

## Institutional report - Aortic and aneurysmal Circulating matrix metalloproteinase-9 concentrations and abdominal aortic aneurysm presence: a meta-analysis

Hisato Takagi\*, Hideaki Manabe, Norikazu Kawai, Shin-nosuke Goto, Takuya Umemoto

Department of Cardiovascular Surgery, Shizuoka Medical Center, 762-1 Nagasawa, Shimizu-cho, Sunto-gun, Shizuoka 411-8611, Japan

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### Abstract

To summarize the present evidence for an association between matrix metalloproteinase-9 (MMP-9) and abdominal aortic aneurysm (AAA) presence, we performed a meta-analysis of case-control studies that compared circulating MMP-9 concentrations between patients with AAA and subjects without AAA. MEDLINE database was searched to identify all case-control studies. For each study, data regarding serum or plasma MMP-9 concentrations in both the AAA and control groups were used to generate standardized mean differences (SMDs) and 95% confidence intervals (CIs). Study-specific estimates were combined using inverse variance-weighted average of logarithmic SMDs in both fixed- and random-effects models. Our search identified eight eligible studies including 580 patients with AAA and 258 subjects without AAA. Pooled analysis demonstrated significantly higher circulating MMP-9 concentrations in the AAA group than those in the control group in random-effect models (SMD, 0.70; 95% CI, 0.23–1.17;  $P=0.004$ ). There was significant study heterogeneity of results ( $P<0.00001$ ) but no evidence of significant publication bias ( $P=0.1376$ ). We found that, based on a systematic review and meta-analysis, circulating MMP-9 concentrations are higher in patients with AAA than those in subjects without AAA. Higher circulating MMP-9 concentrations are associated with AAA presence.

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**Keywords:** Matrix metalloproteinase-9; Aortic aneurysm, abdomen; Biological markers

### 1. Introduction

Circulating concentrations of many kinds of biomarkers have been measured in cases with abdominal aortic aneurysm (AAA) and controls without AAA to assess those possible roles in the pathogenesis or progression of AAA. Circulating biomarkers could play a role in the diagnosis of AAA and may have a role in predicting subsequent progression of AAA [1]. These biomarkers include extracellular matrix markers, matrix-degrading enzymes, proteins associated with thrombosis, lipids, and markers of inflammation. Fragmentation of the extracellular matrix of the aortic media is perhaps the most specific histological hallmark of AAA [1]. Among matrix-degrading enzymes, circulating matrix metalloproteinase-9 (MMP-9) concentrations have been investigated most frequently, but the findings have not been completely consistent. To summarize the present evidence for an association between MMP-9 and AAA presence, we performed a meta-analysis of case-control studies that compared circulating MMP-9 concentrations between patients with AAA and subjects without AAA.

### 2. Methods

#### 2.1. Search strategy

All case-control studies that compared circulating MMP-9 concentrations between patients with AAA and subjects

without AAA were identified using a two-level search strategy. First, a public domain database (MEDLINE) was searched using a Web-based search engine (PubMed). Second, relevant studies were identified through a manual search of secondary sources including references of initially identified articles and a search of reviews and commentaries. All references were downloaded for consolidation, elimination of duplicates, and further analysis. The MEDLINE database was searched from January 1966 to December 2008. MeSH keywords included *matrix metalloproteinase 9*; and *aortic aneurysm, abdomen*. Text keywords included *matrix metalloproteinase 9*, *matrix metalloproteinase-9*, *MMP 9*, *MMP-9*, and *abdominal aortic aneurysm*.

#### 2.2. Study selection and data abstraction

Studies considered for inclusion met the following criteria: the design was a case-control study; the study population was patients with AAA and subjects without AAA; main outcomes included means and standard deviations (S.D.s) of circulating MMP-9 concentrations in the AAA and control groups. Data regarding detailed inclusion criteria and MMP-9 concentrations were abstracted (as available) from each individual study.

