Anadani M, Arthur AS, Tsivgoulis G, et al. Blood Pressure Goals and Clinical Outcomes after Successful Endovascular Therapy: A Multicentre Study. Ann Neurol 2020;87(6):830-9.
107. Anadani M, Arthur AS, Alawieh A, et al. Blood pressure reduction and outcome after endovascular therapy with successful reperfusion: a multicentre study. J Neurointerv Surg 2020;12(10):932-6.
108. Vilela P, Rowley HA. Brain ischaemia: CT and MRI techniques in acute ischaemic stroke. Eur J Radiol 2017;96:162-72.
109. Wardlaw JM, Smith EE, Biessels GJ, et al. Neuroimaging standards for research into small vessel disease and its contribution to ageing and neurodegeneration. Lancet Neurol 2013;12(8):822-38.
110. Wardlaw JM, Mielke O. Early signs of brain infarction at CT: observer reliability and outcome after thrombolytic treatment--systematic review. Radiology 2005;235(2):444-53.
111. Byrne D, Walsh JP, Sugrue G, et al. CT Imaging of Acute Ischaemic Stroke. Can Assoc Radiol J 2020;71(3):266-80.
112. Vert C, Parra-Fariñas C, Rovira À. MR imaging in hyperacute ischaemic stroke. Eur J Radiol. 2017;96:125-132.
113. Provost C, Soudant M, Legrand L, et al. Magnetic Resonance Imaging or Computed Tomography Before Treatment in Acute Ischaemic Stroke. Stroke 2019;50(3):659-64.

114. El-Koussy M, Schroth G, Brekenfeld C, et al. Imaging of acute ischaemic stroke. Eur Neurol 2014;72(5-6):309-16.
115. Puig J, Shankar J, Liebeskind D, et al. From "Time is Brain" to "Imaging is Brain": A Paradigm Shift in the Management of Acute Ischaemic Stroke. J Neuroimaging 2020;30(5):562-71.
116. Powers WJ, Rabinstein AA, Ackerson T, et al. Guidelines for the Early Management of Patients With Acute Ischaemic Stroke: 2019 Update to the 2018 Guidelines for the Early Management of Acute Ischaemic Stroke: A Guideline for Healthcare Professionals From the American Heart Association/American Stroke Association. Stroke 2019;50(12):e344-418.
117. Smith a. B, Dillon WP, Gould R, et al. Radiation dose-reduction strategies for neuroradiology CT protocols. AJNR Am J Neuroradiol 2007;28(9):1628-32.
118. de Lucas EM, Sánchez E, Gutiérrez A, et al. CT protocol for acute stroke: tips and tricks for general radiologists. Radiographics 2008;28(6):1673-87.
119. American College of Radiology. ACR-ASNR-SPR Practice Parameter for the Performance and Interpretation of Cervicocerebral Computed Tomography Angiography (CTA). 2015;1076:1-14.
120. Raman SP, Mahesh M, Blasko RV, et al. CT scan parameters and radiation dose: practical advice for radiologists. J Am Coll Radiol 2013;10(11):840-6.
121. uideline AP, Mark A Guenin, Alan S Brody, et al. ACR Practice Guideline for Imaging Pregnant or Potentially Pregnant Adolescents and Women With Ionizing Radiation. Article 2008;1076: 23-37.
122. Leary MC, Kidwell CS, Villablanca JP, et al. Validation of computed tomographic middle cerebral artery "dot"sign: an angiographic correlation study. Stroke 2003;34(11):2636-40.
123. Farrell C, Chappell F, Armitage PA, et al. Development and initial testing of normal reference MR images for the brain at ages 65-70 and 75-80 years. Eur Radiol 2009;19(1):177-83
124. van Swieten JC, Hijdra A, Koudstaal PJ, et al. Grading white matter lesions on CT and MRI: a simple scale. J Neurol Neurosurg Psychiatry 1990;53(12):1080-3.
125. Mair G, Zhou Z, Yoshimura S, et al. Reader reliability and accuracy of brain imaging interpretation in the ENCHANTED trial: Effectiveness of specialist training for non-expert readers. ESOC 2019 (abstract). DOI: 10.13140/RG.2.2.30630.75840
126. Patel SC, Levine SR, Tilley BC, et al. Lack of clinical significance of early ischaemic changes on computed tomography in acute stroke. JAMA 2001;286(22):2830-8.
127. Torres-Mozqueda F, He J, Yeh IB, et al. An acute ischaemic stroke classification instrument that includes CT or MR angiography: the Boston acute stroke imaging scale. AJNR Am J Neuroradiol 2008;29(6):1111-7.
128. Leiva-Salinas C, Jiang B, Wintermark M. Computed Tomography, Computed Tomography Angiography, and Perfusion Computed Tomography Evaluation of Acute Ischaemic Stroke. Neuroimaging Clin N Am 2018;28(4):565-72.
129. Zhang F, Lu Z, Wang F. Advances in the pathogenesis and prevention of contrast-induced nephropathy. Life Sci 2020;259:118379.
130. Huynh K, Baghdanian AH, Baghdanian AA, et al. Updated guidelines for intravenous contrast use for CT and MRI. Emerg Radiol 2020;27(2):115-26.
131. Beckett KR, Moriarity AK, Langer JM. Safe Use of Contrast Media: What the Radiologist Needs to Know. Radiographics 2015;35(6):1738-50.

132. Byrne D, Sugrue G, Stanley E, et al. Improved detection of anterior circulation occlusions: the ‘‘delayed vessel sign’’ on multiphase CT angiography. AJNR Am J Neuroradiol 2017;38(10):1911-6
133. Yu AY, Zerna C, Assis Z, et al. Multiphase CT angiography increases detection of anterior circulation intracranial occlusion. Neurology 2016;87(6):609-16.
134. Dundamadappa S, Iyer K, Agrawal A, et al. Multiphase CT Angiography: A Useful Technique in Acute Stroke Imaging-Collaterals and Beyond. AJNR Am J Neuroradiol 2021;42(2):221-227.
135. Higashida RT, Furlan AJ, Roberts H, et al. Trial design and reporting standards for intra-arterial cerebral thrombolysis for acute ischaemic stroke. Stroke 2003;34(8):e109-37.
136. Kimura K, Iguchi Y, Shibazaki K, et al. Intravenous tissue plasminogen activator thrombolysis in patients without major arterial occlusion seems to be safe and effective. Eur Neurol 2010;64(5):258-64.
137. Lahoti S, Gokhale S, Caplan L, et al. Thrombolysis in ischaemic stroke without arterial occlusion at presentation. Stroke 2014;45(9):2722-7.
138. Mikulik R, Goldemund D, Reif M, et al. Outcome of patients with negative CT angiography results for arterial occlusion treated with intravenous thrombolysis. Stroke 2009;40(3):868-72.
139. Yeo LL, Kong WY, Paliwal P, et al. Intravenous Thrombolysis for Acute Ischaemic Stroke due to Cervical Internal Carotid Artery Occlusion. J Stroke Cerebrovasc Dis. 2016;25(10):2423-9.
140. Paciaroni M, Agnelli G, Caso V, et al. Intravenous thrombolysis for acute ischaemic stroke associated to extracranial internal carotid artery occlusion: the ICARO-2 study. Cerebrovasc Dis 2012;34(5-6):430-5.
141. Paciaroni M, Balucani C, Agnelli G, et al. Systemic Thrombolysis in Patients With Acute Ischaemic Stroke and Internal Carotid ARtery Occlusion: The ICARO Study. Stroke 2012;43(1):125-30.
142. Ospel JM, Menon BK, Demchuk AM, et al. Clinical Course of Acute Ischaemic Stroke Due to Medium Vessel Occlusion With and Without Intravenous Alteplase Treatment. Stroke 2020;51(11):3232-40
143. Seners P, Delepierre J, Turc G, et al. Thrombus Length Predicts Lack of Post-Thrombolysis Early Recanalisation in Minor Stroke With Large Vessel Occlusion. Stroke 2019;50(3):761-4.
144. Dutra BG, Tolhuisen ML, Alves HCBR, et al. Thrombus Imaging Characteristics and Outcomes in Acute Ischaemic Stroke Patients Undergoing Endovascular Treatment. Stroke 2019;50(8):2057-64.
145. Alves HC, Treurniet KM, Jansen IGH, et al. Thrombus Migration Paradox in Patients With Acute Ischaemic Stroke. Stroke 2019;50(11):3156-63.
146. Huang SJ, Diao SS, Lu Y, et al. Value of thrombus imaging in predicting the outcomes of patients with large-vessel occlusive strokes after endovascular therapy. Neurol Sci 2020;41(6):1451-8.
147. Boulouis G, Lauer A, Siddiqui AK, et al. Clinical imaging factors associated with infarct progression in patients with ischaemic stroke during transfer for mechanical thrombectomy. JAMA Neurol 2017;74(11):1361-7.
148. de Havenon A, Mlynash M, Kim-Tenser MA, et al. Results From DEFUSE 3: Good Collaterals Are Associated With Reduced Ischaemic Core Growth but Not Neurologic Outcome. Stroke 2019;50(3):632-8.

149. Zhang X, Zhang M, Ding W, et al. Distinct predictive role of collateral status on clinical outcome in variant stroke subtypes of acute large arterial occlusion. Eur J Neurol 2018;25(2):293-300.
150. Hochberg AR, Young GS. Cerebral perfusion imaging. Semin Neurol 2012;32(4):454-65.
151. Calamante F, Christensen S, Desmond PM, et al. The physiological significance of the time-to-maximum (Tmax) parameter in perfusion MRI. Stroke 2010;41(6):1169-74.
152. Wintermark M, Reichhart M, Thiran JP, et al. Prognostic accuracy of cerebral blood flow measurement by perfusion computed tomography, at the time of emergency room admission, in acute stroke patients. Ann Neurol 2002;51(4):417-32.
153. Schaefer PW, Barak ER, Kamalian S, et al. Quantitative assessment of core/penumbra mismatch in acute stroke: CT and MR perfusion imaging are strongly correlated when sufficient brain volume is imaged. Stroke 2008;39(11):2986-92.
154. Dani KA, Thomas RG, Chappell FM, et al. Computed tomography and magnetic resonance perfusion imaging in ischaemic stroke: definitions and thresholds. Ann Neurol 2011;70(3):384-401.
155. Bivard A, Levi C, Spratt N, et al. Perfusion CT in acute stroke: a comprehensive analysis of infarct and penumbra. Radiology 2013;267(2):543-50.
156. Butcher K, Parsons M, Baird T, et al. Perfusion thresholds in acute stroke thrombolysis. Stroke 2003;34(9):2159-64.
157. Wintermark M, Rowley HA, Lev MH. Acute stroke triage to intravenous thrombolysis and other therapies with advanced CT or MR imaging: pro CT. Radiology 2009;251(3):619-26.
158. Campbell BC, Christensen S, Levi CR, et al. Cerebral blood flow is the optimal CT perfusion parameter for assessing infarct core. Stroke 2011;42(12):3435-40.
159. Olivot JM, Mlynash M, Thijs VN, et al. Optimal Tmax threshold for predicting penumbral tissue in acute stroke. Stroke 2009;40(2):469-75.
160. Meyer JR, Gutierrez A, Mock B, et al. High-b-value diffusion-weighted MR imaging of suspected brain infarction. AJNR Am J Neuroradiol 2000;21(10):1821-9.
161. Vert C, Parra-Fariñas C, Rovira À. MR imaging in hyperacute ischaemic stroke. Eur J Radiol 2017;96:125-32.
162. Purushotham A, Campbell BC, Straka M, et al. Apparent diffusion coefficient threshold for delineation of ischaemic core. Int J Stroke 2015;10(3):348-53.
163. Nagaraja N, Forder JR, Warach S, et al. Reversible diffusion-weighted imaging lesions in acute ischaemic stroke: A systematic review. Neurology 2020;94(13):571-87.
164. Makin SD, Doubal FN, Dennis MS, et al. Clinically Confirmed Stroke With Negative Diffusion-Weighted Imaging Magnetic Resonance Imaging: Longitudinal Study of Clinical Outcomes, Stroke Recurrence, and Systematic Review. Stroke 2015;46(11):3142-8.
165. Azizyan A, Sanossian N, Mogensen MA, et al. Fluid-attenuated inversion recovery vascular hyperintensities: an important imaging marker for cerebrovascular disease. AJNR Am J Neuroradiol 2011;32(10):1771-5.
166. Cheng B, Ebinger M, Kufner A, et al. Hyperintense vessels on acute stroke fluid-attenuated inversion recovery imaging: associations with clinical and other MRI findings. Stroke 2012;43(11):2957-61.
167. Olindo S, Chausson N, Joux J, Saint - Vil M, et al. Fluid-attenuated inversion recovery vascular hyperintensity: an early predictor of clinical outcome in proximal middle cerebral artery occlusion. Arch Neurol 2012;69(11):1462-8.

168. Mead GE, Wardlaw JM. Detection of intraluminal thrombus in acute stroke by proton density MR imaging. Cerebrovasc Dis 1998;8(2):133-4.
169. Sanossian N, Saver JL, Alger JR, et al. Angiography reveals that fluid-attenuated inversion recovery vascular hyperintensities are due to slow flow, not thrombus. AJNR Am J Neuroradiol 2009;30(3):564-8.
170. Mahdjoub E, Turc G, Legrand L, et al. Do Fluid-Attenuated Inversion Recovery Vascular Hyperintensities Represent Good Collaterals before Reperfusion Therapy? AJNR Am J Neuroradiol 2018;39(1):77-83.
171. Lee KY, Latour LL, Luby M, et al. Distal hyperintense vessels on FLAIR: an MRI marker for collateral circulation in acute stroke? Neurology 2009;72(13):1134-9.
172. Legrand L, Tisserand M, Turc G, et al. Do FLAIR vascular hyperintensities beyond the DWI lesion represent the ischaemic penumbra? AJNR Am J Neuroradiol 2015;36(2):269-74.
173. Thomalla G, Rossbach P, Rosenkranz M, et al. Negative fluid-attenuated inversion recovery imaging identifies acute ischaemic stroke at 3 hours or less. Ann Neurol 2009;65(6):724-32.
174. Koga M, Yamamoto H, Inoue M, et al. Thrombolysis With Alteplase at 0.6 mg/kg for Stroke With Unknown Time of Onset: A Randomised Controlled Trial. Stroke 2020;51(5):1530-8.
175. Toyoda K, Inoue M, Yoshimura S, et al. Magnetic Resonance Imaging-Guided Thrombolysis (0.6 mg/kg) Was Beneficial for Unknown Onset Stroke Above a Certain Core Size: THAWS RCT Substudy. Stroke 2021;52(1):12-9.
176. Greenberg SM, Vernooij MW, Cordonnier C, et al. Cerebral microbleeds: a guide to detection and interpretation. Lancet Neurol 2009;8(2):165-74.
177. Turc G, Sallem A, Moulin S, et al. Microbleed Status and 3-Month Outcome After Intravenous Thrombolysis in 717 Patients With Acute Ischaemic Stroke. Stroke 2015;46(9):2458-63.
178. Charidimou A, Turc G, Oppenheim C, et al. Microbleeds, Cerebral Haemorrhage, and Functional Outcome After Stroke Thrombolysis. Stroke 2017;48(8):2084-90.
179. Charidimou A, Kakar P, Fox Z, et al. Cerebral microbleeds and the risk of intracerebral haemorrhage after thrombolysis for acute ischaemic stroke: systematic review and meta-analysis. J Neurol Neurosurg Psychiatry 2013;84(3):277-80.
180. Ishimaru H, Ochi M, Morikawa M, et al. Accuracy of pre- and postcontrast 3D time-of-flight MR angiography in patients with acute ischaemic stroke: correlation with catheter angiography. AJNR Am J Neuroradiol 2007;28(5):923-6.
181. Fürst G, Saleh A, Wenserski F, et al. Reliability and validity of noninvasive imaging of internal carotid artery pseudo-occlusion. Stroke 1999;30(7):1444-9.
182. Ge X, Zhou Z, Zhao H, et al. Evaluation of carotid plaque vulnerability in vivo: Correlation between dynamic contrast-enhanced MRI and MRI-modified AHA classification. J Magn Reson Imaging 2017;46(3):870-6.
183. Saba L, Micheletti G, Brinjikji W, et al. Carotid Intraplaque-Haemorrhage Volume and Its Association with Cerebrovascular Events. AJNR Am J Neuroradiol 2019;40(10):1731-7.
184. Yamagata S, Yoshida K, Miyamoto S, et al. Prediction of Stroke Risk by Detection of Haemorrhage in Carotid Plaques: Meta-Analysis of Individual Patient Data. JACC Cardiovasc Imaging 2020;13(2 Pt 1):395-406.

185. Forkert ND, Kaesemann P, Treszl A, et al. Comparison of 10 TTP and Tmax estimation techniques for MR perfusion-diffusion mismatch quantification in acute stroke. AJNR Am J Neuroradiol 2013;34(9):1697-703.
186. Lev MH. Acute stroke imaging: what is sufficient for triage to endovascular therapies? AJNR Am J Neuroradiol 2012;33(5):790-2.
187. Soun JE, Chow DS, Nagamine M, et al. Artificial Intelligence and Acute Stroke Imaging. AJNR Am J Neuroradiol 2021;42(1):2-11.
188. Nagel S, Sinha D, Day D, et al. e-ASPECTS software is non-inferior to neuroradiologists in applying the ASPECT score to computed tomography scans of acute ischaemic stroke patients. Int J Stroke 2017;12(6):615-22.
189. Nagel S, Wang X, Carcel C, et al. Clinical Utility of Electronic Alberta Stroke Program Early Computed Tomography Score Software in the ENCHANTED Trial Database. Stroke 2018;49(6):1407-11.
190. Delcourt C, Wang X, Zhou Z, et al. Brain imaging abnormalities and outcome after acute ischaemic stroke: the ENCHANTED trial. J Neurol Neurosurg Psychiatry 2020;91(12):1290-6.

Chapter 2: Thrombolysis Outcomes in Acute Ischaemic Stroke by Fluid-Attenuated Inversion Recovery Hyperintense Arteries

2.1 Link to thesis

In Chapter 1, I presented an overview of current evidence on the efficacy and safety of low-dose intravenous alteplase and early intensive BP lowering in AIS patients, followed by a summary of brain imaging in AIS and associations of imaging signs of 'brain frailty' and acute ischaemia on NCCT with clinical prognosis from the ENCHANTED trial. Although it was not required, baseline or follow-up MRI was collected during the trial conducting and adjudicated centrally for part of the ENCHANTED participants, which provides an opportunity to investigate fluid-attenuated inversion recovery hyperintense arteries (FLAIR-HAs) on MRI and clinical prognosis in thrombolysis-treated AIS patients. Prior studies reported controversial results, and this Chapter reports my efforts to determine the independent factors associated with the presence of FLAIR-HAs on baseline brain MRI and their prediction to key clinical outcomes based on the ENCHANTED data.

I have published this work:
Zhou Z, Yoshimura S, Delcourt C, Lindley RI, You S, Malavera A, Torii-Yoshimura T, Carcel C, Wang X, Chen X, Parsons MW, Demchuk AM, Wardlaw JM, Mair G, Robinson TG, Chalmers J, Xu J, Anderson CS. Thrombolysis outcomes in acute ischaemic stroke by fluid-attenuated inversion recovery hyperintense arteries. *Stroke* 2020;51(7):2240-3.

I presented this work at one international and one international conferences:

1. Zhou Z, Yoshimura S, Delcourt C, Lindley RI, You S, Malavera A, Torii-Yoshimura T, Carcel C, Wang X, Chen X, Parsons MW, Demchuk AM, Wardlaw JM, Mair G, Robinson TG, Chalmers J, Xu J, Anderson CS. Thrombolysis Outcomes in Acute Ischaemic Stroke by Fluid-Attenuated Inversion Recovery Hyperintense Arteries. 5th European Stroke Organisation Conference, Milan, Italy, 22-24 May 2019.
2. Zhou Z, Yoshimura S, Delcourt C, Lindley RI, You S, Malavera A, Torii-Yoshimura T, Carcel C, Wang X, Chen X, Parsons MW, Demchuk AM, Wardlaw JM, Mair G, Robinson TG, Chalmers J, Xu J, Anderson CS. Thrombolysis Outcomes in Acute Ischaemic Stroke

by Fluid-Attenuated Inversion Recovery Hyperintense Arteries. Stroke Society of Australasia scientific meeting 2019, Canberra, Australia, 10-13 September 2019.

2.2 Abstract

Background and purpose: To determine factors associated with fluid-attenuated inversion recovery (FLAIR) hyperintense arteries (FLAIR-HAs) on magnetic resonance imaging and their prognostic significance in thrombolysis-treated patients with acute ischaemic stroke from the ENCHANTED (Enhanced Control of Hypertension and Thrombolysis Stroke Study) trial alteplase-dose arm.

Methods: Patients with acute ischaemic stroke (N=293) with brain magnetic resonance imaging (FLAIR and diffusion-weighted imaging sequences) scanned <4.5 hours of symptom onset were assessed for location and extent (score) of FLAIR-HAs, infarct volume, large vessel occlusion (LVO), and other ischaemic signs. Logistic regression models were used to determine predictors of FLAIR-HAs and the association of FLAIR-HAs with 90-day outcomes: favourable functional outcome (primary; modified Rankin Scale scores, 0-1), other modified Rankin Scale scores, and intracerebral haemorrhage.

Results: Prior atrial fibrillation, LVO, large infarct volume, and anterior circulation infarction were independently associated with FLAIR-HAs. The rate of modified Rankin Scale scores 0 to 1 was numerically lower in patients with FLAIR-HAs versus without (69/152 [45.4%] versus 75/131 [57.3%]), as was the subset of LVO (37/93 [39.8%] versus 9/16 [56.3%]), but not in those without LVO (25/36 [69.4%] versus 60/106 [56.6%]). After adjustment for covariables, FLAIR-HAs were independently associated with increased primary outcome (adjusted odds ratio [95% CI]: overall 4.14 [1.63-10.50]; with LVO 4.92 [0.87-27.86]; no LVO 6.16 [1.57-24.14]) despite an increased risk of haemorrhagic infarct (4.77 [1.12-20.26]).

Conclusions: FLAIR-HAs are more frequent in acute ischaemic stroke with cardioembolic features and indicate potential for a favourable prognosis in thrombolysis-treated patients possibly moderated by LVO.

Registration: URL: https://www.clinicaltrials.gov. Unique identifier: NCT01422616.

2.3 Background

In patients with acute ischaemic stroke (AIS), fluid-attenuated inversion recovery hyperintense arteries (FLAIR-HAs) are often observed (>45%) on brain magnetic resonance imaging (MRI),[1] but their prognostic significance is uncertain. Herein, we report independent factors associated with FLAIR-HAs and their prognostic significance on key clinical outcomes in thrombolysis-treated patients with AIS from the alteplase-dose arm of the ENCHANTED (Enhanced Control of Hypertension and Thrombolysis Stroke Study).

2.4 Methods

2.4.1 Study design

The data that support the findings of this study are available from the first or corresponding authors upon reasonable request. The study design, patient characteristics, and main results of the ENCHANTED alteplase-dose arm have been published.[2] Ethics committee at each participating centre approved the ENCHANTED protocol, and written informed consent was obtained from patients or an appropriate legal surrogate. These analyses pertain to participants with brain MRI including FLAIR and diffusion-weighted imaging (DWI) sequences obtained <4.5 hours of symptom onset.

2.4.2 Imaging analyses

FLAIR-HAs were defined as focal, tubular, or serpentine hyperintensities that corresponded to a typical arterial course on FLAIR sequence (examples shown in appendix 2.1). FLAIR-HAs in middle cerebral artery (MCA), anterior cerebral artery, and posterior circulation (posterior cerebral artery or infratentorial region) were assessed independently by a neuroradiologist (Zien Zhou) and neurologist (Sohei Yoshimura), blind to clinical data; any discrepancies were settled by discussion. FLAIR-HAs in the MCA territory were further subdivided by M1, M2, or M3-6 segments and geographic relationship to the ischaemic lesion (surrounded, facing, or beyond DWI lesion). Infarct volume (slice thickness × sum of diffusion-restricted area on each slice) on DWI, FLAIR-HAs score (0-10), mass effect, old vascular lesions, small vessel disease, and large vessel occlusion (LVO) were measured or assessed according to prior definitions.[3–6]

2.4.3 Outcomes

Primary outcome was favourable functional outcome (mRS scores 0-1) at 90 days after randomisation. Secondary outcomes included other mRS scores (0-2, ordinal shift), any intracerebral haemorrhage (ICH), symptomatic ICH, haemorrhagic infarct, and parenchymal haemorrhage.

2.4.4 Statistical analysis

Logistic regression models were used to determine independent baseline variables associated with FLAIR-HAs, including variables with P<0.2 in univariable analyses. Stepwise removal of nonsignificant covariates identified through a likelihood-ratio test was undertaken until all remaining variables were statistically significant. The association of FLAIR-HAs with 90-day functional outcomes and ICH were estimated in logistic regression models with adjustment of covariates found to be significant in univariable analysis of patients with and without FLAIR-HAs. Proportional odds regression models were used to determine outcome on the ordinal mRS. Data were reported as odds ratios (OR) and 95% CIs, and a 2-sided P<0.05 was considered statistically significant. SAS version 7.1 was used.

2.5 Results

There were 293 patients with AIS included in these analyses, in whom 158 (53.9%) had FLAIR-HAs with MCA being the predominant vessel (appendix 2.2 and 2.3). Appendix 2.4 shows the baseline differences across patients with and without FLAIR-HAs. Independent predictors of FLAIR-HAs were prior atrial fibrillation (OR, 7.61 [95% CI, 2.52–22.93]), LVO (15.85 [6.82–36.83]), anterior circulation infarct (6.73 [2.32–19.50]), and larger infarct volume (1.05 [1.02–1.08]; appendix 2.5).

Patients with FLAIR-HAs versus those without had lower rate of good 90-day functional outcomes, whether defined by mRS scores 0 to 1 (45.4% versus 57.3%), scores 0 to 2 (57.2% versus 75.6%), or ordinal shift (Table 2.1). However, after adjustment of covariables, FLAIR-HAs were significantly associated with increased favourable outcome (mRS 0-1; adjusted OR, 4.14 [95% CI, 1.63-10.50]) and ordinal shift (2.23 [1.13-4.41]). Results were comparable after adjustment for multiple clinical covariables plus infarct volume or LVO, or adjustment for all imaging covariables alone (appendix 2.6). The association of FLAIR-HAs with functional outcome appear to vary numerically by the presence of LVO: more favourable outcomes in

those without LVO (N=145, 69.4% versus 56.6%) compared with LVO (N=115, 39.8% versus 56.3%; appendix 2.7). In the subset with MCA infarct (N=198), association with mRS scores 0 to 1 was driven by FLAIR-HAs located at distal MCA (adjusted OR, 4.27 [95% CI, 1.46-12.44]) and beyond DWI lesion (2.97, 1.19-7.38; appendix 2.8). There was a significant increase in haemorrhagic infarct risk regardless of covariables adjustment (Table 2.1).

Table 2.1. Thrombolysis outcomes by FLAIR-HAs

	FLAIR-HAs		Odds ratio (95% CI)	P value	Adjusted odds ratio (95% CI)*	P value
	With n/N (%)	Without n/N (%)				
mRS 0-1	69/152 (45.4)	75/131 (57.3)	0.62 (0.39, 0.99)	**0.05**	4.14 (1.63, 10.50)	**0.003**
mRS 0-2	87/152 (57.2)	99/131 (75.6)	0.43 (0.26, 0.72)	**0.001**	2.95 (0.99, 8.78)	0.05
mRS 0	31/152 (20.4)	33/131 (25.2)	0.50 (0.33, 0.76)	**0.001**	2.23 (1.13, 4.41)	**0.02**
1	38/152 (25.0)	42/131 (32.1)				
2	18/152 (11.8)	24/131 (18.3)				
3	11/152 (7.2)	21/131 (16.0)				
4	21/152 (13.8)	7/131 (5.3)				
5	20/152 (13.2)	1/131 (0.8)				
6	13/152 (8.6)	3/131 (2.3)				
Any ICH	65/158 (41.1)	15/135 (11.1)	5.59 (3.00, 10.42)	**<0.001**	2.03 (0.81, 5.08)	0.13
sICH†	24/158 (15.2)	7/135 (5.2)	3.27 (1.36, 7.86)	**0.008**	1.39 (0.39, 4.92)	0.61
PH	20/158 (12.7)	7/135 (5.2)	2.65 (1.08, 6.48)	**0.03**	0.78 (0.21, 2.91)	0.71
HI	30/158 (19.0)	3/135 (2.2)	10.31 (3.07, 34.62)	**<0.001**	4.77 (1.12, 20.26)	**0.03**

DWI denotes diffusion-weighted imaging; FLAIR-HAs, fluid-attenuated inversion recovery hyperintense arteries; HI, haemorrhagic infarct; ICH, intracerebral haemorrhage; mRS, modified Rankin scale; NIHSS, National Institute of Health stroke scale; PH, parenchymal haemorrhage; sICH, symptomatic ICH.

*Adjusted for clinical factors (age, systolic blood pressure, NIHSS score, prior atrial fibrillation) and imaging features (DWI volume, ischaemic lesion in anterior/posterior circulation, large vessel occlusion) at baseline, except for sICH, PH, and HI with adjusting imaging features only.

†The National Institutes of Neurological Diseases and Stroke (NINDS) criteria.

2.6 Discussion and conclusion

2.6.1 Discussion

In these post hoc analyses, significant determinants of FLAIR-HAs were related to features of AIS indicative of a cardioembolic etiology. Yet, the presence of FLAIR-HAs was independently associated with favourable 90-day functional outcome after thrombolysis, despite a significant increase in the risk of haemorrhagic infarct.

To our knowledge, a systematic assessment of the determinants of FLAIR-HAs has only been undertaken in one other study,[7] where DWI lesion volume was shown to be of borderline significance in a multivariable model (OR, 0.99 [95% CI, 0.97-1.00]). Discrepancies between the 2 studies may be due to differences in the manner in which infarct volume was defined

according to the standards of eligible MRI scans included in analyses (<12 versus <4.5 hours after symptom onset). Although FLAIR-HAs do not appear to be related to the time interval between stroke onset and MRI scanning in AIS, infarct volume would likely change with time and when large in the context of LVO will have collaterals that are likely to be visible. Thus, FLAIR-HAs are more likely visible when the ischaemic lesion is large or with LVO.

Our study represents the largest group of patients with AIS with regard to the presence of FLAIR-HAs in relation to clinical outcomes after thrombolysis, and where a large range of clinical and other imaging covariables were considered in analyses. Despite a significant increase in favourable outcome after adjustment of covariables, a potential heterogeneity by LVO was seen where early recanalisation remains more important to the prognosis than good collaterals, as reflected by FLAIR-HAs.[8] Unfortunately, few patients with both baseline and 24-hour follow-up angiography images in our study, and limited samples in subsets, hindered further stratification according to recanalisation. Evidence on the association of FLAIR-HAs with ICH after thrombolysis is sparse. A trend towards increased ICH risk was driven by haemorrhagic infarct rather than parenchymal haemorrhage could explain an improved 90-day outcome in those with FLAIR-HAs, even with a high likelihood of haemorrhagic complications after thrombolysis.

2.6.2 Strengths and limitations

Major strengths of this study are the inclusion of patients from a large, prospective, multicentre trial with high quality of data collection. Moreover, the imaging assessment was rigorous. Even so, and aside from the limited recanalisation information, the study was compromised by selection bias: the majority of ENCHANTED participants did not have baseline brain MRI in a pragmatic study, and most were Asians. Moreover, participants with missing imaging data led to a reduction of sample size for multivariable analysis. Our results, however, provide a prompt for clinicians to recognise the presence of FLAIR-HAs where MRI is used in the management of patients with AIS, especially for wake-up stroke patients as evident on the use of MRI in this clinical population recently reported.[9]

2.6.3 Conclusion

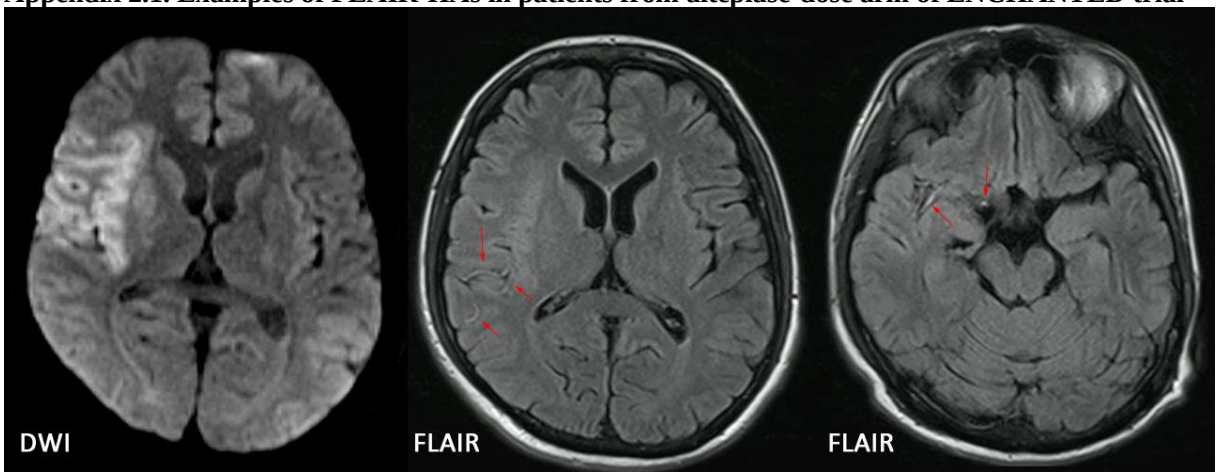
In summary, FLAIR-HAs are more frequently present in patients with AIS with cardioembolic features and indicate potential for a favourable prognosis in thrombolysis-treated patients possibly moderated by LVO.

References

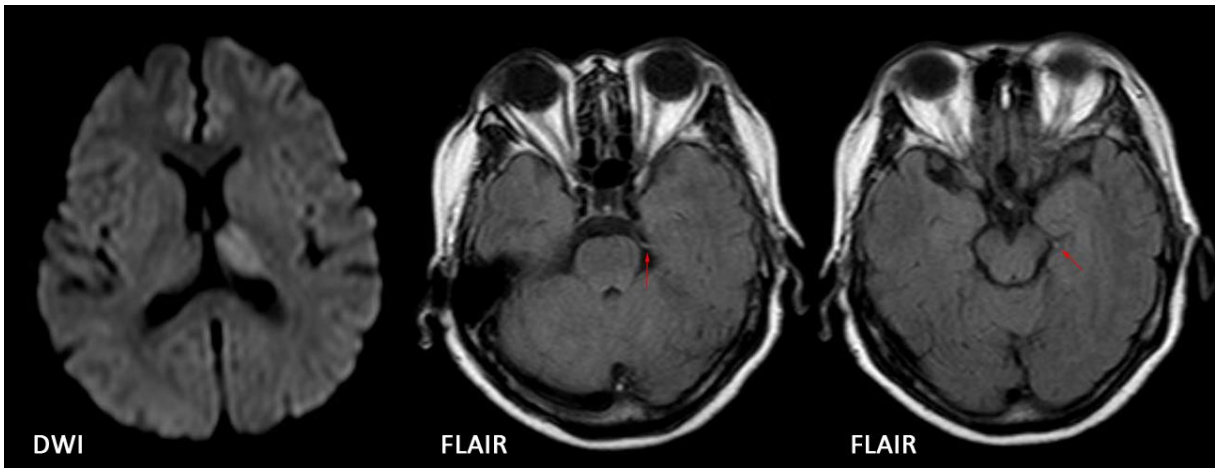
1. Azizyan A, Sanossian N, Mogensen MA, Liebeskind DS. Fluid-attenuated inversion recovery vascular hyperintensities: an important imaging marker for cerebrovascular disease. AJNR Am J Neuroradiol 2011;32(10):1771-5.
2. Anderson CS, Robinson T, Lindley RI, et al. Low-dose versus standard-dose intravenous alteplase in acute ischaemic stroke. N Engl J Med 2016;374(24):2313-23.
3. Olindo S, Chausson N, Joux J, Saint-Vil M, et al. Fluid-attenuated inversion recovery vascular hyperintensity: an early predictor of clinical outcome in proximal middle cerebral artery occlusion. Arch Neurol 2012;69(11):1462-8.
4. Wardlaw JM, Sandercock P, Cohen G, et al. Association between brain imaging signs, early and late outcomes, and response to intravenous alteplase after acute ischaemic stroke in the third International Stroke Trial (IST-3): secondary analysis of a randomised controlled trial. Lancet Neurol 2015;14(5):485-96.
5. Wardlaw JM, Smith EE, Biessels GJ, et al. Neuroimaging standards for research into small vessel disease and its contribution to ageing and neurodegeneration. Lancet Neurol 2013;12(8):822-38.
6. Mair G, von Kummer R, Adami A, et al. Arterial obstruction on computed tomographic or magnetic resonance angiography and response to intravenous thrombolytics in ischaemic stroke. Stroke 2017;48(2):353-60.
7. Cheng B, Ebinger M, Kufner A, et al. Hyperintense vessels on acute stroke fluid-attenuated inversion recovery imaging: associations with clinical and other MRI findings. Stroke 2012;43(11):2957-61.
8. Mahdjoub E, Turc G, Legrand L, et al. Do fluid-attenuated inversion recovery vascular hyperintensities represent good collaterals before reperfusion therapy? AJNR Am J Neuroradiol 2018;39(1):77-83.
9. Thomalla G, Simonsen CZ, Boutitie F, et al. MRI-guided thrombolysis for stroke with unknown time of onset. N Engl J Med 2018;379(7):611-22.

Appendices

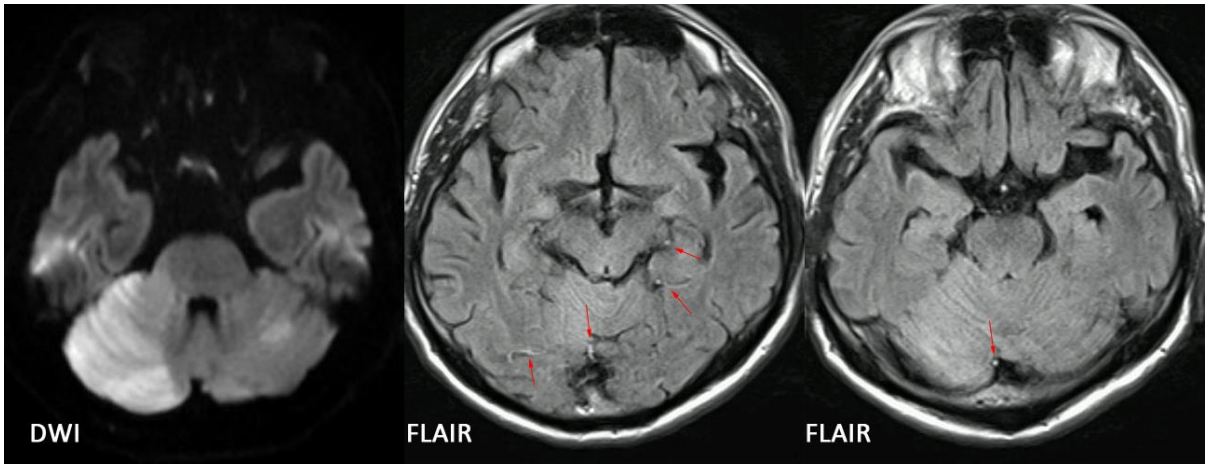
Appendix 2.1. Examples of FLAIR-HAs in patients from alteplase-dose arm of ENCHANTED trial



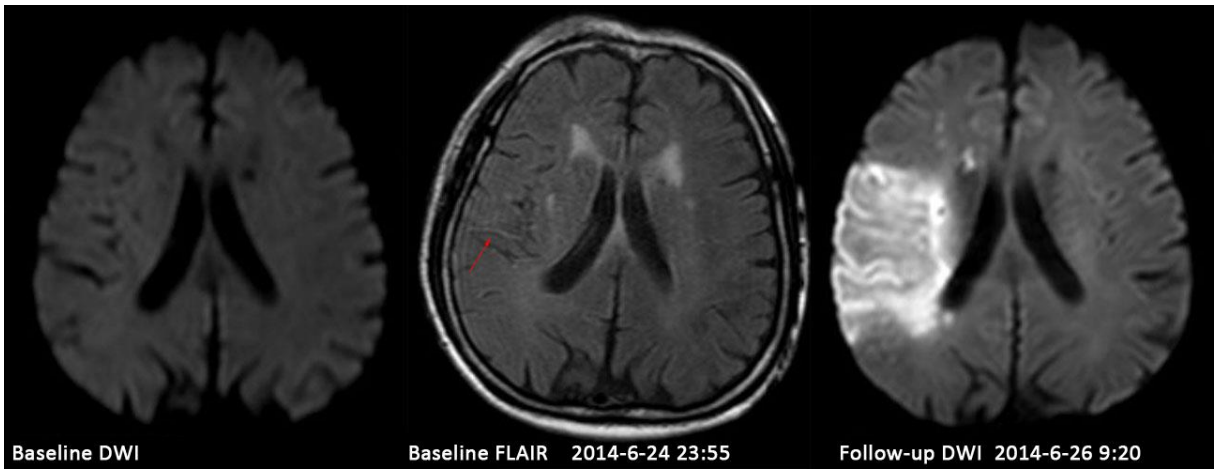
Case 1 Male, 41-year-old, with an ischaemic lesion in right MCA territory on baseline DWI. FLAIR-HAs were present at right proximal (M1 segment), middle (M2 segment/sylvian fissure), and distal (M3-6 segment) MCA on baseline FLAIR sequence (red arrows).



Case 2 Female, 67-year-old, with an ischaemic lesion at left thalamus on baseline DWI. FLAIR-HAs were present at left central branches of the PCA on baseline FLAIR sequence (red arrows).



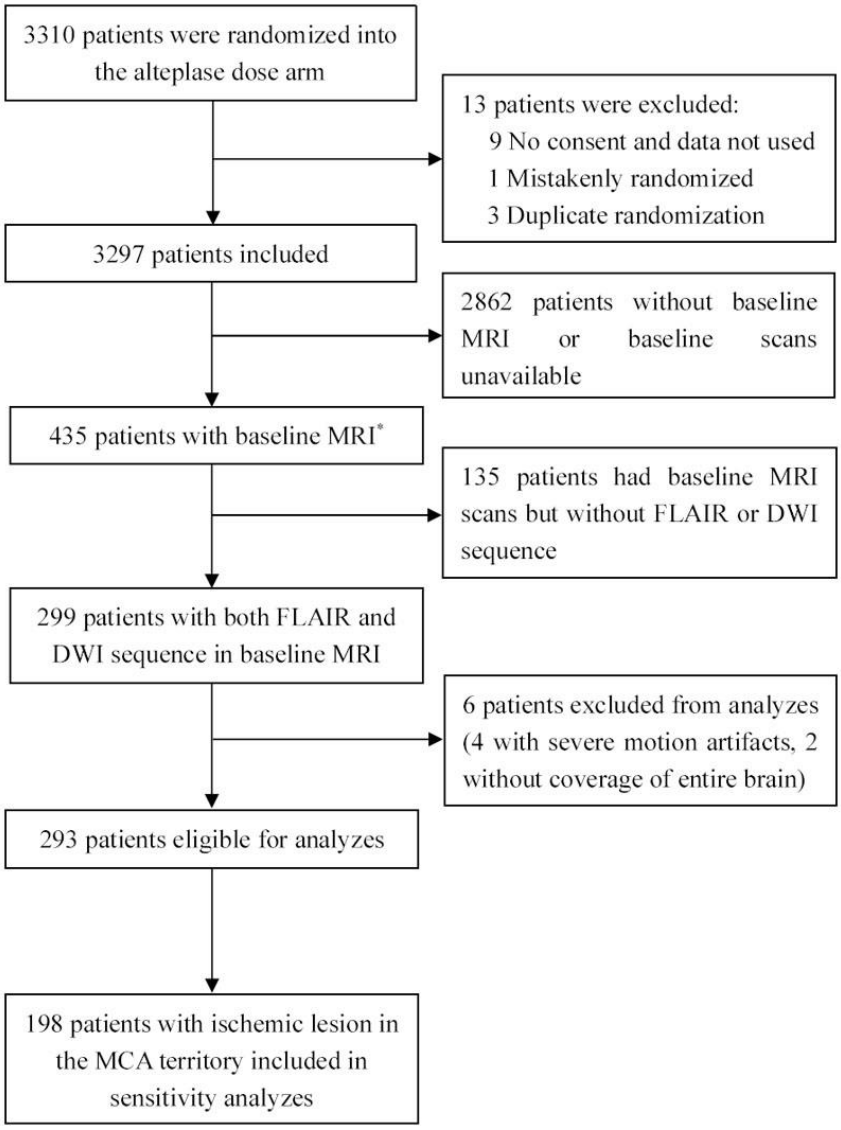
Case 3 Male, 72-year-old, with bilateral ischaemic lesions in the cerebellum on baseline DWI. FLAIR-HAs present in PCA territory and infratentorial regions on baseline FLAIR sequence (red arrows).



Case 4 Male, 70-year-old, without an ischaemic lesion on baseline DWI. FLAIR-HAs present at right distal MCA on baseline FLAIR sequence (red arrow). Ischaemic lesion in the right MCA territory on follow-up DWI.

DWI denotes diffusion-weighted imaging; ENCHANTED, Enhanced Control of Hypertension and Thrombolysis Stroke Study; FLAIR, fluid-attenuated inversion recovery; FLAIR-HAs, FLAIR hyperintense arteries; MCA, middle cerebral artery; PCA, posterior cerebral artery.

Appendix 2.2. Flowchart of patients included in analyses



DWI denotes diffusion-weighted imaging; FLAIR, fluid-attenuated inversion recovery; MCA, middle cerebral artery; MRI, magnetic resonance imaging.
*MRI obtained within 4.5 hours after stroke onset.

Appendix 2.3. Location of FLAIR-HAs and ischaemic lesion, size of ischaemic lesion, and infarct volume in patients with brain MRI including FLAIR and DWI sequences scanned <4.5 hours of symptom onset (N=293)

Patients with FLAIR-HAs (N=158)

Patients with ischemic lesion in the MCA territory were included for sensitivity analyzes (N=136)

Location of FLAIR-HAs	Only ACA	Only MCA	MCA & PCA		Only PCA	Only infratentorial region				PCA & infratentorial region	
n (%)	1 (0.6)	145 (91.8)	2 (1.3)		0 (0.0)	7 (4.4)				3 (1.9)	
Location of ischemic lesion*	AS		PL	MPS-1		No lesion	L-12	S-19	C-17	CS	PS	C-18
n	1		1	1		2	1	2	1	1	2	1
Size of ischemic lesion (IST-3)*	Small		Medium			No lesion	Small		Medium	Large	Small	Large

IST-3 codes (M-1): a) (site) =M; b) (sub-territory site) =1

Location of ischemic lesion*	No lesion	M-1	M-2,3,4a/b,5a/b	M-6,7,8	PS	MAS-4b	MAL-1	MPS-1	MPS-5b	MPS-6,7,8	L-9,11	B-15	S-19
n (%)	8 (5.1)	25 (15.8)	77 (48.7)	13 (8.2)	1 (0.6)	1 (0.6)	1 (0.6)	2 (1.3)	1 (0.6)	7 (4.4)	7 (4.4)	1 (0.6)	1 (0.6)

Size of ischemic lesion (IST-3)*	No lesion	Small (M-1, PS, L, B, S-19)	Medium (M-2,3,4a/b,5a/b, MAS-4b, MAL-1, MPS-1, MPS-5b)	Large (M-6,7,8, MPS-6,7,8)
n (%)	8 (5.1)	35 (22.2)	82 (51.9)	20 (12.7)
Volume of ischemic lesion†(cm³)	0	Tertile 1 (≥0.09 to ≤1.68)	Tertile 2 (>1.68 to ≤10.52)	Tertile 3 (>10.52 to ≤247.90)
n (%)	8 (5.1)	21 (13.3)	46 (29.1)	70 (44.3)

Patients without FLAIR-HAs (N=135)

Patients with ischemic lesion in the MCA territory were included for sensitivity analyzes (N=62)

Location of ischemic lesion*	No lesion	M-1	M-2	AS	AL	PS	MPS-1	MPS-5a	L-9,10,11	L-12	L-13	B-15	C-17	C-18	S-19
n (%)	34 (25.2)	17 (12.6)	15 (11.1)	4 (3.0)	1 (0.7)	5 (3.7)	1 (0.7)	1 (0.7)	27 (20.0)	5 (3.7)	1 (0.7)	1 (0.7)	4 (3.0)	1 (0.7)	18 (13.3)

Size of ischemic lesion*	No lesion	Small (M-1, AS, PS, L, B, S-19)	Medium (M-2, AL, MPS-1, MPS-5a, C-17)	Large (C-18)
n (%)	34 (25.2)	78 (57.8)	22 (16.3)	1 (0.7)
Volume of ischemic lesion†(cm³)	0	Tertile 1 (≥0.09 to ≤1.68)	Tertile 2 (>1.68 to ≤10.52)	Tertile 3 (>10.52 to ≤247.90)
n (%)	34 (25.2)	59 (43.7)	33 (24.4)	9 (6.7)

Among 34 patients without FLAIR-HAs and ischemic lesion on DWI at baseline, 11 were diagnosed as non-stroke at the time of hospital separation. Ischemic lesion was confirmed at the follow-up images in 12 patients.

Among 10 patients with FLAIR-HAs but without ischemic lesion on DWI at baseline, 1 was diagnosed as non-stroke at the time of hospital separation. Ischemic lesion was confirmed at the follow-up images in 7 patients.

ACA denotes anterior cerebral artery; ADC, apparent diffusion coefficient; DWI, diffusion-weighted imaging; FLAIR, fluid-attenuated inversion recovery; FLAIR-HAs, FLAIR hyperintense arteries; IST-3, the third International Stroke Trial; MCA, middle cerebral artery; PCA, posterior cerebral artery.

*Ischaemic lesion was confirmed by diffusion restricted lesion on DWI or ADC map. The site and size of ischaemic lesion was classified according to the IST-3 criteria:

a) site codes: M=MCA#=any lesion in the MCA territory; AS=Infarct of up to half of ACA territory; AL=Infarct of more than half of ACA territory; PS=Infarct of up to half of PCA territory; PL=Infarct of more than half of PCA territory; MAS=M+AS#; MAL=M+AL#; MPS=M+PS#; MPL=M+PL#; MAP=Infarct of whole MCA, ACA and PCA territories; L=Lacunar#; B=Borderzone#; C=Cerebellum#; S=Brainstem#; CS=Cerebellum and brainstem (#code sub-territory sites in b).

b) sub-territory codes: (MCA sub-territory codes) 1=small cortical infarct; 2=basal ganglia infarct (>2x2x2cm); 3= infarct of white matter lateral to the lateral ventricle (>2x2x2cm); 4a=infarct of anterior half of peripheral MCA territory (not involving basal ganglia); 4b=infarct of anterior half of peripheral MCA territory (involving part of basal ganglia); 5a=infarct of the posterior half of peripheral MCA territory (not involving basal ganglia); 5b=infarct of the posterior half of peripheral MCA territory (involving part of basal ganglia); 6=infarct of the whole of peripheral MCA territory; 7=6+infarct of lateral part of basal ganglia; 8=infarct of whole of MCA territory; (Lacunar sub-territory codes) 9=lacune in internal capsule/lentiform; 10=lacune in internal border zone; 11=lacune in centrum semiovale; 12=lacune in thalamus; 13=lacune in brainstem, inc. pons; (Borderzone sub-territory codes) 14=anterior (mainly) border zone; 15=posterior (mainly) border zone; (Cerebellum sub-territory codes) 16=small cortical; 17=<1/2 hemisphere (medium);18=>1/2 hemisphere; (Brainstem sub-territory codes) 19=small, i.e.<1/2 medulla; 20=extensive, i.e. pons + medulla.

†Infarct volume (slice thickness × sum of diffusion-restricted area on each slice, cm3) was measured on DWI.

Appendix 2.4. Baseline characteristics of patients by the presence of FLAIR-HAs

	With FLAIR-HAs (n=158)	Without FLAIR-HAs (n=135)	P value
Age (years)	66.5 (10.8)	62.8 (9.7)	**0.003**
≥80 years	5 (3.2)	0 (0.0)	**0.04**
Female	48 (30.4)	46 (34.1)	0.50
Asian ethnicity	154/157 (98.1)	132 (97.8)	0.85
Clinical features			
Systolic BP (mmHg)	142 (21)	149 (20)	**0.004**
Diastolic BP (mmHg)	83 (13)	87 (11)	**0.02**
Heart rate (beats per minute)	80 (17)	79 (13)	0.70
NIHSS score*	9 (5-15)	5 (3-8)	**<0.001**
NIHSS score ≥14	49 (31.0)	9 (6.7)	**<0.001**
GCS score†	15 (13-15)	15 (15-15)	**<0.001**
Medical history			
Previous stroke	24 (15.2)	18 (13.3)	0.65
Hypertension	88/157 (56.1)	74 (54.8)	0.83
Atrial fibrillation	58/157 (36.9)	6 (4.4)	**<0.001**
Coronary artery disease	22/157 (14.0)	8 (5.9)	**0.02**
Valvular or other heart disease	11/157 (7.0)	3 (2.2)	0.06
Diabetes mellitus	39/157 (24.8)	27 (20.0)	0.32
Hypercholesterolemia	25/157 (15.9)	15 (11.1)	0.23
Current smoker	44/157 (28.0)	55 (40.7)	**0.02**
Pre-stroke asymptomatic function (mRS=0)	16/157 (10.2)	17 (12.6)	0.52
Medication on admission			
Antihypertensive agent(s)	75/157 (47.8)	53 (39.3)	0.14
Warfarin anticoagulation	9/156 (5.8)	1/134 (0.7)	**0.02**
Aspirin/other antiplatelet agent	41/156 (26.3)	24/134 (17.9)	0.09
Statin/other lipid lowering agent	41/156 (26.3)	33/134 (24.6)	0.75
Time from stroke onset to MRI scan (hrs)	1.9 (1.4-2.7)	2.3 (1.7-2.7)	0.05
Imaging features			
With ischaemic lesion on DWI	148 (93.7)	101 (74.8)	**<0.001**
At left side	66/148 (44.6)	61/101 (60.4)	**0.01**
At right side	73/148 (49.3)	34/101 (33.7)	**0.01**
At midline or bilateral side	9/148 (6.1)	6/101 (5.9)	0.96
At anterior circulation	125/148 (84.5)	66/101 (65.3)	**<0.001**
At posterior circulation	12/148 (8.1)	33/101 (32.7)	**<0.001**
At both anterior and posterior circulation	11/148 (7.4)	2/101 (2.0)	0.06
DWI volume (cm^3)	10.5 (3.1-48.9)	1.3 (0.6-3.3)	**<0.001**
Size of ischaemic lesion (IST-3 criteria)			**<0.001**
Small	41/148 (27.7)	78/101 (77.2)	
Medium	85/148 (57.4)	22/101 (21.8)	
Large	22/148 (14.9)	1/101 (1.0)	
With signal change on FLAIR	55 (34.8)	46 (34.1)	0.90
With mass effect‡	67 (42.4)	12 (8.9)	**<0.001**
With old vascular lesions	52 (32.9)	49 (36.3)	0.54
With brain atrophy	124 (78.5)	100 (74.1)	0.38
With white matter changes	132 (83.5)	117 (86.7)	0.46
With microbleeds	30/123 (24.4)	31/106 (29.2)	0.41
Large vessel occlusion§	98/134 (73.1)	17/126 (13.5)	**<0.001**
Time from stroke onset to randomisation (hrs)	1.9 (1.3-2.9)	2.3 (1.6-3.1)	**0.04**
Randomised low-dose treatment	81 (51.3)	67 (49.6)	0.78
Assigned to intensive BP lowing	3 (1.9)	1 (0.7)	0.40
Assigned to standard BP lowing	3 (1.9)	3 (2.2)	0.85
Presumed diagnosis at time of hospital separation			
Non-stroke	1/152 (0.7)	11/134 (8.2)	**0.002**

Large artery occlusion because of atheroma	67/152 (44.1)	47/134 (35.1)	0.12
Small vessel disease	6/152 (3.9)	44/134 (32.8)	**<0.001**
Cardioembolism	59/152 (38.8)	8/134 (6.0)	**<0.001**
Dissection	2/152 (1.3)	2/134 (1.5)	0.90
Other or uncertain pathogenesis	17/152 (11.2)	22/134 (16.4)	0.20

BP denotes blood pressure; DWI, diffusion-weighted imaging; FLAIR, fluid-attenuated inversion recovery; FLAIR-HAs, FLAIR hyperintense arteries; GCS, Glasgow coma scale; IST-3, the third International Stroke Trial; MRA, magnetic resonance angiography; MRI, magnetic resonance imaging; mRS, modified Rankin scale; NIHSS, National Institutes of Health stroke scale.

Data are n (%), mean (SD), or median (Q1, Q3). P values are based on Chi-square, analysis of variance, or Wilcoxon signed-rank test.

*Scores on the NIHSS range from 0 to 42, with higher scores indicating more severe neurological deficits.

†Scores on the GCS range from 15 (normal) to 3 (deep coma).

‡Scored from 1 to 6 in assessment of mass effect on MRI were graded as: 0, no swelling; 1, effacement of the sulci overlying the infarct; 2, 1 + minor effacement of adjacent lateral ventricle; 3, 1 + complete effacement of lateral ventricle; 4, 1 + effacement of the lateral and third ventricle; 5, 4 + shift of the midline away from the side of the ventricle; 6, 5 + effacement of basal cisterns.

§Scored as 0, 1, or 2 for proximal cerebral arteries (A1-2 segment of anterior cerebral artery, internal carotid artery, M1-2 segment of middle cerebral artery, P1-2 segment of posterior cerebral artery, or basilar artery) on raw or reconstructed MRA by a modified Thrombolysis in Cerebral Infarction (TICI) score for abnormal artery: 0, no patency; 1, minimal patency – some contrast penetrates obstruction but no/minimal enters distal artery; 2, patency of <50% of the lumen at the point of obstruction and some filling of branches of the affected artery; 3, patency of >50% of the lumen and filling of most branches of the affected artery; 4, complete patency – normal artery.

Appendix 2.5. Association of baseline variables with the presence of FLAIR-HAs

	Univariate		Multivariate	
	OR (95% CI)	P value	OR (95% CI)	P value
Age (1 year higher)	1.04 (1.01, 1.06)	**0.003**¶		
Female vs. male	0.84 (0.52, 1.38)	0.50		
Asian vs. non-Asian	1.17 (0.23, 5.88)	0.85		
Clinical features				
Systolic BP (1 mmHg greater)	0.98 (0.97, 0.99)	**0.005**¶		
Diastolic BP (1 mmHg greater)	0.98 (0.96, 1.00)	**0.02**#		
Heart rate (1 beat per minute greater)	1.00 (0.99, 1.02)	0.70		
NIHSS score* (1 unit greater)	1.18 (1.12, 1.24)	**<0.001**¶		
GCS score† (1 unit greater)	0.82 (0.73, 0.92)	**0.001**#		
Medical history				
Previous stroke (Yes vs. No)	1.16 (0.60, 2.25)	0.65		
Hypertension (Yes vs. No)	1.05 (0.66, 1.67)	0.83		
Atrial fibrillation (Yes vs. No)	12.60 (5.22, 30.38)	**<0.001**¶	7.61 (2.52, 22.93)	**<0.001**
Coronary artery disease (Yes vs. No)	2.59 (1.11, 6.02)	**0.03**¶		
Valvular or other heart disease (Yes vs. No)	3.32 (0.91, 12.14)	0.07¶		
Diabetes mellitus (Yes vs. No)	1.32 (0.76, 2.30)	0.32		
Hypercholesterolemia (Yes vs. No)	1.52 (0.76, 3.01)	0.24		
Current smoker (Yes vs. No)	0.57 (0.35, 0.92)	**0.02**¶		
Pre-stroke without symptoms (mRS=0) (Yes vs. No)	0.79 (0.38, 1.63)	0.52		
Medication on admission				
Antihypertensive agent (s) (Yes vs. No)	1.42 (0.89, 2.26)	0.14¶		
Warfarin anticoagulation (Yes vs. No)	8.14 (1.02, 65.13)	**0.05**¶		
Aspirin/other antiplatelet agent (Yes vs. No)	1.63 (0.93, 2.88)	0.09¶		
Statin/other lipid lowering agent (Yes vs. No)	1.09 (0.64, 1.85)	0.75		
Time from stroke onset to MRI scan (1 hour late)	0.83 (0.64, 1.08)	0.17¶		
Imaging features				
With ischaemic lesion on DWI (Yes vs. No)	4.98 (2.36, 10.54)	**<0.001**#		
Ischaemic lesion at right vs. left side	1.98 (1.16, 3.39)	**0.01**¶		
Ischaemic lesion at anterior vs. posterior circulation	5.50 (2.67, 11.32)	**<0.001**¶	6.73 (2.32, 19.50)	**<0.001**
DWI volume (1 cm^3 greater)	1.08 (1.04, 1.11)	**<0.001**¶	1.05 (1.02, 1.08)	**0.003**
Ischaemic lesion size‡ (large or medium vs. others)	10.22 (5.84, 17.87)	**<0.001**#		
Large vessel occlusion§ (Yes vs. No)	17.45 (9.22, 33.04)	**<0.001**¶	15.85 (6.82, 36.83)	**<0.001**
With signal change on FLAIR (Yes vs. No)	1.03 (0.64, 1.68)	0.89		
With mass effect‖ (Yes vs. No)	7.54 (3.85, 14.76)	**<0.001**#		

With old vascular lesions (Yes vs. No)	0.86 (0.53, 1.40)	0.54	
With brain atrophy (Yes vs. No)	1.28 (0.74, 2.19)	0.38	
With white matter changes (Yes vs. No)	0.78 (0.41, 1.50)	0.46	
With microbleeds (Yes vs. No)	0.78 (0.43, 1.40)	0.41	C-statistic=0.91

BP denotes blood pressure; DWI, diffusion-weighted imaging; IST-3, the third International Stroke Trial; FLAIR, fluid-attenuated inversion recovery; FLAIR-HAs, FLAIR hyperintense arteries; GCS, Glasgow coma scale; MRI, magnetic resonance imaging; mRS, modified Rankin scale; NIHSS, National Institutes of Health stroke scale; OR, odds ratio.

*Scores on the NIHSS range from 0 to 42, with higher scores indicating more severe neurological deficits.

†Scores on the GCS range from 15 (normal) to 3 (deep coma).

‡Classified as small, medium or large according to the IST-3 criteria.

§Scored as 0, 1, or 2 for proximal cerebral arteries (A1-2 segment of anterior cerebral artery, internal carotid artery, M1-2 segment of middle cerebral artery, P1-2 segment of posterior cerebral artery, or basilar artery) on raw or reconstructed MRA by a modified Thrombolysis in Cerebral Infarction (TICI) score for abnormal artery: 0, no patency; 1, minimal patency – some contrast penetrates obstruction but no/minimal enters distal artery; 2, patency of <50% of the lumen at the point of obstruction and some filling of branches of the affected artery; 3, patency of >50% of the lumen and filling of most branches of the affected artery; 4, complete patency – normal artery.

‖Scored from 1 to 6 in assessment of mass effect on MRI were graded as: 0, no swelling; 1, effacement of the sulci overlying the infarct; 2, 1+minor effacement of adjacent lateral ventricle; 3, 1+complete effacement of lateral ventricle; 4, 1+effacement of the lateral and third ventricle; 5, 4+shift of the midline away from the side of the ventricle; 6, 5+effacement of the basal cisterns.

¶Included for multivariable analysis. Stepwise removal of nonsignificant covariates identified through a likelihood-ratio test was undertaken until all the remaining variables were statistically significant (P <0.05).

#As potential high correlation existed in the spearman correlation test between systolic blood pressure and diastolic blood pressure (r=0.69), NIHSS score and GCS score (r=-0.62), with ischaemic lesion and lesion volume (r=0.62), ischaemic lesion size and lesion volume (r=0.74), mass effect and lesion volume (r=0.64), variables of diastolic blood pressure, GCS score, with ischaemic lesion, ischaemic lesion size, and mass effect were not entered into multivariable analysis. Sensitivity analysis by putting ischaemic lesion size and mass effect instead of lesion volume into the model, variables of history of atrial fibrillation (OR 8.32, 95% CI 2.73-25.40), ischaemic lesion in anterior circulation (OR 5.07, 95% CI 1.70-15.12), large vessel occlusion (OR 14.13, 95% CI 6.03-33.12), large or medium ischaemic lesion (OR 2.63, 95% CI 1.15-5.99) and mass effect (OR 2.76, 95% CI 1.07-7.12) were significant (C statistic=0.91).

Appendix 2.6. Thrombolysis outcomes (mRS 0-1) by FLAIR-HAs after adjusting different covariables

	n/N	Unadjusted OR (with FLAIR-HAs vs. without)	P value
No adjustment	144/283	0.62 (0.39, 0.99)	**0.05**
Adjusted for		**Adjusted OR (with FLAIR-HAs vs. without)**	**P value**
Single clinical covariable			
Age	144/283	0.70 (0.43, 1.14)	0.15
Systolic blood pressure	144/283	0.58 (0.36, 0.93)	**0.02**
NIHSS score	144/283	1.38 (0.79, 2.43)	0.26
Prior atrial fibrillation	144/283	0.81 (0.48, 1.35)	0.41
Single imaging covariable			
Infarct volume on DWI	144/283	1.15 (0.68, 1.95)	0.61
Ischaemic lesion in anterior/posterior circulation	114/241	0.72 (0.42, 1.24)	0.24
Large vessel occlusion	131/251	1.13 (0.60, 2.13)	0.70
Combined clinical covariables			
NIHSS score, prior atrial fibrillation	144/283	1.56 (0.86, 2.86)	0.15
Age, NIHSS score, prior atrial fibrillation	144/283	1.65 (0.89, 3.03)	0.11
All clinical covariables	144/283	1.56 (0.84, 2.91)	0.16
Combined imaging covariables			
Infarct volume on DWI, large vessel occlusion	131/251	1.73 (0.88, 3.42)	0.11
All imaging covariables	103/212	2.36 (1.07, 5.18)	**0.03**
Combined clinical and imaging covariables			
All clinical covariables, infarct volume on DWI	144/283	2.29 (1.17, 4.49)	**0.02**
All clinical covariables, ischaemic lesion in anterior/posterior circulation	114/241	1.69 (0.85, 3.35)	0.14
All clinical covariables, large vessel occlusion	131/251	2.31 (1.06, 5.03)	**0.03**
All clinical covariables, infarct volume on DWI, large vessel occlusion	131/251	3.23 (1.41, 7.40)	**0.006**
All clinical and imaging covariables	103/212	4.14 (1.63, 10.50)	**0.003**

DWI denotes diffusion-weighted imaging; FLAIR, fluid-attenuated inversion recovery; FLAIR-HAs, FLAIR hyperintense arteries; mRS, modified Rankin scale; NIHSS, National Institute of Health stroke scale; OR, odds ratio.

Appendix 2.7. Thrombolysis outcomes by FLAIR-HAs in patients with and without large vessel occlusion at baseline

			FLAIR-HAs		OR	P	P inter-	AOR	P
			With, n/N (%)	Without, n/N (%)	(95% CI)	value	action*	(95% CI)†	value
mRS 0-1	With LVO‡§		37/93 (39.8)	9/16 (56.3)	0.51 (0.18, 1.50)	0.22	0.07	4.92 (0.87, 27.86)	0.07
	No LVO‡		25/36 (69.4)	60/106 (56.6)	1.74 (0.78, 3.90)	0.18		6.16 (1.57, 24.14)	**0.01**
mRS 0-2	With LVO		46/93 (49.5)	11/16 (68.8)	0.44 (0.14, 1.38)	0.16	0.09	2.00 (0.30, 13.37)	0.47
	No LVO		30/36 (83.3)	80/106 (75.5)	1.62 (0.61, 4.34)	0.33		4.81 (0.87, 26.69)	0.07
mRS shift	With LVO	0	14/93 (15.1)	3/16 (18.8)	0.46 (0.18, 1.19)	0.11	**0.03**	2.57 (0.68, 9.67)	0.16
		1	23/93 (24.7)	6/16 (37.5)					
		2	9/93 (9.7)	2/16 (12.5)					
		3	7/93 (7.5)	4/16 (25.0)					
		4	16/93 (17.2)	0/16 (0.0)					
		5	15/93 (16.1)	0/16 (0.0)					
		6	9/93 (9.7)	1/16 (6.3)					
	No LVO	0	12/36 (33.3)	27/106 (25.5)	1.52 (0.77, 3.01)	0.23		3.45 (1.35, 8.82)	**0.01**
		1	13/36 (36.1)	33/106 (31.1)					
		2	5/36 (13.9)	20/106 (18.9)					
		3	2/36 (5.6)	16/106 (15.1)					
		4	1/36 (2.8)	7/106 (6.6)					
		5	2/36 (5.6)	1/106 (0.9)					
		6	1/36 (2.8)	2/106 (1.9)					

AOR denotes adjusted odds ratio; DWI, diffusion-weighted imaging; FLAIR, fluid-attenuated inversion recovery; FLAIR-HAs, FLAIR hyperintense arteries; LVO, large vessel occlusion; mRS, modified Rankin scale; NIHSS, National Institute of Health stroke scale; OR, odds ratio.

*P value for the interaction term of functional outcomes in patients with versus without FLAIR-HAs by the presence of LVO.

†Adjusted for clinical covariables (age, systolic blood pressure, NIHSS score, history of atrial fibrillation) and imaging features (DWI volume, ischaemic lesion in anterior or posterior circulation) at baseline. ‡Among 115 patients with LVO and 145 patients without LVO on magnetic resonance angiography, 6 and 3 patients had missing 90-day mRS scores, respectively. §Scored as 0, 1, or 2 for proximal cerebral arteries (A1-2 segment of anterior cerebral artery, internal carotid artery, M1-2 segment of middle cerebral artery, P1-2 segment of posterior cerebral artery, or basilar artery) on raw or reconstructed MRA by a modified Thrombolysis in Cerebral Infarction (TICI) score for abnormal artery: 0, no patency; 1, minimal patency – some contrast penetrates obstruction but no/minimal enters distal artery; 2, patency of <50% of the lumen at the point of obstruction and some filling of branches of the affected artery; 3, patency of >50% of the lumen and filling of most branches of the affected artery; 4, complete patency – normal artery.

Appendix 2.8. Thrombolysis outcomes in MCA infarct patients by the presence of FLAIR-HAs in the MCA territory, different locations of FLAIR-HAs, FLAIR-HAs score, and whether FLAIR-HAs are surrounded or beyond the boundaries of ischaemic lesion

	n/N	OR (95% CI)	P value	AOR (95% CI)*	P value
mRS 0-1					
FLAIR-HAs in the MCA territory (with vs. without)	90/192	0.69 (0.37, 1.26)	0.22	6.61 (2.04, 21.36)	**0.002**
FLAIR-HAs score in the MCA territory (1 unit greater)	90/192	0.87 (0.79, 0.95)	**0.002**	1.13 (0.93, 1.36)	0.21
(5 to 10 vs. 0 to 4)	90/192	0.40 (0.22, 0.72)	**0.002**	1.40 (0.48, 4.05)	0.54
(7 to 10 vs. 0 to 4)	70/147	0.26 (0.12, 0.56)	**<0.001**	0.71 (0.19, 2.66)	0.61
FLAIR-HAs at proximal MCA (M1 segment) (yes vs. no)	90/192	0.37 (0.19, 0.70)	**0.002**	0.84 (0.30, 2.38)	0.75
At proximal or middle MCA (M1-M2 segment) (yes vs. no)	90/192	0.40 (0.22, 0.72)	**0.002**	1.67 (0.51, 5.45)	0.39
At distal MCA (M3-M6 segment) (yes vs. no)	90/192	0.65 (0.36, 1.19)	0.17	4.27 (1.46, 12.44)	**0.008**
FLAIR-HAs are surrounded by ischaemic lesion (yes vs. no)	90/192	0.31 (0.16, 0.60)	**<0.001**	1.22 (0.41, 3.58)	0.72
Beyond the boundaries of ischaemic lesion (yes vs. no)	90/192	1.62 (0.91, 2.88)	0.10	2.97 (1.19, 7.38)	**0.02**
mRS 0-2					
FLAIR-HAs in the MCA territory (with vs. without)	119/192	0.45 (0.23, 0.87)	**0.02**	4.79 (1.12, 20.56)	**0.04**
FLAIR-HAs score in the MCA territory (1 unit greater)	119/192	0.83 (0.75, 0.91)	**<0.001**	1.02 (0.81, 1.27)	0.89
(5 to 10 vs. 0 to 4)	119/192	0.32 (0.18, 0.60)	**<0.001**	1.02 (0.30, 3.51)	0.97
(7 to 10 vs. 0 to 4)	96/147	0.29 (0.14, 0.61)	**0.001**	0.66 (0.15, 3.01)	0.59
FLAIR-HAs at proximal MCA (M1 segment) (yes vs. no)	119/192	0.35 (0.19, 0.66)	**0.001**	1.05 (0.34, 3.24)	0.93
At proximal or middle MCA (M1-M2 segment) (yes vs. no)	119/192	0.30 (0.16, 0.57)	**<0.001**	1.71 (0.40, 7.44)	0.47
At distal MCA (M3-M6 segment) (yes vs. no)	119/192	0.45 (0.23, 0.86)	**0.02**	3.19 (0.86, 11.75)	0.08
FLAIR-HAs are surrounded by ischaemic lesion (yes vs. no)	119/192	0.21 (0.11, 0.40)	**<0.001**	0.63 (0.20, 2.02)	0.44
Beyond the boundaries of ischaemic lesion (yes vs. no)	119/192	1.68 (0.92, 3.04)	0.09	2.51 (0.89, 7.06)	0.08
Any ICH (reported or adjudicated)					
FLAIR-HAs in the MCA territory (with vs. without)	70/198	5.66 (2.50, 12.78)	**<0.001**	1.88 (0.61, 5.81)	0.28
FLAIR-HAs score in the MCA territory (1 unit greater)	70/198	1.31 (1.18, 1.46)	**<0.001**	1.17 (0.99, 1.39)	0.07
(5 to 10 vs. 0 to 4)	70/198	4.02 (2.15, 7.51)	**<0.001**	1.52 (0.58, 3.94)	0.39
(7 to 10 vs. 0 to 4)	50/151	5.59 (2.66, 11.78)	**<0.001**	2.51 (0.80, 7.86)	0.11
FLAIR-HAs at proximal MCA (M1 segment) (yes vs. no)	70/198	4.24 (2.26, 7.96)	**<0.001**	1.72 (0.70, 4.18)	0.23
At proximal or middle MCA (M1-M2 segment) (yes vs. no)	70/198	6.27 (3.02, 13.03)	**<0.001**	2.55 (0.77, 8.51)	0.13
At distal MCA (M3-M6 segment) (yes vs. no)	70/198	4.52 (2.12, 9.62)	**<0.001**	1.48 (0.53, 4.14)	0.45
FLAIR-HAs are surrounded by ischaemic lesion (yes vs. no)	70/198	3.04 (1.63, 5.69)	**<0.001**	1.09 (0.42, 2.82)	0.86
Beyond the boundaries of ischaemic lesion (yes vs. no)	70/198	1.29 (0.72, 2.32)	0.39	1.06 (0.47, 2.41)	0.89

AOR denotes adjusted odds ratio; CI, confidence interval; DWI, diffusion-weighted imaging; FLAIR, fluid-attenuated inversion recovery; FLAIR-HAs, FLAIR hyperintense arteries; ICH, intracerebral haemorrhage; MCA, middle cerebral artery; MRI, magnetic resonance imaging; mRS, modified Rankin scale; NIHSS, National Institute of Health stroke scale; OR, odds ratio.

*Adjusted for clinical covariables (systolic blood pressure, NIHSS score, history of atrial fibrillation) and imaging features (DWI volume, large vessel occlusion) at baseline.

Chapter 3: Clinical Prognosis of FLAIR Hyperintense Arteries in Ischaemic Stroke Patients: A Systematic Review and Meta-analysis

3.1 Link to thesis

In Chapter 2, I presented a study on identifying independent factors associated with the presence of FLAIR-HAs and their prediction to key clinical outcomes in thrombolysis-treated AIS patients from the ENCHANTED trial. Uncertainty remains in the literature with regard to the clinical prognosis of FLAIR-HAs in AIS patients, which prompted me to conduct a systematic review and meta-analysis on this topic after pooling data from prior studies and the ENCHANTED data. This chapter presents this review.

I have published this work:
Zhou Z, Malavera A, Yoshimura S, Delcourt C, Mair G, Al-Shahi Salman R, Demchuk AM, Wardlaw JM, Lindley RI, Anderson CS. Clinical prognosis of FLAIR hyperintense arteries in ischaemic stroke patients: a systematic review and meta-analysis. *J Neurol Neurosurg Psychiatry* 2020;91(5):475-82.

I presented this work at one international conference:
Zhou Z, Malavera A, Yoshimura S, Delcourt C, Mair G, Al-Shahi Salman R, Demchuk AM, Wardlaw JM, Lindley RI, Anderson CS. Clinical prognosis of FLAIR hyperintense arteries in ischaemic stroke patients: a systematic review and meta-analysis. 2020 European Stroke Organisation and the World Stroke Organization Jointing Conference, Vienna, Austria, 7-9 Nov 2020 (virtual online).

3.2 Abstract

Objective: We performed a systematic review and meta-analysis to determine the association of fluid-attenuated inversion recovery (FLAIR) hyperintense arteries (FLAIR-HAs) on brain MRI and prognosis after acute ischaemic stroke (AIS).

Methods: We searched Medline, Embase and Cochrane Central Register of Controlled Trials for studies reporting clinical or imaging outcomes with presence of FLAIR-HAs after AIS. Two

researchers independently assessed eligibility of retrieved studies and extracted data, including from the Enhanced Control of Hypertension and Thrombolysis Stroke Study (ENCHANTED). Outcomes were unfavourable functional outcome (primary, modified Rankin scale scores 3-6 or 2-6), death, intermediate clinical and imaging outcomes. We performed subgroup analyses by treatment or types of FLAIR-HAs defined by location (at proximal/distal middle cerebral artery (MCA), within/beyond diffusion-weighted imaging [DWI] lesion) or extent.

Results: We included 36 cohort studies (33 prospectively collected) involving 3577 patients. FLAIR-HAs were not associated with functional outcome overall (pooled risk ratio 0.87, 95% CI 0.71 to 1.06), but were significantly associated with better outcome in those receiving endovascular therapy (0.56, 95% CI 0.41 to 0.75). Contrary to FLAIR-HAs at proximal MCA or within DWI lesions, FLAIR-HAs beyond DWI lesions were associated with better outcome (0.67, 95% CI 0.57 to 0.79). FLAIR-HAs favoured recanalisation (1.21, 95% CI 1.06 to 1.38) with increased risk of intracerebral haemorrhage (2.07, 95% CI 1.37 to 3.13) and early neurological deterioration (1.93, 95% CI 1.30 to 2.85).

Conclusions: FLAIR-HAs were not associated with functional outcome overall but were associated with outcome after endovascular therapy for AIS. FLAIR-HAs were also associated with early recanalisation or haemorrhagic complications, and early neurologic deterioration.

PROSPERO registration number: CRD42019131168.

3.3 Background

Fluid-attenuated inversion recovery (FLAIR) hyperintense arteries (FLAIR-HAs) are a common sign (>45%) on brain MRI in patients with acute ischaemic stroke (AIS).[1,2] Although recognised for nearly two decades,[3,4] the prognostic significance of FLAIR-HAs has not been well defined despite reflecting the presence of a large ischaemic penumbra and good collateral circulation from slow retrograde flow distal to the occluded vessel (‘clot’) or ischaemic lesion.[5-7] Potential reasons for this uncertainty in the literature include the limitations of retrospective design, small sample size, varying assessment of FLAIR-HAs, different treatments administered to patients and heterogeneity of endpoints across studies.[8-14]

Our previous analyses of thrombolysis-treated AIS patients (n=293) from the alteplase-dose arm of the international Enhanced Control of Hypertension and Thrombolysis Stroke Study (ENCHANTED)[15] showed that presence of FLAIR-HAs was independently associated with a

favourable 90-day functional outcome, defined on the modified Rankin scale (mRS) (scores 0-1 [n=144], adjusted OR 4.14, 95% CI 1.63 to 10.50) despite an increased risk of haemorrhagic infarct (adjusted OR 4.77, 95% CI 1.12 to 20.26) after adjusting for baseline covariables (unpublished). Herein, we extended this work through a systematic review and meta-analysis of the association of FLAIR-HAs with early vascular recanalisation or reperfusion, intracerebral haemorrhage (ICH), cerebral infarct growth and clinical outcomes in AIS.

3.4 Methods

3.4.1 Search strategy

The protocol followed the Preferred Reporting Items for Systemic Reviews and Meta-Analyses statement,[16] and was registered with PROSPERO international prospective register of systematic reviews. Our search strategy was based on a combination of the following medical subject headings terms or keywords: [‘arterial hyperintensity’ OR ‘hyperintense vessels’ OR ‘vascular hyperintensities’] AND [‘FLAIR’ OR ‘MRI’] AND [‘cerebrovascular disease’ OR ‘stroke’ OR ‘cerebral infarction’] (appendix 3.1). Medline, Embase and the Cochrane Central Register of Controlled Trials were searched from inception to the end of October 2019. The clinicaltrials.gov website was searched for randomised controlled trials (RCTs) registered as completed but not yet published. There was no language restriction. Reference lists of all retrieved studies and related review articles were cross-checked for further relevant studies until no further publications were found.

3.4.2 Eligibility criteria

We included the ENCHANTED trial and other RCTs or cohort studies that recruited AIS patients (age ≥18 years) where baseline brain MRI was performed after admission and before intravenous thrombolysis or endovascular therapy. We aimed to extract data on vascular recanalisation or reperfusion, infarct growth, ICH, short/long-term clinical outcomes (measured by National Institutes of Health Stroke Scale [NIHSS] or mRS score) from reports. We excluded: (i) studies where outcomes were not reported separately for participants who did and did not have FLAIR-HAs; (ii) where the primary aim was to assess the effect of FLAIR-HAs change before and after thrombolysis on clinical prognosis, or to explore the association of FLAIR-HAs with other imaging features such as collateral flow, perfusion status or perfusion-diffusion mismatch; and (iii) reviews, editorials, letters, case or case series reports, guidelines,

technical notes and book chapters. Conference abstracts were not excluded if the data could be extracted.

In this report, we also included 220 ENCHANTED[15,17-19] participants (198 from alteplase-dose arm and 22 from blood pressure lowering arm) who had middle cerebral artery (MCA) infarct identified on MRI obtained within 4.5 hours of symptom onset with both FLAIR and diffusion-weighted imaging (DWI).

3.4.3 Study selection and data extraction

The screen of potentially eligible studies identified by searches was conducted by two authors (Zien Zhou and Alejandra Malavera) to select reports for review in full text. Each full text article was reviewed for eligibility by these authors and, for each included study, data were extracted independently using a standardised electronic form; any disagreement was settled by discussion or in consultation with two other authors (Sohei Yoshimura and Candice Delcourt). Extracted data included: (i) first author, year of publication, publication type, country or region, study design, sample size, inclusion criteria, clinical and imaging characteristics of recruited participants at baseline (age, sex, hypertension, diabetes mellitus, dyslipidaemia, atrial fibrillation [AF], coronary artery disease, history of stroke, current smoking status, NIHSS score, time from stroke onset to MRI, infarct volume, hypoperfusion volume, proximal arterial or large vessel occlusion and collateral grade); (ii) treatment administered to the study patients (intravenous thrombolysis, endovascular therapy, conservative treatment or mixed treatments); (iii) FLAIR-HAs definition or assessment method by location or extent: at proximal (M1 or M1-2 segment) or distal (M3-6 segment) MCA, within or beyond DWI lesion, FLAIR-HAs score (0-10, calculated by using 10 consecutive axial slices from the first slice with an appearance of the M1 segment of MCA, absence of FLAIR-HAs on one slice rated 0 and ≥1 present rated 1 point),[9] modified Alberta Stroke Program Early CT Score (ASPECTS) grade for FLAIR-HAs (0-7, calculated according to the spatial distribution in seven cortical areas of ASPECTS [insula, M1-M6], absence of FLAIR-HAs in one area rated 0 and present rated 1 point)[7]; and (iv) primary and other outcomes in patients who did and did not have FLAIR-HAs on baseline MRI. In cases of missing data, the authors were contacted using details given in articles or identified by internet search. Two authors (Zien Zhou and Alejandra Malavera) also judged the quality of each included studies according to the Cochrane Collaboration's tool for RCT[20] or the Newcastle-Ottawa Scale (NOS) for cohort studies.[21]

3.4.4 Outcomes

The primary outcome of interest was unfavourable functional outcome, defined by mRS scores 2-6 or 3-6. Other outcomes were: 90-day unfavourable functional outcome, any death, intermediate clinical outcomes (early neurological improvement (ENI), early neurological deterioration (END), NIHSS score change before and after treatment), and intermediate imaging outcomes (early vascular recanalisation or reperfusion, ICH, infarct growth).

