C-Reactive protein/interleukin-6 ratio as marker of the size of the uncomplicated thoracic aortic aneurysms

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Abstract

OBJECTIVES: The role of C-reactive protein (CRP) and interleukin-6 (IL-6) as markers in the prognosis of asymptomatic thoracic aortic aneurysm (TAA) patients has not been well established. As such, we evaluated a group of patients for a possible association between serum CRP and IL-6 and aneurysm dimension.

METHODS: Serum CRP and IL-6 were determined and aneurysmal size was measured in 26 patients with TAA.

RESULTS: The mean (SD) CRP and IL-6 were 0.58 (1.07) and 7.47 (17.78) pg/ml, respectively. Serum CRP, IL-6 and the ratio CRP/IL-6 correlated with the descending aortic aneurysmal dimension (r = 0.426, r = 0.743 and r = 0.328, respectively). A significant correlation was also found between values of the ratio above 0.8 and aneurysmal dimension (both ascending and descending aneurysms) (r = 0.785). Additionally, a significant association between smoking, age group above 69 years and dyslipidemia and aneurysm dimension was established (P = 0.002, P = 0.061 and P = 0.070, respectively).

CONCLUSIONS: This report shows that serum CRP, IL-6 levels and the ratio CRP/IL-6 are associated with descending aortic aneurysmal dimensions. Also values of the ratio CRP/IL-6 above 0.8 are associated with aneurysmal dimensions for both ascending and descending aortic aneurysms. It is still early to establish the clinical significance of those findings, and further studies with larger groups of patients with longer follow-up are required in order to truly assess the usefulness of the serum CRP and IL-6 as markers in relation to the progression of the disease.

Keywords: Aneurysm · Aorta · Interleukin-6 · C-Reactive protein

INTRODUCTION

C-Reactive protein (CRP) [1] is a classical plasma protein a marker that is elevated in the acute phase of inflammation, infection and tissue damage. CRP is mainly expressed by hepatocytes and its synthesis is regulated at the post-transcriptional level by cytokines mainly by Interleukin-6 (IL-6) with a synergic effect of IL-1.

Data [2] from both clinical and experimental studies have shown that a high level of plasma CRP is a risk factor as well as marker for cardiovascular disease and emerging evidence indicates that high levels of CRP may potentially be atherogenic.

Recent data from several studies have shown that CRP is produced by the liver as well as other tissues [3]. It has been demonstrated that CRP is expressed and produced in atherosclerotic lesions and aneurysmal tissue and is associated in the latter with aneurysmal size.

The multifactorial cytokine IL-6 has been specifically implicated in aneurysm pathogenesis contributing to both acute and chronic inflammatory process [4]. Elevated levels of circulating IL-6 have been reported in patients with abdominal aortic aneurysm (AAA) and levels appear to correlate with aortic diameter. There is some evidence that AAA and thoracic aortic aneurysm (TAA) may actually secrete IL-6 in the circulation.

The roles of CRP and IL-6 as markers in the prognosis of uncomplicated TAAs has yet to be well investigated and as such, the aim of this study was to examine their roles in the prognosis of these TAAs.

METHODS

Patients with uncomplicated TAAs (n = 26) and a diameter above 4 cm were included in this study. Eighteen (69.2%) patients were
aware of the presence of the aneurysm and for the remaining 8 (30.8%) patients, the presence of the aneurysm was an incidental finding by the cardiologists. Twenty-one patients were hypertensive with well-controlled blood pressures.

All patients were presented at the outpatient clinic of the Cardiovascular Department of the AHEPA University Hospital.

Patients with ruptured/symptomatic TAA, recent infections, active inflammatory disorders and/or serum CRP >5 mg/dl were excluded.

The patients underwent a series of three CT images of the thoracic aorta. The scans were performed at 4 and 8 months after the first one, and at the same time blood samples were obtained for the determination of CRP and IL-6.

Twenty-two patients had involvement of the ascending aorta and 4, involvement of the descending aorta. Two of the patients with involvement of the descending aorta also had aneurysmal involvement of the abdominal aorta. The first patient underwent replacement of the abdominal aorta and the second underwent endovascular stent insertion. Only one patient had aneurysm of the thoracic aorta secondary to type B dissection.