#### 2.3. Statistical analysis

We conducted a meta-analysis of summary statistics from the individual studies because detailed, patient-level data

\*Corresponding author. Tel.: +81 559752000; fax: +81 559752725.

E-mail address: kfgth973@ybb.ne.jp (H. Takagi).

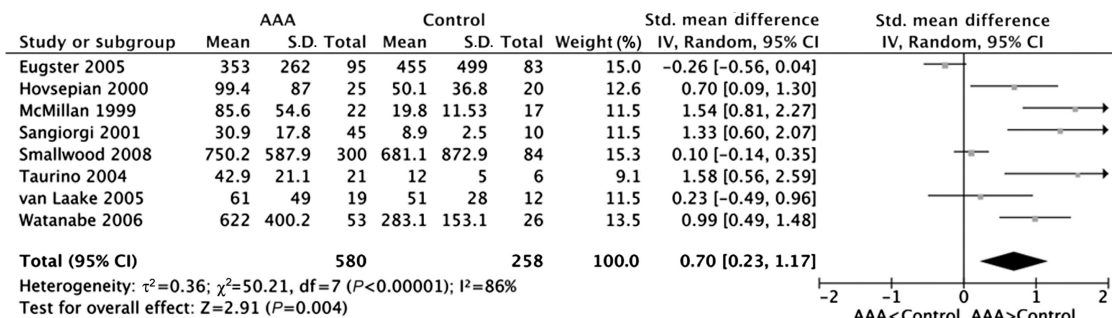


Fig. 1. Forest plot of standardized mean difference of circulating matrix metalloproteinase-9 concentrations between cases with abdominal aortic aneurysm (AAA) and controls without AAA. S.D., standard deviation; IV, inverse variance; CI, confidence interval.

were not available for all studies. For each study, data regarding serum or plasma MMP-9 concentrations in both the AAA and control groups were used to generate standardized mean differences (SMDs) and 95% confidence intervals (CIs). When 95% CIs or standard errors (S.E.s) of the concentrations were reported, we converted them into S.D.s by means of standard formulae in the Cochrane Handbook for Systematic Reviews of Interventions Version 5.0.1 [updated September 2008] (The Cochrane Collaboration, 2008. Available from [www.cochrane-handbook.org](http://www.cochrane-handbook.org)). When the concentrations were stated separately in subgroups of the AAA or control group, we combined them as the values of the AAA or control group. Study-specific estimates were combined using inverse variance-weighted average of logarithmic SMDs in both fixed- and random-effects models. Between-study heterogeneity was analyzed by means of standard  $\chi^2$ -tests. Where significant statistical heterogeneity was identified, the random-effect estimate was used preferentially as the summary measure. Sensitivity analyses were performed to assess the contribution of each study to the pooled estimate by excluding individual studies one at a time and recalculating the pooled SMD estimates for the remaining studies. To assess the impact of differential control selection, pooled estimates were explored exclusively for healthy controls. Publication bias was assessed graphically using a funnel plot and mathematically using an adjusted rank-correlation test, according to the method of Begg and Mazumdar [2]. All analyses were conducted using Review Manager (RevMan) [3] and Microsoft Excel (Version 11.5.0).

### 3. Results

Of 184 titles and abstracts identified through database searches, 64 were excluded because of neither human studies nor English-language publications. Among the remaining 120 abstracts, 12 were also excluded because of the publication type such as editorial, letter, review, case reports, and comment. We selected 108 full-text articles for detailed assessment and further excluded 100 of them because of not being case-control studies ( $n=95$ ) and other reasons ( $n=5$ ). Finally, our meta-analysis included eight relevant case-control studies [4–11] that compared serum or plasma MMP-9 concentrations between patients with AAA and subjects without AAA. In total, our meta-analysis

included data on 580 cases with AAA and 258 controls without AAA.

