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Snoep, J.D.

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ASSOCIATION OF LABORATORY- DEFINED ASPIRIN RESISTANCE WITH A HIGHER RISK OF RECURRENT CARDIOVASCULAR EVENTS: A SYSTEMATIC REVIEW AND META-ANALYSIS

J.D. Snoep, M.M.C. Hovens, J.C.J. Eikenboom,
J.G. van der Bom, M.V. Huisman

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ABSTRACT

Background

The risk of recurrence of cardiovascular events among patients using aspirin (acetylsalicylic acid) for secondary prevention of such events remains high. Persistent platelet reactivity despite aspirin therapy, a laboratory-defined phenomenon called aspirin resistance (hereinafter, laboratory aspirin resistance), might explain this in part, but its actual contribution to the risk remains unclear. The objective of this study was to systematically review all available evidence on whether laboratory aspirin resistance is related to a higher risk of recurrent cardiovascular events.

Methods

Using a predefined search strategy, we searched electronic databases. To be included in our analysis, articles had to report on patients using aspirin for secondary cardiovascular prevention, had to contain a clear description of a method to establish the effects of aspirin on platelet reactivity, and had to report recurrence rates of cardiovascular events. Odds ratios of cardiovascular outcome of eligible studies were pooled in a random-effects model.

Results

We included 15 full-text articles and 1 meeting abstract. Fifteen of these studies revealed an adverse relation between laboratory aspirin resistance and occurrence of cardiovascular events. The pooled odds ratio of all cardiovascular outcomes was 3.8 (95% confidence interval 2.3 to 6.1) for laboratory aspirin resistance.

Conclusions

This systematic review and meta-analysis shows that patients biochemically identified as having laboratory aspirin resistance are more likely to also have “clinically resistance” because they exhibit significantly higher risks of recurrent cardiovascular events than patients who are identified as (laboratory) aspirin sensitive.

INTRODUCTION

Cardiovascular diseases are the most common cause of mortality and morbidity in Western countries in the twenty-first century. In the United States, cardiovascular mortality contributed to nearly 40% of total mortality in 2003.¹ Because aggregation of platelets is pivotal to the development of cardiovascular events, inhibition of this process could play an important role in prevention of cardiovascular disease.²

Nowadays, aspirin (acetylsalicylic acid) forms the cornerstone in the secondary prevention of cardiovascular events. The effect of low-dose aspirin is most likely based on the permanent inactivation of cyclooxygenase-1 (COX-1) through blockade of the COX-1-channel by the acetylation of serine residue 529, which results in an irreversible inhibition of the production of thromboxane A_2 by platelets.³ Because thromboxane A_2 is a potent platelet activator that also causes vasoconstriction and smooth muscle proliferation, a decrease in thromboxane A_2 leads to reduced aggregation of platelets.^{3,4}

The clinical effectiveness of aspirin in the secondary prevention of cardiovascular events has been well established. The Antithrombotic Trialists' Collaboration documented a 22% reduction in death and serious ischemic vascular events by using antiplatelet therapy compared with placebo, in their most recent meta-analysis of 287 randomized trials, which comprised more than 200,000 patients.⁵

However, not all patients benefit to the same extent, which could be explained by a variety of pharmacodynamic, pharmacokinetic and biochemical features.⁶ Addressed biochemically as persistent platelet reactivity *ex vivo* despite the use of aspirin, this phenomenon is called laboratory-defined aspirin resistance (hereinafter, laboratory aspirin resistance). Based on the failure of aspirin to inhibit platelet thromboxane A_2 production or to inhibit tests of platelet function, a variety of laboratory tests to define and quantify aspirin resistance has been proposed. Yet, a uniform and agreed-on definition of laboratory aspirin resistance and its measurement is lacking.⁷⁻⁹ Laboratory aspirin resistance has received much attention, in medical journals^{8,10,11} as well as in the lay media.¹²

A recent meta-analysis of studies addressing the prevalence of persistent platelet reactivity despite use of aspirin in a secondary cardiovascular prevention setting reported a mean prevalence of laboratory aspirin resistance of approximately 25%.¹³ However, the main question – whether patients who are biochemically identified as having laboratory aspirin resistance also exhibit “clinical resistance” to aspirin (i.e., whether they are at a higher risk of recurrent cardiovascular events) – remains largely unanswered hitherto. To try to quantify evidence addressing this topic, we conducted a systematic review and meta-analysis of all reports, to our knowledge, on the clinical consequences of laboratory aspirin resistance among patients using aspirin for secondary prevention of cardiovascular events.

