REVIEW

# A revised systematic review and meta-analysis on the effect of statins on D-dimer levels

Suzanne Schol-Gelok[1] | Francesca Morelli[1] | Lidia R. Arends[2] | Eric Boersma[3] | Marieke J. H. A. Kruip[4] | Jorie Versmissen[1] | Teun van Gelder[1]

[1]Department of Hospital Pharmacy and Internal Medicine, Erasmus MC, University Medical Center Rotterdam, Rotterdam, The Netherlands

[2]Department of Biostatistics, Erasmus MC, University Medical Center Rotterdam, Rotterdam, The Netherlands

[3]Department of Cardiology, Erasmus MC, University Medical Center Rotterdam, Rotterdam, The Netherlands

[4]Department of Hematology, Erasmus MC, University Medical Center Rotterdam, Rotterdam, The Netherlands

**Correspondence**
Suzanne Schol-Gelok, Department of Hospital Pharmacy and Internal Medicine, Room Na-206, Erasmus MC, University Medical Center Rotterdam, 's Gravendijkwal 230, 3015CE Rotterdam, The Netherlands.
Email: s.schol-gelok@erasmusmc.nl

**Funding information**
Suzanne Schol-Gelok received a grant from the Dutch Society for Clinical Pharmacology & Biopharmacy, that allowed her to do her training in Clinical Pharmacology.

## Abstract

**Background:** D-dimers are generated during endogenous fibrinolysis of a blood clot and have a central role in diagnostic algorithms to rule out venous thromboembolism. HMG-CoA reductase inhibitors, more commonly called statins, are known to have effects independent of LDL-cholesterol lowering, including antithrombotic properties. An effect of statins on D-dimer levels has been reported in a prior systematic review and meta-analysis, but methodological shortcomings might have led to an overestimated effect. To re-evaluate the association between statins and D-dimer levels, we systematically reviewed all published articles on the influence of statins on D-dimer levels and conducted a novel meta-analysis (PROSPERO registration number CRD42017058932).

**Materials and methods:** We electronically searched EMBASE, Medline Epub, Cochrane, Web of Science and Google Scholar (100 top relevance) (date of last search: 5 October 2017). We included randomized controlled trials, cohort studies and cross-sectional studies. Two reviewers independently screened all articles retrieved and extracted data on study and patient characteristics, study quality and D-dimer levels.

**Results:** Study-level meta-analysis involving 18,052 study participants showed lower D-dimer levels in those receiving statin treatment than controls (SMD: −0.165, 95% CI −0.234; −0.096, $P = <0.001$). Sensitivity analyses and additional analyses on treatment duration (<12 weeks vs ≥12 weeks) and type of statin (lipophilic or hydrophilic) did not modify this overall result.

**Conclusion:** This meta-analysis suggests an association between use of statins and reduction of D-dimer levels, independent of treatment duration and type of statin used. This effect is small but robust, and should be interpreted with caution.

**KEYWORDS**
D-dimer, fibrin fragment D, hydroxymethylglutaryl-CoA reductase inhibitors, meta-analysis, venous thromboembolism

# 1 | INTRODUCTION

In case of a thromboembolism, D-dimers are generated in the blood clot during fibrinolysis by the sequential action of thrombin, activated factor XIII and plasmin.[1,2] Age, active malignancy, infection, pregnancy and use of anticoagulants are well known to have an influence on D-dimer levels.[3-6] Use of medication with an effect on thrombus formation, such as HMG-CoA reductase inhibitors, more commonly known as statins, may influence D-dimer levels as well. These antithrombotic properties are part of what has been referred to as the cholesterol-independent or "pleiotropic" effects of statins, explaining why the benefits observed with statins appear to exceed what might be expected from changes in cholesterol levels alone.[7-9] In line with these antithrombotic effects, statin treatment might lead to a 15% lower risk of primary venous thrombosis as confirmed in a recent meta-analysis of intervention studies.[7]

In clinical practice, D-dimer levels have a central role in diagnostic algorithms to rule out venous thromboembolism (VTE).[10,11] Several studies have addressed the effect of statins on D-dimer levels, with some of them being evaluated in a systematic review and meta-analysis by Sahebkar et al[12] This meta-analysis included nine randomized controlled trials and reported a significant reduction of 0.988 μg/mL (95%CI: −1.590 to −0.385, $P = 0.001$) in D-dimer levels in statin users. However, this estimate is inappropriate since the used Cohen's d effect size should be dimensionless, while 0.988 μg/mL suggests a tremendous clinical impact of statin use on D-dimer levels. Triggered by this inaccuracy, we further elucidated the used methods and results and found several important shortcomings. Our main concerns next to misuse of Cohen's d are incorrect extraction of data from original studies and unreported assumptions.

Because the research question is of high importance though, we decided to conduct a novel systematic review and meta-analysis on the effect of statins on D-dimer levels, including recent studies.

# 2 | METHODS

## 2.1 | Protocol registration

This study was registered on 10 March 2017 in the PROSPERO international prospective register of systematic reviews (CRD42017058932) and designed according to the guidelines of the 2009 Preferred Reporting Items for Systematic Reviews and Meta-Analysis (PRISMA) statement (Appendix S1 and S2).[13]

## 2.1.1 | Search methods for identification of studies

Together with a biomedical information specialist (see Acknowledgement), SS-G electronically searched the following databases: EMBASE (Ovid SP); Medline Epub (Ovid SP); Cochrane Central Register of Controlled Trials (CENTRAL); Web of Science and Google Scholar (100 top relevance) (date of last search: 5 October 2017). We used search terms as reported in "Appendix S3," in summary: D-dimer OR D-dimers AND statin OR statins OR hydroxymethylglutaryl reductase OR HMG-CoA reductase in combination with individual drug names of statins. To improve sensitivity, we also combined these search terms with the wild-card term "*" and the accessory MeSH terms.

## 2.2 | Data collection and extraction process

Two authors (SS-G and FM) independently screened titles and abstracts retrieved by the electronic survey, and disagreement in selection was resolved by discussion. After consensus was reached, the two reviewers independently selected eligible articles based on the results in full text. Selection of articles was discussed in detail, and in case of disagreement, a third author (TvG) was consulted for final decision. We present a flow diagram to show the decision-making process for including studies in the review (Figure 1).[13] The first reviewer (SS-G) extracted the following data: first author's name, year of publication, study design, country where the study was performed, D-dimer assay used, use of co-medication, number of participants, time of exposure, statin regimen, D-dimer levels with its variation and the conclusions of the individual studies on the effect of statins on the D-dimer levels. Also, all QUADAS-2 items were assessed. If results could not be extracted from original articles (table or well described in the text), authors were requested repeatedly to send their original data. All D-dimer levels were converted to μg/mL. If multiple D-dimer levels were available, we chose to report those values close to 6-month follow-up. All results after extraction were double-checked and confirmed by the second reviewer (FM).

## 2.3 | Selection of studies

We included randomized controlled trials, cohort studies and cross-sectional studies conducted in humans, in which D-dimers levels were described or reported and results could be compared among users or nonusers of statins. For both randomized controlled trials and cohort studies, we defined that statins should be used for at least 7 days in order to achieve a pharmacodynamically relevant effect.[14,15] Also, to prevent interference of the effect of anticoagulant
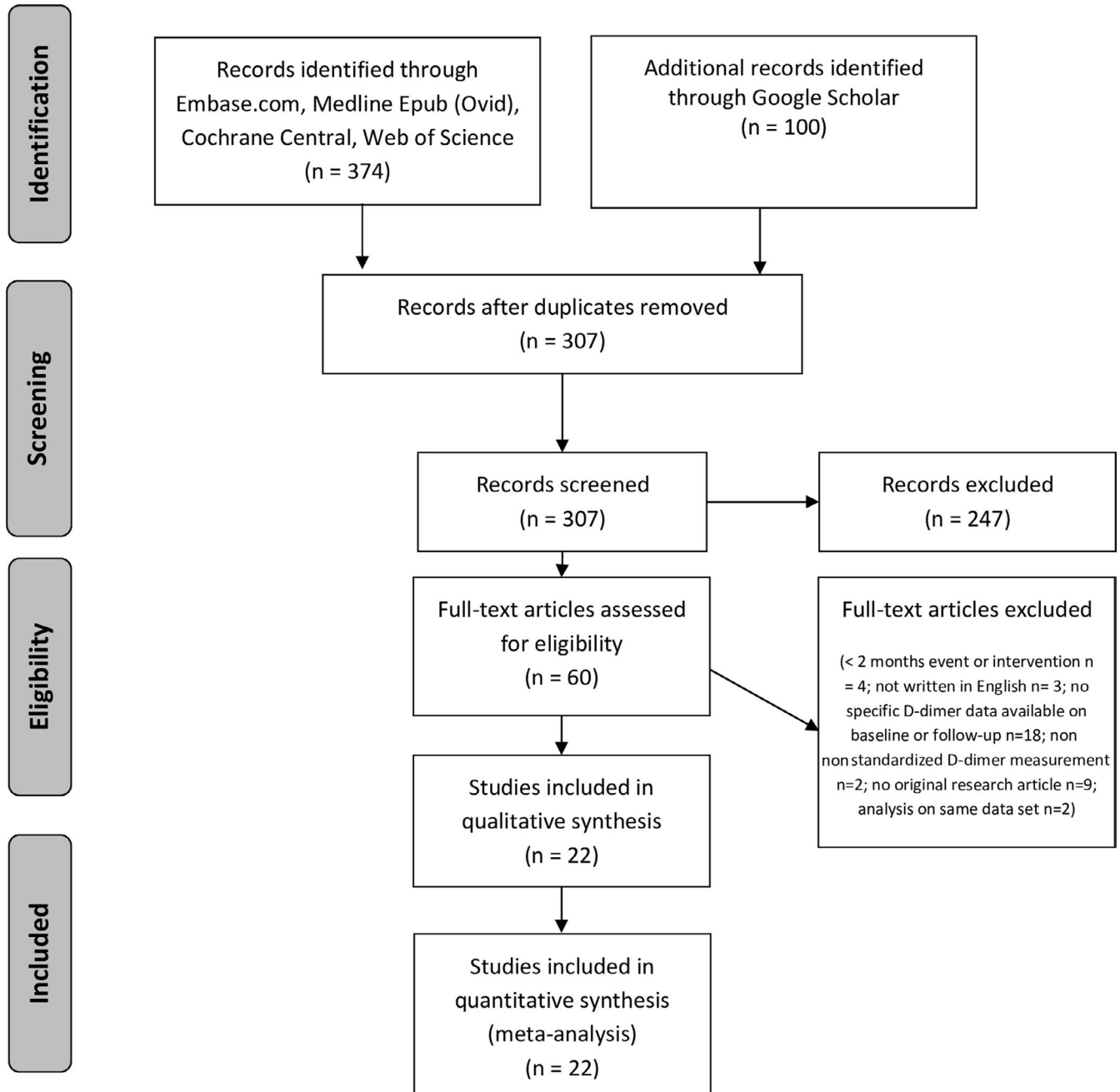
**FIGURE 1** Flow diagram on decision-making process for including studies following PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) statement

drugs on D-dimer levels, we excluded randomized controlled trials or cohorts primary conducted among patients treated with anticoagulant drugs at baseline or during follow-up. Studies in which any medical intervention or cardiovascular event within 2 months between baseline and follow-up measurement of D-dimer levels was part of the inclusion criteria were also excluded to reduce confounding effects on D-dimer levels. Since different D-dimer tests are used in clinical practice, we decided to include only standardized enzyme-linked immunoassays or latex (semi) quantitative tests.[16] Studies without availability of full text

that were also not available after repeated requests to the (corresponding) authors or articles not written in English language were excluded, because the quality of these articles could not be assessed.