Six [5, 7–11] of the eight studies included men and women, whereas remaining two studies [4, 6] included men exclusively. Five [7–11] of the eight studies reported plasma MMP-9 concentrations (ng/l), whereas the remaining three studies [4–6] stated serum concentrations (ng/l). Only Smallwood et al. [4] reported geometric serum MMP-9 concentrations (ng/ml). Five [5–9] of the eight studies stated means and S.D.s of MMP-9 concentrations. Instead of S.D.s, two studies [10, 11] reported S.E.s and Smallwood et al. [4] stated 95% CIs; therefore, we converted them into S.D.s. Three [4, 6, 9] of the eight studies reported MMP-9 concentrations in healthy controls, and van Laake et al. [7] stated them in controls with aorto-iliac occlusive disease. Whereas, remaining four studies reported them separately in healthy controls and controls with atherosclerosis (arterial occlusive disease [5], carotid artery stenosis [8], atherosclerotic occlusive disease [10], or aorto-iliac occlusive disease [11]). In these four studies [5, 8, 10, 11], we combined the separately stated values as those in the control group. Eugster et al. [6] reported MMP-9 concentrations separately in patients with aortic dilatation  $\geq 25$  mm and in those with known AAA that were operated on, whereas Taurino et al. [8] and Sangiorgi et al. [9] stated them separately in patients undergoing open surgical and

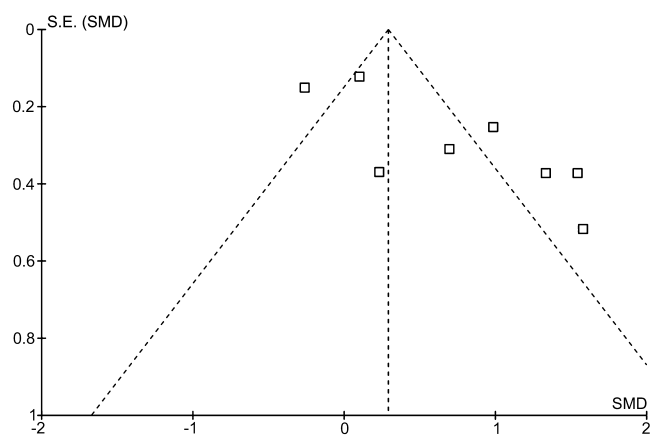


Fig. 2. Funnel plot of standardized mean difference (SMD) of circulating matrix metalloproteinase-9 between cases with abdominal aortic aneurysm (AAA) and controls without AAA. S.E., standard error.

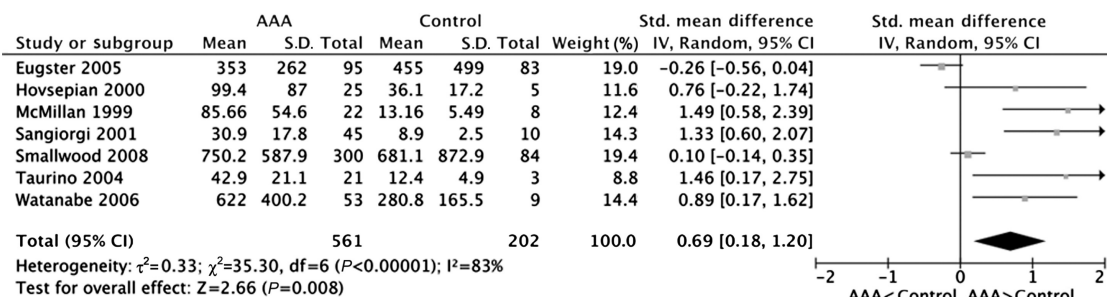


Fig. 3. Forest plot of standardized mean difference of circulating matrix metalloproteinase-9 concentrations between cases with abdominal aortic aneurysm (AAA) and healthy controls without AAA. S.D., standard deviation; IV, inverse variance; CI, confidence interval.

endovascular repair. In these three studies [6, 8, 9], we combined the separately reported values as those in the AAA group.

