SUPPLEMENTAL MATERIAL

Does Statin Increase the Risk of Intracerebral Hemorrhage in Stroke Survivors? A Meta-analysis and Trial Sequential Analysis

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Table S1: PRISMA Checklist

Section/topic	#	Checklist item	Reported on page #
TITLE			
Title	1	Identify the report as a systematic review, meta-analysis, or both.	1
ABSTRACT			
Structured summary	2	Provide a structured summary including, as applicable: background; objectives; data sources; study eligibility criteria, participants, and interventions; study appraisal and synthesis methods; results; limitations; conclusions and implications of key findings; systematic review registration number.	3
INTRODUCTION			
Rationale	3	Describe the rationale for the review in the context of what is already known.	5-6
Objectives	4	Provide an explicit statement of questions being addressed with reference to participants, interventions, comparisons, outcomes, and study design (PICOS).	6
METHODS			
Protocol and registration	5	Indicate if a review protocol exists, if and where it can be accessed (e.g., Web address), and, if available, provide registration information including registration number.	7
Eligibility criteria	6	Specify study characteristics (e.g., PICOS, length of follow-up) and report characteristics (e.g., years considered, language, publication status) used as criteria for eligibility, giving rationale.	7
Information sources	7	Describe all information sources (e.g., databases with dates of coverage, contact with study authors to identify additional studies) in the search and date last searched.	7
Search	8	Present full electronic search strategy for at least one database, including any limits used, such that it could be repeated.	7, Table S2
Study selection	9	State the process for selecting studies (i.e., screening, eligibility, included in systematic review, and, if applicable, included in the meta-analysis).	Figure S1
Data collection process	10	Describe method of data extraction from reports (e.g., piloted forms, independently, in duplicate) and any processes for obtaining and confirming data from investigators.	8
Data items	11	List and define all variables for which data were sought (e.g., PICOS, funding sources) and any assumptions and simplifications made.	8
Risk of bias in individual studies	12	Describe methods used for assessing risk of bias of individual studies (including specification of whether this was done at the study or outcome level), and how this information is to be used in any data synthesis.	8

Summary measures	13	State the principal summary measures (e.g., risk ratio, difference in means).	8
Synthesis of results	14	Describe the methods of handling data and combining results of studies, if done, including measures of consistency ($e.g., I^2$) for each meta-analysis.	8-9

Table S1: PRISMA Checklist

Section/topic	#	Checklist item	Reported on page #
Risk of bias across studies	15	Specify any assessment of risk of bias that may affect the cumulative evidence (e.g., publication bias, selective reporting within studies).	8
Additional analyses	16	Describe methods of additional analyses (e.g., sensitivity or subgroup analyses, meta-regression), if done, indicating which were pre-specified.	8-9
RESULTS			
Study selection	17	Give numbers of studies screened, assessed for eligibility, and included in the review, with reasons for exclusions at each stage, ideally with a flow diagram.	10
Study characteristics	18	For each study, present characteristics for which data were extracted (e.g., study size, PICOS, follow-up period) and provide the citations.	10, Table 1
Risk of bias within studies	19	Present data on risk of bias of each study and, if available, any outcome level assessment (see item 12).	10-11, Figure S2-S3
Results of individual studies	20	For all outcomes considered (benefits or harms), present, for each study: (a) simple summary data for each intervention group (b) effect estimates and confidence intervals, ideally with a forest plot.	11-12, Table 2, Figure 1
Synthesis of results	21	Present results of each meta-analysis done, including confidence intervals and measures of consistency.	11-12
Risk of bias across studies	22	Present results of any assessment of risk of bias across studies (see Item 15).	10-11, Figure S4
Additional analysis	23	Give results of additional analyses, if done (e.g., sensitivity or subgroup analyses, meta-regression [see Item 16]).	11-13, Figure S5-S7
DISCUSSION			
Summary of evidence	24	Summarize the main findings including the strength of evidence for each main outcome; consider their relevance to key groups (e.g., healthcare providers, users, and policy makers).	14-17
Limitations	25	Discuss limitations at study and outcome level (e.g., risk of bias), and at review-level (e.g., incomplete retrieval of identified research, reporting bias).	17-18
Conclusions	26	Provide a general interpretation of the results in the context of other evidence, and implications for future research.	19

FUNDING			
Funding	27	Describe sources of funding for the systematic review and other support (e.g., supply of data); role of funders for the systematic review.	20

From: Moher D, Liberati A, Tetzlaff J, Altman DG, The PRISMA Group (2009). Preferred Reporting Items for Systematic Reviews and Meta-Analyses: The PRISMA Statement. PLoS Med 6(6): e1000097. doi:10.1371/journal.pmed1000097

For more information, visit: **www.prisma-statement.org**.

Table S2. Search strategy of relevant literature

Three electronic databases, MEDLINE, Embase, and the Cochrane Library, were systematically searched without language restrictions.

1. MEDLINE, PreMEDLINE, and other related databases via PubMed search interface

Search **((((((((((((((stroke) OR strokes) OR Stroke[MeSH Terms]) OR ischaemic stroke) OR ischemic stroke) OR hemorrhagic stroke) OR haemorrhagic stroke) OR fatal stroke) OR brain ischaemia) OR brain ischemia) OR brain haemorrhage) OR brain hemorrhage) OR cerebrovascular accident)) AND ((((((((((((statin) OR statins) OR hydroxymethylglutaryl coa reductase inhibitors[MeSH Terms]) OR HMG CoA reductase inhibitor) OR HMG-CoA *) OR simvastatin) OR atorvastatin) OR rosuvastatin) OR fluvastatin) OR lovastatin) OR pitavastatin) OR pravastatin) OR cerivastatin))) AND ((randomized controlled trial[pt] OR controlled clinical trial[pt] OR randomized[tiab] OR placebo[tiab] OR clinical trials as topic[mesh:noexp] OR randomly[tiab] OR trial[ti] NOT (animals[mh] NOT humans [mh])))** Filters: **Randomized Controlled Trial; Humans**