## 2.4 | Risk of bias in individual studies and across studies

The data extraction form incorporated a quality assessment section comprising items from Quality Assessment of

Diagnostic Accuracy Studies-2 (QUADAS-2).[17] Following this revised tool, we omitted and added signalling questions and two independent reviewers (SS and FM) applied the QUADAS-2 score in a small number of studies. After refinement of the tool (as described in detail in Appendix S4) with review-specific signalling questions and appropriate items, grouped into three domains (patient selection, index test, and flow and timing) also scoring conflicts of interest, we applied this tool for all studies. We evaluated the influence of each study on the overall effect size by removing one study each time and repeating the analysis, a so-called leave-one-out method sensitivity analysis.[18] We also performed a subanalysis including only studies with low-risk patient selection bias and low concern about applicability according to the scoring of these QUADAS-2 items and performed a separate subanalysis only including controlled trials. To detect potential publication bias, we visually inspected the distribution of the studies within a funnel plot and also created a funnel plot taking into account the trim-and-fill adjustment of Duval and Tweedie.[19] Also, Begg's rank correlation and Egger's test were used to detect publication bias.[20,21] Furthermore, as another marker of publication bias, we estimated the number of missing studies we would need to retrieve and impute in the meta-analysis to make the p-value nonsignificant using the "fail-safe N" method.[22]

## 2.5 | Quantitative data-synthesis

The meta-analysis was conducted using Comprehensive Meta-analysis (version 3; Biostat). In studies in which participants were exposed to different statin regimens, the different statin-exposed groups were analysed separately and values were compared to the control group in case of (randomized) controlled studies. When medians and interquartile ranges (IQR) were reported, we estimated the average standard deviation (SD) using the following formula: SD = ((75th percentile-25th percentile)/1.35) and in case of reporting medians and full range, we estimated the average SD using the following formula: SD = ((75th percentile-25th percentile)/5.16).[23] If not reported, the mean difference was estimated using the following formula: SD = square root [(SDpre-treatment)2 + (SDpost-treatment)2 − (2R × SDpre-treatment × SDpost-treatment)], assuming a correlation coefficient ($R$) = 0.5. Net changes in measurements (change scores) were calculated for controlled trials, as follows: (value at end of follow-up in the treatment group − value at baseline in the treatment group) − (value at end of follow-up in the control group − value at baseline in the control group). If percentage change in D-dimer levels was reported, we estimated mean or median D-dimer post-treatment levels by multiplying reported mean or median pre-treatment D-dimer levels with 1 +

percentage change and assumed that the post-treatment SD was equal to reported SD before treatment. For crossover studies, we used the reported results of delta mean change and its corresponding SD to prevent artificial widening of confidence intervals of the pooled treatment effect.[24] For cohorts, we calculated change scores by (value at end of follow-up in the treatment group − value at baseline in the treatment group) assuming that in a fictional control group D-dimer would not change during follow-up. For results on cross-sectional studies, we measured change scores by (value in the statin users group − value in the nonexposed group). When the authors adjusted D-dimer levels for other confounding factors, we used the adjusted D-dimer levels for analysis. We expressed effect sizes as a standardized mean difference (SMD) with its corresponding 95% confidence intervals (CIs) using the dimensionless Cohen's d as the summary statistic.[25] To compensate for heterogeneity including study design, population characteristics, statin dose and treatment duration, we used a random-effects model. Post hoc subanalyses were performed to assess the potential effects of treatment duration of statin therapy (<12 weeks vs ≥12 weeks) and type of statin (lipophilic or hydrophilic). Simvastatin, atorvastatin and fluvastatin were classified as lipophilic statins and pravastatin and rosuvastatin as hydrophilic statins.[15]

## 3 | RESULTS

### 3.1 | Study selection and evaluation of bias of individual studies

In total, we screened 307 studies, of which 60 were assessed for eligibility reading full text, and finally, 22 studies were included in this review (Figure 1).[26-47] Reasons for exclusion were an event or intervention <2 months (n = 4), not written in English (n = 3), no specific D-dimer data available on baseline or follow-up (n = 18), nonstandardized D-dimer measurement (n = 2), no original research article (n = 9) and repeated analysis on same data set (n = 2). We included 7 controlled trials, 11 cohort studies and 4 cross-sectional studies. Taken together, this analysis included 22 control groups and 27 statin-exposed groups with a total number of 18 052 study participants (Table 1). The included studies were performed among different study populations. Six studies were performed in subjects with dyslipidaemia, 6 studies in patients with proven cardiovascular disease, 4 studies in HIV-infected patients, 2 in patients with type 2 diabetes mellitus, one in healthy subjects, one in patients diagnosed with lupus, one in COPD patients and one in heart transplant patients. Of all 27 statin-exposed groups, 17 groups were defined as lipophilic-type statin users and 7 as hydrophilic-type statin users, while the other 3 groups comprised of lipophilic-type as well as hydrophilic-type statin users. Of the

23 statin-exposed groups in which we could assess treatment duration, 19 groups were exposed to statins for 12 weeks or longer.

The risk of bias regarding patient selection was regarded low for only 6 of the 22 included studies and for 8 studies we had concerns about applicability of the results based on the specific characteristics of the statin-exposed groups and control groups included in these studies (Figure 2, Table S1). For four studies, the D-dimer test was not clearly described, and we assumed a standardized test.[34,37,38,47]

## 3.2 | Meta-analysis

Study-level meta-analysis involving 18 052 study participants showed significantly lower D-dimer levels in those receiving statin treatment compared to controls (SMD: $-0.165$, 95% CI $-0.234$; $-0.096$, $P \le 0.001$) (Figure 3). The estimated effect sizes were similar in sensitivity analyses that omitted any single study (Figure 4). The 6 studies with low risk of patient selection (SMD: $-0.099$, 95%CI $-0.140$; $-0.058$, $P < 0.001$) and the 16 studies with low risk of limited patient applicability (SMD: $-0.216$, 95%CI $-0.334$; $-0.099$, $P < 0.001$) also resulted in lower D-dimer values after statin treatment. A separate meta-analysis of the 7 controlled trials did not show a different effect on D-dimer levels (SMD: $-0.096$, 95% CI $-0.138$; $-0.055$, $P < 0.001$). Furthermore, treatment duration ($<12$ weeks vs $\ge 12$ weeks) did not influence the effect on D-dimer levels in statin users ($P = 0.887$) (Figure 5) and type of statin (lipophilic or hydrophilic) also did not modify this overall result ($P = 0.167$) (Figure 6).

## 3.3 | Publication bias

A visual inspection of the funnel plot showed asymmetry, suggesting potential publication bias. Using the "trim-and-fill" method with five potentially missing studies imputed, the effect size was estimated to an adjusted SMD with a larger effect ($-0.224$, 95% CI $-0.295$; $-0.153$) than the unadjusted SMD (Figure 7). Begg's rank correlation (Kendall's Tau with continuity correction $= -0.160$, $Z = 1.167$, two-tailed $P = 0.243$) and Egger's test (intercept $-0.611$, 95% CI $-1.447$; $0.226$, two-tailed $P = 0.145$) were both nonsignificant. Following the "fail-safe N" method, we would need to retrieve and impute 422 missing studies in the meta-analysis to make the p-value nonsignificant.

## 4 | DISCUSSION

In this meta-analysis, for which we included randomized controlled trials, cohort and cross-sectional studies conducted in humans, we found that statin treatment is associated with lower D-dimer levels. This effect is small but robust and not driven by any single study. Results from post hoc subanalyses on treatment duration and type of statin therapy were not different from this overall effect.

Our findings are important in further understanding the pleiotropic antithrombotic effects of statins. Statins have been shown to significantly lower the risk of primary VTE and therefore might have a role in the prevention of VTEs.[7,48] Several mechanisms have been described to explain these antithrombotic properties. Statins inhibit platelet activation within hours after intake by upregulation of the nitric oxide synthase and downregulation of phospholipase A2–mediated thromboxane A2 formation and probably also by reduced exposure of platelet-derived microparticles and glycoprotein IIIa, a receptor for fibrinogen and von Willebrand factor.[49-51] Also important, statins interfere directly with the clotting system. In vitro, two lipophilic types of statins decreased tissue factor activity in a dose-dependent manner.[52] As a result, a smaller amount of factor X is activated and generation of thrombin is diminished.[8,53,54] Other ways through which statins interfere with the clotting system are inhibition of isoprenoid intermediates, which indirectly activates the protein C pathway and lowering of the oxidized LDL-induced tissue factor expression. Inhibition of geranylgeranylation of the Rho/Rho kinase pathway is one of the key mechanisms of these anticoagulant effects.[8,55] By inhibition of this pathway, resulting in a shift in the fibrinolytic balance towards increased fibrinolytic activity is suggested by inhibition of the expression of plasminogen activator inhibitor-1 and upregulation of tissue-type plasminogen activator.[56,57]

These mechanisms might consequently result in lower D-dimer levels in statin users. This decrease of D-dimer levels may theoretically be stronger for lipophilic than for hydrophilic type of statin users. Lipophilic type of statins can enter cells in any organ and also penetrate cell membranes. In contrast, cellular uptake of hydrophilic type of statins is dependent on the presence of a specific carrier-mediated mechanism, which is only present in hepatocytes but not in extrahepatic cells.[58] Furthermore, tissue factor activity could in vitro only be decreased by lipophilic type of statins and not by pravastatin, a hydrophilic type of statin.[52] Clinical relevant difference of pleiotropic effects in general between lipophilic and hydrophilic type of statins is however controversial.[9] In our subanalyses on type of statin therapy, for both lipophilic and hydrophilic type of statin users D-dimer levels were significantly lower. This effect was not significantly different among these groups. Probably the clinical anticoagulant effect in vivo is independent on the mechanism of uptake.