Five [5, 8–11] of the eight individual studies demonstrated significantly higher circulating MMP-9 concentrations, and two studies [4, 7] showed non-significantly higher concentrations in the AAA group than those in the control group. Only Eugster et al. [6] demonstrated non-significantly lower MMP-9 concentrations in the AAA group than those in the control group. Pooled analysis of all the eight studies demonstrated significantly higher circulating MMP-9 concentrations in the AAA group than those in the control group in random-effect models [SMD, 0.70; 95% CI, 0.23–1.17;  $P=0.004$  (Fig. 1)]. There was significant study heterogeneity of results ( $P<0.00001$ ) and accordingly a little difference in the pooled result from fixed-effects modeling (SMD, 0.29; 95% CI, 0.13–0.44;  $P=0.0003$ ). To assess publication bias, we generated a funnel plot of the effect size vs. the S.E. for each study (Fig. 2). There was no evidence of significant publication bias ( $P=0.1376$  by Begg adjusted rank-correlation test). To assess the impact of qualitative heterogeneity in trial design and control selection on the pooled effect estimate, we performed several sensitivity analyses. In general, exclusion of any single study from the analysis did not substantively alter the overall result of our analysis. Combining the seven studies [4–6, 8–11] with healthy controls (representing 561 cases and 202 controls; random-effects SMD, 0.69; 95% CI, 0.18–1.20;  $P=0.008$ ) did not substantially change the pooled point estimate (Fig. 3).

#### 4. Discussion

The results of our analysis suggest that circulating MMP-9 concentrations are higher in patients with AAA than those in subjects without AAA. Circulating MMP-9 concentrations could play a role in the diagnosis of AAA. Our analysis, however, must be viewed in the context of its limitations. First, there was substantial qualitative heterogeneity in control selection. The control groups included healthy subjects and patients with atherosclerosis. Our sensitive analyses explored exclusively for healthy controls, however, did not substantially change the pooled point estimate. Second, MMP-9 concentrations were measured in plasma or serum. We generated not mean differences but SMDs. Third, substantial heterogeneity in participant selection was present. Two of the eight studies included men exclu-

sively, whereas remaining six studies included men and women. Despite these acknowledged limitations, we found that, based on a systematic review and meta-analysis, circulating MMP-9 concentrations are higher in cases with AAA than those in controls without AAA. Higher circulating MMP-9 concentrations are associated with AAA presence. Hovsepian et al. [9] found that an elevated MMP-9 had sensitivity of 48% and a specificity of 95% as a diagnostic screening test for the presence or absence of AAA. The false-positive rate was 7.7% and the false-negative rate was 51.8%, for an overall accuracy of 65%. These results indicate that an elevated MMP-9 was a relatively good predictor of the presence of AAA, but that normal MMP-9 was insufficient to exclude the diagnosis [9].

It is postulated that biomarkers measured at diagnosis or during follow-up might provide important prognostic information about subsequent aortic behavior [1]. Hackmann et al. [12] demonstrated a significant positive correlation between the maximum aortic diameter and the plasma MMP-9 level. Lindholt et al. [13] also showed that plasma MMP-9 levels were significantly associated with aneurysmal size and expansion. Meanwhile, more authors demonstrated no significant correlation between circulating MMP-9 concentrations and AAA diameter [5–7, 14] or expansion [5]. Further studies are needed to confirm whether circulating MMP-9 concentrations are associated with AAA diameter or expansion.

Wilson et al. [14] showed that the concentrations of MMP-9 were significantly elevated in the plasma of ruptured AAA compared with non-ruptured AAA. Furthermore, they demonstrated that elevation of MMP-9 was associated with ruptured aneurysm related 30-day mortality. Circulating MMP-9 concentrations may be associated with AAA rupture and be a survival indicator in this group.