#1	MeSH descriptor: [Hydroxymethylglutaryl-CoA Reductase Inhibitors] explode all trees
#2	HMG-CoA*
#3	statin or statins
#4	simvastatin or atorvastatin or rosuvastatin or fluvastatin or lovastatin or pitavastatin or pravastatin or cerivastatin

#5	#1 or #2 or #3 or #4

2. Embase via Elsevier search interface

#1	'hydroxymethylglutaryl coenzyme a reductase inhibitor'/exp
#2	'statin'/exp OR 'statins'/exp
#3	'hmg coa':ab,ti
#4	'simvastatin'/exp OR 'atorvastatin'/exp OR 'rosuvastatin'/exp OR 'fluvastatin'/exp OR 'lovastatin'/exp OR 'pitavastatin'/exp OR 'pravastatin'/exp OR 'cerivastatin'/exp
#5	'hydroxymethylglutaryl-coa reductase inhibitor$' OR 'hmg-coa reductase inhibitor$' OR 'hydroxymethylglutaryl coenzyme a reductase inhibitor$'
#6	#1 OR #2 OR #3 OR #4 OR #5
#7	stroke:ab,ti OR strokes:ab,ti OR 'hemorrhagic stroke':ab,ti OR 'haemorrhagic stroke':ab,ti OR 'ischemic stroke':ab,ti OR 'ischaemic stroke':ab,ti OR 'fatal stroke':ab,ti OR 'brain ischemia':ab,ti OR 'brain hemorrhage':ab,ti OR 'brain ischaemia':ab,ti OR 'brain haemorrhage':ab,ti OR 'cerebrovascular accident':ab,ti
#8	'cerebrovascular accident'/exp
#9	#7 OR #8
#10	#6 AND #9
#11	'crossover procedure':de OR 'double-blind procedure':de OR 'randomized controlled trial':de OR 'single-blind procedure':de OR random*:de,ab,ti OR factorial*:de,ab,ti OR crossover*:de,ab,ti OR ((cross NEXT/1 over*):de,ab,ti) OR placebo*:de,ab,ti OR ((doubl* NEAR/1 blind*):de,ab,ti) OR ((singl* NEAR/1 blind*):de,ab,ti) OR assign*:de,ab,ti OR allocat*:de,ab,ti OR volunteer*:de,ab,ti
#12	#10 AND #11
#13	#10 AND #11 AND [randomized controlled trial]/lim
#14	#10 AND #11 AND [randomized controlled trial]/lim AND [humans]/lim

3. Cochrane Library via Wiley search interface

#6	MeSH descriptor: [Stroke] explode all trees
#7	stroke or strokes or "ischaemic stroke" or "ischemic stroke" or "haemorrhagic stroke" or "hemorrhagic stroke" or "brain ischaemia" or "brain ischemia" or "brain haemorrhage" or "brain hemorrhage" or "cerebrovascular accident"
#8	#6 or #7
#9	#5 and #8

Table S3. Excluded articles with reasons for exclusion

Excluded reference	Reason for exclusion
Min LQ, Shao S, Wu XN, et al. Anti-inflammatory and anti-thrombogenic effects of atorvastatin in acute ischemic stroke. *Neural Regeneration Research* 2013; 8: 2144-2154. Article. DOI: 10.3969/j.issn.1673-5374.2013.23.004.	Irrelevant outcome (C-reactive protein, fibrinogen, D-dimer)
Zhou X, Chen J, Wang C, et al. Anti-inflammatory effects of simvastatin in patients with acute intracerebral hemorrhage in an intensive care unit. *Experimental and therapeutic medicine 14(6)*, http://cochranelibrary-wiley.com/o/cochrane/clcentral/articles/402/CN-01430402/frame.html https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5740808/pdf/etm-14-06-6193.pdf (2017).	Irrelevant outcome (Vasospasm, adverse effects, recurrent convulsions)
Tseng M-Y, Czosnyka M, Richards H, et al. Biological effects of acute pravastatin therapy on cerebral vasospasm, delayed ischemic deficits, and outcome in patients following aneurysmal subarachnoid hemorrhage: a randomised controlled trial. *American association of neurological surgeons annual meeting 2006*, http://cochranelibrarywiley.com/o/cochrane/clcentral/articles/272/CN-00603272/frame.html (2006).	Irrelevant outcome (Vasospasm, delayed ischemic neurological deficit)
Tseng M, Hutchinson P, Turner C, et al. Biological effects of acute pravastatin treatment in patients after aneurysmal subarachnoid hemorrhage: a double-blind, placebo-controlled trial. *Journal of neurosurgery 107(6)*, http://cochranelibrarywiley.com/o/cochrane/clcentral/articles/734/CN-00621734/frame.html (2007).	Irrelevant outcome (Vasospasm, delayed ischemic neurological deficit, laboratory tests)
Yusuf S, Lonn E, Pais P, et al. Blood-Pressure and Cholesterol Lowering in Persons without Cardiovascular Disease. *New england journal of medicine 374(21)*, http://cochranelibrarywiley.com/o/cochrane/clcentral/articles/124/CN-01253124/frame.html (2016).	Non-target population (without cardiovascular disease)
Amiri-Nikpour M, Farid V and Ahmadi-Salmasi B. Effect of atorvastatin on cerebral vasomotor reactivity in patients with ischemic stroke. *Journal of global pharma technology 8(12)*, http://cochranelibrary-wiley.com/o/cochrane/clcentral/articles/725/CN-01416725/frame.html (2016).	Irrelevant outcome (Breath holding index and Apnea Mean flow velocity)