The question of a possible dose-effect of statins in lowering D-dimer levels is also relevant, yet hard to answer because of difference in statin types and dosages that were applied in the included studies. Still, we applied a post hoc analysis, utilizing the previously developed concept of a "statin

**TABLE 1** Characteristics of included studies for meta-analysis on the effect of statins on plasma D-dimer level

| | Location | Population | D-dimer assay | Information about use of co-medication | Age (years) | Time of exposure |
|---|---|---|---|---|---|---|
| **Controlled trials** | | | | | | |
| Chang, 2002 | South Korea | Haemodialysis patients with hypercholesterolaemia5 | ELISA Asserachrom D-Di (Diagnostica Stago, Asnières-sur-Seine, France) | Exclusion cholesterol modifying or oxidation medication | 63 (11) 60 (12) | 8 wk |
| Eckhard, 2014 | USA | Nonhypercholesterolaemic HIV infected | LPIA (Diagnostica Stago, Parsippany, NJ) | On antiretroviral therapy ASA, steroids, NSAIDs, antihypertensive medication | 45.6 (41.1-51.4) 46.9 (39.2-53.6) | 24 wk |
| Kinlay, 2009 | USA | acute coronary syndromes | Not reported | ASA, heparin, nitrates and β-blockers | 64 (12) | 16 wk |
| Nixon, 2016[a] | USA | HIV infected | ELISA (Diagnostica Stago, Asnières-sur-Seine, France) | On antiretroviral therapy Exclusion of immuno-suppressant users | 48 (41-55) | 20 wk |
| Sommeijer, 2004 | The Netherlands | Type 2 diabetes mellitus | LPIA (bioMérieux, Durham, NC)op | Antihypertensive medication, ASA | Overall: 59 (54-64) median (IQR) | 8 wk |
| Tonkin, 2015 | Australia | Acute coronary syndrome | LPIA (Architect c8000, Abbott Diagnostics) | ASA | 62 (55-67) 63 (56-68) | 12 mos |
| Van de Ree, 2003 | The Netherlands | Type 2 diabetes mellitus | ELISA (Dade-Behring, Marburg, Germany) | - | 59.7 (7.6) 60.3 (7.8) 58.6 (7.5) | 30 wk |
| **Cohort studies** | | | | | | |
| Bolaman, 2006 | Turkey | Primary hypercholesterolaemia | ELISA (not otherwise specified) | - | 55 (10) | 24 wk |
| Calza, 2017 | Italy | HIV-1 infected | ELISA (Medical Systems, Genova, Italy) | On antiretroviral therapy Exclusion of steroid, androgen, oestrogen, growth hormone, antihypertensive medication, thyroid preparation and acid-reducing agent users | 46.8 (40.6-55.9) | 6 mos |
| Costejon, 2017 | Spain | Females with sae systemic lupus erythematosus | Not reported | Antimalarials and immunosuppressant | 47 (23-80) | 8 wk |
| Hölschermann, 2000 | Germany | heart transplant recipients receiving oral immunosuppression | ELISA (Asserachrom; Boehringer Mannheim Diagnostics, Mannheim, Germany) | immunosupresssants | 48 (12) (mean (SD)) | 7 d or 1 mos |
| Joukhadar, 2001 | Austria | Hypercholesterolaemia | ELISA (Diagnostica Stago, Asnières, France) | Exclusion of hypolipaemic, anticoagulant, anti-inflammatory or antihypertensive medication users | 55 (9) 52 (9) 55 (8) | 3 mos |
| Lin, 2000 | Taiwan | hypercholesterolaemia | LPIA (Diagnostica Stago, France) | Antihypertensive medication, hormone replacement | 59.8 (7.1) | 8 wk |
| Lin, 2006 | Taiwan | hyperlipidaemia | LPIA (Diagnostica Stago, France) | - | 58.5 (9.7) | 16 wk |
| Seljeflot, 2002 | Norway | dyslipidaemia and history of angina pectoris | ELISA in plasma and serum (Asserachrom D-Di; Stago Diagnostica, Asnières-sur-Seine, France) | Antihypertensive medication, warfarin, ASA, nitrates | Not reported | 12 mos |
| Trifiletti, 2003 | Italy | Hypercholesterolaemia | ELISA (Asserachrom; Diagnostica Stago) | Exclusion of ASA users | 55 (3) | 6 mos |

| Regimen (daily dose) | Participants (number) | D-dimer (µg/mL) before exposure | D-dimer (µg/mL) after exposure | Conclusion | Details |
|---|---|---|---|---|---|
| Simvastatin (20mg) | 28 | 1.05 (0.90) | 0.99 (0.83) | No effect | Open RCT |
| No simvastatin | 30 | 1.12 (1.01) | 1.09 (0.97) | | |
| Rosuvastatin (10 mg) | 67 | 0.19(0.13-0.33) | Baseline + 6.9% (43.8 to −35.0) | No effect | Double-blind RCT |
| Placebo | 69 | 0.18 (0.09, 0.29) | Baseline + 21.9% (−9.1 to 73.3) | | |
| Atorvastatin (80 mg) | 387 | Overall 0.3447 (0.0708 to 5.351) | Baseline + 0.0108 µg/mL (−93,2 to 145) | No effect | Double-blind RCT |
| Placebo | | | Baseline + 0.0244 µg/mL (−0.1097 to 0.1234) | | |
| a)Atorvastatin (10-20 mg) | 37 | 0.1870 (0.1209-0.3196) | 0.219 (0.1352-0.3177) | No difference | Double-blind RCT with crossover design with 4-wk washout period |
| a)Placebo | 37 | 0.1998 (0.1319-0.3383) | 0.2127 (0.1467-0.3393) | | |
| b)Placebo | 36 | 0.1785 (0.1256-0.2545) | 0.1804 (0.1316-0.2250) | | |
| b)Atorvastatin (10-20mg) | 36 | 0.1727 (0.1212-0.3039) | 0.1755 (0.1113-0.2387) | | |
| Pravastatin (40 mg) | 50 | - | Between pravastatin and no pravastatin group change: −0.02 (−0.09 to 0.05) | No effect | Open RCT met crossover design. |
| No pravastatin | 50 | | | | |
| Pravastatin (40mg) | 3941 | 0.172 (0.112-0.269) | 0.166 (0.108-0.263) | Significant reduction | Double-blind RCT |
| Placebo | 3922 | 0.173 (0.112-0.276) | 0.178 (0.115-0.284) | | |
| a)Atorvastatin (10 mg) | 69 | 0.115 (0.086-0.160) | Baseline −7.4% | Significant reduction in both atorvastatin groups | Double-blind RCT |
| b)Atorvastatin (40-80 mg) | 66 | 0.137(0.104-0.186) | Baseline −8.5% | | |
| Placebo | 61 | 0.123 (0.101-0.151) | Baseline + 1.9% | | |
| Atorvastatin (10mg – 20mg) | 44 | 0.195(0.073) | 0.197 (0.085) | No effect | |
| rosuvastatin (10mg) | 57 | 0.345 (0.166-0.445) | 0.275 (0.149-0.381) | Significant reduction | |
| Atorvastatin (20mg) | 37 | 0.49 (0.46) | 0.51 (0.39) | No effect | |
| Simvastatin (10mg) | 15 | 0.695 (total range 0.160-1.580) | 0.490 (total range 0.160-1.470) | Significant reduction | |
| a) Atorvastatin (10 mg) | 24 | 0.42 (0.53) | 0.35 (0.34) | No effect | |
| b) Pravastatin (40 mg) | 24 | 0.29 (0.15) | 0.29(0.16) | | |
| c) Simvastatin (40 mg) | 27 | 0.35 (0.25) | 0.33(0.17) | | |
| Pooled data | 75 | 0.35(0.34) | 0.33(0.23) | | |
| Fluvastatin (40mg) | 23 | 0.38 (0.31) | 0.28 (0.19) | Significant reduction | Exclusion of familial hypercholesterolaemia |
| Simvastatin (20mg-40mg) | 22 | 0.33 (0.17) | 0.29 (0.14) | No effect | |
| a) atorvastatin (20-40mg) | 28 | 0.493 (0.296-0.767) | 0.416 (0.269-0.749) | No effect | Both serum and plasma D-dimers reported. In this review, plasma D-dimer (mostly used assay) reported |
| b) Simvastatin (20-40mg) | 30 | 0.384(0.218-0.657) | 0.385(0.221-0.541) | | |
| Atorvastatin (20mg) | 32 | 0.248 (0.055) | 0.229 (0.042) | No effect | |

(Continues)

**TABLE 1** (Continued)

| | Location | Population | D-dimer assay | Information about use of co-medication | Age (years) | Time of exposur |
|---|---|---|---|---|---|---|
| Wada, 1992 | Japan | Hypercholesterolaemia | Felisa D-dimer (Agen, Brisbane, Australia) | - | 55.2 (14.6) | >3 mos |
| Weiss, 2016 | France | HIV-1–infected receiving c-ART | ELISA (Asserachrom D-Di) | c-ART | 47 (41-54) | 12 wk |
| Cross-sectional studies | | | | | | |
| Adams, 2013 | USA | Caucasian, African American, Hispanic and Chinese, free of cardiovascular diseases or active cancer | LPIA(Liatest D-DI; Diagnostica Stago, Parsippany, NJ) | - | 65.9 (8.7) 61.5 (10.3) | - |
| Kaba, 2004 | USA | ≥2 months post-myocardial infarction | ELISA (American Diagnostica, Greenwich, CT, USA) | ASA, antihypertensive medication, oral anticoagulants | 60 (12) 58 (11) | - |
| Vidula, 2010 | USA | Peripheral artery disease | ELISA (Asserachrom D-Di kit; Diagnostica Stago, Asnières-sur-Seine, France) | - | 72.1 (7.9) 73.0 (8.9) | - |
| Walter, 2010 | Germany | Undergoing elective coronary angiography | ELISA (Asserachrom D-Di, Stago, Asnières, France) | Antihypertensive medication, oral anti-hyperglycaemic drugs, ASA, clopidogrel | 60.6 (10.4) 62.4 (9.0) | - |

*Note*: Data are reported as means (SD) or medians (75th percentile to 25th percentile) unless stated otherwise

Abbreviations: ASA, acetylsalicylic acid; d, days; ELISA, enzyme-linked immunosorbent assay; LPIA, latex-enhanced photometric immunoassays; mg, milligram; mos, months; NA, not available; RCT, randomized controlled trial; wk, weeks.

[a]Original data on effects on D-dimers received and reported.

correction factor," while adjusting for differences in the potency of statin type/dosage on LDL lowering.[59] Following this concept, we visually inspected the relation of the SMD in D-dimer levels against the statin correction factor and found no clear dose-effect relation (Figure S1). An explanation for this lack of dose-effect on D-dimer levels could be that other mechanisms are involved in the anticoagulant effect of statins compared to the cholesterol-dependent effects. The dose-effect relation of statins on D-dimers levels might therefore

be independent of the potency of lowering LDL-cholesterol levels.