METHODS

Study selection, quality assessment and data extraction

We used electronic databases to identify relevant reports. The following databases were searched: MEDLINE (January 1966 to October 2006), EMBASE (January 1974 to October 2006), the Cochrane Central Register of Controlled trials (CENTRAL) (1800 to October 2006) and Web of Science (1945 to October 2006). We used predefined search terms (available from the authors) and used no language restrictions. Furthermore, we tried to identify additional studies by searching the reference lists of relevant studies and reading reviews, editorials, and letters on this topic. Authors of identified appropriate studies were contacted to obtain additional data not reported in the original article. Both full-text articles and meeting abstracts were included.

To be included in the analysis, selected studies had to meet all of the following inclusion criteria: 1) included patients had established coronary artery, cerebrovascular or peripheral artery disease; 2) patients were treated with aspirin for secondary prevention of cardiovascular events; 3) the study contained a clear description of the method used to establish the effects of aspirin on platelet reactivity to compare patients with laboratory aspirin resistance with those without; and 4) the study reported data on recurrence rates of fatal and non-fatal myocardial infarction, fatal and non-fatal stroke or other cardiovascular endpoints as predefined by investigators. For this systematic review, we defined laboratory aspirin resistance as ex vivo non-responsiveness to aspirin according to any test that reflects platelet thromboxane A₂ synthesis or platelet function.

The quality of the identified studies was assessed based largely on quality criteria concerning minimization of bias. In detail, we evaluated information regarding control for confounders, measurement of exposure, completeness of follow-up, and blinding. For case-control studies, we also assessed matching and case definition. No formal scoring system was used. Reviewers were not blinded to journal, author or institution of publication.

We used a prespecified data collection form to extract information for each report regarding year of publication, duration and setting of study, study design, total sample size and study population (baseline characteristics). Concerning our research question, the following variables were collected from each selected study: the dosage of aspirin used, definition of laboratory aspirin resistance and cardiovascular outcomes used, prevalence of laboratory aspirin resistance, and occurrence rates of cardiovascular outcomes.

Selection, quality assessment, and data extraction of studies to be included in this review were all independently performed by two reviewers (JDS and MMCH). Disagreements were resolved by consensus and discussion with a third party (MVH). The κ statistic for agreement between reviewers was performed manually for each process in study selection. The overall κ statistic was calculated as a weighted mean of those different values.

Statistical analyses

To relate laboratory aspirin resistance to clinical outcomes, we calculated odds ratios (ORs) with corresponding 95% confidence intervals (CIs) for each study that reported the proportions of patients with laboratory aspirin resistance with cardiovascular events versus those without laboratory aspirin resistance with cardiovascular events. P-values are calculated with the χ^2 -test or Fisher exact test where appropriate. The ORs from cohort studies were pooled using a random-effects model.¹⁴ This rather conservative method for meta-analysis accounts for the possibility of statistical interstudy heterogeneity. To test for statistical interstudy heterogeneity, the χ^2 -value was calculated for the hypothesis of homogeneity. Quantification of the effect of heterogeneity was assessed by means of I^2 , which demonstrates the percentage of total variation across studies owing to heterogeneity.

We pooled all cohort studies reporting cardiovascular outcomes, as well as several subgroups of cohort studies. These subgroups included studies reporting clinical cardiovascular outcomes as cardiovascular death, myocardial infarction, stroke, acute coronary syndrome and revascularization; studies reporting on (re)occlusion after bypass grafting or angioplasty; and studies providing data on occurrence of myonecrosis represented by creatine kinase-myocardial band elevation after percutaneous coronary intervention. We assessed potential publication bias graphically, using funnel plots on ORs for laboratory aspirin resistance.

Analyses were performed using Cochrane Review Manager Software (version 4.2.8; Cochrane Library Software, Oxford, England). For all analyses, a level of significance of $\alpha=0.05$ was used.

RESULTS

Characteristics of included studies

We included 15 full-text articles¹⁵⁻²⁹ and 1 meeting abstract³⁰ (Figure 1). Overall, the κ was 0.86, indicating good interobserver agreement. Details of included studies are summarized in Table 1 and Table 2. Studies are grouped according to outcomes used. Ten studies used a composite endpoint of clinical cardiovascular events.^{15-23,30} In four reports, the studied outcome was (re)occlusion after bypass grafting or angioplasty.²⁴⁻²⁷ Two studies assessed myonecrosis, defined by elevated creatine kinase-myocardial band levels, after percutaneous coronary intervention.^{28,29}

Aspirin dosages used varied from 80 to 1500 mg daily,^{15,28,30} although nearly all studies used a low to intermediate dosage between 80 and 325 mg daily.¹⁶⁻³⁰ Various methods were used to establish the effects of aspirin on platelet reactivity. Conventional optical light transmittance aggregometry was used in five studies.^{20,23,24,27,29} Multiple agonists were used to induce aggregation. Three studies determined thromboxane B₂, which is a stable metabolite of thromboxane A₂, in plasma or urine.^{18,21,27} Five studies used the Platelet Function Analyzer 100 system (Dade Behring, Deerfield, IL, USA), which measures in vitro shear-stress-induced platelet activation in terms of platelet occlusion of a membrane coated with platelet agonists.^{17,19,22,25,26} In three