3.4.5 Statistical analysis

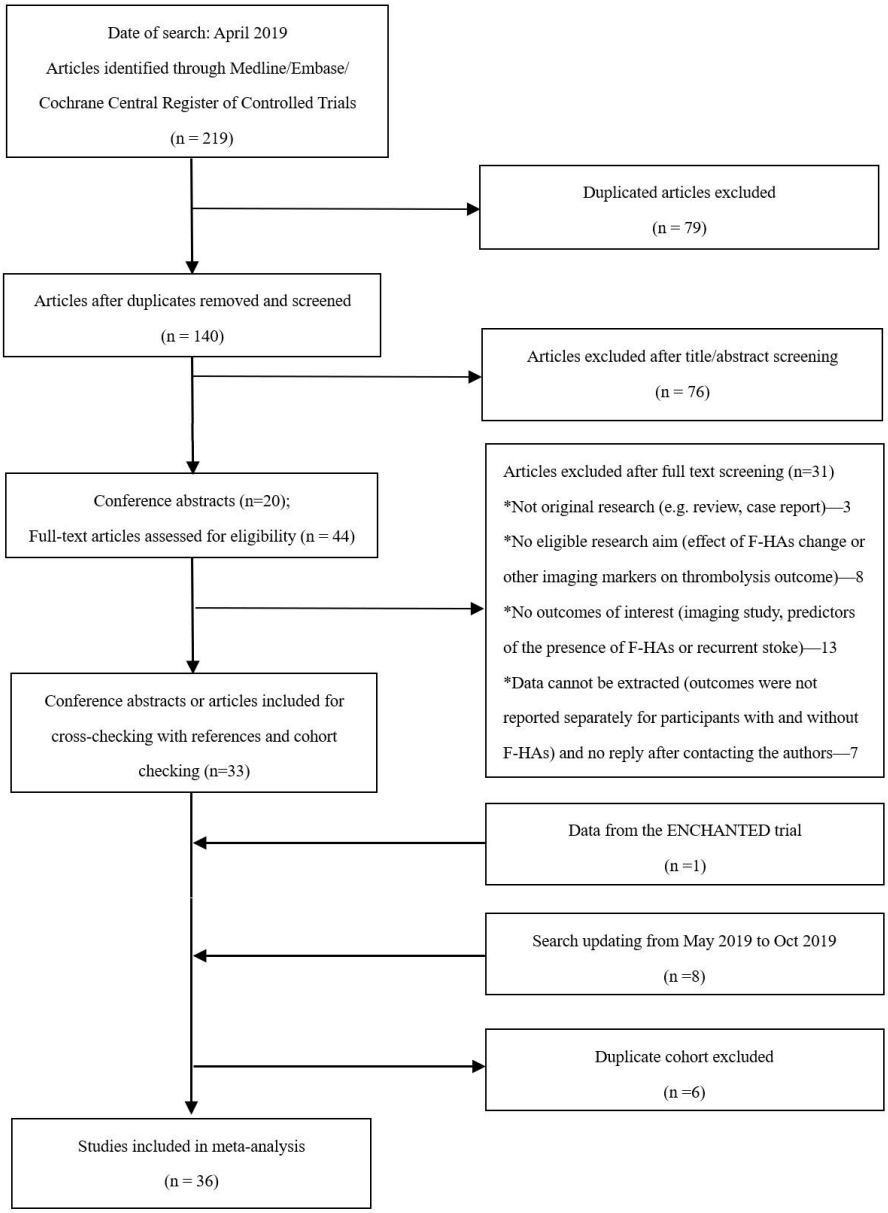
The number of dichotomous outcomes was summarised. Mean values with SD or median values with ranges or IQRs were collated for continuous outcomes. Pooled risk ratios (RR) with 95% CI were estimated for dichotomous outcomes using the DerSimonian and Laird random-effects model.[22] For changes in NIHSS scores and infarct growth, the extracted data were only tabulated, given their non-normal distribution and it was not common to estimate pooled median differences through meta-analysis. In every case, a two-sided p value ≤ 0.05 was deemed significant. The percentage of variability across the pooled estimates attributable to heterogeneity beyond chance was estimated using the I^2 statistic, and by calculating the p value for heterogeneity. I^2 values of 25%, 50% and 75% were regarded as low, moderate and high heterogeneity, respectively. Where there was a high likelihood of heterogeneity in pooled primary outcome, sensitivity analyses were performed by excluding individual studies: those without prospective data collection or studies where participants had late brain MRI (>24 hours after onset). As planned, subgroup analyses were performed for the primary outcome by treatment administered to patients, or types of FLAIR-HAs defined by location or extent. In addition, a random-effects meta-analysis was undertaken to show the association of clinical and imaging factors with the presence of FLAIR-HAs on baseline MRI. Evidence of publication bias was assessed using Egger's regression test for funnel asymmetry in addition to visual inspection of the funnel plots. All statistical analyses were performed using Stata V.12.0.

3.5 Results

3.5.1 Study selection and characteristics

The literature search in April 2019 yielded 219 potentially eligible articles or conference abstracts, of which 44 articles were reviewed in full (figure 3.1). After search updating by the

Figure 3.1. Flow chart of literature search



ENCHANTED denotes Enhanced Control of Hypertension and Thrombolysis Stroke Study; FLAIR, fluid-attenuated inversion recovery; F-HAs, FLAIR hyperintense arteries.

end of October 2019 and including data from the ENCHANTED trial, a total of 36 cohort studies (33 with prospective data collection, 6 from conference abstracts) involving 3577 AIS patients met the inclusion criteria.[5-7,9-13,23-49] Excluded studies included: not original research, no eligible research objectives or outcomes of interest, duplicate cohorts or outcome data not separately obtainable in patients with and without FLAIR-HAs. Included studies were published between 2007 and 2019, and the sample sizes ranged from 30[11] to 325,[40] with MCA infarct being the predominant lesion type (appendix 3.2). Twenty-six studies involving 2681 patients[7,9,11,12,23,25–27,29-33,35,36,38-44,46,48,49] were pooled for an assessment of functional outcome,

in which 23 studies involving 2500 patients had this reported at 90 days.[7,9,12,23,25-27,29,30,32,33,35,36,38-44,46,48] The number of eligible studies (patients) for meta-analysis of recanalisation or reperfusion at 24-48 hours was 17 (n=1487),[5,7,9,12,13,23,25,26,30,31,33-36,46,47] ICH 7 (n=1114),[9,13,28,31,40,45] ENI 5 (n=644),[7,24,30,36] END 4 (n=738)[29,40,48] and any death 3 (n=635).[12,40] Seventeen studies (n=1677)[5-7,12,13,23,25,29,31,34-37,40,42,46] reported infarct volume on admission or within 5 days after admission, in which six studies (n=705)[6,7,12,25,37,40] reported infarct growth. Data from 21 studies[6,7,9,10,12,13,23,25,29,31,33-38,40,42,43,48] were pooled for predictors with the presence of FLAIR-HAs. Only one study[13] had low quality, with more than half of the total items (6/9) being assessed as having a high risk of bias (appendix 3.3).

3.5.2 Association of FLAIR-HAs with functional outcome

Prior AF (pooled RR 1.17, 95% CI 1.01 to 1.36; p=0.04) and proximal arterial occlusion (pooled RR 1.88, 95% CI 1.28 to 2.77; p=0.001) were significantly associated with the presence of FLAIR-HAs (table 3.1). Overall, compared with no FLAIR-HAs, the presence of FLAIR-

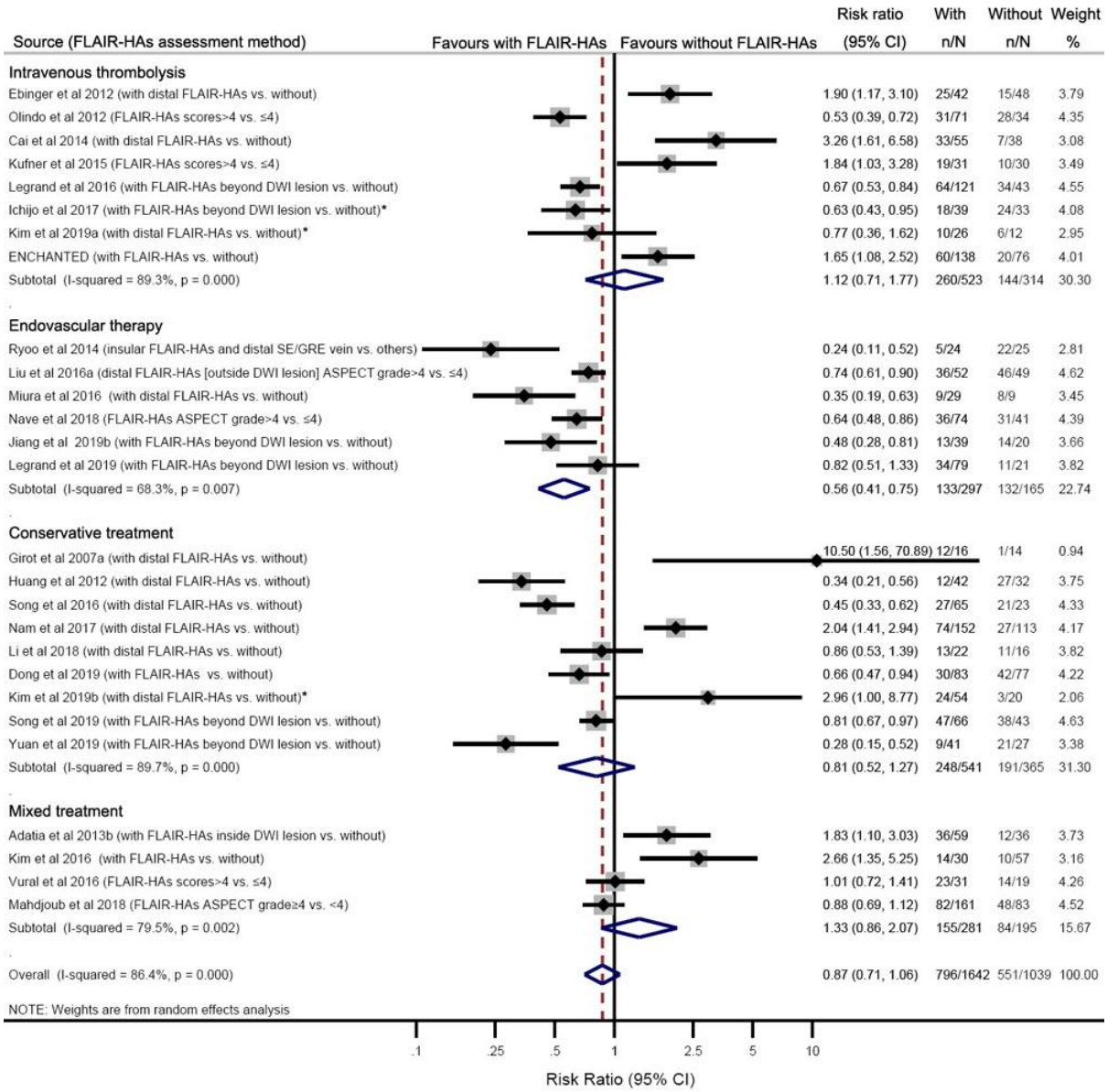
Table 3.1. Meta-analysis of the associations of baseline factors with the presence of FLAIR-HAs

Factors	No. of studies (patients)	Reference number of included studies besides ENCHANTED	Risk ratio or mean difference [95% CI]	Hetero-geneity I^2 (%)
Clinical factors				
Age (year) (with FLAIR-HAs vs. without)	14 (1304)	6,9,10,23,25,31,34-38,42,43	-0.81 (-3.27, 1.66)	61.9
Male vs. female	18 (1950)	6,7,9,10,13,23,29,31,33-38,42,43,48	0.94 (0.87, 1.02)	27.4
With hypertension vs. without	17 (1838)	6,7,9,10,13,23,29,31,33-38,42,48	0.94 (0.87, 1.02)	20.1
With DM vs. without	17 (1839)	6,7,9,10,13,23,29,31,33-38,42,48	0.97 (0.89, 1.06)	0.0
With dyslipidemia vs. without	15 (1594)	6,7,9,10,13,23,29,31,33-37,48	1.03 (0.93, 1.13)	27.0
With AF vs. without	15 (1480)	9,10,12,13,23,29,31,33-35,37,42,43,48	**1.17 (1.01, 1.36)**	64.7
With CAD vs. without	6 (775)	10,31,33,42,43	1.06 (0.87, 1.29)	42.3
With prior stroke/TIA vs. without	6 (694)	13,23,31,43,48	0.87 (0.64, 1.19)	54.2
Smoking vs. no	14 (1608)	6,7,9,23,29,33-37,42,43,48	0.94 (0.85, 1.03)	17.2
Imaging factors				
With proximal arterial occlusion vs. without	12 (1564)	7,10,12,13,23,34,38,40,42,43,48	**1.88 (1.28, 2.77)**	91.3

AF denotes atrial fibrillation; CAD, coronary artery disease; DM, diabetes mellitus; ENCHANTED, Enhanced Control of Hypertension and Thrombolysis Stroke Study; FLAIR-HAs, fluid-attenuated inversion recovery hyperintense arteries; TIA, transient ischaemic attack.

HAs was not associated with poor functional outcome (pooled RR 0.87, 95% CI 0.71 to 1.06; p=0.17). However, there was a significant association with better functional outcome in patients who had endovascular therapy (pooled RR 0.56, 95% CI 0.41 to 0.75; p<0.001) (figure 3.2).

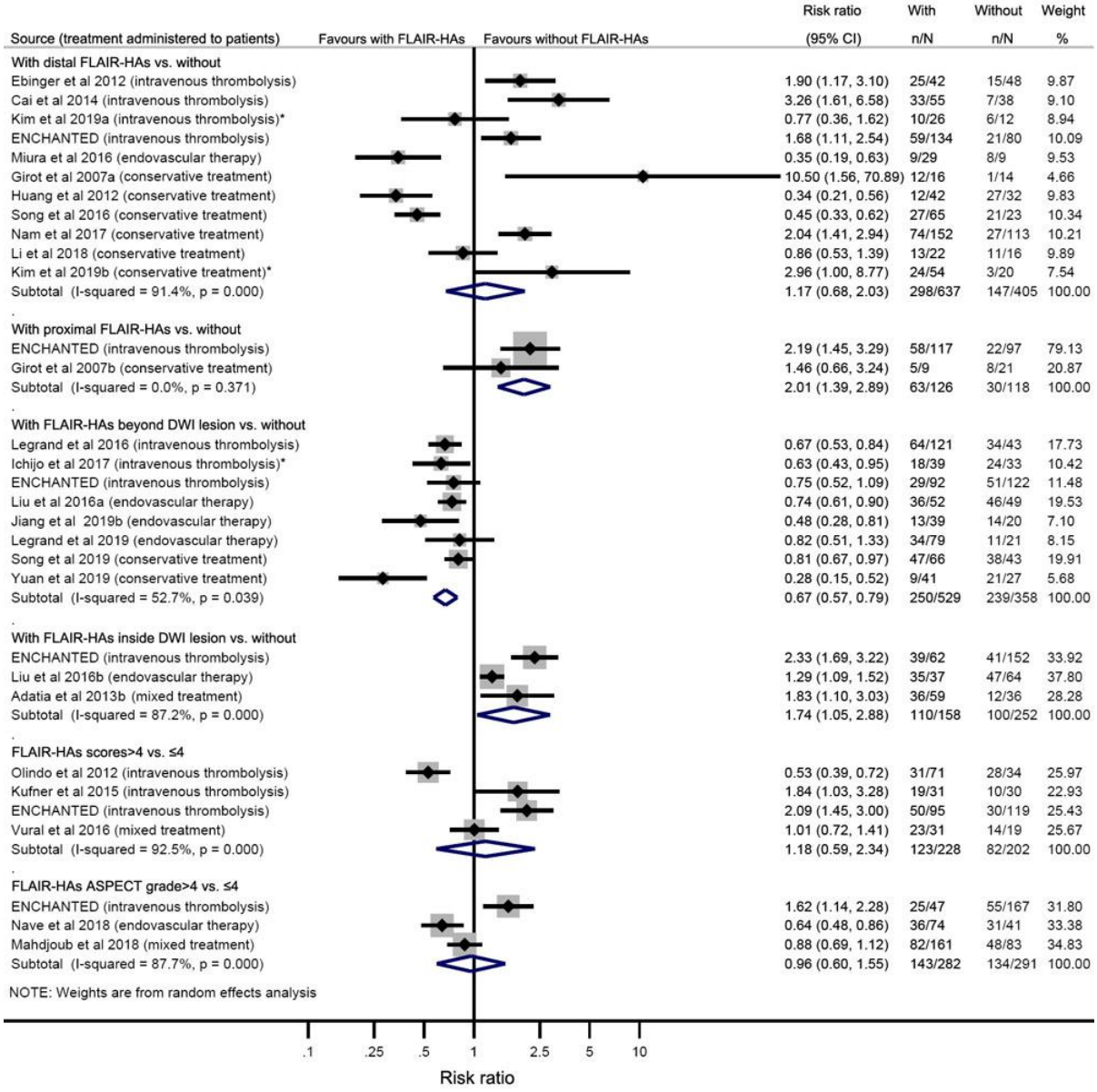
Figure 3.2. Meta-analysis of associations between FLAIR-HAs and unfavourable functional outcome, by type of treatment administered to the study patients



ASPECTS denotes Alberta Stroke Program Early CT Score; ENCHANTED, Enhanced Control of Hypertension and Thrombolysis Stroke Study; DWI, diffusion-weighted imaging; FLAIR, fluid-attenuated inversion recovery; FLAIR-HAs, FLAIR hyperintense arteries; mRS, modified Rankin scale; SE/GRE, spin echo/gradient recalled echo sequence.
*mRS scores 2-6 were regarded as unfavourable functional outcome (mRS scores 3-6 for other studies).

Comparable results were observed in sensitivity analyses for 90-day functional outcome (appendix 3.4) and after excluding studies without prospective data collection and early brain MRI (appendix 3.5). There was high heterogeneity in results pooled for functional outcome

Figure 3.3. Meta-analysis of associations between the location or extent of FLAIR-HAs and unfavourable functional outcome



ASPECTS denotes Alberta Stroke Program Early CT Score; ENCHANTED, Enhanced Control of Hypertension and Thrombolysis Stroke Study; DWI, diffusion-weighted imaging; FLAIR, fluid-attenuated inversion recovery; FLAIR-HAs, FLAIR hyperintense arteries; mRS, modified Rankin scale.
*mRS scores 2-6 was regarded as unfavourable functional outcome (mRS scores 3-6 for other studies).

(I^2=86.4%), which persisted in sensitivity analyses. FLAIR-HAs in relation to proximal MCA (pooled RR 2.01, 95% CI 1.39 to 2.89; p<0.001) and within DWI lesions (pooled RR 1.74, 95% CI 1.05 to 2.88; p=0.03), and associated with better outcome for FLAIR-HAs beyond a DWI lesion (pooled RR 0.67, 95% CI 0.57 to 0.79; p<0.001) (figure 3.3). There was no association with functional outcome for FLAIR-HAs related to distal MCA and with greater extent (FLAIR-HAs scores >4 or FLAIR-HAs ASPECTS grade >4). Heterogeneity for associations

was generally high across studies, except in the assessment of FLAIR-HAs in relation to proximal MCA (I^2=0.0%) or beyond DWI lesion (I^2=52.7%).

3.5.3 Association of FLAIR-HAs with other outcomes

Presence of FLAIR-HAs was associated with recanalisation or reperfusion at 24 to 48 hours (pooled RR 1.21, 95% CI 1.06 to 1.38; p=0.005), increased risk of ICH (pooled RR 2.07, 95% CI 1.37 to 3.13; p=0.001) and END (pooled RR 1.93, 95% CI 1.30 to 2.85; p=0.001), all with low to moderate heterogeneity (I^2 from 21.1% to 51.8%) (figure 3.4). There was no significant association with ENI (pooled RR 1.44, 95% CI 0.83 to 2.49; p=0.19) or death (pooled RR 3.60, 95% CI 0.65 to 19.81; p=0.14). Funnel plots and Egger's regression tests identified no strong evidence of publication bias for all dichotomous outcomes except recanalisation or reperfusion (p=0.01) (appendix 3.6). Data from the ENCHANTED trial as well as several other studies had shown that patients with FLAIR-HAs had greater NIHSS scores at baseline and early follow-up than those without, but there was no apparent imbalance in the level of NIHSS scores according to FLAIR-HAs beyond a DWI lesion (appendix 3.7). Moreover, compared with those without FLAIR-HAs, those with FLAIR-HAs had greater reduction in NIHSS scores between baseline and early follow-up and were more likely to have larger initial and subsequent growth in infarct volumes (appendix 3.8).

3.6 Discussion and conclusion

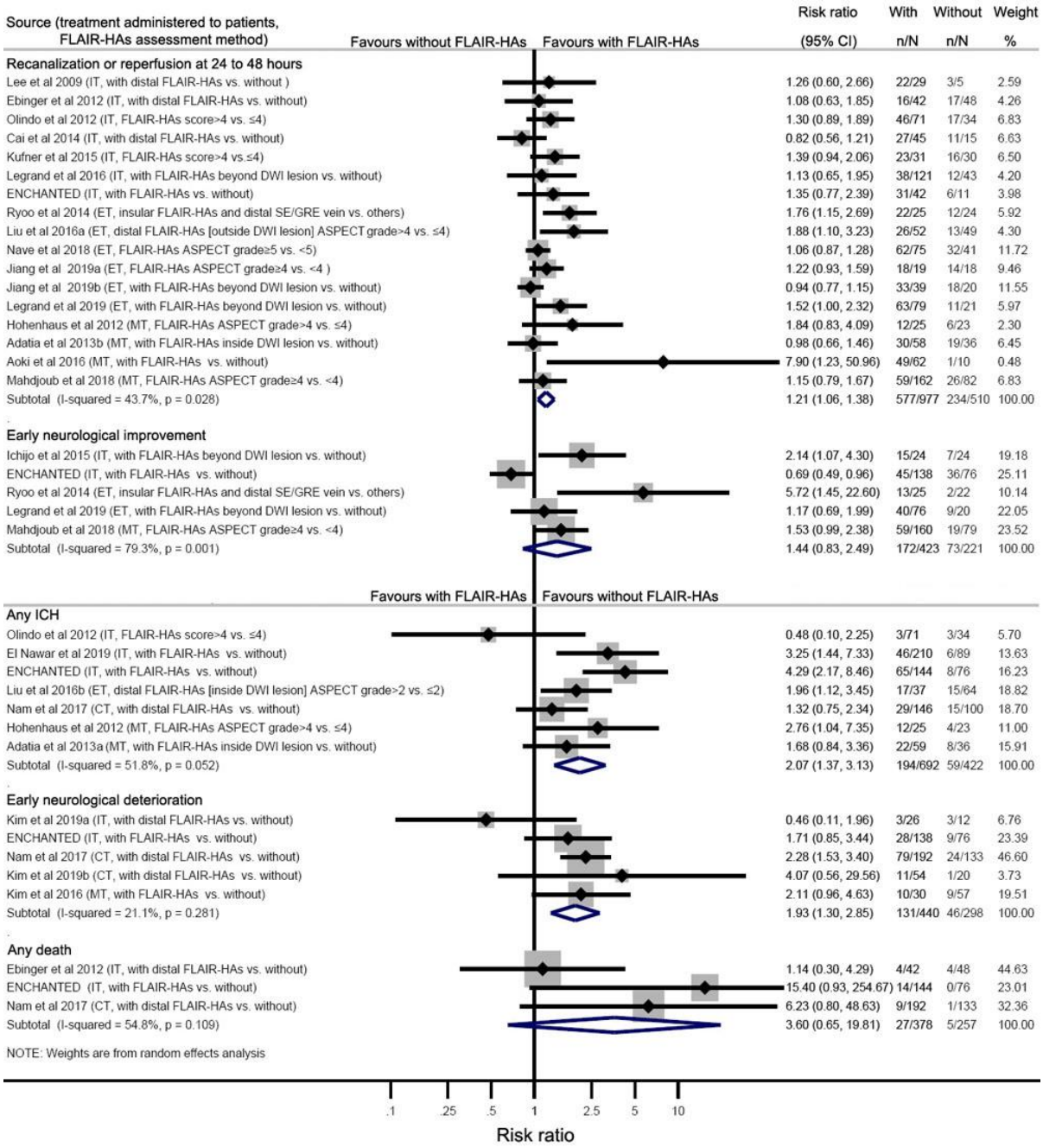
3.6.1 Discussion

In this systematic review and meta-analysis, we have identified the key determinants of FLAIR-HAs on MRI after AIS as prior AF and proximal arterial occlusion. While overall, FLAIR-HAs were not associated with functional outcome, it is associated with favourable recovery when endovascular therapy is performed. FLAIR-HAs were also associated with early recanalisation or haemorrhagic complications, and early neurologic deterioration.

We did not identify a significant association of FLAIR-HAs with functional outcome, but there was an apparent benefit in those with MCA AIS who received endovascular therapy, where the FLAIR-HAs was located beyond a DWI lesion, and when there was no proximal FLAIR-HAs or FLAIR-HAs within the DWI lesion. FLAIR-HAs were more likely to be seen with proximal arterial occlusion, suggesting they indicate retrograde filling in the ischaemic territory by leptomeningeal collateral flow. This explanation also provides a plausible mechanism for

improved outcome in such patients treated with thrombectomy. Data from ENCHANTED showed a possibility that proximal arterial occlusion status moderated the association of FLAIR

Figure 3.4. Meta-analysis of associations between FLAIR-HAs and death, intermediate clinical and imaging outcomes



ASPECTS denotes Alberta Stroke Program Early CT Score; CT, conservative treatment; ENCHANTED, Enhanced Control of Hypertension and Thrombolysis Stroke Study; DWI, diffusion-weighted imaging; ET, endovascular therapy; FLAIR, fluid-attenuated inversion recovery; FLAIR-HAs, FLAIR hyperintense arteries; ICH, intracerebral haemorrhage; IT, intravenous thrombolysis; MT, mixed treatment; SE/GRE, spin echo/gradient recalled echo sequence.

-HAs with 90-day functional outcome. FLAIR-HAs, if present, are also more likely to be assessed as within rather than beyond the DWI lesions in cases where proximal arterial

occlusion tends to cause a large ischaemic lesion on DWI. This may explain the variable associations of FLAIR-HAs within versus beyond the DWI lesion with the functional outcome.

We also found the presence of FLAIR-HAs favoured early recanalisation or reperfusion after reperfusion treatment, which is consistent with prior studies reporting good leptomeningeal collateral flow being associated with recanalisation or reperfusion after either intravenous thrombolysis or endovascular therapy.[50-53] Good leptomeningeal collaterals have been shown to be associated with permeable thrombus,[54] which is easier to lyse based on larger contact surface area between the thrombus and alteplase for use of intravenous thrombolysis or ease of thrombectomy with stent retrievers due to reduced wall tension.[55-57] Association between FLAIR-HAs and ICH could be supported by the concept that reperfusion can lead to haemorrhagic infarct (HI): Miller Fisher found petechial HI around infarcts (mainly occipital lobe) in a small post-mortem study in the 1950s;[58] or that HI is more common with proximal arterial occlusion: another small contemporaneous post-mortem study reported worse HI with proximal arterial occlusion and peripheral collaterals replicated in primates.[59] Our prior analysis of ENCHANTED participants showed that a significant increase in the risk of ICH after intravenous thrombolysis was driven by HI (adjusted OR 4.77, 95% CI 1.12 to 20.26; p=0.03) rather than parenchymal haemorrhage (adjusted OR 0.78, 95% CI 0.21 to 2.91; p=0.71) after adjustment of baseline covariables, which may explain why the clinical prognosis is not influenced even after haemorrhagic complications occur in those with FLAIR-HAs.

It is interesting that the pooled results for ENI and END were contradictory. The small number of studies included in our meta-analysis, and the different definitions of ENI or END across studies (appendix 3.9), limits the robustness of these results and raises the potential influence of the play of chance. The consistent results from the ENCHANTED data were that FLAIR-HAs were associated with an increased risk of END, and a decrease in ENI, which may be related to a greater likelihood of ICH after reperfusion in those with FLAIR-HAs versus without, but this needs further confirmation in the future.

3.6.2 Strengths and limitations

A key limitation of the study is the low number of studies and participants to allow examination of associations in subgroups and for some intermediate outcomes, raising the potential for chance finding. While caution is warranted over the interpretation of the findings in relation to FLAIR-HAs in patients who received endovascular therapy, early recanalisation or reperfusion

remains the most plausible explanatory mechanism for the positive association of FLAIR-HAs and good functional outcome. Evidence on the use of MRI in wake-up stroke patients has been provided recently.[60] Whether FLAIR-HAs may serve as an additional imaging marker to select these and other types of stroke patients with DWI-FLAIR mismatch for endovascular therapy warrants future investigation. Another issue is that we were not able to provide a strong estimate of the relation with NIHSS score change or infarct growth, which were undertaken in the ENCHANTED trial and varied according to types of FLAIR-HAs defined by location or extent. A future collaboration towards producing an individual participant data meta-analysis could provide more insights.

3.6.3 Conclusion

In summary, FLAIR-HAs were not associated with functional outcome overall but were associated with outcome after endovascular therapy for AIS. FLAIR-HAs were also associated with early recanalisation or haemorrhagic complications, and early neurologic deterioration.

References

1. Azizyan A, Sanossian N, Mogensen MA, et al. Fluid-Attenuated inversion recovery vascular hyperintensities: an important imaging marker for cerebrovascular disease. AJNR Am J Neuroradiol 2011;32(10):1771-5.
2. Cheng B, Ebinger M, Kufner A, et al. Hyperintense vessels on acute stroke fluid-attenuated inversion recovery imaging: associations with clinical and other MRI findings. Stroke 2012;43(11):2957-61.
3. Cosnard G, Duprez T, Grandin C, et al. Fast FLAIR sequence for detecting major vascular abnormalities during the hyperacute phase of stroke: a comparison with MR angiography. Neuroradiology 1999;41(5):342-6.
4. Kamran S, Bates V, Bakshi R, et al. Significance of hyperintense vessels on FLAIR MRI in acute stroke. Neurology 2000;55(2):265-9.
5. Lee KY, Latour LL, Luby M, et al. Distal hyperintense vessels on FLAIR: an MRI marker for collateral circulation in acute stroke? Neurology 2009;72(13):1134-9.
6. Legrand L, Tisserand M, Turc G, et al. Do FLAIR vascular hyperintensities beyond the DWI lesion represent the ischaemic penumbra? AJNR Am J Neuroradiol 2015;36(2):269-74.
7. Mahdjoub E, Turc G, Legrand L, et al. Do fluid-attenuated inversion recovery vascular hyperintensities represent good collaterals before reperfusion therapy? AJNR Am J Neuroradiol 2018;39(1):77-83.
8. Huang X, Liu W, Zhu W, et al. Distal hyperintense vessels on FLAIR: a prognostic indicator of acute ischaemic stroke. Eur Neurol 2012;68(4):214-20.
9. Olindo S, Chausson N, Joux J, et al. Fluid-Attenuated inversion recovery vascular hyperintensity: an early predictor of clinical outcome in proximal middle cerebral artery occlusion. Arch Neurol 2012;69(11):1462-8.
10. Pérez de la Ossa N, Hernández-Pérez M, Domènech S, et al. Hyperintensity of distal vessels on FLAIR is associated with slow progression of the infarction in acute ischaemic stroke. Cerebrovasc Dis 2012;34(5-6):376-84.
11. Girot M, Gauvrit J-Y, Cordonnier C, et al. Prognostic value of hyperintense vessel signals on fluid-attenuated inversion recovery sequences in acute cerebral ischaemia. Eur Neurol 2007;57(2):75-9.
12. Ebinger M, Kufner A, Galinovic I, et al. Fluid-Attenuated inversion recovery images and stroke outcome after thrombolysis. Stroke 2012;43(2):539-42.
13. Hohenhaus M, Schmidt WU, Brunecker P, et al. Flair vascular hyperintensities in acute ICA and MCA infarction: a marker for mismatch and stroke severity? Cerebrovasc Dis 2012;34(1):63-9.
14. Schellinger PD, Chalela JA, Kang D-W, et al. Diagnostic and prognostic value of early MR Imaging vessel signs in hyperacute stroke patients imaged <3 hours and treated with recombinant tissue plasminogen activator. AJNR Am J Neuroradiol 2005;26(3):618-24.
15. Anderson CS, Robinson T, Lindley RI, et al. Low-Dose versus standard-dose intravenous alteplase in acute ischaemic stroke. N Engl J Med 2016;374(24):2313-23.
16. Moher D, Liberati A, Tetzlaff J, et al. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. Open Med 2009;3:e123-30.

17. Anderson CS, Huang Y, Lindley RI, et al. Intensive blood pressure reduction with intravenous thrombolysis therapy for acute ischaemic stroke (enchanted): an international, randomised, open-label, blinded-endpoint, phase 3 trial. Lancet 2019;393(10174):877-88.
18. Huang Y, Sharma VK, Robinson T, et al. Rationale, design, and progress of the ENhanced Control of Hypertension ANd Thrombolysis strokE stuDy (ENCHANTED) trial: An international multicentre 2 × 2 quasi-factorial randomised controlled trial of low- vs. standard-dose rt-PA and early intensive vs. guideline-recommended blood pressure lowering in patients with acute ischaemic stroke eligible for thrombolysis treatment. Int J Stroke 2015;10(5):778-88.
19. Anderson CS, Woodward M, Arima H, et al. Statistical analysis plan for evaluating low- vs. standard-dose alteplase in the enhanced control of hypertension and thrombolysis strokE stuDy (enchanted). Int J Stroke 2015;10(8):1313-5.
20. Higgins JPT, Altman DG, Gøtzsche PC, et al. The Cochrane collaboration's tool for assessing risk of bias in randomised trials. BMJ 2011;343:d5928.
21. Wells GA, Shea B, O'Connell D, et al. The Newcastle-Ottawa scale (NOS) for assessing the quality of Nonrandomised studies in meta-analyses. The Ottawa Hospital research Institute. Available: http://ohri.ca/programs/clinical_epidemiology/ oxford.asp [Accessed 20 Feb 2015].
22. DerSimonian R, Laird N. Meta-analysis in clinical trials. Control Clin Trials 1986;7(3):177-88.
23. Cai X, Sun J, Chen Z, et al. Prognostic value of fluid-attenuated inversion recovery imaging vascular hyperintensity in intravenous thrombolysis of acute ischaemic stroke. Chinese Journal of Neurology 2014;47(9):628-32.
24. Ichijo M, Iwasawa E, Numasawa Y, et al. Significance of development and reversion of collaterals on MRI in early neurologic improvement and long-term functional outcome after intravenous thrombolysis for ischaemic stroke. AJNR Am J Neuroradiol 2015;36(10):1839-45.
25. Kufner A, Galinovic I, Ambrosi V, et al. Hyperintense vessels on FLAIR: haemodynamic correlates and response to thrombolysis. AJNR Am J Neuroradiol 2015;36(8):1426-30.
26. Legrand L, Tisserand M, Turc G, et al. Fluid-Attenuated inversion recovery vascular Hyperintensities-Diffusion-Weighted imaging mismatch identifies acute stroke patients most likely to benefit from recanalisation. Stroke 2016;47(2):424-7.
27. Ichijo M, Ishibashi S, Miki K, et al. Significance of hyperintense vessels in negative DWI area as a prognostic factor after thrombolysis (Abstract). World Congress of Neurology 2017.
28. El Nawar R, Yeung J, Labreuche J, et al. MRI-Based predictors of haemorrhagic transformation in patients with stroke treated by intravenous thrombolysis. Front Neurol 2019;10:897.
29. Kim D-H, Lee Y-K, Cha J-K. Prominent FLAIR vascular hyperintensity is a predictor of unfavourable outcomes in Non-thrombolysed ischaemic stroke patients with mild symptoms and large artery occlusion. Front Neurol 2019;10:722.
30. Ryoo S, Kim J, Lee K. Acute neuroimaging prediction of antegrade flow using FLAIR and GRE in hyperacute stroke (Abstract). Istanbul, Turkey: World Stroke Congress, 2014.
31. Liu D, Scalzo F, Rao NM, et al. Fluid-Attenuated inversion recovery vascular hyperintensity topography, novel imaging marker for revascularization in middle cerebral artery occlusion. Stroke 2016;47(11):2763-9.

32. Miura M, Nakajima M, Singu T. Distal hyperintense vessels on FLAIR is a marker of collateral flow grade and outcomes in patients with acute ischaemic stroke (Abstract). Los Angeles, CA United States: International Stroke Conference and State-of-the-Science Stroke Nursing Symposium, 2016.
33. Nave AH, Kufner A, Bücke P, et al. Hyperintense vessels, collateralization, and functional outcome in patients with stroke receiving endovascular treatment. Stroke 2018;49(3):675-81.
34. Jiang L, Chen Y-C, Zhang H, et al. Flair vascular hyperintensity in acute stroke is associated with collateralization and functional outcome. Eur Radiol 2019;29(9):4879-88.
35. Jiang L, Peng M, Geng W, et al. Flair hyperintensities-DWI mismatch in acute stroke: associations with DWI volume and functional outcome. Brain Imaging Behav 2020;14(4):1230-7.
36. Legrand L, Turc G, Edjlali M, et al. Benefit from revascularization after thrombectomy according to FLAIR vascular hyperintensities-DWI mismatch. Eur Radiol 2019;29(10):5567-76.
37. Zhou S-B, Zhang X-M, Gao Y, et al. Diffusion-Weighted imaging volume and diffusion-weighted imaging volume growth in acute stroke: associations with fluid-attenuated inversion recovery hyperintensities-diffusion-weighted imaging mismatch and functional outcome. Neuroreport 2019;30(13):875-81.
38. Huang XJ, Zhu WS, Wang QZ, et al. Prognostic value of fluid-attenuated inversion recovery hyperintense vessel in acute middle cerebral artery occlusion. Chinese Journal of Neurology 2012;45(3):174-8.
39. Song J, Ma Z, Meng H, et al. Distal hyperintense vessels alleviate insula infarction in proximal middle cerebral artery occlusion. Int J Neurosci 2016;126(11):1030-5.
40. Nam K-W, Kwon H-M, Park S-W, et al. Distal hyperintense vessel sign is associated with neurological deterioration in acute ischaemic stroke. Eur J Neurol 2017;24(4):617-23.
41. Li C-C, Hao X-Z, Tian J-Q, et al. Predictors of short-term outcome in patients with acute middle cerebral artery occlusion: unsuitability of fluid-attenuated inversion recovery vascular hyperintensity scores. Neural Regen Res 2018;13(1):69-76.
42. Dong X, Nao J. Fluid-Attenuated inversion recovery vascular hyperintensities in anterior circulation acute ischaemic stroke: associations with cortical brain infarct volume and 90-day prognosis. Neurol Sci 2019;40(8):1675-82.
43. Song L, Lyu C, Shen G, et al. Application of FLAIR vascular Hyperintensity-DWI mismatch in ischaemic stroke depending on semi-quantitative DWI-Alberta stroke program early CT score. Front Neurol 2019;10:994.
44. Yuan T, Ren G, Hu X, et al. Added assessment of middle cerebral artery and atrial fibrillation to FLAIR vascular hyperintensity-DWI mismatch would improve the outcome prediction of acute infarction in patients with acute internal carotid artery occlusion. Neurol Sci 2019;40(12):2617-24.
45. Adatia S, Almekhlafi M, Nambiar V. Flair hyperintense vessel sign on MRI brain (HVS) does not predict any intracranial haemorrhage or symptomatic haemorrhage (SICH) in acute strokes with proximal vessel occlusions treated with intravenous or intra-arterial lytic/ mechanical therapy (Abstract). Montreal, QC Canada: Canadian Stroke Congress, 2013.
46. Adatia S, Nambiar V, Sangll S. New twist to an old turn: hyperintense vessels (proximal and distal) on early FLAIR MRI brain scans help predict final infarct volume at 24 H and

outcome at 3 months in acute ischaemic stroke with proximal vessel occlusions (Abstract). Barcelona Spain: European Neurological Society, 2013.
47. Aoki J, Suzuki K, Sakamoto Y. Negative-FLAIR vascular hyperintensities serves as a marker of chronic major arterial occlusion in acute stroke (Abstract. Los Angeles, CA United States: International Stroke Conference and State-of-the-Science Stroke Nursing Symposium, 2016.
48. Kim SE, Lee BI, Kim SE, et al. Clinical significance of fluid-attenuated inversion recovery vascular hyperintensities in borderzone infarcts. Stroke 2016;47(6):1548-54.
49. Vural A, Gocmen R, Oguz KK, et al. Bright and dark vessels on stroke imaging: different sides of the same coin? Diagn Interv Radiol 2016;22(3):284-90.
50. Nicoli F, Lafaye de Micheaux P, Girard N. Perfusion-Weighted imaging-derived collateral flow index is a predictor of MCA M1 recanalisation after i.v. thrombolysis. AJNR Am J Neuroradiol 2013;34(1):107-14.
51. Calleja AI, Cortijo E, García-Bermejo P, et al. Collateral circulation on perfusion-computed tomography-source images predicts the response to stroke intravenous thrombolysis. Eur J Neurol 2013;20(5):795-802.
52. Liebeskind DS, Tomsick TA, Foster LD, et al. Collaterals at angiography and outcomes in the interventional management of stroke (IMS) II trial. Stroke 2014;45(3):759-64.
53. Nambiar V, Sohn SI, Almekhlafi MA, et al. Cta collateral status and response to recanalisation in patients with acute ischaemic stroke. AJNR Am J Neuroradiol 2014;35(5):884-90.
54. Alves HC, Treurniet KM, Dutra BG, et al. Associations between collateral status and thrombus characteristics and their impact in anterior circulation stroke. Stroke 2018;49(2):391-6.
55. Ahn SH, d'Esterre CD, Qazi EM, et al. Occult anterograde flow is an under-recognised but crucial predictor of early recanalisation with intravenous tissue-type plasminogen activator. Stroke 2015;46(4):968-75.
56. Santos EMM, Dankbaar JW, Treurniet KM, et al. Permeable thrombi are associated with higher intravenous recombinant tissue-type plasminogen activator treatment success in patients with acute ischaemic stroke. Stroke 2016;47(8):2058-65.
57. Menon BK, Al-Ajlan FS, Najm M, et al. Association of clinical, imaging, and thrombus characteristics with recanalisation of visible intracranial occlusion in patients with acute ischaemic stroke. JAMA 2018;320(10):1017-26.
58. Fisher M, Adams RD. Observations on brain embolism with special reference to the mechanism of haemorrhagic infarction. J Neuropathol Exp Neurol 1951;10(1):92-4.
59. Globus JH, Epstein JA. Massive cerebral haemorrhage: spontaneous and experimentally induced. J Neuropathol Exp Neurol 1953;12(2):107-31.
60. Thomalla G, Simonsen CZ, Boutitie F, et al. MRI-Guided thrombolysis for stroke with unknown time of onset. N Engl J Med 2018;379(7):611-22.

Appendices

Appendix 3.1. Search strategy used in the current systematic review and meta-analysis

Medline and Cochrane Central Register of Controlled Trials
1. (arterial hyperintensit* or hyperintense vessel* or vascular hyperintensit*).mp
2. exp Magnetic Resonance Imaging/ or (MRI or fluid-attenuated inversion recovery or FLAIR).mp
3. 1 and 2
4. exp Cerebrovascular Disorders/ or exp Stroke/ or exp Brain Ischaemia/ or exp Cerebral Infarction/ or exp Ischaemic Attack, Transient/
5. (cerebrovascular disorder* or cerebrovascular disease* or cerebrovascular accident* or brain vascular accident* or stroke or apoplexy* or transient isch?emic attack or TIA).mp
6. (brain or cerebr* or cerebell* or hemispher* or intracerebr* or intracran* or cerebrovasc* or brain vasc* or cerebral vasc*) adj5 (infarct* or isch?emi* or thromb* or embol* or occlus*).mp
7. 4 or 5 or 6
8. 3 and 7
9. limit 8 to human (applied for Medline)

Embase
1. (arterial hyperintensit* or hyperintense vessel* or vascular hyperintensit*).mp
2. exp nuclear magnetic resonance imaging/ or (MRI or fluid-attenuated inversion recovery or FLAIR).mp
3. 1 and 2
4. exp cerebrovascular disease/ or exp cerebrovascular accident/ or exp brain ischaemia/ or exp brain infarction/ or exp transient ischaemic attack/
5. (cerebrovascular disorder* or cerebrovascular disease* or cerebrovascular accident* or brain vascular accident* or stroke or apoplexy* or transient isch?emic attack or TIA).mp
6. (brain or cerebr* or cerebell* or hemispher* or intracerebr* or intracran* or cerebrovasc* or brain vasc* or cerebral vasc*) adj5 (infarct* or isch?emi* or thromb* or embol* or occlus*).mp
7. 4 or 5 or 6
8. 3 and 7
9. limit 8 to human

Appendix 3.2. Characteristics of included studies

Study	Design	Inclusion criteria	Analysis method of FLAIR-HAs	Sample size (N)			Factors significantly different between groups at baseline	Outcomes
				All	E	C		
Intravenous thrombolysis								
Lee et al 2009 USA[1]	R cohort (P data collect)	AIS patients with MCA infarct and within 3 hours after onset	With prominent distal F-HAs (M3-6) vs. subtle vs. none	38	32†	6†	NIHSS score, infarct volume, PWI-DWI mismatch	5-day NIHSS score, 2h and 24h recanalisation, 24h infarct volume
Ebinger et al 2012 Germany[2]	R cohort (P data collect)	AIS patients with known time of onset and had MRI before and 1 day after thrombolysis	With F-HAs that were not the main occluded vessel vs. without	90	42	48	AF, NIHSS score, infarct volume, arterial occlusion, hypoperfusion volume, PWI-DWI mismatch	90-day functional outcome, mortality, NIHSS score change, 2-day reperfusion, infarct growth
Olindo et al 2012 France[3]	R cohort (P data collect)	AIS patients with M1-MCA occlusion on MRI obtained within 4.5 h of stroke onset	F-HAs score ≤4 vs. 5-6 vs. ≥7 (in the MCA territory)	105	71‡	34‡	Infarct extent (DWI-ASPECT score)	90-day functional outcome, 24 h recanalisation, infarct growth, ICH
Cai et al 2014 China[4]	R cohort (P data collect)	AIS patients with anterior circulation infarction and had MRI before and 24 h after intravenous thrombolysis.	With F-HAs in distal MCA territory (M3-M6) vs. without	93	55	38	AF, dyslipidemia, NIHSS score, infarct volume, proximal arterial occlusion, hypoperfusion volume	90-day functional outcome, 24 h reperfusion
Ichijo et al 2015 Japan[5]	R cohort (P data collect)	AIS patients with proximal MCA occlusion treated with rtPA and underwent pretreatment MRI	F-HAs score >4 and positive PCA laterality vs. others	48	24	24	Not reported	ENI
Kufner et al 2015 Germany[6]	R cohort (P data collect)	AIS patients had MRI before and within 24 hrs after thrombolysis, being proved vessel occlusion, with CTP	F-HAs score >4 vs. ≤4 (F-HAs distal to the main occluded artery)	62	32	30	NIHSS score, infarct volume, hypoperfusion volume, PWI-DWI mismatch	90-day functional outcome, 2-day NIHSS score, 24 h recanalisation, infarct growth
Legrand et al 2015 France[7]	R cohort (P data collect)	AIS patients with M1-MCA occlusion and had pretreatment and 24 h MRI, with 3-month clinical follow-up	With F-HAs beyond the boundaries of DWI lesion vs. without	141	102	39	Diabetes mellitus, infarct volume, PWI-DWI mismatch	90-day functional outcome, 1-day NIHSS score, 24 h recanalisation, infarct growth
Legrand et al 2016 France[8]	R cohort (P data collect)	AIS patients with M1-MCA occlusion and had pretreatment and 24 h MRI, with 3-month clinical follow-up	With F-HAs beyond the boundaries of DWI lesion vs. without	164	121	43	Onset time to MRI, infarct volume,	90-day functional outcome, 24 h recanalisation, infarct growth
Ichijo et al* 2017 Japan[9]	R cohort (P data collect)	AIS patients with proximal MCA occlusion treated with rtPA and underwent pretreatment MRI	With F-HAs in negative DWI area vs. without	72	39	33	Not reported	long-term functional outcome
El Nawar et al 2019 France[10]	R cohort (P data collect)	AIS patients who were treated by intravenous thrombolysis	With F-HAs vs. without; F-HAs score ≥3 vs. <3	299	210 144	89 155	Not reported	ICH
Kim et al 2019a South Korea[11]	R cohort (P data collect)	AIS patients with initial NIHSS scores ≤5 and MCA occlusion on MRA within 24 h of stroke onset	With prominent distal F-HAs (M3-6) vs. without	38	26	12	No	90-day functional outcome, END
ENCHANT-ED	R cohort (P data collect)	AIS patients with DWI lesion in MCA territory on MRI obtained within 4.5 h after stroke onset	With F-HAs vs. without	220	144	76	NIHSS score, BP, AF, smoking, infarct volume, proximal arterial occlusion	90-day functional outcome, mortality, ENI, END, 1-day NIHSS score change, ICH, recanalisation, infarct growth
Endovascular therapy								

Ryoo et al* 2014 South Korea[12]	Not reported	AIS patients who had ischaemic stroke within 6 h and underwent endovascular therapy	With insular F-HAs (M2 and/or M3) and distal SE/GRE vein vs. others	50	25	25	Not reported	90-day functional outcome, ENI, recanalisation,
Liu et al 2016a, 2016b USA[13]	R cohort (P data collect)	AIS patients who had initial imaging demonstrating occlusion of M1-MCA	F-HAs (outside DWI)-ASPECTS >4 vs. ≤4;	101	52	49	Dyslipidemia, NIHSS score, infarct volume	Discharge functional outcome, recanalisation, ICH
			F-HAs (within DWI)-ASPECTS >2 vs. ≤2	101	37	64		
Miura et al* 2016 Japan[14]	R cohort	AIS patients who had ICA or MCA occlusion and underwent FLAIR sequence before endovascular therapy	With distal F-HAs vs. without	38	29	9	ASITN/SIR collateral grade	90-day functional outcome, 7-day NIHSS score
Nave et al 2018 Germany[15]	R cohort (P data collect)	AIS patients with acute M1-MCA occlusion who received an MRI before endovascular therapy	F-HAs-ASPECTS >4 vs. ≤4	116	75	41	ASITN/SIR collateral grade	90-day and discharge functional outcome, discharge NIHSS score, recanalisation
Jiang et al 2019a China[16]	R cohort (P data collect)	AIS patients with anterior circulation infarct within 4.5 h after onset and had pretreatment MRI	F-HAs-ASPECTS ≥4 vs. <4	37	19	18	NIHSS score, ASITN/SIR collateral grade	Discharge NIHSS score, recanalisation
Jiang et al 2019b China[17]	R cohort (P data collect)	AIS patients with anterior circulation infarct within 6 h after onset and had pretreatment MRI	With F-HAs beyond the boundaries of DWI lesion vs. without	59	39	20	Age, infarct volume, ASITN/SIR collateral grade	Recanalisation
Legrand et al 2019 France[18]	R cohort (P data collect)	AIS patients (18 to 80 years old, NIHSS scores 10 to 25) due to occlusion of M1-MCA.	With F-HAs beyond the boundaries of DWI lesion vs. without	100	79	21	Age, sex, current smoking, infarct volume	90-day functional outcome, recanalisation, ENI
Zhou et al 2019 China[19]	R cohort (P data collect)	AIS patients with anterior circulation infarct within 6 h after onset and had pretreatment MRI	With F-HAs beyond the boundaries of DWI lesion vs. without	38	23	15	Infarct volume, ASITN/SIR collateral grade	24 h infarct growth
Conservative treatment								
Girot et al 2007a, 2007b France[20]	R cohort (P data collect)	Consecutive AIS patients admitted within 12 h after onset of hemispheric cerebral ischaemia.	With distal F-HAs vs. without; with proximal F-HAs vs. without	30	16	14	Not reported	1-month functional outcome
				30	9	21		
Huang et al 2012 China[21]	R cohort (P data collect)	MCA infarct patients with admission within 24 h after onset. MRA/DSA show M1- or M2-MCA occlusion	With distal F-HAs (M3-6) vs. without	74	42	32	Age, hypertension, NIHSS score, infarct volume	10- and 90-day functional outcome, 10-day NIHSS score
Song et al 2016 China[22]	R cohort (P data collect)	First-ever AIS patients with MRA showing complete occlusion in M1-MCA	With prominent distal F-HAs vs. subtle vs. none	88	65†	23†	Not reported	90-day functional outcome
Nam et al 2017 South Korea[23]	R cohort (P data collect)	First-ever ischaemic stroke patients within 24 h after onset. With symptomatic occlusion or severe stenosis in ICA, M1- or M2-MCA	With prominent distal F-HAs vs. subtle vs. none	325	192†	133†	NIHSS score, infarct volume, proximal arterial occlusion	90-day functional outcome, in hospital mortality, END, 72 h and 7-day NIHSS score, ICH, infarct growth
Li et al 2018 China[24]	R cohort (P data collect)	AIS patients with M1-MCA occlusion. MRI/CTP/CTA were obtained within 72 h of stroke onset	With distal F-HAs vs. without	38	22	16	Not reported	90-day functional outcome

Dong et al 2019 China[25]	R cohort (P data collect)	AIS patients with anterior circulation infarct and MRI within 48 h of stroke onset	With F-HAs vs. without	160	83	77	Hypertension, intracranial large artery disease, infarct volume, NIHSS score, anticoagulant use	90-day functional outcome, discharge NIHSS score
Kim et al 2019b South Korea[11]	R cohort (P data collect)	AIS patients with initial NIHSS scores ≤5 and MCA occlusion on MRA within 24 h of stroke onset	With prominent distal F-HAs (M3-6) vs. without	74	54	20	No	90-day functional outcome, END
Song et al 2019 China[26]	R cohort	Ischaemic stroke patients with M1-MCA stenosis confirmed on MRI	With F-HAs beyond the boundaries of DWI lesion vs. without	109	66	43	Age, triglycerides, AF, infarct volume, time interval from stroke onset to MRI	90-day functional outcome, discharge NIHSS score
Yuan et al 2019 China[27]	R cohort (P data collect)	AIS patients with ICA occlusion and MCA territory infarct	With F-HAs beyond the boundaries of DWI lesion (≥3 sections) vs. without	68	41	27	Not reported	90-day functional outcome
Mixed treatment								
Hohenhaus et al 2012 Germany[28]	R cohort (P data collect)	AIS patients with ischaemic lesion on DWI and vessel occlusion or symptomatic stenosis of ICA or MCA	F-HAs-ASPECTS >4 vs. ≤4	84, 48§	41, 25§	43, 23§	Age, prior stroke NIHSS score, infarct volume, hypoperfusion volume, PWI-DWI mismatch	5- to 7-day NIHSS score, 5- to 7-day infarct growth, 2-day recanalisation, ICH
Pérez de la Ossa et al 2012 Spain[29]	R cohort (P data collect)	AIS patients with a large artery occlusion of the anterior circulation in which CT and MRI were performed	With prominent distal F-HAs vs. subtle vs. none	70	57†	13†	Sex, BP	Infarct growth
Adatia et al* 2013a, 2013b Canada[30,31]	R cohort (P data collect)	AIS patients with M1-MCA or ICA occlusion and had pretreatment 24 h MRI	With F-HAs in the region of DWI lesion vs. without	95	59	36	No	90-day functional outcome, recanalisation, ICH
Aoki et al* 2016 Japan[32]	R cohort (P data collect)	AIS patients within 24 h of onset and with major arterial occlusion on MRA	With F-HAs vs. without	72	62	10	NIHSS score	Recanalisation during hospital
Kim et al 2016 South Korea[33]	R cohort (P data collect)	Ischaemic stroke patients who visited the hospital within 7 days of onset and had DWI lesion on MCA territory	With F-HAs vs. without	87	30	57	NIHSS score, proximal arterial occlusion	90-day functional outcome, END, 7-day NIHSS score
Vural et al 2016 Turkey[34]	R cohort (P data collect)	Ischaemic stroke patients with proximal MCA occlusion who underwent SWI and FLAIR within 24 h of onset	F-HAs score >4 vs. ≤4	50	31	19	Not reported	Discharge functional outcome,
Mahdjoub et al 2018 France[35]	R cohort (P data collect)	Acute MCA infarct patients with pretreatment and 24 h follow-up MRI	F-HAs-ASPECTS ≥4 vs. <4	244	162	82	Onset time to MRI, proximal arterial occlusion,hypoperfusion volume, PWI-DWI mismatch	90-day functional outcome, ENI, 24h recanalisation, infarct growth

AF denotes atrial fibrillation; **AIS**, acute ischaemic stroke; **ASPECT**, Alberta Stroke Program Early Computed Tomography score; **ASITN/SIR**, American Society of Intervention and Therapeutic Neuroradiology/Society of Interventional Radiology; **BP**, blood pressure; **C,** control group; **CT**, computed tomography; **CTP**, CT perfusion; **DWI**, diffusion-weighted imaging; **E**, exposure group; **ENCHANTED**, Enhanced Control of Hypertension and Thrombolysis Stroke Study; **END**, early neurological deterioration; **ENI**, early neurological improvement; **FLAIR**, fluid-attenuated inversion recovery; **FLAIR (F)-HAs**, FLAIR hyperintense arteries; **hr**, hour; **ICA,** internal carotid artery; **ICH**, intracerebral haemorrhage; **MCA**, middle cerebral artery; **M1/2/3-6-MCA**, segment 1/2/3-6 of MCA; **MT**, mechanical thrombectomy; **MRA**, magnetic resonance angiography; **MRI,** magnetic resonance imaging; **NIHSS**, National Institutes of Health stroke scale; **PWI**, perfusion-

weighted imaging; **P,** prospectively; **R,** retrospective; **rtPA**, recombinant human tissue plasminogen activator; **SE/GRE**, spin echo/gradient recalled echo sequence; **sICH,** symptomatic ICH.
*Conference abstract.
†Number of patients with prominent or subtle distal FLAIR-HAs vs. no FLAIR-HAs.
‡Number of patients with FLAIR-HAs scores >4 versus FLAIR-HAs scores ≤4.
§Number of follow-up patients.

Appendix 3.3. Quality assessment of included studies

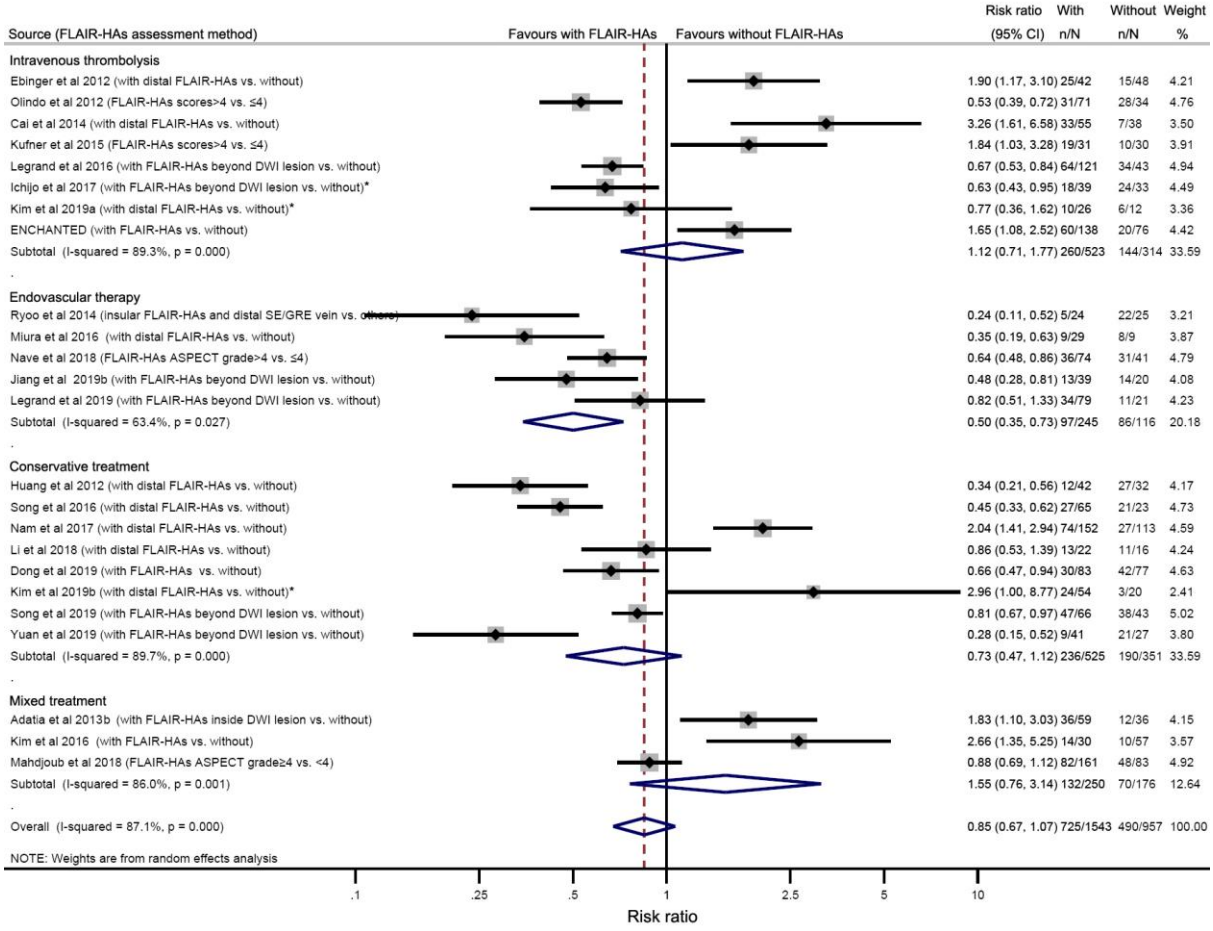
	Selection (4)				Comparability (2)		Outcome (3)		
Study	Representative-ness of having F-HAs cohort	Selection of not having F-HAs cohort	Ascertain-ment of F-HAs	Outcomes were not present at start of study	Control for proximal artery occlusion	Control for other confounders	Assess-ment of outcomes	Was follow-up long enough for outcomes to occur	Adequacy of follow up of cohorts
Intravenous thrombolysis									
Lee et al 2009[1]	*	*	*	*	-	*	*	-	-
Ebinger et al 2012[2]	*	*	*	*	-	-	*	*	*
Olindo et al 2012[3]	-	-	*	*	*	*	*	*	*
Cai et al 2014[4]	*	*	*	*	*	*	*	*	*
Ichijo et al 2015[5]	-	-	*	*	*	*	*	*	*
Kufner et al 2015[6]	-	-	*	*	*	*	*	*	*
Legrand et al 2015[7]	*	*	*	*	*	-	*	*	*
Legrand et al 2016[8]	*	*	*	*	*	-	*	*	*
Ichijo et al 2017[9]	*	*	*	*	*	*	NR	-	-
El Nawar et al 2019[10]	*	*	*	*	-	*	*	*	-
Kim et al 2019a[11]	-	-	*	*	*	*	*	*	*
ENCHANTED	*	*	*	*	*	*	*	*	*
Endovascular therapy									
Ryoo et al 2014[12]	-	-	*	*	-	-	*	*	*
Liu et al 2016a[13], 2016b[13]	-	-	*	*	*	*	*	-	-
Miura et al 2016[14]	*	*	-	*	-	*	*	*	*
Nave et al 2018[15]	-	-	*	*	*	*	*	*	*
Jiang et al 2019a[16]	-	-	*	*	NR	*	*	*	*
Jiang et al 2019b[17]	-	-	*	*	*	*	*	*	*
Legrand et al 2019[18]	-	-	*	*	*	*	*	*	*
Zhou et al 2019[19]	-	-	*	*	NR	-	*	*	*
Conservative treatment									
Girot et al 2007a[20], 2007b[20]	*	*	*	*	-	-	*	-	-
Huang et al 2012[21]	*	*	*	*	*	*	*	*	*
Song et al 2016[22]	*	*	*	*	*	*	*	*	*
Nam et al 2017[23]	*	*	*	*	-	*	*	*	*
Li et al 2018[24]	*	*	*	*	*	*	*	*	*
Dong et al 2019[25]	*	*	*	*	*	*	*	*	*
Kim et al 2019b[11]	-	-	*	*	*	*	*	*	*
Song et al 2019[26]	-	-	*	*	*	-	*	*	*
Yuan et al 2019[27]	-	-	*	*	*	*	*	*	*
Mixed treatment									
Hohenhaus et al 2012[28]	-	-	*	*	-	-	*	-	-
Pérez de la Ossa et al 2012[29]	*	*	*	*	*	*	-	-	-
Adatia et al 2013a[30], 2013b[31]	*	*	*	*	*	*	*	*	*
Aoki et al 2016[32]	*	*	-	*	*	*	-	-	-
Kim et al 2016[33]	*	*	*	*	*	*	*	*	*

Vural et al 2016[34]	-	-	*	*	*	*	*	-	-
Mahdjoub et al 2018[35]	-	-	*	*	-	-	*	*	*

ENCHANTED denotes Enhanced Control of Hypertension and Thrombolysis Stroke Study; **FLAIR**, fluid-attenuated inversion recovery; **F-HAs**, FLAIR hyperintense arteries.

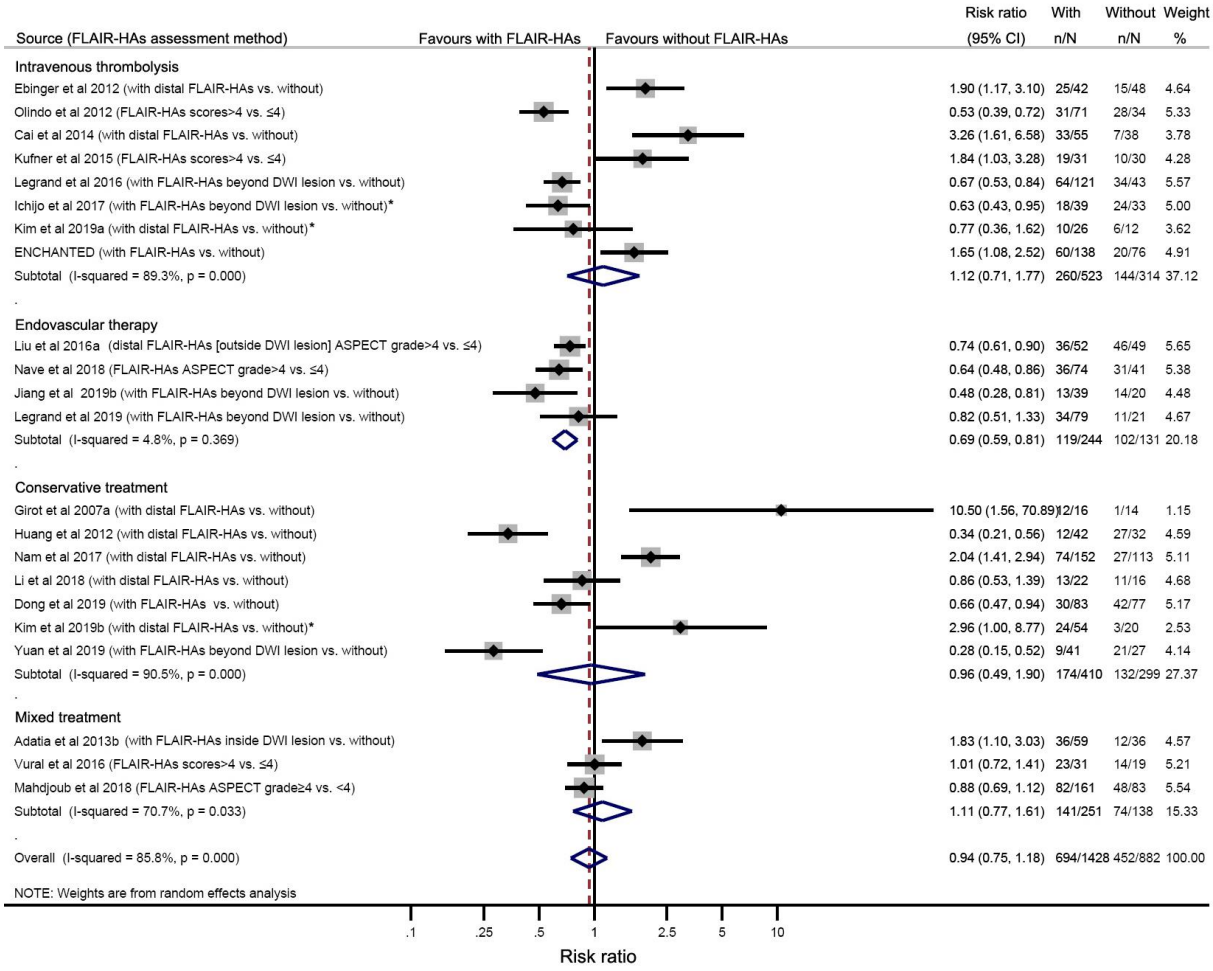
*indicates low risk of bias; - indicates high risk of bias; NR; not report.

Appendix 3.4. Meta-analysis of associations between FLAIR-HAs and 90-day unfavourable functional outcome, by type of treatment administered to the study patients



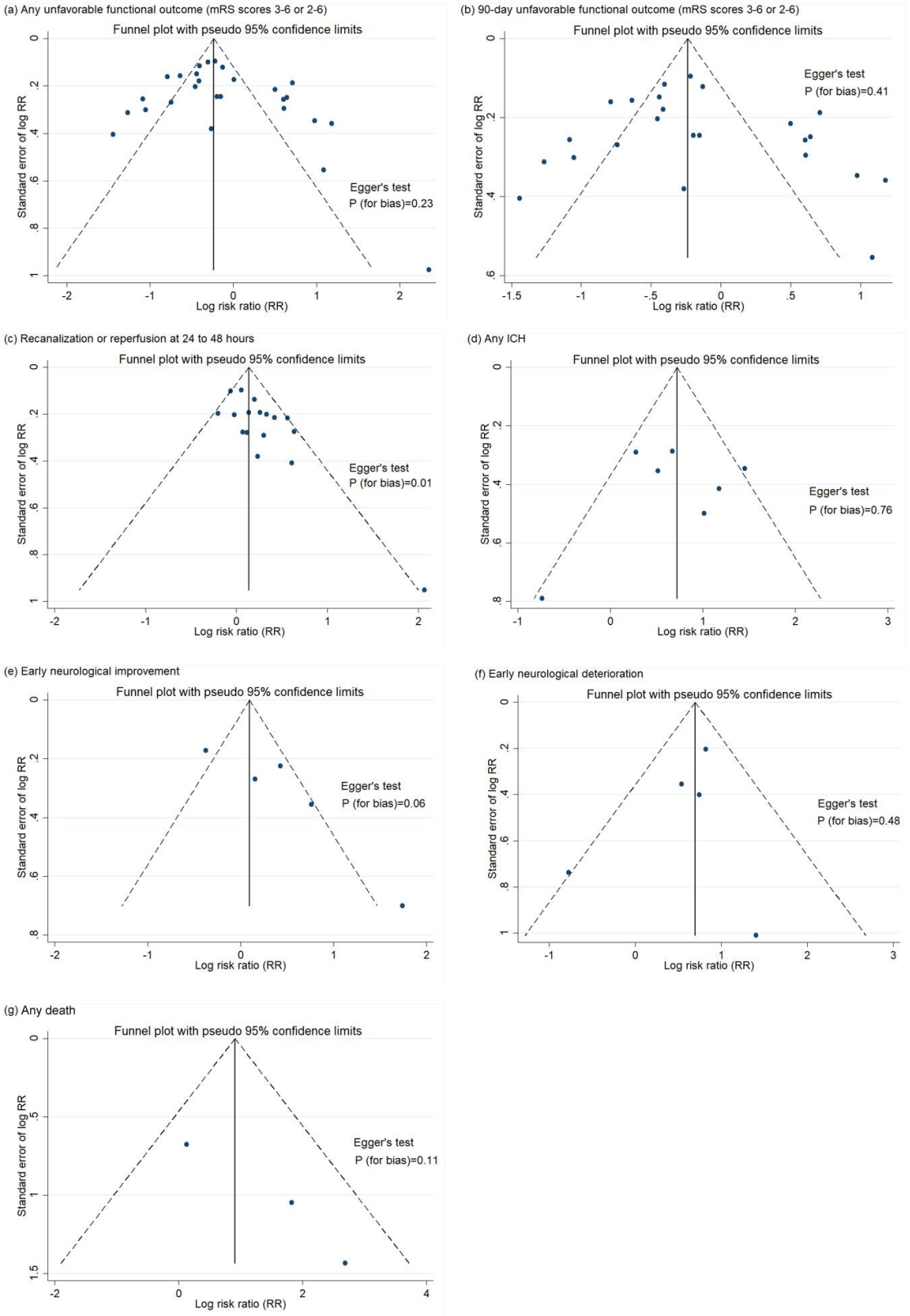
ASPECTS denotes Alberta Stroke Program Early Computed Tomography score; **ENCHANTED**, Enhanced Control of Hypertension and Thrombolysis Stroke Study; **DWI**, diffusion weighted imaging; **FLAIR**, fluid-attenuated inversion recovery; **FLAIR-HAs**, FLAIR hyperintense arteries; **mRS**, modified Rankin scale; **SE/GRE**, spin echo/gradient recalled echo sequence.
*mRS scores 2-6 was regarded as unfavourable functional outcome (mRS scores 3-6 for other studies)

Appendix 3.5. Sensitivity analysis for associations between FLAIR-HAs and unfavourable functional outcome in studies with data collecting prospectively and recruiting acute ischaemic stroke only



ASPECTS denotes Alberta Stroke Program Early Computed Tomography score; **ENCHANTED**, Enhanced Control of Hypertension and Thrombolysis Stroke Study; **DWI**, diffusion weighted imaging; **FLAIR**, fluid-attenuated inversion recovery; **FLAIR-HAs**, FLAIR hyperintense arteries; **mRS**, modified Rankin scale. *mRS scores 2-6 was regarded as unfavourable functional outcome (mRS scores 3-6 for other studies)

Appendix 3.6. Assessment of publication bias



ICH denotes intracerebral haemorrhage; **mRS** denotes modified Rankin Scale.
Funnel plot and Egger's regression test for (a) any unfavourable functional outcome (primary), (b) 90-day unfavourable functional outcome, (c) recanalisation or reperfusion at 24 to 48 hours, (d) any ICH, (e) early neurological improvement, (f) early neurological deterioration, and (g) any death.

Appendix 3.7. NIHSS score (change) by the presence of FLAIR-HAs

Study	Analysis method of FLAIR-HAs (number of patients)	Time of NIHSS score assessment at FU	NIHSS score in patients with FLAIR-HAs		NIHSS score in patients without FLAIR-HAs	
NIHSS score on admission						
Intravenous thrombolysis						
Lee et al 2009[1]	with prominent distal FLAIR-HAs (n=23) vs. without (n=6)	-	9.4 (mean)	6.2 (sd)	18.3 (mean)	7.5 (sd)
Ebinger et al 2012[2]	with distal FLAIR-HAs (n=42) vs. without (n=48)	-	10 (median)	5-17 (iqr)	5 (median)	4-8 (iqr)
Olindo et al 2012[3]	FLAIR-HAs score>4 (n=71) vs. ≤4 (n=34)	-	15.0 (mean)	2-27 (range)	17.1 (mean)	4.3 (sd)
Cai et al 2014[4]	with distal FLAIR-HAs (n=55) vs. without (n=38)	-	11.8 (mean)	6.0 (sd)	7.2 (mean)	4.5 (sd)
Kufner et al 2015[6]	FLAIR-HAs score >4 (n=32) vs. ≤4 (n=30)	-	14.5 (median)	11-18 (iqr)	5 (median)	4-13 (iqr)
Legrand et al 2015[7]	with FLAIR-HAs beyond DWI lesion (n=102) vs. without (n=39)	-	16 (median)	12-20 (iqr)	17 (median)	15-22 (iqr)
Kim et al 2019a[11]	with prominent distal FLAIR-HAs (n=26) vs. without (n=12)	-	4 (median)	2-5 (iqr)	3.5(median)	2-4.5 (iqr)
ENCHANTED	with FLAIR-HAs (n=144) vs. without (n=76)	-	11 (median)	5.5-15.0 (iqr)	5 (median)	3.5-8 (iqr)
	with distal FLAIR-HAs (n=140) vs. without (n=80)	-	11 (median)	5-15 (iqr)	6 (median)	4-8 (iqr)
	with proximal FLAIR-HAs (n=69) vs. without (n=151)	-	13 (median)	9-16 (iqr)	6 (median)	4-10 (iqr)
	with FLAIR-HAs beyond DWI lesion (n=94) vs. without (n=126)	-	9 (median)	4-13 (iqr)	7 (median)	4-13 (iqr)
	with FLAIR-HAs within DWI lesion (n=66) vs. without (n=154)	-	13 (median)	9-17 (iqr)	6 (median)	4-10 (iqr)
	FLAIR-HAs score >4 (n=100) vs. ≤4 (n=120)	-	12 (median)	6.5-16 (iqr)	6 (median)	4-9 (iqr)
	FLAIR-HAs ASPECTS grade >4 (n=50) vs. ≤4 (n=170)	-	13 (median)	7-16 (iqr)	7 (median)	4-11 (iqr)
Endovascular treatment						
Liu et al 2016a[13]	distal FLAIR-HAs (outside DWI) ASPECTS grade >4 (n=52) vs. ≤4 (n=49)	-	14.5 (median)	11-19.8 (iqr)	19 (median)	13-23 (iqr)
Liu et al 2016b[13]	distal FLAIR-HAs (within DWI) ASPECTS grade >2 (n=37) vs. ≤2 (n=64)	-	20 (median)	15.5-23.0 (iqr)	13.5 (median)	10-19.8 (iqr)
Nave et al 2018[15]	FLAIR-HAs ASPECT grade ≥5 (n=75) vs. <5 (n=41)	-	15 (median)	9-18 (iqr)	14 (median)	12-19 (iqr)
Jiang et al 2019a[16]	FLAIR-HAs ASPECTS grade ≥4 (n=19) vs. <4 (n=18)	-	12.1 (mean)	4.3 (sd)	16 (mean)	5.5 (sd)
Jiang et al 2019b[17]	with FLAIR-HAs beyond DWI lesion (n=39) vs. without (n=20)	-	12.9 (mean)	5.0 (sd)	14.2 (mean)	6.3 (sd)
Legrand et al 2019[18]	with FLAIR-HAs beyond DWI lesion (n=79) vs. without (n=21)	-	18 (median)	14-20 (iqr)	19 (median)	17-21 (iqr)
Zhou et al 2019[19]	with FLAIR-HAs beyond DWI lesion (n=23) vs. without (n=15)	-	12.4 (mean)	4.9 (sd)	15.6 (mean)	6.4 (sd)
Conservative treatment						
Huang et al 2012[21]	with distal FLAIR-HAs (n=42) vs. without (n=32)	-	11 (median)	1-22 (range)	15 (median)	6-25 (range)
Nam et al 2017[23]	with prominent distal FLAIR-HAs (n=91) vs. without (n=133)	-	10 (median)	5-18 (iqr)	3 (median)	1-8 (iqr)
Dong et al 2019[25]	with FLAIR-HAs (n=83) vs. without (n=77)	-	7.2 (mean)	4.0 (sd)	5.4 (mean)	4.5 (sd)
Kim et al 2019b[11]	with prominent distal FLAIR-HAs (n=54) vs. without (n=20)	-	2.5 (median)	2-4 (iqr)	2.5(median)	1.5-4 (iqr)
Song et al 2019[26]	with FLAIR-HAs beyond DWI lesion (n=66) vs. without (n=43)	-	18 (median)	16-22 (iqr)	17 (median)	16-22 (iqr)
Mixed treatment						
Hohenhaus et al 2012[28]	FLAIR-HAs ASPECTS grade >4 (n=25) vs. ≤4 (n=23)	-	13 (median)	5-15 (iqr)	4 (median)	1-11 (iqr)
Pérez de la Ossa et al 2012[29]	with prominent distal FLAIR-HAs (n=24) vs. without (n=13)	-	17 (median)	8-21 (range)	18 (median)	13-22 (range)
Aoki et al 2016[32]	with FLAIR-HAs (n=62) vs. without (n=10)	-	10 (median)	4-21 (iqr)	4 (median)	2-8 (iqr)
Kim et al 2016[33]	with FLAIR-HAs (n=30) vs. without (n=57)	-	4 (median)	0-15 (range)	2 (median)	0-11 (range)
Mahdjoub et al 2018[35]	FLAIR-HAs ASPECT grade 6-7 (n=49) vs. 0-1 (n=19)	-	15 (median)	11-21 (iqr)	13 (median)	8-20 (iqr)
NIHSS score at FU						

Intravenous thrombolysis						
Lee et al 2009[1]	with prominent distal FLAIR-HAs (n=23) vs. without (n=6)	5 day	5.2 (mean)	5.8 (sd)	15.3 (mean)	14.4 (sd)
Kufner et al 2015[6]	FLAIR-HAs score >4 (n=32) vs. ≤4 (n=30)	2 day	5 (median)	2-17 (iqr)	3 (median)	1-5 (iqr)
Legrand et al 2015[7]	with FLAIR-HAs beyond DWI lesion (n=102) vs. without (n=39)	24 hours	11 (median)	5-18 (iqr)	16 (median)	10-22 (iqr)
ENCHANTED	with FLAIR-HAs (n=138) vs. without (n=76)	24 hours	6 (median)	3-14 (iqr)	3 (median)	2-6 (iqr)
	with distal FLAIR-HAs (n=134) vs. without (n=80)	24 hours	6 (median)	3-14 (iqr)	3 (median)	2-6 (iqr)
	with proximal FLAIR-HAs (n=67) vs. without (n=147)	24 hours	9 (median)	3-17 (iqr)	4 (median)	2-7 (iqr)
	with FLAIR-HAs beyond DWI lesion (n=92) vs. without (n=122)	24 hours	5 (median)	2-9 (iqr)	5 (median)	2-11 (iqr)
	with FLAIR-HAs within DWI lesion (n=62) vs. without (n=152)	24 hours	10 (median)	6-17 (iqr)	3 (median)	2-6 (iqr)
	FLAIR-HAs score >4 (n=96) vs. ≤4 (n=118)	24 hours	7 (median)	3-16 (iqr)	3 (median)	2-6 (iqr)
	FLAIR-HAs ASPECTS grade >4 (n=46) vs. ≤4 (n=168)	24 hours	7 (median)	2-14 (iqr)	4 (median)	2-8 (iqr)
Endovascular treatment						
Miura et al 2016[14]	with distal FLAIR-HAs (n=29) vs. without (n=9)	7 day	7.8 (median)	-	16.3(median)	-
Nave et al 2018[15]	FLAIR-HAs ASPECT grade ≥5 (n=75) vs. <5 (n=41)	at discharge	4 (median)	1-9 (iqr)	8 (median)	3-15 (iqr)
Jiang et al 2019a[16]	FLAIR-HAs ASPECT grade ≥4 (n=19) vs. <4 (n=18)	at discharge	4.1 (mean)	4.9 (sd)	12.7 (mean)	8.2 (sd)
Legrand et al 2019[18]	with FLAIR-HAs beyond DWI lesion (n=79) vs. without (n=21)	24 hours	8 (median)	4-16 (iqr)	14 (median)	6-24 (iqr)
Conservative treatment						
Huang et al 2012[21]	with distal FLAIR-HAs (n=42) vs. without (n=32)	10 day	7 (median)	0-22 (range)	14 (median)	4-25 (range)
Nam et al 2017[23]	with prominent distal FLAIR-HAs (n=91) vs. without (n=133)	72 hours	12 (median)	5-18 (iqr)	2 (median)	0-8 (iqr)
	with prominent distal FLAIR-HAs (n=91) vs. without (n=133)	7 day	9 (median)	4-16 (iqr)	2 (median)	0-6 (iqr)
Dong et al 2019[25]	with FLAIR-HAs (n=83) vs. without (n=77)	at discharge	6.2 (mean)	3.9 (sd)	5.3 (mean)	3.9 (sd)
Song et al 2019[26]	with FLAIR-HAs beyond DWI lesion (n=66) vs. without (n=43)	at discharge	16 (median)	15-19 (iqr)	16 (median)	13-21 (iqr)
Mixed treatment						
Hohenhaus et al 2012[28]	FLAIR-HAs ASPECTS grade >4 (n=25) vs. ≤4 (n=23)	5-7 day	9 (median)	2-13 (iqr)	2 (median)	0-11 (iqr)
Kim et al 2016[33]	with FLAIR-HAs (n=30) vs. without (n=57)	7 day	3.5 (median)	0-17 (range)	1 (median)	0-12 (range)
Median NIHSS score change (NIHSS score at FU minus that on admission)						
Ebinger et al 2012[2]	with distal FLAIR-HAs (n=42) vs. without (n=48)	not specify	-3 (median)	-4-0 (iqr)	-2 (median)	-5-0 (iqr)
ENCHANTED	with FLAIR-HAs (n=138) vs. without (n=76)	24 hours	-2 (median)	-4-0 (iqr)	-1 (median)	-3-0 (iqr)
	with distal FLAIR-HAs (n=134) vs. without (n=80)	24 hours	-2 (median)	-4-0 (iqr)	-1 (median)	-3-0 (iqr)
	with proximal FLAIR-HAs (n=67) vs. without (n=147)	24 hours	-2 (median)	-5-0 (iqr)	-1 (median)	-4-0 (iqr)
	with FLAIR-HAs beyond DWI lesion (n=92) vs. without (n=122)	24 hours	-2 (median)	-4.5-0 (iqr)	-1 (median)	-4-0 (iqr)
	with FLAIR-HAs within DWI lesion (n=62) vs. without (n=152)	24 hours	-2 (median)	-4-0 (iqr)	-1 (median)	-4-0 (iqr)
	FLAIR-HAs score >4 (n=96) vs. ≤4 (n=118)	24 hours	-1.5 (median)	-5-0 (iqr)	-1 (median)	-3-0 (iqr)
	FLAIR-HAs ASPECTS grade >4 (n=46) vs. ≤4 (n=168)	24 hours	-2 (median)	-6-0 (iqr)	-1 (median)	-4-0 (iqr)
Mean NIHSS score change (NIHSS score at FU minus that on admission) in ENCHANTED						
ENCHANTED	with FLAIR-HAs (n=138) vs. without (n=76)	24 hours	-1.9 (mean)	5.6 (sd)	-1.7 (mean)	3.7 (sd)
	with distal FLAIR-HAs (n=134) vs. without (n=80)	24 hours	-2.0 (mean)	5.7 (sd)	-1.7 (mean)	3.6 (sd)
	with proximal FLAIR-HAs (n=67) vs. without (n=147)	24 hours	-1.7 (mean)	6.9 (sd)	-1.9 (mean)	3.9 (sd)
	with FLAIR-HAs beyond DWI lesion (n=92) vs. without (n=122)	24 hours	-2.3 (mean)	5.3 (sd)	-1.5 (mean)	4.8 (sd)
	with FLAIR-HAs within DWI lesion (n=62) vs. without (n=152)	24 hours	-1.5 (mean)	5.4 (sd)	-2.0 (mean)	4.9 (sd)
	FLAIR-HAs score >4 (n=96) vs. ≤4 (n=118)	24 hours	-2.0 (mean)	6.5 (sd)	-1.8 (mean)	3.5 (sd)

FLAIR-HAs ASPECTS grade >4 (n=46) vs. ≤4 (n=168)	24 hours	-2.4 (mean)	6.0 (sd)	-1.7 (mean)	4.7 (sd)

ASPECTS denotes Alberta Stroke Program Early Computed Tomography score; **DWI**, diffusion weighted imaging; **ENCHANTED**, Enhanced Control of Hypertension and Thrombolysis Stroke Study; **FLAIR**, fluid-attenuated inversion recovery; **FLAIR-HAs**, FLAIR hyperintense arteries; **FU**, follow-up; **iqr**, interquartile range; **NIHSS**, National Institutes of Health Stroke Scale; **sd**, standard deviation.