The baseline characteristics of the patients are shown in Table 1.

### Determination of serum CRP and IL-6

Venous blood samples that were drawn were immediately centrifuged for 8 min at 3000 rpm. Serum was stored at −28°C until analysis. CRP was determined by the rate nephelometry immunoassay in a nephelometer Image 800 (Beckman-Coulter Inc., Fullerton, CA). Plasma IL-6 concentrations were measured by ELISA. The polyclonal and monoclonal antibodies were obtained from R&D (Quantikine HS, R&D Systems Inc., Minneapolis, MN).

### Thoracic aortic aneurysm dimensions

Computer-assisted tomography was used to visualize the aorta and to determine the maximal aneurysm diameter. CT scans used were the Tomoscan SR 700 (Philips) and the Somaton Emotion 16 Siemens. The field of view was 370 cm. Firstly without contrast material, sequential cuts of 10 cm each were done from the lung apices to the lung bases for the anatomic evaluation of the aorta and the presence of any atherosclerotic plaque. Afterwards, with the infusion of 120 ml of any commercially available non-ionic contrast material at a rate 3.2 ml/s and using the spiral technique, cuts of 5 mm were done after the initiation of the infusion of the contrast material in two planes. The first plane was cephalad-caudal from the level of the aortic arch to the lung bases and the second plane was caudo-cephalad from the lung bases to the lung apices. Finally, the measurements of the maximal aortic diameter at any level of the thoracic aorta were taken.

### Statistical analysis

Pearson’s $r$ was computed to explore the correlations between aneurysm size and log-transformed serum CRP, IL-6 and the ratio of CRP/IL-6. Analysis of Variance was used to further study the univariate association between serum CRP, IL-6 and the ratio of CRP/IL-6 and aneurysm dimension and to assess the independent effect of sex, age, smoking, hypertension, dyslipidemia and diabetes mellitus on aneurysm size. For the statistical analysis, the software of JMP (Version 4, SAS Institute Inc., Cary, NC) and KaleidaGraph (Version 4.1.1, Synergy software) were used.

### RESULTS

A total of 26 patients (5 women) aged 61.5 (±18.5) years with an atherosclerotic risk profile (hypertension: 80.8%, diabetes mellitus: 26.9%, smoking: 30.8%, dyslipidemia: 65.4%) were included in this study.

### CRP and IL-6 in aneurysmal disease

The mean (SD), 25th, 50th and 75th percentiles of serum CRP were 0.58 (1.07), 0.24, 0.35 and 0.63 mg/dl, respectively. Moreover, the mean (SD), 25th, 50th and 75th percentiles of serum IL-6 were 7.47 (17.78), 0.80, 3.12 and 5.20 pg/ml, respectively. Finally the mean values of the (SD) 25th, 50th and 75th percentiles of the ratio of CRP/IL-6 were 0.23 (0.24), 0.07, 0.17 and 0.31, respectively (Fig. 1).

The mean (SD) serum CRPs in each of the three measurements are 0.34 (0.451), 0.37 (0.323) and 0.35 (0.342) mg/dl,

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<th>Table 1: Baseline characteristics of the patients</th>
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<td>Demographic profile</td>
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ACE: angiotensin converting enzyme; ARB: angiotensin receptor blocking agent; AAA: aortic abdominal aneurysm.
respectively, with no significant statistical difference between the measurements ($P = NS$).

Aneurysm dimension and serum CPR and IL-6

The mean (SD) maximal aneurysm dimension was 4.66 (0.77) cm. The mean (SD) aneurysm dimensions of the three measurements are 4.4 (0.77), 4.4 (0.77) and 4.4 (0.80) cm, respectively, with no significant statistical difference between the measurements ($P = NS$).

The correlations of serum CRP, IL-6 and the ratio of CRP/IL-6 with the aneurysm dimension for each measurement separately are shown in figure 2.