Table 1 – Details of included studies

Source	Design	Study population (n)	Aspirin dose, mg/day	Method of assessment of LAR
Grotemeyer et al., 993 ¹⁵	Prospective cohort	Stroke (180)	1500	Platelet reactivity index >1.25, using a technique reflecting platelet activation following blood sampling ³⁶
Buchanan et al., 2000 ¹⁶	Prospective cohort	CABG (289)	325	Variation coefficient bleeding time <26% with or without aspirin
Andersen et al., 2002 ¹⁷	Prospective cohort	CAD (71)	160	PFA-100 CEPI-CT ≤196s
Eikelboom et al., 002 ¹⁸	Nested case-control of HOPE study ^{37,38}	CV disease (488 cases) 488 controls)	NR	Urinary TxB ₂ : 4 th quartile (most platelet activation)
Grundmann et al., 003 ¹⁹	Case-control	Stroke (35 cases) 18 controls)	100	PFA-100 CEPI-CT ≤165s
Gum et al., 2003 ²⁰	Prospective cohort	CV disease (326)	325	LTA ≥70% (10 μmol/L ADP) and ≥20% (0.5 mg/mL AA)
Cotter et al., 2004 ²¹	Prospective cohort	CAD (73)	100	Plasma-TxB ₂ > lowest value found in aspirin non-users
Cheng et al., 2005 ³⁰ (abstract)	Prospective cohort	CAD (422)	80-300	VerifyNow ARU ≥550
Pamukcu et al., 2006 ²²	Prospective cohort	CAD (105)	100-300	PFA-100 CEPI-CT <186s
Stejskal et al., 2006 ²³	Prospective cohort	CAD (103)	100	LTA ≥5% (spontaneous) or ≥53% (3μmol/L cationic propyl gallate)
Mueller et al., 1997 ²⁴	Prospective cohort	PAD/PTA ^a (100)	100	LTA (10 and 5 μmol/L ADP and 5 and 2 μg/mL collagen), on average >80% of baseline
Ziegler et al., 2002 ²⁵	Prospective cohort	PAD/PTA ^a (52)	100	PFA-100 CEPI-CT ≤170s
Yilmaz et al., 2005 ²⁶	Case-control	CABG (14 cases) 14 controls)	Cases: 189 ± 100, Controls: 214 ± 90	PFA-100 CEPI-CT ≤193s
Poston et al., 2006 ²⁷	Prospective cohort	CABG (225)	325	Meets 2 of 3 criteria: TEG (0.5 μmol/L AA) >50%, LTA (1 and 5μg/mL collagen) >50%, Plasma-TxB ₂ >25% of baseline
Chen et al., 2004 ²⁸	Prospective cohort	PCI (151)	80-300	VerifyNow ARU ≥ 550
Lev et al., 2006 ²⁹	Prospective cohort	PCI (150)	81-325	Meets 2 of 3 criteria: LTA ≥70% (10 μmol/L ADP), LTA ≥20% (0.5 mg/mL AA), RPFA ARU ≥550

Footnotes on subsequent page ▽

Outcome	Duration follow-up	Comments
CV death, MI, stroke	2 y	Very heterogeneous distribution of withdrawals, LAR determined once, adh NA, adj OCs unblinded
Death, MI, stroke, graft occlusion	2 y	Bleeding time poorly established for this goal, low event rates, LAR determined once
Non-fatal MI, stroke, revascularization	4 y	Small groups, no exclusion criteria (confounding), LAR determined once, adh NA, adj OCs unblinded
CV death, MI, stroke	5 y	Confounders cases/controls: DM, BMI, tension, peripheral artery disease, TxB ₂ could be influenced by recent events, LAR determined once, adh NA
Stroke, transient ischemic attack	> 2 y	Small sample size, LAR cause or result of events? LAR determined once, adh NA
CV death, MI, stroke	679 ± 137 d	Few patients with LAR, few events, follow-up time not specified for aspirin response, LAR determined once, adh NA
CV death, MI, stroke, CV-related admission	1 y	Small groups, no exclusion criteria (confounding), LAR determined once
Death, MI, stroke, admission for UA	Not reported	Follow-up time and absolute event rates NR, LAR determined once, adh NA, adj OCs unblinded
CV death, MI, stroke, UA	1 y	Subjective OC (UA) LAR determined once, adh NA, adj OCs unblinded
MI, stroke, UA	4 y	Subjective OC (UA) LAR determined once, adh NA, adj OCs unblinded
Reocclusion	1.5 y	Reasons for exclusion NR, adj OCs unblinded
Restenosis, reocclusion	1 y	Small sample size, few non-responders, LAR determined once, adh NA, adj OCs unblinded
Graft occlusion	Cases: 7.5 ± 3.9 y, Controls: 6.2 ± 2.5 y	Most cases had ACS at presentation vs. stable angina in controls subjects, LAR determined once, adh NA
Graft occlusion	30 d	Very low event rates, adh NA
Myonecrosis (CK-MB >16 U/L)	6-8 h after PCI	Asian population, LAR determined once, adh NA, adj OCs unblinded
Myonecrosis (CK-MB >5.0 ng/mL)	20-24 h after PCI	CK-MB values not available for 6 patients, adj OCs unblinded