Considering lower D-dimer levels in statin users, the performance of the diagnostic algorithms used for patients with suspected pulmonary embolism or deep vein thrombosis could be different for statin users. In these algorithms, a normal D-dimer level in combination with a low clinical probability of thrombosis safely excludes VTE.[60,61] Most D-dimer cut-offs in these diagnostic algorithms range between 0.5 and 1.0 μg/mL,
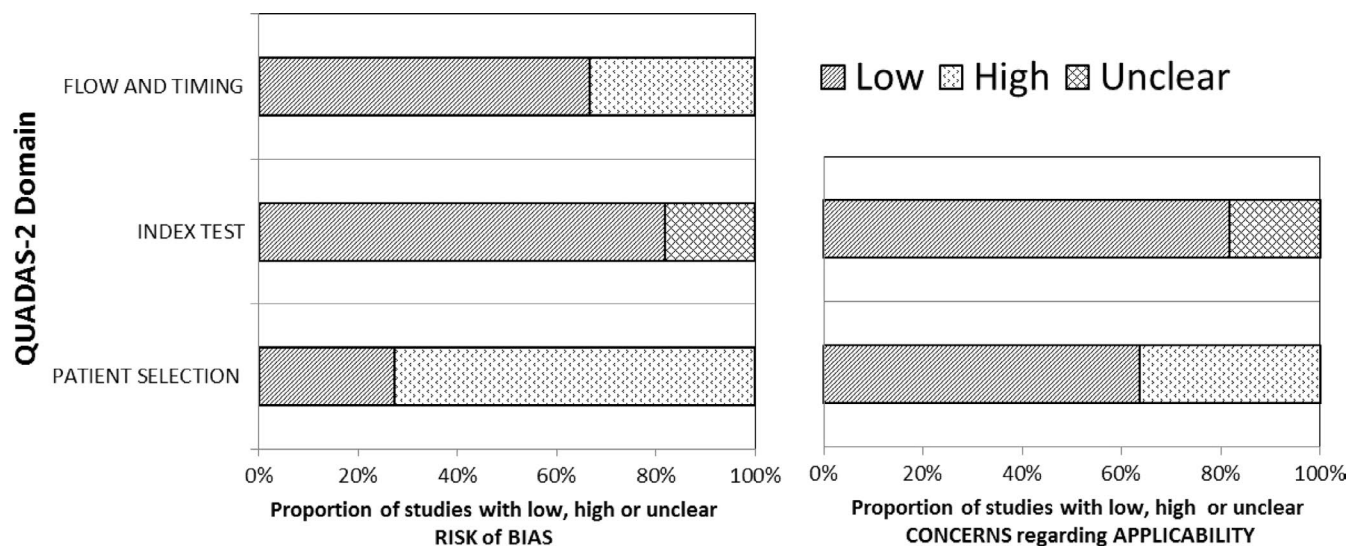


**FIGURE 2** Graphical display for QUADAS-2 results of the 22 studies included

| Regimen (daily dose) | Participants (number) | D-dimer (µg/mL) before exposure | D-dimer (µg/mL) after exposure | Conclusion | Details |
|---|---|---|---|---|---|
| Pravastatin (10mg) | 48 | 0.11 (0.06) | 0.056 (0.039) | Significant reduction | |
| Rosuvastatin (20 mg) | 43 | 0.194 (0.147-0.279) | Baseline +3.7% (−18.2 to +23.3) | No effect | |
| Statin users | 1001 | Not reported | 0.21 | Significant reduction | Adjustment for age, sex, education, individual income and cardiovascular risk factors |
| Nonusers | 5786 | | 0.23 | | |
| Statin users | 644 | 0.487 (0.434) | - | Significant reduction | |
| Nonusers | 401 | 0.731 (1.2) | | | |
| Statin users | 242 | 1.1 (1.4) | - | No effect | |
| Nonusers | 337 | 0.97 (1.4) | | | |
| Atorvastatin (10-40mg) | 54 | 0.466 (0.173) | - | No effect | Matching based on the cholesterol levels |
| Nonusers | 54 | 0.454 (0.182) | | | |

depending on the clinical rule applied.[61,62] These cut-off levels have high sensitivity rates, and therefore, a false negative test in statin users is unlikely to occur. In a recent retrospective post hoc analysis, adjusting D-dimer cut-offs for statin users did not result in a safer diagnostic strategy.[63] However, further validation in a larger prospective cohort is needed.

It is important to note that there are main differences between our methodology and the systematic review and



| Study name | Std diff in means | Standard error | Variance | Lower limit | Upper limit | Z-Value | P-Value |
|---|---|---|---|---|---|---|---|
| Adams et al., 2013 | −0.097 | 0.012 | 0.000 | −0.121 | −0.073 | −7.991 | 0.000 |
| Bolaman et al., 2006 | 0.025 | 0.151 | 0.023 | −0.270 | 0.321 | 0.166 | 0.868 |
| Calza et al., 2017 | −0.887 | 0.156 | 0.024 | −1.193 | −0.580 | −5.673 | 0.000 |
| Chang et al., 2002 | −0.032 | 0.263 | 0.069 | −0.547 | 0.483 | −0.122 | 0.903 |
| Costejon et al., 2017 | 0.047 | 0.164 | 0.027 | −0.276 | 0.369 | 0.283 | 0.777 |
| Eckhard et al., 2014 | −0.178 | 0.172 | 0.030 | −0.514 | 0.159 | −1.033 | 0.301 |
| Hölschermann et al., 2000 | −0.773 | 0.294 | 0.087 | −1.350 | −0.196 | −2.627 | 0.009 |
| Joukhadar et al., 2001a | −0.151 | 0.205 | 0.042 | −0.553 | 0.252 | −0.733 | 0.463 |
| Joukhadar et al., 2001b | 0.000 | 0.204 | 0.042 | −0.400 | 0.400 | 0.000 | 1.000 |
| Joukhadar et al., 2001c | −0.090 | 0.116 | 0.013 | −0.317 | 0.136 | −0.782 | 0.434 |
| Kaba et al., 2004 | −0.300 | 0.064 | 0.004 | −0.425 | −0.174 | −4.686 | 0.000 |
| Kinlay et al., 2009 | −0.078 | 0.102 | 0.010 | −0.277 | 0.121 | −0.766 | 0.444 |
| Lin et al., 2000 | −0.369 | 0.216 | 0.046 | −0.792 | 0.053 | −1.714 | 0.087 |
| Lin et al., 2006 | −0.255 | 0.217 | 0.047 | −0.679 | 0.170 | −1.175 | 0.240 |
| Nixon et al., 2016a | 0.132 | 0.233 | 0.054 | −0.324 | 0.588 | 0.567 | 0.571 |
| Nixon et al., 2016b | 0.009 | 0.236 | 0.056 | −0.453 | 0.471 | 0.037 | 0.971 |
| Seljeflot et al., 2002a | −0.219 | 0.191 | 0.037 | −0.593 | 0.156 | −1.143 | 0.253 |
| Seljeflot et al., 2002b | 0.003 | 0.183 | 0.033 | −0.354 | 0.361 | 0.019 | 0.985 |
| Sommeijer et al., 2004 | −0.193 | 0.143 | 0.020 | −0.473 | 0.087 | −1.351 | 0.177 |
| Tonkin et al., 2015 | −0.092 | 0.023 | 0.001 | −0.136 | −0.048 | −4.078 | 0.000 |
| Trifiletti et al., 2003 | −0.382 | 0.183 | 0.034 | −0.740 | −0.023 | −2.084 | 0.037 |
| van de Ree et al., 2003a | −0.228 | 0.176 | 0.031 | −0.574 | 0.117 | −1.295 | 0.195 |
| van de Ree et al., 2003b | −0.274 | 0.178 | 0.032 | −0.624 | 0.076 | −1.536 | 0.125 |
| Vidula et al., 2010 | 0.093 | 0.084 | 0.007 | −0.072 | 0.258 | 1.101 | 0.271 |
| Wada et al., 1992 | −1.024 | 0.178 | 0.032 | −1.373 | −0.675 | −5.746 | 0.000 |
| Walter et al., 2010 | 0.068 | 0.193 | 0.037 | −0.310 | 0.445 | 0.351 | 0.726 |
| Weiss et al., 2016 | 0.074 | 0.153 | 0.023 | −0.226 | 0.373 | 0.482 | 0.630 |
| | −0.165 | 0.035 | 0.001 | −0.234 | −0.096 | −4.676 | 0.000 |

**FIGURE 3** Forest plot for the effect of statin therapy on plasma D-dimer concentrations. Effect sizes were expressed as standardized mean difference (SMD) with its corresponding 95% confidence intervals (CIs) using Cohen's d as the summary statistic. A random-effects model was used for performance of the meta-analysis