Zare M, Saadatnia M, Mousavi S, et al. The effect of statin therapy in stroke outcome: a double blind clinical trial. *International journal of preventive medicine 3(1)*, http://cochranelibrarywiley.com/o/cochrane/clcentral/articles/017/CN-00898017/frame.html (2012).	Irrelevant outcome (NIHSS score, BARTHEL index)
Cao H, Sun C-K, Zhao J, et al. Effect of statins on neurologic impairment and correlative parameters in serum in patients with cerebral infarction. *Chinese journal of clinical rehabilitation 9(9)*, http://cochranelibrary-wiley.com/o/cochrane/clcentral/articles/427/CN00569427/frame.html (2005).	Irrelevant outcome (NIHSS score, BARTHEL index)

Nishiyama Y, Komaba Y, Ueda M, et al. Effect of statins on plasma levels of asymmetric dimethylarginine in patients with noncardiogenic ischemic stroke. *Journal of cerebral blood flow and metabolism 27 Suppl 1*, http://cochranelibrarywiley.com/o/cochrane/clcentral/articles/436/CN-00885436/frame.html (2007).	Irrelevant outcome (serum asymmetric dimethylarginine levels)
Cao H, Sun C-K, Zhao J, et al. Effect of statins on serum C reactive protein and blood lipids in patients with cerebral infarction: a randomized, double-blind, controlled trial. *Chinese journal of clinical rehabilitation 9(21)*, http://cochranelibrarywiley.com/o/cochrane/clcentral/articles/541/CN-00557541/frame.html (2005).	Irrelevant outcome (C-reactive proteins, blood lipids)
Tseng M, Czosnyka M, Richards H, et al. Effects of acute treatment with statins on cerebral autoregulation in patients after aneurysmal subarachnoid hemorrhage. *Neurosurgical focus 21(3)*, http://cochranelibrary-wiley.com/o/cochrane/clcentral/articles/368/CN-00568368/frame.html (2006).	Irrelevant outcome (autoregulation, mean flow velocity in middle cerebral artery and transient hyperemic response test, delayed ischemic neurological deficit)
The effects of cholesterol lowering with simvastatin on cause-specific mortality and on cancer incidence in 20,536 high-risk people: a randomised placebo-controlled trial [ISRCTN48489393]. *BMC medicine* 2005; 3: 6. 2005/03/18. DOI: 10.1186/1741-7015-3-6.	Non-target population (with vascular disease or diabetes)

Guo C. Efficacy and safety of rosuvastatin in treatment of patients with ischemic stroke for secondary prevention of stroke. *Chinese journal of hospital pharmacy [zhongguo yiyuan yaoxue zazhi] 32(3),* http://cochranelibrary-wiley.com/o/cochrane/clcentral/articles/141/CN00858141/frame.html (2012).	Irrelevant intervention (rosuvastatin, simvastatin, diet control); Irrelevant outcome (blood cholesterol level)
Li X. Efficacy of atorvastatin in preventing symptomatic cerebral vasospasm after subarachnoid hemorrhage. *China tropical medicine 10(7),* http://cochranelibrarywiley.com/o/cochrane/clcentral/articles/987/CN-00883987/frame.html (2010).	Irrelevant outcome (Mean flow velocity of middle cerebral artery, symptomatic cerebral vasospasm and cerebral vascular spasm, delayed cerebral infarction)
Xie J and Zhang CG. Effect of atorvastatin on blood lipid and its safety in preventing ischemic stroke. *Chinese Journal of New Drugs* 2010; 19: 956-958+969. Article.	Irrelevant outcome (Cholesterol levels, side effects)
Jaschinski U, Scherer K, Lichtwarck M, et al. Impact of treatment with pravastatin on delayed ischemic disease and mortality after aneurysmal subarachnoid hemorrhage. *Critical care (london, england) 12 (supp 2),* http://cochranelibrary-wiley.com/o/cochrane/clcentral/articles/525/CN00690525/frame.html (2008).	No full text
Lou M. The Safety and Efficacy Study of High Dose Atorvastatin After Thrombolytic Treatment in Acute Ischemic Stroke (SEATIS). http://cochranelibrarywiley.com/o/cochrane/clcentral/articles/852/CN-01445852/frame.html (2017).	No full text

Wei-Guo T, Song-Bin H, Mao-Jun S, et al. Therapeutic effect of simvastatin in ischemic stroke. *Journal of the neurological sciences 238 (Suppl 1),* http://cochranelibrarywiley.com/o/cochrane/clcentral/articles/184/CN-00604184/frame.html (2005).	No full text
Vergouwen M, Vermeulen M, Meijers J, et al. Biological effects of simvastatin in patients with aneurysmal subarachnoid hemorrhage. *Cerebrovascular diseases (basel, switzerland) 25(Suppl 2),* http://cochranelibrary-wiley.com/o/cochrane/clcentral/articles/963/CN-00660963/frame.html (2008).	Duplicated cohort