Table 1 - Footnotes

^aPatients with PAD undergoing PTA.

AA, arachidonic acid; ACS, acute coronary syndrome; adh, adherence; adj, adjudication; ADP, adenosine diphosphate; ARU, aspirin response unit, VerifyNow Aspirin Assay (Ultegra/Verify Now; Accumetrics, San Diego, CA); BMI, body mass index (calculated as weight in kilograms divided by height in meters squared); CABG, coronary artery bypass graft; CAD, coronary artery disease; CEPI-CT, collagen epinephrine closure time; CK-MB, creatine kinase–myocardial band; CV, cardiovascular; DM, diabetes mellitus; LAR, laboratory-defined aspirin resistance; LTA, light transmission aggregometry; MI, myocardial infarction; NA, not assessed; NR, not reported; OC, outcome; PAD, peripheral artery disease; PCI, percutaneous coronary intervention; PFA-100, Platelet Function Analyzer-100 system (Dade Behring, Deerfield, IL); PTA, percutaneous transluminal angioplasty; TEG, thromboelastography; TxB2, thromboxane B2; UA, unstable angina.

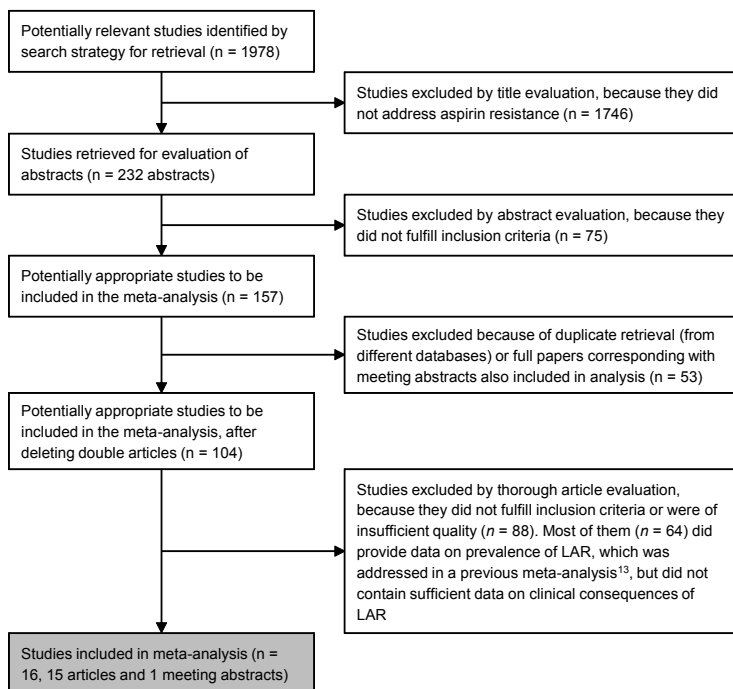


Figure 1 - Flowchart of the process of study selection. LAR indicates laboratory-defined aspirin resistance.

studies platelet function was assessed by the VerifyNow Aspirin Assay (Accumetrics, San Diego, CA, USA), which measures changes in light transmittance related to the rate of aggregation, using a disposable cartridge with fibrinogen-coated beads and a platelet activator.²⁸⁻³⁰ Three studies employed other techniques.^{15,16,27} Duration of follow-up ranged from 6 to 8 hours (for the creatine kinase-myocardial band elevation) to more than 7.5 years.^{26,28}