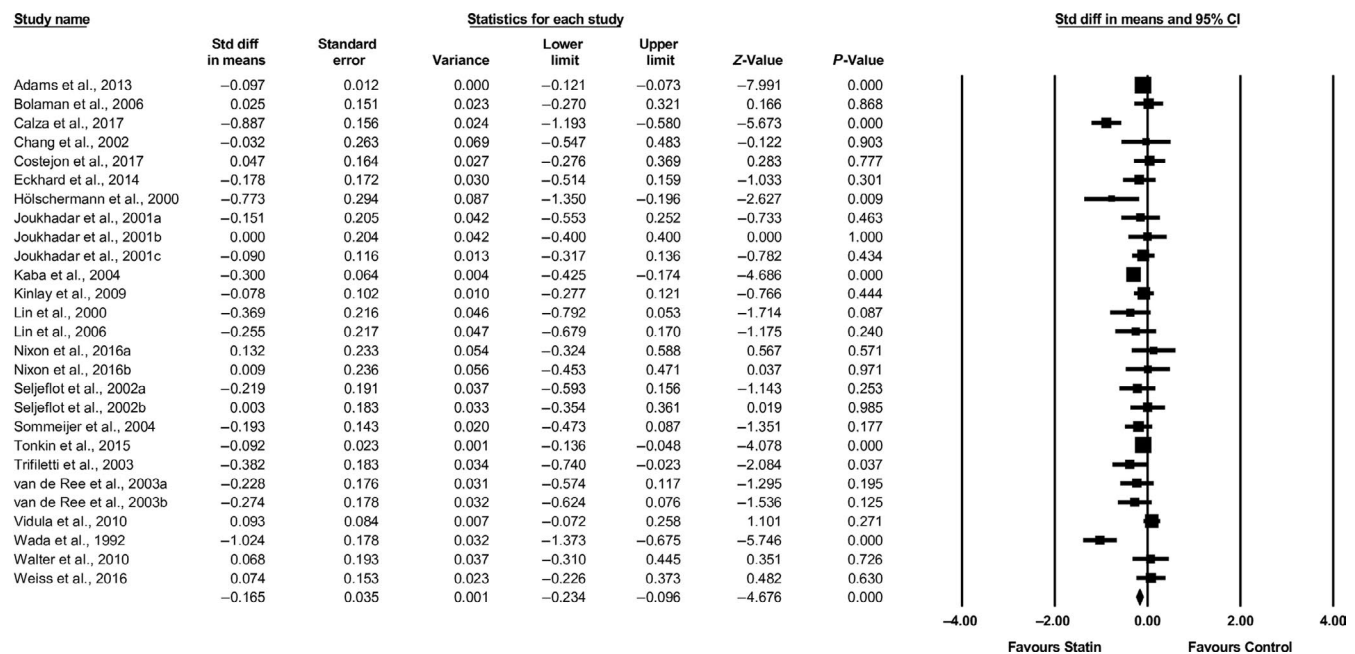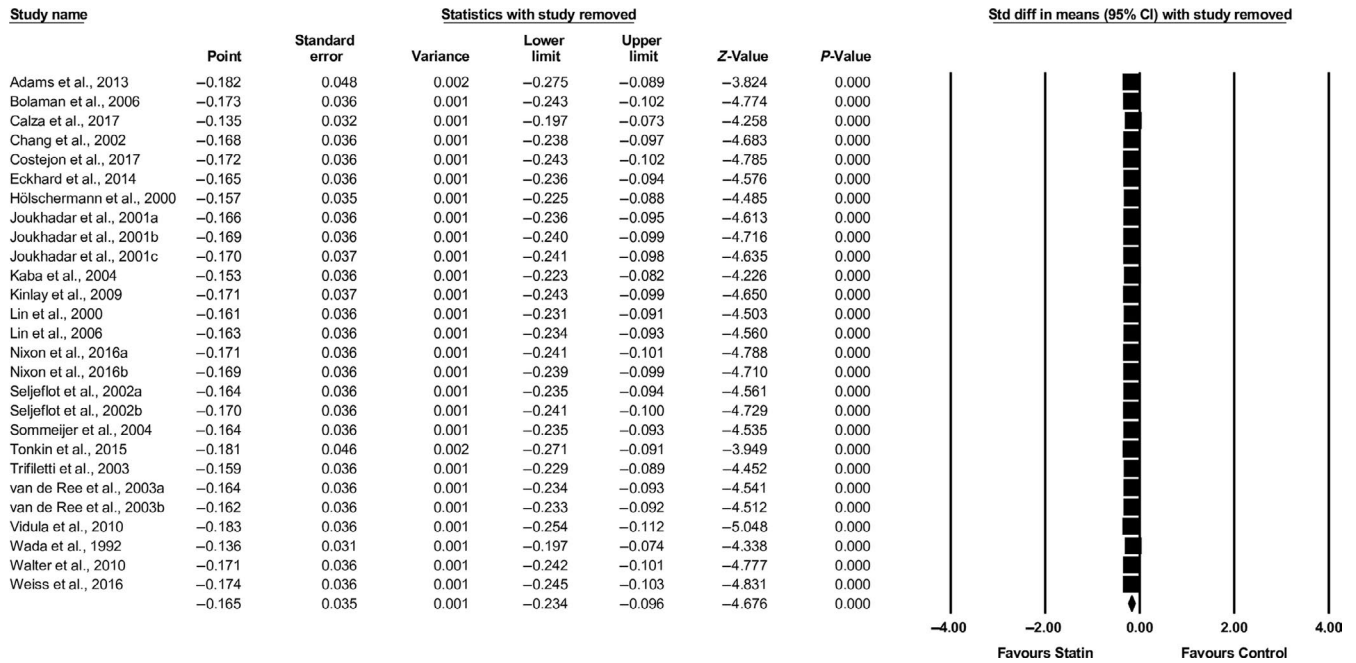| Study name | Point | Standard error | Variance | Lower limit | Upper limit | Z-Value | P-Value |
|---|---|---|---|---|---|---|---|
| Adams et al., 2013 | −0.182 | 0.048 | 0.002 | −0.275 | −0.089 | −3.824 | 0.000 |
| Bolaman et al., 2006 | −0.173 | 0.036 | 0.001 | −0.243 | −0.102 | −4.774 | 0.000 |
| Calza et al., 2017 | −0.135 | 0.032 | 0.001 | −0.197 | −0.073 | −4.258 | 0.000 |
| Chang et al., 2002 | −0.168 | 0.036 | 0.001 | −0.238 | −0.097 | −4.683 | 0.000 |
| Costejon et al., 2017 | −0.172 | 0.036 | 0.001 | −0.243 | −0.102 | −4.785 | 0.000 |
| Eckhard et al., 2014 | −0.165 | 0.036 | 0.001 | −0.236 | −0.094 | −4.576 | 0.000 |
| Hölschermann et al., 2000 | −0.157 | 0.035 | 0.001 | −0.225 | −0.088 | −4.485 | 0.000 |
| Joukhadar et al., 2001a | −0.166 | 0.036 | 0.001 | −0.236 | −0.095 | −4.613 | 0.000 |
| Joukhadar et al., 2001b | −0.169 | 0.036 | 0.001 | −0.240 | −0.099 | −4.716 | 0.000 |
| Joukhadar et al., 2001c | −0.170 | 0.037 | 0.001 | −0.241 | −0.098 | −4.635 | 0.000 |
| Kaba et al., 2004 | −0.153 | 0.036 | 0.001 | −0.223 | −0.082 | −4.226 | 0.000 |
| Kinlay et al., 2009 | −0.171 | 0.037 | 0.001 | −0.243 | −0.099 | −4.650 | 0.000 |
| Lin et al., 2000 | −0.161 | 0.036 | 0.001 | −0.231 | −0.091 | −4.503 | 0.000 |
| Lin et al., 2006 | −0.163 | 0.036 | 0.001 | −0.234 | −0.093 | −4.560 | 0.000 |
| Nixon et al., 2016a | −0.171 | 0.036 | 0.001 | −0.241 | −0.101 | −4.788 | 0.000 |
| Nixon et al., 2016b | −0.169 | 0.036 | 0.001 | −0.239 | −0.099 | −4.710 | 0.000 |
| Seljeflot et al., 2002a | −0.164 | 0.036 | 0.001 | −0.235 | −0.094 | −4.561 | 0.000 |
| Seljeflot et al., 2002b | −0.170 | 0.036 | 0.001 | −0.241 | −0.100 | −4.729 | 0.000 |
| Sommeijer et al., 2004 | −0.164 | 0.036 | 0.001 | −0.235 | −0.093 | −4.535 | 0.000 |
| Tonkin et al., 2015 | −0.181 | 0.046 | 0.002 | −0.271 | −0.091 | −3.949 | 0.000 |
| Trifiletti et al., 2003 | −0.159 | 0.036 | 0.001 | −0.229 | −0.089 | −4.452 | 0.000 |
| van de Ree et al., 2003a | −0.164 | 0.036 | 0.001 | −0.234 | −0.093 | −4.541 | 0.000 |
| van de Ree et al., 2003b | −0.162 | 0.036 | 0.001 | −0.233 | −0.092 | −4.512 | 0.000 |
| Vidula et al., 2010 | −0.183 | 0.036 | 0.001 | −0.254 | −0.112 | −5.048 | 0.000 |
| Wada et al., 1992 | −0.136 | 0.031 | 0.001 | −0.197 | −0.074 | −4.338 | 0.000 |
| Walter et al., 2010 | −0.171 | 0.036 | 0.001 | −0.242 | −0.101 | −4.777 | 0.000 |
| Weiss et al., 2016 | −0.174 | 0.036 | 0.001 | −0.245 | −0.103 | −4.831 | 0.000 |
| | −0.165 | 0.035 | 0.001 | −0.234 | −0.096 | −4.676 | 0.000 |



**FIGURE 4** Leave-one-out sensitivity analysis of the effect of statin therapy on D-dimer

| Study name | Std diff in means | Standard error | Variance | Lower limit | Upper limit | Z-Value | P-Value |
|---|---|---|---|---|---|---|---|
| Chang et al., 2002 | −0.032 | 0.263 | 0.069 | −0.547 | 0.483 | −0.122 | 0.903 |
| Costejon et al., 2017 | 0.047 | 0.164 | 0.027 | −0.276 | 0.369 | 0.283 | 0.777 |
| Hölschermann et al., 2000 | −0.773 | 0.294 | 0.087 | −1.350 | −0.196 | −2.627 | 0.009 |
| Sommeijer et al., 2004 | −0.193 | 0.143 | 0.020 | −0.473 | 0.087 | −1.351 | 0.177 |
| | −0.185 | 0.146 | 0.021 | −0.471 | 0.101 | −1.265 | 0.206 |

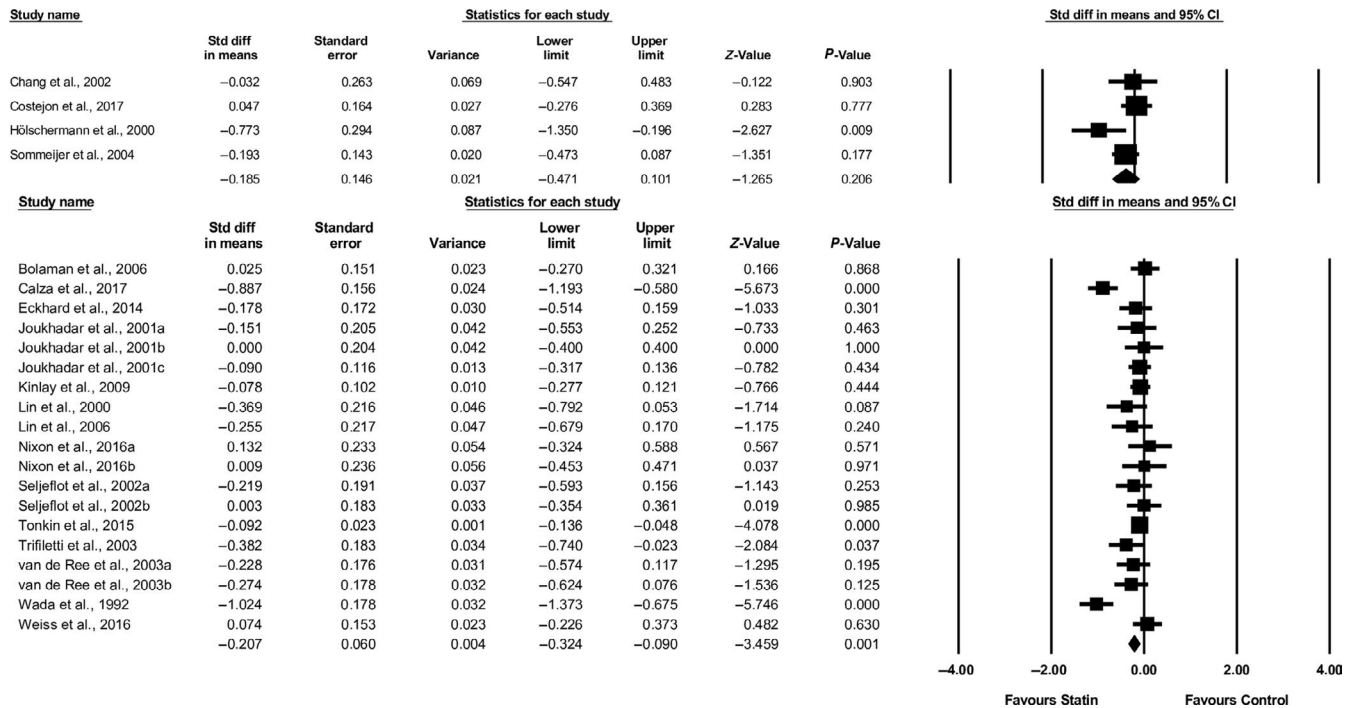| Study name | Std diff in means | Standard error | Variance | Lower limit | Upper limit | Z-Value | P-Value |
|---|---|---|---|---|---|---|---|
| Bolaman et al., 2006 | 0.025 | 0.151 | 0.023 | −0.270 | 0.321 | 0.166 | 0.868 |
| Calza et al., 2017 | −0.887 | 0.156 | 0.024 | −1.193 | −0.580 | −5.673 | 0.000 |
| Eckhard et al., 2014 | −0.178 | 0.172 | 0.030 | −0.514 | 0.159 | −1.033 | 0.301 |
| Joukhadar et al., 2001a | −0.151 | 0.205 | 0.042 | −0.553 | 0.252 | −0.733 | 0.463 |
| Joukhadar et al., 2001b | 0.000 | 0.204 | 0.042 | −0.400 | 0.400 | 0.000 | 1.000 |
| Joukhadar et al., 2001c | −0.090 | 0.116 | 0.013 | −0.317 | 0.136 | −0.782 | 0.434 |
| Kinlay et al., 2009 | −0.078 | 0.102 | 0.010 | −0.277 | 0.121 | −0.766 | 0.444 |
| Lin et al., 2000 | −0.369 | 0.216 | 0.046 | −0.792 | 0.053 | −1.714 | 0.087 |
| Lin et al., 2006 | −0.255 | 0.217 | 0.047 | −0.679 | 0.170 | −1.175 | 0.240 |
| Nixon et al., 2016a | 0.132 | 0.233 | 0.054 | −0.324 | 0.588 | 0.567 | 0.571 |
| Nixon et al., 2016b | 0.009 | 0.236 | 0.056 | −0.453 | 0.471 | 0.037 | 0.971 |
| Seljeflot et al., 2002a | −0.219 | 0.191 | 0.037 | −0.593 | 0.156 | −1.143 | 0.253 |
| Seljeflot et al., 2002b | 0.003 | 0.183 | 0.033 | −0.354 | 0.361 | 0.019 | 0.985 |
| Tonkin et al., 2015 | −0.092 | 0.023 | 0.001 | −0.136 | −0.048 | −4.078 | 0.000 |
| Trifiletti et al., 2003 | −0.382 | 0.183 | 0.034 | −0.740 | −0.023 | −2.084 | 0.037 |
| van de Ree et al., 2003a | −0.228 | 0.176 | 0.031 | −0.574 | 0.117 | −1.295 | 0.195 |
| van de Ree et al., 2003b | −0.274 | 0.178 | 0.032 | −0.624 | 0.076 | −1.536 | 0.125 |
| Wada et al., 1992 | −1.024 | 0.178 | 0.032 | −1.373 | −0.675 | −5.746 | 0.000 |
| Weiss et al., 2016 | 0.074 | 0.153 | 0.023 | −0.226 | 0.373 | 0.482 | 0.630 |
| | −0.207 | 0.060 | 0.004 | −0.324 | −0.090 | −3.459 | 0.001 |