Yakusevich V, Malygin A and Kabanov A. Effect of simvastatin on the prognosis and the changes of the clinical status in patients with acute ischemic stroke. The results of the 12 month randomized, open comparative study. *Rational pharmacotherapy in cardiology 9(4)*, http://cochranelibrary-wiley.com/o/cochrane/clcentral/articles/276/CN-00916276/frame.html (2013).	Duplicated cohort
Matsumoto M. Effects of 3-hydroxy-3-methyglutaryl-coenzyme A (HMG-CoA) reductase inhibitor upon carotid intima-media complex thickness in the post-ischemic patients with hyperlipidemia during the prospective study of J-STARS. *UMIN clinical trials registry (UMINCTR) (http://wwwuminacjp/ctr/)*, http://cochranelibrarywiley.com/o/cochrane/clcentral/articles/371/CN-00604371/frame.html (2005).	Duplicated cohort
Tseng M, Hutchinson P, Czosnyka M, et al. Effects of acute pravastatin treatment on intensity of rescue therapy, length of inpatient stay, and 6-month outcome in patients after aneurysmal subarachnoid hemorrhage. *Stroke; a journal of cerebral circulation 38(5)*, http://cochranelibrarywiley.com/o/cochrane/clcentral/articles/692/CN-00579692/frame.html http://stroke.ahajournals.org/content/strokeaha/38/5/1545.full.pdf (2007).	Duplicated cohort
Amarenco P, Goldstein L, Szarek M, et al. Effects of intense low-density lipoprotein cholesterol reduction in patients with stroke or transient ischemic attack: the Stroke Prevention by Aggressive Reduction in Cholesterol Levels (SPARCL) trial. *Stroke; a journal of cerebral circulation 38(12)*, http://cochranelibrary-wiley.com/o/cochrane/clcentral/articles/394/CN-00704394/frame.html http://stroke.ahajournals.org/content/strokeaha/38/12/3198.full.pdf (2007).	Duplicated cohort
Koga M, Toyoda K, Minematsu K, et al. Long-term effect of pravastatin on carotid intima-media complex thickness: the J-Stars Echo study (Japan statin treatment against recurrent stroke). *Stroke; a journal of cerebral circulation 49(1)*, http://cochranelibrarywiley.com/o/cochrane/clcentral/articles/747/CN-01445747/frame.html http://stroke.ahajournals.org/content/strokeaha/49/1/107.full.pdf (2018).	Duplicated cohort

Heo J, Song D, Nam H, et al. Randomized double-blind placebo-controlled trials of effects of rosuvastatin in preventing recurrence of ischemic stroke. *International journal of stroke 10(Suppl 2)*, http://cochranelibrary-wiley.com/o/cochrane/clcentral/articles/686/CN-01101686/frame.html (2015).	Duplicated cohort