Appendix 3.8. Infarct volume and growth by the presence of FLAIR-HAs

Study	Analysis method of FLAIR-HAs (no. of patients)	Analysis method of infarct volume/growth	Infarct volume in patients with FLAIR-HAs		Infarct volume in patients without FLAIR-HAs	
Infarct volume on admission						
Intravenous thrombolysis						
Lee et al 2009[1]	with prominent distal FLAIR-HAs (n=23) vs. without (n=6)	DWI lesion volume (mL)	22.1 (mean)	21.8 (sd)	127.5 (mean)	77.0 (sd)
Ebinger et al 2012[2]	with distal FLAIR-HAs (n=42) vs. without (n=48)	DWI lesion volume (mL)	5.0 (median)	1.0-18.9 (iqr)	0.5 (median)	0.1-1.7 (iqr)
Cai et al 2014[4]	with distal FLAIR-HAs (n=55) vs. without (n=38)	DWI lesion volume (mL)	5.5 (median)	31.0 (iqr range)	2.0 (median)	3.0 (iqr range)
Kufner et al 2015[6]	FLAIR-HAs score >4 (n=32) vs. ≤4 (n=30)	DWI lesion volume (mL)	8.1 (median)	2.4-20.6 (iqr)	1.6 (median)	0.3-4.9 (iqr)
Legrand et al 2015[7]	with FLAIR-HAs beyond DWI lesion (n=102) vs. without (n=39)	DWI lesion volume (mL)	18 (median)	8-35 (iqr)	108 (median)	57-160 (iqr)
Kim et al 2019a[11]	with prominent distal FLAIR-HAs (n=26) vs. without (n=12)	DWI lesion volume (mL)	7.0 (median)	1.3-15.5 (iqr)	4.3 (median)	1.8-12.5 (iqr)
ENCHANTED	with FLAIR-HAs (n=144) vs. without (n=76)	DWI lesion volume (cm^3)	11.2 (median)	3.9-50.9 (iqr)	1.1 (median)	0.6-2.8 (iqr)
	with distal FLAIR-HAs (n=140) vs. without (n=80)	DWI lesion volume (cm^3)	11.2 (median)	3.9-50.9 (iqr)	1.1 (median)	0.6-3.0 (iqr)
	with proximal FLAIR-HAs (n=69) vs. without (n=151)	DWI lesion volume (cm^3)	16.5 (median)	4.4-60.2 (iqr)	2.7 (median)	0.8-10.5 (iqr)
	with FLAIR-HAs beyond DWI lesion (n=94) vs. without (n=126)	DWI lesion volume (cm^3)	6.9 (median)	2.9-15.0 (iqr)	2.8 (median)	0.8-41.3 (iqr)
	with FLAIR-HAs within DWI lesion (n=66) vs. without (n=154)	DWI lesion volume (cm^3)	51.3 (median)	14.6-107.7 (iqr)	2.3 (median)	0.8-6.3 (iqr)
	FLAIR-HAs score >4 (n=100) vs. ≤4 (n=120)	DWI lesion volume (cm^3)	12.8 (median)	4.2-51.4 (iqr)	1.9 (median)	0.7-7.1 (iqr)
	FLAIR-HAs ASPECTS grade >4 (n=50) vs. ≤4 (n=170)	DWI lesion volume (cm^3)	13.1 (median)	2.7-54.6 (iqr)	3.6 (median)	1.0-12.8 (iqr)
Endovascular treatment						
Liu et al 2016a[13]	distal FLAIR-HAs (outside DWI) ASPECTS grade >4 (n=52) vs. ≤4 (n=49)	DWI lesion volume (mL)	13.7 (median)	6.3-33.7 (iqr)	46.5 (median)	14.6-89.6 (iqr)
Liu et al 2016b[13]	distal FLAIR-HAs (within DWI) ASPECTS grade >2 (n=37) vs. ≤2 (n=64)	DWI lesion volume (mL)	65.0 (median)	30.5-97.5 (iqr)	13.0 (median)	5.9-23.7 (iqr)
Jiang et al 2019a[16]	FLAIR-HAs ASPECTS grade ≥4 (n=19) vs. <4 (n=18)	DWI lesion volume (mL)	17.6 (mean)	21.3 (sd)	38.0 (mean)	43.4 (sd)
Jiang et al 2019b[17]	with FLAIR-HAs beyond DWI lesion (n=39) vs. without (n=20)	DWI lesion volume (mL)	15.8 (mean)	21.3 (sd)	48.7 (mean)	47.9 (sd)
Legrand et al 2019[18]	with FLAIR-HAs beyond DWI lesion (n=79) vs. without (n=21)	DWI lesion volume (mL)	16 (median)	9-34 (iqr)	75 (median)	38-122 (iqr)
Zhou et al 2019[19]	with FLAIR-HAs beyond DWI lesion (n=23) vs. without (n=15)	DWI lesion volume (mL)	15.1 (mean)	23.0 (sd)	56.9 (mean)	51.0 (sd)
Conservative treatment						
Nam et al 2017[23]	with prominent distal FLAIR-HAs (n=91) vs. without (n=133)	DWI lesion volume (mL)	17.2 (median)	3.8-55.1 (iqr)	2.2 (median)	0.3-16.3 (iqr)
Dong et al 2019[25]	with FLAIR-HAs (n=83) vs. without (n=77)	Cortical DWI lesion volume (mL)	13.9 (mean)	25.6 (sd)	6.6 (mean)	13.5 (sd)
		Deep DWI lesion volume (mL)	1.2 (mean)	3.1 (sd)	0.8 (mean)	2.0 (sd)
Kim et al 2019b[11]	with prominent distal FLAIR-HAs (n=54) vs. without (n=20)	DWI lesion volume (mL)	7.1 (median)	1.5-25.7 (iqr)	3.4 (median)	0.2-10.3 (iqr)
Mixed treatment						
Hohenhaus et al 2012[28]	FLAIR-HAs ASPECTS grade >4 (n=25) vs. ≤4 (n=23)	DWI lesion volume (cm^3)	25.2 (median)	6.4-42.7 (iqr)	3.4 (median)	0.7-22.5 (iqr)
Mahdjoub et al 2018[35]	FLAIR-HAs ASPECTS grade 6-7 (n=49) vs. 0-1 (n=19)	DWI lesion volume (mL)	12 (median)	6-34 (iqr)	24 (median)	12-102 (iqr)
Infarct volume at FU						
Intravenous thrombolysis						
Lee et al 2009[1]	with prominent distal FLAIR-HAs (n=23) vs. without (n=6)	24 hours DWI lesion volume (mL)	33.0 (mean)	40.5 (sd)	144.8 (mean)	78.2 (sd)
Legrand et al 2015[7]	with FLAIR-HAs beyond DWI lesion (n=102) vs. without (n=39)	24 hours DWI lesion volume (mL)	36 (median)	19-66 (iqr)	161 (median)	95-230 (iqr)
ENCHANTED	with FLAIR-HAs (n=49) vs. without (n=33)	24-48 hours DWI lesion volume (mL)	35.6 (median)	8.1-105 (iqr)	2.0 (median)	0.9-4.3 (iqr)
	with distal FLAIR-HAs (n=49) vs. without (n=33)	24-48 hours DWI lesion volume (mL)	35.6 (median)	8.1-105 (iqr)	2.0 (median)	0.9-4.3 (iqr)
	with proximal FLAIR-HAs (n=24) vs. without (n=58)	24-48 hours DWI lesion volume (mL)	40.3 (median)	9.1-136.5 (iqr)	4.1 (median)	1.6-13.3 (iqr)
	with FLAIR-HAs beyond DWI lesion (n=32) vs. without (n=50)	24-48 hours DWI lesion volume (mL)	20.7 (median)	5.2-63.7 (iqr)	4.1 (median)	1.6-15.5 (iqr)

	with FLAIR-HAs within DWI lesion (n=21) vs. without (n=61)	24-48 hours DWI lesion volume (mL)	104.5(median)	41.2-142.8 (iqr)	4.0 (median)	1.6-10.8 (iqr)
	FLAIR-HAs score >4 (n=35) vs. ≤4 (n=47)	24-48 hours DWI lesion volume (mL)	41.2 (median)	10.2-128.7 (iqr)	3.2 (median)	1.3-8.6 (iqr)
	FLAIR-HAs ASPECTS grade >4 (n=14) vs. ≤4 (n=68)	24-48 hours DWI lesion volume (mL)	38.4 (median)	3.3-104.5 (iqr)	6.7 (median)	1.9-32.3 (iqr)
Endovascular treatment						
Zhou et al 2019[19]	with FLAIR-HAs beyond DWI lesion (n=23) vs. without (n=15)	24 hours DWI lesion volume (mL)	32.2 (mean)	39.4 (sd)	101.4 (mean)	86.4 (sd)
Conservative treatment						
Nam et al 2017[23]	with prominent distal FLAIR-HAs (n=91) vs. without (n=133)	2.5-5 days DWI lesion volume (mL)	37.4 (median)	8.5-99.5 (iqr)	2.5 (median)	0.3-14.4 (iqr)
Mixed treatment						
Hohenhaus et al 2012[28]	FLAIR-HAs ASPECT grade >4 (n=25) vs. ≤4 (n=23)	5 days FLAIR lesion volume (cm^3)	71.7 (median)	27.0-162.5 (iqr)	11.4 (median)	1.5-62.1 (iqr)
Adatia et al 2013b[31]	with FLAIR-HAs in DWI lesion (n=59) vs. without (n=36)	24 hours DWI lesion volume (mL)	88.7 (mean)	-	37.9 (mean)	-
Mahdjoub et al 2018[35]	FLAIR-HAs ASPECTS grade 6-7 (n=49) vs. 0-1 (n=19)	24 hours DWI lesion volume (mL)	29 (median)	12-52 (iqr)	73 (median)	14-150 (iqr)
Absolute infarct growth (infarct volume on DWI at FU minus that on admission)						
Intravenous thrombolysis						
Ebinger et al 2012[2]	with distal FLAIR-HAs (n=42) vs. without (n=48)	FU – on admission (mL)	3.7 (median)	0.7-28.2 (iqr)	0.3 (median)	0-2.1 (iqr)
Kufner et al 2015[6]	FLAIR-HAs score>4 (n=32) vs. ≤4 (n=30)	24 hours FU – on admission (mL)	12.8 (median)	2.5-43.9 (iqr)	1.2 (median)	0.2-12.7 (iqr)
ENCHANTED	with FLAIR-HAs (n=49) vs. without (n=33)	24-48 hours FU – on admission (cm^3)	11.7 (median)	1.9-54.0 (iqr)	0.5 (median)	0.1-2.0 (iqr)
	with distal FLAIR-HAs (n=49) vs. without (n=33)	24-48 hours FU – on admission (cm^3)	11.7 (median)	1.9-54.0 (iqr)	0.5 (median)	0.1-2.0 (iqr)
	with proximal FLAIR-HAs (n=24) vs. without (n=58)	24-48 hours FU – on admission (cm^3)	26.0 (median)	3.7-66.4 (iqr)	1.4 (median)	0.2-6.8 (iqr)
	with FLAIR-HAs beyond DWI lesion (n=32) vs. without (n=50)	24-48 hours FU – on admission (cm^3)	6.9 (median)	1.5-35.8 (iqr)	1.4 (median)	0.2-9.4 (iqr)
	with FLAIR-HAs within DWI lesion (n=21) vs. without (n=61)	24-48 hours FU – on admission (cm^3)	37.0 (median)	21.0-71.8 (iqr)	1.7 (median)	0.2-6.1 (iqr)
	FLAIR-HAs score >4 (n=35) vs. ≤4 (n=47)	24-48 hours FU – on admission (cm^3)	21.0 (median)	3.7-59.5 (iqr)	0.8 (median)	0.1-3.5 (iqr)
	FLAIR-HAs ASPECTS grade >4 (n=14) vs. ≤4 (n=68)	24-48 hours FU – on admission (cm^3)	16.4 (median)	1.9-57.6 (iqr)	2.1 (median)	0.3-13.8 (iqr)
Endovascular treatment						
Zhou et al 2019[19]	with FLAIR-HAs beyond DWI lesion (n=23) vs. without (n=15)	24 hours FU – on admission (mL)	17.0 (mean)	23.4 (sd)	44.5 (mean)	41.7 (sd)
Conservative treatment						
Nam et al 2017[23]	with prominent distal FLAIR-HAs (n=91) vs. without (n=133)	2.5-5 days FU – on admission (mL)	10.7 (median)	2.5-35.7 (iqr)	0.8 (median)	0.0-4.7 (iqr)
Mixed treatment						
Mahdjoub et al 2018[35]	FLAIR-HAs ASPECTS grade 6-7 (n=49) vs. 0-1 (n=19)	24 hours FU – on admission (mL)	13 (median)	4-28 (iqr)	27 (median)	2-87 (iqr)
Relative infarct growth						
Intravenous thrombolysis						
Ebinger et al 2012[2]	with distal FLAIR-HAs (n=42) vs. without (n=48)	not specify	1.8 (median)	1.4-4.5 (iqr)	2.1 (median)	0.9-4.0 (iqr)
Legrand et al 2015[7]	with FLAIR-HAs beyond DWI lesion (n=102) vs. without (n=39)	24 hours FU ÷ on admission	2.0 (median)	1.1-3.6 (iqr)	1.3 (median)	1.1-1.7 (iqr)
ENCHANTED	with FLAIR-HAs (n=49) vs. without (n=33)	24 hours FU ÷ on admission	1.9 (median)	1.4-4.7 (iqr)	1.6 (median)	1.0-2.8 (iqr)
ENCHANTED	with distal FLAIR-HAs (n=49) vs. without (n=33)	24 hours FU ÷ on admission	1.9 (median)	1.4-4.7 (iqr)	1.6 (median)	1.0-2.8 (iqr)
	with proximal FLAIR-HAs (n=24) vs. without (n=58)	24 hours FU ÷ on admission	2.1 (median)	1.4-4.9 (iqr)	1.7 (median)	1.1-3.0 (iqr)
	with FLAIR-HAs beyond DWI lesion (n=32) vs. without (n=50)	24 hours FU ÷ on admission	1.9 (median)	1.4-4.9 (iqr)	1.7 (median)	1.1-3.0 (iqr)
	with FLAIR-HAs within DWI lesion (n=21) vs. without (n=61)	24 hours FU ÷ on admission	1.7 (median)	1.5-3.3 (iqr)	1.8 (median)	1.1-3.3 (iqr)
	FLAIR-HAs score >4 (n=35) vs. ≤4 (n=47)	24 hours FU ÷ on admission	3.2 (median)	1.5-6.6 (iqr)	1.5 (median)	1.0-2.4 (iqr)
	FLAIR-HAs ASPECTS grade >4 (n=14) vs. ≤4 (n=68)	24 hours FU ÷ on admission	2.1 (median)	1.4-5.4 (iqr)	1.7 (median)	1.1-3.2 (iqr)

ASPECTS denotes Alberta Stroke Program Early Computed Tomography score; **DWI**, diffusion weighted imaging; **ENCHANTED**, Enhanced Control of Hypertension and Thrombolysis Stroke Study; **FLAIR**, fluid-attenuated inversion recovery; **FLAIR-HAs**, FLAIR hyperintense arteries; **FU**, follow-up; **iqr**, interquartile range; **sd**, standard deviation.

Appendix 3.9. Definition of intermediate clinical and imaging outcomes in included studies

Study	Definition
Recanalisation or reperfusion	
Lee et al 2009[1]	Successful recanalisation defined as TIMI grade 2 or 3 at 24 hours
Ebinger et al 2012[2]	Reperfusion (specific definition was not reported) on day 2
Olindo et al 2012[3]	Recanalisation defined as TIMI grade 2 or 3 at 24 hours
Cai et al 2014[4]	Reperfusion (specific definition was not reported) at 24 hours
Kufner et al 2015[6]	Recanalisation defined as an increase in at least 2 TIMI points within 24 hours of treatment based on acute and follow-up MRA.
Legrand et al 2016[8]	Complete recanalisation (specific definition was not reported) on MRA at 24 hours
ENCHANTED	Recanalisation defined as revised AOL score 2b or 3 on MRA within 48 hours after randomisation
Ryoo et al 2014[12]	Successful recanalisation (specific definition was not reported) after endovascular treatment
Liu et al 2016a[13]	Recanalisation defined as original TICI scale 2b or 3 after endovascular treatment
Nave et al 2018[15]	Successful recanalisation defined as modified TICI scale 2b or 3 after endovascular treatment
Jiang et al 2019a[16]	Successful recanalisation defined as modified TICI scale 2b or 3 after endovascular treatment
Jiang et al 2019b[17]	Successful recanalisation defined as modified TICI scale 2b or 3 after endovascular treatment
Legrand et al 2019[18]	Successful early revascularization defined as modified TICI scale 2b or 3 after endovascular treatment
Hohenhaus et al 2012[28]	Recanalisation on follow-up MRI (specific definition was not reported) on day 2
Adatia et al 2013a[30]	Recanalisation defined as TIMI grade 2 or 3
Aoki et al 2016[32]	Recanalisation (specific definition was not reported)
Mahdjoub et al 2018[35]	Complete recanalisation defined as AOL score 3 on 24 hours MRA
Any ICH	
Olindo et al 2012[3]	Symptomatic ICH defined as an NIHSS score increase of ≥4 attributable to a bleed detected on the 24 hours MRI
El Nawar et al 2019[10]	Haemorrhagic transformation on CT or MRI within 24 hours after intravenous thrombolysis (ECASS II classification)
ENCHANTED	Any ICH reported by site investigators as a serious adverse event or noted on brain imaging adjudicated centrally
Liu et al 2016b[13]	Haemorrhagic transformation defined as a new hyper-attenuated region identified on any follow-up CT scan before patient discharge
Nam et al 2017[23]	Haemorrhagic transformation defined as a haemorrhagic lesion on follow-up MRI that was not present on the initial MRI
Hohenhaus et al 2012[28]	Haemorrhagic transformation on follow-up MRI within 2 days after stroke onset (ECASS criteria with HI/PH classification)
Adatia et al 2013a[30]	Any intracranial haemorrhage
Early neurological improvement	
Ichijo et al 2015[5]	A decrease in NIHSS score of ≥10 or NIHSS score ≤2 at 24 hours after alteplase treatment
ENCHANTED	A decrease in NIHSS score of ≥8 or NIHSS score ≤2 at 24 hours after randomisation
Ryoo et al 2014[12]	A decrease in NIHSS score of ≥8 or NIHSS score ≤2 at 24 hours after endovascular treatment
Legrand et al 2019[18]	A decrease in NIHSS score of ≥8 at 24 hours
Mahdjoub et al 2018[35]	A decrease in NIHSS score of ≥8 or NIHSS score ≤1 at 24 hours
Early neurological deterioration	
Kim et al 2019a[11]	An increase in total NIHSS score of ≥2 between hospital days 0 and 5
ENCHANTED	An increase in total NIHSS score of ≥2 within 72 hours after hospital admission
Nam et al 2017[23]	An increase in the motor NIHSS score of ≥1 or ≥2 in total NIHSS score within 72 hours after hospital admission
Kim et al 2019b[11]	An increase in total NIHSS score of ≥2 between hospital days 0 and 5
Kim et al 2016[33]	An increase in the motor NIHSS score of ≥1 after hospital admission

CT denotes computed tomography; **ECASS,** the European-Australian Cooperative Acute Stroke Study; **ENCHANTED**, Enhanced Control of Hypertension and Thrombolysis Stroke Study; **HI**, haemorrhagic infarcts; **ICH**, intracerebral haemorrhage; **MRA**, magnetic resonance angiography; **MRI**, magnetic resonance imaging;

NIHSS, National Institutes of Health Stroke Scale; **PH**, parenchymal haematomas; **AOL**, arterial occlusive lesion score; **TICI**, thrombolysis in cerebral infarction scale; **TIMI**, thrombolysis in myocardial infarction grade.

Appendices References

1. Lee KY, Latour LL, Luby M, et al. Distal hyperintense vessels on FLAIR: an MRI marker for collateral circulation in acute stroke? Neurology. 2009;72(13):1134-9.
2. Ebinger M, Kufner A, Galinovic I, et al. Fluid-attenuated inversion recovery images and stroke outcome after thrombolysis. Stroke. 2012;43(2):539-42.
3. Olindo S, Chausson N, Joux J, et al. Fluid-attenuated inversion recovery vascular hyperintensity: an early predictor of clinical outcome in proximal middle cerebral artery occlusion. Arch Neurol. 2012;69(11):1462-8.
4. Cai X, Sun J, Chen Z, et al. Prognostic value of fluid-attenuated inversion recovery imaging vascular hyperintensity in intravenous thrombolysis of acute ischaemic stroke. Chinese Journal of Neurology. 2014;47(9):628-32.
5. Ichijo M, Iwasawa E, Numasawa Y, et al. Significance of Development and Reversion of Collaterals on MRI in Early Neurologic Improvement and Long-Term Functional Outcome after Intravenous Thrombolysis for Ischaemic Stroke. Am J Neuroradiol. 2015;36(10):1839-45.
6. Kufner A, Galinovic I, Ambrosi V, et al. Hyperintense Vessels on FLAIR: Haemodynamic Correlates and Response to Thrombolysis. Am J Neuroradiol. 2015;36(8):1426-30.
7. Legrand L, Tisserand M, Turc G, et al. Do FLAIR vascular hyperintensities beyond the DWI lesion represent the ischaemic penumbra? Am J Neuroradiol. 2015;36(2):269-74.
8. Legrand L, Tisserand M, Turc G, et al. Fluid-Attenuated Inversion Recovery Vascular Hyperintensities-Diffusion-Weighted Imaging Mismatch Identifies Acute Stroke Patients Most Likely to Benefit From Recanalisation. Stroke. 2016;47(2):424-7.
9. Ichijo M, Ishibashi S, Miki K, et al. Significance of hyperintense vessels in negative DWI area as a prognostic factor after thrombolysis (abstract). 2017. 23rd World Congress of Neurology. Japan.
10. El Nawar R, Yeung J, Labreuche J, et al. MRI-Based Predictors of Haemorrhagic Transformation in Patients With Stroke Treated by Intravenous Thrombolysis. Front Neurol. 2019;10:897.
11. Kim DH, Lee YK, Cha JK. Prominent FLAIR Vascular Hyperintensity Is a Predictor of Unfavourable Outcomes in Non-thrombolysed Ischaemic Stroke Patients With Mild Symptoms and Large Artery Occlusion. Front Neurol. 2019;10:722.
12. Ryoo S, Kim J, Lee K. Acute neuroimaging prediction of antegrade flow using flair and GRE in hyperacute stroke (abstract). 2014. 9th World Stroke Congress. Istanbul, Turkey.
13. Liu D, Scalzo F, Rao NM, et al. Fluid-Attenuated Inversion Recovery Vascular Hyperintensity Topography, Novel Imaging Marker for Revascularization in Middle Cerebral Artery Occlusion. Stroke. 2016;47(11):2763-2769.
14. Miura M, Nakajima M, Singu T, et al. Distal hyperintense vessels on FLAIR is a marker of collateral flow grade and outcomes in patients with acute ischaemic stroke (abstract). 2016. International Stroke Conference and State-of-the-Science Stroke Nursing Symposium. Los Angeles, CA United States.
15. Nave AH, Kufner A, Bücke P, et al. Hyperintense Vessels, Collateralization, and Functional Outcome in Patients With Stroke Receiving Endovascular Treatment. Stroke. 2018;49(3):675-681.
16. Jiang L, Chen YC, Zhang H, et al. FLAIR vascular hyperintensity in acute stroke is associated with collateralization and functional outcome. Eur Radiol. 2019;29(9):4879-4888.
17. Jiang L, Peng M, Geng W, et al. FLAIR hyperintensities-DWI mismatch in acute stroke: associations with DWI volume and functional outcome. Brain Imaging Behav. 2019. doi: 10.1007/s11682-019-00156-x. [Epub ahead of print]
18. Legrand L, Turc G, Edjlali M, et al. Benefit from revascularization after thrombectomy according to FLAIR vascular hyperintensities-DWI mismatch. Eur Radiol. 2019;29(10):5567-5576.
19. Zhou SB, Zhang XM, Gao Y, et al. Diffusion-weighted imaging volume and diffusion-weighted imaging volume growth in acute stroke: associations with fluid-attenuated inversion recovery hyperintensities-diffusion-weighted imaging mismatch and functional outcome. Neuroreport. 2019;30(13):875-881.
20. Girot M, Gauvrit JY, Cordonnier C, et al. Prognostic value of hyperintense vessel signals on fluid-attenuated inversion recovery sequences in acute cerebral ischaemia. Eur Neurol. 2007;57(2):75-9.
21. Huang XJ, Zhu WS, Wang QZ, et al. Prognostic value of fluid-attenuated inversion recovery hyperintense vessel in acute middle cerebral artery occlusion. Chinese Journal of Neurology. 2012;45(3):174-8.
22. Song J, Ma Z, Meng H, et al. Distal hyperintense vessels alleviate insula infarction in proximal middle cerebral artery occlusion. Int J Neurosci. 2016;126(11):1030-5.
23. Nam KW, Kwon HM, Park SW, et al. Distal hyperintense vessel sign is associated with neurological deterioration in acute ischaemic stroke. Eur J Neurol. 2017;24(4):617-623.

24. Li CC, Hao XZ, Tian JQ, et al. Predictors of short-term outcome in patients with acute middle cerebral artery occlusion: unsuitability of fluid-attenuated inversion recovery vascular hyperintensity scores. Neural Regen Res. 2018;13(1):69-76.
25. Dong X, Nao J. Fluid-attenuated inversion recovery vascular hyperintensities in anterior circulation acute ischaemic stroke: associations with cortical brain infarct volume and 90-day prognosis. Neurol Sci. 2019;40(8):1675-1682.
26. Song L, Lyu C, Shen G, et al. Application of FLAIR Vascular Hyperintensity-DWI Mismatch in Ischaemic Stroke Depending on Semi-Quantitative DWI-Alberta Stroke Program Early CT Score. Front Neurol. 2019;10:994.
27. Yuan T, Ren G, Hu X, et al. Added assessment of middle cerebral artery and atrial fibrillation to FLAIR vascular hyperintensity-DWI mismatch would improve the outcome prediction of acute infarction in patients with acute internal carotid artery occlusion. Neurol Sci. 2019. doi: 10.1007/s10072-019-04029-5. [Epub ahead of print]
28. Hohenhaus M, Schmidt WU, Brunecker P, et al. FLAIR vascular hyperintensities in acute ICA and MCA infarction: a marker for mismatch and stroke severity?. Cerebrovasc Dis. 2012;34(1):63-9.
29. Pérez de la Ossa N, Hernández-Pérez M, Domènech S, et al. Hyperintensity of distal vessels on FLAIR is associated with slow progression of the infarction in acute ischaemic stroke. Cerebrovasc Dis. 2012;34(5-6):376-84.
30. Adatia S, Almekhlafi M, Nambiar V, et al. FLAIR hyperintense vessel sign on MRI brain (HVS) does not predict any intracranial haemorrhage or symptomatic haemorrhage (SICH) in acute strokes with proximal vessel occlusions treated with intravenous or intra-arterial lytic/ mechanical therapy (abstract). 2013. 2013 Canadian Stroke Congress. Montreal, QC Canada.
31. Adatia S, Nambiar V, Sangll S, et al. New twist to an old turn: Hyperintense vessels (proximal and distal) on early FLAIR MRI brain scans help predict final infarct volume at 24 h and outcome at 3 months in acute ischaemic stroke with proximal vessel occlusions (abstract). 2013. 23rd Meeting of the European Neurological Society. Barcelona Spain.
32. Aoki J, Suzuki K, Sakamoto Y, et al. Negative-FLAIR vascular hyperintensities serves as a marker of chronic major arterial occlusion in acute stroke (abstract). 2016. International Stroke Conference and State-of-the-Science Stroke Nursing Symposium. Los Angeles, CA United States.
33. Kim SE, Lee BI, Kim SE, et al. Clinical Significance of Fluid-Attenuated Inversion Recovery Vascular Hyperintensities in Borderzone Infarcts. Stroke. 2016;47(6):1548-54.
34. Vural A, Gocmen R, Oguz KK, et al. Bright and dark vessels on stroke imaging: different sides of the same coin? Diagn Interv Radiol. 2016;22(3):284-90.
35. Mahdjoub E, Turc G, Legrand L, et al. Do Fluid-Attenuated Inversion Recovery Vascular Hyperintensities Represent Good Collaterals before Reperfusion Therapy? Am J Neuroradiol. 2018;39(1):77-83.

Chapter 4: Low-Dose versus Standard-Dose Alteplase in Acute Lacunar Ischaemic Stroke: The ENCHANTED Trial

4.1 Link to thesis

In Chapters 2 and 3, I presented data on the association of FLAIR-HAs with clinical prognosis based on the ENCHANTED data alone and a systematic review and meta-analysis. Although subgroup analysis of the ENCHANTED alteplase arm showed no heterogeneity ($P_{interaction}$=0.88) of treatment effects on the primary outcome across different AIS subtypes defined by site investigators using standard TOAST (Trial of ORG 10172 in Acute Stroke Treatment) criteria, there is concern that this approach may over-estimate lacunar AIS, representing a single penetrating artery occlusion. In addition, clinical concern persists over whether the modest risk of thrombolysis-related ICH could offset the modest benefits of intravenous thrombolysis for lacunar AIS. All these prompted me to identify the effects of low-dose versus standard-dose alteplase in lacunar AIS confirmed on brain image (more reliable in diagnosing lacunar AIS than the TOAST classifications). This Chapter presents this study.

I have published this work:
Zhou Z, Delcourt C, Xia C, Yoshimura S, Carcel C, Torii-Yoshimura T, You S, Malavera A, Chen X, Hackett ML, Woodward M, Chalmers J, Xu J, Robinson TG, Parsons MW, Demchuk AM, Lindley RI, Mair G, Wardlaw JM, Anderson CS. Low-dose vs standard-dose alteplase in acute lacunar ischaemic stroke: the ENCHANTED trial. *Neurology* 2021;96(11):e1512-26.

I presented this work at one international conference:
Zhou Z, Delcourt C, Xia C, Yoshimura S, Carcel C, Torii-Yoshimura T, You S, Malavera A, Chen X, Hackett ML, Woodward M, Chalmers J, Xu J, Robinson TG, Parsons MW, Demchuk AM, Lindley RI, Mair G, Wardlaw JM, Anderson CS. Low-Dose vs Standard-Dose Alteplase in Acute Lacunar Ischaemic Stroke: The ENCHANTED Trial. 2021 International Stroke Conference, USA, 17-19 March 2021 (virtual online, the abstract was selected as Paul Dudley White International Scholar Award by the American Heart Association to recognise as the highest ranked abstract from Australia).

4.2 Abstract

Objective: To determine any differential efficacy and safety of low- versus standard-dose intravenous alteplase for lacunar vs nonlacunar acute ischaemic stroke (AIS), we performed post hoc analyses from the Enhanced Control of Hypertension and Thrombolysis Stroke Study (ENCHANTED) alteplase dose arm.

Methods: In a cohort of 3,297 ENCHANTED participants, we identified those with lacunar or nonlacunar AIS with different levels of confidence (definite/probable/possible) according to prespecified definitions based on clinical and adjudicated imaging findings. Logistic regression models were used to determine associations of lacunar AIS with 90-day outcomes (primary, modified Rankin Scale [mRS] scores 2-6; secondary, other mRS scores, intracerebral haemorrhage [ICH], and early neurologic deterioration or death) and treatment effects of low- versus standard-dose alteplase across lacunar and nonlacunar AIS with adjustment for baseline covariables.

Results: Of 2,588 participants with available imaging and clinical data, we classified cases as definite/probable lacunar (n=490) or nonlacunar AIS (n=2,098) for primary analyses. Regardless of alteplase dose received, lacunar AIS participants had favourable functional (mRS 2-6, adjusted odds ratio [95% confidence interval] 0.60 [0.47-0.77]) and other clinical or safety outcomes compared to participants with nonlacunar AIS. Low-dose alteplase (versus standard) had no differential effect on functional outcomes (mRS 2-6, 1.04 [0.87–1.24]) but reduced the risk of symptomatic ICH in all included participants. There were no differential treatment effects of low- versus standard-dose alteplase on all outcomes across lacunar and nonlacunar AIS (all $P_{interaction} \geq 0.07$).

Conclusions: We found no evidence from the ENCHANTED trial that low-dose alteplase had any advantages over standard dose for definite/probable lacunar AIS.

Classification of Evidence: This study provides Class II evidence that for patients with lacunar AIS, low-dose alteplase had no additional benefit or safety over standard-dose alteplase.

Clinical Trial Registration: Clinicaltrials.gov identifier NCT01422616.

4.3 Background

In routine clinical practice, patients with lacunar acute ischaemic stroke (AIS) are eligible to receive intravenous (IV) thrombolysis, given comparable favourable outcomes to other common AIS pathologic subtypes.[1-3] These results were confirmed in a recent subgroup analysis of the Efficacy and Safety of Magnetic Resonance Imaging–Based Thrombolysis in Wake-Up Stroke (WAKE-UP) trial, where the safety and efficacy of standard-dose IV alteplase were comparable between lacunar and nonlacunar subtypes defined on baseline MRI.[4] Similar consistency of effect of IV alteplase between lacunar and nonlacunar AIS, defined by the Oxfordshire Community Stroke Project (OCSP) syndromic classification, was found in the third International Stroke Trial (IST-3).[5] Despite this evidence, some clinical concern persists over whether the modest risk of thrombolysis-related intracerebral haemorrhage (ICH) could offset the modest benefits of IV thrombolysis for lacunar AIS, where the natural course is generally more benign compared to other AIS subtypes[6] from there being no or small thrombotic lytic target on the presumption of a single penetrating artery occlusion.[7,8]

In the alteplase-dose arm of the Enhanced Control of Hypertension and Thrombolysis Stroke Study (ENCHANTED),[9] a lower dose (0.6 mg/kg) of IV alteplase was shown to have a lower risk of ICH compared to standard dose (0.9 mg/kg) in thrombolysis-eligible patients with AIS. Whether it is the same for lacunar AIS is unclear. Herein, we report further analyses of the efficacy and safety of low- versus standard dose IV alteplase in the ENCHANTED participants with lacunar (versus nonlacunar) AIS who were identified by the combination of clinical and adjudicated imaging findings.

4.4 Methods

4.4.1 Primary research question and evidence level

Is there are any differential efficacy and safety of low- versus standard-dose IV alteplase between participants with lacunar and nonlacunar AIS in the alteplase dose arm of the ENCHANTED trial? This study provides Class II evidence that for patients with lacunar AIS, low-dose alteplase has no additional benefit or safety over standard-dose alteplase.

4.4.2 Design and participants

ENCHANTED was an international, multicentre, 2×2 quasifactorial, prospective, randomised, open-label, blinded-endpoint trial that assessed the effectiveness of low-dose (0.6 mg/kg; 15% as bolus, 85% as infusion during 1 hour) versus standard-dose (0.9 mg/kg; 10% as bolus, 90% as infusion during 1 hour) IV alteplase, and more intensive versus guideline-recommended control of blood pressure (BP) in adult participants with AIS. The study design, participant characteristics, and main results of the alteplase-dose arm have been reported[9-11] for 3,310 patients with AIS recruited from 111 centres in 13 countries. Key demographic and clinical characteristics were recorded at the time of enrollment, with clinical severity defined according to the NIH Stroke Scale (NIHSS) at baseline, 24 hours, and at day 7 (or on discharge from hospital if earlier). A final clinical diagnosis of AIS subtypes based upon the opinion of site investigations, generally according to the Trial of Org 10172 in Acute Stroke Treatment (TOAST) classification system,[12] was made at day 7, post-randomisation (or on discharge from hospital, if earlier).

4.4.3 Standard protocol approvals, registrations, and participant consents

The study protocol was approved by the appropriate ethics committee at each participating centre and written informed consent was obtained from participants or an appropriate legal surrogate according to the Declaration of Helsinki. The ENCHANTED trial was registered at ClinicalTrials.gov (Unique identifier: NCT01422616).

4.4.4 Imaging analysis

Uncompressed digital images of all baseline and follow-up digital CT, MRI, and angiographic images were uploaded into the study brain imaging database in Digital Imaging and Communications in Medicine (DICOM) format identified only by the participant's unique study identification number. Images were analysed centrally for any ICH by a trial adjudication panel, blind to clinical data, treatment, date, and sequence of scan. Assessors graded any identified symptomatic ICH (sICH) using a range of standard definitions from the Safe Implementation of Thrombolysis in Stroke-Monitoring Study (SITS-MOST), National Institute of Neurologic Disease and Stroke (NINDS), the European-Australian Cooperative Acute Stroke Study II (ECASS), ECASS III, and IST-3 (appendix 4.1).

The ENCHANTED Imaging Analysis Project was established in August 2016, with the aim of defining the presence, extent, and severity of, and swelling from, acute ischaemic changes (including arterial territory, border zone, small subcortical and brainstem/cerebellar infarcts), coexisting old vascular lesions and their subtypes, white matter lesions, and brain volume loss on all collected images by an imaging analysis team of trained individuals, blind to all clinical data, using an electronic scoring system modified from IST-3.[13] All observed infarct lesions on baseline (pre-randomisation) CT or MRI were coded according to the IST-3 criteria for infarct site and size. Separately and subsequent to primary scan reads, a neuroradiologist (Zien Zhou) and neurosurgeon (Chao Xia) sought the ischaemic lesion on 24-hour follow-up images while viewing the baseline images for those with no infarct lesion identified at baseline. They also assessed large vessel occlusion (LVO) on baseline CT angiography (CTA) or magnetic resonance angiography (MRA) according to a modified Thrombolysis in Cerebral Infarction (TICI) score for an abnormal artery in IST-3.[14] All the imaging data were cross-checked (Zien Zhou) and a final rating made before unmasking the clinical data and randomisation code for analyses.

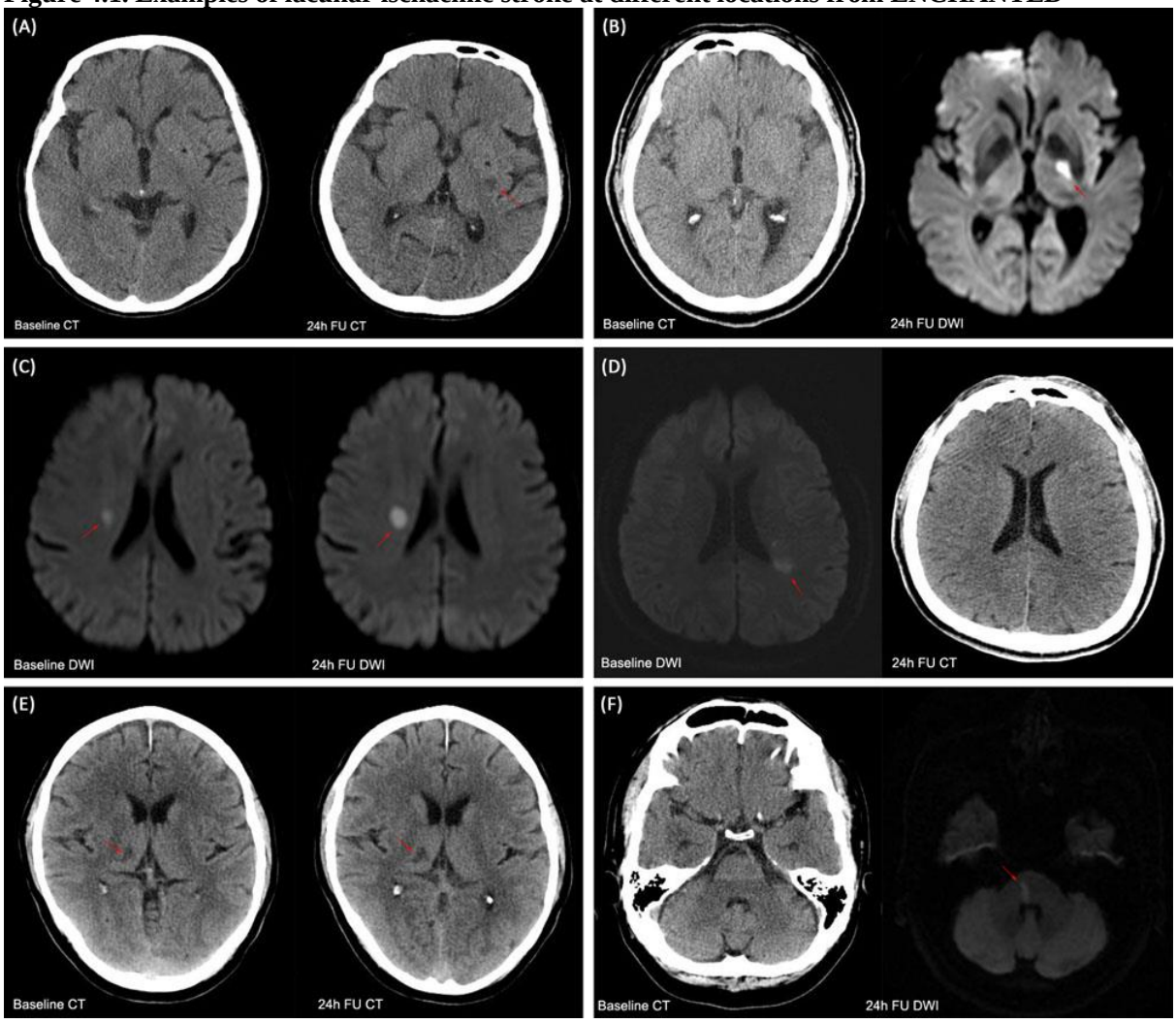
4.4.5 Definitions of lacunar and nonlacunar AIS

Different levels of confidence (definite/probable/possible) were used around the definitions of lacunar and nonlacunar AIS based on adjudicated imaging findings, clinical severity, and clinical diagnosis (appendix 4.2). In brief, definite lacunar AIS was defined when all 4 criteria were met: (1) the presence of acute infarct lesion (maximum diameter ≤20 mm) in the territory of penetrating arteries, with a rounded, ovoid, or tubular shape on axial CT or diffusion-weighted imaging/apparent diffusion coefficient map (figure 4.1)[15]; (2) no LVO adjudicated centrally (on CTA/MRA) or reported by site investigators (on CTA/MRA/digital subtraction angiography); (3) the final diagnosis was reported as "small vessel or perforating vessel 'lacunar' disease" according to the TOAST criteria that involved any of the standard clinical lacunar syndromes with the lack of large vessel atheroma or cerebral cortical dysfunction; and (4) infarct side on images is consistent with that reported by site investigators. Definite nonlacunar AIS was defined as having acute infarct lesion with maximum diameter >20 mm or LVO on angiography. Participants were classified as nonlacunar if they had lacunar and nonlacunar infarcts.

Given that the clinical diagnosis of lacunar syndrome plus baseline NIHSS score <7 had a high specificity to predict imaging-confirmed lacunar stroke in IST-3,[16] probable lacunar and

nonlacunar AIS were discriminated mainly by baseline NIHSS scores and final diagnosis in the situation that there was no acute infarct lesion identified on images or the images were not collected from the sites. For those with conflicting clinical and adjudicated imaging information that compromised the confidence of discrimination, we classified as possible lacunar or nonlacunar AIS according to the clinical diagnosis and LVO status.

Figure 4.1. Examples of lacunar ischaemic stroke at different locations from ENCHANTED



DWI denotes diffusion-weighted imaging; ENCHANTED, Enhanced Control of Hypertension and Thrombolysis Stroke Study.
Lacunar stroke at (A) left lentiform (red arrow) identified on 24-hour follow-up CT; (B) left internal capsule (red arrow) identified on 24-hour follow-up MRI; (C) right centrum semiovale (red arrow) identified on baseline and 24-hour follow-up MRI; (D) left internal border zone (red arrow) identified on baseline MRI; (E) right thalamus (red arrow) identified on baseline and 24-hour follow-up CT; and (F) brainstem (red arrow) identified on 24-hour follow-up MRI.

4.4.6 Outcomes

The primary outcome of these analyses was the composite endpoint of disability or death (modified Rankin Scale [mRS] scores 2-6) at 90 days post-randomisation. Secondary efficacy

outcomes included major disability or death (mRS 3-6), death (mRS 6), and ordinal shift of the full range of mRS scores at 90 days. Secondary safety outcomes were sICH defined according to several criteria from other studies, fatal ICH within 7 days, ICH identified by central adjudicators, and any ICH adjudicated centrally or reported by site investigators. Other clinical outcomes included early neurologic deterioration (END) (≥4-point increase in NIHSS scores) or death within 24 hours or 7 days.

4.4.7 Statistical analysis

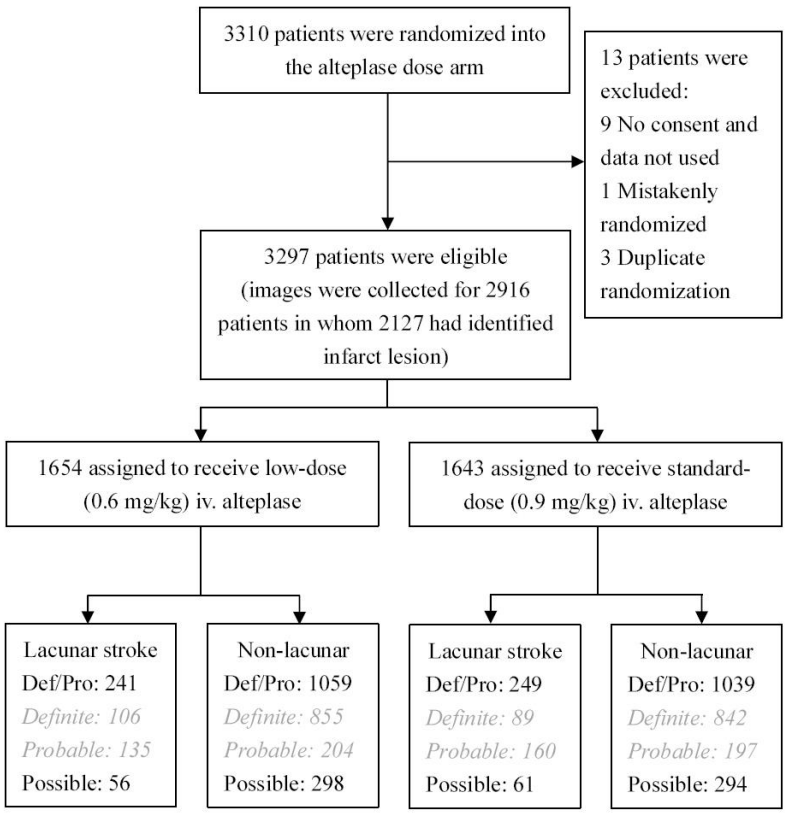
Continuous or categorical variables at baseline were presented as mean (SD), median (interquartile range), or number (percentage). Baseline differences between participants with lacunar and nonlacunar AIS were evaluated using analysis of variance, χ^2 test, or Wilcoxon signed-rank test, as appropriate. Associations of lacunar AIS with 90-day function, safety, and other secondary outcomes were estimated in logistic regression models with adjustment for randomised treatment and key prognostic covariates (age, sex, ethnicity, baseline NIHSS score, time from symptom onset to randomisation, premorbid function [mRS score 0 or 1], prior use of antithrombotic agents, history of diabetes or cardiovascular disease, and assigned to intensive blood pressure-lowering group). The treatment effect of low- versus standard-dose alteplase was determined in logistic regression models and the heterogeneity of alteplase dose effect across participants with lacunar and nonlacunar AIS was estimated by adding an interaction term to statistical models. Proportional odds regression models were used to analyse ordinal mRS scores. The primary analyses pertain to participants with definite/probable lacunar and nonlacunar AIS after excluding those with a possible diagnostic classification. Sensitivity analyses of the treatment effects of low- versus standard-dose alteplase were performed in participants with definite lacunar/nonlacunar AIS and in all participants with possible lacunar/nonlacunar AIS. We also performed an exploratory analysis of the treatment effects in a subset of lacunar AIS identified at baseline (infarct size ≤15 mm and no adjudicated LVO). Data were reported as odds ratios (ORs) and 95% confidence intervals (CIs) and a 2-sided p <0.05 was considered statistically significant. All analyses were performed using SAS version 7.1 and Stata version 12.0.

4.5 Results

4.5.1 Baseline characteristics

Among 3,297 AIS participants in the ENCHANTED alteplase dose arm, 2,588 (78.5%) were classifiable (definite lacunar, n=195; probable lacunar, n=295; definite nonlacunar, n=1,697; and probable nonlacunar, n=401 AIS) for inclusion in the primary analysis (figure 4.2). Compared to the 709 excluded participants, they were more likely to be older, have higher baseline NIHSS scores, be Asian, have a history of cardiovascular disease, and have a final diagnosis of LVO, but they also had shorter time interval from symptom onset to randomisation (appendix 4.3). Table 4.1 shows that all the baseline clinical characteristics were significantly different between definite/probable lacunar and nonlacunar AIS except for sex, history of diabetes, and prior use of statin/other lipid-lowering agents. Participants with lacunar (versus nonlacunar) AIS were younger and had milder neurologic impairment, higher baseline BP, and a lower proportion with conventional cardiovascular risk factors except smoking. In keeping with the lacunar pattern of stroke, few participants had multiple lesions in both anterior and posterior circulation, but they were more likely to have a lesion only in the posterior circulation. They were also less likely to have brain atrophy or a hyperdense vessel sign on CT or hyperintense arteries on MRI.

Figure 4.2. Flowchart of participants included in analyses



Def/Pro denotes definite or probable.

4.5.2 Lacunar AIS and outcomes

Compared to participants with definite/probable nonlacunar AIS, those with definite/probable lacunar AIS had better 90-day functional outcomes, whether defined by the outcome of mRS scores 2-6 (unadjusted OR 0.26, 95% CI 0.21-0.33), mRS scores 3-6 (0.20, 0.15-0.26), ordinal shift in the full range of scores (0.27, 0.23-0.33), or death alone (0.04, 0.01-0.12) (table 4.2). They were also less likely to have ICH and END or death after IV thrombolysis. The findings persisted with adjustment of baseline covariables and randomised alteplase dose.

Table 4.1. Baseline characteristics of participants with definite/probable lacunar and nonlacunar stroke

		Low-dose N=241	**Standard-dose N=249**	**Total N=490**	**P value**[§]
	LACS Non-L	**N=1059**	**N=1039**	**N=2098**	
Age (years)	LACS	63.9 (12.8)	63.1 (12.5)	63.5 (12.7)	**<0.001**
	Non-L	67.8 (12.7)	67.8 (12.6)	67.8 (12.7)	
Female	LACS	82 (34.0)	91 (36.5)	173 (35.3)	0.31
	Non-L	402 (38.0)	390 (37.5)	792 (37.8)	
Asian ethnicity	LACS	179 (74.3)	174 (69.9)	353 (72.0)	**<0.001**
	Non-L	675 (63.7)	663/1038 (63.9)	1338/2097 (63.8)	
Clinical features					
Systolic BP (mmHg)	LACS	151.6 (17.8)	153.7 (19.4)	152.6 (18.7)	**<0.001**
	Non-L	148.0 (19.7)	148.3 (20.2)	148.2 (19.9)	
Diastolic BP (mmHg)	LACS	86.6 (11.7)	86.7 (12.9)	86.6 (12.3)	**<0.001**
	Non-L	84.0 (13.2)	84.2 (13.0)	84.1 (13.1)	
Heart rate (beats per minute)	LACS	76.1 (11.7)	77.7 (12.7)	76.9 (12.2)	**0.001**
	Non-L	79.3 (16.6)	79.7 (16.3)	79.5 (16.5)	
NIHSS score[*]	LACS	4 (3-6)	5 (4-6)	5 (3-6)	**<0.001**
	Non-L	11 (7-16)	11 (7-16)	11 (7-16)	
GCS score[†]	LACS	15 (15-15)	15 (15-15)	15 (15-15)	**<0.001**
	Non-L	14 (12-15)	15 (12-15)	15 (12-15)	
Medical history					
Previous stroke	LACS	39 (16.2)	30 (12.0)	69 (14.1)	**0.03**
	Non-L	185 (17.5)	197 (19.0)	382 (18.2)	
Hypertension	LACS	142 (58.9)	148 (59.4)	290 (59.2)	**0.03**
	Non-L	671 (63.4)	678/1038 (65.3)	1349/2097 (64.3)	
Atrial fibrillation	LACS	9 (3.7)	11 (4.4)	20 (4.1)	**<0.001**
	Non-L	288/1056 (27.3)	259/1038 (25.0)	547/2094 (26.1)	
Coronary artery disease	LACS	23 (9.5)	16 (6.4)	39 (8.0)	**<0.001**
	Non-L	184 (17.4)	171/1038 (16.5)	355/2097 (16.9)	
Valvular/other heart disease	LACS	4 (1.7)	7 (2.8)	11 (2.2)	**<0.001**
	Non-L	92 (8.7)	95/1038 (9.2)	187/2097 (8.9)	
Diabetes mellitus	LACS	50 (20.7)	52 (20.9)	102 (20.8)	0.59
	Non-L	203 (19.2)	211/1038 (20.3)	414/2097 (19.7)	
Hypercholesterolemia	LACS	34 (14.1)	32 (12.9)	66 (13.5)	**0.04**
	Non-L	194 (18.3)	171/1038 (16.5)	365/2097 (17.4)	
Current smoker	LACS	63 (26.1)	85 (34.1)	148 (30.2)	**<0.001**
	Non-L	222/1057 (21.0)	233/1037 (22.5)	455/2094 (21.7)	
Pre-stroke function without disability[‡]	LACS	36 (14.9)	33 (13.3)	69 (14.1)	**0.005**
	Non-L	194/1058 (18.3)	216/1037 (20.8)	410/2095 (19.6)	
Medication on admission					

Antihypertensive agent(s)	LACS	93 (38.6)	103 (41.4)	196 (40.0)	**0.002**
	Non-L	507 (47.9)	496/1038 (47.8)	1003/2097 (47.8)	
Warfarin anticoagulation	LACS	1/240 (0.4)	1 (0.4)	2/489 (0.4)	**<0.001**
	Non-L	39 (3.7)	29/1037 (2.8)	68/2096 (3.2)	
Aspirin/other antiplatelet agent	LACS	46/240 (19.2)	47 (18.9)	93/489 (19.0)	**0.01**
	Non-L	287 (27.1)#	225/1037 (21.7)#	512/2096 (24.4)	
Statin/other lipid lowering agent	LACS	40/240 (16.7)	38 (15.3)	78/489 (16.0)	0.11
	Non-L	215/1058 (20.3)	185/1037 (17.8)	400/2095 (19.1)	
Time from stroke onset to CT/MRI scan (hrs)	LACS	1.8 (1.3-2.5)	1.9 (1.3-2.6)	1.8 (1.3-2.5)	**<0.001**
	Non-L	1.7 (1.1-2.3)	1.6 (1.1-2.3)	1.6 (1.1-2.3)	
Imaging features					
Infarct at left side	LACS	78/153 (51.0)	78/150 (52.0)	156/303 (51.5)	0.14
	Non-L	400/841 (47.6)	384/833 (46.1)	784/1674 (46.8)	
Infarct at right side	LACS	70/153 (45.8)	66/150 (44.0)	136/303 (44.9)	0.11
	Non-L	407/841 (48.4)	428/833 (51.4)	835/1674 (49.9)	
Infarct at midline or bilateral side	LACS	5/153 (3.3)	6/150 (4.0)	11/303 (3.6)	0.76
	Non-L	34/841 (4.0)	21/833 (2.5)	55/1674 (3.3)	
Infarct in anterior circulation only	LACS	117/153 (76.5)	114/150 (76.0)	231/303 (76.2)	0.09
	Non-L	689/841 (81.9)	658/833 (79.0)	1347/1674 (80.5)	
Infarct in posterior circulation only	LACS	35/153 (22.9)	36/150 (24.0)	71/303 (23.4)	**<0.001**
	Non-L	103/841 (12.2)	120/833 (14.4)	223/1674 (13.3)	
Infarct in anterior and posterior circulation	LACS	1/153 (0.7)	0/150 (0.0)	1/303 (0.3)	**<0.001**
	Non-L	49/841 (5.8)	55/833 (6.6)	104/1674 (6.2)	
With FLAIR-HAs or hyperdense vessel sign	LACS	3/151 (2.0)	5/156 (3.2)	8/307 (2.6)	**<0.001**
	Non-L	306/835 (36.6)	305/826 (36.9)	611/1661 (36.8)	
With old vascular lesions	LACS	70/153 (45.8)	59/150 (39.3)	129/303 (42.6)	0.83
	Non-L	362/841 (43.0)	362/833 (43.5)	724/1674 (43.2)	
With brain atrophy	LACS	94/153 (61.4)	79/150 (52.7)	173/303 (57.1)	**<0.001**
	Non-L	574/841 (68.3)	589/833 (70.7)	1163/1674 (69.5)	
With white matter changes	LACS	64/153 (41.8)#	46/150 (30.7)#	110/303 (36.3)	0.82
	Non-L	301/841 (35.8)	318/833 (38.2)	619/1674 (37.0)	
Site reported LVO or assessed centrally	LACS	0 (0.0)	0 (0.0)	0 (0.0)	**<0.001**
	Non-L	270/1041 (25.9)	262/1027 (25.5)	532/2068 (25.7)	
Time from stroke onset to randomisation (hrs)	LACS	3.0 (2.3-3.7)	2.8 (2.2-3.6)	2.9 (2.2-3.6)	**<0.001**
	Non-L	2.6 (1.9-3.3)	2.6 (1.9-3.4)	2.6 (1.9-3.4)	
Assigned to intensive BP lowering	LACS	48 (19.9)	48 (19.3)	96 (19.6)	**<0.001**
	Non-L	127 (12.0)	139 (13.4)	266 (12.7)	
Assigned to standard BP lowering	LACS	41 (17.0)	48 (19.3)	89 (18.2)	**0.008**
	Non-L	145 (13.7)	138 (13.3)	283 (13.5)	

Data are n (%), mean (SD), or median (Q1, Q3). The P values are based on Chi-square, analysis of variance, or Wilcoxon signed-rank test.

BP denotes blood pressure; CTA, computed tomographic angiography; FLAIR, fluid-attenuated inversion recovery; FLAIR-HAs, FLAIR hyperintense arteries; GCS, Glasgow Coma Scale; LACS, lacunar stroke; LVO, large vessel occlusion; MRI, magnetic resonance imaging; mRS, modified Rankin scale; NIHSS, National Institutes of Health stroke scale; Non-L, non-lacunar stroke.

*Scores on the NIHSS range from 0 to 42, with higher scores indicating more severe neurological deficits.

†Scores on the GCS range from 15 (normal) to 3 (deep coma).

‡mRS=0.

§Total lacunar stroke versus total non-lacunar stroke.

#P<0.05 by randomisation treatment.

4.5.3 Lacunar AIS and alteplase dose

The overall treatment effects of low- versus standard-dose alteplase on function, safety, and other outcomes in these 2,588 participants were comparable to the main results of the

Table 4.2. Thrombolysis outcomes in definite/probable lacunar versus nonlacunar stroke

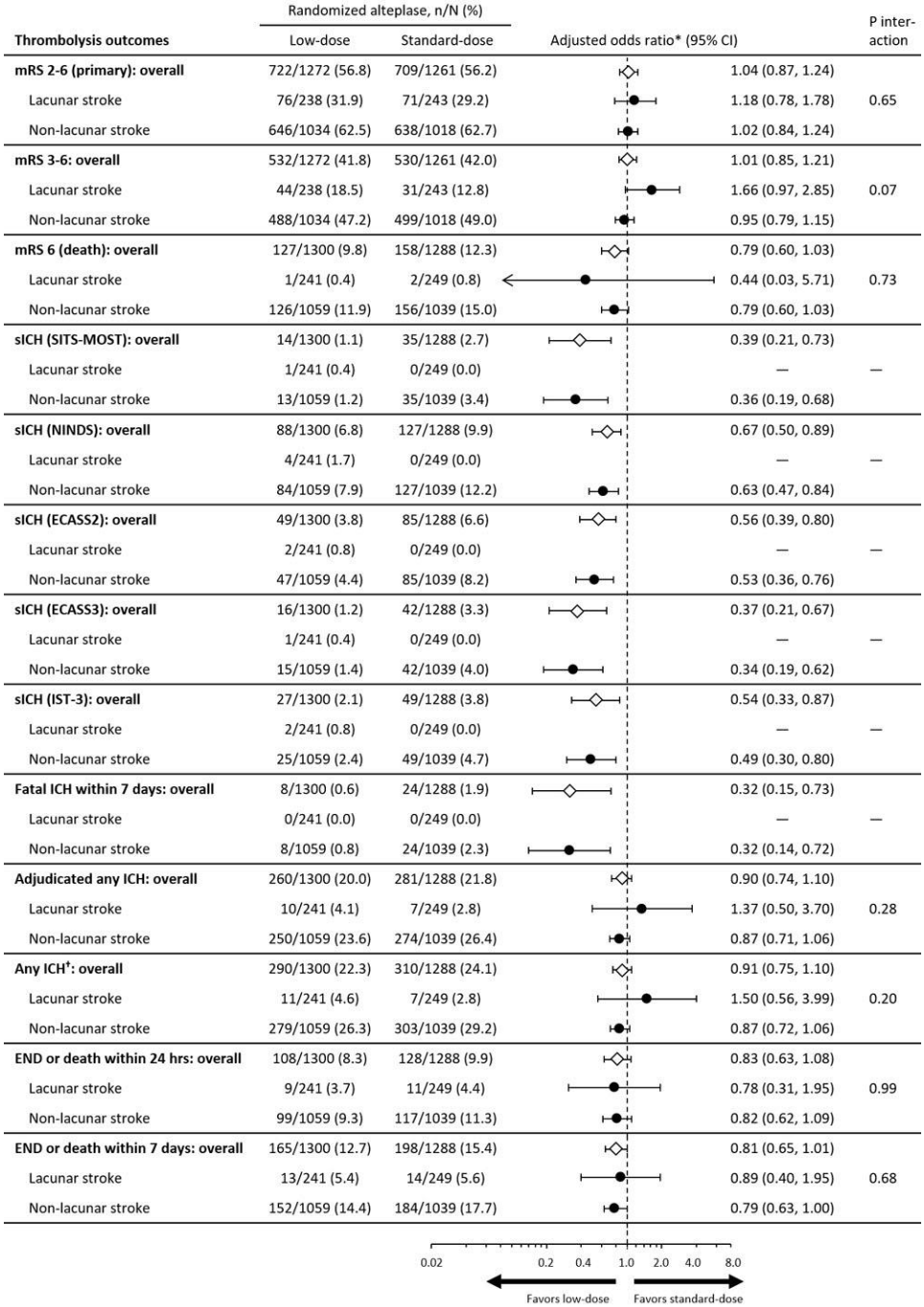
	Lacunar	Non-lacunar	Lacunar versus non-lacunar stroke			
	n/N (%)	n/N (%)	OR (95% CI)*	P value	AOR (95% CI)*†	P value
90-day functional outcomes						
mRS 2–6	147/481 (30.6)	1284/2052 (62.6)	0.26 (0.21, 0.33)	**<0.001**	0.60 (0.47, 0.77)	**<0.001**
mRS 3–6	75/481 (15.6)	987/2052 (48.1)	0.20 (0.15, 0.26)	**<0.001**	0.51 (0.38, 0.69)	**<0.001**
mRS 6	3/490 (0.6)	282/2098 (13.4)	0.04 (0.01, 0.12)	**<0.001**	0.13 (0.04, 0.43)	**<0.001**
mRS 0	185/481 (38.5)	370/2052 (18.0)	0.27 (0.23, 0.33)	**<0.001**	0.64 (0.52, 0.78)	**<0.001**
1	149/481 (31.0)	398/2052 (19.4)				
2	72/481 (15.0)	297/2052 (14.5)				
3	47/481 (9.8)	278/2052 (13.5)				
4	21/481 (4.4)	265/2052 (12.9)				
5	4/481 (0.8)	162/2052 (7.9)				
6	3/481 (0.6)	282/2052 (13.7)				
Safety outcomes (sICH or ICH)						
SITS-MOST	1/490 (0.2)	48/2098 (2.3)	0.09 (0.01, 0.63)	**0.02**	0.09 (0.01, 0.70)	**0.02**
NINDS	4/490 (0.8)	211/2098 (10.1)	0.07 (0.03, 0.20)	**<0.001**	0.10 (0.04, 0.27)	**<0.001**
ECASS2	2/490 (0.4)	132/2098 (6.3)	0.06 (0.02, 0.25)	**<0.001**	0.08 (0.02, 0.31)	**<0.001**
ECASS3	1/490 (0.2)	57/2098 (2.7)	0.07 (0.01, 0.53)	**0.01**	0.08 (0.01, 0.58)	**0.01**
IST-3	2/490 (0.4)	74/2098 (3.5)	0.11 (0.03, 0.46)	**0.002**	0.13 (0.03, 0.54)	**0.005**
Fatal ICH	0/490 (0.0)	32/2098 (1.5)	-	-	-	-
Adjudicated any ICH	17/490 (3.5)	524/2098 (25.0)	0.11 (0.07, 0.18)	**<0.001**	0.18 (0.11, 0.29)	**<0.001**
Any ICH	18/490 (3.7)	582/2098 (27.7)	0.10 (0.06, 0.16)	**<0.001**	0.16 (0.10, 0.27)	**<0.001**
Other secondary outcomes						
END or death						
within 24 hrs	20/490 (4.1)	216/2098 (10.3)	0.37 (0.23, 0.59)	**<0.001**	0.30 (0.18, 0.50)	**<0.001**
within 7 days	27/490 (5.5)	336/2098 (16.0)	0.31 (0.20, 0.46)	**<0.001**	0.36 (0.24, 0.56)	**<0.001**

ECASS denotes the European-Australian Cooperative Acute Stroke Study; END, early neurologic deterioration; ICH, intracerebral haemorrhage; IST-3, the third International Stroke Trial; mRS, modified Rankin scale; NINDS, the National Institutes of Neurological Diseases and Stroke; sICH, symptomatic intracerebral haemorrhage; SITS-MOST, the Safe Implementation of Thrombolysis in Stroke-Monitoring Study.

*Refer to the effect of intravenous thrombolysis in definite/probable lacunar stroke versus non-lacunar stroke after pooling the two groups of randomised alteplase dose as one cohort.

†Adjusted for key prognostic covariates (age; sex; ethnicity; baseline NIHSS score; time from stroke onset to randomisation; pre-morbid function [mRS scores 0 or 1]; prior use of antithrombotic agents [aspirin, other antiplatelet agent, or warfarin]; history of diabetes mellitus or cardiovascular disease [stroke, atrial fibrillation, coronary artery disease, valvular or other heart disease]; assigned to intensive blood pressure lowering group; and randomisation to low-dose alteplase group) for functional outcomes. Adjusted for minimization and key prognostic covariates (age, baseline NIHSS score, time from stroke onset to randomisation, assigned to intensive blood pressure lowering group, and randomisation to low-dose alteplase group) for safety outcomes, and neurologic deterioration within 24 hours and 7 days.

Figure 4.3. Thrombolysis outcomes in participants with definite/probable lacunar and nonlacunar stroke by randomised treatment

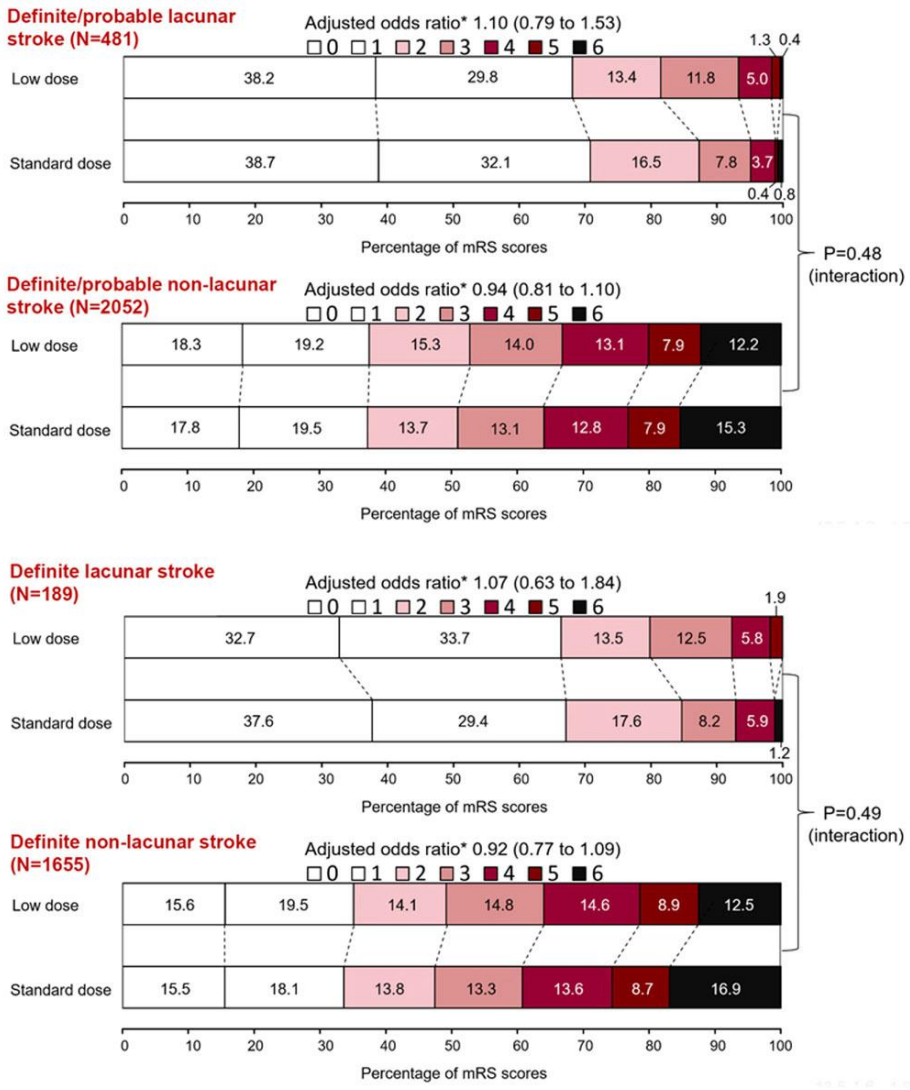


CI denotes confidence interval; ECASS, European-Australian Cooperative Acute Stroke Study; END, early neurologic deterioration; ICH, intracerebral haemorrhage; IST-3, third International Stroke Trial; NINDS, National Institutes of Neurologic Diseases and Stroke; sICH, symptomatic intracerebral haemorrhage; SITS-MOST, Safe Implementation of Thrombolysis in Stroke-Monitoring Study.

*Adjusted for key prognostic covariates (age, sex, ethnicity, baseline NIH Stroke Scale [NIHSS] score, time from stroke onset to randomisation, premorbid function [modified Rankin Scale (mRS) scores 0 or 1], prior use of antithrombotic agents [aspirin, other antiplatelet agent, or warfarin], history of diabetes or cardiovascular disease [stroke, atrial fibrillation, coronary artery disease, valvular or other heart disease], assigned to intensive blood pressure lowering group) for functional outcomes. Adjusted for minimization and key prognostic covariates (age, baseline NIHSS score, time from stroke onset to randomisation, and assigned to intensive blood pressure lowering group) for safety outcomes and neurologic deterioration within 24 hours or 7 days.

†Site reported or adjudicated centrally.

Figure 4.4. Randomised treatment effects on the ordinal modified Rankin scale (mRS) score by lacunar and nonlacunar stroke

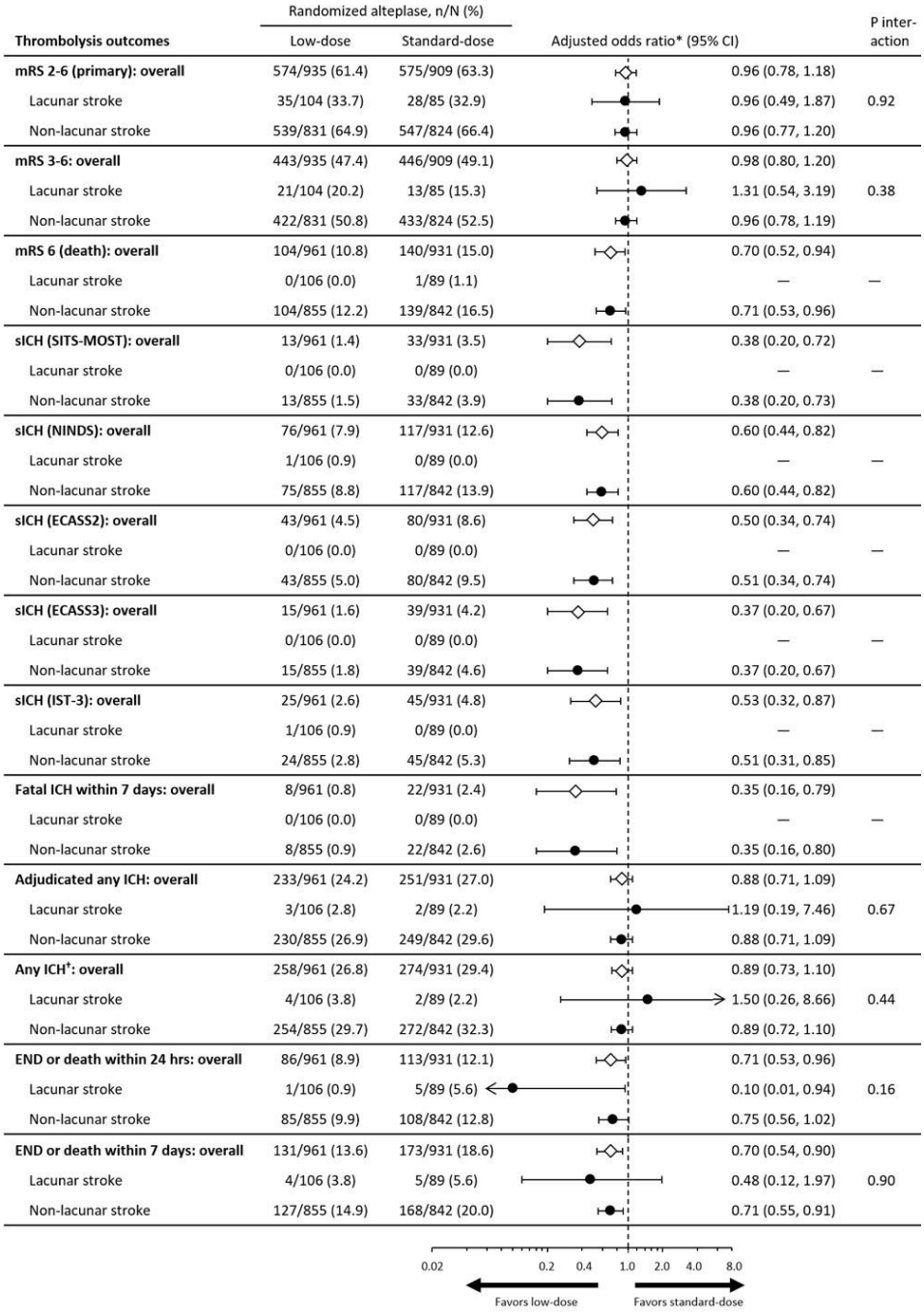


mRS denotes modified Rankin scale

*Adjusted for key prognostic covariates (age; sex; ethnicity; baseline NIHSS score; time from stroke onset to randomisation; pre-morbid function [mRS scores 0 or 1]; prior use of antithrombotic agents [aspirin, other antiplatelet agent, or warfarin]; history of diabetes mellitus or cardiovascular disease [stroke, atrial fibrillation, coronary artery disease, valvular or other heart disease]; assigned to intensive blood pressure lowering group).

ENCHANTED trial, that low-dose versus standard-dose alteplase reduced the risk of sICH (SITS-MOST criteria, adjusted OR 0.39, 95% CI 0.21-0.73; NINDS criteria, 0.67, 0.50-0.89; ECASS II criteria, 0.56, 0.39-0.80; ECASS III criteria, 0.37, 0.21-0.67; IST-3 criteria, 0.54, 0.33-0.87) but with no difference in effect on functional outcomes (mRS 2-6, adjusted OR 1.04, 95% CI 0.87-1.24; mRS 3-6, 1.01, 0.85-1.21). There was no heterogeneity of treatment effects on all outcomes for definite/probable lacunar versus nonlacunar AIS after adjustment for baseline covariables (all $P_{interaction} \geq 0.07$) (figures 4.3 and 4.4). Similar results were seen in the sensitivity analyses for definite lacunar and nonlacunar AIS (all $P_{interaction} \geq 0.16$) (figures 4.4

Figure 4.5. Thrombolysis outcomes in participants with definite lacunar and nonlacunar stroke by randomised treatment



CI denotes confidence interval; ECASS, European-Australian Cooperative Acute Stroke Study; END, early neurologic deterioration; ICH, intracerebral haemorrhage; IST-3, third International Stroke Trial; NINDS, National Institutes of Neurologic Diseases and Stroke; sICH, symptomatic intracerebral haemorrhage; SITS-MOST, Safe Implementation of Thrombolysis in Stroke-Monitoring Study.

*Adjusted for key prognostic covariates (age, sex, ethnicity, baseline NIH Stroke Scale [NIHSS] score, time from stroke onset to randomisation, premorbid function [modified Rankin Scale (mRS) scores 0 or 1], prior use of antithrombotic agents [aspirin, other antiplatelet agent, or warfarin], history of diabetes or cardiovascular disease [stroke, atrial fibrillation, coronary artery disease, valvular or other heart disease], assigned to intensive blood pressure lowering group) for functional outcomes. Adjusted for minimization and key prognostic covariates (age, baseline NIHSS score, time from stroke onset to randomisation, and assigned to intensive blood pressure lowering group) for safety outcomes and neurologic deterioration within 24 hours or 7 days.

†Site reported or adjudicated centrally.

and 5.5) and definite/probable/possible lacunar and nonlacunar AIS (all $P_{interaction} \geq 0.12$) (data available on request).

Specifically, in the definite subgroup of lacunar AIS, there were no significant differences on the primary efficacy outcome (mRS 2-6) (33.7% vs 32.9%, adjusted OR 0.96, 95% CI 0.49-1.87) or major disability or death (mRS 3-6) (20.2% vs 15.3%, adjusted OR 1.31, 95% CI 0.54-3.19) between low dose and standard-dose alteplase groups (figure 4.5). There was one case of sICH (0.9%) meeting NINDS and IST-3 criteria in participants with definite lacunar AIS treated by low-dose alteplase, but no case of sICH was observed after use of standard-dose alteplase. In participants with definite lacunar AIS who received low-dose alteplase, 3 (2.8%) had adjudicated ICH and 1 more had ICH reported by a site investigator, while any ICH occurred in 2 (2.2%) participants with definite lacunar AIS assigned to the standard-dose group. In a small subset of definite lacunar AIS identified at baseline with size <15 mm and no adjudicated LVO, 4 of the 9 participants (44.4%) in the low-dose group and 2 of the 7 participants (28.6%) in the standard-dose group had mRS 2-6 at 90 days post-randomisation, and no ICH occurred in either treatment group (appendix 4.4).