#### References

- [1] Golledge J, Tsao PS, Dalman RL, Norman PE. Circulating markers of abdominal aortic aneurysm presence and progression. *Circulation* 2008;118:2382–2392.
- [2] Begg CB, Mazumdar M. Operating characteristics of a rank correlation test for publication bias. *Biometrics* 1994;50:1088–1101.
- [3] Review Manager (RevMan) [Computer program]. Version 5.0. Copenhagen: The Nordic Cochrane Centre, The Cochrane Collaboration, 2008.
- [4] Smallwood L, Allcock R, van Bockxmeer F, Warrington N, Palmer LJ, Iacopetta B, Golledge J, Norman PE. Polymorphisms of the matrix metalloproteinase 9 gene and abdominal aortic aneurysm. *Br J Surg* 2008;95:1239–1244.

- [5] Watanabe T, Sato A, Sawai T, Uzuki M, Goto H, Yamashita H, Akamatsu D, Sato H, Shimizu T, Miyama N, Nakano Y, Satomi S. The elevated level of circulating matrix metalloproteinase-9 in patients with abdominal aortic aneurysms decreased to levels equal to those of healthy controls after an aortic repair. *Ann Vasc Surg* 2006;20:317–321.
- [6] Eugster T, Huber A, Obeid T, Schwegler I, Gürke L, Stierli P. Aminoterminal propeptide of type III procollagen and matrix metalloproteinases-2 and -9 failed to serve as serum markers for abdominal aortic aneurysm. *Eur J Vasc Endovasc Surg* 2005;29:378–382.
- [7] van Laake LW, Vainas T, Dammers R, Kitslaar PJ, Hoeks AP, Schurink GW. Systemic dilation diathesis in patients with abdominal aortic aneurysms: a role for matrix metalloproteinase-9? *Eur J Vasc Endovasc Surg* 2005;29:371–377.
- [8] Taurino M, Visco V, Raffa S, Ricci B, Ruggiero M, Torrisi MR, Fiorani P. Matrix metalloproteinase 9 activity in patients before and after endovascular or surgical repair of abdominal aortic aneurysms. *Vascular* 2004;12:312–317.
- [9] Sangiorgi G, D'Averio R, Mauriello A, Bondio M, Pontillo M, Castelvechchio S, Trimarchi S, Tolva V, Nano G, Rampoldi V, Spagnoli LG, Inglese L. Plasma levels of metalloproteinases-3 and -9 as markers of successful abdominal aortic aneurysm exclusion after endovascular graft treatment. *Circulation* 2001;104(12 Suppl 1):I288–I295.
- [10] Hovsepian DM, Ziporin SJ, Sakurai MK, Lee JK, Curci JA, Thompson RW. Elevated plasma levels of matrix metalloproteinase-9 in patients with abdominal aortic aneurysms: a circulating marker of degenerative aneurysm disease. *J Vasc Interv Radiol* 2000;11:1345–1352.
- [11] McMillan WD, Pearce WH. Increased plasma levels of metalloproteinase-9 are associated with abdominal aortic aneurysms. *J Vasc Surg* 1999;29:122–129.
- [12] Hackmann AE, Rubin BG, Sanchez LA, Geraghty PA, Thompson RW, Curci JA. A randomized, placebo-controlled trial of doxycycline after endoluminal aneurysm repair. *J Vasc Surg* 2008;48:519–526.
- [13] Lindholt JS, Vammen S, Fasting H, Henneberg EW, Heickendorff L. The plasma level of matrix metalloproteinase 9 may predict the natural history of small abdominal aortic aneurysms. A preliminary study. *Eur J Vasc Endovasc Surg* 2000;20:281–285.
- [14] Wilson WR, Anderton M, Choke EC, Dawson J, Loftus IM, Thompson MM. Elevated plasma MMP1 and MMP9 are associated with abdominal aortic aneurysm rupture. *Eur J Vasc Endovasc Surg* 2008;35:580–584.