Table 2 – Statistical Details of Included Studies

Source	Patients with LAR, (%)		Occurrence of cardiovascular events		P-value
	Patients with LAR, n/N (%) ^a	Patients without LAR, n/N (%) ^b OR (95%CI)	Patients with LAR, n/N (%) ^a	Patients without LAR, n/N (%) ^b OR (95%CI)	
Grottemeyer et al., 1993 ¹⁵	60 (33)	24/60 (40)	5/114 (4)	14.5 (5.2 to 40.9)	<0.001
Buchanan et al., 2000 ¹⁶	158 (55)	15/158 (10)	9/131 (7)	1.4 (0.6 to 3.4)	0.42
Andersen et al., 2002 ¹⁷	25 (35)	9/25 (36)	11/46 (24)	1.8 (0.6 to 5.2)	0.28
Eikelboom et al., 2002 ¹⁸	NR	NR	NR	1.8 (1.2 to 2.9) ^b	0.01
Grundmann et al., 2003 ¹⁹	12 (23)	12/35 (34) ^c	0/18 (0) ^d	6.8 (1.8 to 26.2) ^e	0.004
Gum et al., 2003 ²⁰	17 (5)	4/17 (24)	30/309 (10)	2.9 (0.9 to 9.3)	0.09
Cotter et al., 2004 ²¹	21 (29)	6/21 (29)	3/52 (6)	6.5 (1.5 to 29.3)	0.01
Cheng et al., 2005 ³⁰ (abstract)	113 (27)	NR	NR	2.9 (1.5 to 5.7) ^f	0.002
Pamukcu et al., 2006 ²²	20 (19)	9/20 (45)	10/85 (12)	6.1 (2.0 to 18.5)	<0.001
Stejskal et al., 2006 ²³	57 (55)	50/57 (88)	21/46 (46)	8.5 (3.2 to 22.7)	<0.001
Mueller et al., 1997 ²⁴	65 (65)	8/65 (12)	0/35 (0)	10.5 (0.6 to 187.5)	0.048
Ziegler et al., 2002 ²⁵	5 (10)	0/5 (0)	13/47 (28)	0.2 (0.0 to 4.5)	0.31
Yilmaz et al., 2005 ²⁶	8 (29)	7/14 (50) ^c	1/14 (7) ^d	13.0 (1.3 to 128.1) ^e	0.03
Poston et al., 2006 ²⁷	22 (10)	4/22 (18)	12/203 (6)	3.5 (1.0 to 12.1)	0.06
Chen et al., 2004 ²⁸	29 (19)	15/29 (52)	30/122 (25)	3.3 (1.4 to 7.6)	0.004
Lev et al., 2006 ²⁹	19 (13)	7/18 (39)	23/126 (18)	2.9 (1.0 to 8.1)	0.045

^aThe numerators are the patients with cardiovascular events, and the denominators are the total number of patients with and without LAR.

^bReported OR of upper vs. lower quartile.

^cPrevalence of LAR in cases.

^dPrevalence of LAR in controls.

^eOdds ratio for patients with LAR.

^fOdds ratio not reported; reported as HR.

CI, confidence interval; HR, hazard ratio; LAR, laboratory-defined aspirin resistance; NR, not reported; OR, odds ratio

Relation between laboratory aspirin resistance and cardiovascular outcome

The prevalence of laboratory aspirin resistance ranged from 5 to 65%.^{20,24} In the 12 studies eligible for pooling,^{15-17,20-25,27-29} comprising 1813 patients, the mean prevalence of laboratory aspirin resistance was 27%. The total variation (I^2) among these studies, likely reflecting aforementioned differences, was 49%, resulting in a significant statistical heterogeneity among studies ($P=0.03$).

The ORs of cardiovascular outcome varied from 0.2 (95%CI 0.0 to 4.5) to 14.5 (95%CI 5.2 to 40.9) for laboratory aspirin resistance.^{15,25} We pooled the ORs of several groups of studies, the results of which are graphically presented in Figure 2. When studies with clinical cardiovascular outcomes were pooled,^{15-17,20-23} the resultant OR for laboratory aspirin resistance was 4.4 (95%CI 2.2 to 8.7). In three cohort studies addressing (re)occlusion after interventional procedures,^{24,25,27} the pooled OR was 2.4 (95%CI 0.4 to 14.3). The OR of myonecrosis after PCI was 3.1 (95%CI 1.6 to 6.0).^{28,29}