4.6 Discussion and conclusion

4.6.1 Discussion

In these post-hoc analyses of the ENCHANTED trial, we did not identify any benefit, nor any harm, from the use of low-dose alteplase versus standard-dose alteplase to treat patients with lacunar AIS compared to those with other subtypes of AIS. As well as having a range of significantly different characteristics, the 90-day outcomes were better for those with lacunar than nonlacunar AIS, which provided some internal consistency for the classifications used in our study. However, given the low event rate of sICH, with fewer than 5 events in the primary analysis for definite or probable lacunar AIS, we are limited in the conclusions that can be drawn as to whether a lower dose of IV alteplase should be preferred because of the good prognosis for lacunar AIS.

Our results on thrombolysis outcomes for lacunar AIS are consistent with prior observational studies.[2,17-21] However, the net benefit of thrombolysis for lacunar AIS is still debated, mainly because the evidence is drawn from subgroup analyses of trials, such as WAKE-UP[4] and IST-3,[5] where there is low statistical power. In addition, accurate identification of lacunar AIS is challenging, especially in the absence of an acute lesion on the initial CT, and even MRI (in

nearly one third of patients with nondisabling stroke).[22] The pragmatic approach of applying a lacunar syndrome classification system in studies has moderate diagnostic sensitivity and specificity,[15] which may potentially mix patients with nonlacunar AIS with the target population of lacunar AIS, and nondifferentially bias results towards IV thrombolysis.

We were unable to confirm in ENCHANTED participants any benefit of low-dose over standard-dose alteplase in lacunar AIS. The fact that there were few cases of sICH in the low-dose alteplase group, and no sICH in the standard-dose group, highlights the potential for chance and imprecise estimates of treatment effects when there are few events. Even with current imaging techniques and clinical criteria, it is difficult to discriminate lacunar AIS due to occlusion of a deep penetrating arteriole presumed caused by progressive lipohyalinosis from thrombosis related to atherosclerosis or embolus. Platelet activation triggered by disintegration of the endothelium from intrinsic cerebral small vessel disease (CSVD) may also be relevant in this type of AIS.[8] It is possible, therefore, that IV thrombolysis may have a differential effect dependent on the cause of lacunar stroke, being more effective when there is underlying thromboembolism. In lacunar AIS, we noted a significant imbalance in the frequency of background white matter lesions between the low-dose and standard-dose alteplase groups (41.8% vs 30.7%), which could partly account for more ICH in the former (appendix 4.5).[23] Again, however, due to the few sICH events in patients with lacunar AIS, we cannot confirm whether the increase in sICH by low-dose alteplase was confounded by CSVD.

4.6.2 Strengths and limitations

Some strengths of our study include the large, prospective, multicentre cohort of patients with AIS who had systematic, complete, and high-quality data collected prospectively, where we were able to adjust for multiple covariables in statistical models. Furthermore, the imaging assessment was completed blind to clinical features and other data, using a rigorously defined approach developed for the IST-3 study. However, we acknowledge limitations that include insufficient statistical power and inevitable selection bias from the data being derived from a clinical trial where a large number of participants were from Asia and had mild to moderate stroke. Moreover, given the pragmatic nature of ENCHANTED, few participants had a baseline brain MRI, and the identification of lacunar AIS required analysis of follow-up images with comparison to those obtained at baseline. Whereas this approach may have altered the imaging appearances of acute ischaemic lesions after use of IV thrombolysis[24] and limited the identification of all true lacunar AIS, our results are comparable with previous work showing

that nearly one-third of patients with nondisabling AIS lack an infarct lesion on acute MRI (median 4 days poststroke).[22] In the ENCHANTED alteplase arm, 27.1% (789/2,916) of participants had no infarct lesion on either the baseline or 24-hour follow-up images. Thus, we had to use a combination of clinical and adjudicated imaging data to classify as many cases as possible into lacunar and nonlacunar AIS, which likely closely represents that used in routine practice. Relatively small samples in lacunar AIS compromised the power of a reliable assessment of any interaction, especially for sICH. Moreover, regarding the outcomes of major disability or death, a $P_{interaction}$ of 0.07 might have been due to chance rather than true differential treatment effects of low- versus standard-dose alteplase across definite/probable lacunar and nonlacunar AIS. Future research in systematic reviews and clinical registries may be required to confirm or refute these findings.

4.6.3 Conclusion

We found no clear evidence that low-dose IV alteplase was any better or safer than standard-dose alteplase in the ENCHANTED participants who had lacunar AIS. According to standard eligibility criteria, patients with lacunar AIS should receive standard dose IV alteplase as with other AIS subtypes.

References

1. Hsia AW, Sachdev HS, Tomlinson J, et al. Efficacy of IV tissue plasminogen activator in acute stroke: does stroke subtype really matter? Neurology 2003;61(1):71-5.
2. Fuentes B, Martínez-Sánchez P, Alonso de Leciñana M, et al. Efficacy of intravenous thrombolysis according to stroke subtypes: the Madrid Stroke Network data. Eur J Neurol 2012;19(12):1568-74.
3. Mustanoja S, Meretoja A, Putaala J, et al. Outcome by stroke etiology in patients receiving thrombolytic treatment: descriptive subtype analysis. Stroke 2011;42(1):102-6.
4. Barow E, Boutitie F, Cheng B, et al. Functional outcome of intravenous thrombolysis in patients with lacunar infarcts in the WAKE-UP trial. JAMA Neurol 2019;76(6):641-9.
5. Lindley RI, Wardlaw JM, Whiteley WN, et al. Alteplase for acute ischaemic stroke: outcomes by clinically important subgroups in the third International Stroke Trial. Stroke 2015;46(3):746-56.
6. Bamford J, Sandercock P, Jones L, et al. The natural history of lacunar infarction: the Oxfordshire Community Stroke Project. Stroke 1987;18(3):545-51.
7. Norrving B. Lacunar infarcts: no black holes in the brain are benign. Pract Neurol 2008;8(4):222-8.
8. Regenhardt RW, Das AS, Lo EH, Caplan LR. Advances in Understanding the Pathophysiology of Lacunar Stroke: A Review. JAMA Neurol 2018;75(10):1273-81.
9. Anderson CS, Robinson T, Lindley RI, et al. Low-dose versus standard-dose intravenous alteplase in acute ischaemic stroke. N Engl J Med 2016;374(24):2313-23.
10. Huang Y, Sharma VK, Robinson T, et al. Rationale, design, and progress of the ENhanced Control of Hypertension ANd Thrombolysis strokE stuDy (ENCHANTED) trial: an international multicentre 2×2 quasi-factorial randomised controlled trial of low- vs. standard-dose rt-PA and early intensive vs. guideline-recommended blood pressure lowering in patients with acute ischaemic stroke eligible for thrombolysis treatment. Int J Stroke 2015;10(5):778-88.
11. Anderson CS, Woodward M, Arima H, et al. Statistical analysis plan for evaluating low- vs. standard-dose alteplase in the ENhanced Control of Hypertension and Thrombolysis strokE stuDy (ENCHANTED). Int J Stroke 2015;10(8):1313-5.
12. Adams HP Jr, Bendixen BH, Kappelle LJ, et al. Classification of subtype of acute ischaemic stroke. Definitions for use in a multicentre clinical trial. TOAST. Trial of Org 10172 in Acute Stroke Treatment. Stroke 1993;24(1):35-41.
13. Wardlaw JM, Sandercock P, Cohen G, et al. Association between brain imaging signs, early and late outcomes, and response to intravenous alteplase after acute ischaemic stroke in the third International Stroke Trial (IST-3): secondary analysis of a randomised controlled trial. Lancet Neurol 2015;14(5):485-96.
14. Mair G, von Kummer R, Adami A, et al. Arterial obstruction on computed tomographic or magnetic resonance angiography and response to intravenous thrombolytics in ischaemic stroke. Stroke 2017;48(2):353-60.
15. Wardlaw JM, Smith EE, Biessels G, et al. Neuroimaging standards for research into small vessel disease and its contribution to ageing and neurodegeneration. Lancet Neurol 2013;12(8):822-38.
16. Arba F, Mair G, Phillips S, et al. Improving clinical detection of acute lacunar stroke: analysis from the IST-3. Stroke 2020;51(5):1411-8.

17. Matusevicius M, Paciaroni M, Caso V, et al. Outcome after intravenous thrombolysis in patients with acute lacunar stroke: an observational study based on SITS international registry and a meta-analysis. Int J Stroke 2019;14(9):878-86.
18. Zivanovic Z, Gubi M, Vlahovic D, et al. Patients with acute lacunar infarction have benefit from intravenous thrombolysis. J Stroke Cerebrovasc Dis 2019;28(2):435-40.
19. Eggers CCJ, Bocksrucker C, Seyfang L, et al. The efficacy of thrombolysis in lacunar stroke - evidence from the Austrian Stroke Unit Registry. Eur J Neurol 2017;24(6):780-7.
20. Shobha N, Fang J, Hill MD. Do lacunar strokes benefit from thrombolysis? Evidence from the registry of the Canadian Stroke Network. Int J Stroke 2013;8(Suppl A100):45-9.
21. Fluri F, Hatz F, Rutgers MP, et al. Intravenous thrombolysis in patients with stroke attributable to small artery occlusion. Eur J Neurol 2010;17(8):1054-60.
22. Makin SD, Doubal FN, Dennis MS, et al. Clinically confirmed stroke with negative diffusion-weighted imaging magnetic resonance imaging: longitudinal study of clinical outcomes, stroke recurrence, and systematic review. Stroke 2015;46(11):3142-8.
23. Pantoni L, Fierini F, Poggesi A. Thrombolysis in acute stroke patients with cerebral small vessel disease. Cerebrovasc Dis 2014;37(1):5-13.
24. Nagaraja N, Forder JR, Warach S, et al. Reversible diffusion-weighted imaging lesions in acute ischaemic stroke: a systematic review. Neurology 2020;94(13):571-87.

Appendices

Appendix 4.1. Definitions of symptomatic intracerebral haemorrhage

For intracerebral haemorrhage, bleeding was coded as HI1 (small petechiae along infarct margins), HI2 (confluent petechiae within infarcted area without space-occupying effect), PH1 (blood clot(s) in <30% of infarcted area with slight space-occupying effect) and PH2 (blood clot(s) in >30% of infarcted area with substantial space-occupying effect). In addition, independent assessors were asked to adjudicate if haemorrhage was the predominant cause of neurological worsening, and if there was evidence of midline shift. These assessments enabled the following definitions of symptomatic intracerebral haemorrhage (sICH) to be adjudicated: Safe Implementation of Thrombolysis in Stroke Monitoring Study (SITS-MOST): large or remote parenchymal ICH (type 2, defined as greater than 30% of the infarcted area affected by haemorrhage with mass effect or extension outside the infarct) combined with neurological deterioration (>4 points on the NIHSS) or leading to death within 24 to 36 hours [Wahlgren et al, 2007]; any ICH associated with neurological deterioration (>1 point change in NIHSS score) from baseline or death within 24 to 36 hours (NINDS) [NINDS Study Group, 1995]; any ICH with neurological deterioration (>4 points on the NIHSS) from baseline or death within 24 to 36 hours (ECASS2) [Hacke et al, 1998]; any ICH with neurological deterioration (>4 points increase on the NIHSS) from baseline or death within 36 hours (ECASS3) [Hacke et al, 2008]; either significant ICH (local or distant from the infarct) or significant haemorrhagic transformation of an infarct on brain imaging with clinically significant deterioration or death within the first 7 days of treatment (IST3) [IST-3 Collaborative Group, 2012]; and fatal ICH, any type 2 parenchymal ICH and death within 7 days.

References

Wahlgren N, Ahmed N, Davalos S, et al. Thrombolysis with alteplase for acute ischaemic stroke in the Safe Implementation of Thrombolysis in Stroke-Monitoring Study (SITS-MOST): an observational study Lancet 2007; 369: 275-282.

The National Institute of Neurological Disorders and Stroke rt-PA Stroke Study Group. Tissue plasminogen activator for acute ischaemic stroke. N Engl J Med 1995; 333:1581-1587.

Hacke W, Kaste M, Fieschi C, et al. Randomised double-blind placebo controlled trial of thrombolytic therapy with intravenous alteplase in acute ischaemic stroke (ECASS II). Lancet 1998; 352: 1245-1251.

Hacke W, Kaste M, Bluhmki E, et al. Thrombolysis with alteplase 3 to 4.5 hours after acute ischaemic stroke. N Engl J Med 2008; 359: 1317-1329.

The IST-3 Collaborative Group. The benefits and harms of intravenous thrombolysis with recombinant tissue plasminogen activator within 6 h of acute ischaemic stroke (the third international stroke trial [IST-3]): a randomised controlled trial. Lancet 2012; 379:2352-2363.

Appendix 4.2. Definitions and samples of lacunar and nonlacunar stroke

1. Definite lacunar stroke (N=195)
Meet all following 4 points
(1) Subcortical or brainstem lesions (maximum diameter ≤20 mm) in the territory of penetrating arteries, with a rounded, ovoid, or tubular shape on axial CT or diffusion-weighted imaging (DWI)/apparent diffusion coefficient (ADC) map. For participants without MRI at baseline or 24 hours follow-up, baseline and follow-up CT scans need to be compared so as to exclude old lesion with no change in lesion size/shape and attenuation between baseline and follow-up scans.
(2) No large vessel occlusion (LVO) on cerebral computed tomographic angiography (CTA)/magnetic resonance angiography (MRA) assessed centrally* or on cerebral CTA/MRA/digital subtraction angiography (DSA) reported by site investigators.
(3) The final diagnosis by site investigators is "Small vessel or perforating vessel 'lacunar' disease" according to the TOAST (Trial of ORG 10172 in Acute Stroke Treatment) classification.
(4) Side of infarct lesion assessed on brain images centrally is consistent to that reported by site investigators.

2. Probable lacunar stroke (criterion 1 or 2 or 3)
Criterion 1 (N=17): Meet the criteria of definite lacunar stroke except the consistency of infarct side assessed centrally on brain images and that reported by site investigators.

Criterion 2 (N=91):
Meet all following 4 points
(1) Subcortical or brainstem lesions (maximum diameter ≤20 mm) in the territory of penetrating arteries, with a rounded, ovoid, or tubular shape on axial CT or diffusion-weighted imaging (DWI)/apparent diffusion coefficient (ADC) map. For participants without MRI at baseline or 24 hours follow-up, baseline and follow-up CT scans need to be compared so as to exclude old lesion with no change in lesion size/shape and attenuation between baseline and follow-up scans.
(2) No large vessel occlusion (LVO) on cerebral angiography assessed centrally or reported by site investigators.
(3) The final diagnosis by site investigators is **NOT** "Small vessel or perforating vessel 'lacunar' disease" according to the TOAST classification.
(4) Baseline National Institutes of Health stroke scale (NIHSS) scores <7.

Criterion 3 (N=187):
Meet all following 4 points
(1) No acute infarct lesion identified on brain images or the brain images were not collected at the George Institute.
(2) No LVO on cerebral angiography assessed centrally or reported by site investigators.
(3) The final diagnosis by site investigators is "Small vessel or perforating vessel 'lacunar' disease" according to the TOAST classification.
(4) Baseline NIHSS scores <7.

3. Definite non-lacunar stroke (N=1697)
Acute infarct lesion with maximum diameter >20 mm or LVO identified on brain images. For participants without MRI at baseline or 24 hours follow-up, baseline and follow-up CT scans need to be compared so as to exclude old lesion with no change in lesion size/shape and attenuation between baseline and follow-up scans.

4. Probable non-lacunar stroke (N=401)
Meet all following 3 points
(1) No acute infarct lesion identified on brain images or the brain images were not collected at the George Institute.
(2) With LVO on cerebral angiography reported by site investigators or the final diagnosis by site investigators is "Large artery occlusion because of atheroma", "Cardio-emboli", "Dissection", "Other definite pathological mechanism", or "Uncertain aetiology" according to the TOAST classification.
(3) Baseline NIHSS scores ≥7.

5. Possible lacunar stroke (N=117)

Meet all following 3 points
(1) Does not meet the criteria of definite and probable lacunar or non-lacunar stroke.
(2) The final diagnosis by site investigators is "Small vessel or perforating vessel 'lacunar' disease" according to the TOAST classification.
(3) No LVO reported by site investigators.

6. Possible non-lacunar stroke (N=592)
Meet all following 2 points
(1) Does not meet the criteria of definite and probable lacunar or non-lacunar stroke.
(2) The final diagnosis by site investigators is **NOT** "Small vessel or perforating vessel 'lacunar' disease" according to the TOAST classification **OR** there is LVO reported by site investigators (including unknown LVO status).

*LVO assessed centrally on brain CTA or MRA: >50% stenosis (IST-3 Angiography Score for stenosis/occlusion) at anterior cerebral artery, M1 and M2 segment of middle cerebral artery, posterior cerebral artery, basilar artery, vertebral artery, and intracranial and extracranial Internal carotid artery.

References
Adams HP Jr, Bendixen BH, Kappelle LJ, et al. Classification of subtype of acute ischaemic stroke. Definitions for use in a multicentre clinical trial. TOAST. Trial of Org 10172 in Acute Stroke Treatment. Stroke. 1993;24(1):35-41.

Arba F, Mair G, Phillips S, et al. Improving Clinical Detection of Acute Lacunar Stroke: Analysis From the IST-3. Stroke. 2020. 51(5):1411-1418.

Heldner MR, Hsieh K, Broeg-Morvay A, et al. Clinical prediction of large vessel occlusion in anterior circulation stroke: mission impossible? J Neurol. 2016;263(8):1633-40.

Appendix 4.3. Baseline characteristics of ENCHANTED rtPA arm participants included and excluded for primary analyses

	Included (N=2588)	Excluded (N=709)	P value
Age (years)	67.0 (12.8)	64.0 (12.8)	**<0.001**
≥80 years	383 (14.8)	89 (12.6)	0.13
Female	965 (37.3)	283 (39.9)	0.20
Asian ethnicity	1691/2587 (65.4)	388/704 (55.1)	**<0.001**
Clinical features			
Systolic BP (mmHg)	149.0 (19.8)	150.2 (19.9)	0.14
Diastolic BP (mmHg)	84.6 (13.0)	85.0 (12.5)	0.44
Heart rate (beats per minute)	79.0 (15.8)	79.1 (13.8)	0.89
NIHSS score*	9 (5-15)	6 (4-10)	**<0.001**
NIHSS score ≥14	776 (30.0)	77 (10.9)	**<0.001**
GCS score†	15 (13-15)	15 (14-15)	**<0.001**
Medical history			
Previous stroke	451 (17.4)	138 (19.5)	0.21
Hypertension	1639/2587 (63.4)	426/701 (60.8)	0.21
Atrial fibrillation	567/2584 (21.9)	69/701 (9.8)	**<0.001**
Coronary artery disease	394/2587 (15.2)	85/701 (12.1)	**0.04**
Valvular/other heart disease	198/2587 (7.7)	37/701 (5.3)	**0.03**
Diabetes mellitus	516/2587 (19.9)	130/701 (18.5)	0.41
Hypercholesterolaemia	431/2587 (16.7)	124/701 (17.7)	0.52
Current smoker	603/2584 (23.3)	167/700 (23.9)	0.77
Pre-stroke function without symptoms (mRS=0)	479/2585 (18.5)	133/701 (19.0)	0.79
Medication on admission			
Antihypertensive agent(s)	1199/2587 (46.3)	299/701 (42.7)	0.08
Warfarin anticoagulation	70/2585 (2.7)	12/700 (1.7)	0.14
Aspirin/other antiplatelet agent	605/2585 (23.4)	147/700 (21.0)	0.18
Statin/other lipid lowering agent	478/2584 (18.5)	137/700 (19.6)	0.52
Time from onset to randomisation (hrs)	2.7 (2.0-3.4)	2.8 (2.1-3.5)	**0.005**
Large vessel occlusion‡	532/2558 (20.8)	59/689 (8.6)	**<0.001**
Randomised low-dose treatment	1300 (50.2)	354 (49.9)	0.89
Assigned to intensive BP lowering	362 (14.0)	94 (13.3)	0.62
Assigned to standard BP lowering	372 (14.4)	107 (15.1)	0.63
Presumed diagnosis at time of hospital separation			
Non-stroke	10/2560 (0.4)	87/674 (12.9)	**<0.001**
Large artery occlusion due to significant atheroma	1025/2560 (40.0)	245/674 (36.4)	0.08
Small vessel disease	548/2560 (21.4)	125/674 (18.5)	0.10
Cardioembolism	571/2560 (22.3)	70/674 (10.4)	**<0.001**
Dissection	20/2560 (0.8)	5/674 (0.7)	0.92
Other or uncertain pathogenesis	386/2560 (15.1)	142/674 (21.1)	**<0.001**

Data are n (%), mean (SD), or median (Q1, Q3). P values are based on Chi-square, analysis of variance, or Wilcoxon signed-rank test

BP denotes blood pressure; GCS, Glasgow coma scale; mRS, modified Rankin scale; NIHSS, National Institutes of Health stroke scale.

*Scores on the NIHSS range from 0 to 42, with higher scores indicating more severe neurological deficits.

†Scores on the GCS range from 15 (normal) to 3 (deep coma).

‡Reported by site investigators (on cerebral computed tomographic angiography [CTA], magnetic resonance angiography [MRA], or digital subtraction angiography) or assessed centrally (on cerebral CTA or MRA).

Appendix 4.4. Thrombolysis outcomes in participants with definite lacunar stroke (≤15 mm, no LVO adjudicated centrally) identified on baseline images* by randomised treatment

	Randomised alteplase, n/N (%)		OR (95% CI)	P
	Low-dose	Standard-dose	(Low vs. standard)	value
Death or disability (mRS 2-6, primary)	4/9 (44.4)	2/7 (28.6)	2.00 (0.24, 16.36)	0.52
Death or major disability (mRS 3-6)	2/9 (22.2)	2/7 (28.6)	0.71 (0.07, 6.92)	0.77
mRS categories (unadjusted) 0	2/9 (22.2)	3/7 (42.9)	1.64 (0.27, 9.89)	0.59
1	3/9 (33.3)	2/7 (28.6)		
2	2/9 (22.2)	0/7 (0.0)		
3	2/9 (22.2)	1/7 (14.3)		
4	0/9 (0.0)	1/7 (14.3)		
sICH				
SITS-MOST criteria	0/9 (0.0)	0/7 (0.0)	-	-
NINDS criteria	0/9 (0.0)	0/7 (0.0)	-	-
ECASS2 criteria	0/9 (0.0)	0/7 (0.0)	-	-
ECASS3 criteria	0/9 (0.0)	0/7 (0.0)	-	-
IST-3 criteria	0/9 (0.0)	0/7 (0.0)	-	-
Fatal ICH within 7 days	0/9 (0.0)	0/7 (0.0)	-	-
Adjudicated any ICH	0/9 (0.0)	0/7 (0.0)	-	-
Any ICH (clinical report or adjudicated)	0/9 (0.0)	0/7 (0.0)	-	-
END or death within 24 hours	0/9 (0.0)	0/7 (0.0)	-	-
END or death within 7 days	0/9 (0.0)	0/7 (0.0)	-	-

ECASS denotes the European-Australian Cooperative Acute Stroke Study; END, early neurologic deterioration; ICH, intracerebral haemorrhage; IST-3, the third International Stroke Trial; LVO, large vessel occlusion; mRS, modified Rankin scale; NINDS, the National Institutes of Neurological Diseases and Stroke; sICH, symptomatic intracerebral haemorrhage; SITS-MOST, the Safe Implementation of Thrombolysis in Stroke-Monitoring Study.

*Among 16 participants with lacunar stroke (≤15 mm, no LVO adjudicated centrally) identified at baseline, 14 were confirmed on MRI and 2 were confirmed on CT.

Appendix 4.5. Thrombolysis outcomes in participants with definite/probable lacunar and non-lacunar stroke, in whom the presence of white matter lesion was eligible to assess on collected images, by randomised treatment

	Randomised aleplase, n/N (%)		$OR_{unadjusted}$ (95% CI)	P	$OR_{adjusted}$* (95% CI)	P	$OR_{adjusted}$† (95% CI)	P
	Low-dose	Standard-dose	(Low vs. standard)	INT	(Low vs. standard)	INT	(Low vs. standard)	INT
90-day functional outcomes								
Death or disability (mRS 2–6, primary)								
Overall	591/967 (61.1)	595/960 (62.0)	0.96 (0.80, 1.16)		0.98 (0.80, 1.20)		0.98 (0.80, 1.20)	
Lacunar	60/150 (40.0)	51/145 (35.2)	1.23 (0.77, 1.97)	0.28	1.17 (0.70, 1.93)	0.44	1.15 (0.69, 1.91)	0.47
Non-lacunar	531/817 (65.0)	544/815 (66.7)	0.92 (0.75, 1.14)		0.95 (0.76, 1.18)		0.95 (0.76, 1.19)	
Death or major disability (mRS 3–6)								
Overall	450/967 (46.5)	453/960 (47.2)	0.97 (0.81, 1.17)		1.00 (0.82, 1.23)		1.00 (0.82, 1.23)	
Lacunar	35/150 (23.3)	23/145 (15.9)	1.61 (0.90, 2.90)	0.08	1.59 (0.84, 3.00)	0.13	1.57 (0.83, 2.97)	0.14
Non-lacunar	415/817 (50.8)	430/815 (52.8)	0.92 (0.76, 1.12)		0.95 (0.76, 1.18)		0.95 (0.77, 1.18)	
Death (mRS 6)								
Overall	103/994 (10.4)	140/983 (14.2)	0.70 (0.53, 0.91)		0.69 (0.51, 0.94)		0.69 (0.51, 0.94)	
Lacunar	0/153 (0.0)	2/150 (1.3)	-	0.95	-	0.94	-	0.94
Non-lacunar	103/841 (12.2)	138/833 (16.6)	0.70 (0.53, 0.93)		0.71 (0.53, 0.96)		0.71 (0.52, 0.96)	
mRS categories (unadjusted)								
Overall 0	170/967 (17.6)	175/960 (18.2)	0.92 (0.79, 1.08)		0.94 (0.80, 1.10)		0.94 (0.80, 1.10)	
1	206/967 (21.3)	190/960 (19.8)						
2	141/967 (14.6)	142/960 (14.8)						
3	146/967 (15.1)	121/960 (12.6)						
4	128/967 (13.2)	119/960 (12.4)						
5	73/967 (7.5)	73/960 (7.6)						
6	103/967 (10.7)	140/960 (14.6)						
Lacunar 0	42/150 (28.0)	50/145 (34.5)	1.32 (0.87, 1.99)	0.12	1.15 (0.75, 1.75)	0.27	1.14 (0.75, 1.73)	0.29
1	48/150 (32.0)	44/145 (30.3)						
2	25/150 (16.7)	28/145 (19.3)						
3	24/150 (16.0)	12/145 (8.3)						
4	9/150 (6.0)	8/145 (5.5)						
5	2/150 (1.3)	1/145 (0.7)						
6	0/150 (0.0)	2/145 (1.4)						
Non- 0	128/817 (15.7)	125/815 (15.3)	0.88 (0.74, 1.04)		0.90 (0.76, 1.07)		0.91 (0.76, 1.08)	
lacunar 1	158/817 (19.3)	146/815 (17.9)						
2	116/817 (14.2)	114/815 (14.0)						
3	122/817 (14.9)	109/815 (13.4)						
4	119/817 (14.6)	111/815 (13.6)						

5	71/817 (8.7)	72/815 (8.8)						
6	103/817 (12.6)	138/815 (16.9)						
Safety outcomes								
sICH (SITS-MOST criteria)								
Overall	13/994 (1.3)	33/983 (3.4)	0.38 (0.20, 0.73)		0.38 (0.20, 0.73)		0.38 (0.20, 0.73)	
Lacunar	0/153 (0.0)	0/150 (0.0)	-	-	-	-	-	-
Non-lacunar	13/841 (1.5)	33/833 (4.0)	0.38 (0.20, 0.73)		0.38 (0.20, 0.73)		0.38 (0.20, 0.73)	
sICH (NINDS criteria)								
Overall	77/994 (7.7)	117/983 (11.9)	0.62 (0.46, 0.84)		0.63 (0.46, 0.85)		0.62 (0.46, 0.85)	
Lacunar	3/153 (2.0)	0/150 (0.0)	-	0.95	-	0.95	-	0.95
Non-lacunar	74/841 (8.8)	117/833 (14.0)	0.59 (0.43, 0.80)		0.59 (0.44, 0.81)		0.59 (0.43, 0.81)	
sICH (ECASS2 criteria)								
Overall	43/994 (4.3)	80/983 (8.1)	0.51 (0.35, 0.75)		0.51 (0.35, 0.75)		0.51 (0.35, 0.75)	
Lacunar	1/153 (0.7)	0/150 (0.0)	-	0.96	-	0.96	-	0.96
Non-lacunar	42/841 (5.0)	80/833 (9.6)	0.49 (0.34, 0.73)		0.50 (0.34, 0.73)		0.50 (0.34, 0.73)	
sICH (ECASS3 criteria)								
Overall	15/994 (1.5)	39/983 (4.0)	0.37 (0.20, 0.68)		0.37 (0.20, 0.68)		0.37 (0.20, 0.68)	
Lacunar	0/153 (0.0)	0/150 (0.0)	-	-	-	-	-	-
Non-lacunar	15/841 (1.8)	39/833 (4.7)	0.37 (0.20, 0.68)		0.37 (0.20, 0.68)		0.37 (0.20, 0.68)	
sICH (IST-3 criteria)								
Overall	25/994 (2.5)	45/983 (4.6)	0.54 (0.33, 0.88)		0.54 (0.33, 0.89)		0.54 (0.33, 0.89)	
Lacunar	1/153 (0.7)	0/150 (0.0)	-	0.95	-	0.95	-	0.95
Non-lacunar	24/841 (2.9)	45/833 (5.4)	0.51 (0.31, 0.85)		0.52 (0.31, 0.86)		0.52 (0.31, 0.86)	
Fatal ICH within 7 days								
Overall	8/994 (0.8)	22/983 (2.2)	0.35 (0.16, 0.80)		0.36 (0.16, 0.80)		0.35 (0.16, 0.80)	
Lacunar	0/153 (0.0)	0/150 (0.0)	-	-	-	-	-	-
Non-lacunar	8/841 (1.0)	22/833 (2.6)	0.35 (0.16, 0.80)		0.35 (0.16, 0.80)		0.35 (0.16, 0.80)	
Adjudicated any ICH								
Overall	235/994 (23.6)	253/983 (25.7)	0.89 (0.73, 1.10)		0.90 (0.73, 1.11)		0.90 (0.73, 1.11)	
Lacunar	6/153 (3.9)	5/150 (3.3)	1.18 (0.35, 3.96)	0.64	1.09 (0.32, 3.72)	0.63	1.00 (0.29, 3.44)	0.60
Non-lacunar	229/841 (27.2)	248/833 (29.8)	0.88 (0.71, 1.09)		0.89 (0.71, 1.10)		0.88 (0.71, 1.09)	
Any ICH (clinical report or adjudicated)								
Overall	260/994 (26.2)	276/983 (28.1)	0.91 (0.74, 1.11)		0.91 (0.74, 1.12)		0.91 (0.74, 1.12)	
Lacunar	7/153 (4.6)	5/150 (3.3)	1.39 (0.43, 4.48)	0.46	1.26 (0.38, 4.14)	0.46	1.18 (0.36, 3.92)	0.43
Non-lacunar	253/841 (30.1)	271/833 (32.5)	0.89 (0.73, 1.10)		0.90 (0.73, 1.11)		0.89 (0.72, 1.10)	

Other secondary outcomes								
Neurologic deterioration or death within 24 hours								
Overall	89/994 (9.0)	118/983 (12.0)	0.72 (0.54, 0.96)		0.72 (0.54, 0.97)		0.72 (0.54, 0.97)	
Lacunar	6/153 (3.9)	11/150 (7.3)	0.52 (0.19, 1.43)	0.50	0.45 (0.15, 1.30)	0.48	0.46 (0.16, 1.32)	0.46
Non-lacunar	83/841 (9.9)	107/833 (12.8)	0.74 (0.55, 1.01)		0.75 (0.55, 1.01)		0.75 (0.55, 1.02)	
Neurologic deterioration or death within 7 days								
Overall	133/994 (13.4)	179/983 (18.2)	0.69 (0.54, 0.89)		0.70 (0.55, 0.89)		0.70 (0.55, 0.89)	
Lacunar	9/153 (5.9)	12/150 (8.0)	0.72 (0.29, 1.76)	0.93	0.63 (0.25, 1.59)	0.97	0.65 (0.26, 1.63)	0.99
Non-lacunar	124/841 (14.7)	167/833 (20.0)	0.69 (0.53, 0.89)		0.70 (0.54, 0.90)		0.70 (0.54, 0.90)	

ECASS denotes the European-Australian Cooperative Acute Stroke Study; ICH, intracerebral haemorrhage; INT, interaction; IST-3, the third International Stroke Trial; mRS, modified Rankin scale; NINDS, the National Institutes of Neurological Diseases and Stroke; sICH, symptomatic intracerebral haemorrhage; SITS-MOST, the Safe Implementation of Thrombolysis in Stroke-Monitoring Study.

*Adjusted for key prognostic covariates (age; sex; ethnicity; baseline NIHSS score; time from stroke onset to randomisation; pre-morbid function [mRS scores 0 or 1]; prior use of antithrombotic agents [aspirin, other antiplatelet agent, or warfarin]; history of diabetes mellitus or cardiovascular disease [stroke, atrial fibrillation, coronary artery disease, valvular or other heart disease]; and assigned to intensive blood pressure lowering group) for functional outcomes. Adjusted for minimization and key prognostic covariates (age, baseline NIHSS score, time from stroke onset to randomisation, and assigned to intensive blood pressure lowering group) for safety outcomes, death or neurologic deterioration within 24 hours and 7 days.

[†]Adding the adjustment for the presence of white matter lesion.

Chapter 5: Intensive versus Guideline-recommended Blood Pressure Reduction in Acute Lacunar Stroke with Intravenous Thrombolysis Therapy: The ENCHANTED Trial

5.1 Link to thesis

Chapter 4 presented a post-hoc analysis from the ENCHANTED alteplase arm on low-dose versus standard-dose intravenous alteplase in acute lacunar ischaemic stroke. I provided Class II evidence that for patients with lacunar AIS, low-dose alteplase had no additional benefit or safety over standard-dose alteplase. Subsequently, I performed counterpart analysis in the ENCHANTED BP arm to identify the effects of intensive versus guideline-recommended BP lowering in thrombolysis-treated patients with lacunar AIS identified by a combination of clinical and centrally adjudicated imaging findings. The work is presented in this Chapter.

I have published this work:
Zhou Z, Xia C, Carcel C, Yoshimura S, Wang X, Delcourt C, Malavera A, Chen X, Mair G, Woodward M, Chalmers J, Demchuk AM, Lindley RI, Robinson TG, Parsons MW, Wardlaw JM, Anderson CS. Intensive versus guideline-recommended blood pressure reduction in acute lacunar stroke with intravenous thrombolysis therapy: the ENCHANTED trial. *Eur J Neurol* 2021;28(3):783-93.

5.2 Abstract

Objective: This was an investigation of the differential effects of early intensive versus guideline-recommended blood pressure (BP) lowering between lacunar and non-lacunar acute ischaemic stroke (AIS) in the BP arm of the Enhanced Control of Hypertension and Thrombolysis Stroke Study (ENCHANTED).

Methods: In 1,632 participants classified as having definite or probable lacunar (n=454 [27.8%]) or non-lacunar AIS according to pre-specified definitions based upon clinical and adjudicated imaging findings, mean BP changes over days 0-7 were plotted, and systolic BP differences by treatment between subgroups were estimated in generalised linear models. Logistic regression models were used to estimate the BP treatment effects on 90-day outcomes (primary, an ordinal

shift of modified Rankin scale scores) across lacunar and non-lacunar AIS after adjustment for baseline covariables.

Results: Most baseline characteristics, acute BP and other management differed between lacunar and non-lacunar AIS, but mean systolic BP differences by treatment were comparable at each time point (all $P_{interaction}$ >0.12) and over 24 h post-randomisation (−5.5, 95% CI −6.5, −4.4 mmHg in lacunar AIS versus −5.6, 95% CI −6.3, −4.8 mmHg in non-lacunar AIS, $P_{interaction}$=0.93). The neutral effect of intensive BP lowering on functional outcome and the beneficial effect on intracranial haemorrhage were similar for the two subgroups (all $P_{interaction}$ >0.19).

Conclusions: There were no differences in the treatment effect of early intensive versus guideline-recommended BP lowering across lacunar and non-lacunar AIS.

5.3 Background

Optimal blood pressure (BP) management in acute ischaemic stroke (AIS) patients who are candidates for intravenous thrombolysis is uncertain. Current guideline recommendations (BP <185/110 and <180/105 mmHg, prior to and for 24 h after alteplase, respectively) are based on expert opinion given a clear association of elevated BP and increased risk of symptomatic intracerebral haemorrhage (ICH).[1,2] The completed BP arm of the Enhanced Control of Hypertension and Thrombolysis Stroke Study (ENCHANTED) was the first large randomised control trial to determine the effectiveness of more intensive (target systolic BP [SBP] 130-140 mmHg within 1 h after randomisation) versus guideline-recommended BP management (SBP <180 mmHg) in thrombolysis AIS patients.[3] The results showed a neutral effect of intensive BP lowering versus guideline recommendation on functional outcome, but with a significant beneficial effect on intracranial haemorrhage.

Prior studies suggest differences in BP levels at the time of stroke onset,[4] time courses before and after ictus,[5,6] and in relation to BP lowering and prognosis[7,8] between lacunar and non-lacunar AIS as well as across other AIS subtypes, raising the possibility that BP management in AIS may need stratification according to underlying aetiology. BP lowering might increase the risk of harm in AIS patients with large vessel occlusion (LVO) if greater infarct growth before recanalisation offsets the potential benefit of intensive BP lowering in reducing the risk of reperfusion ICH.[9,10] However, there may be no need to have the same concern for lacunar AIS given a single penetrating artery occlusion with a small infarct lesion. Although subgroup

analysis of the ENCHANTED BP arm showed no heterogeneity ($P_{interaction}$=0.90) of treatment effects across different AIS subtypes defined by site investigators using standard Trial of ORG 10172 in Acute Stroke Treatment (TOAST) criteria, there is concern that this approach may over-estimate the frequency of lacunar AIS and of diagnostic reliability being influenced by the experience of assessors.[11-13] Herein, investigation of the effects of intensive BP lowering in thrombolysis-treated patients in the BP arm of ENCHANTED across lacunar and non-lacunar AIS identified by a combination of clinical and centrally adjudicated imaging findings was reported.

5.4 Methods

5.4.1 Design and patients

The study design, patient characteristics and main results of the ENCHANTED BP arm have been reported.[3,14,15] Ethics committees at each participating centre approved the ENCHANTED protocol, and written informed consent was obtained from patients or an appropriate legal surrogate. Key demographic and clinical characteristics were recorded at the time of enrolment, with the severity of neurological symptoms defined according to the National Institutes of Health Stroke Scale (NIHSS) at baseline and at 24 and 72 h after the start of alteplase treatment. The clinical diagnosis of AIS subtypes according to TOAST criteria was made by site investigators at day 7 (or on discharge from hospital, if earlier). Non-invasive BP monitoring was performed using an automated device applied to the non-hemiparetic arm (or right arm in situations of coma or tetraparesis), with the participant resting supine for at least 3 min. After receiving alteplase, BP measurements were recorded every 15 min for 1 h, hourly from hours 1 to 6, 6 hourly from hours 6 to 24, and twice daily from days 2 to 7 (or until death or discharge from hospital, if earlier). All patients were followed up to 90 days by trained and certified researchers at each site to assess functional outcomes and health-related quality of life, blind to treatment allocation.

5.4.2 Imaging assessment

Brain images at baseline (pre-randomisation) and 24-h follow-up were collected and adjudicated centrally for any intracranial haemorrhage by an imaging adjudication panel, blind to clinical information. Assessors graded any identified symptomatic ICH using a range of standard definitions (appendix 5.1). Since August 2016, the nonhaemorrhagic components of the brain imaging were assessed by a research team with a background in stroke after receiving

standardized training (www.ed.ac.uk/edinb urgh-imagi ng/access).[16] All observed infarct lesions on baseline computed tomography or magnetic resonance imaging were coded by the team according to the third International Stroke Trial (IST-3) criteria for infarct site and size.[17] Separately and subsequent to primary scan reads, a neuroradiologist (Zien Zhou) and neurosurgeon (Chao Xia) sought the ischaemic lesion on 24-h follow-up images whilst viewing baseline images in parallel for those with no infarct lesion identified at baseline. LVO on angiography at baseline was assessed (by Zien Zhou and Chao Xia) according to a modified Thrombolysis in Cerebral Infarction score for abnormal artery used in IST-3.[18] The definitions of lacunar and non-lacunar AIS were pre-specified with different levels of confidence (definite/probable/possible) based on adjudicated imaging findings, the severity of neurological symptoms at baseline and clinical diagnosis of AIS subtypes (appendix 5.2). All the imaging data included in these analyses were cross-checked (Zien Zhou) and a final rating was made before unmasking the clinical data and the randomisation code for statistical analyses.

5.4.3 Outcomes

An ordinal shift of the full range of scores on the modified Rankin scale (mRS) at 90 days post-randomisation, the pre-specified primary outcome in the ENCHANTED BP arm, was adopted as the primary outcome for these analyses. Secondary efficacy outcomes included disability or death (mRS scores 2-6), major disability or death (mRS scores 3-6) and death at 90 days, and neurological deterioration (≥4 points increase in NIHSS scores) or death within 24 and 72 h. Secondary safety outcomes were any intracranial haemorrhage adjudicated centrally or reported by site investigators within 7 days after randomisation, any ICH identified by central adjudicators, and symptomatic ICH defined according to the criteria from the Safe Implementation of Thrombolysis in Stroke Monitoring Study (SITS-MOST).[19]

5.4.4 Statistical analysis

These analyses pertain to participants with ‘definite’ or ‘probable’ lacunar and non-lacunar AIS. Baseline differences between lacunar and non-lacunar AIS were evaluated using analysis of variance, chi-squared test or Wilcoxon signed-rank test, where appropriate. BP changes over the acute period in lacunar and non-lacunar AIS by treatment group were plotted for visual inspection, and between group SBP differences up to 7 days post-randomisation were estimated using the generalised linear model, with BP treatment as the exploratory variable and SBP at each time point as the dependent variable. From days 2 to 7, mean BP values of the two

measurements per day were included for analysis. The treatment effects of intensive versus guideline-recommended BP reduction were determined in logistic regression models with adjustment of key prognostic covariates (age, sex, ethnicity, baseline NIHSS score, time from stroke onset to randomisation, pre-morbid function [mRS scores 0 or 1], prior use of antithrombotic agents, history of diabetes mellitus or cardiovascular disease, and assigned to low-dose alteplase group). Heterogeneity of treatment effect across patients with lacunar and non-lacunar AIS was estimated by adding an interaction term to statistical models. A proportional odds regression model was used to determine outcome on the ordinal mRS. Sensitivity analyses were also performed in participants with 'definite' lacunar/non-lacunar AIS and an exploratory analysis was performed of the treatment effects in a subset of lacunar AIS with infarct size ≤15 mm on 24-h follow-up images. Data were reported as odds ratios (OR) and 95% confidence intervals (CI), and a two-sided p value <0.05 was considered statistically significant. All analyses were performed using SAS version 7.1 and Stata version 12.0.

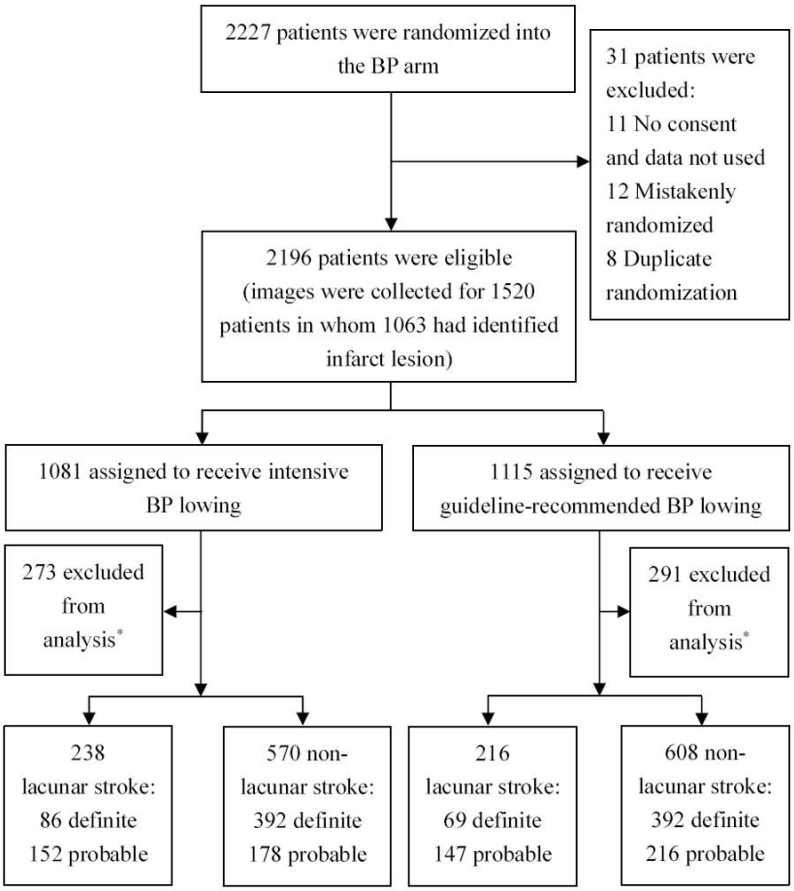
5.5 Results

5.5.1 Baseline characteristics

Amongst 2,196 AIS patients in the ENCHANTED BP arm, there were 1,632 (74.3%) patients with definite/probable lacunar or nonlacunar AIS (Figure 5.1). Compared with 564 remaining participants (defined as 'possible' lacunar or non-lacunar AIS) excluded from analyses, they were older, had more severe neurological impairment at baseline, more likely to have a history of cardiovascular disease, no pre-morbid symptoms of functional impairment, treatment with antihypertensive agents, LVO, and allocation to the alteplase arm, although they had a shorter time interval from symptom onset to randomisation (appendix 5.3). The final diagnoses of AIS subtypes were different between included and excluded participants, except for AIS attributed to dissection. There were 454 (27.8%) participants with lacunar AIS (definite 155; probable 299). The clinical characteristics at baseline were all significantly different between those with lacunar and non-lacunar AIS except sex, SBP, history of hypertension, current smoker, and allocation to the alteplase arm (table 5.1). Regarding the imaging features, patients with lacunar AIS were less likely to have infarction involving both anterior and posterior circulation, hyperdense vessel sign on computed tomography or brain atrophy (appendix 5.4).

5.5.2 Blood pressure and other management within 1 week

Figure 5.1. Flowchart of participants included in analyses



*Classified as 'possible' lacunar or non-lacunar stroke.

There was no difference regarding the alteplase dose given to patients with lacunar and non-lacunar AIS, but patients with lacunar AIS received alteplase treatment later than those with non-lacunar AIS (appendix 5.5). Most aspects of BP and other managements within 1 week of post-randomisation were different between lacunar and non-lacunar AIS, except time from symptom onset to treatment, antithrombotic agent use in the first 24 h and intravenous steroid administration. In terms of the use of BP lowering agents, intravenous labetalol, nitroglycerin, frusemide and other intravenous medications, and oral beta-blocker, were less likely to be prescribed in the management of lacunar AIS compared with non-lacunar AIS; but intravenous nicardipine was more likely to be administered in patients with lacunar AIS (appendix 5.6). The use of other BP lowering agents was comparable across lacunar and non-lacunar AIS. As shown in the plots of BP change over time, difference in mean SBP by treatment looks smaller in lacunar than non-lacunar AIS within 30 min post-randomisation but greater in lacunar AIS after 24 h (figure 5.2). However, there was no heterogeneity across the two subgroups in mean SBP difference by treatment at each time point up to 7 days (30 min, −3.1 [95% CI −5.9, −0.3] mmHg in lacunar stroke versus −4.6 [95% CI −6.6, −2.5] mmHg in non-lacunar stroke; 1 h,

−7.3 [−10.2, −4.4] mmHg versus −6.0 [−8.0, −3.9] mmHg; 24 h, −7.2 [−9.8, −4.5] mmHg versus −4.3 [−6.3, −2.4] mmHg) and over 24 h after randomisation (−5.5 [−6.5, −4.4] mmHg versus −5.6 [−6.3, −4.8] mmHg) (all $P_{interaction}$ >0.12) (table 5.2).

Table 5.1. Clinical characteristics of participants with lacunar and nonlacunar stroke

	Lacunar stroke (N=454)	Non-lacunar stroke (N=1178)	P value
Age (years)	64.1 (12.1)	69.0 (12.0)	**<0.001**
Female	174 (38.3)	461 (39.1)	0.76
Asian ethnicity	381 (83.9)	808 (68.6)	**<0.001**
Clinical features			
Systolic BP (mmHg)	165.3 (8.7)	165.2 (9.2)	0.83
Diastolic BP (mmHg)	92.1 (10.3)	90.6 (11.8)	**0.02**
Heart rate (beats per minute)	77.9 (12.5)	80.0 (16.3)	**0.01**
NIHSS score*	4 (3-6)	11 (7-15)	**<0.001**
GCS score†	15 (15-15)	14 (12-15)	**<0.001**
Medical history			
Previous stroke	66 (14.5)	233 (19.8)	**0.01**
Hypertension	314 (69.2)	859 (72.9)	0.13
Atrial fibrillation	14 (3.1)	253/1176 (21.5)	**<0.001**
Coronary artery disease	31 (6.8)	213 (18.1)	**<0.001**
Valvular or other heart disease	6 (1.3)	76 (6.5)	**<0.001**
Diabetes mellitus	88 (19.4)	283 (24.0)	**0.05**
Hypercholesterolemia	29 (6.4)	164 (13.9)	**<0.001**
Current smoker	98 (21.6)	232/1176 (19.7)	0.40
Pre-stroke function without symptoms (mRS=0)	43 (9.5)	208/1177 (17.7)	**<0.001**
Medication on admission			
Antihypertensive agent(s)	188 (41.4)	589 (50.0)	**0.002**
Warfarin anticoagulation	0 (0.0)	25 (2.1)	**0.002**
Aspirin/other antiplatelet agent	65 (14.3)	231 (19.6)	**0.01**
Statin/other lipid lowering agent	43 (9.5)	202 (17.1)	**<0.001**
Site reported LVO or assessed centrally	0 (0.0)	211/1177 (17.9)	**<0.001**
Time from stroke onset to randomisation (hrs)	3.5 (2.7-4.1)	3.2 (2.5-4.0)	**0.001**
Assigned to low-dose alteplase group	88 (19.4)	270 (22.9)	0.12
Assigned to standard-dose alteplase group	96 (21.1)	273 (23.2)	0.38
Final clinical diagnosis			
Non-stroke	2 (0.4)	3/1162 (0.3)	0.55
Large artery occlusion because of atheroma	39 (8.6)	644/1162 (55.4)	**<0.001**
Small vessel disease	396 (87.2)	106/1162 (9.1)	**<0.001**
Cardioembolism	2 (0.4)	245/1162 (21.1)	**<0.001**
Dissection	0 (0.0)	5/1162 (0.4)	0.16
Other or uncertain pathogenesis	15 (3.3)	159/1162 (13.7)	**<0.001**

BP denotes blood pressure; GCS, Glasgow Coma Scale; LVO, large vessel occlusion; mRS, modified Rankin scale; NIHSS, National Institutes of Health stroke scale.
Data are n (%), mean (SD), or median (Q1, Q3). P values are based on Chi-square, analysis of variance, or Wilcoxon signed-rank test.
*Scores on the NIHSS range from 0 to 42, with higher scores indicating more severe neurological deficits.
†Scores on the GCS range from 15 (normal) to 3 (deep coma).

5.5.3 Lacunar AIS and intensive BP reduction

The overall treatment effect of intensive BP reduction versus guideline-recommended BP management on functional, safety and other clinical outcomes was comparable to the main

results of the ENCHANTED trial. There was no heterogeneity of treatment effect on primary and other outcomes across the subgroups of lacunar and non-lacunar AIS, after adjusting for baseline covariables (all $P_{interaction}$ >0.19) (Figures 5.3 and 5.4).

Table 5.2. Mean SBP difference (mmHg) up to 7 days by randomisation in lacunar and nonlacunar stroke

Time point or period	Intensive BP lowering		Guideline BP lowering		Least-square mean difference (95% CI)*	P INT*
	n	Mean (SD)	n	Mean (SD)		
Randomisation						
Lacunar stroke	238	165.5 (8.9)	216	165.2 (8.5)	0.3 (-1.3, 1.9)	0.79
Non-lacunar stroke	570	165.3 (9.3)	608	165.2 (9.2)	0.1 (-1.0, 1.1)	
15 minutes						
Lacunar stroke	236	155.8 (15.4)	216	156.8 (15.9)	-1.0 (-3.9, 1.9)	0.54
Non-lacunar stroke	557	156.1 (17.1)	596	158.2 (17.7)	-2.1 (-4.1, -0.1)	
30 minutes						
Lacunar stroke	235	150.4 (15.5)	216	153.4 (14.7)	-3.1 (-5.9, -0.3)	0.42
Non-lacunar stroke	559	152.3 (17.1)	588	156.9 (18.1)	-4.6 (-6.6, -2.5)	
45 minutes						
Lacunar stroke	235	147.6 (15.2)	214	151.8 (16.3)	-4.3 (-7.2, -1.3)	0.88
Non-lacunar stroke	554	149.8 (16.9)	589	154.4 (17.8)	-4.5 (-6.6, -2.5)	
1 hour						
Lacunar stroke	238	144.5 (15.1)	216	151.8 (16.4)	-7.3 (-10.2, -4.4)	0.48
Non-lacunar stroke	561	147.4 (17.9)	592	153.4 (17.5)	-6.0 (-8.0, -3.9)	
6 hours						
Lacunar stroke	238	137.4 (13.3)	215	144.1 (15.2)	-6.7 (-9.4, -4.1)	0.12
Non-lacunar stroke	560	138.4 (15.1)	597	148.0 (18.4)	-9.5 (-11.5, -7.6)	
12 hours						
Lacunar stroke	238	136.6 (12.2)	215	143.9 (16.7)	-7.3 (-10.0, -4.6)	0.72
Non-lacunar stroke	560	137.9 (15.4)	593	144.6 (17.2)	-6.7 (-8.6, -4.8)	
18 hours						
Lacunar stroke	238	136.6 (13.6)	215	142.3 (15.7)	-5.6 (-8.3, -2.9)	0.65
Non-lacunar stroke	555	138.3 (15.4)	588	144.8 (17.8)	-6.5 (-8.4, -4.5)	
24 hours						
Lacunar stroke	238	136.7 (12.7)	212	143.8 (16.0)	-7.2 (-9.8, -4.5)	0.12
Non-lacunar stroke	547	140.4 (15.6)	586	144.8 (18.2)	-4.3 (-6.3, -2.4)	
Day 3						
Lacunar stroke	230	137.9 (12.9)	206	145.6 (15.0)	-7.7 (-10.3, -5.1)	0.12
Non-lacunar stroke	536	138.6 (14.7)	575	143.6 (16.8)	-5.0 (-6.9, -3.1)	
Day 5						
Lacunar stroke	205	138.9 (13.3)	182	143.0 (15.5)	-4.1 (-7.0, -1.2)	0.69
Non-lacunar stroke	481	137.7 (14.3)	505	141.1 (17.5)	-3.4 (-5.4, -1.4)	
Day 7						
Lacunar stroke	183	137.0 (12.5)	159	142.0 (14.6)	-5.0 (-7.9, -2.1)	0.84
Non-lacunar stroke	437	135.9 (14.7)	462	140.5 (16.7)	-4.6 (-6.7, -2.6)	
Over 24 hours after randomisation						
Lacunar stroke	1896†	143.2 (15.8)	1719†	148.5 (16.7)	-5.5 (-6.5, -4.4)‡	0.93‡
Non-lacunar stroke	4453†	145.1 (17.7)	4729†	150.6 (18.6)	-5.6 (-6.3, -4.8)‡	

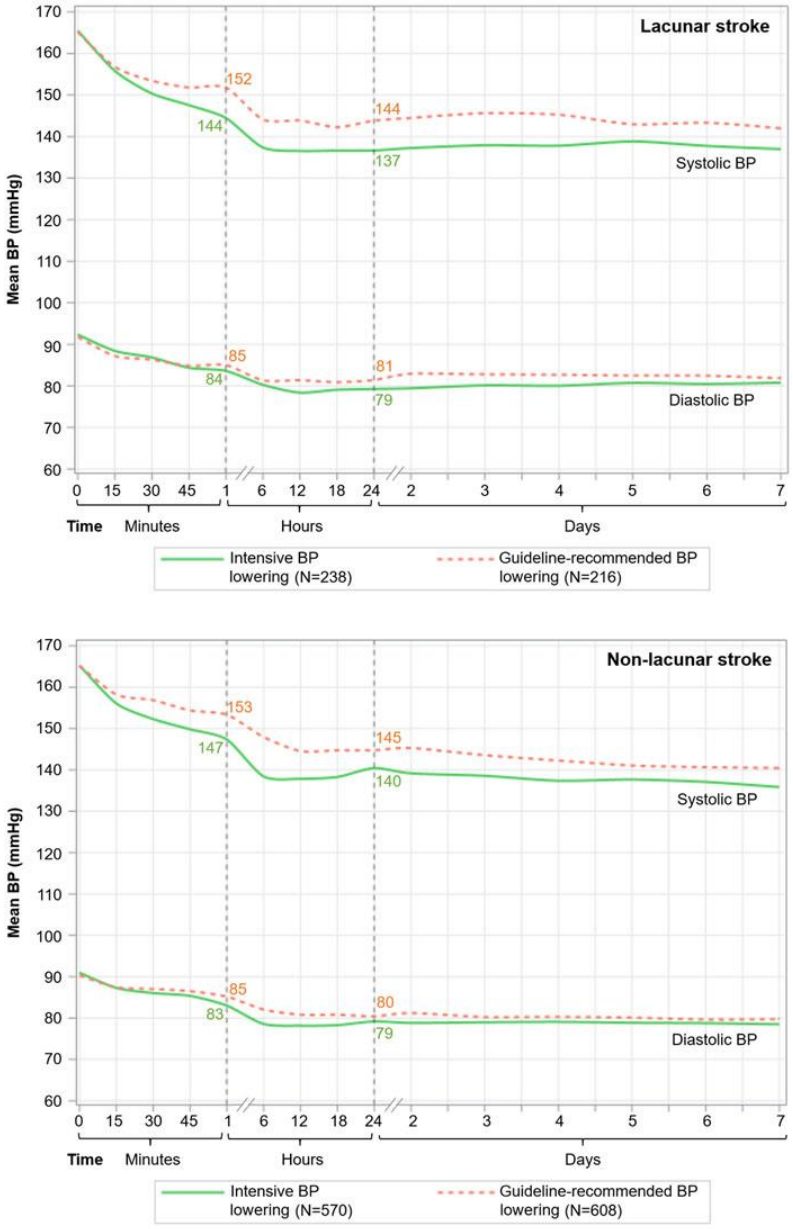
BP denotes blood pressure; P-INT, P value for interaction; SBP, systolic BP.

*The mean treatment difference of intensive compared to guideline-recommended BP lowering in the least squares means and associated 95% CIs were estimated from the generalised linear model. P values for interaction across subgroups of lacunar and non-lacunar stroke were estimated from the interaction item of treatment×subgroup after adding items of subgroup and treatment×subgroup into the model.

†Number of SBP records over 24 hours after randomisation.

‡Adding to adjust SBP at randomisation as a covariable in the model.

Figure 5.2. Mean systolic and diastolic blood pressure (mmHg) from randomisation to day 7 by treatment in lacunar and nonlacunar stroke

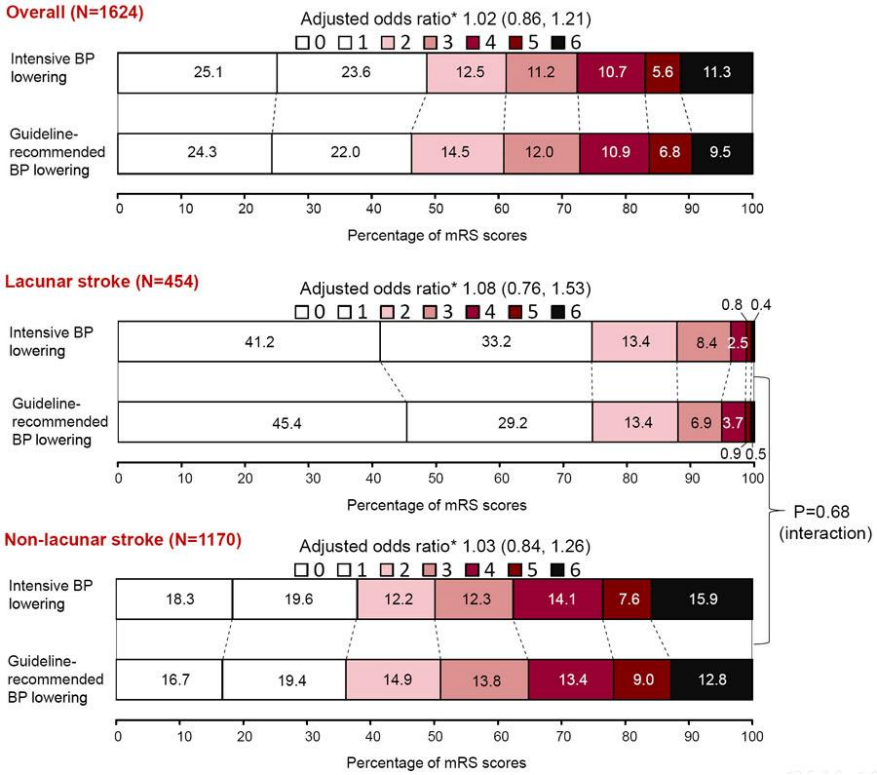


BP denotes blood pressure.

In the lacunar AIS subgroup, there was no significant difference on any outcomes by randomisation, although point estimates of intensive BP lowering effect were consistently above unity for any intracranial haemorrhage (adjusted OR 1.59, 95% CI 0.65-3.89), any adjudicated ICH (1.57, 0.57-4.32) and neurological deterioration or death within 24 h (1.40, 0.60-3.25). Comparable baseline characteristics and results were seen in sensitivity analyses for definite lacunar and non-lacunar AIS, except an increased risk of death by intensive BP lowering versus guideline recommendation in non-lacunar AIS (1.53, 1.01-2.32) (appendices

5.7 to 5.9). There was also no difference in all outcomes by randomisation in lacunar AIS participants with infarct size ≤15 mm on 24-h follow-up images (appendix 5.10).

Figure 5.3. Randomised treatment effects on ordinal mRS score in overall included participants and in the subgroups of lacunar and nonlacunar stroke



*Adjusted for key prognostic covariates (age; sex; ethnicity; baseline NIHSS score; time from stroke onset to randomisation; pre-morbid function [mRS scores 0 or 1]; prior use of antithrombotic agents [aspirin, other antiplatelet agents or warfarin]; history of diabetes or cardiovascular disease [stroke, atrial fibrillation, coronary artery disease, valvular or other heart disease]; assigned to low-dose alteplase group).

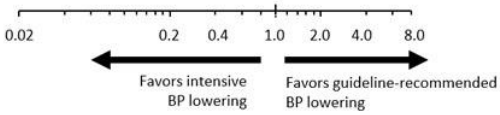
5.6 Discussion and conclusion

5.6.1 Discussion

In these post-hoc analyses of the ENCHANTED trial, the neutral effects of intensive versus guideline-recommended BP lowering on functional outcome and a significant beneficial effect of intensive BP lowering on intracranial haemorrhage in AIS were comparable across lacunar and non-lacunar stroke. These findings were observed on top of baseline characteristics, BP change from baseline to 7 days, and other aspects of acute management across the two subgroups. Overall, the results of the ENCHANTED BP arm did not support a major shift towards the intensive BP lowering for patients with mild-to-moderate AIS who received alteplase, but they left open the potential for variability in the effects across pathological subtypes, especially lacunar AIS.

Figure 5.4. Thrombolysis outcomes in participants with lacunar and non-lacunar stroke by randomised treatment of BP reduction

Thrombolysis outcomes	BP lowering group, n/N (%) Intensive	Guideline	Odds ratio (95% CI)	P inter-action
mRS 2-6 (primary): overall	413/805 (51.3)	440/819 (53.7)	0.92 (0.74, 1.14)*	
Lacunar stroke	61/238 (25.6)	55/216 (25.5)	0.98 (0.63, 1.53)*	0.67*
Non-lacunar stroke	352/567 (62.1)	385/603 (63.8)	0.92 (0.71, 1.18)*	
mRS 3-6: overall	312/805 (38.8)	321/819 (39.2)	1.01 (0.81, 1.26)*	
Lacunar stroke	29/238 (12.2)	26/216 (12.0)	1.00 (0.55, 1.81)*	0.97*
Non-lacunar stroke	283/567 (49.9)	295/603 (48.9)	1.04 (0.82, 1.33)*	
mRS 6 (death): overall	91/808 (11.3)	78/824 (9.5)	1.21 (0.88, 1.67)	
Lacunar stroke	1/238 (0.4)	1/216 (0.5)	0.91 (0.06, 14.59)	0.80
Non-lacunar stroke	90/570 (15.8)	77/608 (12.7)	1.29 (0.93, 1.80)	
sICH (SITS-MOST): overall	12/808 (1.5)	18/824 (2.2)	0.68 (0.32, 1.41)	
Lacunar stroke	1/238 (0.4)	0/216 (0.0)	—	—
Non-lacunar stroke	11/570 (1.9)	18/608 (3.0)	0.65 (0.30, 1.38)	
Adjudicated ICH: overall	125/808 (15.5)	153/824 (18.6)	0.79 (0.60, 1.03)†	
Lacunar stroke	10/238 (4.2)	7/216 (3.2)	1.57 (0.57, 4.32)†	0.32†
Non-lacunar stroke	115/570 (20.2)	146/608 (24.0)	0.77 (0.58, 1.03)†	
Intracranial hemorrhage‡: overall	140/808 (17.3)	178/824 (21.6)	0.74 (0.57, 0.96)†	
Lacunar stroke	13/238 (5.5)	9/216 (4.2)	1.59 (0.65, 3.89)†	0.19†
Non-lacunar stroke	127/570 (22.3)	169/608 (27.8)	0.72 (0.55, 0.94)†	
END or death within 24 hrs: overall	92/808 (11.4)	87/824 (10.6)	1.11 (0.82, 1.52)†	
Lacunar stroke	14/238 (5.9)	10/216 (4.6)	1.40 (0.60, 3.25)†	0.67†
Non-lacunar stroke	78/570 (13.7)	77/608 (12.7)	1.12 (0.80, 1.57)†	
END or death within 72 hrs: overall	125/808 (15.5)	113/824 (13.7)	1.18 (0.89, 1.56)†	
Lacunar stroke	15/238 (6.3)	14/216 (6.5)	1.06 (0.49, 2.27)†	0.61†
Non-lacunar stroke	110/570 (19.3)	99/608 (16.3)	1.25 (0.93, 1.70)†	



END denotes early neurological deterioration; ICH, intracerebral haemorrhage; mRS, modified Rankin scale; sICH, symptomatic intracerebral haemorrhage; SITS-MOST, the Safe Implementation of Thrombolysis in Stroke Monitoring Study.
*Adjusted for key prognostic covariates (age; sex; ethnicity; baseline NIHSS score; time from stroke onset to randomisation; pre-morbid function [mRS scores 0 or 1]; prior use of antithrombotic agents [aspirin, other antiplatelet agents or warfarin]; history of diabetes or cardiovascular disease [stroke, atrial fibrillation, coronary artery disease, valvular or other heart disease]; assigned to low-dose alteplase group) for functional outcomes. †Adjusted for minimization and key prognostic covariates (age, baseline NIHSS score, time from stroke onset to randomisation, and assigned to low-dose alteplase group) for safety outcomes, and neurological deterioration within 24 or 72 h.
‡Site reported or adjudicated centrally.

Evidence of the effects of BP management for lacunar AIS is mainly derived from studies of secondary prevention where, for example, the Secondary Prevention of Small Subcortical Strokes (SPS3) trial suggested reduced recurrent stroke from long-term intensive BP lowering (systolic <130 mmHg) in patients with recent lacunar AIS.[20] To our knowledge, there is no

dedicated clinical trial that has explored the association of BP management in the acute or subacute phase of lacunar AIS. A post hoc analysis of the Scandinavian Candesartan Acute Stroke Trial (SCAST) suggested variable effects of BP lowering treatment (by candesartan versus placebo for 1 week after randomisation) on functional outcome at 6 months across different AIS subtypes of hypertensive (SBP ≥140 mmHg) patients who presented within 30 h of onset[8] which was not seen in our ENCHANTED data regardless of the AIS classification by TOAST or combination of clinical and adjudicated imaging findings. These conflicting results might reflect many differences between SCAST and ENCHANTED in terms of trial design, including eligible participants, use of BP lowering agents, start time of BP lowering, administration of alteplase and the time point of outcome assessment. Two other recent trials, Efficacy of Nitric Oxide in Stroke (ENOS) and phase 3 Rapid Intervention with Glyceryl Trinitrate in Hypertensive Stroke Trial (RIGHT-2), also showed that the neutral effect of early BP lowering by transdermal glyceryl trinitrate was comparable across AIS subtypes.[21-23]

Contrary to prior reports,[4,24] no significant differences in baseline SBP and history of hypertension were seen between lacunar and non-lacunar AIS. However, giving priority to adjudicated imaging findings in our pre-specified definition of lacunar AIS might have improved upon limitations of TOAST in including risk factors of hypertension or diabetes mellitus as one of the criteria.[25] Apart from baseline characteristics, it was shown that BP difference by treatment over 1 week was comparable across lacunar and non-lacunar AIS patients, which increases the reliability of our results regarding the treatment effects of early intensive BP lowering on safety and functional outcomes across subgroups. Although there was no differential treatment effect, it is interesting to see numerically more events of intracranial haemorrhage and adjudicated ICH after early intensive compared with guideline-recommended BP lowering in the lacunar AIS subgroup. Haemorrhage is one of the pathological features during the process of lacune formation with underlying mechanisms of endothelial dysfunction and blood-brain barrier disruption.[26] It seems unlikely that intensive BP lowering would increase the risk of ICH for lacunar AIS after thrombolysis given the lack of collateral circulation to small penetrating arterioles. However, whether a dramatic BP reduction in acute phase may aggravate the damage to fragile arterioles and increase blood-brain permeability is unclear. Given the small number of haemorrhage events, the safety or efficacy of intensive BP lowering in the lacunar AIS subgroup could not be reliably demonstrated.

5.6.2 Strengths and limitations

This study is strengthened by the inclusion of patients from a large, prospective, multicentre, clinical trial with systematic and high quality of data collection, and the ability to adjust for multiple covariables in statistical models. The BP data from randomisation to day 7 were recorded frequently, the imaging assessment was rigorous and based upon IST-3 methodology, and our pre-specified definitions of lacunar and non-lacunar AIS used a combination of clinical and adjudicated imaging information to reduce any misclassification. However, there are limitations that include an insufficient sample of lacunar AIS that limited the statistical precision of the estimated treatment effect, and there is some selection bias as most ENCHANTED participants were from Asia with mild-to-moderate severity of stroke. The actual SBP difference over 24 h after baseline (Δ5.5 mmHg) by randomised treatment was smaller than envisaged, raising the question of whether the treatment effect might be different with greater BP lowering. Baseline magnetic resonance imaging was not requested in our pragmatic trial design and there were challenges in identifying lacunar AIS through comparison of baseline and follow-up images. Finally, the sample size was further compromised as approximately one-quarter of the ENCHANTED BP arm participants were excluded due to insufficient confidence in discriminating lacunar and non-lacunar AIS, although post hoc analyses with all participants with possible lacunar and non-lacunar AIS showed comparable results (data available on request).

5.6.3 Conclusion

In summary, these secondary analyses of the ENCHANTED dataset extend knowledge in relation to early BP management for lacunar AIS, by showing comparable neutral effects of intensive versus guideline-recommended BP lowering on functional outcomes and comparable beneficial effects on intracranial haemorrhage across lacunar and non-lacunar AIS.

References

1. Appleton JP, Sprigg N, Bath PM. Blood pressure management in acute stroke. Stroke Vasc Neurol 2016;1(2):72-82.
2. Ahmed N, Wahlgren N, Brainin M, et al. Relationship of blood pressure, antihypertensive therapy, and outcome in ischaemic stroke treated with intravenous thrombolysis: retrospective analysis from Safe Implementation of Thrombolysis in Stroke-International Stroke Thrombolysis Register (SITS-ISTR). Stroke 2009; 40(7): 2442-9.
3. Anderson CS, Huang Y, Lindley RI, et al. Intensive blood pressure reduction with intravenous thrombolysis therapy for acute ischaemic stroke (ENCHANTED): an international, randomised, open-label, blinded-endpoint, phase 3 trial. Lancet 2019; 393(10174):877-88.
4. Altmann M, Thommessen B, Rønning OM, et al. Blood pressure differences between patients with lacunar and nonlacunar infarcts. Brain Behav. 2015;5(8):e00353.
5. Li L, Welch SJV, Gutnikov SA, et al. Time Course of Blood Pressure Control Prior to Lacunar TIA and Stroke: Population-based Study. Neurology 2018;90(20):e1732-41.
6. Toyoda K, Okada Y, Fujimoto S, et al. Blood Pressure Changes During the Initial Week After Different Subtypes of Ischaemic Stroke. Stroke 2006;37(10):2637-9.
7. Ntaios G, Lambrou D, Michel P. Blood Pressure Changes in Acute Ischaemic Stroke and Outcome With Respect to Stroke Etiology. Neurology 2012;79(14):1440-8.
8. Sandset EC, Jusufovic M, Sandset PM, et al. Effects of blood pressure-lowering treatment in different subtypes of acute ischaemic stroke. Stroke 2015;46(3):877-9.
9. Petersen NH, Ortega-Gutierrez S, Wang A, et al. Decreases in blood pressure during thrombectomy are associated with larger infarct volumes and worse functional outcome. Stroke 2019;50(7):1797-804.
10. Minhas JS, Wang X, Lindley RI, et al. Comparative effects of intensive-blood pressure versus standard-blood pressure-lowering treatment in patients with severe ischaemic stroke in the ENCHANTED trial. J Hypertens 2021;39(2):280-5.
11. Landau WM, Nassief A. Editorial Comment--Time to Burn the TOAST. Stroke 2005;36(4):902-4.
12. Selvarajah JR, Glaves M, Wainwright J, et al. Classification of minor stroke: intra- and inter-observer reliability. Cerebrovasc Dis 2009;27(3):209-14.
13. Yang XL, Zhu DS, Lv HH, et al. Etiological Classification of Cerebral Ischaemic Stroke by the TOAST, SSS-TOAST, and ASCOD Systems: The Impact of Observer's Experience on Reliability. Neurologist 2019;24(4):111-4.
14. Huang Y, Sharma VK, Robinson T, et al. Rationale, design, and progress of the ENhanced Control of Hypertension ANd Thrombolysis strokE stuDy (ENCHANTED) trial: an international multicentre 2×2 quasi-factorial randomised controlled trial of low- vs. standard-dose rt-PA and early intensive vs. guideline-recommended blood pressure lowering in patients with acute ischaemic stroke eligible for thrombolysis treatment. Int J Stroke 2015;10(5):778-88.
15. Anderson CS, Woodward M, Arima H, et al. Statistical analysis plan for evaluating different intensities of blood pressure control in the ENhanced Control of Hypertension And Thrombolysis strokE stuDy. Int J Stroke 2019;14(5):555-8.

16. Delcourt C, Wang X, Zhou Z, et al. Brain imaging abnormalities and outcome after acute ischaemic stroke: the ENCHANTED trial. J Neurol Neurosurg Psychiatry. 2020;91(12):1290-6.
17. The IST-3 collaborative group. Association between brain imaging signs, early and late outcomes, and response to intravenous alteplase after acute ischaemic stroke in the third International Stroke Trial (IST-3): secondary analysis of a randomised controlled trial. Lancet Neurol 2015;14(5):485-96.
18. Mair G, von Kummer R, Adami A, et al. Arterial Obstruction on Computed Tomographic or Magnetic Resonance Angiography and Response to Intravenous Thrombolytics in Ischaemic Stroke. Stroke 2017;48(2):353-60.
19. Wahlgren N, Ahmed N, Davalos S, et al. Thrombolysis with alteplase for acute ischaemic stroke in the Safe Implementation of Thrombolysis in Stroke-Monitoring Study (SITS-MOST): an observational study. Lancet 2007;369(9558):275-82.
20. Benavente OR, Coffey CS, Conwit R, et al. Blood-pressure targets in patients with recent lacunar stroke: the SPS3 randomised trial. Lancet 2013;382(9891):507-15.
21. ENOS Trial Investigators. Efficacy of nitric oxide, with or without continuing antihypertensive treatment, for management of high blood pressure in acute stroke (ENOS): a partial-factorial randomised controlled trial. Lancet 2015;385(9968):617-28.
22. Woodhouse L, Scutt P, Krishnan K, et al. Effect of Hyperacute Administration (Within 6 Hours) of Transdermal Glyceryl Trinitrate, a Nitric Oxide Donor, on Outcome After Stroke: Subgroup Analysis of the Efficacy of Nitric Oxide in Stroke (ENOS) Trial. Stroke 2015;46(11):3194-201.
23. RIGHT-2 Investigators. Prehospital Transdermal Glyceryl Trinitrate in Patients With Ultra-Acute Presumed Stroke (RIGHT-2): An Ambulance-Based, Randomised, Sham-Controlled, Blinded, Phase 3 Trial. Lancet 2019;393(10175):1009-20.
24. Arba F, Mair G, Phillips S, et al. Improving Clinical Detection of Acute Lacunar Stroke: Analysis From the IST-3. Stroke 2020;51(5):1411-8.
25. Jackson C, Sudlow C. Are Lacunar Strokes Really Different? A Systematic Review of Differences in Risk Factor Profiles Between Lacunar and Nonlacunar Infarcts. Stroke 2005;36(4):891-901.
26. Regenhardt RW, Das AS, Lo EH, Caplan LR. Advances in Understanding the Pathophysiology of Lacunar Stroke: A Review. JAMA Neurol 2018;75(10):1273-81.

Appendices

Appendix 5.1. Definitions of symptomatic intracerebral haemorrhage

For intracerebral haemorrhage, bleeding was coded as HI1 (small petechiae along infarct margins), HI2 (confluent petechiae within infarcted area without space-occupying effect), PH1 (blood clot(s) in <30% of infarcted area with slight space-occupying effect) and PH2 (blood clot(s) in >30% of infarcted area with substantial space-occupying effect). In addition, independent assessors were asked to adjudicate if haemorrhage was the predominant cause of neurological worsening, and if there was evidence of midline shift. These assessments enabled the following definitions of symptomatic intracerebral haemorrhage (sICH) to be adjudicated: Safe Implementation of Thrombolysis in Stroke Monitoring Study (SITS-MOST): large or remote parenchymal ICH (type 2, defined as greater than 30% of the infarcted area affected by haemorrhage with mass effect or extension outside the infarct) combined with neurological deterioration (>4 points on the NIHSS) or leading to death within 24 to 36 hours [Wahlgren et al, 2007]; any ICH associated with neurological deterioration (>1 point change in NIHSS score) from baseline or death within 24 to 36 hours (NINDS) [NINDS Study Group, 1995]; any ICH with neurological deterioration (>4 points on the NIHSS) from baseline or death within 24 to 36 hours (ECASS2) [Hacke et al, 1998]; any ICH with neurological deterioration (>4 points increase on the NIHSS) from baseline or death within 36 hours (ECASS3) [Hacke et al, 2008]; either significant ICH (local or distant from the infarct) or significant haemorrhagic transformation of an infarct on brain imaging with clinically significant deterioration or death within the first 7 days of treatment (IST3) [IST-3 Collaborative Group, 2012]; and fatal ICH, any type 2 parenchymal ICH and death within 7 days.

References

Wahlgren N, Ahmed N, Davalos S, et al. Thrombolysis with alteplase for acute ischaemic stroke in the Safe Implementation of Thrombolysis in Stroke-Monitoring Study (SITS-MOST): an observational study Lancet 2007; 369: 275-282.

The National Institute of Neurological Disorders and Stroke rt-PA Stroke Study Group. Tissue plasminogen activator for acute ischaemic stroke. N Engl J Med 1995; 333:1581-1587.

Hacke W, Kaste M, Fieschi C, et al. Randomised double-blind placebo controlled trial of thrombolytic therapy with intravenous alteplase in acute ischaemic stroke (ECASS II). Lancet 1998; 352: 1245-1251.

Hacke W, Kaste M, Bluhmki E, et al. Thrombolysis with alteplase 3 to 4.5 hours after acute ischaemic stroke. N Engl J Med 2008; 359: 1317-1329.

The IST-3 Collaborative Group. The benefits and harms of intravenous thrombolysis with recombinant tissue plasminogen activator within 6 h of acute ischaemic stroke (the third international stroke trial [IST-3]): a randomised controlled trial. Lancet 2012; 379:2352-2363.

Appendix 5.2. Definitions of lacunar and nonlacunar stroke

1. Definite lacunar stroke
Meet all following 4 points
(1) Subcortical or brainstem lesions (maximum diameter ≤20 mm) in the territory of penetrating arteries, with a rounded, ovoid, or tubular shape on axial CT or diffusion-weighted imaging (DWI)/apparent diffusion coefficient (ADC) map. For participants without MRI at baseline or 24 hours follow-up, baseline and follow-up CT scans need to be compared so as to exclude old lesion with no change in lesion size/shape and attenuation between baseline and follow-up scans.
(2) No large vessel occlusion (LVO)* on cerebral angiography assessed centrally or reported by site investigators.
(3) The final diagnosis by site investigators is "Small vessel or perforating vessel 'lacunar' disease" according to the TOAST (Trial of ORG 10172 in Acute Stroke Treatment) classification.
(4) Side of infarct lesion assessed on brain images centrally is consistent with that reported by site investigators.

2. Probable lacunar stroke (criterion 1 or 2 or 3)
Criterion 1: Meet the criteria of definite lacunar stroke except the consistency of infarct side assessed centrally on brain images and that reported by site investigators

Criterion 2:
Meet all following 4 points
(1) Subcortical or brainstem lesions (maximum diameter ≤20 mm) in the territory of penetrating arteries, with a rounded, ovoid, or tubular shape on axial CT or diffusion-weighted imaging (DWI)/apparent diffusion coefficient (ADC) map. For participants without MRI at baseline or 24 hours follow-up, baseline and follow-up CT scans need to be compared so as to exclude old lesion with no change in lesion size/shape and attenuation between baseline and follow-up scans.
(2) No large vessel occlusion (LVO) on cerebral angiography assessed centrally or reported by site investigators.
(3) The final diagnosis by site investigators is **NOT** "Small vessel or perforating vessel 'lacunar' disease" according to the TOAST classification.
(4) Baseline National Institutes of Health stroke scale (NIHSS) scores <7.

Criterion 3:
Meet all following 4 points
(1) No acute infarct lesion identified on brain images or the brain images were not collected at the George Institute.
(2) No LVO on cerebral angiography assessed centrally or reported by site investigators.
(3) The final diagnosis by site investigators is "Small vessel or perforating vessel 'lacunar' disease" according to the TOAST classification.
(4) Baseline NIHSS scores <7.

3. Definite non-lacunar stroke
Acute infarct lesion with maximum diameter >20 mm or LVO identified on brain images. For participants without MRI at baseline or 24 hours follow-up, baseline and follow-up CT scans need to be compared so as to exclude old lesion with no change in lesion size/shape and attenuation between baseline and follow-up scans.