**FIGURE 5** Forest plot of the effect of statin therapy on D-dimer with treatment durations of < 12 weeks (above) and > 12 weeks (below). Effect sizes were expressed as standardized mean difference (SMD) with its corresponding 95% confidence intervals (CIs) using Cohen's d as the summary statistic. A random-effects model was used for performance of the meta-analysis

meta-analysis by Sahebkar et al (2). First, in both studies effect sizes are expressed as standardized mean difference (SMD) using Cohen's d. However, Cohen's d is a dimensionless quantity, calculated as the ratio of the difference between the means of two samples and their pooled standard deviation. Thus, Cohen's d can be interpreted as a standardized difference.[64] Cohen's d was developed to compare effects across studies (even) when outcome variables vary, and results could be interpreted by referring to benchmarks with small (Cohen's d = 0.2), medium (0.5) and large (0.8)
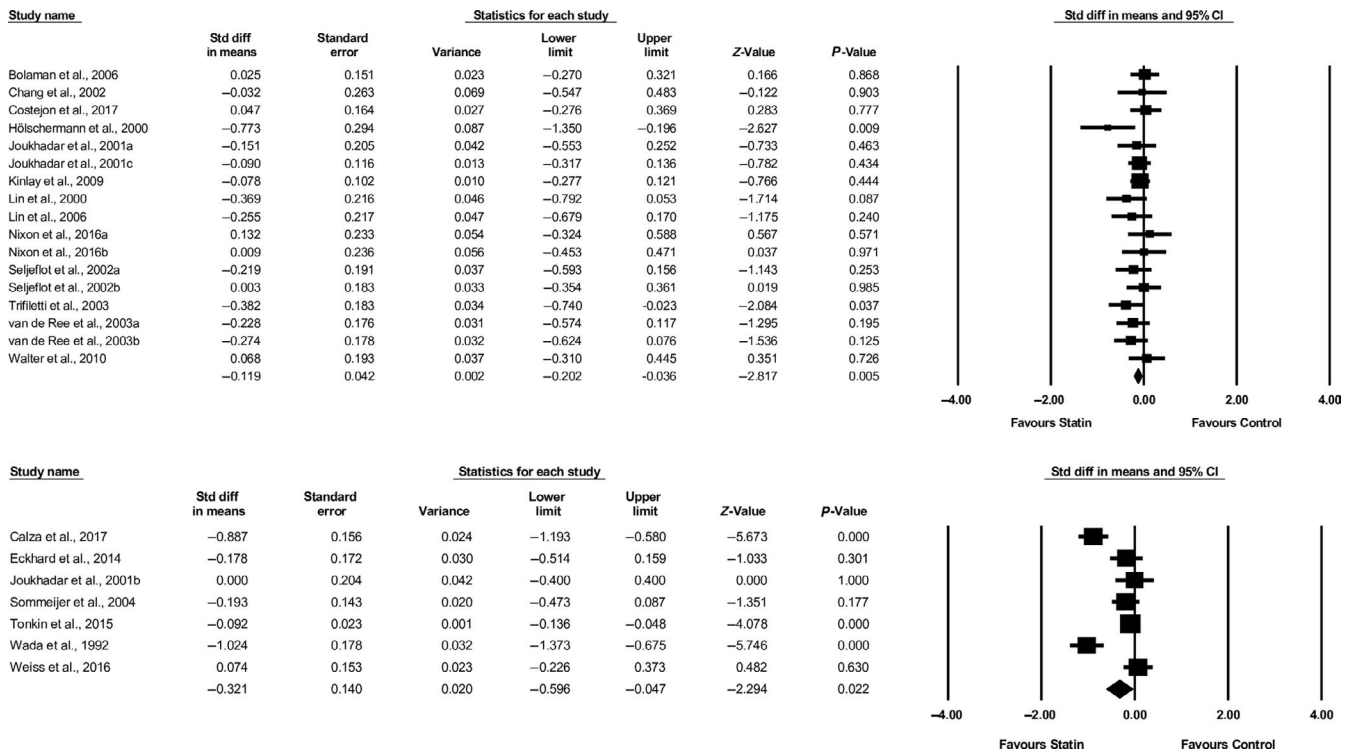
| Study name | Std diff in means | Standard error | Variance | Lower limit | Upper limit | Z-Value | P-Value |
|---|---|---|---|---|---|---|---|
| Bolaman et al., 2006 | 0.025 | 0.151 | 0.023 | −0.270 | 0.321 | 0.166 | 0.868 |
| Chang et al., 2002 | −0.032 | 0.263 | 0.069 | −0.547 | 0.483 | −0.122 | 0.903 |
| Costejon et al., 2017 | 0.047 | 0.164 | 0.027 | −0.276 | 0.369 | 0.283 | 0.777 |
| Hölschermann et al., 2000 | −0.773 | 0.294 | 0.087 | −1.350 | −0.196 | −2.627 | 0.009 |
| Joukhadar et al., 2001a | −0.151 | 0.205 | 0.042 | −0.553 | 0.252 | −0.733 | 0.463 |
| Joukhadar et al., 2001c | −0.090 | 0.116 | 0.013 | −0.317 | 0.136 | −0.782 | 0.434 |
| Kinlay et al., 2009 | −0.078 | 0.102 | 0.010 | −0.277 | 0.121 | −0.766 | 0.444 |
| Lin et al., 2000 | −0.369 | 0.216 | 0.046 | −0.792 | 0.053 | −1.714 | 0.087 |
| Lin et al., 2006 | −0.255 | 0.217 | 0.047 | −0.679 | 0.170 | −1.175 | 0.240 |
| Nixon et al., 2016a | 0.132 | 0.233 | 0.054 | −0.324 | 0.588 | 0.567 | 0.571 |
| Nixon et al., 2016b | 0.009 | 0.236 | 0.056 | −0.453 | 0.471 | 0.037 | 0.971 |
| Seljeflot et al., 2002a | −0.219 | 0.191 | 0.037 | −0.593 | 0.156 | −1.143 | 0.253 |
| Seljeflot et al., 2002b | 0.003 | 0.183 | 0.033 | −0.354 | 0.361 | 0.019 | 0.985 |
| Trifiletti et al., 2003 | −0.382 | 0.183 | 0.034 | −0.740 | −0.023 | −2.084 | 0.037 |
| van de Ree et al., 2003a | −0.228 | 0.176 | 0.031 | −0.574 | 0.117 | −1.295 | 0.195 |
| van de Ree et al., 2003b | −0.274 | 0.178 | 0.032 | −0.624 | 0.076 | −1.536 | 0.125 |
| Walter et al., 2010 | 0.068 | 0.193 | 0.037 | −0.310 | 0.445 | 0.351 | 0.726 |
| | −0.119 | 0.042 | 0.002 | −0.202 | −0.036 | −2.817 | 0.005 |

| Study name | Std diff in means | Standard error | Variance | Lower limit | Upper limit | Z-Value | P-Value |
|---|---|---|---|---|---|---|---|
| Calza et al., 2017 | −0.887 | 0.156 | 0.024 | −1.193 | −0.580 | −5.673 | 0.000 |
| Eckhard et al., 2014 | −0.178 | 0.172 | 0.030 | −0.514 | 0.159 | −1.033 | 0.301 |
| Joukhadar et al., 2001b | 0.000 | 0.204 | 0.042 | −0.400 | 0.400 | 0.000 | 1.000 |
| Sommeijer et al., 2004 | −0.193 | 0.143 | 0.020 | −0.473 | 0.087 | −1.351 | 0.177 |
| Tonkin et al., 2015 | −0.092 | 0.023 | 0.001 | −0.136 | −0.048 | −4.078 | 0.000 |
| Wada et al., 1992 | −1.024 | 0.178 | 0.032 | −1.373 | −0.675 | −5.746 | 0.000 |
| Weiss et al., 2016 | 0.074 | 0.153 | 0.023 | −0.226 | 0.373 | 0.482 | 0.630 |
| | −0.321 | 0.140 | 0.020 | −0.596 | −0.047 | −2.294 | 0.022 |