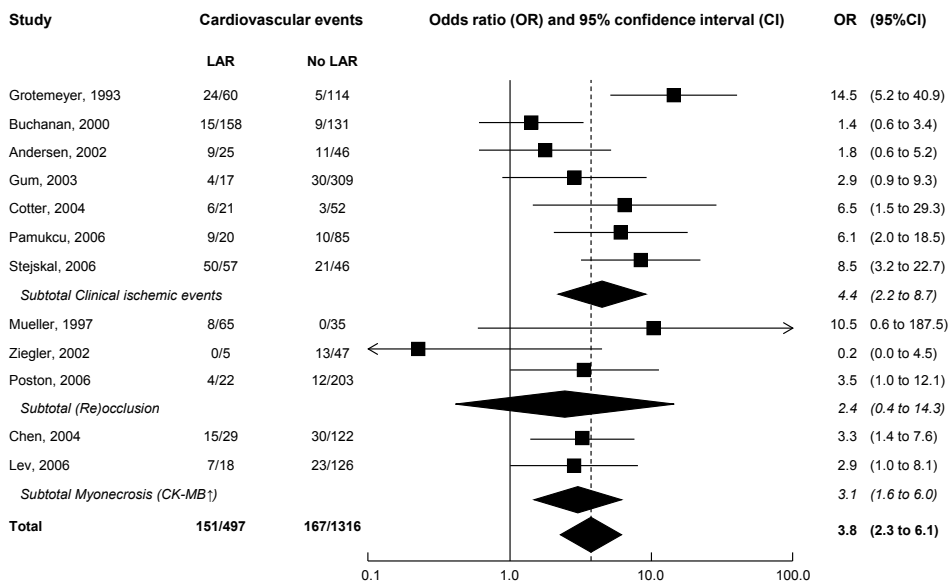


Figure 2 - Forest plots of odds ratios (ORs) of the cardiovascular outcome for patients with laboratory-defined aspirin resistance (LAR) versus those without LAR from eligible studies. Studies are grouped by the outcome parameter used: group 1 presents a composite outcome of clinical ischemic events, including cardiovascular death, myocardial infarction, stroke, acute coronary syndrome, and revascularization procedure; group 2, (re)occlusion after bypass grafting or angioplasty; and group 3, myonecrosis after percutaneous coronary intervention (PCI), represented by a creatine kinase–myocardial band elevation. In the lower part of the figure, all studies on these cardiovascular outcomes are pooled together. The black squares represent ORs for the association between aspirin resistance and cardiovascular outcomes of individual studies. Horizontal lines represent corresponding 95% confidence intervals (95CIs). The 95CIs of the totals are indicated by the black diamonds.

When all of these studies were combined, the pooled OR of cardiovascular outcome was 3.8 (95%CI 2.3 to 6.1) for laboratory aspirin resistance. We also stratified for the aspirin dosage used (≤ 100 mg, 101 to 300 mg and ≥ 300 mg); however, no differences among the dosage groups were found.

All studies not included in the analysis because they were not a cohort study or because they did not report proportions of patients having laboratory aspirin resistance versus those without, showed an association between persistent platelet reactivity despite use of aspirin and occurrence of cardiovascular events as well (Table 1 and Table 2).^{18,19,26,30}

DISCUSSION

We conducted a systematic review and meta-analysis to quantify evidence regarding the question of whether patients with laboratory aspirin resistance have a higher risk of recurrent cardiovascular events. We showed that patients with laboratory aspirin resistance have an increased risk of cardiovascular events. Among studies eligible for meta-analysis, the pooled OR of cardiovascular outcome was 3.8 (95%CI 2.3 to 6.1). The studies not included in the analysis strengthen this conclusion, because they all indicate an association between persistent platelet reactivity despite use of aspirin and occurrence of cardiovascular events.

The studies in our systematic review varied in many ways. The patients included in the studies had different cardiovascular diseases and experienced a variety of risks of recurrent events. Furthermore, the studies differed in aspirin dosage used, duration of follow-up time, laboratory methods used to establish the effects of aspirin and definition of outcome. Despite these clinical and methodological diversities, almost all included studies suggested a positive association between the risk of cardiovascular events and the presence of laboratory aspirin resistance. We therefore decided that it could be informative to pool the findings from the cohort studies with a random-effects model, which partly accounts for statistical heterogeneity between the studies.¹⁴

Beside these heterogeneities, several methodological limitations of included studies require comment. In a most of the studies, endpoints were not adjudicated in a blinded fashion for laboratory aspirin resistance, making them more susceptible for bias.^{15,17,22-25,28-30} In one study, 45 patients were excluded for reasons that were not mentioned,²⁴ and in another study allocation to either aspirin or clopidogrel was not randomized but based on patients' concerns. Moreover, use of non-steroidal anti-inflammatory drugs, which may have differed between studies as it was no formal exclusion criterion in nine of them,^{15-18,21,25-27,29,30} could have influenced the prevalence of laboratory aspirin resistance.³¹⁻³³ Furthermore, laboratory aspirin resistance was only determined on a single occasion in all but four studies,^{23,24,27,29} which may have led to misclassification. For example, persistent platelet reactivity may be more common after coronary artery bypass grafting owing to increased platelet turnover.³⁴ This temporal 'resistance' was recently observed in a population of patients who had undergone coronary bypass surgery.²⁷ Although noncompliance with treatment is an

important cause of laboratory aspirin resistance,^{21,35} patient adherence to treatment was assessed in only three studies.^{16,21,24} Cotter et al. have suggested that after exclusion of non-adherent patients, laboratory aspirin resistance is no longer related to recurrent events.²¹

The strength of our study lies in the systematic nature of the reviewing process. By prespecifying inclusion criteria and using a sensitive search strategy, we were able to review all retrievable studies with a minimum risk of bias. Thus, we were able to provide an extensive and, to our knowledge, complete overview of available data on cardiovascular consequences of laboratory aspirin resistance in patients with cardiovascular disease. In contrast, previous reviews included only selected studies on cardiovascular consequences of aspirin resistance. Many individual studies were relatively small, making extrapolation difficult. However, by pooling available studies, we found a strong association between laboratory aspirin resistance and recurrent cardiovascular events.