4. Probable non-lacunar stroke
Meet all following 3 points
(1) No acute infarct lesion identified on brain images or the brain images were not collected at the George Institute.
(2) With LVO on cerebral angiography reported by site investigators or the final diagnoses by site investigators are "Large artery occlusion because of atheroma", "Cardio-emboli", "Dissection", "Other definite pathological mechanism", or "Uncertain aetiology" according to the TOAST classification.
(3) Baseline NIHSS scores ≥7.

5. Possible lacunar stroke
Meet all following 3 points
(1) Does not meet the criteria of definite and probable lacunar or non-lacunar stroke.
(2) The final diagnosis by site investigators is "Small vessel or perforating vessel 'lacunar' disease" according to the TOAST classification.
(3) No LVO reported by site investigators.

6. Possible non-lacunar stroke
Meet all following 2 points
(1) Does not meet the criteria of definite and probable lacunar or non-lacunar stroke.
(2) The final diagnosis by site investigators is **NOT** "Small vessel or perforating vessel 'lacunar' disease" according to the TOAST classification, **OR** there is LVO reported by site investigators (including unknown LVO status).

*LVO assessed centrally on brain CTA or MRA: >50% stenosis (IST-3 Angiography Score for stenosis/occlusion) at anterior cerebral artery, M1 and M2 segment of middle cerebral artery, posterior cerebral artery, basilar artery, vertebral artery, and intracranial and extracranial Internal carotid artery.

References
Adams HP Jr, Bendixen BH, Kappelle LJ, et al. Classification of subtype of acute ischaemic stroke. Definitions for use in a multicentre clinical trial. TOAST. Trial of Org 10172 in Acute Stroke Treatment. Stroke. 1993;24(1):35-41.

Arba F, Mair G, Phillips S, et al. Improving Clinical Detection of Acute Lacunar Stroke: Analysis From the IST-3. Stroke. 2020;51(5):1411-1418.

Heldner MR, Hsieh K, Broeg-Morvay A, et al. Clinical prediction of large vessel occlusion in anterior circulation stroke: mission impossible? J Neurol. 2016;263(8):1633-40.

Appendix 5.3. Baseline characteristics of ENCHANTED BP arm participants included and excluded for analyses in this study

	Include (N=1632)	Exclude (N=564)	P value
Age (years)	67.6 (12.2)	64.8 (11.9)	**<0.001**
≥ 80 years	259 (15.9)	60 (10.6)	**0.002**
Female	635 (38.9)	200 (35.5)	0.15
Asian ethnicity	1189 (72.9)	429/562 (76.3)	0.11
Clinical features			
Systolic BP (mmHg)	165.3 (9.1)	165.3 (9.5)	0.97
Diastolic BP (mmHg)	91.1 (11.5)	90.6 (11.4)	0.45
Heart rate (beats per minute)	79.4 (15.3)	79.0 (13.1)	0.53
NIHSS score*	8 (5-13)	6 (4-9)	**<0.001**
NIHSS score ≥14	396 (24.3)	41 (7.3)	**<0.001**
GCS score†	15 (13-15)	15 (14-15)	**<0.001**
Medical history			
Previous stroke	299 (18.3)	115 (20.4)	0.28
Hypertension	1173 (71.9)	395/560 (70.5)	0.54
Atrial fibrillation	267/1630 (16.4)	45/560 (8.0)	**<0.001**
Coronary artery disease	244 (15.0)	65/560 (11.6)	**0.05**
Valvular or other heart disease	82 (5.0)	12/560 (2.1)	**0.004**
Diabetes mellitus	371 (22.7)	125/560 (22.3)	0.84
Hypercholesterolemia	193 (11.8)	56/560 (10.0)	0.24
Current smoker	330/1630 (20.2)	114/560 (20.4)	0.95
Pre-stroke function without symptoms (mRS=0)	251/1631 (15.4)	63/560 (11.3)	**0.02**
Medication on admission			
Antihypertensive agent(s)	777 (47.6)	235/560 (42.0)	**0.02**
Warfarin anticoagulation	25 (1.5)	4/560 (0.7)	0.14
Aspirin/other antiplatelet agent	296 (18.1)	90/560 (16.1)	0.27
Statin/other lipid lowering agent	245 (15.0)	93 (16.6)	0.37
Time from onset to randomisation (hrs)	3.3 (2.5-4.1)	3.4 (2.7-4.2)	**0.03**
Large vessel occlusion‡	211/1631 (12.9)	32/558 (5.7)	**<0.001**
Randomised intensive BP lowering treatment	808 (49.5)	273 (48.4)	0.65
Assigned to low-dose alteplase group	358 (21.9)	98 (17.4)	**0.02**
Assigned to standard-dose alteplase group	369 (22.6)	100 (17.7)	**0.01**
Presumed diagnosis at time of hospital separation			
Non-stroke	5/1616 (0.3)	28/544 (5.1)	**<0.001**
Large artery occlusion due to significant atheroma	683/1616 (42.3)	269/544 (49.4)	**0.004**
Small vessel disease	502/1616 (31.1)	121/544 (22.2)	**<0.001**
Cardioembolism	247/1616 (15.3)	42/544 (7.7)	**<0.001**
Dissection	5/1616 (0.3)	2/544 (0.4)	0.84
Other or uncertain pathogenesis	174/1616 (10.8)	82/544 (15.1)	**0.007**

BP denotes blood pressure; GCS, Glasgow coma scale; mRS, modified Rankin scale; NIHSS, National Institutes of Health stroke scale.

Data are n (%), mean (SD), or median (Q1, Q3). P values are based on Chi-square, analysis of variance, or Wilcoxon signed-rank test.

*Scores on the NIHSS range from 0 to 42, with higher scores indicating more severe neurological deficits.

†Scores on the GCS range from 15 (normal) to 3 (deep coma).

‡Reported by site investigators or assessed centrally on computed tomographic or magnetic resonance angiography.

Appendix 5.4. Baseline characteristics of participants with definite/probable lacunar and nonlacunar stroke

	Definite/probable lacunar stroke			Definite/probable non-lacunar stroke			P value[‖]
	Intensive BP (N=238)	Standard BP (N=216)	Total (N=454)	Intensive BP (N=570)	Standard BP (N=608)	Total (N=1178)	
Age (years)	63.4 (11.9)	64.9 (12.3)	64.1 (12.1)	68.7 (12.4)	69.2 (11.7)	69.0 (12.0)	**<0.001**
≥80 years	18 (7.6)	28 (13.0)	46 (10.1)	100 (17.5)	113 (18.6)	213 (18.1)	**<0.001**
Female	92 (38.7)	82 (38.0)	174 (38.3)	213 (37.4)	248 (40.8)	461 (39.1)	0.76
Asian ethnicity	202 (84.9)	179 (82.9)	381 (83.9)	386 (67.7)	422 (69.4)	808 (68.6)	**<0.001**
Clinical features							
Systolic BP (mmHg)	165.5 (8.9)	165.2 (8.5)	165.3 (8.7)	165.3 (9.3)	165.2 (9.2)	165.2 (9.2)	0.83
Diastolic BP (mmHg)	92.4 (10.6)	91.8 (10.0)	92.1 (10.3)	91.0 (11.7)	90.3 (12.0)	90.6 (11.8)	**0.02**
Heart rate (beats per minute)	79.0 (12.1)	76.7 (13.0)	77.9 (12.5)	79.7 (16.0)	80.3 (16.5)	80.0 (16.3)	**0.01**
NIHSS score[*]	5 (3-6)[¶]	4 (3-5)[¶]	4 (3-6)	11 (7-15)	10 (7-15)	11 (7-15)	**<0.001**
NIHSS score ≥14	3 (1.3)	1 (0.5)	4 (0.9)	194 (34.0)	198 (32.6)	392 (33.3)	**<0.001**
GCS score[†]	15 (15-15)	15 (15-15)	15 (15-15)	14 (12-15)	14 (12-15)	14 (12-15)	**<0.001**
Medical history							
Previous stroke	38 (16.0)	28 (13.0)	66 (14.5)	113 (19.8)	120 (19.7)	233 (19.8)	**0.01**
Hypertension	167 (70.2)	147 (68.1)	314 (69.2)	415 (72.8)	444 (73.0)	859 (72.9)	0.13
Atrial fibrillation	7 (2.9)	7 (3.2)	14 (3.1)	109 (19.1)	144/606 (23.8)	253/1176 (21.5)	**<0.001**
Coronary artery disease	16 (6.7)	15 (6.9)	31 (6.8)	105 (18.4)	108 (17.8)	213 (18.1)	**<0.001**
Valvular or other heart disease	4 (1.7)	2 (0.9)	6 (1.3)	33 (5.8)	43 (7.1)	76 (6.5)	**<0.001**
Diabetes mellitus	43 (18.1)	45 (20.8)	88 (19.4)	131 (23.0)	152 (25.0)	283 (24.0)	**0.05**
Hypercholesterolemia	15 (6.3)	14 (6.5)	29 (6.4)	77 (13.5)	87 (14.3)	164 (13.9)	**<0.001**
Current smoker	54 (22.7)	44 (20.4)	98 (21.6)	112/569 (19.7)	120/607 (19.8)	232/1176 (19.7)	0.40
Pre-stroke function without symptoms[‡]	21 (8.8)	22 (10.2)	43 (9.5)	104 (18.2)	104/607 (17.1)	208/1177 (17.7)	**<0.001**
Medication on admission							
Antihypertensive agent(s)	95 (39.9)	93 (43.1)	188 (41.4)	281 (49.3)	308 (50.7)	589 (50.0)	**0.002**
Warfarin anticoagulation	0 (0.0)	0 (0.0)	0 (0.0)	12 (2.1)	13 (2.1)	25 (2.1)	**0.002**
Aspirin/other antiplatelet agent	33 (13.9)	32 (14.8)	65 (14.3)	106 (18.6)	125 (20.6)	231 (19.6)	**0.01**
Statin/other lipid lowering agent	22 (9.2)	21 (9.7)	43 (9.5)	94 (16.5)	108 (17.8)	202 (17.1)	**<0.001**
Imaging features							
Infarct at left side	48/112 (42.9)	48/101 (47.5)	96/213 (45.1)	184/388 (47.4)	192/387 (49.6)	376/775 (48.5)	0.37
Infarct at right side	61/112 (54.5)	50/101 (49.5)	111/213 (52.1)	181/388 (46.6)	175/387 (45.2)	356/775 (45.9)	0.11
Infarct at midline or bilateral side	3/112 (2.7)	3/101 (3.0)	6/213 (2.8)	23/388 (5.9)	20/387 (5.2)	43/775 (5.5)	0.10
Infarct in anterior circulation only	87/112 (77.7)	75/101 (74.3)	162/213 (76.1)	274/388 (70.6)	279/387 (72.1)	553/775 (71.4)	0.17
Infarct in posterior circulation only	23/112 (20.5)	24/101 (23.8)	47/213 (22.1)	78/388 (20.1)	76/387 (19.6)	154/775 (19.9)	0.48
At both anterior and posterior circulation	2/112 (1.8)	2/101 (2.0)	4/213 (1.9)	36/388 (9.3)	32/387 (8.3)	68/775 (8.8)	**<0.001**
With hyperdense vessel sign on CT	2/161 (1.2)	4/156 (2.6)	6/317 (1.9)	107/440 (24.3)	107/443 (24.2)	214/883 (24.2)	**<0.001**
LVO on baseline CTA or MRA[§]	0/24 (0.0)	0/16 (0.0)	0/40 (0.0)	58/88 (65.9)	40/74 (54.1)	98/162 (60.5)	**<0.001**
With old vascular lesions	82/169 (48.5)	74/158 (46.8)	156/327 (47.7)	221/446 (49.6)	230/451 (51.0)	451/897 (50.3)	0.43
With brain atrophy	81/169 (47.9)	83/158 (52.5)	164/327 (50.2)	311/446 (69.7)	306/451 (67.8)	617/897 (68.8)	**<0.001**
With white matter changes	66/169 (39.1)	62/158 (39.2)	128/327 (39.1)	192/446 (43.0)	192/451 (42.6)	384/897 (42.8)	0.25
Site reported LVO or assessed centrally	0 (0.0)	0 (0.0)	0 (0.0)	114/570 (20.0)	97/607 (16.0)	211/1177 (17.9)	**<0.001**

Time from stroke onset to randomisation (hrs)	3.4 (2.6-4.1)	3.5 (2.7-4.2)	3.5 (2.7-4.1)	3.2 (2.4-4.0)	3.2 (2.6-4.0)	3.2 (2.5-4.0)	**0.001**
Assigned to low-dose alteplase group	47 (19.7)	41 (19.0)	88 (19.4)	127 (22.3)	143 (23.5)	270 (22.9)	0.12
Assigned to standard-dose alteplase group	48 (20.2)	48 (22.2)	96 (21.1)	135 (23.7)	138 (22.7)	273 (23.2)	0.38
Presumed diagnosis at time of hospital separation							
Non-stroke	1 (0.4)	1 (0.5)	2 (0.4)	2/565 (0.4)	1/597 (0.2)	3/1162 (0.3)	0.55
Large artery occlusion because of atheroma	20 (8.4)	19 (8.8)	39 (8.6)	307/565 (54.3)	337/597 (56.4)	644/1162 (55.4)	**<0.001**
Small vessel disease	212 (89.1)	184 (85.2)	396 (87.2)	59/565 (10.4)	47/597 (7.9)	106/1162 (9.1)	**<0.001**
Cardioembolism	1 (0.4)	1 (0.5)	2 (0.4)	117/565 (20.7)	128/597 (21.4)	245/1162 (21.1)	**<0.001**
Dissection	0 (0.0)	0 (0.0)	0 (0.0)	3/565 (0.5)	2/597 (0.3)	5/1162 (0.4)	0.16
Other or uncertain pathogenesis	4 (1.7)¶	11 (5.1)¶	15 (3.3)	77/565 (13.6)	82/597 (13.7)	159/1162 (13.7)	**<0.001**

BP denotes blood pressure; CTA, computed tomography angiography; GCS, Glasgow Coma Scale; LVO, large vessel occlusion; MRA, magnetic resonance angiography; mRS, modified Rankin scale; NIHSS, National Institutes of Health stroke scale.

Data are n (%), mean (SD), or median (Q1, Q3). The P values are based on Chi-square, analysis of variance, or Wilcoxon signed-rank test.

*Scores on the NIHSS range from 0 to 42, with higher scores indicating more severe neurological deficits.

†Scores on the GCS range from 15 (normal) to 3 (deep coma).

‡mRS=0.

§Scored as 0, 1, or 2 for proximal cerebral arteries (A1-2 segment of anterior cerebral artery, internal carotid artery, M1-2 segment of middle cerebral artery, P1-2 segment of posterior cerebral artery, or basilar artery) on raw or reconstructed CTA or MRA by a modified Thrombolysis in Cerebral Infarction (TICI) score for abnormal artery: 0, no patency; 1, minimal patency – some contrast penetrates obstruction but no/minimal enters distal artery; 2, patency of <50% of the lumen at the point of obstruction and some filling of branches of the affected artery; 3, patency of >50% of the lumen and filling of most branches of the affected artery; 4, complete patency – normal artery.

‖Total lacunar stroke versus total non-lacunar stroke.

¶P<0.05 by randomisation treatment.

Appendix 5.5. Use of alteplase, BP lowering treatment within 24 hours, and other management from randomisation to Day 7 in participants with definite/probable lacunar and nonlacunar stroke

	Definite/probable lacunar stroke			Definite/probable non-lacunar stroke			P value[†]
	Intensive BP (N=238)	Standard BP (N=216)	Total (N=454)	Intensive BP (N=570)	Standard BP (N=608)	Total (N=1178)	
Alteplase treatment							
Any given	236 (99.2)	214 (99.1)	450 (99.1)	567 (99.5)	605 (99.5)	1172 (99.5)	0.39
Bolus dose, mg	5.9 (1.2)	5.9 (1.2)	5.9 (1.2)	6.2 (3.1)	6.0 (1.4)	6.1 (2.4)	0.13
Infusion over 60 mins dose, mg	48.3 (13.2)	48.0 (13.2)	48.1 (13.2)	48.8 (13.7)	48.6 (14.2)	48.7 (13.9)	0.44
Time from randomisation to treatment, mins	-5.0 (-33.6-7.2)	-12.1 (-42.9-5.2)	-7.6 (-39.0-5.7)	-0.2 (-37.6-8.5)	-0.1 (-35.6-8.4)	-0.1 (-36.6-8.5)	**0.008**
Time from stroke onset to treatment, mins	185 (142-230)	192 (147-235)	190 (144-230)	175 (135-220)	182 (140-225)	180 (139-220)	**0.03**
BP Management							
Any BP medication taken in the first 24 hours	188 (79.0)[‡]	92/215 (42.8)[‡]	280/453 (61.8)	470/567 (82.9)[‡]	357/606 (58.9)[‡]	827/1173 (70.5)	**<0.001**
Time from alteplase to treatment, mins	20 (0-64)	7 (-8-70)	16 (0-64.5)	18 (0-85)[‡]	50 (0-204)[‡]	23 (0-120)	**0.03**
Time from randomisation to treatment, mins	7.2 (-7.1-34.7)	-5.2 (-40.3-41.4)	6.0 (-18.8-40.4)	11.7 (-0.6-45.9)[‡]	29.8 (-9.3-184.4)[‡]	15.4 (-3.1-79.4)	**<0.001**
Time from stroke onset to treatment, mins	210 (157-265)	230 (170-296)	213 (160-270)	216 (157-275)[‡]	255 (180-390)[‡]	225 (164-301)	0.17
Method of iv medication administration							
Bolus	56 (23.5)[‡]	29/215 (13.5)[‡]	85/453 (18.8)	192/565 (34.0)[‡]	99/606 (16.3)[‡]	291/1171 (24.9)	**0.009**
Infusion	84 (35.3)[‡]	35/215 (16.3)[‡]	119/453 (26.3)	299/566 (52.8)[‡]	192/606 (31.7)[‡]	491/1172 (41.9)	**<0.001**
Number of different iv medications taken							
0	115 (48.3)[‡]	156/215 (72.6)[‡]	271/453 (59.8)	144/567 (25.4)[‡]	339/606 (55.9)[‡]	483/1173 (41.2)	**<0.001**
1	105 (44.1)[‡]	55/215 (25.6)[‡]	160/453 (35.3)	268/567 (47.3)[‡]	180/606 (29.7)[‡]	448/1173 (38.2)	
2	14 (5.9)[‡]	3/215 (1.4)[‡]	17/453 (3.8)	117/567 (20.6)[‡]	69/606 (11.4)[‡]	186/1173 (15.9)	
≥3	4 (1.7)[‡]	1/215 (0.5)[‡]	5/453 (1.1)	38/567 (6.7)[‡]	18/606 (3.0)[‡]	56/1173 (4.8)	
Systolic BP at 24 hours, mmHg	137 (13)[‡]	144 (16)[‡]	140 (15)	140 (16)[‡]	145 (18)[‡]	143 (17)	**0.004**
Average systolic BP within 24 hours, mmHg	143 (9)[‡]	149 (11)[‡]	146 (10)	145 (10)[‡]	151 (12)[‡]	148 (12)	**<0.001**
Any iv BP lowing treatment in first 24 hours	120 (50.4)[‡]	57/215 (26.5)[‡]	177/453 (39.1)	406/567 (71.6)[‡]	240/606 (39.6)[‡]	646/1173 (55.1)	**<0.001**
Any iv BP lowing treatment in days 2-7	64 (26.9)[‡]	29/215 (13.5)[‡]	93/453 (20.5)	256/562 (45.6)[‡]	175/596 (29.4)[‡]	431/1158 (37.2)	**<0.001**
Other management							
Cerebral angiogram undertaken	9 (3.8)	3 (1.4)	12 (2.6)	41 (7.2)	41 (6.7)	82 (7.0)	**<0.001**
Occluded cerebral vessel identified	0/9 (0.0)	0/3 (0.0)	0/12 (0.0)	32/40 (80.0)	26/41 (63.4)	58/81 (71.6)	**<0.001**
Endovascular clot retrieval used	0/9 (0.0)	0/3 (0.0)	0/12 (0.0)	25/41 (61.0)	17/41 (41.5)	42/82 (51.2)	**<0.001**
Intubation and ventilation	2 (0.8)	1/215 (0.5)	3/453 (0.7)	43/562 (7.7)	37/596 (6.2)	80/1158 (6.9)	**<0.001**
Fever occurrence	11 (4.6)	7/215 (3.3)	18/453 (4.0)	146/562 (26.0)	161/596 (27.0)	307/1158 (26.5)	**<0.001**
Fever treated	10/221 (4.5)	5/191 (2.6)	15/412 (3.6)	130/515 (25.2)	144/548 (26.3)	274/1063 (25.8)	**<0.001**
Nasogastric feeding given	2 (0.8)	5/215 (2.3)	7/453 (1.5)	159/562 (28.3)	165/596 (27.7)	324/1158 (28.0)	**<0.001**
Patient mobilized by therapist	72 (30.3)	58/215 (27.0)	130/453 (28.7)	247/562 (44.0)	279/596 (46.8)	526/1158 (45.4)	**<0.001**
Compression stockings used	12 (5.0)	10/215 (4.7)	22/453 (4.9)	65/562 (11.6)	54/596 (9.1)	119/1158 (10.3)	**<0.001**
Subcutaneous heparin used	19 (8.0)[‡]	33 (15.3)[‡]	52 (11.5)	147 (25.8)	134 (22.0)	281 (23.9)	**<0.001**
Any antithrombotic agent[*] used in first 24 hours	21 (8.8)	28 (13.0)	49 (10.8)	79 (13.9)	79/607 (13.0)	158/1177 (13.4)	0.15
iv traditional Chinese medicine administered	126 (52.9)	115/215 (53.5)	241/453 (53.2)	229/562 (40.7)	243/596 (40.8)	472/1158 (40.8)	**<0.001**
iv steroids administered	3 (1.3)	2/215 (0.9)	5/453 (1.1)	19/562 (3.4)	12/596 (2.0)	31/1158 (2.7)	0.05
iv mannitol administered	6 (2.5)	4/215 (1.9)	10/453 (2.2)	92/567 (16.2)	107/606 (17.7)	199/1173 (17.0)	**<0.001**

Hemicraniectomy performed	0 (0.0)	0/215 (0.0)	0/453 (0.0)	9/562 (1.6)	12/596 (2.0)	21/1158 (1.8)	**0.004**
Any neurosurgery performed	3 (1.3)	1 (0.5)	4 (0.9)	14 (2.5)	25 (4.1)	39 (3.3)	**0.006**
Any stroke unit admission	94 (39.5)	86/215 (40.0)	180/453 (39.7)	279/562 (49.6)	286/596 (48.0)	565/1158 (48.8)	**0.001**
Any intensive care unit admission	30 (12.6)	25/215 (11.6)	55/453 (12.1)	149/562 (26.5)	142/596 (23.8)	291/1158 (25.1)	**<0.001**
Any rehabilitation given	102 (42.9)	77/215 (35.8)	179/453 (39.5)	292/562 (52.0)	330/596 (55.4)	622/1158 (53.7)	**<0.001**
Decision to withdrawal active care	4 (1.7)	3/215 (1.4)	7/453 (1.5)	25/562 (4.4)	16/596 (2.7)	41/1158 (3.5)	**0.03**

BP denotes blood pressure; iv, intravenous.

Data are n (%), mean (SD), or median (Q1, Q3). The P values are based on Chi-square, analysis of variance, or Wilcoxon signed-rank test.

*Including antiplatelet agent or heparin.

†Total lacunar stroke versus total non-lacunar stroke.

‡ P<0.05 by randomisation treatment.

Appendix 5.6. Details on BP lowering treatment within 24 hours, and in days 2-7 post-randomisation in participants with definite/probable lacunar and nonlacunar stroke

	Definite/probable lacunar stroke			Definite/probable non-lacunar stroke			P value*
	Intensive BP (N=238)	Standard BP (N=216)	Total (N=454)	Intensive BP (N=570)	Standard BP (N=608)	Total (N=1178)	
BP lowering in the first 24 hours after randomisation							
Minimum (SD) SBP within 24 hours, mmHg	126 (11)†	131 (13)†	128 (12)	125 (12)†	131 (15)†	128 (14)	0.95
Maximum (SD) SBP within 24 hours, mmHg	161 (14)†	166 (14)†	164 (14)	165 (16)†	169 (16)†	167 (16)	**<0.001**
Intravenous agent used							
Labetalol	18 (7.6)†	3/215 (1.4)†	21/453 (4.6)	82/567 (14.5)†	42/606 (6.9)†	124/1173 (10.6)	**<0.001**
Metoprolol	1 (0.4)	0/215 (0.0)	1/453 (0.2)	5/567 (0.9)	3/606 (0.5)	8/1173 (0.7)	0.26
Atenolol	0 (0.0)	0/215 (0.0)	0/453 (0.0)	1/567 (0.2)	1/606 (0.2)	2/1173 (0.2)	0.38
Nicardipine	26 (10.9)†	11/215 (5.1)†	37/453 (8.2)	35/567 (6.2)	24/606 (4.0)	59/1173 (5.0)	**0.02**
Clevidipine	0 (0.0)	0/215 (0.0)	0/453 (0.0)	1/567 (0.2)	0/606 (0.0)	1/1173 (0.1)	0.53
Nimodipine	4 (1.7)	1/215 (0.5)	5/453 (1.1)	15/567 (2.6)†	6/606 (1.0)†	21/1173 (1.8)	0.32
Nifedipine	0 (0.0)	0/215 (0.0)	0/453 (0.0)	5/567 (0.9)	4/606 (0.7)	9/1173 (0.8)	0.06
Urapidil	23 (9.7)	17/215 (7.9)	40/453 (8.8)	92/567 (16.2)†	28/606 (4.6)†	120/1173 (10.2)	0.40
Sodium nitroprusside	37 (15.5)†	19/215 (8.8)†	56/453 (12.4)	107/567 (18.9)†	58/606 (9.6)†	165/1173 (14.1)	0.37
Nitroglycerin	11 (4.6)	4/215 (1.9)	15/453 (3.3)	78/567 (13.8)†	21/606 (3.5)†	99/1173 (8.4)	**<0.001**
Isosorbide dinitrate	1 (0.4)	0/215 (0.0)	1/453 (0.2)	7/567 (1.2)	5/606 (0.8)	12/1173 (1.0)	0.10
Frusemide	4 (1.7)	0/215 (0.0)	4/453 (0.9)	47/567 (8.3)	43/606 (7.1)	90/1173 (7.7)	**<0.001**
Prazosin	2 (0.8)	0/215 (0.0)	2/453 (0.4)	0/567 (0.0)	1/606 (0.2)	1/1173 (0.1)	0.13
Hydralazine	2 (0.8)	1/215 (0.5)	3/453 (0.7)	12/567 (2.1)	6/606 (1.0)	18/1173 (1.5)	0.16
Clonidine	3 (1.3)	0/215 (0.0)	3/453 (0.7)	7/567 (1.2)	2/606 (0.3)	9/1173 (0.8)	0.82
Enalapril	2 (0.8)	0/215 (0.0)	2/453 (0.4)	2/567 (0.4)	3/606 (0.5)	5/1173 (0.4)	0.97
Other medication(s)	8 (3.4)	4/215 (1.9)	12/453 (2.6)	36/567 (6.3)	25/606 (4.1)	61/1173 (5.2)	**0.03**
Topical nitrates used	12 (5.0)	4/215 (1.9)	16/453 (3.5)	36/566 (6.4)†	17/606 (2.8)†	53/1172 (4.5)	0.37
Oral agents used							
ACEI/angiotensin II receptor antagonist	69 (29.0)†	25/215 (11.6)†	94/453 (20.8)	112/567 (19.8)†	83/606 (13.7)†	195/1173 (16.6)	0.05
Diuretic	16 (6.7)†	5/215 (2.3)†	21/453 (4.6)	35/567 (6.2)	37/606 (6.1)	72/1173 (6.1)	0.24
Beta blocker	5 (2.1)	9/215 (4.2)	14/453 (3.1)	48/567 (8.5)	65/606 (10.7)	113/1173 (9.6)	**<0.001**
Calcium channel blocker	71 (29.8)†	30/215 (14.0)†	101/453 (22.3)	133/567 (23.5)†	80/606 (13.2)†	213/1173 (18.2)	0.06
Oral sympathetic antagonist	1 (0.4)	3/215 (1.4)	4/453 (0.9)	3/567 (0.5)	6/606 (1.0)	9/1173 (0.8)	0.81
Other medication(s)	10 (4.2)	8/215 (3.7)	18/453 (4.0)	28/567 (4.9)	40/606 (6.6)	68/1173 (5.8)	0.14
BP lowering treatment on Day 7							
Any BP medication taken	166 (69.7)†	105/215 (48.8)†	271/453 (59.8)	425/562 (75.6)†	420/596 (70.5)†	845/1158 (73.0)	**<0.001**
Any iv BP lowering treatment	69 (29.0)†	34/215 (15.8)†	103/453 (22.7)	283/562 (50.4)†	221/596 (37.1)†	504/1158 (43.5)	**<0.001**
Number of different iv medications taken							
0	176 (73.9)†	183/215 (85.1)†	359/453 (79.2)	288/562 (51.2)†	378/596 (63.4)†	666/1158 (57.5)	**<0.001**
1	48 (20.2)†	28/215 (13.0)†	76/453 (16.8)	154/562 (27.4)†	139/596 (23.3)†	293/1158 (25.3)	
2	9 (3.8)†	2/215 (0.9)†	11/453 (2.4)	86/562 (15.3)†	61/596 (10.2)†	147/1158 (12.7)	
≥3	5 (2.1)†	2/215 (0.9)†	7/453 (1.5)	34/562 (6.0)†	18/596 (3.0)†	52/1158 (4.5)	

ACEI denotes angiotensin converting enzyme inhibitor; BP, blood pressure; iv, intravenous; SBP, systolic BP; SD, standard deviation.

Data are n (%), or mean (SD). The P values are based on Chi-square or analysis of variance.
*Total lacunar stroke versus total non-lacunar stroke.
† P<0.05 by randomisation treatment.

Appendix 5.7. Baseline characteristics of participants with definite lacunar and nonlacunar stroke

	Definite lacunar stroke			Definite non-lacunar stroke			P
	Intensive BP lowing (N=86)	Standard BP lowing (N=69)	Total (N=155)	Intensive BP lowing (N=392)	Standard BP lowing (N=392)	Total (N=784)	value[‖]
Age (years)	63.4 (12.6)	62.0 (11.9)	62.7 (12.3)	69.3 (12.4)	69.7 (11.2)	69.5 (11.8)	**<0.001**
≥80 years	6 (7.0)	3 (4.3)	9 (5.8)	80 (20.4)	67 (17.1)	147 (18.8)	**<0.001**
Female	30 (34.9)	19 (27.5)	49 (31.6)	148 (37.8)	162 (41.3)	310 (39.5)	0.06
Asian ethnicity	68 (79.1)	55 (79.7)	123 (79.4)	236 (60.2)	247 (63.0)	483 (61.6)	**<0.001**
Clinical features							
Systolic BP (mmHg)	165.4 (9.4)	166.1 (9.4)	165.7 (9.4)	165.7 (9.5)	165.5 (9.1)	165.6 (9.3)	0.89
Diastolic BP (mmHg)	92.0 (9.4)	93.3 (10.9)	92.6 (10.1)	91.0 (12.0)	89.6 (12.4)	90.3 (12.2)	**0.03**
Heart rate (beats per minute)	79.1 (12.6)	78.8 (13.9)	78.9 (13.1)	79.7 (15.8)	80.5 (17.2)	80.1 (16.5)	0.42
NIHSS score[*]	6 (4-8)	5 (3-7)	5 (4-7)	10 (6-16)	10 (6-15)	10 (6-15)	**<0.001**
NIHSS score ≥14	3 (3.5)	1 (1.4)	4 (2.6)	133 (33.9)	131 (33.4)	264 (33.7)	**<0.001**
GCS score[†]	15 (15-15)[¶]	15 (15-15)[¶]	15 (15-15)	15 (13-15)	15 (13-15)	15 (13-15)	**<0.001**
Medical history							
Previous stroke	16 (18.6)[¶]	5 (7.2)[¶]	21 (13.5)	66 (16.8)	70 (17.9)	136 (17.3)	0.25
Hypertension	61 (70.9)	44 (63.8)	105 (67.7)	290 (74.0)	289 (73.7)	579 (73.9)	0.12
Atrial fibrillation	3 (3.5)	2 (2.9)	5 (3.2)	79 (20.2)[¶]	105/391 (26.9)[¶]	184/783 (23.5)	**<0.001**
Coronary artery disease	5 (5.8)	6 (8.7)	11 (7.1)	67 (17.1)	71 (18.1)	138 (17.6)	**0.001**
Valvular or other heart disease	0 (0.0)	1 (1.4)	1 (0.6)	28 (7.1)	30 (7.7)	58 (7.4)	**0.002**
Diabetes mellitus	17 (19.8)	15 (21.7)	32 (20.6)	84 (21.4)	100 (25.5)	184 (23.5)	0.45
Hypercholesterolemia	7 (8.1)	4 (5.8)	11 (7.1)	61 (15.6)	69 (17.6)	130 (16.6)	**0.003**
Current smoker	25 (29.1)	17 (24.6)	42 (27.1)	80/391 (20.5)	86/391 (22.0)	166/782 (21.2)	0.11
Pre-stroke function without symptoms[‡]	6 (7.0)	7 (10.1)	13 (8.4)	78 (19.9)	70 (17.9)	148 (18.9)	**0.002**
Medication on admission							
Antihypertensive agent(s)	41 (47.7)	29 (42.0)	70 (45.2)	212 (54.1)	210 (53.6)	422 (53.8)	**0.05**
Warfarin anticoagulation	0 (0.0)	0 (0.0)	0 (0.0)	9 (2.3)	7 (1.8)	16 (2.0)	0.07
Aspirin/other antiplatelet agent	14 (16.3)	12 (17.4)	26 (16.8)	76 (19.4)	92 (23.5)	168 (21.4)	0.19
Statin/other lipid lowering agent	12 (14.0)	7 (10.1)	19 (12.3)	77 (19.6)	80 (20.4)	157 (20.0)	**0.02**
Imaging features							
Infarct at left side	36 (41.9)	30 (43.5)	66 (42.6)	184/388 (47.4)	192/387 (49.6)	376/775 (48.5)	0.18
Infarct at right side	49 (57.0)	36 (52.2)	85 (54.8)	181/388 (46.6)	175/387 (45.2)	356/775 (45.9)	**0.04**
Infarct at midline or bilateral side	1 (1.2)	3 (4.3)	4 (2.6)	23/388 (5.9)	20/387 (5.2)	43/775 (5.5)	0.12
Infarct at anterior circulation	71 (82.6)	51 (73.9)	122 (78.7)	274/388 (70.6)	279/387 (72.1)	553/775 (71.4)	0.06
Infarct at posterior circulation	15 (17.4)	17 (24.6)	32 (20.6)	78/388 (20.1)	76/387 (19.6)	154/775 (19.9)	0.83
At both anterior and posterior circulation	0 (0.0)	1 (1.4)	1 (0.6)	36/388 (9.3)	32/387 (8.3)	68/775 (8.8)	**<0.001**
With hyperdense vessel sign on CT	0/78 (0.0)	1/67 (1.5)	1/145 (0.7)	104/384 (27.1)	100/384 (26.0)	204/768 (26.6)	**<0.001**
LVO on baseline CTA or MRA[§]	0/16 (0.0)	0/10 (0.0)	0/26 (0.0)	58/78 (74.4)[¶]	40/69 (58.0)[¶]	98/147 (66.7)	**<0.001**
With old vascular lesions	38 (44.2)	36 (52.2)	74 (47.7)	193/390 (49.5)	190 (48.5)	383/782 (49.0)	0.78
With brain atrophy	38 (44.2)	34 (49.3)	72 (46.5)	272/390 (69.7)	261 (66.6)	533/782 (68.2)	**<0.001**
With white matter changes	37 (43.0)	23 (33.3)	60 (38.7)	165/390 (42.3)	164 (41.8)	329/782 (42.1)	0.44
Site reported LVO or assessed centrally	0 (0.0)	0 (0.0)	0 (0.0)	100 (25.5)	80/391 (20.5)	180/783 (23.0)	**<0.001**

Time from stroke onset to randomisation (hrs)	3.5 (2.6-4.2)	3.7 (2.7-4.2)	3.5 (2.7-4.2)	3.1 (2.4-3.9)	3.1 (2.5-3.9)	3.1 (2.4-3.9)	**<0.001**
Assigned to low-dose alteplase group	20 (23.3)	18 (26.1)	38 (24.5)	102 (26.0)	109 (27.8)	211 (26.9)	0.54
Assigned to standard-dose alteplase group	19 (22.1)	12 (17.4)	31 (20.0)	101 (25.8)	108 (27.6)	209 (26.7)	0.08
Presumed diagnosis at time of hospital separation							
Non-stroke	0 (0.0)	0 (0.0)	0 (0.0)	2/387 (0.5)	1/382 (0.3)	3/769 (0.4)	0.44
Large artery occlusion because of atheroma	0 (0.0)	0 (0.0)	0 (0.0)	174/387 (45.0)	180/382 (47.1)	354/769 (46.0)	**<0.001**
Small vessel disease	86 (100.0)	69 (100.0)	155 (100.0)	58/387 (15.0)	47/382 (12.3)	105/769 (13.7)	**<0.001**
Cardioembolism	0 (0.0)	0 (0.0)	0 (0.0)	93/387 (24.0)	97/382 (25.4)	190/769 (24.7)	**<0.001**
Dissection	0 (0.0)	0 (0.0)	0 (0.0)	3/387 (0.8)	2/382 (0.5)	5/769 (0.7)	0.31
Other or uncertain pathogenesis	0 (0.0)	0 (0.0)	0 (0.0)	57/387 (14.7)	55/382 (14.4)	112/769 (14.6)	**<0.001**

BP denotes blood pressure; CTA, computed tomography angiography; GCS, Glasgow Coma Scale; LVO, large vessel occlusion; MRA, magnetic resonance angiography; mRS, modified Rankin scale; NIHSS, National Institutes of Health stroke scale.

Data are n (%), mean (SD), or median (Q1, Q3). The P values are based on Chi-square, analysis of variance, or Wilcoxon signed-rank test.

*Scores on the NIHSS range from 0 to 42, with higher scores indicating more severe neurological deficits.

†Scores on the GCS range from 15 (normal) to 3 (deep coma).

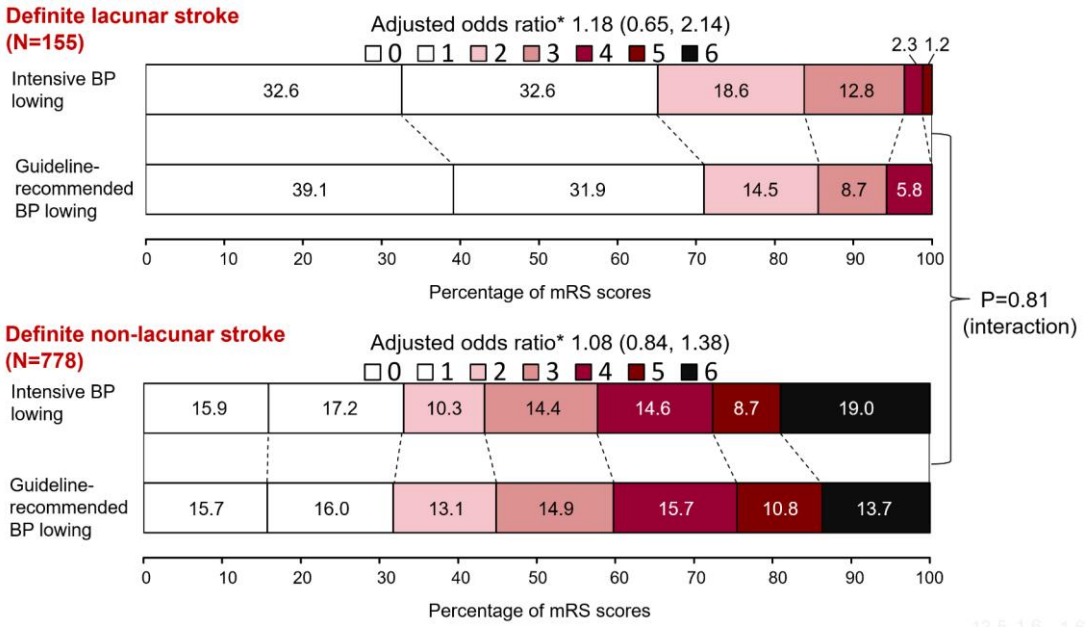
‡mRS=0.

§Scored as 0, 1, or 2 for proximal cerebral arteries (A1-2 segment of anterior cerebral artery, internal carotid artery, M1-2 segment of middle cerebral artery, P1-2 segment of posterior cerebral artery, or basilar artery) on raw or reconstructed CTA or MRA by a modified Thrombolysis in Cerebral Infarction (TICI) score for abnormal artery: 0, no patency; 1, minimal patency – some contrast penetrates obstruction but no/minimal enters distal artery; 2, patency of <50% of the lumen at the point of obstruction and some filling of branches of the affected artery; 3, patency of >50% of the lumen and filling of most branches of the affected artery; 4, complete patency – normal artery.

‖Total lacunar infarcts versus total non-lacunar stroke.

¶P<0.05 by randomisation treatment.

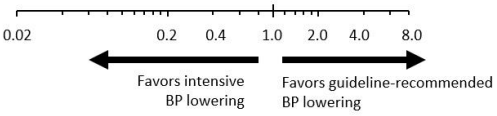
Appendix 5.8. Randomised treatment effects on ordinal mRS score in definite lacunar and nonlacunar stroke



BP denotes blood pressure.
*Adjusted for key prognostic covariates (age; sex; ethnicity; baseline NIHSS score; time from stroke onset to randomisation; pre-morbid function [mRS scores 0 or 1]; prior use of antithrombotic agents [aspirin, other antiplatelet agents, or warfarin]; history of diabetes or cardiovascular disease [stroke, atrial fibrillation, coronary artery disease, valvular or other heart disease]; assigned to low-dose alteplase group).

Appendix 5.9. Thrombolysis outcomes in participants with definite lacunar and nonlacunar stroke by randomised treatment of BP reduction

Thrombolysis outcomes	BP lowering group, n/N (%)		Odds ratio (95% CI)	P inter-action
	Intensive	Guideline		
mRS 2-6: overall	291/476 (61.1)	285/457 (62.4)	0.93 (0.70, 1.25)*	
Definite lacunar stroke	30/86 (34.9)	20/69 (29.0)	1.13 (0.54, 2.39)*	0.41*
Definite non-lacunar stroke	261/390 (66.9)	265/388 (68.3)	0.90 (0.66, 1.25)*	
mRS 3-6: overall	235/476 (49.4)	224/457 (49.0)	1.02 (0.77, 1.36)*	
Definite lacunar stroke	14/86 (16.3)	10/69 (14.5)	1.01 (0.38, 2.71)*	0.99*
Definite non-lacunar stroke	221/390 (56.7)	214/388 (55.2)	1.06 (0.78, 1.43)*	
mRS 6 (death): overall	74/478 (15.5)	53/461 (11.5)	1.46 (0.97, 2.21)*	
Definite lacunar stroke	0/86 (0.0)	0/69 (0.0)	—	—
Definite non-lacunar stroke	74/392 (18.9)	53/392 (13.5)	1.53 (1.01, 2.32)*	
sICH (SITS-MOST): overall	11/478 (2.3)	13/461 (2.8)	0.79 (0.35, 1.79)†	
Definite lacunar stroke	0/86 (0.0)	0/69 (0.0)	—	—
Definite non-lacunar stroke	11/392 (2.8)	13/392 (3.3)	0.83 (0.37, 1.88)†	
Adjudicated ICH: overall	89/478 (18.6)	107/461 (23.2)	0.76 (0.55, 1.04)	
Definite lacunar stroke	1/86 (1.2)	2/69 (2.9)	0.39 (0.03, 4.44)	0.58
Definite non-lacunar stroke	88/392 (22.4)	105/392 (26.8)	0.79 (0.57, 1.10)	
Intracranial hemorrhage‡: overall	96/478 (20.1)	122/461 (26.5)	0.70 (0.51, 0.95)	
Definite lacunar stroke	1/86 (1.2)	3/69 (4.3)	0.26 (0.03, 2.55)	0.38
Definite non-lacunar stroke	95/392 (24.2)	119/362 (30.4)	0.73 (0.54, 1.01)	
END or death within 24 hrs: overall	66/478 (13.8)	60/461 (13.0)	1.07 (0.74, 1.56)	
Definite lacunar stroke	4/86 (4.7)	2/69 (2.9)	1.63 (0.29, 9.20)	0.65
Definite non-lacunar stroke	62/392 (15.8)	58/392 (14.8)	1.08 (0.73, 1.60)	
END or death within 72 hrs: overall	94/478 (19.7)	76/461 (16.5)	1.24 (0.89, 1.73)	
Definite lacunar stroke	4/86 (4.7)	3/69 (4.3)	1.07 (0.23, 4.96)	0.81
Definite non-lacunar stroke	90/392 (23.0)	73/392 (18.6)	1.30 (0.92, 1.84)	



END denotes early neurologic deterioration; ICH, intracerebral haemorrhage; mRS, modified Rankin scale; sICH, symptomatic intracerebral haemorrhage; SITS-MOST, the Safe Implementation of Thrombolysis in Stroke-Monitoring Study.

*Adjusted for key prognostic covariates (age; sex; ethnicity; baseline NIHSS score; time from stroke onset to randomisation; pre-morbid function [mRS scores 0 or 1]; prior use of antithrombotic agents [aspirin, other antiplatelet agents, or warfarin]; history of diabetes or cardiovascular disease [stroke, atrial fibrillation, coronary artery disease, valvular or other heart disease]; assigned to low-dose alteplase group) for functional outcomes.

†Adjusted for minimization and key prognostic covariates (age, baseline NIHSS score, time from stroke onset to randomisation, and assigned to low-dose alteplase group) for safety outcomes, and neurologic deterioration within 24 or 72 hours.

‡Site reported or adjudicated centrally.

Appendix 5.10. Thrombolysis outcomes in participants with definite lacunar stroke and the infarct size ≤15 mm on 24-hour follow-up images by randomised treatment

		Randomised BP lowering, n/N (%)		OR (95% CI) (intensive vs. guideline)	P value	Adjusted OR (95% CI)* (intensive vs. guideline)	P value
		Intensive	Guideline				
mRS shift	0	19/52 (36.5)	19/39 (48.7)	1.65 (0.77, 3.55)	0.20	1.09 (0.48, 2.48)	0.84
	1	14/52 (26.9)	10/39 (25.6)				
	2	10/52 (19.2)	6/39 (15.4)				
	3	7/52 (13.5)	2/39 (5.1)				
	4	2/52 (3.8)	2/39 (5.1)				
Death or disability (mRS 2–6)		19/52 (36.5)	10/39 (25.6)	1.67 (0.67, 4.16)	0.27	0.95 (0.32, 2.82)	0.93
Death or major disability (mRS 3–6)		9/52 (17.3)	4/39 (10.3)	1.83 (0.52, 6.45)	0.35	1.46 (0.35, 6.10)	0.60
Death (mRS 6)		0/52 (0.0)	0/39 (0.0)	-	-	-	-
sICH (SITS-MOST criteria)		0/52 (0.0)	0/39 (0.0)	-	-	-	-
Adjudicated ICH		1/52 (1.9)	1/39 (2.6)	0.75 (0.05, 12.29)	0.84		
Intracranial haemorrhage		1/52 (1.9)	1/39 (2.6)	0.75 (0.05, 12.29)	0.84	-	-
END or death within 24 hours		2/52 (3.8)	2/39 (5.1)	0.74 (0.10, 5.50)	0.77	-	-
END or death within 7 days		2/52 (3.8)	2/39 (5.1)	0.74 (0.10, 5.50)	0.77	-	-

END denotes early neurologic deterioration; ICH, intracerebral haemorrhage; mRS, modified Rankin scale; SITS-MOST, the Safe Implementation of Thrombolysis in Stroke-Monitoring Study.

*Adjusted for key prognostic covariates (age; sex; ethnicity; baseline NIHSS score; time from stroke onset to randomisation; pre-morbid function [mRS scores 0 or 1]; prior use of antithrombotic agents [aspirin, other antiplatelet agents, or warfarin]; history of diabetes or cardiovascular disease [stroke, atrial fibrillation, coronary artery disease, valvular or other heart disease]; assigned to low-dose alteplase group).

Chapter 6: Thrombolysis Outcomes According to Arterial Characteristics of Acute Ischaemic Stroke by Alteplase Dose and Blood Pressure Target

6.1 Link to thesis

In Chapters 4 and 5, I presented post-hoc analyses from the ENCHANTED trial on low-dose versus standard-dose alteplase and intensive versus guideline-recommended BP lowering in patients with acute lacunar ischaemic stroke who usually have no vascular occlusion on cerebral angiography. Intravenous thrombolysis provides benefit to AIS patients with large vessel obstruction but depends on the site of obstruction. There is still little evidence of thrombolysis outcomes in AIS patients with obstruction at the internal carotid artery or medium cerebral artery. During the trial, the steering committee requested each site to upload participants' baseline (before randomisation) and 24-hour follow-up brain images of non-contrast CT or MRI for central adjudication and encouraged centres to upload angiography and perfusion images if available. This provides an opportunity to study thrombolysis outcomes according to vascular obstruction status/sites from a large-scale trial and explore associations between randomised alteplase dose or BP target and outcomes across participants with different vascular obstruction status/sites. This Chapter presents vascular analyses from the ENCHANTED trial.

I have published this work:
Zhou Z, Xia C, Mair G, Delcourt C, Yoshimura S, Liu X, Chen Z, Malavera A, Carcel C, Chen X, Wang X, Al-Shahi Salman R, Robinson TG, Lindley RI, Chalmers J, Wardlaw JM, Parsons MW, Demchuk AM, Anderson CS. Thrombolysis outcomes according to arterial characteristics of acute ischaemic stroke by alteplase dose and blood pressure target. *Int J Stroke* 2021:17474930211025436 (Online ahead of print).

6.2 Abstract

Objective: We explored the influence of low-dose intravenous alteplase and intensive blood pressure (BP) lowering on outcomes of acute ischaemic stroke (AIS) according to status/location of vascular obstruction in participants of the Enhanced Control of Hypertension and Thrombolysis Stroke Study (ENCHANTED).

Methods: ENCHANTED was a multicentre, quasi-factorial, randomised trial to determine efficacy and safety of low- versus standard-dose intravenous alteplase and intensive- versus guideline-recommended BP lowering in AIS patients. In those who had baseline CT or MRI angiography, the degree of vascular occlusion was grouped according to being no (NVO), medium (MVO), or large (LVO). Logistic regression models were used to determine 90-day outcomes (modified Rankin scale [mRS] shift [primary], other mRS cut-scores, intracranial haemorrhage, early neurologic deterioration [END], and recanalisation) by vascular obstruction status/site. Heterogeneity in associations for outcomes across subgroups was estimated by adding an interaction term to the models.

Results: There were 940 participants: 607 in alteplase arm only, 243 in BP arm only, and 90 assigned to both arms. Compared to the NVO group, functional outcome was worse in LVO (mRS shift, adjusted OR [95% CI] 2.13 [1.56-2.90] but comparable in MVO (1.34 [0.96-1.88]) groups. There were no differences in associations of alteplase dose or BP lowering and outcomes across NVO/MVO/LVO groups (mRS shift: low versus standard alteplase dose 0.84 [0.54-1.30]/0.48 [0.25-0.91]/0.99 [0.75-2.09], $P_{interaction}$=0.28; intensive versus standard BP lowering 1.32 [0.74-2.38]/0.78 [0.31-1.94]/1.24 [0.64-2.41], $P_{interaction}$=0.41), except for a borderline significant difference for intensive BP lowering and increased END (0.63 [0.14-2.72]/0.17 [0.02-1.47]/2.69 [0.90-8.04], $P_{interaction}$=0.05).

Conclusions: Functional outcome by dose of alteplase or intensity of BP lowering is not modified by vascular obstruction status/site according to analyses from ENCHANTED, although these results are compromised by low statistical power.

Clinical Trial Registration: http://www.clinicaltrials.gov. Unique identifiers: NCT01422616

6.3 Background

There is increasing recognition of the importance of multimodal brain images with vascular and perfusion information in guiding management of patients with acute ischaemic stroke (AIS).[1] Intravenous thrombolysis provides benefit to AIS patients with large vessel obstruction (LVO), but is dependent on the site of obstruction.[2] Meanwhile, evidence of thrombolysis outcomes in AIS patients without vascular obstruction or with obstruction at internal carotid artery (ICA) or medium cerebral artery is limited, with most data derived from observational studies.[3-5]

The Enhanced Control of Hypertension and Thrombolysis Stroke Study (ENCHANTED) was the first large-scale, multicentre, quasi-factorial, randomised trial that assessed the effectiveness of low-dose (0.6 mg/kg) versus standard-dose (0.9 mg/kg) intravenous alteplase (alteplase arm), and more intensive blood pressure (BP) lowering (target systolic BP [SBP] 130-140 mmHg within 1 hour post-randomisation) versus guideline-recommended BP control (SBP <180 mmHg) (BP arm) in AIS patients.[6,7] The trial did not show noninferiority between the two alteplase dose groups with respect to 90-day death and disability post-randomisation, and indicated a neutral effect of intensive versus guideline-recommended BP lowering on functional recovery. Low-dose alteplase significantly reduced the risk of symptomatic intracerebral haemorrhage (sICH) and intensive BP lowering significantly reduced the risk of any intracranial haemorrhage. Herein, we report our post-hoc analyses of ENCHANTED in relation to randomised alteplase dose and BP target, and key clinical outcomes, according to vascular obstruction status/site.

6.4 Methods

6.4.1 Patients

The study design, patient characteristics, and main results of ENCHANTED have been reported.[6,7] The study was approved by ethics committees at participating centres, and written informed consent was obtained from all patients or an appropriate legal surrogate. A key inclusion criterion was a clinical diagnosis of AIS confirmed by brain imaging that fulfilled guideline criteria for thrombolysis treatment <4.5 hours of symptom onset. These analyses pertain to ENCHANTED participants with baseline CT angiography (CTA) or MRI angiography (MRA) of moderate to good quality (appendix 6.1) or with proximal arterial occlusion according to reports by site investigators; the small number of participants who received endovascular therapy were excluded.

6.4.2 Imaging analysis

The analysis for non-contrast CT/MRI images collected from sites has been outlined elsewhere.[8] Since September 2019, we further assessed vascular occlusion parameters according to the presence, location, extent (third International Stroke Trial [IST-3] angiography score and modified Thrombolysis in Cerebral Infarction [TICI] score[3]), clot burden score,[9] visual residual flow,[10] and collateral score[11] on baseline CTA or MRA, and recanalisation (revised Arterial Occlusive Lesion [rAOL] score[10]) in participants with both baseline and

follow-up angiography (appendix 6.2). All methods of imaging assessment were pre-specified, and readers were blind to all clinical, treatment and outcome data except time, where they were allowed to simultaneously view baseline and follow-up scans; these data were cross-checked (Zien Zhou) before unblinding for analyses.

Vascular obstruction was defined as IST-3 angiography scores 0 to 2b and TICI scores 0 to 2a, after excluding the mildest grades of obstruction (patency of >50% of vascular lumen and filling of most branches). In the primary analyses, eligible participants were grouped as having no vascular obstruction (NVO), medium vascular obstruction (MVO) (M2/M3 segment of middle cerebral artery [M2/M3-MCA], anterior cerebral artery [ACA], or posterior cerebral artery [PCA] obstruction) and LVO (M1 segment of MCA [M1-MCA], vertebral artery, basilar artery, or ICA obstruction) according to the IST-3 classification,[3] by which site-reported angiography data were classifiable, with more samples after involving both adjudicated and site-reported angiography data (appendix 6.3). Participants were classified as LVO if there was both LVO and MVO. Given the definition of MVO has not been unified, two sets of sensitivity analyses were undertaken after re-classifying MVO according to criteria used in a recent study:[5] MVO in sensitivity analysis 1 (M2/M3-MCA, A2/A3-ACA, or P2/P3-PCA obstruction) and MVO in sensitivity analysis 2 (distal M2/M3-MCA, A2/A3-ACA, or P2/P3-PCA obstruction); those with A1-ACA, P1-PCA, or proximal M2-MCA obstruction were classified as LVO accordingly.

6.4.3 Outcomes

The primary outcome for these analyses was an ascending ordinal shift in the full range of scores on the modified Rankin scale (mRS) at 90 days post-randomisation. Secondary efficacy outcomes included early neurological deterioration (END; ≥4 points increase in NIHSS score or death) within 24 hours, recanalisation (rAOL scores 2b or 3) within 48 hours, and disability or death (mRS scores 2-6), major disability or death (mRS scores 3-6) and death (mRS score 6) at 90 days. Secondary safety outcomes were any intracranial haemorrhage adjudicated centrally or reported by site investigators within 7 days post-randomisation, and sICH defined according to the criteria from the second European-Australian Cooperative Acute Stroke Study (ECASS2).[12]

6.4.4 Statistical analysis

Associations of MVO or LVO versus NVO, with 90-day function, safety, and other secondary outcomes, were estimated in logistic regression models after adjustment for randomised

treatment (alteplase dose and BP lowering) and key clinical covariates (post-hoc defined) that were significantly different in univariable analysis and/or associated with prognosis according to literature and theoretical considerations. Associations of randomised alteplase dose or BP target with outcomes were determined in logistic regression models, with adjustment for randomised treatment and minimized key prognostic covariates (age, sex, Asian ethnicity, baseline NIHSS score, time from stroke onset to randomisation) for outcomes with ≥20 events. Heterogeneity across NVO/MVO/LVO groups was estimated by adding an interaction term to the statistical models. Ordinal logistic regression model was used to analyse mRS shift. Data are reported as odds ratios (OR) and 95% confidence intervals (CI), and a 2-sided P <0.05 was considered statistically significant. All analyses were performed using SAS version 7.1.

6.5 Results

6.5.1 Baseline characteristics

We included 940 ENCHANTED participants (607 in alteplase arm only, 243 in BP arm only, and 90 were assigned to both arms) for the primary analyses (appendix 6.4); including 641 evaluated by collected CTA (n=488) or MRA (n=153), and 299 classified from site-reported angiography data. Recanalisation was assessed in 74 participants who had vascular obstruction at baseline (22, 51 and 1 on follow-up CTA, MRA, and digital subtraction angiography, respectively). In the alteplase dose arm, compared to participants excluded from analyses, those included were older and less Asian, with lower BP and history of prior stroke, but higher heart rate and greater proportion of hypercholesterolemia and use of antihypertensive, antiplatelet and lipid lowering agents, and shorter time interval from symptom onset to randomisation (appendix 6.5). There were also differences between participants from the BP arm included and excluded in relation to age, proportion of Asians, baseline BP level, heart rate, history of atrial fibrillation (AF), hypercholesterolemia, and pre-stroke independence (appendix 6.6).

Across participants with different vascular obstruction status/site, there were differences according to ethnicity, baseline SBP level, NIHSS score, GCS score, prior AF, hypercholesterolemia and treatment with lipid lowering agents, time from stroke onset to randomisation, confirmed infarct lesion, presence of hyperdense vessel sign on CT, and presumed diagnosis of AIS. Neurological impairment was mildest for NVO (median [Q1-Q3] NIHSS 6 [4-9]) and greatest for LVO (12 [7-17]) (appendix 6.7). Most of the baseline characteristics were comparable by randomised alteplase dose or BP lowering across the

subgroups of NVO/MVO/LVO (appendices 6.8 and 6.9). Distribution of alteplase dose received, and mean BP within the first week post-randomisation, in participants with different obstruction status/site are shown in appendices 6.10 and 6.11, respectively.

6.5.2 Vascular obstruction and outcomes

Compared to patients with NVO, those with LVO had worse 90-day functional recovery independent of baseline covariables (age, sex, ethnicity, SBP, NIHSS score, prior AF, time from symptom onset to randomisation) and randomised alteplase dose and BP control; whether defined by ordinal shift in the full range of mRS scores (adjusted OR [95% CI] 2.13 [1.56-2.90]), mRS scores 2-6 (1.62 [1.11-2.36]), mRS scores 3-6 (2.58 [1.74-3.82]), death alone (4.12 [1.89-9.11]), and END (3.27 [1.70-6.28]) (table 6.1). There was no significant difference in functional outcomes and END for MVO versus NVO, after adjustment of covariables and randomised treatment (mRS shift 1.34 [0.96-1.88]; END 2.17 [0.999-4.70]). Haemorrhagic complications were more likely to occur after thrombolysis in either MVO or LVO compared to NVO (any intracranial haemorrhage MVO 2.27 [1.34-3.83], LVO 2.01 [1.25-3.26]; sICH MVO 3.42 [1.02-11.48], LVO 4.38 [1.65-11.61]). Recanalisation within 48 hours post-thrombolysis was higher in MVO compared to LVO (56.7% versus 31.8%; P=0.03). In sensitivity analysis, the results were comparable except for no difference in haemorrhagic complications for MVO versus NVO (data not shown).

6.5.3 Vascular obstruction and alteplase dose

Associations of randomised alteplase dose with functional outcomes in 697 included participants from the alteplase dose arm were comparable to the main results of the ENCHANTED trial: that is, the low-dose versus standard-dose groups had comparable functional outcomes (mRS shift, N=679, adjusted OR [95% CI] 0.86 [0.66-1.13]) (Figure 6.1). However, the benefit of low-dose alteplase on sICH was not confirmed in the present analysis (N=697, OR [95% CI] 0.52 [0.26-1.04]). There were no differential associations with mRS shift and other functional, safety, and intermediate outcomes across the subgroups of NVO/MVO/LVO (all $P_{interaction} \geq 0.12$), although MVO patients had a significant reduction in mRS shift (N=130, 0.48 [0.25-0.91]) and mRS 2-6 (N=130, 0.45 [0.21-0.97]) from low-dose alteplase treatment (Figures 6.1 and 6.2) (not seen in sensitivity analyses [appendices 6.12-6.14]).

Table 6.1. Thrombolysis outcomes by vascular obstruction status and sites

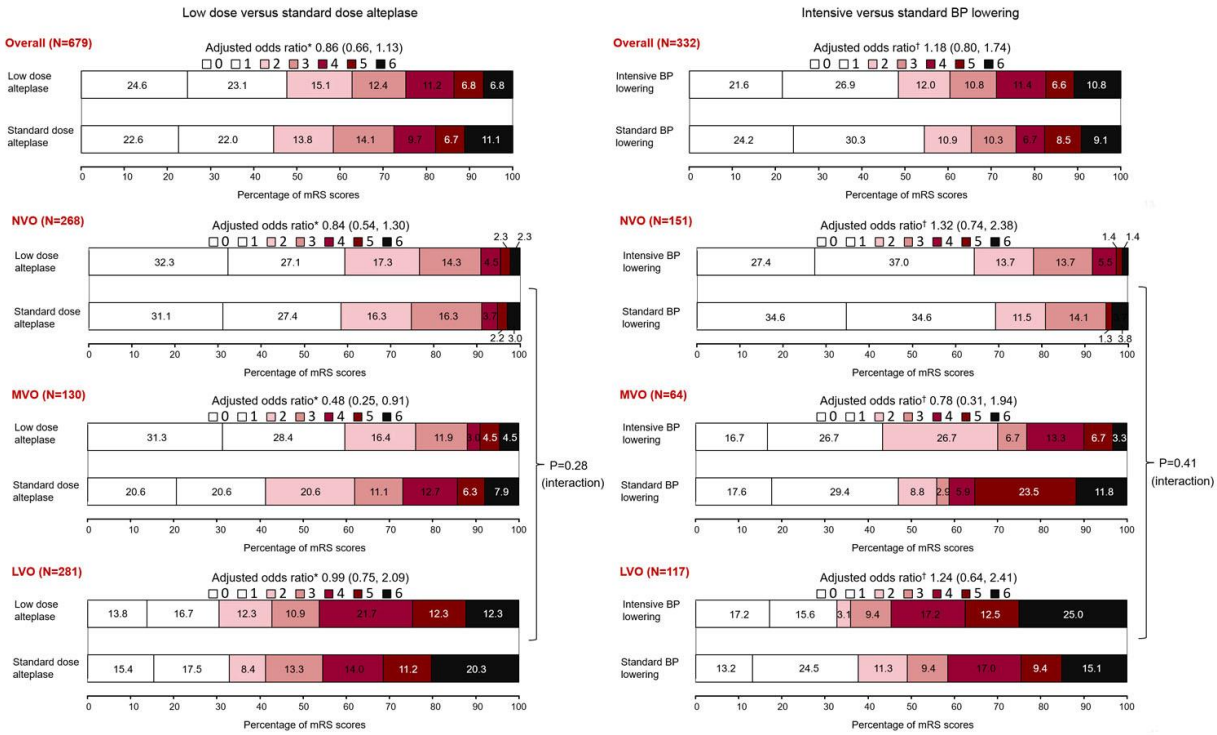
	NVO	MVO	LVO	P trend†
90-day functional outcomes				
mRS shift				
n/N (%) 0	117/382 (30.6)	39/179 (21.8)	57/360 (15.8)	
1	120/382 (31.4)	47/179 (26.3)	65/360 (18.1)	
2	59/382 (15.4)	33/179 (18.4)	35/360 (9.7)	
3	56/382 (14.7)	18/179 (10.1)	43/360 (11.9)	
4	14/382 (3.7)	16/179 (8.9)	61/360 (16.9)	
5	7/382 (1.8)	13/179 (7.3)	38/360 (10.6)	
6	9/382 (2.4)	13/179 (7.3)	61/360 (16.9)	
OR (95% CI)	Reference	**1.86 (1.35, 2.56)**	**3.94 (3.01, 5.16)**	**<0.001**
AOR (95% CI)*	Reference	1.34 (0.96, 1.88)	**2.13 (1.56, 2.90)**	**<0.001**
mRS 2–6				
n/N (%)	145/382 (38.0)	93/179 (52.0)	238/360 (66.1)	
OR (95% CI)	Reference	**1.77 (1.23, 2.53)**	**3.19 (2.36, 4.31)**	**<0.001**
AOR (95% CI)*	Reference	1.23 (0.82, 1.85)	**1.62 (1.11, 2.36)**	**0.007**
mRS 3–6				
n/N (%)	86/382 (22.5)	60/179 (33.5)	203/360 (56.4)	
OR (95% CI)	Reference	**1.74 (1.17, 2.57)**	**4.45 (3.24, 6.12)**	**<0.001**
AOR (95% CI)*	Reference	1.12 (0.71, 1.76)	**2.58 (1.74, 3.82)**	**<0.001**
mRS 6				
n/N (%)	9/388 (2.3)	13/183 (7.1)	61/369 (16.5)	
OR (95% CI)	Reference	**3.22 (1.35, 7.68)**	**8.34 (4.08, 17.06)**	**<0.001**
AOR (95% CI)*	Reference	1.82 (0.70, 4.79)	**4.12 (1.86, 9.11)**	**<0.001**
Safety outcomes within 7 days				
Any intracranial haemorrhage				
n/N (%)	37/388 (9.5)	41/183 (22.4)	100/369 (27.1)	
OR (95% CI)	Reference	**2.74 (1.69, 4.45)**	**3.53 (2.34, 5.31)**	**<0.001**
AOR (95% CI)*	Reference	**2.27 (1.34, 3.83)**	**2.01 (1.25, 3.26)**	**0.001**
sICH (ECASS2 criteria)				
n/N (%)	6/388 (1.5)	9/183 (4.9)	33/369 (8.9)	
OR (95% CI)	Reference	**3.29 (1.15, 9.40)**	**6.25 (2.59, 15.11)**	**<0.001**
AOR (95% CI)*	Reference	**3.42 (1.02, 11.48)**	**4.38 (1.65, 11.61)**	**0.002**
Other intermediate outcomes				
END or death within 24 hours				
n/N (%)	17/388 (4.4)	16/183 (8.7)	47/369 (12.7)	
OR (95% CI)	Reference	**2.09 (1.03, 4.24)**	**3.19 (1.79, 5.66)**	**<0.001**
AOR (95% CI)*	Reference	2.17 (0.999, 4.70)	**3.27 (1.70, 6.28)**	**<0.001**
Recanalisation within 48 hours				
n/N (%)	-	17/30 (56.7)	14/44 (31.8)	-

CI denotes confidence interval; END, early neurologic deterioration; ICH, intracerebral haemorrhage; LVO, large vessel obstruction; mRS, modified Rankin scale; MVO, medium vessel obstruction; NVO, no vessel obstruction; ECASS, the European-Australian Cooperative Acute Stroke Study; OR, odds ratio; AOR, adjusted odds ratio; sICH, symptomatic intracerebral haemorrhage.

*Adjusted for key prognostic covariates (age; sex; ethnicity; baseline systolic blood pressure; baseline NIHSS score; time from stroke onset to randomisation; history of atrial fibrillation; history of hypercholesterolemia; assigned to intensive blood pressure lowering group; and assigned to low-dose alteplase group).

†P trend for the odds ratios of MVO and LVO with the reference to NVO.

Figure 6.1. Association of randomised alteplase dose or BP lowering with ordinal mRS score across participants stratified by vascular obstruction status and site



BP denotes blood pressure; LVO, large vascular obstruction; mRS, modified Rankin scale; MVO, medium vascular obstruction; NVO, no vascular obstruction.
*Adjusted for minimized key prognostic covariates (age, sex, Asian ethnicity, baseline NIHSS score, time from stroke onset to randomisation) and assigned to intensive BP lowering group.
†Adjusted for minimized key prognostic covariates (age, sex, Asian ethnicity, baseline NIHSS score, time from stroke onset to randomisation) and assigned to low-dose alteplase group.

6.5.4 Vascular obstruction and intensive BP reduction

Associations of randomised BP target with functional outcomes in 333 included participants from the BP arm were also comparable to the main results of ENCHANTED (mRS shift, N=332 adjusted OR [95% CI] 1.18 [0.80-1.74]) (Figure 6.1), but the reduced risk of any intracranial haemorrhage by intensive BP lowering in the main trial was not confirmed (N=333, OR [95% CI] 0.88 [0.33-2.33]) (Figure 6.3). Associations of intensive versus guideline-recommended BP lowering with all outcomes were comparable across NVO/MVO/LVO groups, except for a borderline significant difference in the association with END across the subgroups ($P_{interaction}$=0.05), and for a trend towards increased END after thrombolysis in LVO (N=117, OR [95% CI] 2.69 [0.90-8.04], P=0.08) (Figures 6.1 and 6.3). However, this was not confirmed in sensitivity analyses (appendices 6.12, 6.15 and 6.16).

6.6 Discussion and conclusion

6.6.1 Discussion

In these post-hoc observational and subgroup analyses of the ENCHANTED trial, we were able to confirm prior reports that AIS patients with LVO had poor outcomes from thrombolysis compared to those with NVO, whereas outcomes were comparable between MVO and NVO. Although better functional recovery was evident after low-dose alteplase in patients with MVO, there were no differences in outcome between low- versus standard-dose alteplase across NVO/MVO/LVO groups. Similar associations for outcomes were seen for intensive versus guideline-recommended BP lowering across these subgroups, except for there being borderline significant difference for END. The results were inconsistent in sensitivity analyses, using different definitions of MVO, but where statistical power was reduced after the exclusion of those with site-reported angiography data.

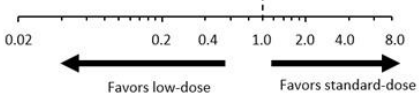
We undertook these vascular analyses to provide more insights into any differential thrombolysis outcomes according to the differential use of low-dose alteplase and intensive BP lowering. After merging eligible participants from both arms and randomised groups into a single cohort, the baseline characteristics of ENCHANTED participants were generally consistent with other studies according to any vascular obstruction.[4,13,14] As reported elsewhere,[3,5,9,13,14] thrombolysis outcomes were worse in those with LVO compared to NVO, with nearly half and one third of ENCHANTED participants having mRS scores 2-6 and 3-6 at 90 days for MVO. Low statistical power from there being a limited number of participants with both baseline and follow-up CTA or MRA hindered reliability of our recanalisation estimates. However, the higher frequency of recanalisation in MVO than LVO was as expected, as efficacy of alteplase is likely to be greater in MVO than LVO. Thrombus migration from LVO to MVO after intravenous alteplase administration is another explanation.

We have been unable to define who will benefit from low-dose intravenous alteplase in AIS. Several post-hoc subgroup analyses of the ENCHANTED trial have shown that various clinical factors (age, ethnicity, severity, prior stroke, diabetes mellitus, use of lipid lowering agents, and kidney function) cannot modify the neutral effects of low- versus standard-dose alteplase on 90-day functional outcomes, except for prior use of antiplatelets.[15] Acute or chronic cerebral ischaemic signs on non-contrast CT were also explored without any significant findings.[8] These analyses suggest low-dose alteplase may improve functional outcome in thrombolysis-treated AIS patients with MVO. Despite support for a lower dose of alteplase being effective in MVO

on the basis of smaller diameter of occluded vessel compared to LVO, we have been cautious in interpreting our findings as spurious findings may have arisen when the main trial produced a neutral result and there were small sample sizes for these subgroups.

Figure 6.2. Other thrombolysis outcomes by randomised alteplase dose across participants stratified by vascular obstruction status and site

Thrombolysis outcomes	Alteplase dose group, n/N (%) Low	Standard	Odds ratio (95% CI)		P inter-action
mRS 2-6: overall	177/338 (52.4)	189/341 (55.4)		0.91 (0.65, 1.29)*	
NVO	54/133 (40.6)	56/135 (41.5)		0.94 (0.55, 1.60)*	0.12*
MVO	27/67 (40.3)	37/63 (58.7)		0.45 (0.21, 0.97)*	
LVO	96/138 (69.6)	96/143 (67.1)		1.29 (0.74, 2.27)*	
mRS 3-6: overall	126/338 (37.3)	142/341 (41.6)		0.84 (0.59, 1.20)*	
NVO	31/133 (23.3)	34/135 (25.2)		0.81 (0.45, 1.48)*	0.46*
MVO	16/67 (23.9)	24/63 (38.1)		0.49 (0.20, 1.22)*	
LVO	79/138 (57.2)	84/143 (58.7)		1.03 (0.61, 1.76)*	
mRS 6: overall	23/350 (6.6)	38/347 (11.0)		0.57 (0.33, 0.98)	
NVO	3/138 (2.2)	4/136 (2.9)		0.73 (0.16, 3.34)	0.94
MVO	3/68 (4.4)	5/65 (7.7)		0.55 (0.13, 2.42)	
LVO	17/144 (11.8)	29/146 (19.9)		0.54 (0.28, 1.03)	
Intracranial hemorrhage†: overall	70/350 (20.0)	73/347 (21.0)		0.96 (0.66, 1.40)*	
NVO	14/138 (10.1)	16/136 (11.8)		0.79 (0.36, 1.72)*	0.81*
MVO	14/68 (20.6)	15/65 (23.1)		0.89 (0.38, 2.08)*	
LVO	42/144 (29.2)	42/146 (28.8)		1.11 (0.66, 1.87)*	
sICH (ECASS2): overall	13/350 (3.7)	24/347 (6.9)		0.52 (0.26, 1.04)	
NVO	1/138 (0.7)	3/136 (2.2)		0.32 (0.03, 3.15)	0.48
MVO	1/68 (1.5)	5/65 (7.7)		0.18 (0.02, 1.58)	
LVO	11/144 (7.6)	16/146 (11.0)		0.67 (0.30, 1.50)	
END or death (24 hrs): overall	25/350 (7.1)	31/347 (8.9)		0.78 (0.45, 1.36)	
NVO	6/138 (4.3)	6/136 (4.4)		0.98 (0.31, 3.13)	0.31
MVO	2/68 (2.9)	7/65 (10.8)		0.25 (0.05, 1.26)	
LVO	17/144 (11.8)	18/146 (12.3)		0.95 (0.47, 1.93)	
No recanalization (48 hrs): overall	11/28 (39.3)	21/33 (63.6)		0.37 (0.13, 1.04)	
NVO	—	—		—	—
MVO	1/10 (10.0)	6/12 (50.0)		0.11 (0.01, 1.17)	
LVO	10/18 (55.6)	15/21 (71.4)		0.50 (0.13, 1.88)	

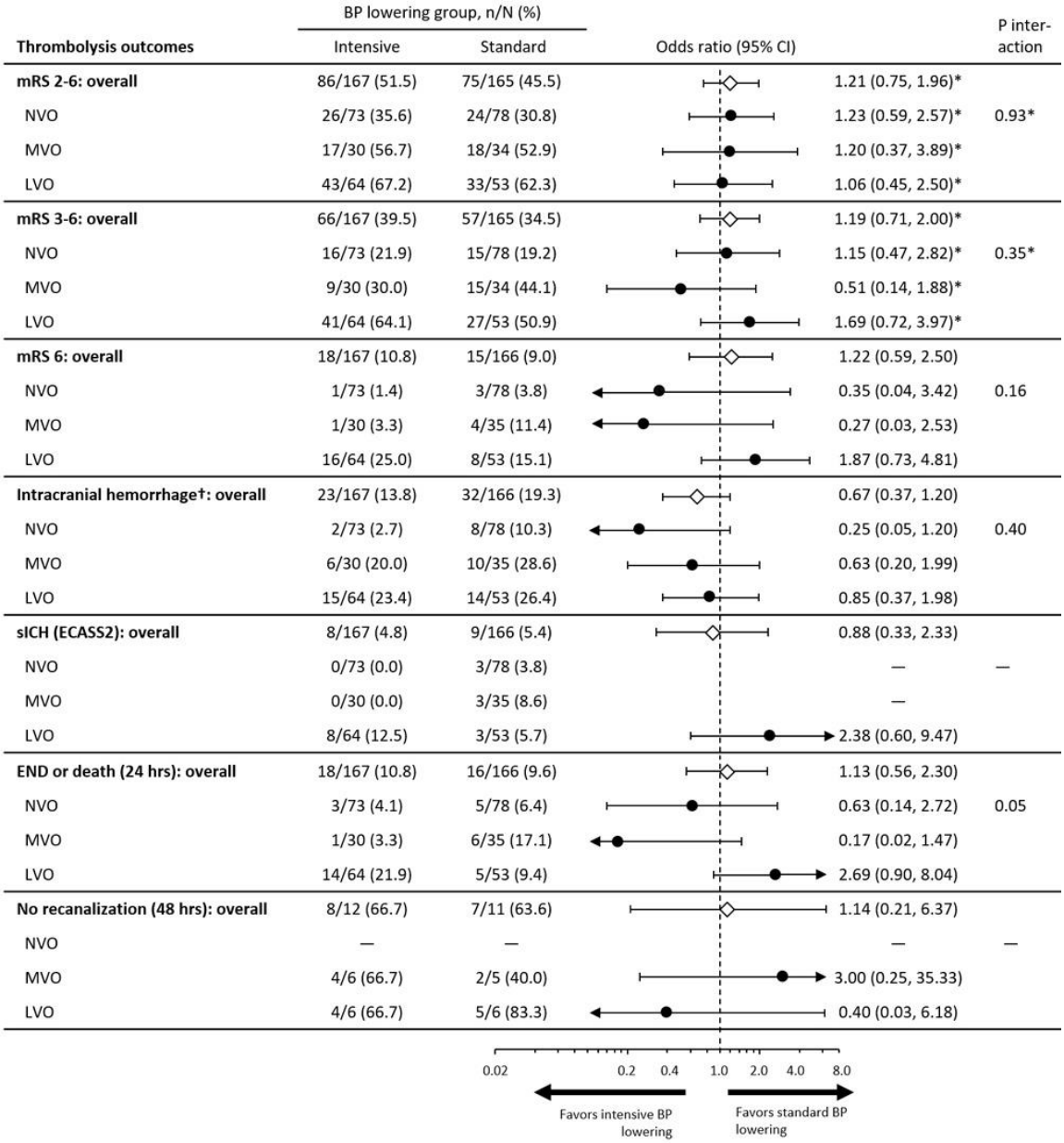


BP denotes blood pressure; END, early neurologic deterioration; ICH, intracerebral haemorrhage; LVO, large vessel obstruction; mRS, modified Rankin scale; MVO, medium vessel obstruction; ECASS, the European-Australian Cooperative Acute Stroke Study; NVO, no vascular obstruction; sICH, symptomatic intracerebral haemorrhage.

*Adjusted for minimized key prognostic covariates (age, sex, Asian ethnicity, baseline NIHSS score, time from stroke onset to randomisation) and assigned to intensive BP lowering group.

†Adjudicated centrally or reported by site investigators.

Figure 6.3. Other thrombolysis outcomes by randomised BP lowering across participants stratified by vascular obstruction status and site



BP denotes blood pressure; END, early neurologic deterioration; ICH, intracerebral haemorrhage; LVO, large vessel obstruction; mRS, modified Rankin scale, MVO: medium vessel obstruction; ECASS, the European-Australian Cooperative Acute Stroke Study; NVO, no vascular obstruction; sICH, symptomatic intracerebral haemorrhage.
*Adjusted for minimized key prognostic covariates (age, sex, Asian ethnicity, baseline NIHSS score, time from stroke onset to randomisation) and assigned to low-dose alteplase group.
†Adjudicated centrally or reported by site investigators.

There are ongoing concerns that BP lowering may increase the risk of harm in AIS patients with LVO or large infarct lesion.[16] Our results suggest greater END after thrombolysis for LVO participants allocated to intensive BP lowering treatment, but the play of chance cannot be excluded in relation to the small subset of participants in these particular analyses. However, our recent post-hoc analysis with larger samples (N=1311) that included ENCHANTED BP

arm participants with severe AIS (defined by the combination of LVO and baseline NIHSS score >10),[17] as well as another analysis in participants with definite non-lacunar AIS (N=784),[18] both showed an increased risk of death in those who were allocated to intensive BP lowering, which adds to the concern of large BP falls in these types of AIS patients.

6.6.2 Strengths and limitations

Strengths of this study were the inclusion of patients from a large, prospective, multicentre, clinical trial with systematic and complete data collection, where vascular assessments were completed blind to clinical data, using a pre-specified approach. However, several limitations include the small subgroup samples on opportunistic baseline CTA or MRA which were not pre-specified in our pragmatic trial design. Although adjustments were made for key prognostic covariables in analyses of randomised treatment effects and for assessments of interaction by subgroups, there is still the concerns over bias, residual confounding and chance. There were few patients with cervical CTA or MRA, which compromised the ability to identify and separate extracranial ICAO from LVO, with the former considered to have a better prognosis than intracranial ICAO with tandem obstructions.[19] We also included site-reported angiography data (not adjudicated centrally) in the primary analysis to enhance statistical power, but this did not allow reclassification of MVO and they were removed in sensitivity analyses. Furthermore, we did not identify early recanalisation and reperfusion status (within 6 hours post-thrombolysis), which are important in determining functional recovery. Nonetheless, our study might give light to future research in exploring whether AIS patients with MVO would benefit from low-dose alteplase and whether caution should be advised for intensive BP lowering in thrombolysis-treated LVO patients.

6.6.3 Conclusion

In summary, among thrombolysis eligible and treated AIS participants with baseline CTA or MRA from ENCHANTED, clinical prognosis is worse in those with LVO, and comparable for MVO, compared to others with NVO. Functional outcomes after low- versus standard-dose alteplase or intensive versus guideline-recommended BP lowering are not modified by different vascular obstruction status/site according to these post-hoc subgroup analyses from ENCHANTED which were statistically underpowered. Whether prognosis in MVO AIS patients is better after low- versus standard-dose alteplase treatment requires further research, as dose intensive BP lowering in LVO.