**FIGURE 6** Forest plot for the post hoc analysis on the effect of type of statin therapy on plasma D-dimer concentrations with lipophilic statins above and hydrophilic statins below. Effect sizes were expressed as standardized mean difference (SMD) with its corresponding 95% confidence intervals (CIs) using Cohen's d as the summary statistic. A random-effects model was used for performance of the meta-analysis
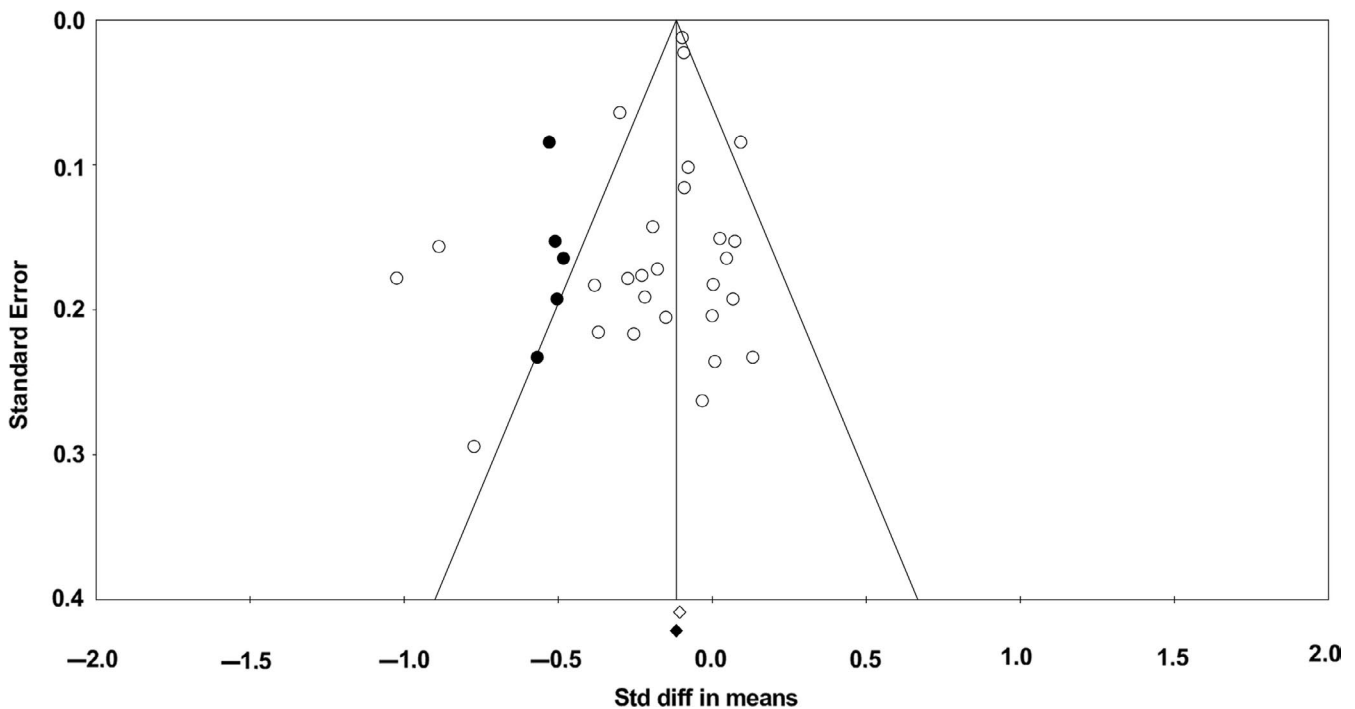


**FIGURE 7** Funnel plot representing publication bias within literature analysed with Duval and Tweedie's trim-and-fill method about the effect of statin therapy on D-dimer levels. Observed studies are shown as open circles, and imputed studies are shown as filled circles

effect sizes.[25,64] Effect sizes should also be set in clinical perspective, incorporating that small effects could have large implications in clinical settings. In the article by Sahebkar et al therefore, the overall effect of statins on the plasma D-dimer levels could have been interpreted as a large effect (d = −0.988), but not as a reduction of D-dimer levels by

0.988 µg/mL (which would be an extremely large effect). Second, in the meta-analysis by Sahebkar et al we found inconsistencies in data extracted from the incorporated studies (Table S2). In seven of the nine studies, differences in mean (standard deviation [SD]) D-dimer levels were reported incorrectly in Table 1 of their meta-analysis.[12] For example, in both studies of Sommeijer et al and Walter et al, D-dimer values after treatment were reported as D-dimer changes.[26,31] Third, in our meta-analysis we explained essential assumptions with respect to the interpretation of the original data. In the meta-analysis by Sahebkar et al on the other side, it remains unclear how exactly means or SDs were estimated if not reported in the study manuscripts. Because of concerns on the validity of the reported D-dimer results, due to inconsistent calculation of D-dimer changes, results of sensitivity analyses and unstandardized D-dimer measurement, one could argue about inclusion of the studies of Dangas et al, Min et al and Undas et al.[65-67] In our meta-analysis, we excluded these three studies.

The results of our meta-analysis should of course also be interpreted with caution. In this meta-analysis, we did not only include randomized controlled trials, but also cohort and cross-sectional studies. In the two latter types of studies, we scored the risk of bias to be high and heterogeneity between individual studies will be higher. The meta-analysis was not limited to randomized controlled trials only, because we would then have ignored a large number of observational evidence.[68] It is however important to note that within the group of cross-sectional studies, there are some differences in the retrieved data. The study of Adams adjusted results of D-dimer levels in statin users and nonusers for the following potential confounding factors: age, sex, education, individual income, race, smoking status, current alcohol use, body mass index, diabetes status, hypertension, use of acetylsalicylic acid and hormone therapy use among women.[46] On the other hand, Walter et al matched users of atorvastatin with controls according to their total cholesterol levels and Kaba et al and Vidula et al did not adjust D-dimer levels for any confounding factors.[28,31,35] However, age and sex, two of the most influencing confounding factors, were not significantly different among statin users and nonusers in these studies. Also, duration of statin treatment was not assessed in these cross-sectional data. The described between-study heterogeneity is unlikely to have had a large impact on the results of our meta-analysis. In the subanalyses of the 6 controlled trials with low risk of patient selection and the 16 studies with low risk of limited patient applicability, change in D-dimer levels was not significantly different from the overall effect with all studies included. Also, a separate subanalysis only including the controlled trials did not differ from these results and resulted in lower D-dimer values after statin treatment. Moreover, the post hoc analyses on treatment duration and statin type did not show a difference. Another concern might be that the included studies were heterogeneous

in the characteristics of study participants. Studies were performed in patients with proven cardiovascular disease, HIV infection, type 2 diabetes mellitus, lupus and COPD and in heart transplant patients. All these conditions could have influenced D-dimer levels. By running our meta-analysis with a random-effects model, we assumed the studies to be heterogeneous and our sensitivity analysis was robust. Furthermore, we could not fully exclude that publication bias has had an effect on the results of the meta-analysis. The adjusted effect size using the trim-and-fill method though was even larger than what we had observed, indicating that the effect size of reduction of D-dimer levels in statin users is more likely to be an underestimation rather than nonsignificant. Also, Begg's rank correlation and Egger's test were nonsignificant, indicating no publication bias and many missing studies (n = 422) would be needed and imputed in our meta-analysis to come to a nonsignificant effect.

In conclusion, in this meta-analysis use of statins was associated with a reduction of D-dimer levels, independent of treatment duration and type of statin used. This antithrombotic effect is part of the "pleiotropic" effects of statins and contributes to the benefits of statins on cardiovascular outcomes. The reduction of D-dimer levels in statin users may affect the performance of diagnostic algorithms on suspected VTE in this specific patient group, and prospective studies investigating the impact of statin use on these diagnostic algorithms are recommended.

## CONFLICT OF INTEREST
None of the authors reports a conflict of interest with regard to this manuscript.

## AUTHOR CONTRIBUTIONS
SS-G, MK and TvG designed the study. SS-G and FM selected the articles and managed the study with support and input from all other authors. SS-G, JV, HB and LRA verified the data, which was analysed by LRA and interpreted by all other authors. SS-G wrote the first draft of the manuscript, which was reviewed, modified and approved by all other authors.

**ORCID**

*Suzanne Schol-Gelok* (iD) https://orcid.org/0000-0003-3897-5113

*Jorie Versmissen* (iD) https://orcid.org/0000-0003-0674-7765

**REFERENCES**

1. Adam SS, Key NS, Greenberg CS. D-dimer antigen: current concepts and future prospects. *Blood*. 2009;113:2878-2887.

2. Weitz JI, Fredenburgh JC, Eikelboom JW. A test in context: D-dimer. *J Am Coll Cardiol*. 2017;70:2411-2420.

3. Douma R, van Sluis G, Kamphuisen P, et al. Clinical decision rule and D-dimer have lower clinical utility to exclude pulmonary embolism in cancer patients. Explanations and potential ameliorations. *Thromb Haemost*. 2010;104:831-836.

4. Harb TS, Zareba W, Moss AJ, et al. Association between inflammatory markers, hemostatic, and lipid factors in postinfarction patients. *Am J Cardiol*. 2003;91:1120-1123.

5. Chabloz P, Reber G, Boehlen F, Hohlfeld P, de Moerloose P. TAFI antigen and D-dimer levels during normal pregnancy and at delivery. *Br J Haematol*. 2001;115:150-152.

6. Crop MJ, Siemes C, Berendes P, van der Straaten F, Willemsen S, Levin MD. Influence of C-reactive protein levels and age on the value of D-dimer in diagnosing pulmonary embolism. *Eur J Haematol*. 2014;92:147-155.

7. Kunutsor SK, Seidu S, Khunti K. Statins and primary prevention of venous thromboembolism: a systematic review and meta-analysis. *Lancet Haematol*. 2017;4:e83-e93.

8. Violi F, Calvieri C, Ferro D, Pignatelli P. Statins as antithrombotic drugs. *Circulation*. 2013;127:251-257.

9. Bonetti PO, Lerman LO, Napoli C, Lerman A. Statin effects beyond lipid lowering–are they clinically relevant? *Eur Heart J*. 2003;24:225-248.

10. Konstantinides SV, Torbicki A, Agnelli G, et al. 2014 ESC guidelines on the diagnosis and management of acute pulmonary embolism. *Eur Heart J*. 2014;35(3033–69):69a–69k.

11. Raja AS, Greenberg JO, Qaseem A, et al. Evaluation of patients with suspected acute pulmonary embolism: best practice advice from the Clinical Guidelines Committee of the American College of Physicians. *Ann Intern Med*. 2015;163:701-711.

12. Sahebkar A, Serban C, Mikhailidis D, et al. Association between statin use and plasma d-dimer levels: a systematic review and meta-analysis of randomised controlled trials. *Thromb Haemost*. 2015;114:546-557.

13. Moher D, Liberati A, Tetzlaff J, Altman DG, PRISMA Group. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. *Ann Intern Med*. 2009;151(49):264–269.

14. Cilla DD Jr, Whitfield LR, Gibson DM, Sedman AJ, Posvar EL. Multiple-dose pharmacokinetics, pharmacodynamics, and safety of atorvastatin, an inhibitor of HMG-CoA reductase, in healthy subjects. *Clin Pharmacol Ther*. 1996;60:687–695.

15. Schachter M. Chemical, pharmacokinetic and pharmacodynamic properties of statins: an update. *Fundam Clin Pharmacol*. 2005;19:117–125.

16. Konstantinides SV. 2014 ESC Guidelines on the diagnosis and management of acute pulmonary embolism. *Eur Heart J*. 2014;35:3145–3146.

17. Whiting PF, Rutjes AW, Westwood ME, et al. QUADAS-2: a revised tool for the quality assessment of diagnostic accuracy studies. *Ann Intern Med*. 2011;155:529–536.

18. Cooper H, Hedges LV. *The Handbook of Research Synthesis*. New York, NY: Russell Sage Foundation; 1994.

19. Duval S, Tweedie R. Trim and fill: A simple funnel-plot-based method of testing and adjusting for publication bias in meta-analysis. *Biometrics*. 2000;56:455–463.