As in all systematic reviews, our results could have been influenced by several forms of bias. However, we tried to minimize selection bias by applying no formal language restriction and including both full-text articles and meeting abstracts. Furthermore, we used a funnel plot in which there was no inverse relationship between size of individual studies and ORs of cardiovascular outcomes, which argues against existence of publication and reporting bias. However, these forms of bias could not be completely excluded owing to the relatively small number of included studies. Moreover, we assumed laboratory aspirin resistance to be categorical variable. This may not be the case because there is no standardized definition of laboratory aspirin resistance. However, even when laboratory aspirin resistance should be seen as a continuous variable, it is likely that a categorical definition would be also predictive and that just the strength of the association might differ.

In conclusion, our systematic review and meta-analysis strongly indicates that laboratory aspirin resistance is a clinically important phenomenon. Patients biochemically identified as having aspirin resistance are more likely to also have "clinical resistance" to aspirin because they exhibit a considerably increased risk of recurrent cardiovascular events compared with patients identified as (laboratory) aspirin sensitive. Because cardiovascular diseases are very prevalent and associated with considerable mortality and morbidity, there is a clear need for future studies to thoroughly evaluate individual determinants of laboratory aspirin resistance, predictive value of the various laboratory methods and possible solutions for individual patients.

REFERENCES

1. Thom T, Haase N, Rosamond W et al. Heart disease and stroke statistics--2006 update: A report from the American Heart Association statistics committee and stroke statistics subcommittee. *Circulation*. 2006;113:e85-151.
2. Ruggeri ZM. Platelets in atherothrombosis. *Nat Med*. 2002;8:1227-1234.
3. Patrono C, Garcia Rodriguez LA, Landolfi R, Baigent C. Low-dose aspirin for the prevention of atherothrombosis. *N Engl J Med*. 2005;353:2373-2383.
4. Catella-Lawson F. Vascular biology of thrombosis: Platelet-vessel wall interactions and aspirin effects. *Neurology*. 2001;57:5S-7.
5. Antithrombotic Trialists' Collaboration. Collaborative meta-analysis of randomised trials of antiplatelet therapy for prevention of death, myocardial infarction, and stroke in high risk patients. *BMJ*. 2002;324:71-86.
6. Weber AA, Przytulski B, Schanz A, Hohlfeld T, Schror K. Towards a definition of aspirin resistance: a typological approach. *Platelets*. 2002;13:37-40.
7. Patrono C. Aspirin resistance: definition, mechanisms and clinical read-outs. *J Thromb Haemost*. 2003;1:1710-1713.
8. Hankey GJ, Eikelboom JW. Aspirin resistance. *BMJ*. 2004;328:477-479.
9. Michelson AD, Cattaneo M, Eikelboom JW et al. Aspirin resistance: position paper of the Working Group on Aspirin Resistance. *J Thromb Haemost*. 2005;3:1309-1311.
10. Sanderson S, Emery J, Baglin T, Kinmonth AL. Narrative review: aspirin resistance and its clinical implications. *Ann Intern Med*. 2005;142:370-380.
11. Hankey GJ, Eikelboom JW. Aspirin resistance. *Lancet*. 2006;367:606-617.
12. Pollack A. For some, Aspirin may not help hearts. *New York, New York Times*, July 20, 2004.
13. Hovens MMC, Snoep JD, Eikenboom JCJ, van der Bom JG, Mertens BJA, Huisman MV. Prevalence of persistent platelet reactivity despite use of aspirin: a systematic review. *Am Heart J*. 2007;153:175-181.
14. DerSimonian R, Laird N. Meta-analysis in clinical trials. *Control Clin Trials*. 1986;7:177-188.
15. Grottemeyer KH, Scharafinski HW, Husstedt IW. Two-year follow-up of aspirin responder and aspirin non responder. A pilot-study including 180 post-stroke patients. *Thromb Res*. 1993;71:397-403.
16. Buchanan MR, Schwartz L, Bourassa M, Brister SJ, Peniston CM. Results of the BRAT study--a pilot study investigating the possible significance of ASA nonresponsiveness on the benefits and risks of ASA on thrombosis in patients undergoing coronary artery bypass surgery. *Can J Cardiol*. 2000;16:1385-1390.
17. Andersen K, Hurlen M, Arnesen H, Seljeflot I. Aspirin non-responsiveness as measured by PFA-100 in patients with coronary artery disease. *Thromb Res*. 2002;108:37-42.
18. Eikelboom JW, Hirsh J, Weitz JI, Johnston M, Yi Q, Yusuf S. Aspirin-resistant thromboxane biosynthesis and the risk of myocardial infarction, stroke, or cardiovascular death in patients at high risk for cardiovascular events. *Circulation*. 2002;105:1650-1655.
19. Grundmann K, Jaschonek K, Kleine B, Dichgans J, Topka H. Aspirin non-responder status in patients with recurrent cerebral ischemic attacks. *J Neurol*. 2003;250:63-66.
20. Gum PA, Kottke-Marchant K, Welsh PA, White J, Topol EJ. A prospective, blinded determination of the natural history of aspirin resistance among stable patients with cardiovascular disease. *J Am Coll Cardiol*. 2003;41:961-965.
21. Cotter G, Shemesh E, Zehavi M et al. Lack of aspirin effect: aspirin resistance or resistance to taking aspirin? *Am Heart J*. 2004;147:293-300.
22. Pamukcu B, Oflaz H, Oncul A et al. The role of aspirin resistance on outcome in patients with acute coronary syndrome and the effect of clopidogrel therapy in the prevention of major cardiovascular events. *J Thromb Thrombolysis*. 2006;V22:103-110.
23. Stejskal D, Vaclavik J, Lacnak B, Proskova J. Aspirin resistance measured by cationic propyl gallate platelet aggregometry and recurrent cardiovascular events during 4 years of follow-up. *Eur J Intern Med*. 2006;17:349-354.
24. Mueller MR, Salat A, Stangl P et al. Variable platelet response to low-dose ASA and the risk of limb deterioration in patients submitted to peripheral arterial angioplasty. *Thromb Haemost*. 1997;78:1003-1007.