References

1. Puig J, Shankar J, Liebeskind D, et al. From "Time is Brain" to "Imaging is Brain": A Paradigm Shift in the Management of Acute Ischaemic Stroke. J Neuroimaging 2020;30(5):562-71.
2. De Silva DA, Brekenfeld C, Ebinger M, et al. The benefits of intravenous thrombolysis relate to the site of baseline arterial occlusion in the Echoplanar Imaging Thrombolytic Evaluation Trial (EPITHET). Stroke 2010;41(2):295-9.
3. Mair G, von Kummer R, Adami A, et al. Arterial Obstruction on Computed Tomographic or Magnetic Resonance Angiography and Response to Intravenous Thrombolytics in Ischaemic Stroke. Stroke 2017;48(2):353-60.
4. Kimura K, Iguchi Y, Shibazaki K, et al. Intravenous tissue plasminogen activator thrombolysis in patients without major arterial occlusion seems to be safe and effective. Eur Neurol 2010;64(5):258-64.
5. Ospel JM, Menon BK, Demchuk AM, et al. Clinical Course of Acute Ischaemic Stroke Due to Medium Vessel Occlusion With and Without Intravenous Alteplase Treatment. Stroke 2020; 51(11):3232-40.
6. Anderson CS, Robinson T, Lindley RI, et al. Low-dose versus standard-dose intravenous alteplase in acute ischaemic stroke. N Engl J Med 2016;374(24):2313-23.
7. Anderson CS, Huang Y, Lindley RI, et al. Intensive blood pressure reduction with intravenous thrombolysis therapy for acute ischaemic stroke (ENCHANTED): an international, randomised, open-label, blinded-endpoint, phase 3 trial. Lancet 2019;393(10174):877-88.
8. Delcourt C, Wang X, Zhou Z, et al. Brain imaging abnormalities and outcome after acute ischaemic stroke: the ENCHANTED trial. J Neurol Neurosurg Psychiatry 2020;91(12):1290-6.
9. Tan IY, Demchuk AM, Hopyan J, et al. CT Angiography Clot Burden Score and Collateral Score: Correlation With Clinical and Radiologic Outcomes in Acute Middle Cerebral Artery Infarct. Am J Neuroradiol 2009;30(3):525-31.
10. Menon BK, Al-Ajlan FS, Najm M, et al. Association of Clinical, Imaging, and Thrombus Characteristics With Recanalisation of Visible Intracranial Occlusion in Patients With Acute Ischaemic Stroke. JAMA 2018;320(10):1017-26.
11. Menon BK, d'Esterre CD, Qazi EM, et al. Multiphase CT Angiography: A New Tool for the Imaging Triage of Patients With Acute Ischaemic Stroke. Radiology 2015;275(2):510-20.
12. Hacke W, Kaste M, Fieschi C, et al. Randomised double-blind placebo controlled trial of thrombolytic therapy with intravenous alteplase in acute ischaemic stroke (ECASS II). Lancet 1998;352(9136):1245-51.
13. Rai A, Cline B, Williams E, et al. Intravenous thrombolysis outcomes in patients presenting with large vessel acute ischaemic strokes--CT angiography-based prognosis. J Neuroimaging 2015;25(2):238-42.
14. Giralt-Steinhauer E, Ois A, Abilleira S, et al. Frequency and outcome of total anterior circulation strokes without intracranial large-vessel occlusion. Eur J Neurol 2017;24(1):11-17.

15. Robinson TG, Wang X, Arima H, et al. Low- Versus Standard-Dose Alteplase in Patients on Prior Antiplatelet Therapy: The ENCHANTED Trial (Enhanced Control of Hypertension and Thrombolysis Stroke Study). Stroke 2017;48(7):1877-83.
16. Petersen NH, Ortega-Gutierrez S, Wang A, et al. Decreases in blood pressure during thrombectomy are associated with larger infarct volumes and worse functional outcome. Stroke 2019;50(7):1797-804.
17. Minhas JS, Wang X, Lindley RI, et al. Comparative effects of intensive-blood pressure versus standard-blood pressure-lowering treatment in patients with severe ischaemic stroke in the ENCHANTED trial. J Hypertens 2021;39(2):280-5.
18. Zhou Z, Xia C, Carcel C, et al. Intensive versus guideline-recommended blood pressure reduction in acute lacunar stroke with intravenous thrombolysis therapy: the ENCHANTED trial. Eur J Neurol 2021;28(3):783-93.
19. Malhotra K, Goyal N, Tsivgoulis G. Internal Carotid Artery Occlusion: Pathophysiology, Diagnosis, and Management. Curr Atheroscler Rep 2017;19(10):41.

Appendices

Appendix 6.1. Scan quality of computed tomographic or magnetic resonance angiography

Good: meet all following conditions.
Moderate: meet 2 to 3 of following conditions and it is probably to have a reliable assessment.
Poor: meet 1 to 2 of following conditions and it is unlikely to have a reliable assessment.

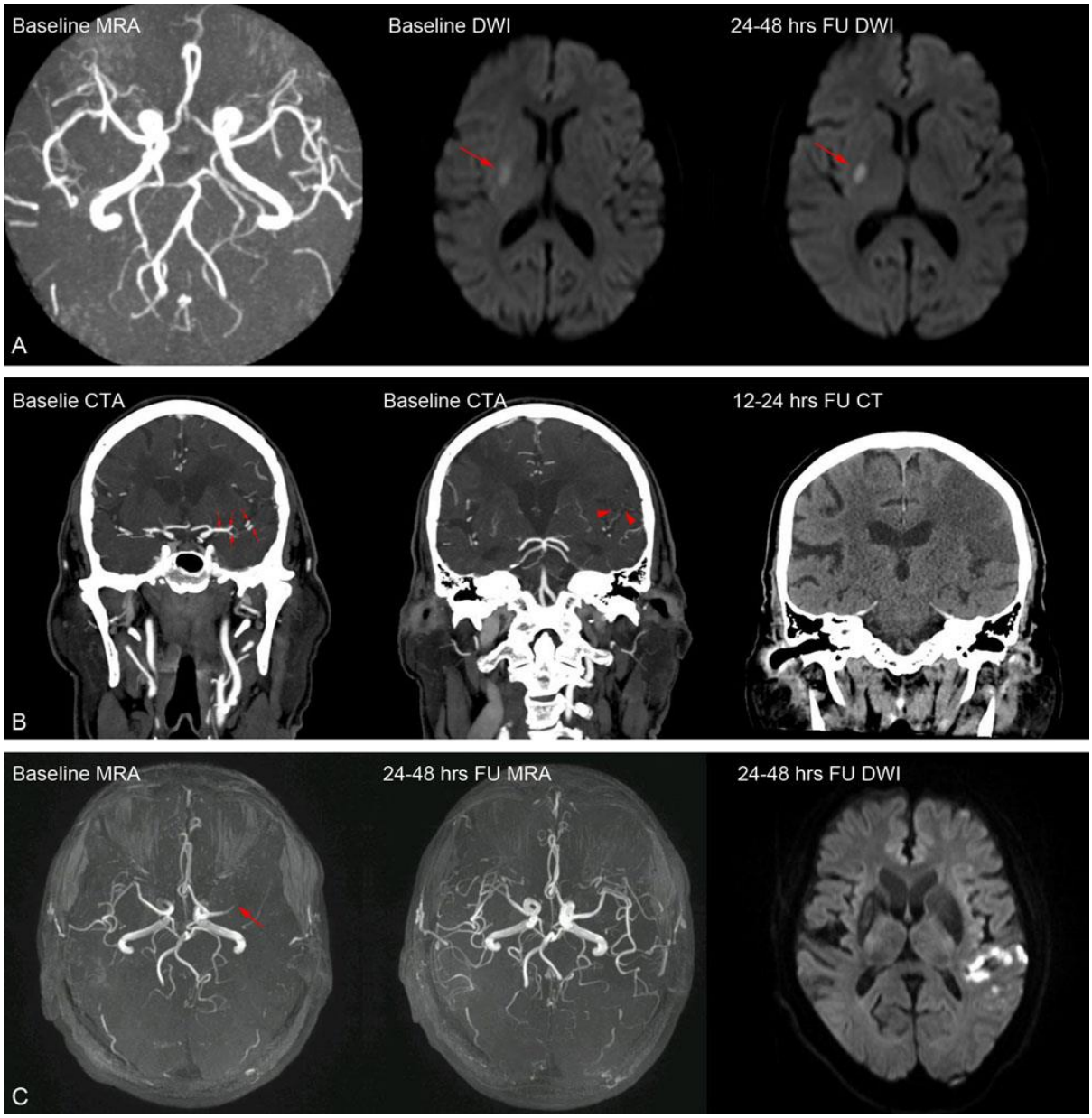
1. No motion artifact.
2. No mal-positioning of patients in scanner.
3. Having raw images besides reconstructed maximum intensity projection (MIP) images.
4. Relative high signal-to-noise ratio (SNR).

Appendix 6.2. Scoring for angiogram characteristics in ENCHANTED

Imaging characteristic	Options presented to imaging reader
Presence of vascular stenosis or occlusion?	1. Yes 2. No
Which side is affected?	1. Right 2. Left 3. Both or midline
Location of the stenosis/occlusion (intracranial arteries): (Select 1 to 3 largest arteries involved)	1. Anterior Cerebral Artery (ACA) 2. M1 segment of Middle Cerebral Artery (MCA) 3. M2 segment of MCA* 4. Terminal Internal Carotid Artery (ICA)† 5. Petrous to supraclinoid segment of ICA 6. Posterior Cerebral Artery (PCA) 7. Basilar Artery (BA) 8. Vertebral Artery (VA) *Further classified as proximal M2 segment or distal M2/M3 segment separately † Further classified as L/T or I occlusion separately
Location of the stenosis/occlusion (extracranial carotid artery):	1. Cervical segment of ICA 2. Bifurcation of common carotid artery 3. Common carotid artery
IST-3 Angiography Score[3]	0 = No patency 1 = Contrast penetrates obstruction but minimal enters distal artery 2a = Luminal patency of <50% with partial filling (< half) of major branches 2b = Luminal patency of <50% with partial filling (> half) of major branches 3 = Patency of >50% of lumen and filling of most branches 4 = Complete patency – normal
Modified TICI Score[3]	0 = No flow/patency 1 = Minimal flow/patency 2a = Partial flow <50% of expected territory 2b = Partial flow >50% of expected territory 3 = Complete flow/patency
Clot Burden Score[9] (Anterior circulation only)	Six arterial locations are assessed for the presence of clot: Infraclinoid (1 point) and supraclinoid ICA (2 points) Proximal (2 points) and distal M1 MCA (2 points) M2 branches of MCA (1 point each) ACA (1 point)
Visual Residual Flow[10]	1. Grade 0 (no residual flow) 2. Grade 1 3. Grade 2 4. Not eligible for assessment
Collateral Supply (MCA)[11]	Grade 0 (poor): When compared with the asymptomatic contralateral hemisphere, there are no vessels visible within the ischaemic territory. Grade 1 (poor): When compared with the asymptomatic contralateral hemisphere, there are just a few vessels visible in the occluded vascular territory. Grade 2 (intermediate): When compared with the asymptomatic contralateral hemisphere, there is decreased prominence and extent and regions with no vessels within the ischaemic territory in the symptomatic hemisphere. Grade 3 (intermediate): When compared with the asymptomatic contralateral hemisphere, there is moderately reduced prominence and extent of pial vessels within the ischaemic territory in the symptomatic hemisphere. Grade 4 (good): When compared with the asymptomatic contralateral hemisphere, there is slightly reduced prominence and extent of pial vessels within the ischaemic territory in the symptomatic hemisphere.

	Grade 5 (good): When compared with asymptomatic contralateral hemisphere, there is increased or normal prominence and extent of pial vessels within the ischaemic territory in the symptomatic hemisphere.
Revised Arterial Occlusive Lesion [rAOL] Score for Recanalisation[10]	Score 0a: Primary occlusive lesion gets worsen with severer stenosis/occlusion degree or more segments of vessel being involved. Score 0b: Primary occlusive lesion remains same. Score 1: Debulking of thrombus without recanalisation. Score 2a: Partial or complete recanalisation of the primary lesion with thrombus/occlusion in major vascular branch*. Score 2b: Partial or complete recanalisation of the primary lesion with thrombus/occlusion in minor vascular branch†, or partial recanalisation of the primary lesion with no thrombus in the vascular tree at or beyond the primary occlusive lesion. Score 3: Complete recanalisation of the primary occlusion with no clot in the vascular tree at or beyond The primary occlusive lesion. *Major vascular branch: ICA, M1 segment of MCA, Functional M1(thrombus in both proximal M2s of MCA), A1 segment of ACA, BA, P1 segment of PCA. †Minor vascular branch: other distal vessels.

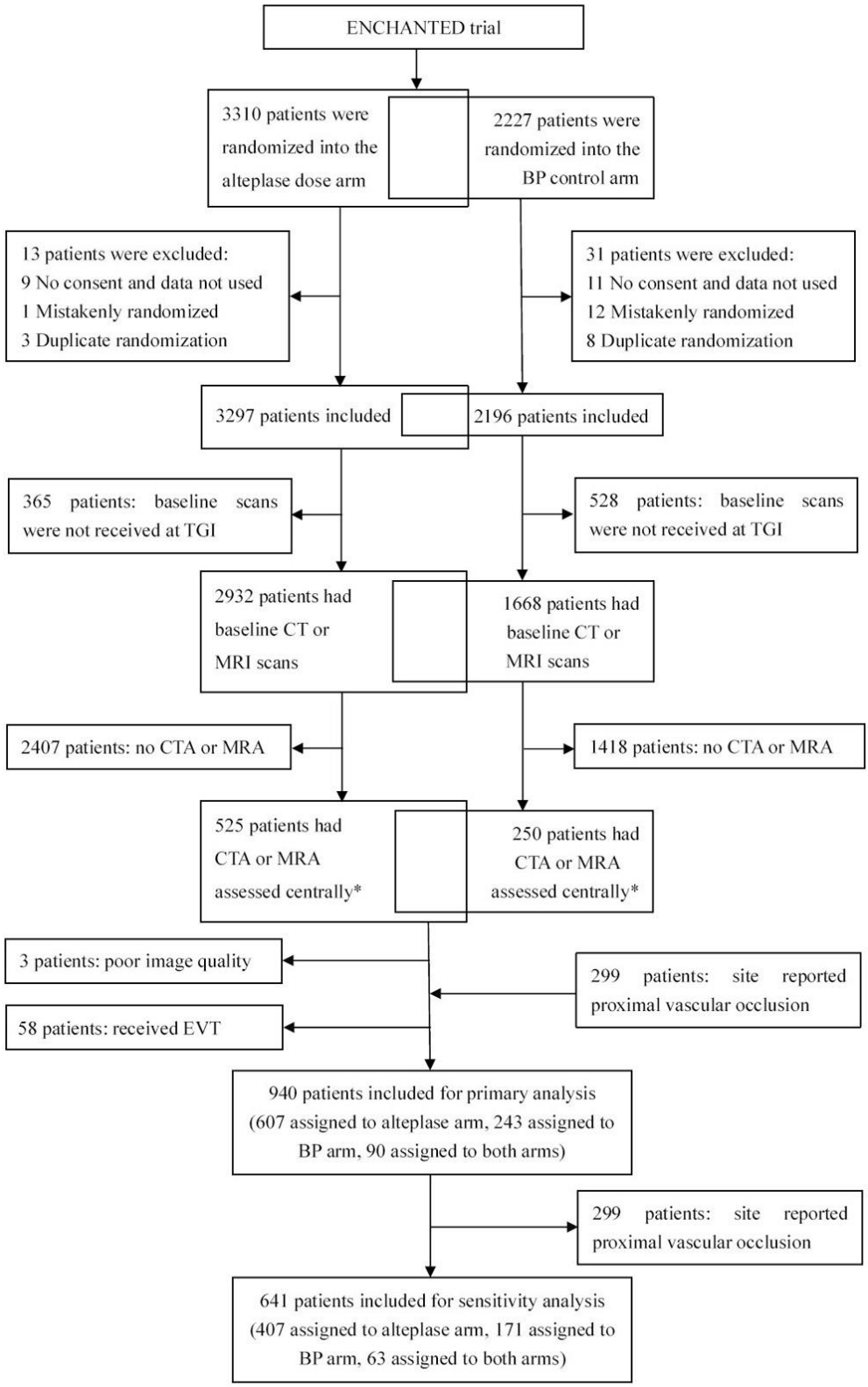
Appendix 6.3. Examples of ENCHANTED participants with different vascular obstruction status and site



CTA denotes computed tomographic angiography; DWI, diffusion-weighted imaging; ENCHANTED, Enhanced Control of Hypertension and Thrombolysis Stroke Study; FU, follow-up; LVO, large vascular obstruction; MCA, middle cerebral artery; MRA, magnetic resonance angiography; MVO, medium vascular obstruction; NVO, no vascular obstruction.

(A) case of NVO: no vascular obstruction on baseline MRA and lacunar infarct at right basal ganglia (red arrow) is identified on baseline and follow-up DWI; (B) case of MVO: left M1 and proximal M2 segment of MCA is shown on baseline CTA (red arrow) with fewer sylvian branches (red arrowhead) compared to contralateral side. Infarct at left temporal lobe is seen on follow-up CT; (C) case of LVO: obstruction at left M1 segment of MCA with recanalisation and infarct at left temporal lobe identified on follow-up MRA and DWI.

Appendix 6.4. Flowchart of participants included in analyses



BP denotes blood pressure; CT, computed tomography; CTA, CT angiography; EVT, endovascular therapy; MRA, magnetic resonance angiography; MRI, magnetic resonance imaging; TGI, the George Institute for Global Health.
*702 patients in total with 452 assigned to alteplase arm, 177 assigned to BP arm, and 73 assigned to both arms.

Appendix 6.5. Baseline characteristics of ENCHANTED rtPA arm participants included and excluded for vascular analyses

	Included (N=697)	Excluded (N=2600)	P value
Age (years)	67.6 (12.8)	66.3 (12.8)	**0.01**
Female	251 (36.0)	997 (38.3)	0.26
Asian ethnicity	347 (49.8)	1732/2594 (66.8)	**<0.001**
Clinical features			
Systolic BP (mmHg)	147.1 (20.6)	149.8 (19.5)	**0.001**
Diastolic BP (mmHg)	83.3 (12.7)	85.0 (12.9)	**0.002**
Heart rate (beats per minute)	80.3 (16.3)	78.7 (15.1)	**0.01**
NIHSS score*	8 (5-14)	8 (5-14)	0.71
GCS score†	15 (14-15)	15 (14-15)	0.36
Medical history			
Previous stroke	104 (14.9)	485 (18.7)	**0.02**
Hypertension	435 (62.4)	1630/2591 (62.9)	0.81
Atrial fibrillation	149 (21.4)	487/2588 (18.8)	0.13
Coronary artery disease	101 (14.5)	378/2591 (14.6)	0.95
Valvular/other heart disease	58 (8.3)	177/2591 (6.8)	0.18
Diabetes mellitus	124 (17.8)	522/2591 (20.1)	0.16
Hypercholesterolaemia	179 (25.7)	376/2591 (14.5)	**<0.001**
Current smoker	170/696 (24.4)	600/2588 (23.2)	0.49
Pre-stroke function without symptoms (mRS=0)	145 (20.8)	467/2589 (18.0)	0.10
Medication on admission			
Antihypertensive agent(s)	344 (49.4)	1154/2591 (44.5)	**0.02**
Warfarin anticoagulation	24/696 (3.4)	58/2589 (2.2)	0.07
Aspirin/other antiplatelet agent	194/696 (27.9)	558/2589 (21.6)	**<0.001**
Statin/other lipid lowering agent	182/696 (26.1)	433/2588 (16.7)	**<0.001**
Time from onset to randomisation (hrs)	2.4 (1.8-3.2)	2.7 (2.1-3.5)	**<0.001**
Randomised low-dose treatment	350 (50.2)	1304 (50.2)	0.98

Data are n (%), mean (SD), or median (Q1, Q3). P values are based on Chi-square, analysis of variance, or Wilcoxon signed-rank test

BP denotes blood pressure; GCS, Glasgow coma scale; mRS, modified Rankin scale; NIHSS, National Institutes of Health stroke scale.

*Scores on the NIHSS range from 0 to 42, with higher scores indicating more severe neurological deficits.

†Scores on the GCS range from 15 (normal) to 3 (deep coma).

Appendix 6.6. Baseline characteristics of ENCHANTED BP arm participants included and excluded for vascular analyses

	Included (N=333)	Excluded (N=1863)	P value
Age (years)	69.7 (12.5)	66.4 (12.1)	**<0.001**
Female	129 (38.7)	706 (37.9)	0.77
Asian ethnicity	133 (39.9)	1485/1861 (79.8)	**<0.001**
Clinical features			
Systolic BP (mmHg)	167.4 (9.1)	164.9 (9.2)	**<0.001**
Diastolic BP (mmHg)	89.7 (11.9)	91.2 (11.4)	**0.03**
Heart rate (beats per minute)	80.9 (16.3)	79.0 (14.5)	**0.03**
NIHSS score*	8 (5-13)	7 (4-12)	0.28
GCS score†	15 (14-15)	15 (14-15)	0.94
Medical history			
Previous stroke	60 (18.0)	354 (19.0)	0.67
Hypertension	227 (68.2)	1341/1859 (72.1)	0.14
Atrial fibrillation	60 (18.0)	252/1857 (13.6)	**0.03**
Coronary artery disease	46 (13.8)	263/1859 (14.1)	0.87
Valvular/other heart disease	18 (5.4)	76/1859 (4.1)	0.27
Diabetes mellitus	79 (23.7)	417/1859 (22.4)	0.60
Hypercholesterolaemia	79 (23.7)	170/1859 (9.1)	**<0.001**
Current smoker	58 (17.4)	386/1857 (20.8)	0.16
Pre-stroke function without symptoms (mRS=0)	81 (24.3)	233/1858 (12.5)	**<0.001**
Medication on admission			
Antihypertensive agent(s)	189 (56.8)	823/1859 (44.3)	**<0.001**
Warfarin anticoagulation	8 (2.4)	21/1859 (1.1)	0.06
Aspirin/other antiplatelet agent	102 (30.6)	284/1859 (15.3)	**<0.001**
Statin/other lipid lowering agent	106 (31.8)	232/1859 (12.5)	**<0.001**
Time from onset to randomisation (hrs)	3.2 (2.5-4.1)	3.4 (2.6-4.1)	0.49
Randomised intensive BP lowering treatment	167 (50.2)	914 (49.1)	0.71

Data are n (%), mean (SD), or median (Q1, Q3). P values are based on Chi-square, analysis of variance, or Wilcoxon signed-rank test

BP denotes blood pressure; GCS, Glasgow coma scale; mRS, modified Rankin scale; NIHSS, National Institutes of Health stroke scale.

*Scores on the NIHSS range from 0 to 42, with higher scores indicating more severe neurological deficits.

†Scores on the GCS range from 15 (normal) to 3 (deep coma).

Appendix 6.7. Baseline characteristics and presumed diagnosis of participants by vascular obstruction status and site

	NVO (N=388)	MVO (N=183)	LVO (N=369)	P value[§]
Age (years)	67.0 (13.3)	69.4 (11.9)	68.4 (12.5)	0.08
Female	140 (36.1)	73 (39.9)	135 (36.6)	0.66
Asian ethnicity	130 (33.5)	94 (51.4)	222 (60.2)	**<0.001**
Clinical features				
Systolic BP (mmHg)	154.7 (19.5)	153.2 (18.4)	149.5 (21.9)	**0.002**
Diastolic BP (mmHg)	85.8 (12.9)	85.3 (12.2)	83.8 (12.9)	0.09
Heart rate (beats per minute)	80.9 (14.9)	78.6 (15.9)	81.1 (18.0)	0.21
NIHSS score[*]	6 (4-9)	9 (5-12)	12 (7-17)	**<0.001**
GCS score[†]	15 (15-15)	15 (13-15)	15 (13-15)	**<0.001**
Medical history				
Previous stroke	65 (16.8)	31 (16.9)	51 (13.8)	0.47
Hypertension	230 (59.3)	126 (68.9)	238 (64.5)	0.07
Atrial fibrillation	45 (11.6)	39 (21.3)	103 (27.9)	**<0.001**
Coronary artery disease	50 (12.9)	29 (15.8)	52 (14.1)	0.63
Valvular or other heart disease	26 (6.7)	17 (9.3)	25 (6.8)	0.49
Diabetes mellitus	73 (18.8)	39 (21.3)	74 (20.1)	0.77
Hypercholesterolemia	116 (29.9)	48 (26.2)	65 (17.6)	**<0.001**
Current smoker	81/387 (20.9)	33 (18.0)	95 (25.7)	0.09
Pre-stroke function without symptoms[‡]	83 (21.4)	47 (25.7)	69 (18.7)	0.17
Medication on admission				
Antihypertensive agent(s)	191 (49.2)	102 (55.7)	186 (50.4)	0.34
Warfarin anticoagulation	11 (2.8)	8 (4.4)	9/368 (2.4)	0.45
Aspirin/other antiplatelet agent	120 (30.9)	52 (28.4)	92/368 (25.0)	0.19
Statin/other lipid lowering agent	136 (35.1)	48 (26.2)	78/368 (21.2)	**<0.001**
Time from onset to randomisation (hrs)	2.9 (2.1-3.6)	2.7 (1.9-3.6)	2.6 (1.8-3.5)	**0.04**
Imaging features				
Infarct location confirmed	225/385 (58.4)	147/162 (90.7)	299/333 (89.8)	**<0.001**
Infarct on left side	104/225 (46.2)	77/147 (52.4)	129/299 (43.1)	0.18
Infarct on right side	111/225 (49.3)	68/147 (46.3)	156/299 (52.2)	0.49
Infarct at midline or bilateral	10/225 (4.4)	2/147 (1.4)	14/299 (4.7)	0.20
Infarct in anterior circulation	164/225 (72.9)	108/147 (73.5)	236/299 (78.9)	0.22
Infarct in posterior circulation	53/225 (23.6)	32/147 (21.8)	37/299 (12.4)	**0.002**
Both anterior and posterior circulation	8/225 (3.6)	7/147 (4.8)	26/299 (8.7)	**0.04**
With hyperdense vessel sign on CT	19/340 (5.6)	34/147 (23.1)	116/305 (38.0)	**<0.001**
With old vascular lesions	166/385 (43.1)	61/162 (37.7)	151/333 (45.3)	0.27
With brain atrophy	260/385 (67.5)	109/162 (67.3)	235/333 (70.6)	0.63
With white matter changes	163/385 (42.3)	63/162 (38.9)	141/333 (42.3)	0.72
Clot burden score	-	9 (9-9), N=64	6 (4-7), N=122	**<0.001**
With visual residual flow (grade 1 or 2)	-	65/93 (69.9)	79/156 (50.6)	**0.003**
Good collaterals (grade 3 to 5)	-	64/93 (68.8)	88/155 (56.8)	0.06
Assigned to low-dose alteplase group	138/274 (50.4)	68/133 (51.1)	144/290 (49.7)	0.96
Assigned to intensive BP lowering group	73/151 (48.3)	30/65 (46.2)	64/117 (54.7)	0.45
Presumed clinical diagnosis				
Non-stroke	27/383 (7.0)	1 (0.5)	2/363 (0.6)	**<0.001**
LVO because of atheroma	69/383 (18.0)	73 (39.9)	204/363 (56.2)	**<0.001**
Small vessel disease	119/383 (31.1)	15 (8.2)	16/363 (4.4)	**<0.001**
Cardioembolism	68/383 (17.8)	54 (29.5)	89/363 (24.5)	**0.004**
Dissection	2/383 (0.5)	0 (0.0)	6/363 (1.7)	0.09
Other or uncertain pathogenesis	98/383 (25.6)	40 (21.9)	46/363 (12.7)	**<0.001**

BP denotes blood pressure; CT, computed tomography; GCS, Glasgow Coma Scale; LVO, large vessel obstruction; MVO, medium vessel obstruction; mRS, modified Rankin scale; NIHSS, National Institutes of Health stroke scale; NVO, no vessel obstruction.

Data are n (%), mean (SD), or median (Q1, Q3). P values are based on Chi-square, analysis of variance, or Wilcoxon signed-rank test.
*Scores on the NIHSS range from 0 to 42, with higher scores indicating more severe neurological deficits.
†Scores on the GCS range from 15 (normal) to 3 (deep coma).
‡mRS=0.
§P values for the comparisons across participants with NVO, MVO and LVO.

Appendix 6.8. Baseline characteristics of participants by randomised alteplase dose in different vascular obstruction status and site

	Low-dose NVO (n=138), MVO (n=68), LVO (n=144)	Standard-dose NVO (n=136), MVO (n=65), LVO (n=146)	Total NVO (n=274), MVO (n=133), LVO (n=290)	P value[§]
Age (years)				
NVO	66.9 (13.5)	66.1 (13.6)	66.5 (13.5)	0.17
MVO	68.7 (11.2)	69.0 (12.5)	68.8 (11.8)	
LVO	68.4 (12.0)	67.8 (13.0)	68.1 (12.5)	
Female				
NVO	48 (34.8)	49 (36.0)	97 (35.4)	0.71
MVO	30 (44.1)	22 (33.8)	52 (39.1)	
LVO	47 (32.6)	55 (37.7)	102 (35.2)	
Asian ethnicity				
NVO	51 (37.0)	42 (30.9)	93 (33.9)	**<0.001**
MVO	37 (54.4)	33 (50.8)	70 (52.6)	
LVO	90 (62.5)	94 (64.4)	184 (63.4)	
Clinical features				
Systolic BP (mmHg)				
NVO	149.0 (19.1)	149.2 (20.8)	149.1 (19.9)	**0.04**
MVO	148.8 (18.5)	147.1 (18.5)	148.0 (18.5)	
LVO	144.1 (22.7)	145.5 (21.4)	144.8 (22.0)	
Diastolic BP (mmHg)				
NVO	84.0 (13.3)	84.7 (12.3)	84.3 (12.8)	0.09
MVO	84.1 (11.0)	83.6 (12.3)	83.9 (11.6)	
LVO	81.2 (12.5)	82.9 (13.6)	82.0 (13.1)	
Heart rate (beats per minute)				
NVO	80.8 (14.4)	81.0 (15.0)	80.9 (14.6)	0.36
MVO	80.1 (18.4)	76.9 (14.2)	78.5 (16.5)	
LVO	81.3 (18.3)	80.0 (17.0)	80.7 (17.7)	
NIHSS score[*]				
NVO	5.5 (4-9)	5 (4-8)	5 (4-8)	**<0.001**
MVO	7.5 (5-12)	9 (6-12)	9 (6-12)	
LVO	12 (8-17)	14 (8-18)	13 (8-18)	
GCS score[†]				
NVO	15 (14-15)	15 (15-15)	15 (15-15)	**<0.001**
MVO	15 (14-15)	15 (12-15)	15 (13-15)	
LVO	14 (12.5-15)	15 (13-15)	14 (13-15)	
Medical history				
Previous stroke				
NVO	18 (13.0)	27 (19.9)	45 (16.4)	0.66
MVO	7 (10.3)	12 (18.5)	19 (14.3)	
LVO	20 (13.9)	20 (13.7)	40 (13.8)	
Hypertension				
NVO	83 (60.1)	77 (56.6)	160 (58.4)	0.16
MVO	45 (66.2)	45 (69.2)	90 (67.7)	
LVO	87 (60.4)	98 (67.1)	185 (63.8)	
Atrial fibrillation				
NVO	23 (16.7)[#]	11 (8.1)[#]	34 (12.4)	**<0.001**
MVO	19 (27.9)	12 (18.5)	31 (23.3)	
LVO	42 (29.2)	42 (28.8)	84 (29.0)	
Coronary artery disease				
NVO	22 (15.9)	17 (12.5)	39 (14.2)	0.98
MVO	11 (16.2)	9 (13.8)	20 (15.0)	
LVO	22 (15.3)	20 (13.7)	42 (14.5)	
Valvular or other heart disease				

NVO	13 (9.4)	11 (8.1)	24 (8.8)	0.64
MVO	6 (8.8)	7 (10.8)	13 (9.8)	
LVO	10 (6.9)	11 (7.5)	21 (7.2)	
Diabetes mellitus				
NVO	22 (15.9)	27 (19.9)	49 (17.9)	0.99
MVO	13 (19.1)	11 (16.9)	24 (18.0)	
LVO	23 (16.0)	28 (19.2)	51 (17.6)	
Hypercholesterolemia				
NVO	46 (33.3)	48 (35.3)	94 (34.3)	**<0.001**
MVO	18 (26.5)	15 (23.1)	33 (24.8)	
LVO	31 (21.5)	21 (14.4)	52 (17.9)	
Current smoker				
NVO	36 (26.1)	33/135 (24.4)	69/273 (25.3)	0.09
MVO	9 (13.2)	14 (21.5)	23 (17.3)	
LVO	41 (28.5)	37 (25.3)	78 (26.9)	
Pre-stroke function without symptoms[‡]				
NVO	31 (22.5)	34 (25.0)	65 (23.7)	0.10
MVO	17 (25.0)	14 (21.5)	31 (23.3)	
LVO	22 (15.3)	27 (18.5)	49 (16.9)	
Medication on admission				
Antihypertensive agent(s)				
NVO	67 (48.6)	64 (47.1)	131 (47.8)	0.46
MVO	39 (57.4)	33 (50.8)	72 (54.1)	
LVO	71 (49.3)	70 (47.9)	141 (48.6)	
Warfarin anticoagulation				
NVO	7 (5.1)[#]	1 (0.7)[#]	8 (2.9)	0.20
MVO	3 (4.4)	5 (7.7)	8 (6.0)	
LVO	6 (4.2)	2/145 (1.4)	8/289 (2.8)	
Aspirin/other antiplatelet agent				
NVO	49 (35.5)	34 (25.0)	83 (30.3)	0.52
MVO	22 (32.4)	13 (20.0)	35 (26.3)	
LVO	40 (27.8)	36/145 (24.8)	76/289 (26.3)	
Statin/other lipid lowering agent				
NVO	49 (35.5)	43 (31.6)	92 (33.6)	**0.001**
MVO	17 (25.0)	16 (24.6)	33 (24.8)	
LVO	34 (23.6)	23/145 (15.9)	57/289 (19.7)	
Time from onset to randomisation (hrs)				
NVO	2.4 (1.8-3.2)	2.7 (2.0-3.4)	2.6 (2.0-3.3)	**0.05**
MVO	2.3 (1.7-3.3)	2.3 (1.6-3.0)	2.3 (1.7-3.2)	
LVO	2.5 (1.7-3.2)	2.3 (1.7-3.1)	2.4 (1.7-3.1)	
Imaging features				
Infarct location confirmed				
NVO	87 (63.0)	74/133 (55.6)	161/271 (59.4)	**<0.001**
MVO	56/63 (88.9)	56/59 (94.9)	112/122 (91.8)	
LVO	117/134 (87.3)	120/131 (91.6)	237/265 (89.4)	
Infarct on left side				
NVO	46/87 (52.9)	36/74 (48.6)	82/161 (50.9)	**0.04**
MVO	32/56 (57.1)	27/56 (48.2)	59/112 (52.7)	
LVO	48/117 (41.0)	48/120 (40.0)	96/237 (40.5)	
Infarct on right side				
NVO	37/87 (42.5)	35/74 (47.3)	72/161 (44.7)	0.05
MVO	23/56 (41.1)	29/56 (51.8)	52/112 (46.4)	
LVO	64/117 (54.7)	69/120 (57.5)	133/237 (56.1)	
Infarct at midline or bilateral				
NVO	4/87 (4.6)	3/74 (4.1)	7/161 (4.3)	0.26
MVO	1/56 (1.8)	0/56 (0.0)	1/112 (0.9)	
LVO	5/117 (4.3)	3/120 (2.5)	8/237 (3.4)	

Infarct in anterior circulation				
NVO	59/87 (67.8)	58/74 (78.4)	117/161 (72.7)	**0.009**
MVO	44/56 (78.6)	39/56 (69.6)	83/112 (74.1)	
LVO	95/117 (81.2)	105/120 (87.5)	200/237 (84.4)	
Infarct in posterior circulation				
NVO	26/87 (29.9)	13/74 (17.6)	39/161 (24.2)	**<0.001**
MVO	11/56 (19.6)	11/56 (19.6)	22/112 (19.6)	
LVO	11/117 (9.4)	9/120 (7.5)	20/237 (8.4)	
Both anterior and posterior circulation				
NVO	2/87 (2.3)	3/74 (4.1)	5/161 (3.1)	0.22
MVO	1/56 (1.8)	6/56 (10.7)	7/112 (6.3)	
LVO	11/117 (9.4)	6/120 (5.0)	17/237 (7.2)	
With hyperdense vessel sign on CT				
NVO	9/125 (7.2)	9/115 (7.8)	18/240 (7.5)	**<0.001**
MVO	10/55 (18.2)	15/54 (27.8)	25/109 (22.9)	
LVO	44/123 (35.8)	51/117 (43.6)	95/240 (39.6)	
With old vascular lesions				
NVO	48 (34.8)	57/133 (42.9)	105/271 (38.7)	0.17
MVO	25/63 (39.7)	16/59 (27.1)	41/122 (33.6)	
LVO	57/134 (42.5)	58/131 (44.3)	115/265 (43.4)	
With brain atrophy				
NVO	98 (71.0)	93/133 (69.9)	191/271 (70.5)	0.67
MVO	44/63 (69.8)	39/59 (66.1)	83/122 (68.0)	
LVO	97/134 (72.4)	95/131 (72.5)	192/265 (72.5)	
With white matter changes				
NVO	54 (39.1)	55/133 (41.4)	109/271 (40.2)	0.55
MVO	20/63 (31.7)	22/59 (37.3)	42/122 (34.4)	
LVO	52/134 (38.8)	51/131 (38.9)	103/265 (38.9)	
Clot burden score				
NVO	-	-	-	**<0.001**
MVO	9 (9-9), n=27	9 (9-9), n=29	9 (9-9), n=56	
LVO	6 (4-7), n=54	6 (4-7), n=48	6 (4-7), n=102	
With visual residual flow (grade 1 or 2)				
NVO	-	-	-	**0.008**
MVO	26/37 (70.3)	27/36 (75.0)	53/73 (72.6)	
LVO	32/64 (50.0)	32/56 (57.1)	64/120 (53.3)	
Good collaterals (grade 3 to 5)				
NVO	-	-	-	**0.03**
MVO	27/36 (75.0)	25/37 (67.6)	52/73 (71.2)	
LVO	33/64 (51.6)	34/56 (60.7)	67/120 (55.8)	
Assigned to intensive BP lowering group				
NVO	9 (6.5)	6 (4.4)	15 (5.5)	0.76
MVO	4 (5.9)	5 (7.7)	9 (6.8)	
LVO	10 (6.9)	10 (6.8)	20 (6.9)	
Assigned to standard BP lowering group				
NVO	8 (5.8)	14 (10.3)	22 (8.0)	0.38
MVO	5 (7.4)	1 (1.5)	6 (4.5)	
LVO	10 (6.9)	8 (5.5)	18 (6.2)	

BP denotes blood pressure; CT, computed tomography; GCS, Glasgow Coma Scale; LVO, large vessel obstruction; MVO, medium vessel obstruction; mRS, modified Rankin scale; NIHSS, National Institutes of Health stroke scale; NVO, no vessel obstruction.

Data are n (%), mean (SD), or median (Q1, Q3). P values are based on Chi-square, analysis of variance, or Wilcoxon signed-rank test.

*Scores on the NIHSS range from 0 to 42, with higher scores indicating more severe neurological deficits.

†Scores on the GCS range from 15 (normal) to 3 (deep coma).

‡mRS=0. §P values for the comparisons among participants with NVO, MVO and LVO.

#P<0.05 by randomised alteplase dose.

Appendix 6.9. Baseline characteristics of participants by randomised BP lowering in different vascular obstruction status and site

	Low-dose NVO (n=73), MVO (n=30), LVO (n=64)	Standard-dose NVO (n=78), MVO (n=35), LVO (n=53)	Total NVO (n=151), MVO (n=65), LVO (n=117)	P value[§]
Age (years)				
NVO	68.4 (13.0)	68.4 (12.9)	68.4 (12.9)	0.16
MVO	70.1 (13.6)	73.4 (11.0)	71.9 (12.3)	
LVO	70.8 (13.8)	69.4 (9.9)	70.1 (12.1)	
Female				
NVO	26 (35.6)	31 (39.7)	57 (37.7)	0.87
MVO	12 (40.0)	15 (42.9)	27 (41.5)	
LVO	25 (39.1)	20 (37.7)	45 (38.5)	
Asian ethnicity				
NVO	21 (28.8)	22 (28.2)	43 (28.5)	**<0.001**
MVO	12 (40.0)	18 (51.4)	30 (46.2)	
LVO	33 (51.6)	27 (50.9)	60 (51.3)	
Clinical features				
Systolic BP (mmHg)				
NVO	168.5 (7.8)	168.1 (10.6)	168.3 (9.3)	0.24
MVO	166.7 (9.4)	167.9 (8.6)	167.3 (8.9)	
LVO	167.2 (8.7)	165.4 (8.9)	166.4 (8.8)	
Diastolic BP (mmHg)				
NVO	89.6 (11.7)	89.2 (13.0)	89.4 (12.3)	0.82
MVO	92.4 (12.5)	87.0 (12.1)	89.5 (12.5)	
LVO	90.8 (11.3)	89.7 (10.7)	90.3 (11.0)	
Heart rate (beats per minute)				
NVO	80.8 (13.7)	80.9 (16.1)	80.8 (15.0)	0.74
MVO	78.3 (15.2)	80.9 (17.0)	79.7 (16.1)	
LVO	81.4 (18.3)	82.1 (17.7)	81.7 (18.0)	
NIHSS score[*]				
NVO	6 (4-10)	6 (5-9)	6 (4-10)	**<0.001**
MVO	7.5 (4-14)	8 (5-12)	8 (5-12)	
LVO	12 (7-17)	10 (5-17)	11 (5-17)	
GCS score[†]				
NVO	15 (15-15)	15 (15-15)	15 (15-15)	**<0.001**
MVO	15 (13-15)	15 (12-15)	15 (13-15)	
LVO	14 (11-15)	15 (13-15)	14 (12-15)	
Medical history				
Previous stroke				
NVO	14 (19.2)	15 (19.2)	29 (19.2)	0.65
MVO	5 (16.7)	8 (22.9)	13 (20.0)	
LVO	9 (14.1)	9 (17.0)	18 (15.4)	
Hypertension				
NVO	46 (63.0)	51 (65.4)	97 (64.2)	0.33
MVO	23 (76.7)	25 (71.4)	48 (73.8)	
LVO	39 (60.9)[#]	43 (81.1)[#]	82 (70.1)	
Atrial fibrillation				
NVO	6 (8.2)	11 (14.1)	17 (11.3)	**0.001**
MVO	2 (6.7)	8 (22.9)	10 (15.4)	
LVO	17 (26.6)	16 (30.2)	33 (28.2)	
Coronary artery disease				
NVO	6 (8.2)	8 (10.3)	14 (9.3)	**0.05**
MVO	4 (13.3)	10 (28.6)	14 (21.5)	
LVO	10 (15.6)	8 (15.1)	18 (15.4)	
Valvular or other heart disease				

NVO	0 (0.0)	4 (5.1)	4 (2.6)	0.05
MVO	2 (6.7)	5 (14.3)	7 (10.8)	
LVO	1 (1.6)[#]	6 (11.3)[#]	7 (6.0)	
Diabetes mellitus				
NVO	14 (19.2)	17 (21.8)	31 (20.5)	0.46
MVO	8 (26.7)	9 (25.7)	17 (26.2)	
LVO	16 (25.0)	15 (28.3)	31 (26.5)	
Hypercholesterolemia				
NVO	19 (26.0)	19 (24.4)	38 (25.2)	0.13
MVO	8 (26.7)	12 (34.3)	20 (30.8)	
LVO	12 (18.8)	9 (17.0)	21 (17.9)	
Current smoker				
NVO	5 (6.8)	12 (15.4)	17 (11.3)	**0.01**
MVO	7 (23.3)	5 (14.3)	12 (18.5)	
LVO	14 (21.9)	15 (28.3)	29 (24.8)	
Pre-stroke function without symptoms[‡]				
NVO	13 (17.8)	15 (19.2)	28 (18.5)	0.06
MVO	10 (33.3)	11 (31.4)	21 (32.3)	
LVO	17 (26.6)	15 (28.3)	32 (27.4)	
Medication on admission				
Antihypertensive agent(s)				
NVO	39 (53.4)	43 (55.1)	82 (54.3)	0.69
MVO	20 (66.7)	19 (54.3)	39 (60.0)	
LVO	34 (53.1)	34 (64.2)	68 (58.1)	
Warfarin anticoagulation				
NVO	3 (4.1)	2 (2.6)	5 (3.3)	0.61
MVO	1 (3.3)	0 (0.0)	1 (1.5)	
LVO	1 (1.6)	1 (1.9)	2 (1.7)	
Aspirin/other antiplatelet agent				
NVO	27 (37.0)	26 (33.3)	53 (35.1)	0.19
MVO	9 (30.0)	11 (31.4)	20 (30.8)	
LVO	12 (18.8)	17 (32.1)	29 (24.8)	
Statin/other lipid lowering agent				
NVO	28 (38.4)	33 (42.3)	61 (40.4)	**0.006**
MVO	10 (33.3)	9 (25.7)	19 (29.2)	
LVO	15 (23.4)	11 (20.8)	26 (22.2)	
Time from onset to randomisation (hrs)				
NVO	3.2 (2.3-4.2)	3.2 (2.3-3.9)	3.2 (2.3-4.0)	0.51
MVO	3.6 (2.7-4.4)	3.6 (2.8-4.2)	3.6 (2.7-4.2)	
LVO	3.6 (2.6-4.2)	3.1 (2.6-3.7)	3.2 (2.6-4.1)	
Imaging features				
Infarct location confirmed				
NVO	42 (57.5)	47 (60.3)	89/151 (58.9)	**<0.001**
MVO	22/24 (91.7)	26/30 (86.7)	48/54 (88.9)	
LVO	53/57 (93.0)	38/45 (84.4)	91/102 (89.2)	
Infarct on left side				
NVO	14/42 (33.3)	25/47 (53.2)	39/89 (43.8)	0.51
MVO	10/22 (45.5)	16/26 (61.5)	26/48 (54.2)	
LVO	27/53 (50.9)	17/38 (44.7)	44/91 (48.4)	
Infarct on right side				
NVO	27/42 (64.3)[#]	20/47 (42.6)[#]	47/89 (52.8)	0.36
MVO	12/22 (54.5)	9/26 (34.6)	21/48 (43.8)	
LVO	24/53 (45.3)	15/38 (39.5)	39/91 (42.9)	
Infarct at midline or bilateral				
NVO	1/42 (2.4)	2/47 (4.3)	3/89 (3.4)	0.14
MVO	0/22 (0.0)	1/26 (3.8)	1/48 (2.1)	
LVO	2/53 (3.8)[#]	6/38 (15.8)[#]	8/91 (8.8)	

Infarct in anterior circulation				
NVO	31/42 (73.8)	33/47 (70.2)	64/89 (71.9)	0.23
MVO	18/22 (81.8)	19/26 (73.1)	37/48 (77.1)	
LVO	35/53 (66.0)	23/38 (60.5)	58/91 (63.7)	
Infarct in posterior circulation				
NVO	10/42 (23.8)	11/47 (23.4)	21/89 (23.6)	0.93
MVO	3/22 (13.6)	7/26 (26.9)	10/48 (20.8)	
LVO	10/53 (18.9)	11/38 (28.9)	21/91 (23.1)	
Both anterior and posterior circulation				
NVO	1/42 (2.4)	3/47 (6.4)	4/89 (4.5)	**0.02**
MVO	1/22 (4.5)	0/26 (0.0)	1/48 (2.1)	
LVO	8/53 (15.1)	4/38 (10.5)	12/91 (13.2)	
With hyperdense vessel sign on CT				
NVO	1/66 (1.5)	3/70 (4.3)	4/136 (2.9)	**<0.001**
MVO	4/21 (19.0)	7/30 (23.3)	11/51 (21.6)	
LVO	21/54 (38.9)	13/44 (29.5)	34/98 (34.7)	
With old vascular lesions				
NVO	36 (49.3)	41 (52.6)	77 (51.0)	0.74
MVO	7/24 (29.2)#	20/30 (66.7)#	27/54 (50.0)	
LVO	27/57 (47.4)	20/45 (44.4)	47/102 (46.1)	
With brain atrophy				
NVO	46 (63.0)	52 (66.7)	98 (64.9)	0.71
MVO	18/24 (75.0)	20/30 (66.7)	38/54 (70.4)	
LVO	43/57 (75.4)	27/45 (60.0)	70/102 (68.6)	
With white matter changes				
NVO	36 (49.3)	31 (39.7)	67 (44.4)	0.19
MVO	15/24 (62.5)	13/30 (43.3)	28/54 (51.9)	
LVO	36/57 (63.2)	21/45 (46.7)	57/102 (55.9)	
Clot burden score				
NVO	-	-	-	**<0.001**
MVO	9 (9-9), n=10	9 (9-9), n=8	9 (9-9), n=18	
LVO	5.5 (3-7), n=20	7 (6-7), n=13	6 (5-7), n=33	
With visual residual flow (grade 1 or 2)				
NVO	-	-	-	0.23
MVO	8/16 (50.0)	12/15 (80.0)	20/31 (64.5)	
LVO	11/28 (39.3)	15/23 (65.2)	26/51 (51.0)	
Good collaterals (grade 3 to 5)				
NVO	-	-	-	0.72
MVO	8/16 (50.0)	10/15 (66.7)	18/31 (58.1)	
LVO	11/27 (40.7)	16/23 (69.6)	27/50 (54.0)	
Assigned to low-dose alteplase group				
NVO	9 (12.3)	8 (10.3)	17 (11.3)	0.39
MVO	4 (13.3)	5 (14.3)	9 (13.8)	
LVO	10 (15.6)	10 (18.9)	20 (17.1)	
Assigned to standard-dose alteplase group				
NVO	6 (8.2)	14 (17.9)	20 (13.2)	0.50
MVO	5 (16.7)	1 (2.9)	6 (9.2)	
LVO	10 (15.6)	8 (15.1)	18 (15.4)	

BP denotes blood pressure; CT, computed tomography; GCS, Glasgow Coma Scale; LVO, large vessel obstruction; MVO, medium vessel obstruction; mRS, modified Rankin scale; NIHSS, National Institutes of Health stroke scale; NVO, no vessel obstruction.

Data are n (%), mean (SD), or median (Q1, Q3). P values are based on Chi-square, analysis of variance, or Wilcoxon signed-rank test.

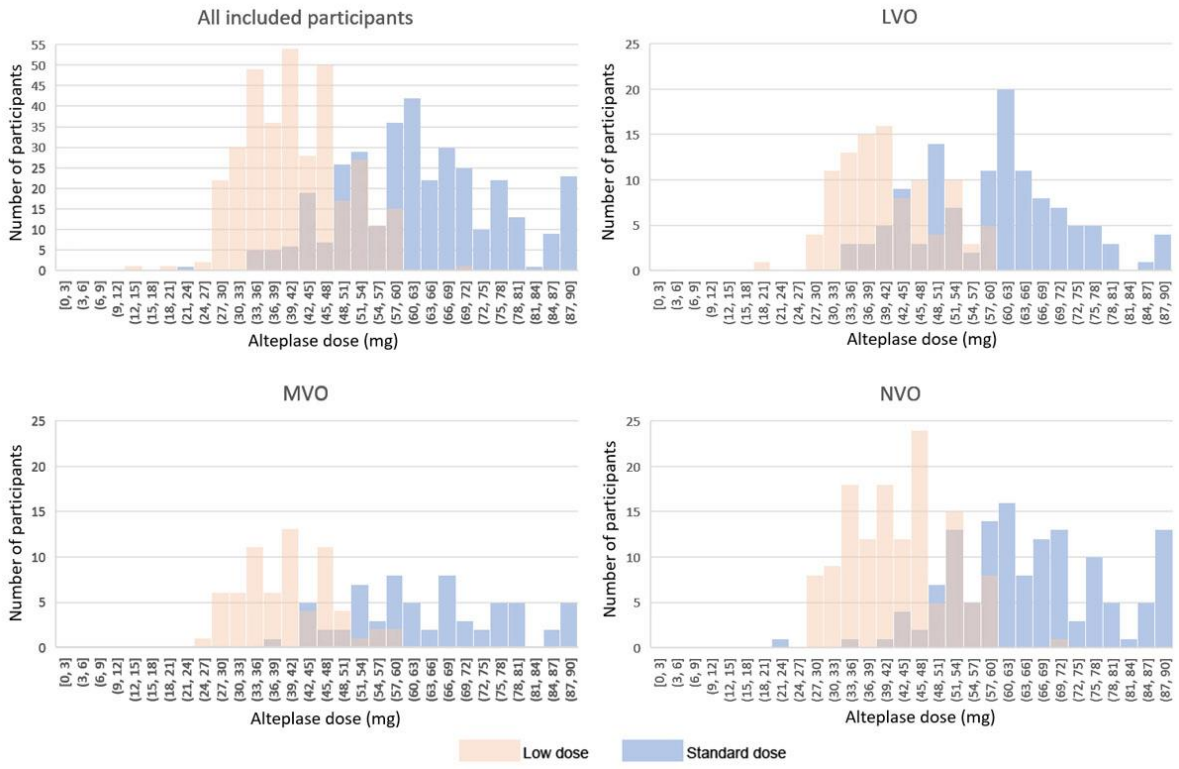
*Scores on the NIHSS range from 0 to 42, with higher scores indicating more severe neurological deficits.

†Scores on the GCS range from 15 (normal) to 3 (deep coma).

‡mRS=0. §P values for the comparisons among participants with NVO, MVO and LVO.

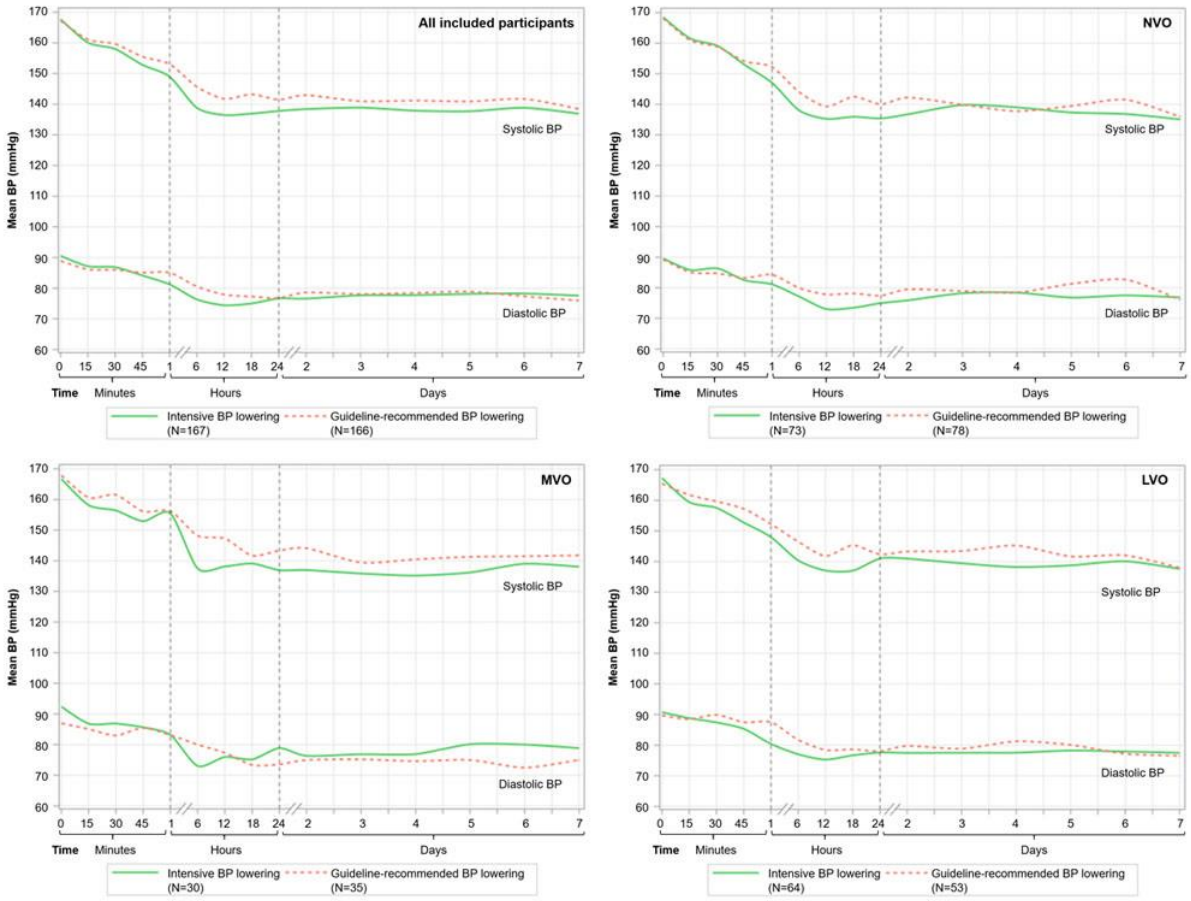
#P<0.05 by randomised BP lowering.

Appendix 6.10. Distribution of alteplase dose based on measured body weight by randomised group in participants with and without vascular obstruction, and different sites of obstruction



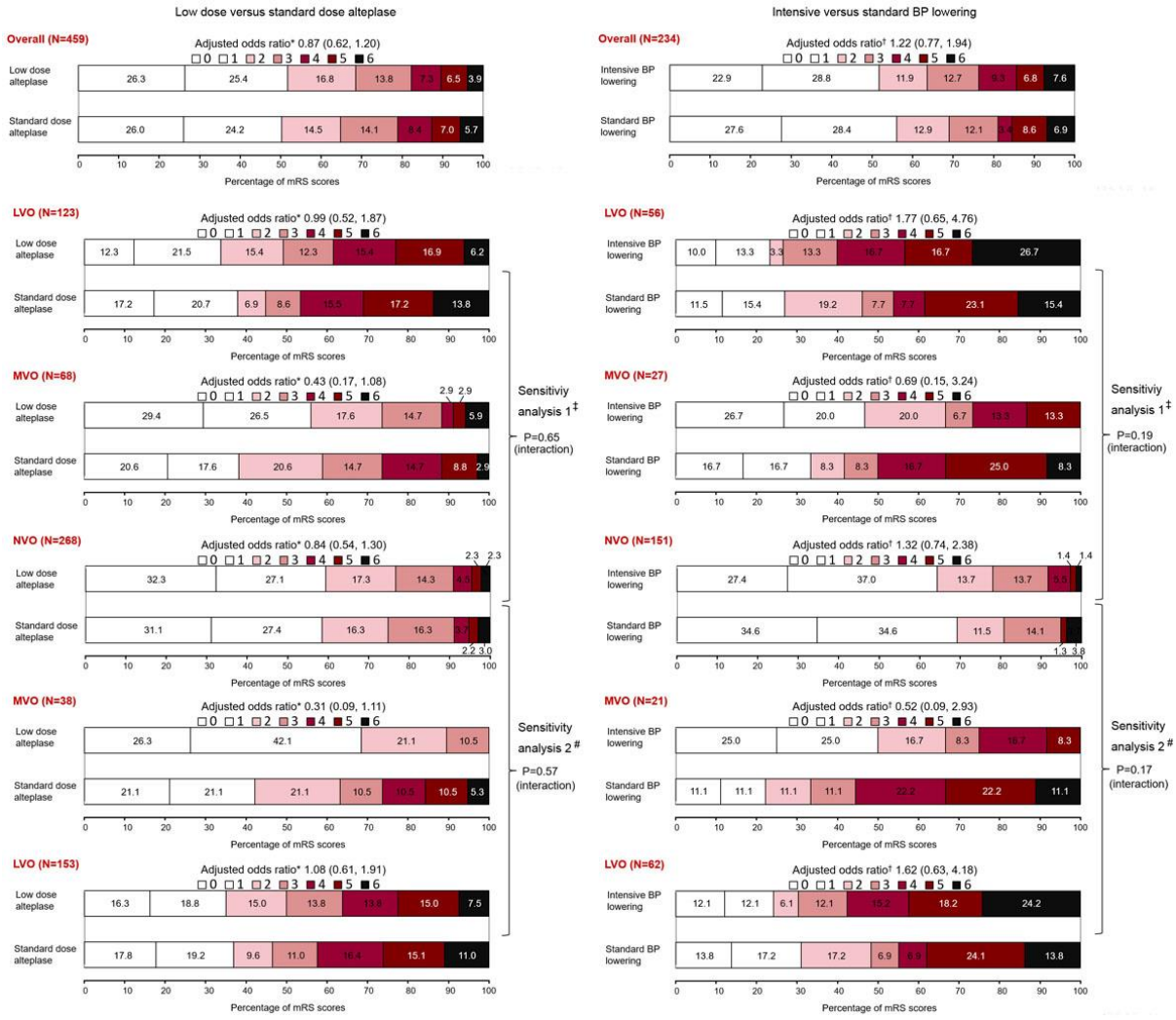
LVO, large vascular obstruction; MVO, medium vascular obstruction; NVO, no vascular obstruction.

Appendix 6.11. Mean blood pressure (mmHg) from randomisation to day 7 by randomised group of BP lowering in participants with and without vascular obstruction, and different sites of obstruction



BP denotes blood pressure; LVO, large vascular obstruction; MVO, medium vascular obstruction; NVO, no vascular obstruction.

Appendix 6.12. Sensitivity analyses for association of randomised alteplase dose or BP lowering with ordinal mRS score across participants stratified by vascular obstruction status and site



ACA denotes anterior cerebral artery; BP, blood pressure; LVO, large vascular obstruction; mRS, modified Rankin scale; MCA, middle cerebral artery; MVO, medium vascular obstruction; NVO, no vascular obstruction; PCA, posterior cerebral artery.

*Adjusted for minimized key prognostic covariates (age, sex, Asian ethnicity, baseline NIHSS score, time from stroke onset to randomisation) and assigned to intensive BP lowering group.

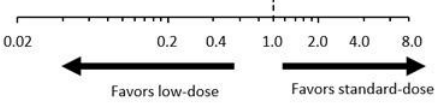
†Adjusted for minimized key prognostic covariates (age, sex, Asian ethnicity, baseline NIHSS score, time from stroke onset to randomisation) and assigned to low-dose alteplase group.

‡Involved participants with adjudicated angiography data only. Those with obstruction at M2/M3 segment of MCA, A2/A3 segment of ACA, P2/P3 segment of PCA were classified as MVO. Those with obstruction at M1 segment of MCA, A1 segment of MCA, P1 segment of PCA, vertebral artery, basilar artery, or internal carotid artery were classified as LVO.

#Involved participants with adjudicated angiography data only. Those with obstruction at distal M2/M3 segment of MCA, A2/A3 segment of ACA, P2/P3 segment of PCA were classified as MVO. Those with obstruction at M1/proximal M2 segment of MCA, A1 segment of MCA, P1 segment of PCA, vertebral artery, basilar artery, or internal carotid artery were classified as LV3.

Appendix 6.13. Sensitivity analyses 1* for other thrombolysis outcomes by randomised alteplase dose across participants stratified by vascular obstruction status and site

Thrombolysis outcomes	Alteplase dose group, n/N (%) Low	Standard	Odds ratio (95% CI)		P inter-action
mRS 2-6: overall	112/232 (48.3)	113/227 (49.8)		0.94 (0.63, 1.42)†	
NVO	54/133 (40.6)	56/135 (41.5)		0.94 (0.55, 1.60)†	0.29†
MVO	15/34 (44.1)	21/34 (61.8)		0.47 (0.15, 1.44)†	
LVO	43/65 (66.2)	36/58 (62.1)		1.62 (0.67, 3.91)†	
mRS 3-6: overall	73/232 (31.5)	80/227 (35.2)		0.82 (0.53, 1.27)†	
NVO	31/133 (23.3)	34/135 (25.2)		0.81 (0.45, 1.48)†	0.83†
MVO	9/34 (26.5)	14/34 (41.2)		0.44 (0.11, 1.72)†	
LVO	33/65 (50.8)	32/58 (55.2)		0.93 (0.40, 2.16)†	
mRS 6: overall	9/240 (3.8)	13/230 (5.7)		0.65 (0.27, 1.55)	
NVO	3/138 (2.2)	4/136 (2.9)		0.73 (0.16, 3.34)	0.47
MVO	2/34 (5.9)	1/35 (2.9)		2.12 (0.19, 24.59)	
LVO	4/68 (5.9)	8/59 (13.6)		0.40 (0.11, 1.40)	
Intracranial hemorrhage‡: overall	41/240 (17.1)	40/230 (17.4)		0.98 (0.61, 1.58)	
NVO	14/138 (10.1)	16/136 (11.8)		0.85 (0.40, 1.81)	0.72
MVO	7/34 (20.6)	9/35 (25.7)		0.75 (0.24, 2.31)	
LVO	20/68 (29.4)	15/59 (25.4)		1.22 (0.56, 2.68)	
sICH (ECASS2): overall	7/240 (2.9)	10/230 (4.3)		0.66 (0.25, 1.77)	
NVO	1/138 (0.7)	3/136 (2.2)		0.32 (0.03, 3.15)	—
MVO	0/34 (0.0)	2/35 (5.7)		—	
LVO	6/68 (8.8)	5/59 (8.5)		1.05 (0.30, 3.62)	
END or death (24 hrs): overall	16/240 (6.7)	19/230 (8.3)		0.79 (0.40, 1.58)	
NVO	6/138 (4.3)	6/136 (4.4)		0.98 (0.31, 3.13)	0.70
MVO	1/34 (2.9)	3/35 (8.6)		0.32 (0.03, 3.27)	
LVO	9/68 (13.2)	10/59 (16.9)		0.75 (0.28, 1.99)	
No recanalization (48 hrs): overall	11/28 (39.3)	21/33 (63.6)		0.37 (0.13, 1.04)	
NVO	—	—		—	0.36
MVO	1/9 (11.1)	6/12 (50.0)		0.13 (0.01, 1.33)	
LVO	10/19 (52.6)	15/21 (71.4)		0.44 (0.12, 1.64)	



BP denotes blood pressure; END, early neurologic deterioration; ICH, intracerebral haemorrhage; LVO, large vessel obstruction; mRS, modified Rankin scale, MVO: medium vessel obstruction; NINDS, the National Institutes of Neurological Diseases and Stroke; NVO, no vascular obstruction; sICH, symptomatic intracerebral haemorrhage.

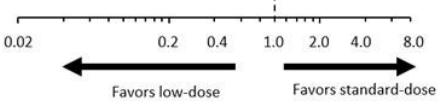
*Involved participants with adjudicated angiography data only. Those with obstruction at M2/M3 segment of MCA, A2/A3 segment of ACA, P2/P3 segment of PCA were classified as MVO. Those with obstruction at M1 segment of MCA, A1 segment of MCA, P1 segment of PCA, vertebral artery, basilar artery, or internal carotid artery were classified as LVO.

†Adjusted for minimized key prognostic covariates (age, sex, Asian ethnicity, baseline NIHSS score, time from stroke onset to randomisation) and assigned to intensive BP lowering group.

‡Adjudicated centrally or reported by site investigators.

Appendix 6.14. Sensitivity analyses 2* for other thrombolysis outcomes by randomised alteplase dose across participants stratified by vascular obstruction status and site

Thrombolysis outcomes	Alteplase dose group, n/N (%) Low	Standard	Odds ratio (95% CI)	P inter-action
mRS 2-6: overall	112/232 (48.3)	113/227 (49.8)	0.94 (0.65, 1.36)	
NVO	54/133 (40.6)	56/135 (41.5)	0.96 (0.59, 1.57)	0.29
MVO	6/19 (31.6)	11/19 (57.9)	0.34 (0.09, 1.27)	
LVO	52/80 (65.0)	46/73 (63.0)	1.09 (0.56, 2.11)	
mRS 3-6: overall	73/232 (31.5)	80/227 (35.2)	0.84 (0.57, 1.24)	
NVO	31/133 (23.3)	34/135 (25.2)	0.90 (0.52, 1.58)	0.27
MVO	2/19 (10.5)	7/19 (36.8)	0.20 (0.04, 1.15)	
LVO	40/80 (50.0)	39/73 (53.4)	0.87 (0.46, 1.65)	
mRS 6: overall	9/240 (3.8)	13/230 (5.7)	0.65 (0.27, 1.55)	
NVO	3/138 (2.2)	4/136 (2.9)	0.73 (0.16, 3.34)	—
MVO	0/19 (0.0)	1/19 (5.3)	—	
LVO	6/83 (7.2)	8/75 (10.7)	0.65 (0.22, 1.98)	
Intracranial hemorrhage†: overall	41/240 (17.1)	40/230 (17.4)	0.98 (0.61, 1.58)	
NVO	14/138 (10.1)	16/136 (11.8)	0.85 (0.40, 1.81)	0.92
MVO	4/19 (21.1)	4/19 (21.1)	1.00 (0.21, 4.76)	
LVO	23/83 (27.7)	20/75 (26.7)	1.05 (0.52, 2.13)	
sICH (ECASS2): overall	7/240 (2.9)	10/230 (4.3)	0.66 (0.25, 1.77)	
NVO	1/138 (0.7)	3/136 (2.2)	0.32 (0.03, 3.15)	—
MVO	0/19 (0.0)	2/19 (10.5)	—	
LVO	6/83 (7.2)	5/75 (6.7)	1.09 (0.32, 3.73)	
END or death (24 hrs): overall	16/240 (6.7)	19/230 (8.3)	0.79 (0.40, 1.58)	
NVO	6/138 (4.3)	6/136 (4.4)	0.98 (0.31, 3.13)	0.67
MVO	1/19 (5.3)	3/19 (15.8)	0.30 (0.03, 3.14)	
LVO	9/83 (10.8)	10/75 (13.3)	0.79 (0.30, 2.07)	
No recanalization (48 hrs): overall	11/28 (39.3)	21/33 (63.6)	0.37 (0.13, 1.04)	
NVO	—	—	—	—
MVO	0/3 (0.0)	1/5 (20.0)	—	
LVO	11/25 (44.0)	20/28 (71.4)	0.31 (0.10, 0.98)	



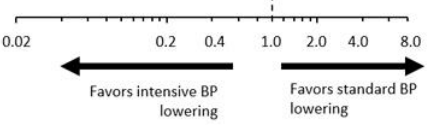
BP denotes blood pressure; END, early neurologic deterioration; ICH, intracerebral haemorrhage; LVO, large vessel obstruction; mRS, modified Rankin scale, MVO: medium vessel obstruction; NINDS, the National Institutes of Neurological Diseases and Stroke; NVO, no vascular obstruction; sICH, symptomatic intracerebral haemorrhage.

*Involved participants with adjudicated angiography data only. Those with obstruction at distal M2/M3 segment of MCA, A2/A3 segment of ACA, P2/P3 segment of PCA were classified as MVO. Those with obstruction at M1/proximal M2 segment of MCA, A1 segment of MCA, P1 segment of PCA, vertebral artery, basilar artery, or internal carotid artery were classified as LVO.

†Adjudicated centrally or reported by site investigators.

Appendix 6.15. Sensitivity analyses 1* for other thrombolysis outcomes by randomised BP lowering across participants stratified by vascular obstruction status and site

Thrombolysis outcomes	BP lowering group, n/N (%)		Odds ratio (95% CI)	P inter-action
	Intensive	Standard		
mRS 2-6: overall	57/118 (48.3)	51/116 (44.0)	1.19 (0.71, 1.99)	
NVO	26/73 (35.6)	24/78 (30.8)	1.24 (0.63, 2.45)	0.67
MVO	8/15 (53.3)	8/12 (66.7)	0.57 (0.12, 2.75)	
LVO	23/30 (76.7)	19/26 (73.1)	1.21 (0.36, 4.06)	
mRS 3-6: overall	43/118 (36.4)	36/116 (31.0)	1.27 (0.74, 2.19)	
NVO	16/73 (21.9)	15/78 (19.2)	1.18 (0.53, 2.60)	0.16
MVO	5/15 (33.3)	7/12 (58.3)	0.36 (0.07, 1.72)	
LVO	22/30 (73.3)	14/26 (53.8)	2.36 (0.77, 7.21)	
mRS 6: overall	9/118 (7.6)	8/116 (6.9)	1.11 (0.41, 3.00)	
NVO	1/73 (1.4)	3/78 (3.8)	0.35 (0.04, 3.42)	—
MVO	0/15 (0.0)	1/12 (8.3)	—	
LVO	8/30 (26.7)	4/26 (15.4)	2.00 (0.52, 7.62)	
Intracranial hemorrhage†: overall	12/118 (10.2)	15/116 (12.9)	0.76 (0.34, 1.71)	
NVO	2/73 (2.7)	8/78 (10.3)	0.25 (0.05, 1.20)	0.25
MVO	3/15 (20.0)	2/12 (16.7)	1.25 (0.17, 9.02)	
LVO	7/30 (23.3)	5/26 (19.2)	1.28 (0.35, 4.65)	
sICH (ECASS2): overall	5/118 (4.2)	4/116 (3.4)	1.24 (0.32, 4.73)	
NVO	0/73 (0.0)	3/78 (3.8)	—	—
MVO	0/15 (0.0)	0/12 (0.0)	—	
LVO	5/30 (16.7)	1/26 (3.8)	5.00 (0.54, 45.92)	
END or death (24 hrs): overall	11/118 (9.3)	10/116 (8.6)	1.09 (0.44, 2.67)	
NVO	3/73 (4.1)	5/78 (6.4)	0.63 (0.14, 2.72)	—
MVO	0/15 (0.0)	0/12 (0.0)	—	
LVO	8/30 (26.7)	5/26 (19.2)	1.53 (0.43, 5.42)	
No recanalization (48 hrs): overall	8/12 (66.7)	7/11 (63.6)	1.14 (0.21, 6.37)	
NVO	—	—	—	—
MVO	3/5 (60.0)	1/4 (25.0)	4.50 (0.25, 80.57)	
LVO	5/7 (71.4)	6/7 (85.7)	0.42 (0.03, 6.06)	



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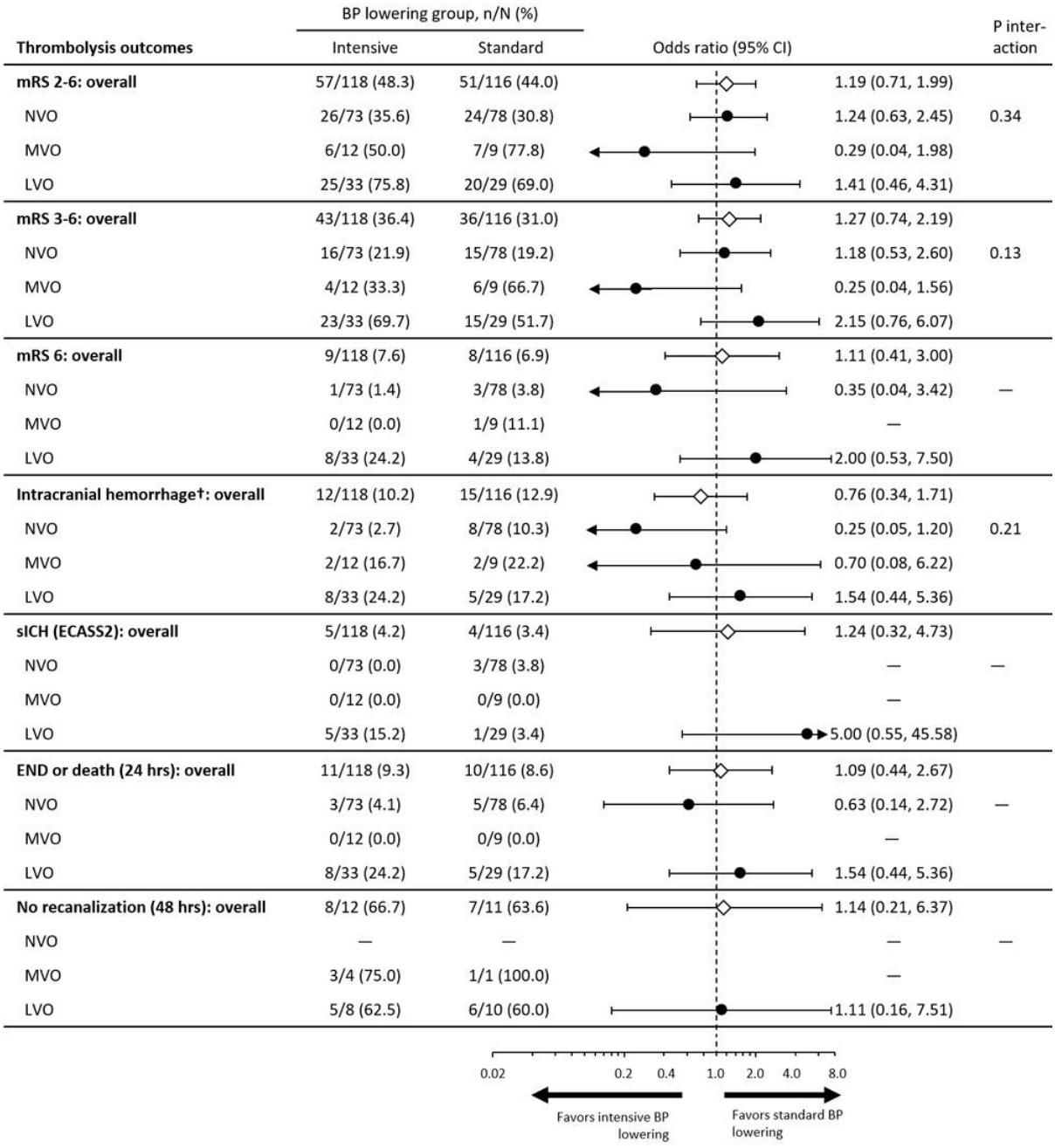
*Involved participants with adjudicated angiography data only. Those with obstruction at M2/M3 segment of MCA, A2/A3 segment of ACA, P2/P3 segment of PCA were classified as MVO. Those with obstruction at M1 segment of MCA, A1 segment of MCA, P1 segment of PCA, vertebral artery, basilar artery, or internal carotid artery were classified as LVO.

†Adjudicated centrally or reported by site investigators.

Appendix 6.16. Sensitivity analyses 2* for other thrombolysis outcomes by randomised BP lowering across participants stratified by vascular obstruction status and site



BP denotes blood pressure; END, early neurologic deterioration; ICH, intracerebral haemorrhage; LVO, large vessel obstruction; mRS, modified Rankin scale, MVO: medium vessel obstruction; NINDS, the National Institutes of Neurological Diseases and Stroke; NVO, no vascular obstruction; sICH, symptomatic intracerebral haemorrhage.

*Involved participants with adjudicated angiography data only. Those with obstruction at distal M2/M3 segment of MCA, A2/A3 segment of ACA, P2/P3 segment of PCA were classified as MVO. Those with obstruction at M1/proximal M2 segment of MCA, A1 segment of MCA, P1 segment of PCA, vertebral artery, basilar artery, or internal carotid artery were classified as LVO.

†Adjudicated centrally or reported by site investigators.

Chapter 7: Concluding Remarks

In this thesis, I explored the associations of brain (micro-) circulation imaging changes and thrombolysis outcomes in AIS patients, and tried to identify potential imaging markers that might modify the treatment effects of intravenous alteplase dose and BP-lowering treatments. I was able to do this by using the unique and comprehensive dataset from the alteplase-dose and BP-lowering arms of the ENCHANTED trial. This thesis explored and elucidated the significance of FLAIR-HAs, lacunar versus non-lacunar infarction, and vascular obstruction with different status/sites, in predicting functional recovery and sICH in thrombolysis-treated AIS patients. In addition, heterogeneity in associations of randomised treatment with outcomes across subgroups stratified by some imaging factors was estimated. In doing so, I have contributed new knowledge and highlighted the potential importance of imaging biomarkers in developing and implementing novel strategies to manage AIS.

7.1 Summary of results

Chapters 2 showed that prior AF, LVO, larger infarct volume, and anterior circulation infarction, were independently associated with FLAIR-HAs in 293 ENCHANTED participants from the alteplase-dose arm who had brain MRI with FLAIR and DWI sequences scanned <4.5 hours after stroke onset. Patients with FLAIR-HAs versus those without had a lower rate of good 90-day functional outcomes, as defined by mRS scores 0 to 1 (45.4% versus 57.3%), scores 0 to 2 (57.2% versus 75.6%), or ordinal shift. However, after adjustment of covariables, FLAIR-HAs were significantly associated with an increased favourable outcome. The association of FLAIR-HAs with functional outcome appear to vary numerically by the presence of LVO: more favourable outcomes in those without LVO (N=145, 69.4% versus 56.6%) compared with LVO (N=115, 39.8% versus 56.3%). There was a significant increase in haemorrhagic infarct risk regardless of covariables adjustment.

Chapter 3 extended the work outlined in Chapter 2 through the presentation of a systematic review and meta-analysis. I included 36 eligible cohort studies (33 prospectively collected, including data from the ENCHANTED trial) involving 3577 patients after literature screening from Medline, Embase, and the Cochrane Central Register of Controlled Trials, searched from inception to the end of October 2019. FLAIR-HAs were not associated with the functional outcome overall, but were significantly associated with better outcomes in those receiving

endovascular therapy. Contrary to FLAIR-HAs at proximal MCA or within DWI lesions, FLAIR-HAs beyond DWI lesions were associated with better outcomes. FLAIR-HAs favoured recanalisation with an increased risk of ICH and early neurological deterioration.

Chapter 4 summarised the findings of low-dose versus standard-dose intravenous alteplase in lacunar AIS. Of 2588 ENCHANTED participants with available imaging and clinical data from the alteplase-dose arm, I classified cases as definite/probable lacunar (n=490) or non-lacunar AIS (n=2098) for the primary analysis. Regardless of the alteplase dose received, lacunar AIS participants had favourable functional and other clinical or safety outcomes compared to participants with non-lacunar AIS. Low-dose alteplase (versus standard) had no differential effect on functional outcomes but reduced the risk of sICH in all included participants. There were no differential treatment effects of low- versus standard-dose alteplase on all outcomes across lacunar and non-lacunar AIS. Comparable findings were seen in the sensitivity analyses for definite lacunar and non-lacunar AIS and definite/probable/possible lacunar and non-lacunar AIS.

In Chapter 5, counterpart analyses of lacunar AIS were performed based on data from the ENCHANTED BP arm. According to pre-specified definitions, I classified 1632 participants as having definite/probable lacunar (n=454 [27.8%]) or non-lacunar AIS based on clinical and adjudicated imaging findings. Most baseline characteristics, acute BP, and aspects of management differed between lacunar and non-lacunar AIS, but mean systolic BP differences by treatment were comparable at each time point and over 24 hours post-randomisation. The neutral effect of intensive BP lowering on functional outcome and the beneficial effect on intracranial haemorrhage were similar for the two subgroups. The results were consistent in the sensitivity analysis for definite lacunar and non-lacunar AIS.

Finally, in Chapter 6, I analysed those with baseline CTA or MRA of moderate to good quality or proximal arterial occlusion according to reports by site investigators (N=940, 607 in alteplase arm only, 243 in BP arm only, and 90 assigned to both arms). The degree of vascular occlusion was grouped according to being no (NVO) (n=388), medium (MVO) (n=183), or large (LVO) (n=369). Compared to the NVO group, functional outcome was worse in LVO but comparable in MVO groups. There were no differences in associations of alteplase dose or BP lowering and outcomes across NVO/MVO/LVO groups, except for a borderline significant difference for intensive BP lowering and increased END. The results were inconsistent in sensitivity analysis,

using different definitions of MVO, but where statistical power was reduced after excluding those with site-reported angiography data.

7.2 Strengths, limitations, and future studies

The key strengths of the studies outlined in this thesis include the use of data derived from AIS patients who participated in a large-scale, international, multi-centre clinical trial with prospective, systematic, complete, and high-quality data collection. The imaging assessment was completed blind to clinical features and other data, using rigorously pre-specified approaches developed in prior studies. Trained site investigators assessed clinical outcomes, and sICH was adjudicated centrally by an adjudication panel with experienced stroke physicians during the trial. The statistical techniques used in this thesis are reliable in that they complied with traditional and widely accepted analytical methods. All these provide reassurance over precision and reliability around the estimates of associations between imaging changes and clinical outcomes.

However, there are several limitations to these analyses that should be acknowledged. The agreement of acute ischaemic signs assessed by the less expert imaging panel was only fair to moderate between raters, which may have underestimated the prognostic value of some imaging features. In addition, the heterogeneous parameters across scanners from different hospitals may have introduced bias that on the one hand reduced predictive power but on the other added real-world relevance.