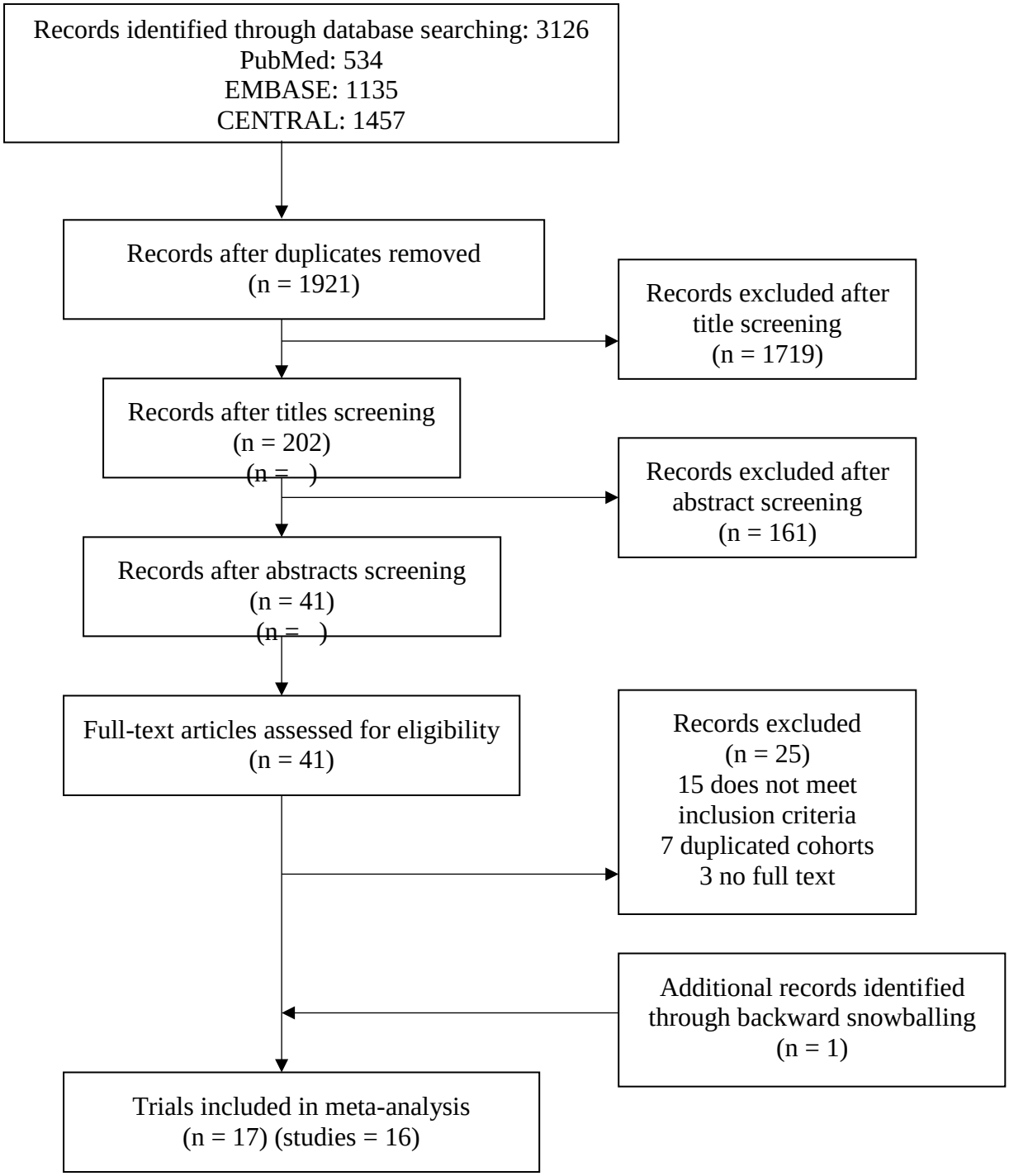


Figure S1. Flow chart of literature selection

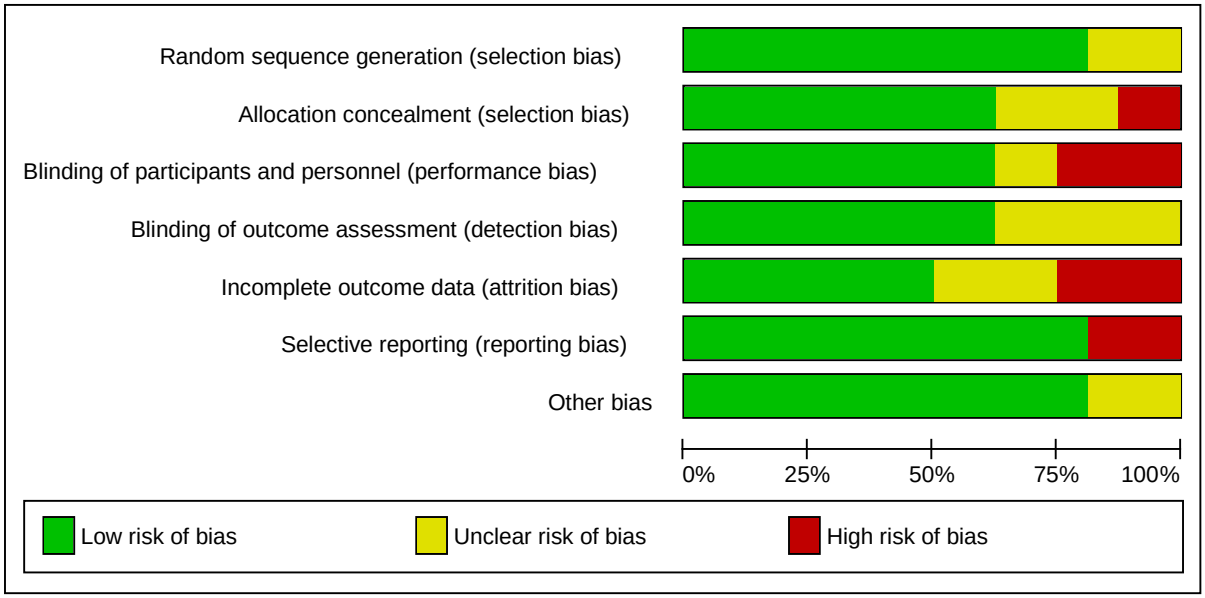


Figure S2. Risk-of-bias graph for the judgement on each methodological quality item that is presented as percentages across all included studies

Random sequence generation (selection bias)

Allocation concealment (selection bias)

Blinding of participants and personnel (performance bias)

Blinding of outcome assessment (detection bias)

Incomplete outcome data (attrition bias)

Selective reporting (reporting bias)

Other bias

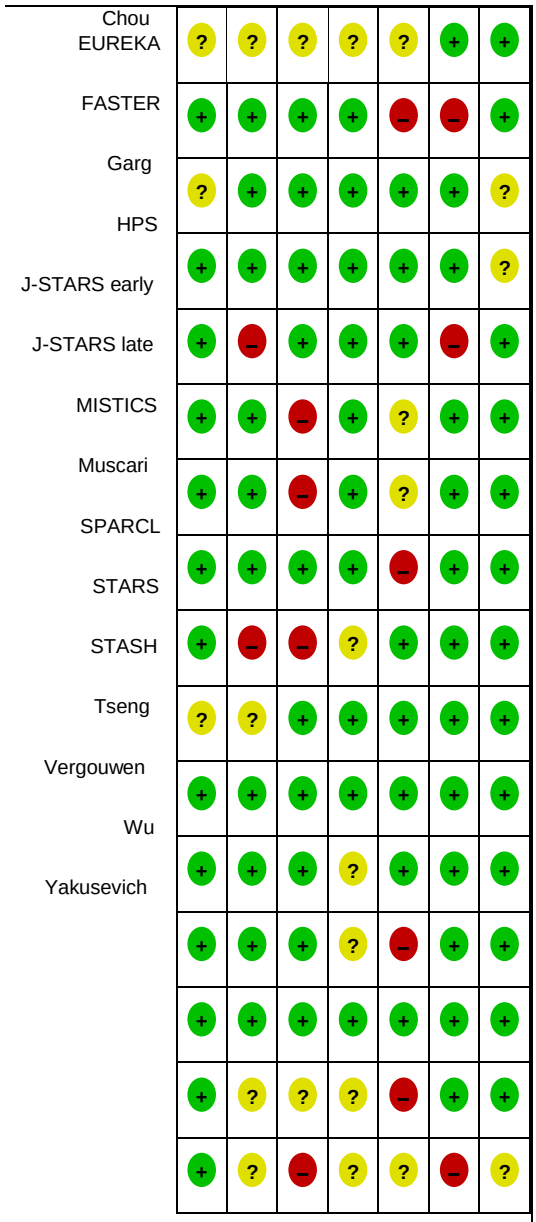
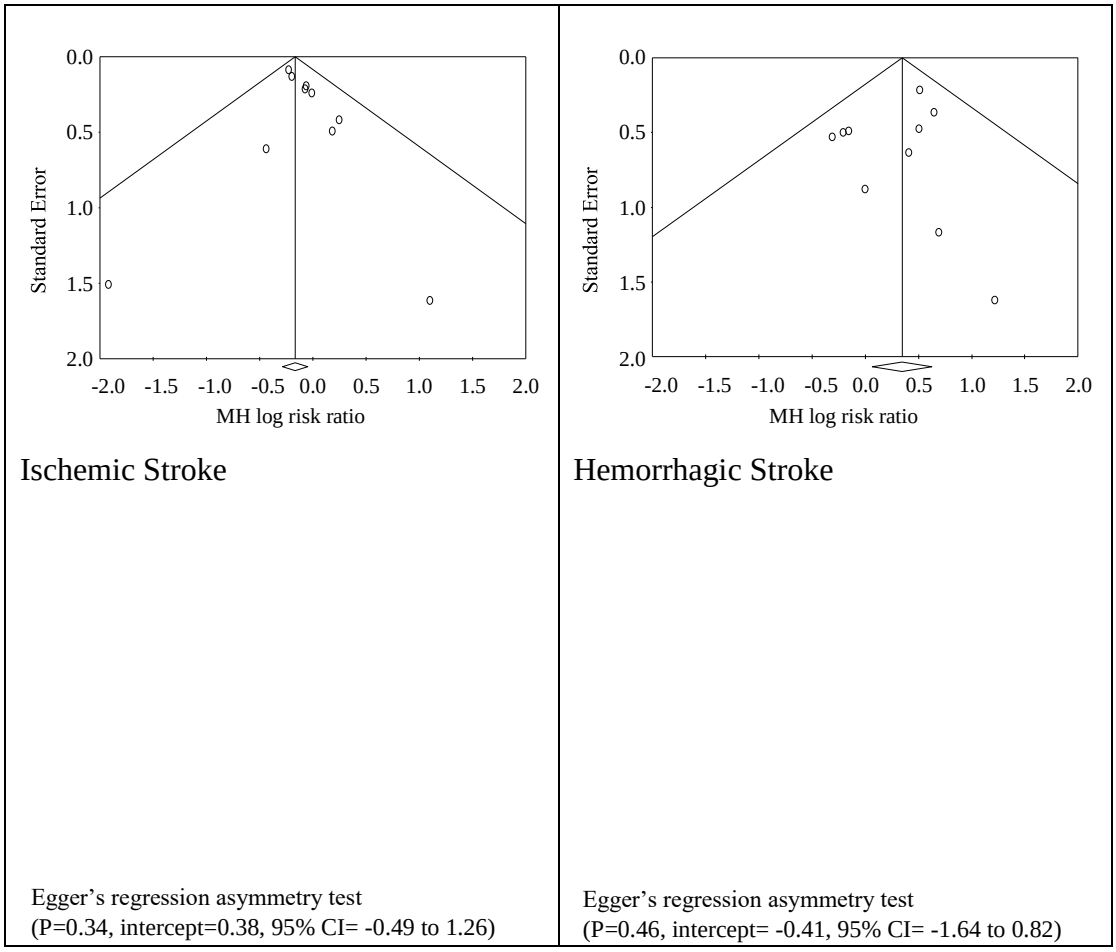
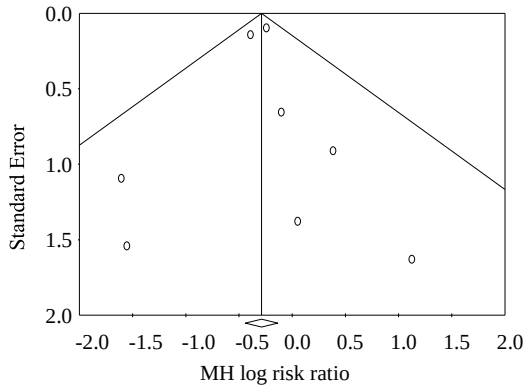