25. Ziegler S, Maca T, Alt E, Speiser W, Schneider B, Minar E. Monitoring of antiplatelet therapy with the PFA-100 in peripheral angioplasty patients. *Platelets*. 2002;13:493-497.
26. Yilmaz MB, Balbay Y, Caldir V et al. Late saphenous vein graft occlusion in patients with coronary bypass: possible role of aspirin resistance. *Thromb Res*. 2005;115:25-29.
27. Poston RS, Gu J, Brown JM et al. Endothelial injury and acquired aspirin resistance as promoters of regional thrombin formation and early vein graft failure after coronary artery bypass grafting. *J Thorac Cardiovasc Surg*. 2006;131:122-130.
28. Chen WH, Lee PY, Ng W, Tse HF, Lau CP. Aspirin resistance is associated with a high incidence of myonecrosis after non-urgent percutaneous coronary intervention despite clopidogrel pretreatment. *J Am Coll Cardiol*. 2004;43:1122-1126.
29. Lev EI, Patel RT, Maresh KJ et al. Aspirin and clopidogrel drug response in patients undergoing percutaneous coronary intervention: the role of dual drug resistance. *J Am Coll Cardiol*. 2006;47:27-33.
30. Cheng X, Chen WH, Lee PY, Ng W, Kwok YY, Lau CP. Prevalence, profile, predictors, and natural history of aspirin resistance measured by the ultegra rapid platelet function assay-ASA in patients with coronary heart disease. *Circulation*. 2005;111:E339.
31. Catella-Lawson F, Reilly MP, Kapoor SC et al. Cyclooxygenase inhibitors and the antiplatelet effects of aspirin. *N Engl J Med*. 2001;345:1809-1817.
32. Capone ML, Sciulli MG, Tacconelli S et al. Pharmacodynamic interaction of naproxen with low-dose aspirin in healthy subjects. *J Am Coll Cardiol*. 2005;45:1295-1301.
33. MacDonald TM, Wei L. Effect of ibuprofen on cardioprotective effect of aspirin. *The Lancet*. 2003;361:573-574.
34. Zimmermann N, Wenk A, Kim U et al. Functional and biochemical evaluation of platelet aspirin resistance after coronary artery bypass surgery. *Circulation*. 2003;108:542-547.
35. Schwartz KA, Schwartz DE, Ghosheh K, Reeves MJ, Barber K, DeFranco A. Compliance as a critical consideration in patients who appear to be resistant to aspirin after healing of myocardial infarction. *Am J Cardiol*. 2005;95:973-975.
36. Grottemeyer KH. The platelet-reactivity-test--a useful "by-product" of the blood-sampling procedure? *Thromb Res*. 1991;61:423-431.
37. The Heart Outcomes Prevention Evaluation study investigators. Effects of an angiotensin-converting-enzyme inhibitor, ramipril, on cardiovascular events in high-risk patients. *N Engl J Med*. 2000;342:145-153.
38. The Heart Outcomes Prevention Evaluation study investigators. Vitamin E supplementation and cardiovascular events in high-risk patients. *N Engl J Med*. 2000;342:154-160.