As most ENCHANTED participants did not have brain MRI in a pragmatic study, the sample size for analyses was hindered in Chapter 2. Furthermore, I did not strictly define baseline brain MRI as images obtained before thrombolysis; rather I included patients with brain MRI performed shortly after the commencement of thrombolysis but within the 4.5-hour time window from stroke onset to enrich the sample with most eligible participants. However, a prior study[1] and my analyses did not show that the detection of FLAIR-HAs is dependent on the time interval between stroke onset and MRI scanning in AIS. Additional analyses of thrombolysis outcomes limited to 164 patients who had brain MRI prior to receipt of intravenous alteplase showed comparable results (data not shown). Nonetheless, our data provide a rationale for further investigating associations of FLAIR-HAs with LVO, grade of collateral circulation, perfusion status, and outcomes after intravenous thrombolysis or endovascular therapy in AIS

patients. This could be undertaken through overviews including additional data from prior studies, or through dedicated observational studies with large samples.

A key limitation of the study in Chapter 3 is the low number of studies and participants to allow a reliable examination of the associations in subgroups and for some intermediate outcomes, which raises the potential for chance findings. The study-level meta-analysis also hindered a firm estimate to be made of the relation with NIHSS score change or infarct growth, which were undertaken in the ENCHANTED trial and varied according to types of FLAIR-HAs defined by location or extent. Thus, a future collaboration based on individual participant data meta-analysis is required. Moreover, for the association of FLAIR-HAs with thrombolysis outcomes in posterior circulation AIS, there is a need for consensus regarding the most appropriate method to assess FLAIR-HAs. In addition, whether FLAIR-HAs may serve as an additional imaging marker to select these and other types of stroke patients with DWI-FLAIR mismatch for endovascular therapy warrants future investigation.

In Chapters 4 and 5, misidentification of true lacunar AIS cannot be avoided by comparing baseline and follow-up images, since imaging appearances of acute ischaemic lesions after intravenous thrombolysis might have altered.[2] Future research with the identification of lacunar AIS on baseline MRI is required to confirm or refute these findings. Most of the ENCHANTED participants were from Asia with mild-to-moderate severity of stroke compromises the generalisability of the results to patients from non-Asian countries or with severe AIS. The actual SBP difference over 24 hours after baseline (Δ5.5 mmHg) by randomised treatment was smaller than envisaged, raising the question as to whether the treatment effect might be different with greater BP lowering. Future investigations out of this topic are awaited.

The vascular analyses in Chapter 6 might give light to future research in exploring whether AIS patients with MVO would benefit from low-dose alteplase. Moreover, whether caution should be advised for intensive BP lowering in thrombolysis-treated LVO patients, although the results were compromised by several factors: low statistical power; use of site-reported angiography data; an inability to identify and separate extracranial internal carotid artery occlusion from LVO as few participants had cervical CTA or MRA; and not identification of ultra-early reperfusion status (within 6 hours post-thrombolysis). Due to the relatively small sample size and insufficient statistical power, efficacy and safety of low-dose intravenous alteplase and intensive BP lowering in individual subgroups and potential heterogeneity across subgroups in Chapters 4 to 6 should be regarded as observational and hypothesis-generating only. However,

corresponding data can be used for sample size calculation if dedicated trials are designed in the future.

Finally, I did not explore other imaging features which are associated with thrombolysis outcomes in AIS and might modify the effects of randomised alteplase dose or BP lowering. These include infarct location, clot length, residual flow grade, collateral circulation status, perfusion parameters, cortical superficial siderosis, and co-existence of microbleeds. All these factors await further investigation.

7.3 Implications for future clinical practice

In translating the results of my thesis to future clinical practice, it is crucial to recognise the characteristics of participants in the ENCHANTED trial. Most of the AIS patients enrolled in the trial were Asians with mild to moderate neurologic dysfunction. Those who were unlikely to benefit from intravenous thrombolysis, due to pre-existing disability or a very high likelihood of death within 24 hours, were excluded. Accordingly, my results do not apply to severe AIS patients, and caution is warranted to generalising them to non-Asian patients.

NCCT is a predominant imaging examination for a suspected AIS patient in current clinical practice, where there is the advantage of quickly excluding primary ICH, but the limitation is low sensitivity in detecting early ischaemic changes. Prior analyses from the ENCHANTED trial indicate that acute ischaemic changes and brain frailty, interpreted by non-expert readers on NCCT, are associated with sICH, mortality, and functional recovery in the ENCHANTED participants. Although these associations were set, especially for an interesting finding that atrophy was associated with a reduced risk of 7-day death, it is far too soon to apply these results in making treatment decisions and defining the individual outcome. Similar conclusions apply to the prognostic significance of imaging features of the brain (micro) circulation, as outlined in this thesis. This is because the threshold value is lacking, inter-reader agreement on imaging assessments was only fair to moderate, and the absolute risk differences were small for some outcomes. In the future, considering both imaging and clinical factors may be valuable in forming management recommendations for particular individual AIS patients.

During recent decades, MRI technology has advanced with improvements in magnetic field intensity enhancement, sequence design optimisation, radiofrequency energy absorption reduction, and mitigating peripheral nerve stimulation. These all offer the potential for wide clinical application in detecting AIS very early after stroke onset. A routine brain MRI scan

which includes the FLAIR sequence, takes no more than five minutes. Besides no signal change on the FLAIR sequence indicating eligibility of intravenous thrombolysis in wake-up stroke patients, FLAIR-HAs may reflect LVO with good collaterals. FLAIR-HAs are common in AIS. They can be easily detected after specific training. My findings on the associations of FLAIR-HAs with thrombolysis outcomes provide additional support to prompt clinicians to actively seek the presence of FLAIR-HAs, where MRI is used in the management of AIS patients, and considerations are given towards reperfusion therapy to salvage the ischaemic penumbra. The results of a subsequent meta-analysis in Chapter 3 suggest that AIS patients with FLAIR-HAs may benefit from endovascular therapy. Therefore, it is worthwhile further investigating the value of FLAIR-HAs in selecting candidates for endovascular therapy, given their faster acquirement with no need for radiocontrast agent compared to the current method based on CTA and CTP.

The ENCHANTED trial suggested that low-dose alteplase significantly reduced the risk of sICH, and that intensive BP lowering significantly reduced the risk of any intracranial haemorrhage in AIS patients. However, guideline recommendations to receive these treatments may not be supported, since neither the non-inferiority of low-dose versus standard-dose alteplase or the superiority of intensive to guideline-recommended BP lowering following intravenous thrombolysis, were shown with respect to the prespecified 90-day primary functional outcome, although the non-inferiority for a prespecified secondary outcome (mRS scores shift) was achieved in the alteplase arm. It is worth considering who may benefit from low-dose alteplase or early intensive BP lowering through comprehensive post-hoc subgroup analyses of the main trial. For instance, low-dose alteplase improves thrombolysis outcomes in AIS patients on prior antiplatelet therapy, indicating low-dose alteplase may be an option for the AIS patients with a high risk of ICH, such as those with HAS-BLED scores (hypertension, abnormal renal or liver function, prior stroke, prior bleeding, labile international normalised ratio, >65 years, drugs or alcohol) ≥3 or with high cerebral microbleeds burden on MRI.

My findings in Chapters 4 and 5 provide evidence that for patients with lacunar AIS, low-dose alteplase and early intensive BP lowering have no additional benefit or safety over standard-dose alteplase and guideline-recommended BP control. This is helpful to dispel concerns over the risk of thrombolysis-related ICH offsetting the benefits of standard-dose intravenous alteplase for lacunar AIS, where the prognosis is usually better than other AIS subtypes. The assumed formation mechanisms of lacunar AIS, occlusion of a single penetrating artery with

no or a tiny clot, causes debate that conservative treatments rather than standard-dose alteplase may be more applicable for lacunar AIS. However, discrimination of true lacunar AIS from non-lacunar infarct at a very early stage is not so straightforward, even with MRI, given that infarct lesions can hardly be detected in nearly one-third of AIS patients, as shown in the ENCHANTED and IST-3 trials. In addition, misdiagnosis cannot be avoided if lacunar AIS is only adjudicated by clinical manifestations (lacunar syndrome). The current AHA/ASA guideline recommends intravenous alteplase to treat lacunar AIS due to the comparable rates of good functional recovery following intravenous alteplase across lacunar and non-lacunar AIS patients. My results further support the use of standard-dose alteplase and do not support a shift towards intensive BP lowering in lacunar AIS.

My study in Chapter 6 shows no heterogeneity in the treatment effects on functional outcomes across the ENCHANTED subgroups by vascular occlusion status/site. However, the point estimate favours low-dose alteplase with respect to the primary outcome, and favours standard BP lowering with respect to the END or death. Thus, low-dose alteplase for MVO and intensive BP lowering for LVO should not be recommended before more supporting evidence appears. However, close monitoring of BP levels in LVO patients is necessary, and a considerable BP reduction before intravenous thrombolysis or endovascular therapy should be avoided.

7.4 Conclusions

Several brain (micro-) circulation imaging features were explored regarding their associations with prognosis in thrombolysis-treated AIS patients, and their influence on the effects of randomised alteplase dose or BP lowering target in the ENCHANTED trial. In Chapter 2, the study shows that prior AF, proximal arterial occlusion, anterior circulation, or more extensive infarct, determine the presence of FLAIR-HAs on baseline brain MRI in AIS patients. Despite a trend towards an increased risk of ICH, FLAIR-HAs are not a reason for withholding thrombolysis, and indicate the potential for a favourable prognosis in thrombolysis-treated AIS patients, including those with large clots burden or LVO. The meta-analysis in Chapter 3 shows that FLAIR-HAs are not associated with the functional outcome overall, but are associated with outcomes after endovascular therapy for AIS. FLAIR-HAs are also associated with early recanalisation or haemorrhagic complications, and early neurologic deterioration. In Chapter 4, no evidence was found from the ENCHANTED trial that low-dose alteplase has an advantage over standard doses for definite/probable lacunar AIS. In Chapter 5, there are no differences in the treatment effect of early intensive versus guideline-recommended BP lowering across

lacunar and non-lacunar AIS. In Chapter 6, functional outcome by the dose of alteplase or intensity of BP lowering is also not modified by vascular obstruction status/site.

Overall, brain (micro-) circulation imaging features on CT or MRI are as important as some clinical characteristics and other NCCT imaging markers in predicting functional recovery and sICH in thrombolysis-treated AIS patients. Finally, I did not identify any imaging markers modifying the treatment effects of randomised alteplase dose and BP lowering in these analyses from the ENCHANTED trial.

References

1. Cheng B, Ebinger M, Kufner A, et al. Hyperintense vessels on acute stroke fluid-attenuated inversion recovery imaging: associations with clinical and other MRI findings. Stroke 2012;43(11):2957-61.
2. Nagaraja N, Forder JR, Warach S, et al. Reversible diffusion-weighted imaging lesions in acute ischaemic stroke: a systematic review. Neurology 2020;94(13):571-87.

Thesis Appendices

Appendix A. The main paper of the ENCHANTED alteplase dose arm

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Low-Dose versus Standard-Dose Intravenous Alteplase in Acute Ischemic Stroke

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ABSTRACT

BACKGROUND

Thrombolytic therapy for acute ischemic stroke with a lower-than-standard dose of intravenous alteplase may improve recovery along with a reduced risk of intracerebral hemorrhage.

METHODS

Using a 2-by-2 quasi-factorial open-label design, we randomly assigned 3310 patients who were eligible for thrombolytic therapy (median age, 67 years; 63% Asian) to low-dose intravenous alteplase (0.6 mg per kilogram of body weight) or the standard dose (0.9 mg per kilogram); patients underwent randomization within 4.5 hours after the onset of stroke. The primary objective was to determine whether the low dose would be noninferior to the standard dose with respect to the primary outcome of death or disability at 90 days, which was defined by scores of 2 to 6 on the modified Rankin scale (range, 0 [no symptoms] to 6 [death]). Secondary objectives were to determine whether the low dose would be superior to the standard dose with respect to centrally adjudicated symptomatic intracerebral hemorrhage and whether the low dose would be noninferior in an ordinal analysis of modified Rankin scale scores (testing for an improvement in the distribution of scores). The trial included 935 patients who were also randomly assigned to intensive or guideline-recommended blood-pressure control.

RESULTS

The primary outcome occurred in 855 of 1607 participants (53.2%) in the low-dose group and in 817 of 1599 participants (51.1%) in the standard-dose group (odds ratio, 1.09; 95% confidence interval [CI], 0.95 to 1.25; the upper boundary exceeded the noninferiority margin of 1.14; P=0.51 for noninferiority). Low-dose alteplase was noninferior in the ordinal analysis of modified Rankin scale scores (unadjusted common odds ratio, 1.00; 95% CI, 0.89 to 1.13; P=0.04 for noninferiority). Major symptomatic intracerebral hemorrhage occurred in 1.0% of the participants in the low-dose group and in 2.1% of the participants in the standard-dose group (P=0.01); fatal events occurred within 7 days in 0.5% and 1.5%, respectively (P=0.01). Mortality at 90 days did not differ significantly between the two groups (8.5% and 10.3%, respectively; P=0.07).

CONCLUSIONS

This trial involving predominantly Asian patients with acute ischemic stroke did not show the noninferiority of low-dose alteplase to standard-dose alteplase with respect to death and disability at 90 days. There were significantly fewer symptomatic intracerebral hemorrhages with low-dose alteplase. (Funded by the National Health and Medical Research Council of Australia and others; ENCHANTED ClinicalTrials.gov number, NCT01422616.)

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Thrombolytic therapy with intravenous alteplase (recombinant tissue-type plasminogen activator) at a dose of 0.9 mg per kilogram of body weight is an effective treatment for acute ischemic stroke, despite increasing the risk of intracerebral hemorrhage.[1-3] However, the Japanese drug safety authority has approved the use of alteplase at a dose of 0.6 mg per kilogram after an uncontrolled, open-label study showed that this dose resulted in equivalent clinical outcomes and a lower risk of intracerebral hemorrhage than that reported in published studies in which the 0.9-mg-per-kilogram dose was used.[4] Other registry studies in Asia[5-11] have shown inconsistent results, but a high risk of symptomatic intracerebral hemorrhage was observed among Asian patients treated with 0.9 mg of alteplase per kilogram in the United States.[12] Differing perceived risks of intracerebral hemorrhage and treatment affordability have led to variations in the doses of intravenous alteplase used to treat patients with acute ischemic stroke in Asia.[8-11]

The Enhanced Control of Hypertension and Thrombolysis Stroke Study (ENCHANTED) was designed to compare low-dose with standard-dose intravenous alteplase in patients with acute ischemic stroke. Using a quasi-factorial design, we are also assessing the effects of early intensive lowering of blood pressure as compared with guideline-recommended management in patients with elevated blood pressure; this part of the trial is scheduled to be completed in 2018. We report the results of the alteplase part of the trial, which was completed in December 2015.

METHODS

TRIAL DESIGN AND OVERSIGHT

In an international, multicenter, prospective, randomized, open-label trial with blinded outcome evaluation, two doses of intravenous alteplase were compared in patients with an acute ischemic stroke who were eligible for thrombolytic therapy; administration of the drug was commenced within 4.5 hours after the onset of the stroke. Patients with elevated systolic blood pressure (range, 150 to 220 mm Hg) could also be randomly assigned to early and intensive lowering of blood pressure (target systolic blood pressure <140 mm Hg within 1 hour) or conventional guideline-directed management of blood pressure (target systolic blood pressure <180 mm Hg) with the use of locally available intravenous agents.

Details of the design and statistical analysis plan of the trial have been published previously.[13,14] An international steering committee, whose members designed the trial with an advisory committee, was responsible for the conduct and reporting of the trial. The George Institute for Global Health coordinated the trial, managed the database, and performed the analyses. The study drug used (alteplase) was that available for routine use at clinical centers; there was no commercial input into any aspect of the trial. The first author wrote the first and subsequent drafts of the manuscript. All the authors commented on drafts of the manuscript, approved the decision to submit the manuscript for publication, and vouch for the accuracy and completeness of the data and for the fidelity of this report to the trial protocol (available with the full text of this article at NEJM.org).

The trial protocol was approved by all appropriate regulatory authorities and ethics committees at the participating centers. All participants, or an approved surrogate for those who were too unwell to comprehend the information, provided written informed consent. Details of the monitoring procedures are provided in the Supplementary Appendix (available at NEJM.org).

PATIENTS AND PROCEDURES

Patients were recruited at 111 clinical centers in 13 countries. Patients were eligible if they were 18 years of age or older, had an acute ischemic stroke, and met guideline-recommended criteria for treatment with intravenous alteplase. For details of the inclusion and exclusion criteria, see the Supplementary Appendix.

After confirmation of patient eligibility, randomization was performed centrally with the use of a minimization algorithm according to center, time from stroke onset (<3 vs. ≥3 hours), and severity of neurologic impairment (score of <10 vs. ≥10 on the National Institutes of Health Stroke Scale [NIHSS]; range, 0 to 42, with higher scores indicating greater severity of stroke). Participants were randomly assigned to receive either a standard dose of intravenous alteplase (0.9 mg per kilogram of estimated, or measured, body weight; 10% as a bolus and 90% as an infusion over a period of 60 minutes; maximum dose, 90 mg) or a low dose (0.6 mg per kilogram, 15% as a bolus and 85% as an infusion over a period of 60 minutes; maximum dose, 60 mg), to be commenced within 4.5 hours after symptom onset. Concomitant therapy followed national practice guidelines,

including the use of endovascular thrombectomy devices, where approved.

Demographic and clinical data were obtained at the time of randomization. Follow-up data were obtained at 24 and 72 hours (including repeat NIHSS scores and measured body weight) and at 7 days (or hospital discharge, if sooner), 28 days, and 90 days, unless death occurred earlier. The 28-day and 90-day evaluations were conducted in person or by telephone, by trained and certified staff who remained unaware of the randomized treatment assignments. Brain imaging was performed at trial entry and at 24 hours, and additionally if clinically indicated, and was analyzed centrally for any hemorrhage by expert assessors who were unaware of the treatment assignments (see the Supplementary Appendix).

OUTCOMES

The prespecified primary outcome was the combined end point of death or disability at 90 days, which was defined by scores of 2 to 6 on the modified Rankin scale,[15] a global seven-level measure of functioning in which scores of 0 or 1 indicate a good outcome with no or minimal neurologic symptoms, scores of 2 to 5 indicate a poor outcome with increasing degree of disability, and 6 indicates death. The key secondary outcome, which was also designated as a safety outcome, was intracerebral hemorrhage, defined according to criteria from a number of other studies (see the Supplementary Appendix); the main definition of intracerebral hemorrhage that we used was the definition in the Safe Implementation of Thrombolysis in Stroke-Monitoring Study (SITS-MOST)[16]: a large local or remote parenchymal pattern (>30% of the infarcted area affected by hemorrhage, with mass effect or extension outside the infarct) and neurologic deterioration from baseline (increase of ≥4 points in the NIHSS score) or death within 36 hours. This definition was finalized and described in the statistical analysis plan of our trial after publication of the original protocol.

Other secondary efficacy outcomes were the distribution of modified Rankin scale scores at 90 days,[17] major disability (modified Rankin scale score >2) at 90 days, deaths at 7 days and 90 days, neurologic deterioration (increase of ≥4 points in the NIHSS score) during the 72 hours after randomization, death and neurologic deterioration (increase of ≥4 points in the NIHSS score) during the 72 hours after randomization, health-related quality of life on the EuroQoL Group 5-Dimension Self-Report Questionnaire (EQ-5D; summary health utility scores range from −0.109 to 1, with higher scores indicating better health)[18] at 90 days (for details on scoring, see the Supplementary Appendix), length of initial hospital stay, recurrent acute myocardial infarction and recurrent ischemic stroke, admission to a long-term residential care facility at 90 days, and use of health services (for economic analyses that have not yet been conducted). Prespecified safety outcomes were all serious adverse events reported until trial completion. Tertiary outcomes included all-cause mortality, place of death, trends in modified Rankin scale scores during follow-up, length of stay in the intensive care unit (ICU), rate of thrombectomy, and individual items of the EQ-5D.

STATISTICAL ANALYSIS

We designed the trial to assess the effects of two treatment variables — alteplase dose and intensity of blood-pressure control — on clinical outcomes, accounting for their potential interaction.[14] Differential patient recruitment resulted in the part of the trial dealing with alteplase dose being completed faster than the part dealing with intensity of blood-pressure control. For the primary analysis, we used an unadjusted logistic-regression model to test whether low-dose alteplase was noninferior to the standard dose. To satisfy the noninferiority hypothesis, the upper boundary of the 95% confidence interval for the odds ratio of the outcome with low-dose alteplase as compared with standard-dose alteplase had to fall below a margin of 1.14; this noninferiority margin was derived from a Cochrane meta-analyses of alteplase trials with effects on poor outcomes reported.[19,20] We estimated that a sample size of 3300 patients would provide at least 90% power to evaluate noninferiority, assuming 5% dropout and potential negative interaction between intensive blood-pressure control and low-dose alteplase, and would also provide at least 80% power to detect superiority of low-dose alteplase in achieving a 40% lower risk of symptomatic intracerebral hemorrhage than that with standard-dose alteplase, with 5% dropout. Consistency of treatment effect across 10 prespecified subgroups was assessed through tests for interaction.

A secondary efficacy analysis was a comparison of ordinal scores on the modified Rankin scale to test for the noninferiority of the low dose to the standard dose with the use of ordi-

Table 1. Characteristics of the Patients at Baseline and Their Treatment.*

Variable	Low-Dose Alteplase (N=1654)	Standard-Dose Alteplase (N=1643)
Age — yr		
Median	68	67
IQR	58–76	58–76
Female sex — no. (%)	634 (38.3)	614 (37.4)
Region of recruitment — no. (%)		
China	708 (42.8)	711 (43.3)
United Kingdom, continental Europe, or Australia	445 (26.9)	439 (26.7)
Asia, other than China	336 (20.3)	334 (20.3)
South America	165 (10.0)	159 (9.7)
Asian race — no./total no. (%)†	1043/1651 (63.2)	1036/1640 (63.2)
Medical history — no./total no. (%)‡		
Hypertension	1031/1648 (62.6)	1034/1640 (63.0)
Any stroke	287/1654 (17.4)	302/1643 (18.4)
Coronary artery disease	256/1648 (15.5)	223/1640 (13.6)
Atrial fibrillation	330/1645 (20.1)	306/1640 (18.7)
Diabetes mellitus	325/1648 (19.7)	321/1640 (19.6)
Hypercholesterolemia	297/1648 (18.0)	258/1640 (15.7)
Current cigarette use	377/1646 (22.9)	393/1638 (24.0)
Modified Rankin scale score of 0 before stroke§	1349/1647 (81.9)	1325/1639 (80.8)
Use of antihypertensive agent	755/1648 (45.8)	743/1640 (45.3)
Use of statin or other lipid-lowering agent	333/1646 (20.2)	282/1638 (17.2)
Use of aspirin or other antiplatelet agent	407/1647 (24.7)	345/1638 (21.1)
Warfarin anticoagulation	48/1647 (2.9)	34/1638 (2.1)
Blood pressure — mm Hg		
Systolic	149±20	150±20
Diastolic	84±13	85±13
NIHSS score — median (IQR)¶	8 (5–14)	8 (5–14)
Signs of cerebral ischemia on brain imaging — no./total no. (%)‖	388/1648 (23.5)	383/1640 (23.4)
Proximal vessel occlusion on CTA or MRA — no./total no. (%)‖	258/1622 (15.9)	248/1624 (15.3)
Final diagnosis at time of hospital discharge — no./total no. (%)**		
Nonstroke diagnosis	50/1625 (3.1)	47/1609 (2.9)
Large-artery occlusion due to clinically significant atheroma	622/1625 (38.3)	648/1609 (40.3)
Small-vessel or perforator lacunar disease	334/1625 (20.6)	339/1609 (21.1)
Cardioembolism	324/1625 (19.9)	317/1609 (19.7)
Dissection	14/1625 (0.9)	11/1609 (0.7)
Other or uncertain cause of stroke	281/1625 (17.3)	247/1609 (15.4)
Time from stroke onset to alteplase administration — min††		
Median	170	170
IQR	125–218	127–219
Estimated body weight before alteplase administration — kg	69.6±14.4	69.9±14.4

Table 1. (Continued.)

Variable	Low-Dose Alteplase (N=1654)	Standard-Dose Alteplase (N=1643)
Any alteplase given to patients — no. (%)	1628 (98.4)	1617 (98.4)
Bolus dose — mg	6.2±1.2	6.3±2.1
Infusion dose — mg	35.5±7.3	56.0±11.3
Concurrent inclusion in part of trial dealing with blood-pressure control — no. (%)	460 (27.8)	475 (28.9)
Assigned to intensive blood-pressure lowering	224 (13.5)	232 (14.1)
Assigned to standard blood-pressure lowering	236 (14.3)	243 (14.8)

* Plus–minus values are means ±SD. There were no significant differences between trial groups in the characteristics listed, except in the administered dose of alteplase as a bolus (P=0.05) and as an infusion (P<0.001). CTA denotes computed tomographic angiography, IQR interquartile range, and MRA magnetic resonance angiography.
† Race was self-reported.
‡ Medical history was based on self-report, with the exception of the presence of atrial fibrillation, which was based on findings on electrocardiography performed at the time of presentation.
§ The modified Rankin scale evaluates global disability; scores range from 0 (no symptoms) to 6 (death).
¶ Scores on the National Institutes of Health Stroke Scale (NIHSS) range from 0 to 42, with higher scores indicating more severe neurologic deficits. Scores ranged from 0 to 40 in the low-dose group and from 0 to 42 in the standard-dose group.
‖ This finding was reported by clinician investigators.
** Diagnoses were determined by clinician investigators.
†† Times ranged from 28 to 1673 minutes (0.47 to 27.88 hours) in the low-dose group and from 11 to 1678 minutes (0.18 to 27.97 hours) in the standard-dose group.

nal logistic regression, after the assumption of proportionality of the odds was confirmed in a likelihood-ratio test.[17] A new assumption-free approach[21] was used to confirm the conclusion. We also performed secondary analyses of the primary outcome with adjustment for minimization and key prognostic covariates[14], as well as secondary analyses in the per-protocol population according to criteria outlined in the Supplementary Appendix. Multiple imputation was to be used if more than 10% of observations were missing. An independent data and safety monitoring board monitored the trial progress and safety with the use of formal stopping boundaries. For ease of interpretation, all reported P values for noninferiority are multiplied by 2 so that an alpha of 0.05 can be used in analyses. The other P values (those for superiority) are two-sided. All P values were prespecified not to be adjusted. SAS software, version 9.3 (SAS Institute), was used for analyses.

RESULTS

PATIENTS

From March 2012 through August 2015, a total of 3310 of the 69,325 patients who were screened underwent randomization (Fig. S1 and Table S1 in the Supplementary Appendix); 1654 patients were assigned to low-dose alteplase and 1643 to standard-dose alteplase. There were no significant differences between the low-dose group and the standard-dose group in baseline demographic and clinical characteristics (Table 1, and Table S2 in the Supplementary Appendix), nor in the number of patients assigned to each of the blood-pressure-lowering groups. The median age of the patients was 67 years (14% were ≥80 years of age), and 38% were women. Approximately two thirds of the patients were Asian, and 43% were recruited from China. The median NIHSS score before treatment was 8 (range, 0 to 42; interquartile range, 5 to 14), and the median time from stroke onset to randomization was 2.7 hours (interquartile range, 2.0 to 3.5).

INTERVENTIONS

In both groups, the mean time from the onset of stroke to administration of alteplase was 170 minutes; the mean dose of alteplase administered as an infusion was 35.5 mg in the low-dose group and 56.0 mg in the standard-dose group (P<0.001) (Table 1). The distribution of dosing and the protocol violations are outlined in Figure

S2 and Tables S3 and S4 in the Supplementary Appendix. Postrandomization management, including endovascular thrombectomy and rates of recanalization, was similar in the two groups during the first 7 days (Table S5 in the Supplementary Appendix). Outcome at 90 days was assessed by telephone in 84.6% of the patients in the low-dose group and in 83.6% of the patients in the standard-dose group; the rest of the patients were assessed by in-person examination. The sources of information for the modified Rankin scale scores were balanced between the groups (Table S6 in the Supplementary Appendix).

BLOOD-PRESSURE CONTROL

In the part of the trial dealing with blood-pressure control that included 935 patients with elevated systolic blood pressure (range, 150 to 220 mm Hg), 224 patients in the low-dose group (13.5%) and 232 in the standard-dose group (14.1%) were assigned to rapid blood-pressure reduction. In both alteplase dose groups, the mean systolic blood pressure levels were significantly lower, by 7 to 9 mm Hg, with intensive blood-pressure control than with standard blood-pressure management at 1 hour and 6 hours after randomization (Table S7 in the Supplementary Appendix).

PRIMARY OUTCOME

Scores on the modified Rankin scale for assessment of the primary outcome could not be obtained, owing to withdrawal of consent or loss to follow-up, in 47 of the patients assigned to low-dose alteplase and 44 assigned to standard-dose alteplase (Fig. S1 in the Supplementary Appendix). In the modified intention-to-treat analysis, the primary outcome (scores of 2 to 6 on the modified Rankin scale) occurred in 855 of 1607 patients (53.2%) in the low-dose group and in 817 of 1599 patients (51.1%) in the standard-dose group (odds ratio, 1.09; 95% confidence interval [CI], 0.95 to 1.25; the upper boundary of the 95% confidence interval exceeded the prespecified boundary for noninferiority of 1.14; one-sided P=0.51 for noninferiority) (Table 2, and Fig. S3 in the Supplementary Appendix). In an adjusted analysis of the intention-to-treat population, the rate was 53.3% in the low-dose group and 50.9% in the standard-dose group (odds ratio, 1.13; 95% CI, 0.97 to 1.31; P=0.88 for noninferiority); in an adjusted analysis in the per-protocol population, the rates were 53.5% and 51.3%, respectively (odds ratio, 1.13; 95% CI, 0.96 to 1.32; one-sided P=0.89 for noninferiority). There was no heterogeneity of effect between patients who began alteplase treatment less than 3 hours after stroke onset and those who began treatment 3 or more hours after stroke onset (Fig. S5 in the Supplementary Appendix), and there was no significant interaction between intensity of blood-pressure lowering and alteplase dose (P=0.29).

SECONDARY OUTCOMES

In an unadjusted ordinal analysis of the distribution of modified Rankin scale scores in the two groups, the odds ratio with low-dose alteplase as compared with standard-dose alteplase was 1.00 (95% CI, 0.89 to 1.13; P=0.04 for noninferiority) (Table 2). The assumption-free, adjusted, and per-protocol alternative approaches were consistent in showing no significant difference in the treatment effect for overall functional outcome on the modified Rankin scale between doses of alteplase (Fig. 1, and Figs. S3, S4, and S6 and Tables S8, S9, and S10 in the Supplementary Appendix).

Major symptomatic intracerebral hemorrhage according to SITS-MOST criteria occurred in 17 of 1654 patients (1.0%) in the low-dose group and in 35 of 1643 patients (2.1%) in the standard-dose group (odds ratio, 0.48; 95% CI, 0.27 to 0.86; P=0.01) (Table 2). There was no significant interaction between intensive blood-pressure lowering and alteplase dose group with respect to the risk of major symptomatic intracerebral hemorrhage (P=0.71). Symptomatic intracerebral hemorrhage according to other criteria also occurred significantly less frequently in the low-dose group than in the standard-dose group (Table 2, and Fig. S7 in the Supplementary Appendix); for example, the rate of fatal events within 7 days was 0.5% in the low-dose group and 1.5% in the standard-dose group (odds ratio, 0.37; 95% CI, 0.17 to 0.80; P=0.01). There was no heterogeneity in the effect of alteplase dose on the risk of symptomatic intracerebral hemorrhage between Asians and non-Asians (Fig. S8 in the Supplementary Appendix).

Mortality at 7 days was 3.6% in the low-dose group versus 5.3% in the standard-dose group (odds ratio, 0.67; 95% CI, 0.48 to 0.94; P=0.02), and mortality at 90 days was 8.5% versus 10.3%

Table 2. Primary and Secondary Outcomes at 3 Months.*

Outcome	Low-Dose Alteplase (N=1654)	Standard-Dose Alteplase (N=1643)	Odds Ratio with Low-Dose Alteplase (95% CI)	P Value†	P Value for Noninferiority‡
Primary outcome: death or disability — no./total no. (%)§	855/1607 (53.2)	817/1599 (51.1)	1.09 (0.95 to 1.25)		0.51
Secondary outcomes					
Symptomatic intracerebral hemorrhage — no. (%)					
By SITS-MOST criteria¶	17 (1.0)	35 (2.1)	0.48 (0.27 to 0.86)	0.01	
By NINDS criteria‖	98 (5.9)	131 (8.0)	0.73 (0.55 to 0.95)	0.02	
Score on the modified Rankin scale — no./total no. (%)			1.00 (0.89 to 1.13)**		0.04
0: No symptoms at all	403/1607 (25.1)	397/1599 (24.8)			
1: No substantive disability despite symptoms	349/1607 (21.7)	385/1599 (24.1)			
2: Slight disability	250/1607 (15.6)	225/1599 (14.1)			
3: Moderate disability requiring some help	211/1607 (13.1)	181/1599 (11.3)			
4: Moderate–severe disability requiring assistance with daily living	165/1607 (10.3)	154/1599 (9.6)			
5: Severe disability, bed-bound and incontinent	89/1607 (5.5)	87/1599 (5.4)			
6: Death	140/1607 (8.7)	170/1599 (10.6)			
Death or major disability — no./total no. (%)††	605/1607 (37.6)	592/1599 (37.0)	1.03 (0.89 to 1.19)	0.73	
Death within 90 days — no. (%)	140 (8.5)	170 (10.3)	0.80 (0.63 to 1.01)	0.07	
Overall health utility score on the EQ-5D‡‡:	0.64±0.40	0.64±0.41	0.00 (–0.03 to 0.03)§§	0.86	
Admission to residential care — no./total no. (%)	36/1513 (2.4)	43/1476 (2.9)	0.81 (0.52 to 1.27)	0.36	
Median duration of hospitalization (IQR) — days	10 (5 to 17)	10 (5 to 18)	–0.47 (–1.93 to 1.00)§§	0.53	
Death or neurologic deterioration in 72 hr — no. (%)¶¶	177 (10.7)	192 (11.7)	0.91 (0.73 to 1.12)	0.37	
Serious adverse event — no. (%)‖‖	415 (25.1)	448 (27.3)	0.89 (0.76 to 1.04)	0.16	

* Plus–minus values are means ±SD. CI denotes confidence interval.
† The P values are for the comparison of the low-dose group with the standard-dose group.
‡ The noninferiority margin was 1.14 (i.e., an upper boundary of the 95% confidence interval for the odds ratio with low-dose alteplase as compared with standard-dose alteplase of less than 1.14).
§ Disability was defined by a score of 2 to 5 on the modified Rankin scale, with higher scores indicating a greater degree of disability. Death was defined by a modified Rankin scale score of 6.
¶ The main definition of symptomatic intracerebral hemorrhage used in the study was the definition from the Safe Implementation of Thrombolysis in Stroke-Monitoring Study (SITS-MOST): a large local or remote parenchymal intracerebral hemorrhage (>30% of the infarcted area affected by hemorrhage with mass effect or extension outside the infarct) in combination with neurologic deterioration from baseline (increase of ≥4 in the NIHSS score) or death within 36 hours.
‖ Symptomatic intracerebral hemorrhage was also assessed according to National Institute of Neurological Diseases and Stroke (NINDS) trial criteria: any intracerebral hemorrhage with neurologic deterioration (increase of ≥1 in the NIHSS score) from baseline or death within 36 hours.
** The common odds ratio was estimated from an ordinal logistic-regression model and indicates the odds of a decrease of 1 in the modified Rankin scale score, with a common odds ratio greater than 1 favoring standard-dose alteplase.
†† Major disability was defined by a score of 3 to 5 on the modified Rankin scale.
‡‡ The EuroQoL Group 5-Dimension Self-Report Questionnaire (EQ-5D) covers five domains of health-related quality of life: mobility, self-care, usual activities, pain–discomfort, and anxiety–depression. Scores from these levels are combined to provide an overall health utility score ranging from –0.109 to 1, with higher scores indicating better health. Data were available for 1594 patients in the low-dose group and 1591 patients in the standard-dose group.
§§ Shown in the estimated difference with 95% confidence interval.
¶¶ Neurologic deterioration was defined as an increase from baseline of at least 4 in the NIHSS score.
‖‖ Serious adverse events were defined by standard criteria and included those that resulted in death, were life-threatening, required inpatient hospitalization or prolongation of an existing hospitalization, resulted in persistent or substantive disability or incapacity, or resulted in a medical or surgical intervention to prevent permanent impairment to body structure or function. Details on serious adverse events are provided in Table S12 in the Supplementary Appendix.

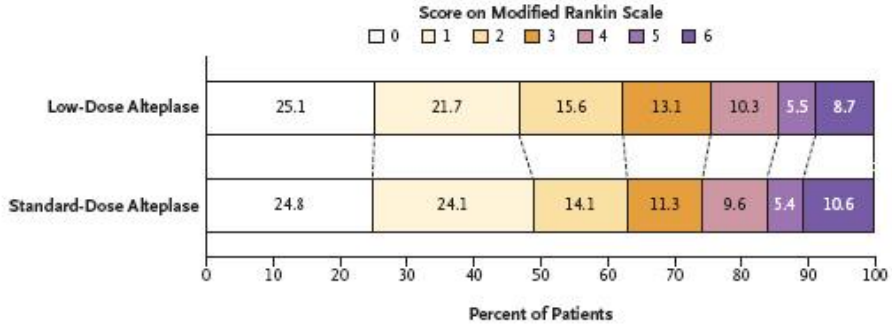

Figure 1. Functional Outcomes at 90 Days, According to Score on the Modified Rankin Scale.
Shown is the raw distribution of scores on the modified Rankin scale at 90 days in the group that received a low dose of alteplase (0.6 mg per kilogram of body weight) and the group that received the standard dose of alteplase (0.9 mg per kilogram). Scores on the modified Rankin scale range from 0 to 6, with 0 indicating no symptoms, 1 symptoms without clinically significant disability, 2 slight disability, 3 moderate disability, 4 moderately severe disability, 5 severe disability, and 6 death.

(odds ratio, 0.80; 95% CI, 0.63 to 1.01; P=0.07) (Fig. S9 and Table S11 in the Supplementary Appendix). No significant between-group differences were evident in other secondary outcomes (Table 2). Other outcomes, including length of stay in the ICU, recurrent vascular events, and individual components of the EQ-5D, are not reported here.

SUBGROUP ANALYSES

There was no significant heterogeneity of treatment effect on the primary outcome across prespecified subgroups (Fig. 2). The interaction between alteplase dose and aspirin or other antiplatelet therapy was not significant (P=0.05); however, this interaction was significant (P=0.02) in a post hoc unadjusted ordinal analysis of modified Rankin scale scores (Fig. S10 in the Supplementary Appendix). Post hoc analyses showed consistency in the effect of alteplase dose on death at 90 days across subgroups and no clear relation with baseline NIHSS score (Fig. S11 and S12 in the Supplementary Appendix).

SAFETY

There was no significant difference between the low-dose group and the standard-dose group in the overall reported rate of serious adverse events, which occurred in 25.1% and 27.3% of the patients, respectively (P=0.16). However, significantly fewer patients in the low-dose alteplase group than in the standard-dose alteplase group had (unadjudicated) fatal cerebral hemorrhage events (1.3% vs. 2.5%, P=0.02). All serious adverse events are listed in Table S12 in the Supplementary Appendix.

DISCUSSION

In patients with acute ischemic stroke who met guideline-recommended criteria for thrombolytic reperfusion treatment, a dose of 0.6 mg of alteplase per kilogram was not shown to be noninferior to a dose of 0.9 mg of alteplase per kilogram with respect to the primary outcome of death or disability at 90 days. Our trial was driven by concern about high risks of intracerebral hemorrhage with alteplase, particularly among Asians, because preliminary studies have had differing results with respect to the effectiveness and risks of this treatment.[8,11] Using several definitions of symptomatic intracerebral hemorrhage,[22] we observed fewer clinically important cases in the group assigned to low-dose alteplase than in the group assigned to standard-dose alteplase, and the difference in risk was consistent in Asians and non-Asians.

The distribution of modified Rankin scale scores in the two groups at 90 days indicates that the lower rate of death with low-dose alteplase than with standard-dose alteplase was accompanied by more patients surviving with mild to moderately severe grades of residual disability. There was no heterogeneity of treatment effect on the primary outcome in prespecified subgroups, but these analyses had low statistical

Subgroup	Low-Dose Alteplase	Standard-Dose Alteplase	Odds Ratio (95% CI)	P Value for Interaction
	no. (%)			
Age				0.20
<65 yr	302 (43.9)	301 (44.3)	0.98 (0.79–1.22)	
≥65 yr	553 (60.2)	516 (56.1)	1.18 (0.98–1.42)	
Sex				0.53
Male	503 (50.9)	480 (48.0)	1.12 (0.94–1.34)	
Female	352 (57.0)	337 (56.4)	1.02 (0.82–1.29)	
Race				0.82
Asian	527 (51.5)	500 (49.0)	1.10 (0.93–1.31)	
Non-Asian	328 (56.4)	317 (54.7)	1.07 (0.85–1.35)	
Time from stroke onset to randomization				0.63
<3 hr	536 (54.5)	497 (51.8)	1.12 (0.93–1.34)	
≥3 hr	319 (51.1)	320 (50.1)	1.04 (0.84–1.30)	
Baseline systolic blood pressure				0.08
≤150 mm Hg	404 (50.6)	402 (51.6)	0.96 (0.79–1.17)	
>150 mm Hg	451 (55.7)	415 (50.6)	1.23 (1.01–1.49)	
Baseline NIHSS score				0.31
≤8	282 (34.9)	282 (34.6)	1.01 (0.83–1.24)	
>8	573 (71.8)	535 (68.3)	1.18 (0.95–1.46)	
Final diagnosis of ischemic stroke				0.88
Large-artery atheroma occlusion	356 (58.0)	362 (56.7)	1.05 (0.84–1.32)	
Small-vessel disease	114 (34.4)	110 (33.0)	1.07 (0.77–1.47)	
Cardioembolism	212 (67.5)	193 (61.7)	1.29 (0.93–1.79)	
Other definite or uncertain cause	138 (51.1)	117 (49.2)	1.08 (0.76–1.53)	
Cerebral infarction on CT				0.66
Yes	205 (58.9)	220 (58.0)	1.04 (0.77–1.39)	
No	649 (51.7)	597 (48.9)	1.12 (0.95–1.31)	
Use of antiplatelet agent				0.05
Yes	222 (56.3)	204 (60.7)	0.84 (0.62–1.12)	
No	632 (52.3)	612 (48.5)	1.16 (0.99–1.36)	
Evidence of atrial fibrillation				0.34
Yes	249 (66.9)	233 (68.3)	0.94 (0.69–1.28)	
No	603 (49.1)	584 (46.4)	1.11 (0.95–1.30)	

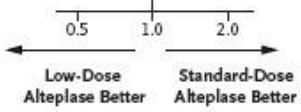

Figure 2. Effects of Low-Dose Alteplase as Compared with Standard-Dose Alteplase on the Primary Efficacy Outcome, According to Prespecified Subgroups.

The primary efficacy outcome was death or disability at 90 days, defined by scores of 2 to 6 on the modified Rankin scale (range, 0 [no symptoms] to 6 [death]). Scores on the National Institutes of Health Stroke Scale (NIHSS) range from 0 to 42, with higher scores indicating more severe neurologic deficits. For subcategories, black squares represent point estimates (with the area of the square proportional to the number of events), and horizontal lines represent 95% confidence intervals. For systolic blood pressure and NIHSS score, values are equal to or above the median of distribution versus below the median of distribution. CT denotes computed tomography.

power. One fifth of our trial population were receiving antiplatelet therapy. Previous studies have shown an increased risk of intracerebral hemorrhage with standard-dose alteplase among patients receiving antiplatelet therapy.[23,24] In our prespecified analysis, there was no significant interaction between alteplase dose and antiplatelet treatment with respect to a poor outcome; however, the interaction was significant in a post hoc ordinal analysis of modified Rankin scale scores. The ongoing part of this trial is testing the effectiveness of intensive and early reduction of elevated systolic blood pressure on outcomes in patients with ischemic stroke. Analyses did not indicate any significant interaction between early intensive blood-pressure lowering and alteplase dose.

Efforts to minimize reporting biases in this

open-label trial included the measurement of body weight, blinded central adjudication of intracerebral hemorrhage, and blinded evaluation of clinical outcomes with the use of established criteria. Imprecision in estimates of the treatment effect may have arisen from interobserver variability in determining the scores on the modified Rankin scale,[15,25] which was administered principally by telephone. Analysis of the net change in functional outcome was based on equal weights assigned to each score (0 through 6) on the modified Rankin scale, but patient and provider assessments can vary across health transitions,[26] and functional recovery can continue beyond 90 days.[27] In our trial, selection bias was due to the inclusion of patients who had a predominantly mild severity of neurologic impairment and who were treated at a later time point after symptom onset than in other trials[1,3,4] and than in quality-assurance studies[12,16,28] of the use of alteplase in patients with acute ischemic stroke. The high percentage of Asian participants and concurrent intensive blood-pressure control in some patients may also raise concerns about generalizability, despite the finding that there were no significant interactions observed between Asians and non-Asians, nor with intensity of blood-pressure control.

In conclusion, in a group of predominantly Asian patients with acute ischemic stroke who were eligible for thrombolysis reperfusion therapy, a dose of alteplase of 0.6 mg per kilogram was not shown to be noninferior to the standard dose of 0.9 mg per kilogram with respect to the primary outcome of death and disability. Fewer patients treated with low-dose alteplase than with standard-dose alteplase (1% vs. 2%) had the secondary outcome of symptomatic intracerebral hemorrhage.

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APPENDIX

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REFERENCES

1. Emberson J, Lees KR, Lyden P, et al. Effect of treatment delay, age, and stroke severity on the effects of intravenous thrombolysis with alteplase for acute ischaemic stroke: a meta-analysis of individual patient data from randomised trials. Lancet 2014;384:1929-35.
2. Wardlaw JM, Murray V, Berge E, et al. Recombinant tissue plasminogen activator for acute ischaemic stroke: an updated systematic review and meta-analysis. Lancet 2012;379:2364-72.
3. The National Institute of Neurological Disorders and Stroke rt-PA Stroke Study Group. Tissue plasminogen activator for acute ischemic stroke. N Engl J Med 1995;333:1581-7.
4. Yamaguchi T, Mori E, Minematsu K, et al. Alteplase at 0.6 mg/kg for acute ischemic stroke within 3 hours of onset: Japan Alteplase Clinical Trial (J-ACT). Stroke 2006;37:1810-5.
5. Toyoda K, Koga M, Naganuma M, et al. Routine use of intravenous low-dose recombinant tissue plasminogen activator in Japanese patients: general outcomes and prognostic factors from the SAMURAI register. Stroke 2009;40:3591-5.
6. Nakagawara J, Minematsu K, Okada Y, et al. Thrombolysis with 0.6 mg/kg intravenous alteplase for acute ischemic stroke in routine clinical practice: the Japan post-Marketing Alteplase Registration Study (J-MARS). Stroke 2010;41:1984-9.
7. Chao AC, Hsu HY, Chung CP, et al. Outcomes of thrombolytic therapy for acute ischemic stroke in Chinese patients: the Taiwan Thrombolytic Therapy for Acute Ischemic Stroke (TTT-AIS) study. Stroke 2010;41:885-90.
8. Kim EJ, Han MK, Park TH, et al. Low- versus standard-dose alteplase for ischemic strokes within 4.5 hours: a comparative effectiveness and safety study. Stroke 2015;46:2541-8.
9. Liao X, Wang Y, Pan Y, et al. Standard-dose intravenous tissue-type plasminogen activator for stroke is better than low doses. Stroke 2014;45:2354-8.
10. Wang Y, Liao X, Zhao X, et al. Using recombinant tissue plasminogen activator to treat acute ischemic stroke in China: analysis of the results from the Chinese National Stroke Registry (CNSR). Stroke 2011;42:1658-64.
11. Sharma VK, Ng KW, Venketasubramanian N, et al. Current status of intravenous thrombolysis for acute ischemic stroke in Asia. Int J Stroke 2011;6:523-30.
12. Menon BK, Saver JL, Prabhakaran S, et al. Risk score for intracranial hemorrhage in patients with acute ischemic stroke treated with intravenous tissue-type plasminogen activator. Stroke 2012;43:2293-9.
13. Huang Y, Sharma VK, Robinson T, et al. Rationale, design, and progress of the ENhanced Control of Hypertension ANd Thrombolysis strokE stuDy (ENCHANTED) trial: an international multicenter 2×2 quasi-factorial randomized controlled trial of low- vs. standard-dose rt-PA and early intensive vs. guideline-recommended blood pressure lowering in patients with acute ischaemic stroke eligible for thrombolysis treatment. Int J Stroke 2015;10:778-88.
14. Anderson CS, Woodward M, Arima H, et al. Statistical analysis plan for evaluating low- vs. standard-dose alteplase in the ENhanced Control of Hypertension and Thrombolysis strokE stuDy (ENCHANTED). Int J Stroke 2015;10:1313-5.
15. Quinn TJ, Dawson J, Walters MR, Lees KR. Functional outcome measures in contemporary stroke trials. Int J Stroke 2009;4:200-5.
16. Wahlgren N, Ahmed N, Dávalos A, et al. Thrombolysis with alteplase for acute ischaemic stroke in the Safe Implementation of Thrombolysis in Stroke-Monitoring Study (SITS-MOST): an observational study. Lancet 2007;369:275-82.
17. Bath PM, Lees KR, Schellinger PD, et al. Statistical analysis of the primary outcome in acute stroke trials. Stroke 2012;43:1171-8.
18. Rabin R, de Charro F. EQ-5D: a measure of health status from the EuroQol Group. Ann Med 2001;33:337-43.
19. Wardlaw JM, Murray V, Berge E, Del Zoppo GJ. Thrombolysis for acute ischaemic stroke. Cochrane Database Syst Rev 2009;4:CD000213.
20. Wardlaw JM, Murray V, Berge E, del Zoppo GJ. Thrombolysis for acute ischaemic stroke. Cochrane Database Syst Rev 2014;7:CD000213.
21. Howard G, Waller JL, Voeks JH, et al. A simple, assumption-free, and clinically interpretable approach for analysis of modified Rankin outcomes. Stroke 2012;43:664-9.
22. von Kummer R, Broderick JP, Campbell BCV, et al. The Heidelberg bleeding classification: classification of bleeding events after ischemic stroke and reperfusion therapy. Stroke 2015;46:2981-6.
23. Diener HC, Foerch C, Riess H, Röther J, Schroth G, Weber R. Treatment of acute ischaemic stroke with thrombolysis or thrombectomy in patients receiving antithrombotic treatment. Lancet Neurol 2013;12:677-88.
24. Whiteley WN, Slot KB, Fernandes P, Sandercock P, Wardlaw J. Risk factors for intracranial hemorrhage in acute ischemic stroke patients treated with recombinant tissue plasminogen activator: a systematic review and meta-analysis of 55 studies. Stroke 2012;43:2904-9.
25. Quinn TJ, Dawson J, Walters MR, Lees KR. Reliability of the modified Rankin Scale: a systematic review. Stroke 2009;40:3393-5.
26. Chaisinanunkul N, Adeoye O, Lewis RJ, et al. Adopting a patient-centered approach to primary outcome analysis of acute stroke trials using a utility-weighted modified Rankin scale. Stroke 2015;46:2238-43.
27. Sandercock P, Wardlaw JM, Dennis M, et al. Effect of thrombolysis with alteplase within 6 h of acute ischaemic stroke on long-term outcomes (the third International Stroke Trial [IST-3]): 18-month follow-up of a randomised controlled trial. Lancet Neurol 2013;12:768-76.
28. Saver JL, Fonarow GC, Smith EE, et al. Time to treatment with intravenous tissue plasminogen activator and outcome from acute ischemic stroke. JAMA 2013;309:2480-8.

Appendix B. The main paper of the ENCHANTED BP arm

Intensive blood pressure reduction with intravenous thrombolysis therapy for acute ischaemic stroke (ENCHANTED): an international, randomised, open-label, blinded-endpoint, phase 3 trial

Craig S Anderson, Yining Huang*, Richard I Lindley, Xiaoying Chen, Hisatomi Arima, Guofang Chen, Qiang Li, Laurent Billot, Candice Delcourt, Philip M Bath, Joseph P Broderick, Andrew M Demchuk, Geoffrey A Donnan, Alice C Durham, Pablo M Lavados, Tsong-Hai Lee, Christopher Levi, Sheila O Martins, Veronica V Olavarria, Jeyaraj D Pandian, Mark W Parsons, Octavio M Pontes-Neto, Stefano Ricci, Shoichiro Sato, Vijay K Sharma, Federico Silva, Lili Song, Nguyen H Thang, Joanna M Wardlaw, Ji-Guang Wang, Xia Wang, Mark Woodward, John Chalmers†, Thompson G Robinson†, for the ENCHANTED Investigators and Coordinators‡*

Summary

Background Systolic blood pressure of more than 185 mm Hg is a contraindication to thrombolytic treatment with intravenous alteplase in patients with acute ischaemic stroke, but the target systolic blood pressure for optimal outcome is uncertain. We assessed intensive blood pressure lowering compared with guideline-recommended blood pressure lowering in patients treated with alteplase for acute ischaemic stroke.

Methods We did an international, partial-factorial, open-label, blinded-endpoint trial of thrombolysis-eligible patients (age ≥18 years) with acute ischaemic stroke and systolic blood pressure 150 mm Hg or more, who were screened at 110 sites in 15 countries. Eligible patients were randomly assigned (1:1, by means of a central, web-based program) within 6 h of stroke onset to receive intensive (target systolic blood pressure 130–140 mm Hg within 1 h) or guideline (target systolic blood pressure <180 mm Hg) blood pressure lowering treatment over 72 h. The primary outcome was functional status at 90 days measured by shift in modified Rankin scale scores, analysed with unadjusted ordinal logistic regression. The key safety outcome was any intracranial haemorrhage. Primary and safety outcome assessments were done in a blinded manner. Analyses were done on intention-to-treat basis. This trial is registered with ClinicalTrials.gov, number NCT01422616.

Findings Between March 3, 2012, and April 30, 2018, 2227 patients were randomly allocated to treatment groups. After exclusion of 31 patients because of missing consent or mistaken or duplicate randomisation, 2196 alteplase-eligible patients with acute ischaemic stroke were included: 1081 in the intensive group and 1115 in the guideline group, with 1466 (67·4%) administered a standard dose among the 2175 actually given intravenous alteplase. Median time from stroke onset to randomisation was 3·3 h (IQR 2·6–4·1). Mean systolic blood pressure over 24 h was 144·3 mm Hg (SD 10·2) in the intensive group and 149·8 mm Hg (12·0) in the guideline group (p<0·0001). Primary outcome data were available for 1072 patients in the intensive group and 1108 in the guideline group. Functional status (mRS score distribution) at 90 days did not differ between groups (unadjusted odds ratio [OR] 1·01, 95% CI 0·87–1·17, p=0·8702). Fewer patients in the intensive group (160 [14·8%] of 1081) than in the guideline group (209 [18·7%] of 1115) had any intracranial haemorrhage (OR 0·75, 0·60–0·94, p=0·0137). The number of patients with any serious adverse event did not differ significantly between the intensive group (210 [19·4%] of 1081) and the guideline group (245 [22·0%] of 1115; OR 0·86, 0·70–1·05, p=0·1412). There was no evidence of an interaction of intensive blood pressure lowering with dose (low *vs* standard) of alteplase with regard to the primary outcome.

Interpretation Although intensive blood pressure lowering is safe, the observed reduction in intracranial haemorrhage did not lead to improved clinical outcome compared with guideline treatment. These results might not support a major shift towards this treatment being applied in those receiving alteplase for mild-to-moderate acute ischaemic stroke. Further research is required to define the underlying mechanisms of benefit and harm resulting from early intensive blood pressure lowering in this patient group.

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Research in context

Evidence before this study

We searched MEDLINE (from Jan 1, 1946) and Embase (from Jan 1, 1966) on Aug 20, 2018, with relevant text words and medical subject headings in any language that included "ischaemic stroke", "thrombolysis", and "blood pressure lowering". Studies were eligible for inclusion if they assessed the effect of blood pressure lowering treatment on the risk of clinical outcome. We identified no randomised trials or meta-analyses. Randomised trials of blood pressure lowering treatment in patients with acute ischaemic stroke without thrombolysis treatment suggest a benefit from very early treatment with glyceryl trinitrate patch within 6 h of the onset of symptoms, but no benefit at times thereafter for this or any type of blood pressure lowering treatment.

Added value of this study

ENCHANTED is the only randomised controlled trial of intensive versus guideline-recommended blood pressure lowering treatment during and for up to 72 h after intravenous thrombolysis for acute ischaemic stroke. The primary outcome of functional status (measured on the modified Rankin scale) at 90 days did not differ significantly between groups. The key safety outcome of any intracranial haemorrhage was significantly less frequent after intensive than after guideline-recommended blood pressure lowering treatment, and consistent reductions in adjudicated symptomatic intracerebral haemorrhage across a range of definitions were observed, albeit without statistical significance.

Implications of all the available evidence

Overall, these results will reassure clinicians that intensive blood pressure control is not associated with an increased risk of death or disability from adverse effects on the cerebral ischaemic penumbra in patients with acute ischaemic stroke receiving intravenous thrombolytic treatment. Such treatment could potentially reduce the risk of major intracranial haemorrhage, but further research is required to define the underlying mechanisms of benefit and harm resulting from early intensive blood pressure lowering in cases of hyperacute acute ischaemic stroke. Moreover, further trials with a greater difference in blood pressure between treatment groups are required to provide more definitive evidence to support the treatment in patients with more severe acute ischaemic stroke requiring thrombolysis or endovascular reperfusion therapy.

Introduction

Timely administration of intravenous thrombolytic treatment is the mainstay of hyperacute reperfusion treatment in patients with acute ischaemic stroke, even with the advent of mechanical thrombectomy for those with proximal large vessel occlusion.[1] The evidence strongly suggests that administration of intravenous alteplase (recombinant tissue plasminogen activator) within 4·5 h of acute ischaemic stroke onset results in a net benefit over harm from intracranial haemorrhage.[2,3] Ongoing research seeks to improve the efficacy and safety of mechanical and pharmacological reperfusion therapies in eligible patients with acute ischaemic stroke.

The alteplase dose-assessment arm of the Enhanced Control of Hypertension and Thrombolysis Stroke Study (ENCHANTED) was unable to clearly show non-inferiority of low-dose intravenous alteplase compared with the standard dose with respect to death and dependency at 90 days, despite significant reductions in early (7-day) mortality and symptomatic intracerebral haemorrhage.[4] However, controversy persists regarding control of peri-thrombolysis blood pressure, for which guidelines consistently contraindicate the use of alteplase in patients with systolic blood pressure of more than 185 mm Hg.[5] Two large registries[6,7] have reported a positive association between increasing systolic blood pressure and increased risk of symptomatic intracerebral haemorrhage, even below this threshold: frequency of symptomatic intracerebral haemorrhage was four times higher in patients with systolic blood pressure of more than 170 mm Hg than it was in those with systolic blood pressure of 141–150 mm Hg.[7] A U-shaped association for death and dependency is also evident, with the best outcomes associated with systolic blood pressure of 141–150 mm Hg. An ongoing concern, however, has been that rapid blood pressure reduction in the absence of reperfusion might worsen cerebral ischaemia due to hypoperfusion in failing collateral circulation into the ischaemic penumbra.[8]

The second, blood pressure control-assessment arm of the ENCHANTED trial was driven by uncertainty over whether any potential benefits related to a reduced risk of thrombolysis-related intracranial haemorrhage could be offset by worsened cerebral ischaemia associated with intensive blood pressure lowering. Herein, we report the results of the blood pressure control arm of the ENCHANTED trial, which tested the hypotheses that, following use of intravenous alteplase, a strategy of intensive blood pressure lowering (target systolic blood pressure 130–140 mm Hg) is superior to guideline-recommended blood pressure lowering (target systolic blood pressure <180 mm Hg) for improving functional recovery and reducing the risk of intracranial haemorrhage in patients with acute ischaemic stroke.

Methods

Study design and participants

ENCHANTED was an international, multicentre, prospective, randomised, open-label, blinded-endpoint trial with a 2×2 partial-factorial design, at 110 sites in 15 countries, to assess the effectiveness of low-dose versus standard-dose alteplase (previously published),[5]

and intensive versus guideline-recommended blood pressure control (described here). Details of the study design and rationale have been published elsewhere,[9] and the protocol is available in the appendix. The statistical analysis plan was submitted for publication before study unmasking.[10]

Adult patients (aged ≥18 years) with acute ischaemic stroke and systolic blood pressure 150 mm Hg or more were eligible if they fulfilled standard criteria for thrombolysis with intravenous alteplase, and if the treating clinician had uncertainty over the benefit and risk of the intensity of blood pressure control during and for up to 72 h (or hospital discharge or death, if this event occurred earlier) after thrombolytic treatment. Although there was no specified upper systolic blood pressure threshold, patients were required to comply with guidelines for the use of thrombolysis, which included having a systolic blood pressure of 185 mm Hg or lower before administration of intravenous alteplase. Participants were randomly assigned to a strategy of intensive blood pressure lowering (intensive group; target systolic blood pressure 130–140 mm Hg within 60 min of randomisation) or guideline-recommended blood pressure control (guideline group; target systolic blood pressure <180 mm Hg) after commencement of intravenous alteplase. On Nov 12, 2013, the protocol was amended as follows: systolic blood pressure target was reduced from 140–150 mm Hg to 130–140 mm Hg in the intensive group to enhance the systolic blood pressure difference between groups; time of randomisation to the blood pressure arm was increased from within 4·5 h to within 6 h of stroke onset to avoid trial-related procedures delaying the achievement of 1 h door-to-needle-time quality performance in the administration of intravenous alteplase as part of routine practice; time to achieve the target systolic blood pressure was increased from 60 min from the commencement of alteplase to 60 min from randomisation; to increase study power, the key secondary outcome was changed from whether intensive blood pressure lowering reduced symptomatic intracerebral haemorrhage to whether it reduced any intracranial haemorrhage; and sample size was reduced from 3300 to 2304 participants. Furthermore, a final protocol amendment on Feb 16, 2017, changed the primary outcome from a conventional binary assessment of poor clinical outcome (modified Rankin scale [mRS] scores of 3–6) to an ordinal shift analysis of the full range of category scores (0–6) of the mRS at 90 days to increase study power; and this change resulted in a further reduction in sample size to 2100 participants consequent upon this change in the primary outcome. Until the conclusion of the alteplase dose arm on Aug 17, 2015, participants could additionally be randomised to low-dose (0·6 mg/kg, maximum of 60 mg; 15% as bolus, 85% as infusion over 1 h) or standard-dose (0·9 mg/kg, maximum of 90 mg; 10% as bolus, 90% as infusion over 1 h) intravenous alteplase. Subsequently, the attending clinician investigator could choose the dose of intravenous alteplase to use according to their interpretation of the evidence.

Key exclusion criteria were that a patient was unlikely to benefit from thrombolysis (eg, had advanced dementia); had a very high likelihood of death within 24 h; had substantial comorbidity that would interfere with the outcome assessments or follow-up (known pre-stroke disability, with estimated scores 2–5 on the mRS); had a specific contraindication to alteplase or any of the blood pressure lowering drugs to be used; or was participating in another clinical trial of a pharmacological agent (see appendix for full inclusion and exclusion criteria).

The trial protocol was approved by appropriate regulatory and ethical authorities at participating centres. Written consent was obtained from each participant or from their approved surrogate for patients who were too unwell to comprehend the information.

Randomisation and masking

After confirmation of patient eligibility, 1:1 randomisation was done centrally via a password-protected, web-based program at The George Institute for Global Health (Sydney, Australia). A minimisation algorithm was used to achieve approximate balance in randomisation according to three key prognostic factors: site of recruitment, time from the onset of symptoms (<3 h or ≥3 h), and severity of neurological impairment according to the National Institutes of Health Stroke Scale (NIHSS) score (<10 points or ≥10 points). Final follow-up was done at 90 days, in person or by telephone, by trained and certified staff who were unaware of the randomised treatment assignment. Central adjudication of safety outcomes was also done by assessors unaware of treatment allocation or clinical details.

Procedures

We sought to assess a management strategy of blood pressure lowering to achieve and maintain intensive (130–140 mm Hg) and guideline-recommended (<180 mm Hg) systolic blood pressure targets. Therefore, local treatment protocols based on available intravenous (bolus and infusion), oral, and topical medications were used (outlined in the appendix). All patients were to be managed in an acute stroke unit, or alternative environment with appropriate staffing and monitoring, and to receive active care and best-practice management according to local guidelines. The use of endovascular thrombectomy, which increased in clinical practice during the course of the trial, was permitted.

Non-invasive blood pressure monitoring was done using an automated device applied to the non-hemiparetic arm (or right arm in situations of coma or tetraparesis), with the patient resting supine for at least 3 min in accordance with a standard protocol. Following thrombolysis, blood pressure measurements were recorded every 15 min for 1 h, hourly from hours 1 to 6, and 6-hourly from hours 6 to 24. Thereafter, blood

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See Online for appendix

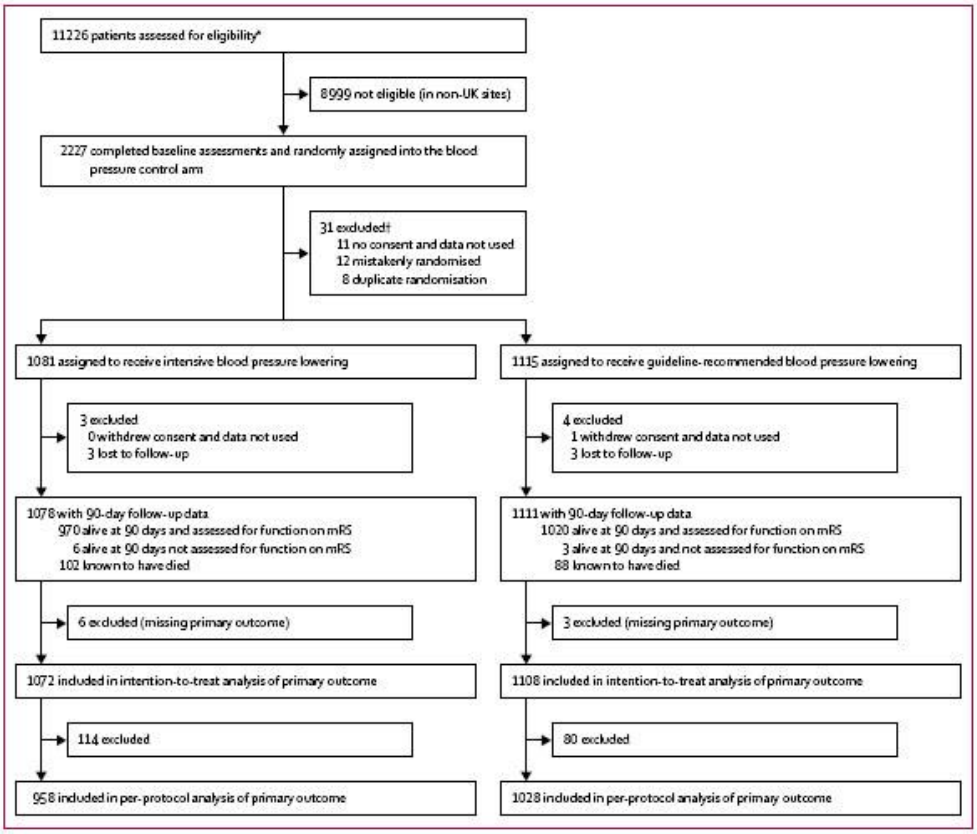

Figure 1: Trial profile
mRS=modified Rankin scale. *Screening logs not used at UK sites. †15 for intensive blood pressure lowering group, 8 for guideline-recommended blood pressure lowering group, and 8 for alteplase-dose arm (patients mistakenly randomised).

pressure was recorded twice daily for 1 week (or until hospital discharge or death, if earlier). Neurological status, measured on the NIHSS and Glasgow coma scale, was assessed at baseline, and 24 h and 72 h after start of alteplase treatment. Brain imaging (CT, MRI, or both) was done at baseline and at 24 h, and at additional timepoints if clinically indicated. Early cerebral ischaemia or infarction, and hyperdense artery sign, as identified by a local investigator, were recorded. Analyses were done centrally for diagnoses of categories of intracranial haemorrhage by expert assessors (appendix).

A detailed list of the assessment schedule is contained in the study protocol. In brief, screening logs with details of key reasons for excluding potentially eligible patients were maintained at all sites, except in the UK (where this activity is not required by the health authority). Sociodemographic and clinical details were obtained at randomisation. Follow-up data were collected at 24 h, 72 h, 7 days (or at hospital discharge if earlier), 28 days, and 90 days. Remote and on-site quality-control monitoring and data verification were done throughout the study (appendix).

Outcomes

The prespecified primary outcome, assessed at 90 days in the intention-to-treat population, was a shift in measures of functioning according to the full range of scores on the mRS,[11] a global, seven-level assessment of disability, in which scores of 0 or 1 indicate a favourable outcome without or with symptoms but no disability, scores of 2 to 5 indicate increasing levels of disability (and dependency), and a score of 6 indicates death. Other secondary efficacy outcomes were assessed by the conventional dichotomous analysis of mRS scores at 90 days: 2–6 (disability or death) versus less than 2, or 3–6 (major disability or death) versus less than 3. The following outcomes were also assessed: cause-specific mortality within 90 days; death or neurological deterioration (≥4 points decline in NIHSS) within 24 h and 72 h;

	Intensive blood pressure lowering group (n=1081)	Guideline-recommended blood pressure lowering group (n=1115)
Time from the onset of symptoms to randomisation, h	3·4 (2·5–4·1)	3·3 (2·6–4·1)
Sex		
Female	401/1081 (37·1%)	434/1115 (38·9%)
Male	680/1081 (62·9%)	681/1115 (61·1%)
Age, years	66·7 (12·4)	67·1 (12·0)
≥80	149/1081 (13·8%)	170/1115 (15·2%)
<80	932/1081 (86·2%)	945/1115 (84·8%)
Ethnicity		
Asian	795/1080 (73·6%)	823/1114 (73·9%)
Non-Asian	285/1080 (26·4%)	291/1114 (26·1%)
Clinical features		
Systolic blood pressure, mm Hg	165·4 (9·1)	165·2 (9·2)
Diastolic blood pressure, mm Hg	91·2 (11·6)	90·7 (11·3)
Heart rate, beats per minute	79·4 (14·6)	79·2 (15·0)
NIHSS score*	7 (4–12)	8 (4–12)
GCS score†	15 (14–15)	15 (14–15)
Medical history		
Hypertension	773/1078 (71·7%)	795/1114 (71·4%)
Currently treated hypertension	493/1078 (45·7%)	519/1114 (46·6%)
Previous stroke (ischaemic, haemorrhagic, or uncertain)	205/1081 (19·0%)	209/1115 (18·7%)
Coronary artery disease	154/1078 (14·3%)	155/1114 (13·9%)
Other heart disease (valvular or other)	42/1078 (3·9%)	52/1114 (4·7%)
Atrial fibrillation confirmed on electrocardiogram	140/1078 (13·0%)	172/1112 (15·5%)
Diabetes	230/1078 (21·3%)	266/1114 (23·9%)
Hypercholesterolaemia	120/1078 (11·1%)	129/1114 (11·6%)
Current smoker	218/1077 (20·2%)	226/1113 (20·3%)
Estimated premorbid function (mRS)		
No symptoms (score 0)	924/1078 (85·7%)	953/1113 (85·6%)
Symptoms without any disability (score 1)	154/1078 (14·3%)	160/1113 (14·4%)
(Table 1 continues in next column)		
(Continued from previous column)		
Medication at time of admission		
Warfarin anticoagulation	14/1078 (1·3%)	15/1114 (1·3%)
Aspirin or other antiplatelet agent	174/1078 (16·1%)	212/1114 (19·0%)
Statin or other lipid lowering agent	154/1078 (14·3%)	184/1114 (16·5%)
Brain imaging features		
CT scan used	1056/1078 (98·0%)	1096/1114 (98·4%)
MRI scan used	81/1078 (7·5%)	78/1114 (7·0%)
Visible early ischaemic changes	160/1078 (14·8%)	175/1114 (15·7%)
Visible cerebral infarction	176/1078 (16·3%)	167/1114 (15·0%)
CT or magnetic resonance angiogram showed proximal vessel occlusion	97/1076 (9·0%)	91/1113 (8·2%)
Final diagnosis‡		
Non-stroke mimic	16/1074 (1·5%)	17/1093 (1·6%)
Presumed stroke cause		
Large artery disease due to significant intracranial atheroma	387/1067 (36·3%)	416/1093 (38·1%)
Large artery disease due to significant extracranial atheroma	70/1067 (6·6%)	79/1093 (7·2%)
Small vessel disease	333/1067 (31·2%)	290/1093 (26·5%)
Cardioembolic	139/1067 (13·0%)	150/1093 (13·7%)
Dissection	4/1067 (0·4%)	3/1093 (0·3%)
Other or uncertain cause	118/1067 (11·1%)	138/1093 (12·6%)

Data are n (%), mean (SD), or median (IQR). NIHSS=National Institutes of Health Stroke Scale. GCS=Glasgow coma scale. mRS=modified Rankin scale. *Scores range from 0 to 42, with higher scores indicating more severe neurological deficit. †Scores range from 15 (normal) to 3 (deep coma). ‡Diagnosis according to the clinician's interpretation of clinical features and results of investigations at the time of separation from hospital.

Table 1: Baseline characteristics

primary cause of death; duration of initial hospitalisation in days; and health-related quality of life, as assessed on the EuroQoL group EQ-5D-3L,[12] according to an overall health utility score at 90 days.

The key safety outcome was any intracranial haemorrhage reported by investigators or after central adjudication of relevant brain imaging within 7 days after randomisation. This outcome included intracerebral haemorrhage, subarachnoid haemorrhage, and other forms of haemorrhage within the cranium identified on an adjudicated scan; any intracranial haemorrhage reported by an investigator with a description of the results of brain imaging without central verification; and any coding according to Medical Dictionary for Regulatory Activities (MedDRA) definitions of intracranial haemorrhage reported as a serious adverse event. Another safety outcome was the topography of intracerebral haemorrhage identified on centrally adjudicated brain images in association with a patient's symptoms (ie, symptomatic intracerebral haemorrhage, in which intracerebral haemorrhage was associated with substantial neurological deterioration or death). The key measure of symptomatic intracerebral haemorrhage was from the Safe Implementation of Thrombolysis in Stroke-Monitoring Study (SITS-MOST), defined as large or remote parenchymal intracerebral haemorrhage (type 2, defined as >30% of the infarcted area affected by haemorrhage with mass effect or extension outside the infarct) combined with neurological deterioration

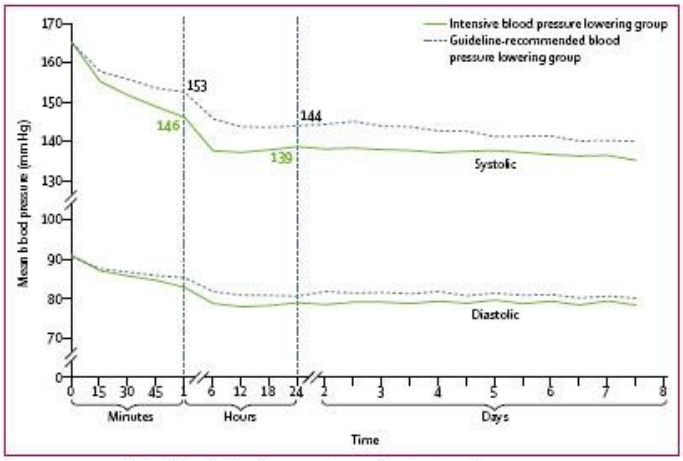

Figure 2: Mean systolic and diastolic blood pressure from randomisation to day 7
Blood pressure values are shown for intensive and guideline-recommended blood pressure lowering groups based on recordings at 15-min intervals for the first hour after randomisation (time 0), hourly from hours 1 to 6, 6-hourly until 24 h, and twice daily until day 7. Mean between-group difference in systolic blood pressure over 24 h was 5·5 mm Hg (95% CI 4·5–6·4).

(≥4 points on the NIHSS) or leading to death within 24–36 h.[6] Other criteria for symptomatic intracerebral haemorrhage that were used in other studies are outlined in the appendix. Other prespecified safety outcomes included all-cause and cause-specific serious adverse events (overall and by vital status) until trial completion, coded according to MedDRA definitions. Outcomes were assessed in both intention-to-treat and per-protocol populations. We also did post-hoc analyses on the between-group systolic blood pressure differences over the study period, a comparison of the characteristics of patients assigned to the guideline-recommended blood pressure management group according to receipt of any intravenous blood pressure lowering, and the effects of treatment on the NIHSS as a continuous measure.

Statistical analysis

Power calculations were based on the estimated treatment effects on a conventional binary assessment of poor outcome (mRS scores 3–6). Assuming poor outcomes of 43% in the intensive blood pressure lowering group and 50% in the guideline-recommended blood pressure lowering group, a sample size of 2304 (1152 per group) was estimated to provide more than 90% power (using a two-sided α=0·05) to detect a 14% relative reduction in poor outcome in the intensive group,[7] taking account of a 5% drop-out and potential negative interaction between low-dose alteplase and intensive blood pressure lowering. However, as the ordinal shift approach provides efficiency gains, a re-estimation of the sample size based on an ordinal analysis of mRS scores indicated that the estimated treatment effect could be detected with a sample size of 2100.[10] This sample size was also estimated to provide more than a 40% reduction in any intracranial haemorrhage associated with a 15 mm Hg difference in systolic blood pressure between randomised groups on the basis of Safe Implementation of Thrombolysis in Stroke-International Stroke Thrombolysis Register (SITS-ISTR) data.[7]

Statistical analyses were done on an intention-to-treat basis. We did shift analyses using ordinal logistic regression for the primary efficacy outcome, and dichotomous logistic regression analyses for all other outcomes. Treatment effects were presented as odds ratios (ORs) with 95% CIs. A priori,[10] the primary analysis for superiority of intensive versus guideline-recommended blood pressure lowering was unadjusted, but we also did prespecified sensitivity analyses of the treatment effects on all outcomes adjusted for minimisation and key prognostic covariates (age; sex; ethnicity; premorbid function [mRS scores 0 or 1]; premorbid use of antithrombotic agents [aspirin, other antiplatelet agent, or warfarin]; history of stroke, coronary artery disease, diabetes mellitus, and atrial fibrillation; and randomised alteplase dose), as well as a per-protocol analysis. Consistency of treatment effect across ten prespecified subgroups was assessed through tests for interaction, obtained from adding interaction terms to statistical models with main effects only. An independent data and safety monitoring committee monitored progress of the trial every 6 months. All tests were two-sided and the nominal level of α was 5%. No adjustment was made for multiplicity. SAS software version 9.3 was used for analyses.

This trial is registered with ClinicalTrials.gov, number NCT01422616, and is now closed at all participating sites.

Role of the funding source

The funders of the study had no role in the study design, data collection, data analysis, data interpretation or writing of the report. The corresponding author had full access to all data in the study and had overall responsibility for the decision to submit for publication.

Results

From March 3, 2012, to April 30, 2018, 2227 patients with acute ischaemic stroke were randomly allocated (figure 1, appendix). 31 patients were excluded because of missing consent, or mistaken or duplicate randomisation, leaving 2196 included in the intention-to-treat analysis: 1081 (49·2%) in the intensive group and 1115 (50·8%) in the guideline group. 925 (42·1%) participants were also enrolled in the alteplase dose arm of the trial (456 [20·8%] receiving low-dose alteplase and 469 [21·4%] standard-dose alteplase). Treatment groups were well balanced with respect to baseline demographic and clinical characteristics (table 1). The overall mean age was 66·9 years (SD 12·2) and 835 (38·0%) participants were female. 1618 (73·7%) patients were recruited in Asia (1428 [65·0%] in China). Median NIHSS score before

treatment was 7 (range 0–42, IQR 4–12). 1012 (46·2% of 2192 participants with available data) were receiving antihypertensive treatment at the time of enrolment, and mean systolic blood pressure before treatment was 165·3 mm Hg (SD 9·2). The median time from stroke onset to randomisation was 3·3 h (IQR 2·6–4·1). Only 42 (1·9%) patients received endovascular thrombectomy treatment.

Adherence to assigned treatment was high and did not differ between groups: 2175 (99·0%) patients received any intravenous alteplase, of whom 1466 (67·4%) received a standard dose (0·9 mg/kg body mass), including 469 (32·0%) who participated in the alteplase dose arm, and 997 (68·0%) who did not participate in the alteplase dose arm but whose actual administered dose was more than the 0·75 mg/kg cutoff dose (appendix). The median time from the initiation of treatment with intravenous alteplase to commencement of any intravenous blood pressure lowering treatment was 20 min (IQR 0–85) in the intensive group and 30 min (0–153) in the guideline group (p=0·0925). 2140 (97·4%) of the 2196 participants received blood pressure lowering treatment in accordance with the assigned protocol (appendix). In the intensive group, the proportions of patients administered any blood pressure lowering treatment (858 [80·1%] of 1071 *vs* 602 [54·3%] of 1108 with available data; p<0·0001) and administered intravenous blood pressure lowering drugs (671 [62·7%] of 1071 *vs* 391 [35·3%] of 1108; p<0·0001) during the first 24 h post-randomisation were significantly higher than those in the guideline group (appendix). Additionally, a greater proportion of patients in the intensive (772 [72·6%] of 1063) than in the guideline group (689 [63·2%] of 1091) received blood pressure lowering therapy over days 2–7 in hospital (p<0·0001; appendix). Mean systolic blood pressure levels were 146·2 mm Hg in the intensive group and 152·7 mm Hg in the guideline group (mean difference −6·4 mm Hg, 95% CI −7·9 to −5·0) at 1 h, and 138·8 mm Hg in the intensive group and 144·1 mm Hg in the guideline group (mean difference −5·3 mm Hg, −6·7 to −3·9) at 24 h; figure 2, appendix). Overall mean systolic blood pressure levels within 24 h were significantly lower in the intensive than in the guideline group (144·3 mm Hg [SD 10·2] *vs* 149·8 mm Hg [12·0], p<0·0001; appendix). Systolic blood pressure remained lower in the intensive than in the guideline group for the subsequent 6 days (figure 2, appendix). There were no significant differences in other clinical management over the 7-day post-randomisation period (appendix).