Chou EUREKA
FASTER
Garg
HPS
J-STARS early
J-STARS late
MISTICS
Muscari
SPARCL
STARS
STASH
Tseng
Vergouwen
Wu
Yakusevich

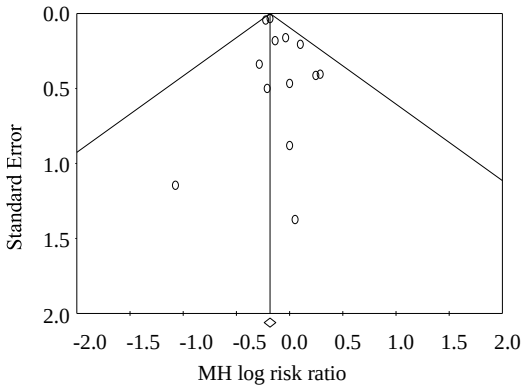
Figure S3. Risk-of-bias summary for all judgements on risk of bias in all included studies Publication bias





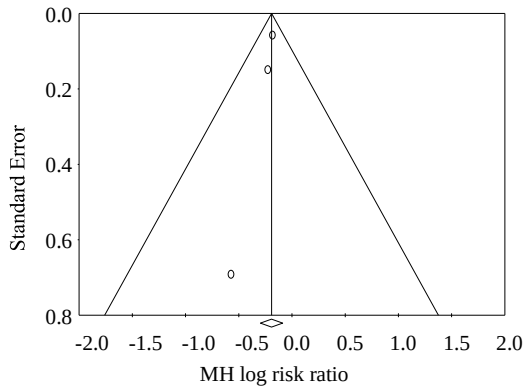
Myocardial infarction

Egger's regression asymmetry test
(P=0.93, intercept= -0.03, 95% CI= -0.99 to 0.92)



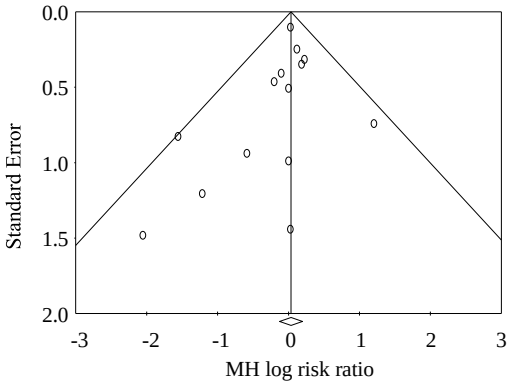
Net clinical benefit

Egger's regression asymmetry test
(P=0.11, intercept=0.43, 95% CI= -0.11 to 0.96)



Cardiovascular mortality

Egger's regression asymmetry test



All-cause death

Egger's regression asymmetry test
(P=0.23, intercept= -0.42, 95% CI= -1.14 to 0.31)

(P=0.07, intercept= -0.58, 95% CI= -1.36 to 0.21)	

Figure S4. Funnel plots and Egger's regression asymmetry test in assessing publication bias