20. Egger M, Davey Smith G, Schneider M, Minder C. Bias in meta-analysis detected by a simple, graphical test. *BMJ*. 1997;315:629–634.

21. Begg CB, Mazumdar M. Operating characteristics of a rank correlation test for publication bias. *Biometrics*. 1994;50:1088–1101.

22. Rosenthal R. The file drawer problem and tolerance for null results. *Psychol Bull*. 1979;86:638–641.

23. Wan X, Wang W, Liu J, Tong T. Estimating the sample mean and standard deviation from the sample size, median, range and/or interquartile range. *BMC Med Res Methodol*. 2014;14:135.

24. Nolan SJ, Hambleton I, Dwan K. The use and reporting of the cross-over study design in clinical trials and systematic reviews: a systematic assessment. *PLoS ONE*. 2016;11:e0159014.

25. Lakens D. Calculating and reporting effect sizes to facilitate cumulative science: a practical primer for t-tests and ANOVAs. *Front Psychol*. 2013;4:863.

26. Sommeijer DW, MacGillavry MR, Meijers J, Van Zanten AP, Reitsma PH, Ten Cate H. Anti-inflammatory and anticoagulant effects of pravastatin in patients with type 2 diabetes. *Diabetes Care*. 2004;27:468–473.

27. Tonkin AM, Blankenberg S, Kirby A, et al. Biomarkers in sae coronary heart disease, their modulation and cardiovascular risk: The LIPID biomarker study. *Int J Cardiol*. 2015;201:499–507.

28. Vidula H, Tian LU, Liu K, et al. Comparison of effects of statin use on mortality in patients with peripheral arterial disease with versus without elevated C-reactive protein and D-dimer levels. *Am J Cardiol*. 2010;105:1348–1352.

29. Van De Ree MA, De Maat MPM, Kluft C, Meinders AE, Princen HMG, Huisman MV. Princen HM and Huisman MV. Decrease of hemostatic cardiovascular risk factors by aggressive vs. conventional atorvastatin treatment in patients with Type 2 diabetes mellitus. *J Thromb Haemost*. 2003;1:1753–1757.

30. Eckard AR, Jiang Y, Debanne SM, Funderburg NT, McComsey GA. Effect of 24 weeks of statin therapy on systemic and vascular inflammation in HIV-infected subjects receiving antiretroviral therapy. *J Infect Dis*. 2014;209:1156–1164.

31. Walter T, Szabo S, Suselbeck T, et al. Effect of atorvastatin on haemostasis, fibrinolysis and inflammation in normocholesterolaemic patients with coronary artery disease: A post hoc analysis of data from a prospective, randomized, double-blind study. *Clin Drug Invest*. 2010;30:453–460.

32. Lin TH, Huang CH, Voon WC, et al. The effect of fluvastatin on fibrinolytic factors in patients with hypercholesterolemia. *Kaohsiung J Med Sci*. 2000;16:600–606.

33. Nixon DE, Bosch RJ, Chan ES, et al. Effects of atorvastatin on biomarkers of immune activation, inflammation, and lipids in virologically suppressed, human immunodeficiency virus-1-infected individuals with low-density lipoprotein cholesterol <130 mg/dL (AIDS Clinical Trials Group Study A5275). *J Clin Lipidol*. 2016;11(1):61–69.

34. Bolaman Z, Kadikoylu G, Özgel N, Yenisey C. Effects of atorvastatin on coagulation parameters and homocysteine in patients with primary hypercholesterolemia. *J Natl Med Assoc*. 2006;98:1273–1277.

35. Kaba NK, Francis CW, Moss AJ, et al. Effects of lipids and lipid-lowering therapy on hemostatic factors in patients with myocardial infarction. *J Thromb Haemost*. 2004;2:718–725.

36. Chang JW, Yang WS, Min WK, Lee SK, Park JS, Kim SB. Effects of simvastatin on high-sensitivity C-reactive protein and serum albumin in hemodialysis patients. *Am J Kidney Dis*. 2002;39:1213–1217.

37. Kinlay S, Schwartz GG, Olsson AG, et al. Endogenous tissue plasminogen activator and risk of recurrent cardiac events after an acute coronary syndrome in the MIRACL study. *Atherosclerosis*. 2009;206:551–555.

38. Wada H, Mori Y, Kaneko T, et al. Hypercoagulable state in patients with hypercholesterolemia: Effects of pravastatin. *Clin Ther*. 1992;14:829–834.

39. Seljeflot I, Tonstad S, Hjermann I, Arnesen H. Improved fibrinolysis after 1-year treatment with HMG CoA reductase inhibitors in patients with coronary heart disease. *Thromb Res*. 2002;105:285–290.

40. Trifiletti A, Lasco A, Scamardi R, et al. Long-term hemostatic effects of cholesterol-lowering therapy with atorvastatin. *Pathophysiol Haemost Thromb*. 2003;33:84–87.

41. Lin T-H, Voon W-C, Yen H-W, et al. Randomized comparative study of the effects of treatment with once-daily, niacin extended-release/lovastatin and with simvastatin on lipid profile and fibrinolytic parameters in Taiwan. *Kaohsiung J Med Sci*. 2006;22:257–265.

42. Weiss L, Chevalier MF, Assoumou L, et al. Rosuvastatin is effective to decrease CD8 T-cell activation only in HIV-infected patients with high residual T-cell activation under antiretroviral therapy. *J Acquired Immune Defic Syndr*. 2016;71:390–398.

43. Calza L, Colangeli V, Magistrelli E, et al. Significant decrease in plasma levels of D-dimer, interleukin-8, and interleukin-12 after a 12-month treatment with rosuvastatin in HIV-infected patients under antiretroviral therapy. *AIDS Res Hum Retroviruses*. 2017;33:126–132.

44. Joukhadar C, Klein N, Prinz M, et al. Similar effects of atorvastatin, simvastatin and pravastatin on thrombogenic and inflammatory parameters in patients with hypercholesterolemia. *Thromb Haemost*. 2001;85:47–51.

45. Hölschermann H, Hilgendorff A, Kemkes-Matthes B, et al. Simvastatin attenuates vascular hypercoagulability in cardiac transplant recipients. *Transplantation*. 2000;69:1830–1836.

46. Adams NB, Lutsey PL, Folsom AR, et al. Statin therapy and levels of hemostatic factors in a healthy population: The Multi-Ethnic study of atherosclerosis. *J Thromb Haemost*. 2013;11:1078–1084.

47. Castejon R, Castañeda A, Sollet A, et al. Short-term atorvastatin therapy improves arterial stiffness of middle-aged systemic lupus erythematosus patients with pathological pulse wave velocity. *Lupus*. 2017;26:355–364.

48. Glynn RJ, Danielson E, Fonseca FAH, et al. A randomized trial of rosuvastatin in the prevention of venous thromboembolism. *N Engl J Med*. 2009;360:1851–1861.

49. Pignatelli P, Carnevale R, Pastori D, et al. Immediate antioxidant and antiplatelet effect of atorvastatin via inhibition of Nox2. *Circulation*. 2012;126:92–103.

50. Tannous M, Cheung R, Vignini A, Mutus B. Atorvastatin increases ecNOS levels in human platelets of hyperlipidemic subjects. *Thromb Haemost*. 1999;82:1390–1394.

51. Sommeijer DW, Joop K, Leyte A, Reitsma PH, ten Cate H. Pravastatin reduces fibrinogen receptor gpIIIa on platelet-derived microparticles in patients with type 2 diabetes. *J Thromb Haemost*. 2005;3:1168–1171.

52. Colli S, Eligini S, Lalli M, Camera M, Paoletti R, Tremoli E. Vastatins inhibit tissue factor in cultured human macrophages. A novel mechanism of protection against atherothrombosis. *Arterioscler Thromb Vasc Biol*. 1997;17:265–272.

53. Owens AP 3rd, Mackman N. The antithrombotic effects of statins. *Annu Rev Med*. 2014;65:433–445.

54. Undas A, Brummel-Ziedins KE, Mann KG. Anticoagulant effects of statins and their clinical implications. *Thromb Haemost*. 2014;111:392–400.

55. Oesterle A, Laufs U, Liao JK. Pleiotropic effects of statins on the cardiovascular system. *Circ Res*. 2017;120:229–243.

56. Bourcier T, Libby P. HMG CoA reductase inhibitors reduce plasminogen activator inhibitor-1 expression by human vascular smooth muscle and endothelial cells. *Arterioscler Thromb Vasc Biol*. 2000;20:556–562.

57. Essig M, Nguyen Geneviève, Prié D, Escoubet B, Sraer J-D, Friedlander Gérard. 3-Hydroxy-3-methylglutaryl coenzyme A reductase inhibitors increase fibrinolytic activity in rat aortic endothelial cells. Role of geranylgeranylation and Rho proteins. *Circ Res*. 1998;83:683–690.

58. van Vliet AK, van Thiel GC, Huisman RH, Moshage H, Yap SH, Cohen LH. Different effects of 3-hydroxy-3-methylglutaryl-coenzyme A reductase inhibitors on sterol synthesis in various human cell types. *Biochim Biophys Acta*. 1995;1254:105–111.

59. Law MR, Wald NJ, Rudnicka AR. Quantifying effect of statins on low density lipoprotein cholesterol, ischaemic heart disease, and stroke: systematic review and meta-analysis. *BMJ*. 2003;326:1423.

60. Douma RA, Mos IC, Erkens PM, et al. Performance of 4 clinical decision rules in the diagnostic management of acute pulmonary embolism: a prospective cohort study. *Ann Intern Med*. 2011;154:709–718.

61. van der Hulle T, Cheung WY, Kooij S, et al. Simplified diagnostic management of suspected pulmonary embolism (the YEARS study): a prospective, multicentre, cohort study. *Lancet*. 2017;390:289–297.

62. van Es N, Kraaijpoel N, Klok FA, et al. The original and simplified Wells rules and age-adjusted D-dimer testing to rule out pulmonary embolism: an individual patient data meta-analysis. *J Thromb Haemost*. 2017;15:678–684.

63. Schol-Gelok S, van der Hulle T, Biedermann JS, et al. Clinical effects of antiplatelet drugs and statins on D-dimer levels. *Eur J Clin Invest*. 2018;48:e12944.

64. Cohen J. *Statistical Power Analysis for the Behavioral Sciences*. Hillsdale, NJ: Lawrence Erlbaum Associates; 1988.

65. Dangas G, Badimon JJ, Smith DA, et al. Pravastatin therapy in hyperlipidemia: effects on thrombus formation and the systemic hemostatic profile. *J Am Coll Cardiol*. 1999;33:1294–1304.

66. Min L, Shao S, Wu X, et al. Anti-inflammatory and anti-thrombogenic effects of atorvastatin in acute ischemic stroke. *Neural Regen Res*. 2013;8:2144–2154.

67. Undas A, Kaczmarek P, Sladek K, et al. Fibrin clot properties are altered in patients with chronic obstructive pulmonary disease:

Beneficial effects of simvastatin treatment. *Thromb Haemost*. 2009;102:1176–1182.

68. Sutton AJ, Abrams KR. Bayesian methods in meta-analysis and evidence synthesis. *Stat Methods Med Res*. 2001;10:277–303.

## SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of the article.