The primary outcome of mRS at 90 days was assessed in 2180 (99·3%) participants (1072 in the intensive group and 1108 in the guideline group), mostly by telephone (figure 1, appendix). The proportional odds assumption was tested and was not significant (p=0·6036). There was no significant difference (shift) in the 90-day mRS score distribution (table 2, figure 3). These results were consistent after adjustment for minimisation and key prognostic variables (table 2). No heterogeneity of treatment effect on primary outcome was found across prespecified subgroups (figure 4). In particular, alteplase

	Intensive blood pressure lowering group	Guideline-recommended blood pressure lowering group	Treatment effect	p value
Improvement in mRS according to category* at day 90	..	..	1·01 (0·87–1·17)†, 0·97 (0·83–1·13)†‡	0·8702†, 0·7171†‡
0	307/1072 (28·6%)	312/1108 (28·2%)	..	..
1	267/1072 (24·9%)	264/1108 (23·8%)	..	..
2	138/1072 (12·9%)	160/1108 (14·4%)	..	..
3	110/1072 (10·3%)	120/1108 (10·8%)	..	..
4	98/1072 (9·1%)	104/1108 (9·4%)	..	..
5	50/1072 (4·7%)	60/1108 (5·4%)	..	..
6	102/1072 (9·5%)	88/1108 (7·9%)	..	..
Death or disability (mRS score 2–6) within 90 days				
Intention-to-treat analysis				
Unadjusted	498/1072 (46·5%)	532/1108 (48·0%)	0·94 (0·79–1·11)	0·4660
Adjusted	498/1072 (46·5%)	531/1106 (48·0%)	0·94 (0·78–1·14)‡	0·5508
Per-protocol analysis				
Unadjusted	451/958 (47·1%)	499/1028 (48·5%)	0·94 (0·79–1·12)	0·5141
Adjusted	451/958 (47·1%)	498/1026 (48·5%)	0·96 (0·79–1·16)‡	0·6595
Death or major disability (mRS score 3–6) within 90 days				
Unadjusted	360/1072 (33·6%)	372/1108 (33·6%)	1·00 (0·84–1·20)	0·9968
Adjusted	360/1072 (33·6%)	371/1106 (33·5%)	1·01 (0·83–1·24)‡	0·9090
Death or neurological deterioration§				
In first 24 h	110/1081 (10·2%)	108/1115 (9·7%)	1·06 (0·80–1·40)	0·7013
In first 72 h	146/1081 (13·5%)	139/1115 (12·5%)	1·10 (0·85–1·41)	0·4687
Death within 90 days				
Unadjusted	102/1081 (9·4%)	88/1115 (7·9%)	1·22 (0·90–1·64)	0·1989
Adjusted	102/1078 (9·5%)	88/1113 (7·9%)	1·18 (0·86–1·64)‡	0·3077

Frequency data are n/N (%). Treatment effect is presented as odds ratio (95% CI) of intensive versus guideline-recommended blood pressure lowering, analysed by unadjusted binary logistic regression unless stated otherwise. mRS=modified Rankin scale. *The primary outcome was an assessment of scores across all seven levels of the mRS (ranging from 0 [no symptoms] to 6 [death]), done using a shift analysis of the ordinal data. †Calculated with ordinal logistic regression. ‡Adjusted for minimisation and key prognostic covariates (age; sex; ethnicity; pre-morbid function [mRS scores 0 or 1]; pre-morbid use of antithrombotic agents [aspirin, other antiplatelet agent, or warfarin]; history of stroke, coronary artery disease, diabetes, and atrial fibrillation; and randomised alteplase dose). §Defined by an increase between baseline and 24 h of ≥4 on the National Institutes of Health Stroke Scale or a decline of ≥2 on the Glasgow coma scale.

Table 2: Efficacy outcomes

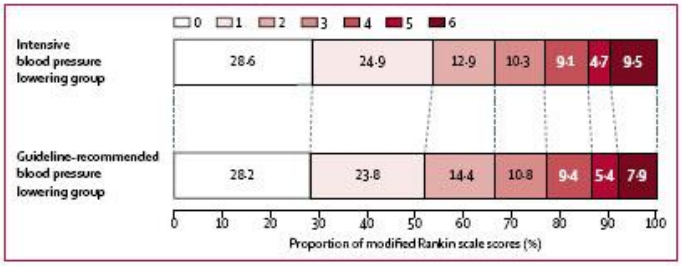

Figure 3: Distribution of modified Rankin scale scores at 90 days by treatment group
Raw distribution of scores is shown. Scores range from 0 to 6: 0=no symptoms, 1=symptoms without clinically significant disability, 2=slight disability, 3=moderate disability, 4=moderately severe disability, 5=severe disability, and 6=death.

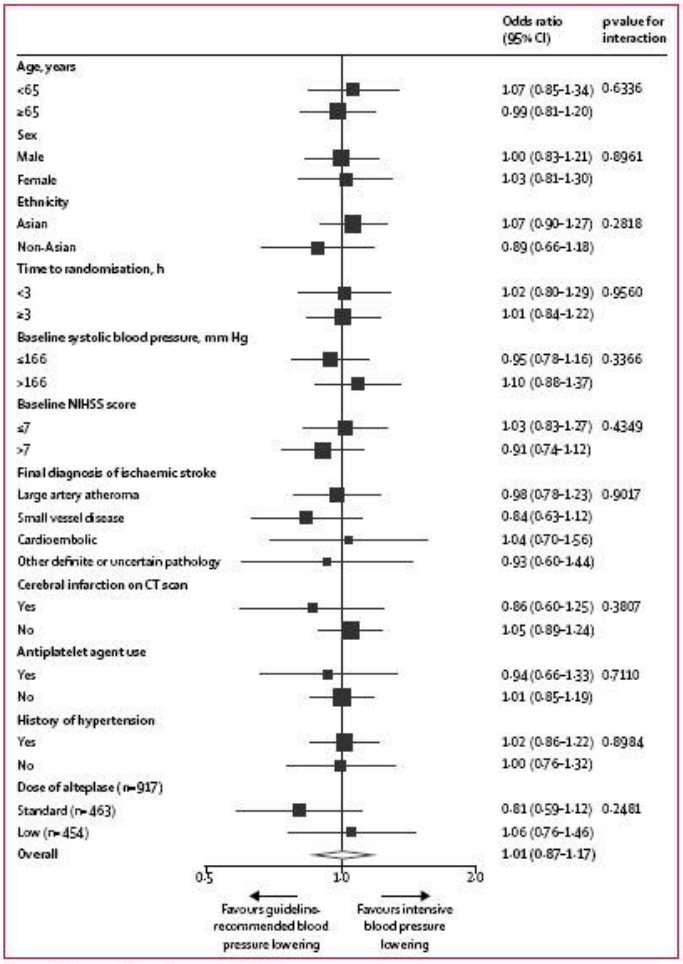

Figure 4: Primary outcome by prespecified subgroups

The primary efficacy outcome was shift in the modified Rankin scale score distribution (range 0 [no symptoms] to 6 [death]) at 90 days. For subcategories, black squares represent point estimates (with the area of the square proportional to the number of events), and horizontal lines represent 95% CIs. Scores on the NIHSS range from 0 to 42, with higher scores indicating more severe neurological deficits. For systolic blood pressure and NIHSS score, subgroups were dichotomised by median value. Dose of alteplase is divided into low (0·6 mg/kg; 15% as bolus, 85% as infusion over 1 h) or standard (0·9 mg/kg; 10% as bolus, 90% as infusion over 1 h). The marginal effect for factorial design (n=917 participants) for intensive versus guideline blood pressure lowering was odds ratio 0·92 (95% CI 0·73–1·16, p=0·4901). NIHSS=National Institutes of Health Stroke Scale.

dose and intensity of blood pressure lowering showed no significant interaction in the 917 patients recruited into both randomisation arms (p=0·2481; figure 4, appendix).

No significant differences were seen in the odds of death or disability at 90 days, whether defined by an mRS score of 2–6 or 3–6 (table 2). The unadjusted and adjusted per-protocol analyses were also consistent in showing no significant differences between the groups in treatment effect for overall functional outcome on mRS (table 2). Death or substantial neurological deterioration within 24 h and death within 90 days occurred in similar proportions of patients in the intensive and the guideline groups (table 2). No significant differences were evident in any of the other secondary clinical outcomes, including the primary cause of death, duration of initial hospitalisation, and health-related quality of life as an overall health utility score (appendix). Post-hoc analysis showed no heterogeneity in the treatment effect on the primary outcome according to quartiles of baseline NIHSS scores (appendix).

Assessment of the key safety outcome, any intracranial haemorrhage, was derived from adjudicated brain scans in 323 (87·5%) patients and other reports in 164 (51·0%) patients (appendix). This outcome was significantly less frequent in the intensive group than in the guideline group (table 3). The absolute difference was 3·9% (95% CI 0·8–7·1; p=0·0141) and the number need to treat to benefit was 25. MedDRA-coded clinician-reported intracranial haemorrhage as a serious adverse event was also significantly less frequent in the intensive group than in the guideline group (table 3). Furthermore, the intensive group had lower frequencies of adjudicated symptomatic intracerebral haemorrhage across a broad range of definitions than did the guideline group (table 3), and adjudicated large parenchymal intracerebral haemorrhage was less frequent in the intensive group (56 [5·2%]) than in the guideline group (80 [7·2%]; OR 0·71, 0·50–1·01, p=0·0535; appendix), although these differences were not significant.

The overall number of serious adverse events was similar in the intensive group (277) and the guideline group (334), and the number of patients with any serious adverse event did not differ significantly between the groups (210 [19·4%] of 1081 *vs* 245 [22·0%] of 1115; OR 0·86, 0·70–1·05, p=0·1412; appendix). However, compared with the guideline-recommended strategy, intensive blood pressure lowering was associated with significantly lower frequencies of intracranial haemorrhage (66 [6·1%] *vs* 105 [9·4%]; OR 0·63, 0·45–0·86, p=0·0040) and intracerebral haemorrhage (59 [5·5%] *vs* 100 [9·0%]; OR 0·59, 0·42–0·82, p=0·0017) reported as serious adverse events, and these events were predominantly non-fatal (appendix). The overall frequency of serious adverse events that the clinician attributed to intensive blood pressure lowering was less than 2·0% (appendix).

A post-hoc analysis of blood pressure management over the course of the study showed that systolic blood pressure difference between the two groups tended to decline over time. Mean systolic blood pressure levels at 1 h were 145 mm Hg in the intensive and 153 mm Hg in the guideline group (mean difference –8·2 mm Hg, 95% CI –10·4 to –6·0) before the end of the alteplase dose arm of the trial (Aug 17, 2015), and 148 mm Hg in the intensive and 153 mm Hg in the guideline group after the end of the alteplase dose arm, with a significantly decreased mean

difference (–5·1 mm Hg, –6·7 to –3·2, p=0·0352; appendix). Similarly, the mean 1 h systolic blood pressure difference significantly decreased from –9·9 mm Hg (–16·9 to –2·9) to –4·2 mm Hg (–10·7 to 2·3) between the first and last years of the study (appendix).

Post hoc, the clinical characteristics of patients in the guideline group were reclassified according to use of intravenous blood pressure lowering treatment. Compared with patients who did not receive any blood pressure lowering treatment in the first 24 h post-randomisation, among the 602 patients who did receive such treatment were significantly higher proportions of non-Asian patients, patients with a history of hypertension, coronary artery disease, and atrial fibrillation, and patients with evidence of proximal clot occlusion on the initial CT scan; higher initial systolic blood pressure and neurological impairment; and fewer patients with small vessel disease on final diagnosis (appendix). All efficacy and safety outcomes were significantly worse for treated than for non-treated patients allocated to the guideline group in adjusted analyses (appendix).

	Intensive blood pressure lowering group	Guideline-recommended blood pressure lowering group	Treatment effect	p value
Any intracranial haemorrhage*	160/1081 (14·8%)	209/1115 (18·7%)	0·75 (0·60–0·94)	0·0137
Any intracranial haemorrhage reported as a serious adverse event	59/1081 (5·5%)	100/1115 (9·0%)	0·59 (0·42–0·82)	0·0017
Major intracerebral haemorrhage based on central adjudication of brain imaging				
Symptomatic intracerebral haemorrhage, SITS-MOST criteria†	14/1081 (1·3%)	22/1115 (2·0%)	0·65 (0·33–1·28)	0·2143
Symptomatic intracerebral haemorrhage, NINDS criteria‡	70/1081 (6·5%)	84/1115 (7·5%)	0·85 (0·61–1·18)	0·3321
Symptomatic intracerebral haemorrhage, ECASS2 criteria§	46/1081 (4·3%)	57/1115 (5·1%)	0·82 (0·55–1·23)	0·3431
Symptomatic intracerebral haemorrhage, ECASS3 criteria¶	21/1081 (1·9%)	30/1115 (2·7%)	0·72 (0·41–1·26)	0·2467
Symptomatic intracerebral haemorrhage, IST-3 criteria	24/1081 (2·2%)	37/1115 (3·3%)	0·66 (0·39–1·11)	0·1198
Large parenchymal intracerebral haemorrhage‖	56/1081 (5·2%)	80/1115 (7·2%)	0·71 (0·50–1·01)	0·0535
Any intracerebral haemorrhage on brain imaging within 7 days	143/1081 (13·2%)	180/1115 (16·1%)	0·79 (0·62–1·00)	0·0542
Fatal intracerebral haemorrhage within 7 days	5/1081 (0·5%)	14/1115 (1·3%)	0·37 (0·13–1·02)	0·0541

Frequency data are n/N (%). Treatment effect is presented as odds ratio (95% CI) of intensive versus guideline-recommended blood pressure lowering, analysed by unadjusted binary logistic regression. Intracranial haemorrhage includes intracerebral haemorrhage, subarachnoid haemorrhage, and subdural and extradural haemorrhage. SITS-MOST=Safe Implementation of Thrombolysis in Stroke-Monitoring Study. NINDS=National Institutes of Neurological Diseases and Stroke. ECASS=European Cooperative Acute Stroke Study. IST=International Stroke Trial. NIHSS=National Institutes of Health Stroke Scale. *Any reported intracranial haemorrhage noted on a local brain imaging report within 7 days after randomisation, any haemorrhage noted on a centrally adjudicated scan, and any intracranial haemorrhage reported by a clinician as a serious adverse event. †Large or remote parenchymal intracerebral haemorrhage (type 2, defined as >30% of the infarcted area affected by haemorrhage with mass effect or extension outside the infarct) combined with neurological deterioration (≥4 points on the NIHSS) or leading to death within 24–36 h. ‡Any intracerebral haemorrhage associated with neurological deterioration (≥1 point change in NIHSS score) from baseline, or death within 24–36 h. §Any intracerebral haemorrhage with neurological deterioration (≥4 points on the NIHSS) from baseline, or death within 24–36 h. ¶Any intracerebral haemorrhage with neurological deterioration (≥4 points increase on the NIHSS) from baseline, or death within 36 h. Either significant intracerebral haemorrhage (local or distant from the cerebral infarct) or significant haemorrhagic transformation of a cerebral infarct on brain imaging with clinically significant deterioration or death within the first 7 days of treatment. ‖Any type 2 parenchymal haematoma of intracerebral haemorrhage.

Table 3: Safety outcomes at day 90

Discussion

Our trial was driven by uncertainty over whether any benefit of intensive blood pressure lowering in terms of improving outcome in patients with acute ischaemic stroke, gained largely from a reduced risk of thrombolysis-related intracerebral haemorrhage, could be offset by the harm of promoting cerebral ischaemia. The main finding was that, in thrombolysis-treated patients with acute ischaemic stroke of predominantly mild-to-moderate severity, a strategy of intensive blood pressure lowering (target systolic blood pressure 130–140 mm Hg within 1 h) compared with current guideline-recommended blood pressure management (target <180 mm Hg) after intravenous alteplase therapy was not associated with a significant difference in functional recovery, as assessed by a shift in the distribution of mRS scores at 90 days. This result was consistent in sensitivity and per-protocol analyses, and across key prespecified subgroups. However, intensive blood pressure lowering was associated with a significant reduction in the incidence of intracranial haemorrhage, as well as slight (non-significant) reductions in major intracerebral haemorrhage, consistent across different measures.

The ENCHANTED trial adds important new information on the role of early intensive blood pressure lowering in the context of thrombolysed patients with acute ischaemic stroke, but it also highlights some of the challenges of doing an open trial in a critical illness with temporal change in the level of equipoise. Although we recruited to our target sample size and achieved a high rate of follow-up over 90 days, the average systolic blood pressure difference of 6 mm Hg between randomised groups was much smaller than the 15 mm Hg envisaged, and decreased as the trial progressed. In part, this finding reflected a shift in clinician behaviour towards targeting lower systolic blood pressure in the guideline group than is recommended in guidelines derived from the protocol of the National Institutes of Neurological Diseases and Stroke (NINDS) recombinant tissue plasminogen activator trial in acute ischaemic stroke.[13] It also relates to complexities in the titration of systolic blood pressure to the target according to the study protocol for patients in the intensive group: this target might have been considered too low for some clinicians, or reflected difficulties of aggressive blood pressure lowering in acute ischaemic stroke.

Systolic blood pressure is an important prognostic factor after acute stroke, with a systolic blood pressure target of 140–150 mm Hg being associated with best outcome in several observational studies.[14,15] To date, randomised evaluations of blood pressure lowering treatment in acute ischaemic stroke with a broad time window from the onset of symptoms and modest systolic blood pressure reductions have been neutral.[16] By

contrast, post-hoc analysis of the pivotal NINDS trial showed that the use of blood pressure lowering therapy after randomisation in hypertensive patients in the recombinant tissue plasminogen activator group was associated with less favourable outcome compared with that of patients who did not receive any such treatment.[15] However, blood pressure elevations are higher in patients who are less likely to reperfuse, have bigger strokes, and are thus more likely to get blood pressure lowering treatment. Conversely, post-hoc analysis from the more recent Multicenter Randomized Clinical Trial of Endovascular Treatment for Acute Ischemic Stroke in the Netherlands (MR CLEAN), specifically in patients with large vessel occlusion, showed a U-shaped relationship between baseline systolic blood pressure and outcome, with a systolic blood pressure nadir of 120 mm Hg being associated with best outcome.[17]

Many clinicians are concerned that rapid blood pressure reductions in the absence of mechanical or pharmacological reperfusion might worsen cerebral ischaemia from potential hypoperfusion with compromised autoregulation and collateral flow.[8] In our trial, any benefit from intensive blood pressure reduction on outcome due to reduction in intracranial haemorrhage might have been offset by hypoperfusion of the ischaemic penumbra. However, we observed no significant heterogeneity of treatment effect in subgroups where large vessel occlusion might be anticipated, including acute ischaemic stroke subtypes classified on the basis of clinician-diagnosis of large vessel disease, cardioemboli, or lacunar acute ischaemic stroke, and in post-hoc analysis of stroke severity based on quartiles of increasing NIHSS score. Since CT or magnetic resonance angiography was not mandated in this pragmatic study, artery status was not assessed in most patients and large vessel occlusion was only confirmed in 97 patients in the intensive group on CT or magnetic resonance angiography. Thus, further studies of intensive blood pressure lowering in the context of mechanical and pharmacological reperfusion therapy in cases of proven large vessel occlusion are required.

In the ENCHANTED trial, we also assessed the potential benefit of intensive blood pressure control in terms of the incidence of intracranial haemorrhage. From the SITS-ISTR of 11080 patients, Ahmed and colleagues' reported a linear association between systolic blood pressure and symptomatic intracerebral haemorrhage up to 24 h after thrombolysis. Similarly, in a post-hoc analysis of the third International Stroke Trial (IST-3), Berge and colleagues[18] reported an association between each 10 mm Hg higher baseline systolic blood pressure and risk of symptomatic intracerebral haemorrhage, with large systolic blood pressure declines over 24 h significantly associated with decreased risk of symptomatic intracerebral haemorrhage. As the only randomised trial of intensive blood pressure reduction in thrombolysis-treated acute ischaemic stroke patients, ENCHANTED suggests that there are benefits in lowering the risk of intracranial haemorrhage, despite no observed statistically significant decrease in adjudicated symptomatic intracerebral haemorrhage. This finding might reflect variable benefit of intensive blood pressure reduction on petechial, alteplase-associated, intracerebral haemorrhage in a hypertensive population with evidence of brain vessel fragility compared with large, space-occupying, alteplase-associated, parenchymal intracerebral haemorrhage, as previously suggested by Butcher and colleagues.[19] However, as ENCHANTED recruited mainly patients with acute ischaemic stroke of mild-to-moderate severity, the study was under-powered to assess the effects of treatment on symptomatic intracerebral haemorrhage, for which the frequencies of death and major neurological deterioration were low. Even so, the lower incidence of symptomatic intracerebral haemorrhage was consistent across all classifications in the intensive group versus the guideline group, and there were non-significant reductions in both petechial (haemorrhagic infarction 1 and 2) and space-occupying (parenchymal haemorrhage 1 and 2) intracerebral haemorrhage, and borderline significant reduction in any parenchymal haemorrhage, in adjudicated brain images. Finally, it is important to note that the ENCHANTED trial excluded patients with systolic blood pressure of more than 185 mm Hg, in keeping with the licensed indication for the use of intravenous alteplase, and thus no comment can be made with respect to the risk of intracranial haemorrhage or the benefit of blood pressure reduction in severely hypertensive patients. However, others have reported that such protocol violations are associated with significantly more frequent symptomatic intracerebral haemorrhage.[20]

The key strengths of this randomised controlled trial were its large size and international recruitment, which enhance the generalisability of the results and the possibility of influencing clinical practice worldwide. We used robust methodologies to ensure masking during assessment of the key efficacy measure (through central coordination of mRS follow-up by staff unaware of treatment allocation) and of the safety outcomes (with central adjudication of intracranial haemorrhage by assessors masked to clinical details and group allocation). Nonetheless, the study had several potential limitations.

First, the trial involved patients with acute ischaemic stroke of predominantly mild-to-moderate severity, with a median NIHSS score of 7, in contrast to previous trial and registry data of patients with acute ischaemic stroke with median NIHSS scores of 12 and 13, respectively.[2,3] However, with increasing use of intravenous thrombolysis, an NIHSS score of 7 is more reflective of the usual treated acute ischaemic stroke population, including those in clinical trials. For example, in a comparison of tenecteplase with alteplase, published in 2017, the median NIHSS was 4.[21] Even so, our results are potentially influenced by selection bias: clinicians might have excluded cases of severe stroke with high perceived risks from intensive blood pressure lowering treatment, but the effects of

intravenous alteplase are modest in mild acute ischaemic stroke. Second, there might be concerns about the generalisability of the trial results to all populations because nearly three-quarters of patients in the sample were Asian. We acknowledge reduced statistical power in the subgroup analyses; however, importantly, there was no heterogeneity of treatment effect by ethnicity, even though the high prevalence of intracranial atherosclerosis (and related intracranial stenosis) and of cerebral small vessel disease present in Asian populations might have increased the risks of hypoperfusion related to intensive blood pressure control.[22] In addition, the increased prevalence of hypertension and associated small vessel disease in Asian patients could have increased the risk of symptomatic intracerebral haemorrhage.[23] Finally, the smaller-than-anticipated systolic blood pressure difference between groups probably resulted in the trial being underpowered. In part, this reduced difference might be attributed to a natural fall in systolic blood pressure following recanalisation and reperfusion in both groups, but probably also reflected the effect of the high proportion (54·3%) of participants in the guideline group who received some form of blood pressure lowering therapy, and 35·3% who received any intravenous therapy in the first 24 h; and these patients had worse outcomes than those who did not receive treatment. The use of post-randomisation intravenous blood pressure lowering agents might reflect increased familiarity with local blood pressure lowering protocols in stroke units since the publication and international guideline adoption of the results of the main Intensive Blood Pressure Reduction in Acute Cerebral Haemorrhage Trial (INTERACT2), albeit in intracerebral haemorrhage patients.[24] Although most participants in the intensive group of our trial had blood pressure lowering treatment initiated soon after administration of intravenous alteplase, when the risk of reperfusion-related intracerebral haemorrhage is greatest, uncertainty remains over the most appropriate timing, approach, and agent(s) for blood pressure lowering, pre-thrombolysis and post-thrombolysis.

Intensive blood pressure lowering during and for up to 72 h after intravenous thrombolysis in predominantly Asian patients with acute ischaemic stroke of mild-to-moderate severity did not improve functional outcome at 90 days compared with that of patients who received guideline-recommended blood pressure management. Overall, the results indicate that intensive blood pressure lowering is safe in this patient group, with significantly decreased incidence of intracranial haemorrhage compared with that of the guideline group, and consistency in the reduced frequency of major intracerebral haemorrhage. However, these results might not support a major shift in clinical practice towards more intensive blood pressure lowering in those receiving thrombolysis for acute ischaemic stroke of mild-to-moderate severity. Because the observed reduction in intracerebral haemorrhage did not improve clinical outcome, further research is required to understand the underlying mechanisms of benefit and harm resulting from early intensive blood pressure lowering in patients with hyperacute acute ischaemic stroke.

Contributors

CSA, JC, RIL, TGR, and YH conceived the trial. CSA was the chief investigator. CSA, RIL, XC, JC, TGR, and ACD were responsible for the day-to-day running of the trial. RIL led the adjudication of neuroimaging. QL did the statistical analysis with supervision from LB. TGR, CSA, JC, and YH wrote the first draft of the manuscript; all authors revised this draft. All authors read and approved the final version.

Declaration of interests

CSA has received grants from the National Health and Medical Research Council (NHMRC) of Australia and Takeda China, honoraria for advisory board activities for Boehringer Ingelheim and Amgen, and speaker fees from Takeda. RIL and MWP have received research grants from the NHMRC of Australia. HA has received lecture fees from Bayer, Daiichi-Sankyo, Fukuda Denshi, Takeda and Teijin, and personal fees for consultancy to Kyowa-Kirin. PMB has received honoraria for advisory board activities from DiaMedica, Moleac, Nestlé, Phagenesis and ReNeuron. JPB has received grants from the National Institute of Neurological Diseases and Stroke, and Genentech. AMD has received speaker fees from Medtronic. PML has received research grants from Bayer, Boehringer Ingelheim, Conicyt, The George Institute for Global Health, and Clinica Alemana. CL has received research grants from NHMRC and honoraria from Boehringer Ingelheim. SOM has received speaker fees from Boehringer Ingelheim, Pfizer, Bayer, and Medtronic. VVO has received research grants from Clinica Alemana de Santiago, The George Institute for Global Health, Boehringer Ingelheim, Lundbeck Chile, and Conicyt. GAD has received advisory committee and speaker fees from Allergan, Amgen, Boehringer Ingelheim, Moleac, and Servier. OMP-N has received speaker fees from Boehringer Ingelheim, Pfizer, and Medtronic. SR has received travel support from Bayer. SS has worked as a medical expert for Bayer, Japan, from the end of the study. MW has received personal fees for consultancy to Amgen. JC has received research grants from NHMRC and Idorsia. TGR and JMW have received research grants from the UK Stroke Association. YH, XC, GC, QL, LB, CD, ACD, T-HL, JDP, VKS, FS, LS, NHT, J-GW, and XW declare no competing interests.

Data sharing statement

Individual, de-identified participant data used in these analyses will be shared by request from any qualified investigator following approval of a protocol and signed data access agreement via the Research Office of The George Institute for Global Health, Australia.

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References

1 Goyal M, Menon BK, van Zwam WH, et al. Endovascular thrombectomy after large-vessel ischaemic stroke: a meta-analysis of individual patient data from five randomised trials. *Lancet* 2016; 387: 1723–31.

2 Emberson J, Lees KR, Lyden P, et al. Effect of treatment delay, age, and stroke severity on the effects of intravenous thrombolysis with alteplase for acute ischaemic stroke: a meta-analysis of individual patient data from randomised trials. *Lancet* 2014; 384: 1929–35.

3 Wardlaw JM, Murray V, Berge E, et al. Recombinant tissue plasminogen activator for acute ischaemic stroke: an updated systematic review and meta-analysis. *Lancet* 2012; 379: 2364–72.

4 Anderson CS, Robinson T, Lindley RI, et al. Low-dose versus standard-dose intravenous alteplase in acute ischemic stroke. *N Engl J Med* 2016; 374: 2313–23.

5 Intercollegiate Stroke Working Party. National clinical guideline for stroke, 5th edn. London: Royal College of Physicians, 2016.

6 Wahlgren N, Ahmed N, Eriksson N, et al. Multivariable analysis of outcome predictors and adjustment of main outcome results to baseline data profile in randomised controlled trials: safe implementation of thrombolysis in stroke-monitoring study (SITS-MOST). *Stroke* 2008; 39: 3316–22.

7 Ahmed N, Wahlgren N, Brainin M, et al. Relationship of blood pressure, antihypertensive therapy, and outcome in ischemic stroke treated with intravenous thrombolysis: retrospective analysis from safe implementation of thrombolysis in stroke-international stroke thrombolysis registry (SITS-ISTR). *Stroke* 2009; 40: 2442–49.

8 Aries MJ, Elting JW, De Keyser J, Kremer BP, Vroomen PC. Cerebral autoregulation in stroke: a review of transcranial Doppler studies. *Stroke* 2010; 41: 2697–704.

9 Huang Y, Sharma VK, Robinson T, et al. Rationale, design, and progress of the enhanced control of hypertension and thrombolysis stroke study (ENCHANTED) trial: an international multicenter 2× 2 quasi-factorial randomized controlled trial of low- vs. standard-dose rt-PA and early intensive vs. guideline-recommended blood pressure lowering in patients with acute ischemic stroke eligible for thrombolysis treatment. *Int J Stroke* 2015; 10: 778–88.

10 Anderson C, Woodward M, Arima H, et al. Statistical analysis plan for evaluating different intensities of blood pressure control in the enhanced control of hypertension and thrombolysis stroke study (ENCHANTED). *Int J Stroke* 2018; published online Oct 9. DOI:10.1177/1747493018806170.

11 Quinn TJ, Dawson J, Walters MR, Lees KR. Reliability of the modified Rankin scale: a systematic review. *Stroke* 2009; 40: 3393–95.

12 Rabin R, de Charro F. EQ-5D: a measure of health status from the EuroQol Group. *Ann Med* 2001; 33: 337–43.

13 Brott T, Lu M, Kothari R, et al. Hypertension and its treatment in the NINDS rt-PA stroke trial. *Stroke* 1998; 29: 1504–09.

14 Geeganage CM, Bath PM. Relationship between therapeutic changes in blood pressure and outcomes in acute stroke: a metaregression. *Hypertension* 2009; 54: 775–81.

15 Tikhonoff V, Zhang H, Richart T, Staessen JA. Blood pressure as a prognostic factor after acute stroke. *Lancet Neurol* 2009; 8: 938–48.

16 Lee M, Ovbiagele B, Hong KS, et al. Effect of blood pressure lowering in early ischemic stroke: meta-analysis. *Stroke* 2015; 46: 1863–89.

17 Mulder M, Ergezen S, Lingsma H, et al. Baseline blood pressure effect on the benefit and safety of intra-arterial treatment in MR CLEAN (multicenter randomized clinical trial of endovascular treatment of acute ischemic stroke in the Netherlands). *Stroke* 2017; 48: 1869–76.

18 Berge E, Cohen G, Lindley RI, et al. Effects of blood pressure and blood pressure-lowering treatment during the first 24 hours among patients in the third international stroke trial of thrombolytic treatment for acute ischemic stroke. *Stroke* 2015; 46: 3362–69.

19 Butcher K, Christensen S, Parsons M, et al. Postthrombolysis blood pressure elevation is associated with hemorrhagic transformation. *Stroke* 2010; 41: 72–77.

20 Waltimo T, Haapaniemi E, Surakka I, et al. Post-thrombolytic blood pressure and symptomatic intracerebral hemorrhage. *Eur J Neurol* 2016; 23: 1757–62.

21 Logallo N, Novotny V, Assmus J, et al. Tenecteplase versus alteplase for management of acute ischaemic stroke (NOR-TEST): a phase 3, randomised, open-label, blinded endpoint trial. *Lancet Neurol* 2017; 16: 781–88.

22 Liao X, Wang Y, Pan Y, et al. Standard-dose intravenous tissue-type plasminogen activator for stroke is better than low doses. *Stroke* 2014; 45: 2354–58.

23 IST3 collaborative group. Association between brain imaging signs, early and late outcomes, and response to intravenous alteplase after acute ischaemic stroke in the third international stroke trial (IST-3). *Lancet Neurol* 2015; 14: 485–96.

24 Anderson CS, Heeley E, Huang Y, et al. Rapid blood-pressure lowering in patients with acute intracerebral hemorrhage. *N Engl J Med* 2013; 368: 2355–65.

Appendix C. The ENCHANTED image interpretation form for brain NCCT and MRI

ENCHANTED RTPA ARM BRAIN IMAGING READING FORMS

Enchanted

Enhanced Control of Hypertension and Thrombolysis Stroke Study

Image interpretation form – Adapted from the Third International Stroke Trial (IST-3)

CT ☐ Q.1-6,8-11,13,14,16,18-21 MRI ☐ Q.1-5,7-10,12,13,15,17-20,22-24

1.SCAN ID:

2. DATE OF READING: 3.DATE OF SCAN:

4.SCAN QUALITY: Good ☐ Moderate ☐ Poor ☐ 5.Comment on quality: ☐

6.TYPE OF SCAN (CT): CT Plain: ☐ CT Perfusion: ☐ CTA: ☐

7.TYPE OF SCAN (MRI): T2 and axial FLAIR: (Q.12a, 17a) ☐ T2 and coronal FLAIR: (Q.12b,17b) ☐ Diffusion Weighted Imaging: ☐

SWI/GRE/T2*: (Q.23, 24) ☐ MR Perfusion: ☐ MRA: ☐

Please tick Yes or No. Please do not leave blanks. Thank you.

		Y	N	
8a.	Are all the scan sequences completely normal? (That is, no acute or chronic ischemic changes or vascular changes on any of the available images.)	☐	☐	*If YES go to Q.18*
8b.	Are there any old vascular lesions?	☐	☐	

8c. Classify old vascular lesion(s):

i) old cortical infarct(s) ☐

ii) old striatocapsular infarct(s) ☐

iii) old borderzone infarct(s) ☐

iv) old lacunar infarct(s) ☐

v) old brainstem/cerebellar infarct(s) ☐

vi) probable old haemorrhage ☐

9. **Ischaemic Changes**

Is there any sign of acute ischaemic change on any sequence? If in doubt as to whether acute or old, code as old and choose No.

Y ☐ N ☐ *If No go to Q.16*

10. **Which side of the brain shows ischaemic change?** (Tick R and L if both)

R ☐ L ☐ **mid** ☐

11. **Classify signs of ischaemic change in the main lesions (if more than one recent lesion) on CT.**

	Y	N	N/A
a) Loss of grey/white matter cortex definition.	☐	☐	
b) Loss of basal ganglia outline.	☐	☐	☐
c) Hypodensity present (so that the lesion appears less dense than white matter).	☐	☐	

12a. **Classify ischaemic change on DWI and axial FLAIR**

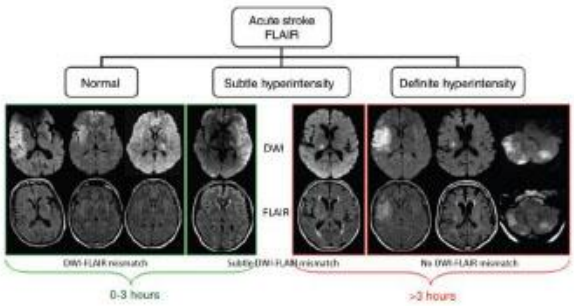
a) Faint hyperintensity on DWI but no lesion visible on FLAIR. (DWI-FLAIR mismatch) ☐

b) Bright hyperintensity on DWI but no lesion visible on FLAIR. (DWI-FLAIR mismatch) ☐

c) Bright or faint hyperintensity on DWI and seems faint hyperintensity on FLAIR. (Subtle DWI-FLAIR mismatch) ☐

d) Lesion clearly visible on FLAIR as well as on DWI. (No DWI-FLAIR mismatch) ☐

e) Lesion visible on FLAIR only. ☐



From Petkova M et al. Radiology.2010;257(3):782-92

12b. Classify ischaemic change on DWI, T2WI and coronal FLAIR

	Y	N
Is there signal change on DWI in ischemic region?	☐	☐
Is there signal change on T2WI in ischemic region?	☐	☐
Is there signal change on FLAIR in ischemic region?	☐	☐

13. Classify site and size of ischaemic lesion(s) on plain CT or MRI (see examples)

a) site (enter most appropriate code in box) ☐

M =MCA* = any lesion in the MCA territory
AS =Infarct of up to half of ACA territory
AL =Infarct of more than half of ACA territory
PS =Infarct of up to half of PCA territory
PL =Infarct of more than half of PCA territory
MAS=M+AS*
MAL=M+AL*
MPS=M+PS*
MPL=M+PL*
MAP=Infarct of whole MCA, ACA and PCA territories
L =Lacunar*
B =Borderzone*
C =Cerebellum*
S =Brainstem*
CS =Cerebellum and brainstem
* code sub-territory sites in b

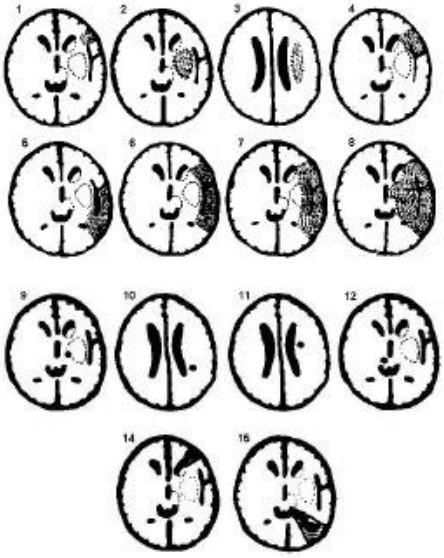
b) MCA sub-territory codes

1=small cortical infarct	☐
2=basal ganglia infarct (>2x2x2cm)	☐
3= infarct of white matter lateral to the lateral ventricle (>2x2x2cm)	☐
4a=infarct of anterior half of peripheral MCA territory (not involving basal ganglia)	☐
4b=infarct of anterior half of peripheral MCA territory (involving part of basal ganglia)	☐
5a=infarct of the posterior half of peripheral MCA territory (not involving basal ganglia)	☐
5b=infarct of the posterior half of peripheral MCA territory (involving part of basal ganglia)	☐
6=infarct of the whole of peripheral MCA territory	☐
7=6+infarct of lateral part of basal ganglia	☐
8=infarct of whole of MCA territory	☐

Lacunar/Borderzone sub-territory codes
9=lacune in internal capsule/lentiform ☐
10=lacune in internal border zone ☐
11=lacune in centrum semiovale ☐
12=lacune in thalamus ☐
13=lacune in brainstem, inc. pons (not shown) ☐
14=anterior (mainly) border zone ☐
15=posterior (mainly) border zone ☐

cerebellum sub-territory codes
16=small cortical (not shown) ☐
17=<1/2 hemisphere (medium) (not shown ☐
18=>1/2 hemisphere (not shown) ☐

Brainstem sub-territory codes
19=small, i.e.<1/2 medulla (not shown) ☐
20=extensive, i.e. pons + medulla (not shown) ☐



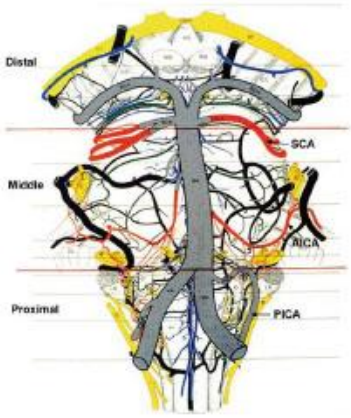
Diagrams ©J Wardlaw, Univ of Edinburgh

c) Posterior Circulation

i. Proximal Y ☐ N ☐ (disappear)
- Right PICA Y ☐ N ☐
- Left PICA Y ☐ N ☐
- BA Y ☐ N ☐

ii. Middle Y ☐ N ☐ (disappear)
- BA Y ☐ N ☐
- Right SCA Y ☐ N ☐
- Left SCA Y ☐ N ☐
- Right AICA Y ☐ N ☐
- Left AICA Y ☐ N ☐

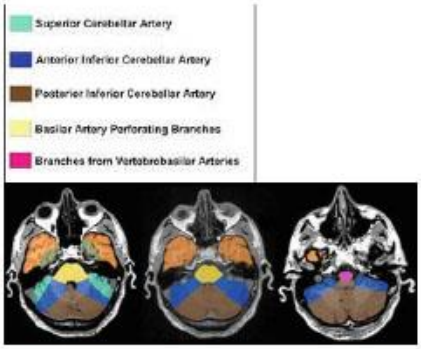
iii. Distal Y ☐ N ☐ (disappear)
- BA Y ☐ N ☐
- Right SCA Y ☐ N ☐
- Left SCA Y ☐ N ☐
- Right PCA Y ☐ N ☐
- Left PCA Y ☐ N ☐



d) anatomical territory sites

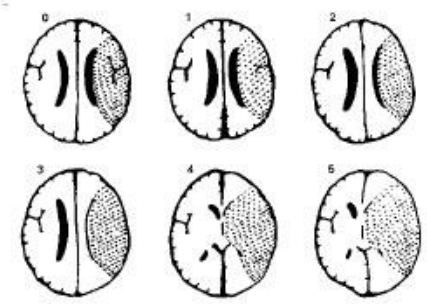
i. Frontal infarct Y ☐ N ☐ (disappear)
ii. Parietal infarct Y ☐ N ☐ (disappear)
iii. Temporal infarct Y ☐ N ☐ (disappear)
iv. Occipital infarct Y ☐ N ☐ (disappear)
v. Brainstem infarct Y ☐ N ☐ (disappear)
- Midbrain Y ☐ N ☐
- Pons Y ☐ N ☐
- Medulla Y ☐ N ☐

vi. Cerebellum infarct Y ☐ N ☐ (disappear)
- Midline Y ☐ N ☐
- R hemisphere Y ☐ N ☐
- L hemisphere Y ☐ N ☐



e) degree of mass effect ☐

Mass effect grading
0=no swelling
1=effacement of the sulci overlying the infarct
2=1+minor effacement of adjacent lateral ventricle
3=1+complete effacement of lateral ventricle
4=1+effacement of the lateral and third ventricle
5=4+shift of the midline away from the side of the ventricle
6=5+effacement of the basal cisterns



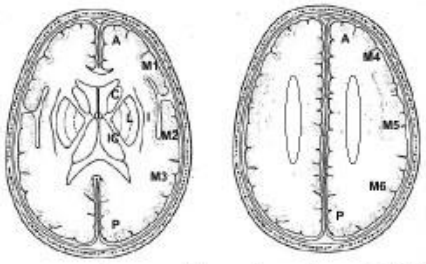
Diagrams ©J Wardlaw, Univ of Edinburgh

14. ASPECTS for the MCA territory

ASPECT Score: _____

Please indicate if each of the MCA areas shown opposite, in the hemisphere that you think is ischaemic, are normal or show some signs of an infarct (abnormal).
(NB: Does not include areas A or P)

	Norm	Abnorm
Caudate (C)	☐	☐
Lentiform (L)	☐	☐
Insula (I)	☐	☐
Internal Capsule (IC)	☐	☐
MCA1 (M1)	☐	☐
MCA2 (M2)	☐	☐
MCA3 (M3)	☐	☐
MCA4 (M4)	☐	☐
MCA5 (M5)	☐	☐
MCA6 (M6)	☐	☐



Diagrams and score taken from Lancet 2000;355:1670-1674

15. Infarct volume (Please measure on DWI and keep two decimals)

a) Supratentorial infarct volume____ cm³

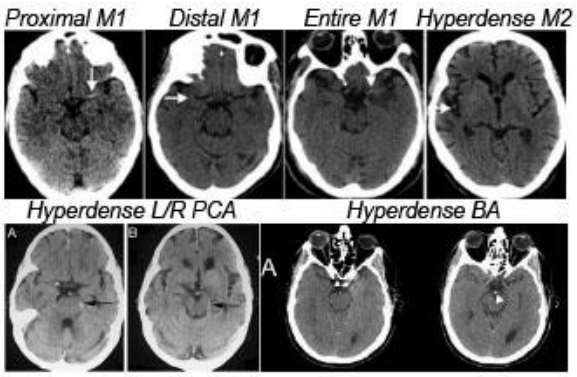
b) Infratentorial infarct volume____ cm³

Brainstem infarct volume____ cm³

Cerebellum infarct volume____ cm³

16. CT hyperattenuated/Abnormal Vessel Sign

Is there a hyperattenuated artery on plain CT (with an absolute density of >43 HU) **Y** ☐ **N** ☐



a) MCA Y ☐ N ☐ (disappear)
R ☐ L ☐
Length: _____ mm
(Please keep two decimals)

Location: Proximal M1 ☐
Distal M1 ☐
Entire M1 ☐
Hyperdense M2 ☐

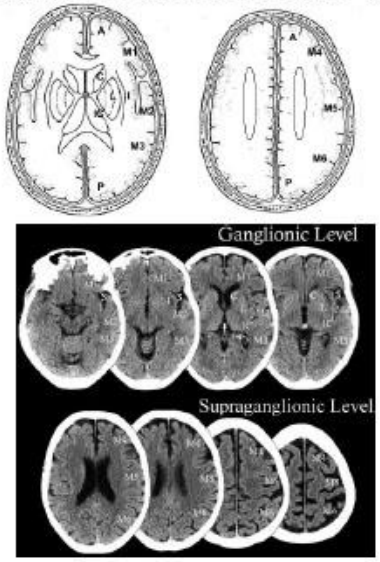
b) PCA Y ☐ N ☐ (disappear)
R ☐ L ☐

c) BA Y ☐ N ☐ (disappear)

17a. Axial FLAIR vessel hyperintensities (FVHs)

Are there FVHs in MCA territory? **Y** ☐ **N** ☐

Diagrams from Lancet 2000;355:1670-1674



a) Anatomic location of FVHs

Proximal MCA (M1 segment) ☐

Middle MCA (M2 segment/sylvian fissure) ☐

Distal MCA (M3 - M6 segment) ☐

Temporal lobe ☐ Frontal lobe ☐
Parietal lobe ☐

b) MCA-ASPECT location of FVHs

Insula (I) / sylvian fissure (S) ☐

M1 ☐ M2 ☐ M3 ☐
M4 ☐ M5 ☐ M6 ☐

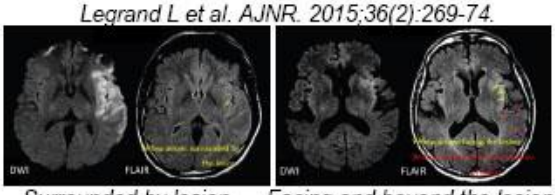
c) Are FVHs surrounded by lesion, facing the lesion or beyond the boundaries of DWI hyperintensity?

Surrounded by lesion ☐

Facing the lesion ☐

Beyond the boundaries of lesion ☐

Legrand L et al. AJNR. 2015;36(2):269-74.

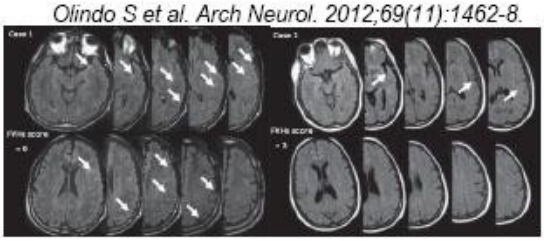


Surrounded by lesion *Facing and beyond the lesion*

d) The FVHs score (0 - 10): ____

(Analyzed from the first M1-MCA appearance to the 10th image. Absence of FVH on 1 slice is rated 0 points. One or more FVHs recognized on 1 slice are rated 1 point.)

Olindo S et al. Arch Neurol. 2012;69(11):1462-8.



Are there FVHs on FLAIR in ACA or PCA territories?

Y ☐ N ☐

ACA territory ☐ PCA territory ☐

17b. Coronal FLAIR vessel hyperintensities (FVHs)

Are there FVHs in MCA territory?

Y ☐ N ☐

Anatomic location of FVHs

Proximal MCA (M1 segment) ☐

Middle MCA (M2 segment/sylvian fissure) ☐

Distal MCA (M3 - M6 segment) ☐

Temporal lobe ☐ Frontal lobe ☐

Parietal lobe ☐

Are there FVHs on FLAIR in ACA or PCA territories?

Y ☐ N ☐

ACA territory ☐ PCA territory ☐

18. **Reduction in brain tissue volume (atrophy)**

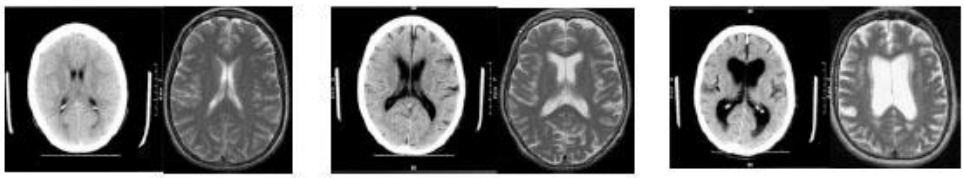
Is there any reduction in brain tissue volume? Y ☐ N ☐

19. **Classify atrophy (see examples and pick nearest likeness):**

	None	Mod	Severe
Central	☐	☐	☐
Cortical	☐	☐	☐

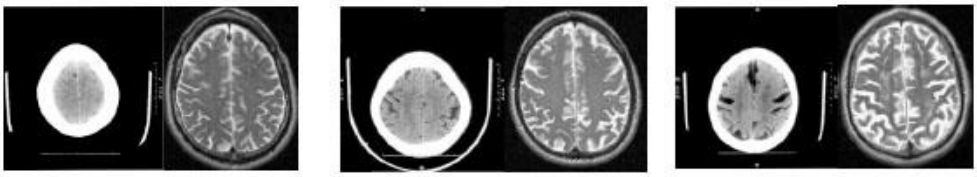
CENTRAL reduction in brain tissue volume

None Modest Severe



CORTICAL reduction in brain tissue

None Modest Severe



Images ©J Wardlaw, Univ of Edinburgh

Approach validated in Eur Radiol 2008;19:177-83

20. **Periventricular white matter changes (lucencies/hyperintensities) on CT or MRI**

Are there any periventricular white matter changes? Y ☐ N ☐ N/A ☐

If No stop here for CT or go to Q. 23 for MRI, if N/A go to Q.23

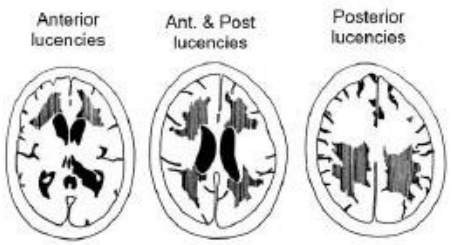
21. **Rate white matter changes using van Swieten scale (CT)**

a. Anterior white matter 0,1,2 ☐

0= no lucency
1= lucency restricted to region adjoining ventricles
2= lucency covering entire region from lateral ventricle to cortex

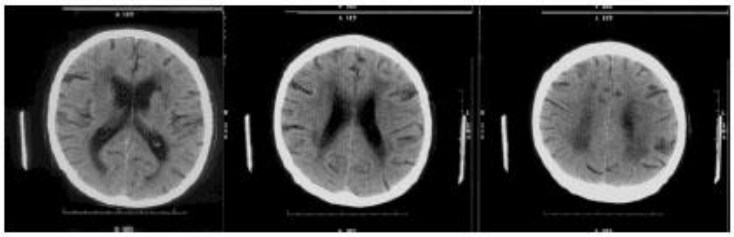
b. Posterior white matter
0= no lucency
1= lucency restricted to region adjoining ventricles
2= lucency covering entire region from lateral ventricle to cortex

0,1,2 ☐

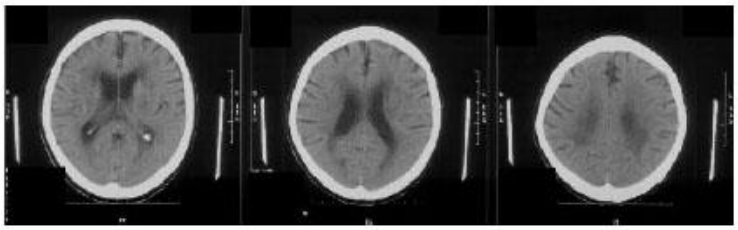


(diagram from van Swieten et al. JNNP 1990;53:1080-1083)

AWM=2 PWM=1



AWM=1 PWM=0



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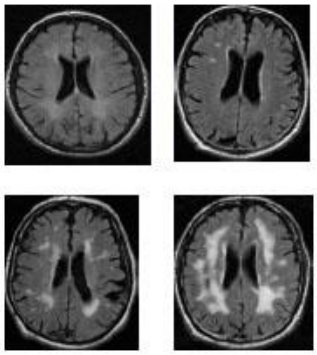
Fazekas et al (1987). AJNR, 8:421-6

22. Rate white matter changes using Fazekas scale (MRI)

a) Periventricular white matter 0,1,2,3 ☐

b) Deep white matter 0,1,2,3 ☐



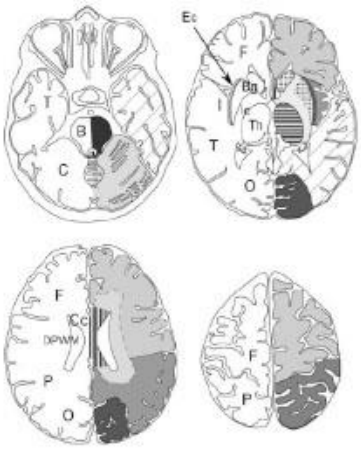
Images ©J Wardlaw, Univ of Edinburgh

23. Microbleeds

Are there any microbleeds in the brain? Y ☐ N ☐

a) Location of microbleeds:

		Definite		Possible	
		R	L	R	L
Infratentorial	Brainstem(B)	☐	☐	☐	☐
	Cerebellum(C)	☐	☐	☐	☐
Deep	Basal Ganglia(Bg)	☐	☐	☐	☐
	Thalamus(Th)	☐	☐	☐	☐
	Internal Capsule(Ic)	☐	☐	☐	☐
	External Capsule(Ec)	☐	☐	☐	☐
	Corpus Callosum(Cc)	☐	☐	☐	☐
	Deep and Periventricular white matter(DPWM)	☐	☐	☐	☐
Lobar	Frontal(F)	☐	☐	☐	☐
	Parietal(P)	☐	☐	☐	☐
	Temporal(T)	☐	☐	☐	☐
	Occipital(O)	☐	☐	☐	☐
	Insula(I)	☐	☐	☐	☐



From Gregoire SM et al. Neurology 2009;73:1759-66.

b) Number of microbleeds <5 mm

Infratentorial	B	C				
	___	___				
Deep	Bg	Th	Ic	Ec	Cc	DPWM
	___	___	___	___	___	___
Lobar	F	P	T	O	I	
	___	___	___	___	___	

c) Number of microbleeds 5-10 mm

Infratentorial	B	C				
	___	___				
Deep	Bg	Th	Ic	Ec	Cc	DPWM
	___	___	___	___	___	___
Lobar	F	P	T	O	I	
	___	___	___	___	___	

Adapted from Cordonnier et al. BOMBS Stroke 2009;49:94-99 and Gregoire SM et al.MARS Neurology 2009;73:1759-66.

24. Cortical Superficial Siderosis

	Y	N
Is there any cortical superficial siderosis?	☐	☐

Location of cortical superficial siderosis

		R	L
Supratentorial	Frontal	☐	☐
	Parietal	☐	☐
	Temporal	☐	☐
	Occipital	☐	☐
	Insula	☐	☐
Infratentorial	Cerebellum	☐	☐
	Brainstem	☐	☐

Number of cortical superficial siderosis

Focal (3 or less sulci) ☐ Disseminated (4 or more sulci) ☐

Appendix D. The ENCHANTED image interpretation form for brain angiography

ENCHANTED ANGIOGRPAPHY IMAGING READING FORMS

Enchanted

Enhanced Control of Hypertension and Thrombolysis Stroke Study

Angiography Image interpretation form – Adapted from the IST-3 trial and INTERRSeCT study

1. Patient No.: 2. BASELINE SCAN ID: 3. DATE OF READING:

4. BASELINE SCAN TYPE: CTA ☐ MRA ☐ DSA or others ☐ 5.DATE/TIME OF BASELINE SCAN:

6.SCAN QUALITY: Good ☐ Moderate ☐ Poor ☐ 7.CTA or MRA phase: ‘

Single-phase or TOF-MRA ☐ Multi-phase ☐ Not applicable (N/A) ☐

(Please select N/A for DSA or others)

8. RANGE OF CTA/MRA: Whole head: ☐ Part of the head: (Willis circle is scanned) ☐

Whole carotid: (The bifurcation is scanned) ☐ Part of the carotid: (only internal carotid artery is scanned) ☐

9.TYPE OF IMAGE: Raw images: ☐ MIP reconstruction images: ☐ MPR or other reconstruction images: ☐

Please tick Yes or No. Please do not leave blanks. Thank you.

10. Is there any appearance of vessel stenosis/occlusion? (comparing with the contralateral side) Y ☐ N ☐ *If No go to Q.17*

11. Which side is affected? R ☐ L ☐ Both ☐

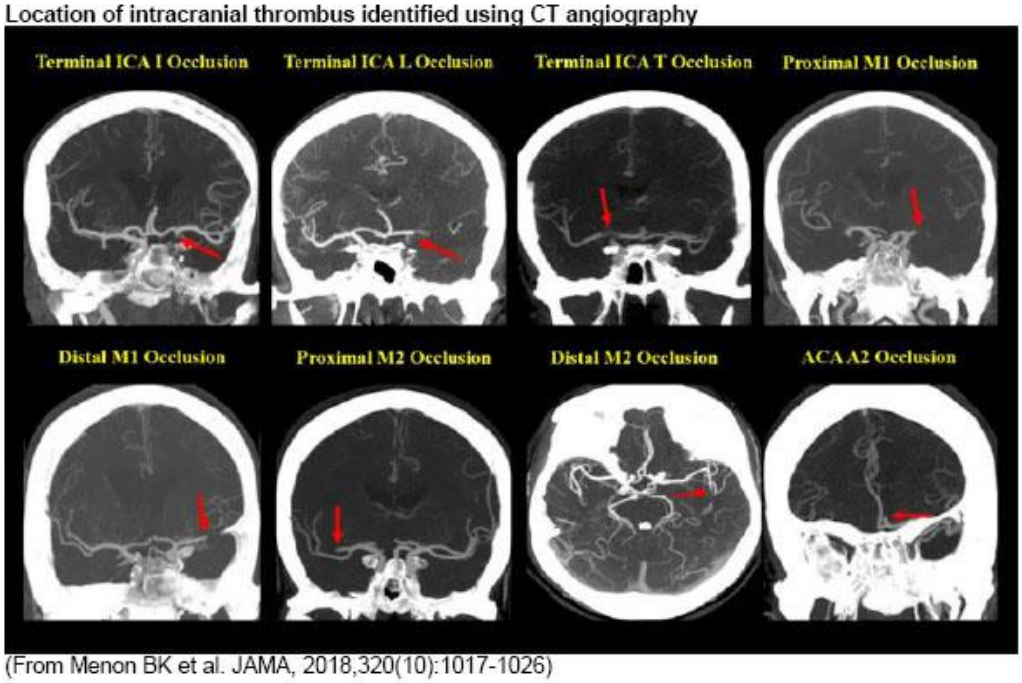
12.1 Location of the stenosis/occlusion (intracranial arteries): (Select 1 to 3 largest arteries involved)

ACA ☐ M1-MCA ☐ M2-MCA ☐ T-ICA ☐ (terminal-segment)

PCA ☐ BA ☐ VA ☐ PtoS-ICA ☐ (petrous- to supra-clinoid-segment)

12.2 Location of the stenosis/occlusion (extracranial carotid artery): Com-CA ☐ Bif-CA ☐ C-ICA ☐ (cervical-segment)

Location of intracranial thrombus identified using CT angiography



(From Menon BK et al. JAMA, 2018,320(10):1017-1026)

Assessment of the vessel stenosis/occlusion:

13.1 IST-3 Angiography Score for stenosis/occlusion degree: ☐

Grade 0: No patency.

Grade 1: Minimal patency – some contrast penetrates stenosis/occlusion but no/minimal enters distal artery.

Grade 2a/b: Patency of less than half of the lumen at the point of stenosis/occlusion and a) only partly filling (<½) or b) incomplete filling but ½ of the major branches of the affected artery.

Grade 3: Patency of >50% of the lumen and filling of most branches of the affected artery.

Grade 4: Complete patency – normal artery.

(Refer to Grant Mair et al. Stroke. 2017; 48(2): 353–360)

13.2 Modified Thrombolysis in Cerebral Infarction (TICI) scale: ☐

0 = No flow/patency

1 = Minimal flow/patency

2a = Partial flow <50% of expected territory

2b = Partial flow >50% of expected territory

3 = Complete flow/patency

14.1 Eligible for clot assessment: Y ☐ N ☐ *If No go to Q.15.1*

(Please select No if thinking stenosis/occlusion is due to chronic vessel change [atheroma or atherosclerosis] rather than acute change [atherosclerotic plaque rupture or cardioembolism])

14.2 Clot Burden Score: ☐

(in occluded ICA/MCA only, please select N/A if not eligible for assessment)

From a total score for normal arteries of 10, two points are subtracted for thrombus found on CTA in the supraclinoid ICA and each of the proximal and distal halves of the MCA trunk. One point is subtracted for thrombus found in the infraclinoid ICA and A1 segment and for each affected M2 branch.

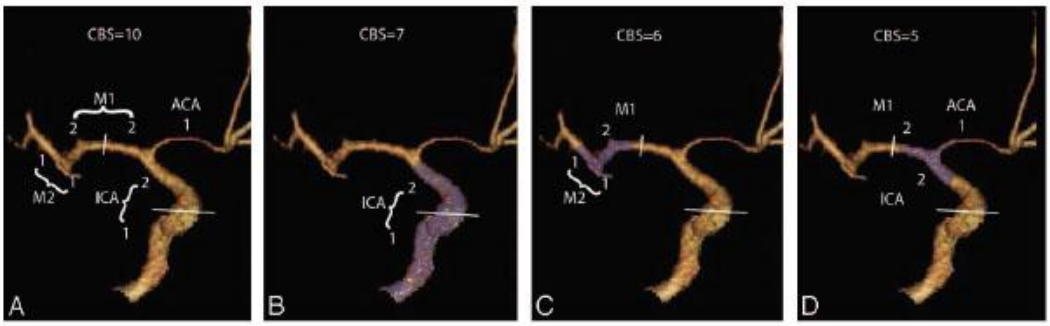


Fig 1. Illustration of CBS. *A*, A 10-point score is normal, implying absence of thrombus. Two points (as indicated) are subtracted for thrombus found on CTA in the supraclinoid ICA and each of the proximal and distal halves of the MCA trunk. One point is subtracted for thrombus found in the infraclinoid ICA and A1 segment and for each affected M2 branch. *B*, Occlusion of infra- and supraclinoid ICAs with a CBS of 7. *C*, Distal M1 and 2 M2 branch occlusions produce a CBS of 6. *D*, Occlusion of the terminal ICA, proximal M1, and A1, with a resultant CBS of 5.

(From Tan IY et al. AJNR, 2009,30(3):525-31)

14.3 Eligible for clot length measurement: Y ☐ N ☐ *If No go to Q.15.1*

(Please select No if lacking axial images with thickness of ≤3mm, or the distal end of clot cannot be identified)

14.4 Can the proximal end of clot be identified: Y ☐ N ☐

(Please select No if the proximal end of clot is not scanned)

14.5 Can the clot length be measured on plain CT before enhancement:

Y ☐ N ☐ *If No go to Q.14.7*

(Please select No if there is no hyperattenuated arteries)

14.6 Clot length on plain CT before enhancement: _____ cm

(Please keep 2 decimals)

14.7 Can the clot length be measured on CTA after enhancement:

Y ☐ N ☐ *If No go to Q.14.9*

14.8 Clot length on CTA after enhancement: _____ cm

(Please measure on raw images of CTA [choose the 3rd phase of multi-phase CTA if possible] and keep 2 decimals)

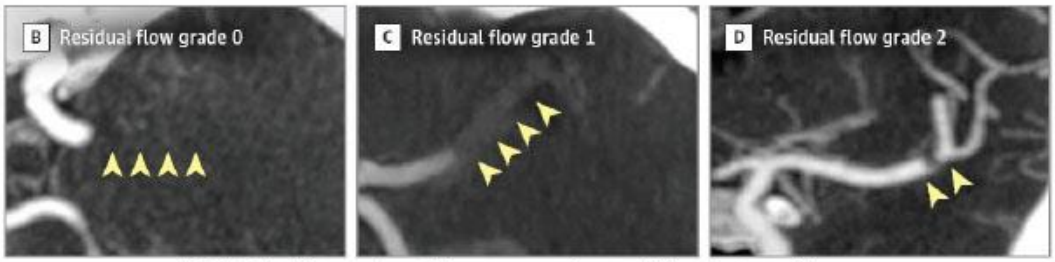
14.9 **Can the clot length be measured on MRA:** Y ☐ N ☐ *If No go to Q.14.11*

14.10 **Clot length on MRA: _____ cm**
(Please measure on raw images of MRA and keep 2 decimals)

14.11 **Eligible for distance measurement from the origin of M1-MCA to the proximal clot interface:** Y ☐ N ☐ *If No go to Q.15.1*
(Please select No if no stenosis/occlusion in ICA/MCA, lacking axial images with thickness of ≤3mm, or the proximal end of clot in ICA/MCA cannot be identified)

14.12 **Distance from the origin of M1-MCA to the proximal clot interface: _____ cm**
(Please keep 2 decimals)

15.1 **Visual grade of residual flow:**
Grade 0 ☐ Grade 1 ☐ Grade 2 ☐ N/A (not eligible for assessment) ☐ (Please select N/A for chronic vessel change)



B-D, Residual flow grade on CTA, a measure of permeability of intracranial thrombus: grade 0 on axial CTA (yellow arrowheads, proximal M1 segment MCA density similar to surrounding brain parenchyma), residual flow grade 1 on axial CTA (yellow arrowheads, distal M1 segment MCA denser than surrounding brain parenchyma), and residual flow grade 2 on coronal CTA (yellow arrowheads, tiny hairline lumen or streak of well-defined contrast within the distal M1 segment MCA thrombus).

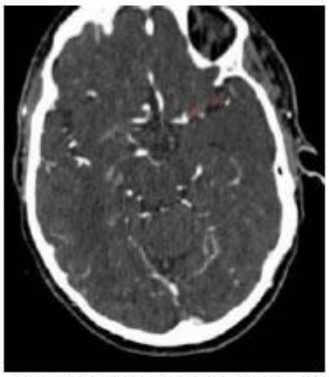
(From Menon BK et al. JAMA, 2018,320(10):1017-1026)

15.2 **Eligible for quantitative residual flow measurement:** Y ☐ N ☐ *If No go to Q.16.1*
(Please select No for chronic vessel change or lacking CTA with thickness of ≤3mm)

15.3 **Maximal CT value of ROI1 (area of 1-3 mm²) in the proximal 1/3 of clot at arterial phase: _____ HU**

15.4 **Maximal CT value of ROI2 (area of 1-3 mm²) in the distal 2/3 of clot at arterial phase: _____ HU**

15.5 **Quantitative residual flow: _____ HU**
(will get automatically by the larger value of *Q.15.3* and *Q15.4*. Method from Prof. Andrew Demchuk, personal communication)



(Quantitative residual flow measurement example of one participant from ENCHANTED)

16.1 **Eligible for collateral circulation assessment:** Y ☐ N ☐ *If No go to Q.17*

16.2 **Angiography type used for collateral circulation assessment:**

Multi-phase CTA	Single-phase CTA	Multi-phase CE-MRA	Single-phase CE-MRA
☐	☐	☐	☐

TOF-MRA	DSA	Others
☐	☐	☐

16.3 **Collateral scoring:** ☐

Collaterals measured **on multi-phase CTA** by comparing backfilling arteries beyond the blocked artery to similar arteries in the opposite unaffected hemisphere. Vascular enhancement distal to occlusion is scored in anterior and posterior MCA territories as:

Grade 0 (poor): Compared to asymptomatic contralateral hemisphere there are no vessels visible in any phase within the occluded vascular territory.

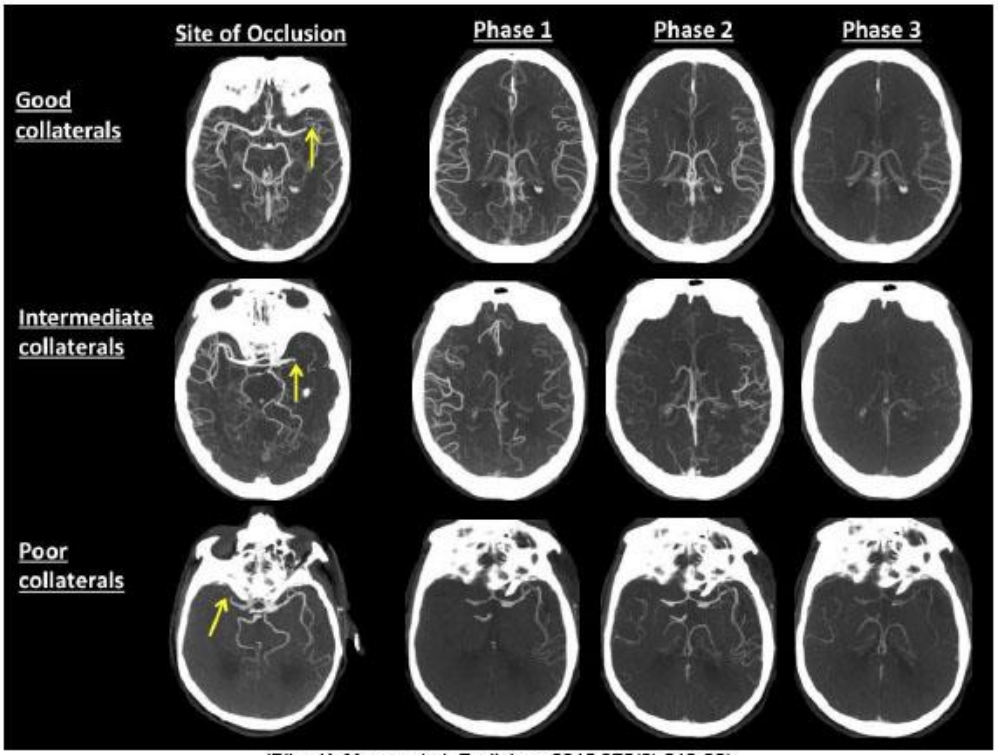
Grade 1 (poor): Compared to asymptomatic contralateral hemisphere there are just a few vessels visible in any phase within the occluded vascular territory.

Grade 2 (intermediate): Compared to asymptomatic contralateral hemisphere there is a delay of two phases in filling in of peripheral vessels and decreased prominence and extent or a one phase delay and some regions with no vessels in some part of the territory occluded.

Grade 3 (intermediate): Compared to asymptomatic contralateral hemisphere there is a delay of two phases in filling in of peripheral vessels but prominence and extent is the same or there is a one phase delay and decreased prominence (thinner vessels) / reduced number of vessels in some part of the territory occluded.

Grade 4 (good): Compared to asymptomatic contralateral hemisphere there is a delay of one phase in filling in of peripheral vessels but prominence and extent is the same.

Grade 5 (good): Compared to asymptomatic contralateral hemisphere, there is no delay and normal or increased prominence of peripheral vessels/ normal extent within the occluded arteries territory within the symptomatic hemisphere.



(Bijoy K. Menon et al. Radiology.2015,275(2):510-20)

Collaterals measured **on single-phase CTA/MRA or TOF MRA**:

Grade 0 (poor): When compared with the asymptomatic contralateral hemisphere, there are no vessels visible within the ischemic territory.

Grade 1 (poor): When compared with the asymptomatic contralateral hemisphere, there are just a few vessels visible in the occluded vascular territory.

Grade 2 (intermediate): When compared with the asymptomatic contralateral hemisphere, there is decreased prominence and extent and regions with no vessels within the ischemic territory in the symptomatic hemisphere.

Grade 3 (intermediate): When compared with the asymptomatic contralateral hemisphere, there is moderately reduced prominence and extent of pial vessels within the ischemic territory in the symptomatic hemisphere.

Grade 4 (good): When compared with the asymptomatic contralateral hemisphere, there is slightly reduced prominence and extent of pial vessels within the ischemic territory in the symptomatic hemisphere.

Grade 5 (good): When compared with asymptomatic contralateral hemisphere, there is increased or normal prominence and extent of pial vessels within the ischemic territory in the symptomatic hemisphere.

Assessment of recanalization:

17 **Number of follow-up scans with angiography?** **0** ☐ **1** ☐ **2** ☐ **3** ☐

If 0 go to Q.21; if 1 complete Q.18; if 2 complete Q.18-19; if 3 complete Q.18-20

18.1 1st FOLLOW-UP SCAN ID: _____

18.2 1st FOLLOW-UP SCAN TYPE: CTA ☐ MRA ☐ DSA or others ☐

18.3 DATE/TIME OF 1st FOLLOW-UP SCAN: _____

18.4 **Date/time interval between baseline and 1st follow-up scan: _____ hrs**
(will calculate automatically according to *Q.18.3* minus *Q.5*)

18.5 **IST-3 Angiography Score for stenosis/occlusion degree (1st follow-up scan):** ☐

Grade 0: No patency.

Grade 1: Minimal patency – some contrast penetrates stenosis/occlusion but no/minimal enters distal artery.

Grade 2a/b: Patency of less than half of the lumen at the point of stenosis/occlusion and a) only partly filling (<½) or b) incomplete filling but ½ of the major branches of the affected artery.

Grade 3: Patency of >50% of the lumen and filling of most branches of the affected artery.

Grade 4: Complete patency – normal artery.

(Refer to Grant Mair et al. Stroke. 2017; 48(2): 353–360)

18.6 **Modified Thrombolysis in Cerebral Infarction (TICI) scale (1st follow-up scan):** ☐

0 = No flow/patency

1 = Minimal flow/patency

2a = Partial flow <50% of expected territory

2b = Partial flow >50% of expected territory

3 = Complete flow/patency

18.7 **Eligible for recanalization assessment (1st follow-up scan):** Y ☐ N ☐ *If No go to Q.18.17*

(Please select No if *Q.10* or *Q.14.1* is assessed as No)

18.8 **Recanalization measurement (1st follow-up scan):** ☐
(revised Arterial Occlusive Lesion [rAOL] score)

Score 0a: Primary occlusive lesion gets worsen with severer stenosis/occlusion degree or more segments of vessel being involved.

Score 0b: Primary occlusive lesion remains same.

Score 1: Debulking of thrombus without recanalization.

Score 2a: Partial or complete recanalization of the primary lesion with thrombus/occlusion in major vascular branch*.

Score 2b: Partial or complete recanalization of the primary lesion with thrombus/occlusion in minor vascular branch**, or partial recanalization of the primary lesion with no thrombus in the vascular tree at or beyond the primary occlusive lesion.

Score 3: Complete recanalization of the primary occlusion with no clot in the vascular tree at or beyond The primary occlusive lesion.

*Major vascular branch: ICA, M1 segment of MCA, Functional M1(thrombus in both proximal M2s of MCA), A1 segment of ACA, Basilar artery, P1 segment of PCA.
**Minor vascular branch: other distal vessels.
** When conventional angiography used for recanalization assessment, the rAOL score was based on the arterial phase of the first angiographic run.
(Refer to Menon BK et al. JAMA, 2018,320(10):1017-1026)

18.9 Eligible for clot length measurement on 1st follow-up scan:

Y ☐ N ☐ *If No go to Q.18.17*

(Please select No if lacking axial images with thickness of ≤3mm, rAOL is scored as 2b or 3, or the distal end of clot cannot be identified)

18.10 Can the proximal end of clot be identified (1st follow-up scan):

Y ☐ N ☐

(Please select No if the proximal end of clot is not scanned)

18.11 Can the clot length be measured on plain CT before enhancement (1st follow-up scan):

Y ☐ N ☐ *If No go to Q.18.13*

(Please select No if there is no hyperattenuated arteries)

18.12 Clot length on plain CT before enhancement (1st follow-up scan): _____ cm
(Please keep 2 decimals)

18.13 Can the clot length be measured on CTA after enhancement (1st follow-up scan):

Y ☐ N ☐ *If No go to Q.18.15*

18.14 Clot length on CTA after enhancement (1st follow-up scan): _____ cm
(Please measure on raw images of CTA [choose the 3rd phase of multi-phase CTA if possible] and keep 2 decimals)

18.15 Can the clot length be measured on MRA (1st follow-up scan):

Y ☐ N ☐ *If No go to Q.18.17*

18.16 Clot length on MRA (1st follow-up scan): _____ cm
(Please measure on raw images of MRA and keep 2 decimals)

18.17 If normal vessel appearance at baseline (*Q.10=N*), is there new clot on 1st follow-up scan:

Y ☐ N ☐ N/A ☐

(Please select N/A if *Q.18.7* is selected as Yes)

19.1 2nd FOLLOW-UP SCAN ID: _____

19.2 2nd FOLLOW-UP SCAN TYPE: CTA ☐ MRA ☐ DSA or others ☐

19.3 DATE/TIME OF 2nd FOLLOW-UP SCAN: _____

19.4 **Date/time interval between baseline and 2nd follow-up scan: _____ hrs**
(will calculate automatically according to *Q.19.3* minus *Q.5*)

19.5 **IST-3 Angiography Score for stenosis/occlusion degree (2nd follow-up scan):** ☐

Grade 0: No patency.

Grade 1: Minimal patency – some contrast penetrates stenosis/occlusion but no/minimal enters distal artery.

Grade 2a/b: Patency of less than half of the lumen at the point of stenosis/occlusion and a) only partly filling (<½) or b) incomplete filling but ½ of the major branches of the affected artery.

Grade 3: Patency of >50% of the lumen and filling of most branches of the affected artery.

Grade 4: Complete patency – normal artery.

(Refer to Grant Mair et al. Stroke. 2017; 48(2): 353–360)

19.6 **Modified Thrombolysis in Cerebral Infarction (TICI) scale (2nd follow-up scan):** ☐

0 = No flow/patency

1 = Minimal flow/patency

2a = Partial flow <50% of expected territory

2b = Partial flow >50% of expected territory

3 = Complete flow/patency

19.7 **Eligible for recanalization assessment (2nd follow-up scan):** Y ☐ N ☐ *If No go to Q.19.17*
(Please select No if *Q.10* or *Q.14.1* is assessed as No)

19.8 **Recanalization measurement (2nd follow-up scan):** ☐
(revised Arterial Occlusive Lesion [rAOL] score)

Score 0a: Primary occlusive lesion gets worsen with severer stenosis/occlusion degree or more segments of vessel being involved.

Score 0b: Primary occlusive lesion remains same.

Score 1: Debulking of thrombus without recanalization.

Score 2a: Partial or complete recanalization of the primary lesion with thrombus/occlusion in major vascular branch*.

Score 2b: Partial or complete recanalization of the primary lesion with thrombus/occlusion in minor vascular branch**, or partial recanalization of the primary lesion with no thrombus in the vascular tree at or beyond the primary occlusive lesion.

Score 3: Complete recanalization of the primary occlusion with no clot in the vascular tree at or beyond The primary occlusive lesion.

*Major vascular branch: ICA, M1 segment of MCA, Functional M1(thrombus in both proximal M2s of MCA), A1 segment of ACA, Basilar artery, P1 segment of PCA.
**Minor vascular branch: other distal vessels.
** When conventional angiography used for recanalization assessment, the rAOL score was based on the arterial phase of the first angiographic run.
(Refer to Menon BK et al. JAMA, 2018,320(10):1017-1026)

19.9 Eligible for clot length measurement on 2nd follow-up scan:

Y ☐ N ☐ *If No go to Q.19.17*

(Please select No if lacking axial images with thickness of ≤3mm, rAOL is scored as 2b or 3, or the distal end of clot cannot be identified)

19.10 Can the proximal end of clot be identified (2nd follow-up scan):

Y ☐ N ☐

(Please select No if the proximal end of clot is not scanned)

19.11 Can the clot length be measured on plain CT before enhancement (2nd follow-up scan):

Y ☐ N ☐ *If No go to Q.19.13*

(Please select No if there is no hyperattenuated arteries)

19.12 Clot length on plain CT before enhancement (2nd follow-up scan): _____ cm
(Please keep 2 decimals)

19.13 Can the clot length be measured on CTA after enhancement (2nd follow-up scan):

Y ☐ N ☐ *If No go to Q.19.15*

19.14 Clot length on CTA after enhancement (2nd follow-up scan): _____ cm
(Please measure on raw images of CTA [choose the 3rd phase of multi-phase CTA if possible] and keep 2 decimals)

19.15 Can the clot length be measured on MRA (2nd follow-up scan):

Y ☐ N ☐ *If No go to Q.19.17*

19.16 Clot length on MRA (2nd follow-up scan): _____ cm
(Please measure on raw images of MRA and keep 2 decimals)

19.17 If normal vessel appearance at baseline (*Q.10=N*), is there new clot on 2nd follow-up scan:

Y ☐ N ☐ N/A ☐

(Please select N/A if *Q.19.7* is selected as Yes)

20.1 3rd FOLLOW-UP SCAN ID: _____

20.2 3rd FOLLOW-UP SCAN TYPE: CTA ☐ MRA ☐ DSA or others ☐

20.3 DATE/TIME OF 3rd FOLLOW-UP SCAN: _____

20.4 Date/time interval between baseline and 3rd follow-up scan: _____ hrs
(will calculate automatically according to *Q.20.3* minus *Q.5*)

20.5 **IST-3 Angiography Score for stenosis/occlusion degree (3rd follow-up scan):** ☐

Grade 0: No patency.

Grade 1: Minimal patency – some contrast penetrates stenosis/occlusion but no/minimal enters distal artery.

Grade 2a/b: Patency of less than half of the lumen at the point of stenosis/occlusion and a) only partly filling (<½) or b) incomplete filling but ½ of the major branches of the affected artery.

Grade 3: Patency of >50% of the lumen and filling of most branches of the affected artery.

Grade 4: Complete patency – normal artery.

(Refer to Grant Mair et al. Stroke. 2017; 48(2): 353–360)

20.6 **Modified Thrombolysis in Cerebral Infarction (TICI) scale (3rd follow-up scan):** ☐

0 = No flow/patency

1 = Minimal flow/patency

2a = Partial flow <50% of expected territory

2b = Partial flow >50% of expected territory

3 = Complete flow/patency

20.7 **Eligible for recanalization assessment (3rd follow-up scan):** Y ☐ N ☐ *If No go to Q.20.17*

(Please select No if *Q.10* or *Q.14.1* is assessed as No)

20.8 **Recanalization measurement (3rd follow-up scan):** ☐
(revised Arterial Occlusive Lesion [rAOL] score)

Score 0a: Primary occlusive lesion gets worsen with severer stenosis/occlusion degree or more segments of vessel being involved.

Score 0b: Primary occlusive lesion remains same.

Score 1: Debulking of thrombus without recanalization.

Score 2a: Partial or complete recanalization of the primary lesion with thrombus/occlusion in major vascular branch*.

Score 2b: Partial or complete recanalization of the primary lesion with thrombus/occlusion in minor vascular branch**, or partial recanalization of the primary lesion with no thrombus in the vascular tree at or beyond the primary occlusive lesion.

Score 3: Complete recanalization of the primary occlusion with no clot in the vascular tree at or beyond The primary occlusive lesion.

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**Minor vascular branch: other distal vessels.
** When conventional angiography used for recanalization assessment, the rAOL score was based on the arterial phase of the first angiographic run.
(Refer to Menon BK et al. JAMA, 2018,320(10):1017-1026)

20.9 Eligible for clot length measurement on 3rd follow-up scan:

Y ☐ N ☐ *If No go to Q.20.17*

(Please select No if lacking axial images with thickness of ≤3mm, rAOL is scored as 2b or 3, or the distal end of clot cannot be identified)

20.10 Can the proximal end of clot be identified (3rd follow-up scan):

Y ☐ N ☐

(Please select No if the proximal end of clot is not scanned)

20.11 Can the clot length be measured on plain CT before enhancement (3rd follow-up scan):

Y ☐ N ☐ *If No go to Q.20.13*

(Please select No if there is no hyperattenuated arteries)

20.12 Clot length on plain CT before enhancement (3rd follow-up scan): _____ cm
(Please keep 2 decimals)

20.13 Can the clot length be measured on CTA after enhancement (3rd follow-up scan):

Y ☐ N ☐ *If No go to Q.20.15*

20.14 Clot length on CTA after enhancement (3rd follow-up scan): _____ cm
(Please measure on raw images of CTA [choose the 3rd phase of multi-phase CTA if possible] and keep 2 decimals)

20.15 Can the clot length be measured on MRA (3rd follow-up scan):

Y ☐ N ☐ *If No go to Q.20.17*

20.16 Clot length on MRA (3rd follow-up scan): _____ cm
(Please measure on raw images of MRA and keep 2 decimals)

20.17 If normal vessel appearance at baseline (*Q.10=N*), is there new clot on 3rd follow-up scan:

Y ☐ N ☐ N/A ☐

(Please select N/A if *Q.20.7* is selected as Yes)

21 Additional comments:

22 Complete? Y ☐ N ☐ Unverified ☐ (submit to panel discuss)