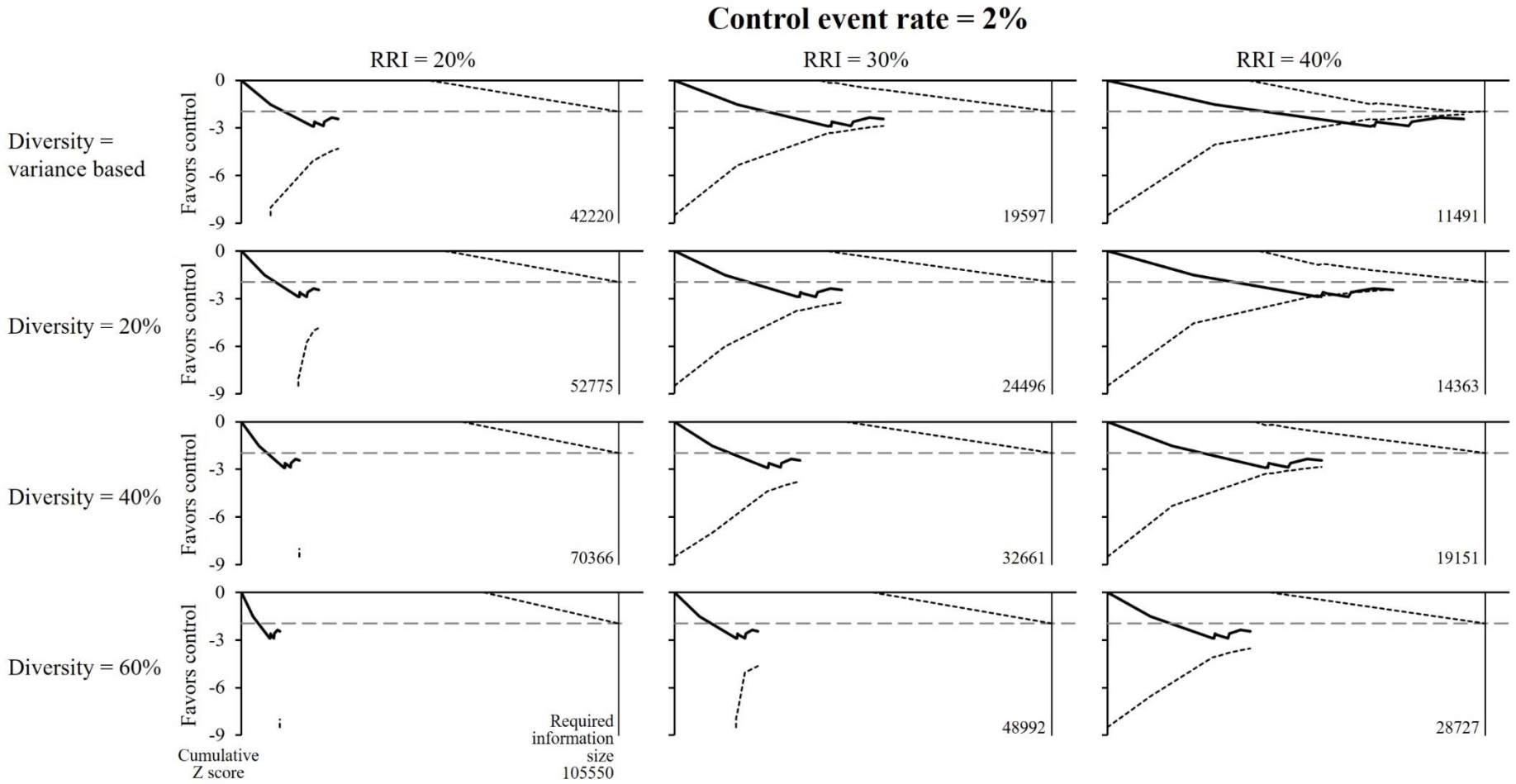


Figure S5. Trial sequential analysis of ten trials reporting the effects of statin on the risk of hemorrhagic stroke in patients with previous stroke

The required information size was calculated based on α of 0.05 (two sided), β of 0.20, a control event rate of 2%, and other different conditions which assumes a range of relative risk increases (RRI of 20%, 30%, or 40%) and various degrees of heterogeneity adjustment (diversity of 0% [model variance based], 20%, 40%, or 60%). The cumulative Z curve (bold solid line) was constructed using a random-effects model. Horizontal dashed line at cumulative Z= -1.96 indicates a conventional level of statistical significance. Converged dot line and diverged dot line represent trial

sequential significance boundary and futility boundary, respectively. These monitoring boundaries were constructed based on the O'Brien-Fleming method.

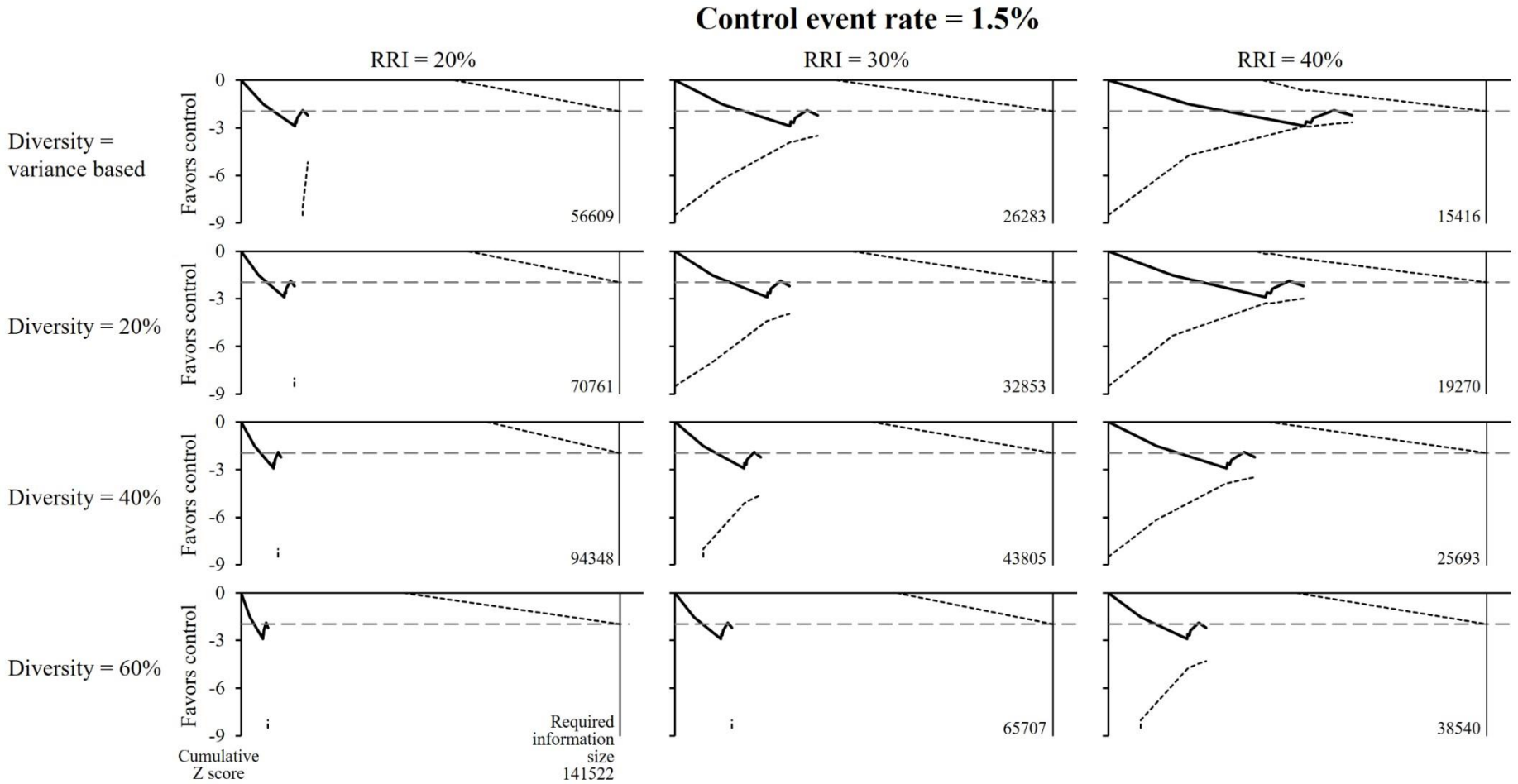


Figure S6. Trial sequential analysis of seven trials reporting the effects of statin on the risk of hemorrhagic stroke in patients with ischemic stroke

The required information size was calculated based on α of 0.05 (two sided), β of 0.20, a control event rate of 1.5%, and other different conditions which assumes a range of relative risk increases (RRI of 20%, 30%, or 40%) and various degrees of heterogeneity adjustment (diversity of 0% [model variance based], 20%, 40%, or 60%). The cumulative Z curve (bold solid line) was constructed using a random-effects model. Horizontal

dashed line at cumulative Z= -1.96 indicates a conventional level of statistical significance. Converged dot line and diverged dot line represent trial sequential significance boundary and futility boundary, respectively. These monitoring boundaries were constructed based on the O'Brien-Fleming method.

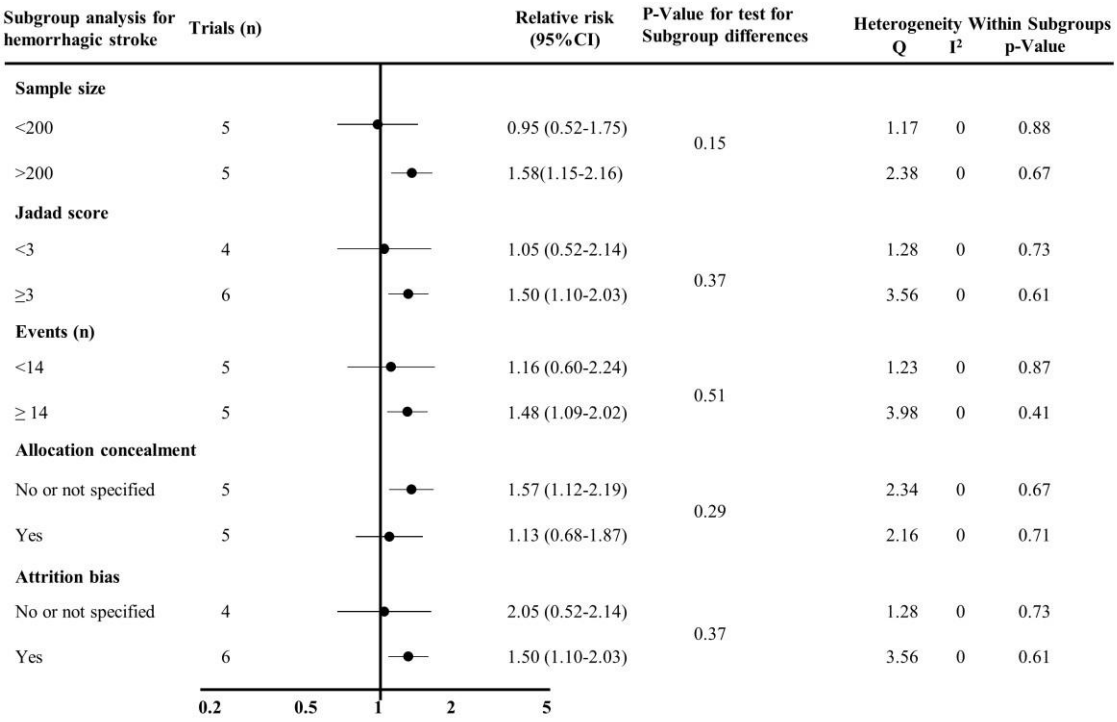


Figure S7. Subgroup analyses relating to study designs across published studies