On the first measurement, the correlation for CRP is $r = 0.296$ ($P = 0.509$), for IL-6, it is $r = 0.486$ ($P = 0.907$) and for the ratio CRP/IL-6, it is $r = 0.136$ ($P = 0.142$). For the second measurement for CRP, it is $r = 0.112$ ($P = 0.56$), for IL-6, it is $r = 0.555$ ($P = 0.823$) and for the ratio CRP/IL-6, it is $r = 0.110$ ($P = 0.591$). For the third measurement, the correlation for CRP is $r = 0.124$ ($P = 0.564$), for IL-6, it is $r = 0.210$ ($P = 0.885$) and for the ratio CRP/IL-6 $r = 0.07$ ($P = 0.324$).

Table 2 shows the correlations of all possible combinations between all the variables, serum CRP and IL-6, ratio of CRP/IL-6 and the aneurysm dimensions (both ascending and descending aortic aneurysms).

Aneurysm size (both ascending and descending aortic aneurysms) did not correlate with serum CRP and IL-6 ($r = -0.0694$ and $r = -0.0070$, respectively) (Table 2).

On the other hand, there was a strong linear correlation between the aneurysmal size and the ratio of CRP/IL-6 for values above 0.8 ($r = 0.785$, Fig. 3). Above this value of 0.8, any increase of the aortic aneurysmal size is associated with an increase of the ratio of CRP/IL-6 and vice versa. Below this cut-off value of 0.8, there is no correlation between the aneurysmal size and the ratio of CRP/IL-6.

Taking into account the extent of the aneurysm, we have the following results. The mean diameter of the aneurysms of this study of the ascending aorta is 4.54 cm and that of the descending aorta is 5.28 cm ($P = 0.999$).

The mean serum CRP levels of the ascending and descending aortic aneurysms are 0.59 and 0.50 mg/dl, respectively ($P = 0.398$). The mean serum IL-6 levels of the ascending and descending aortic aneurysms are 7.58 and 6.86 pg/ml, respectively ($P = 0.449$).

The mean values of the ratio CRP/IL-6 for ascending and descending aortic aneurysms are 0.25 and 0.10 ($P = 0.023$, which is statistically significant).

The correlations between the aneurysm diameter and the levels of the serum CRP, IL-6 and the ratio CRP/IL-6 for ascending and descending aortic aneurysms are the following:

- Concerning the aneurysms of the ascending aorta, there is no correlation between the aneurysm diameter and the serum CRP, IL-6 and the ratio CRP/IL-6 with $r = 0.159$, $r = 0.115$ and $r = 0.184$, respectively (Fig. 4).
- Concerning the aneurysms of the descending aorta, there is a correlation between the aneurysm size and the serum CRP ($r = 0.426$) and the ratio CRP/IL-6 ($r = 0.328$). Interestingly there is also a strong correlation between the aneurysm size and the serum IL-6 levels ($r = 0.743$) (Fig. 5).
From the risk factors for cardiovascular disease, only smoking, age above 69 years and dyslipidemia showed an association with the aneurysm size. The aneurysm diameter mean 95% Confidence Intervals, CI of the patients in the age group above 69 years and those who were smokers tended to be elevated compared with the other patients. Patients aged above 69 years had a mean diameter of 4.88 cm vs 4.39 cm for younger patients ($P = 0.067$). In this age group, there is a tendency ($P = 0.235$) to have higher mean values of serum CRP (0.80 mg/dl vs 0.51 mg/dl for younger patients). As far as serum IL-6 levels are concerned, patients younger than 69 years had the tendency ($P = 0.176$) to have higher values (13.36 pg/ml vs 6.82 pg/ml for patients above 69 years).

Smokers had larger mean aneurysm dimensions ($P = 0.0023$) than non-smokers (5.09 cm vs 4.5 cm for non-smokers). Also, there was no significant statistical difference ($P = 0.779$ and $P = 0.443$) between the levels of serum CRP and IL-6, respectively, between the two groups of patients. The mean values for serum CRP were 0.52 mg/dl for smokers vs 0.598 mg/dl for non-smokers and the mean values for IL-6 were 4.92 pg/ml for smokers vs 8.44 pg/ml for non-smokers.

The aneurysm diameter (mean 95% CI) of the dyslipidemic patients tended to be smaller ($P = 0.0704$). The mean diameter was 4.53 cm for dyslipidemic patients vs 4.84 cm for non-dyslipidemic patients. There was no significant statistical difference between the two groups of patients concerning the serum CRP and a tendency of dyslipidemic patients to have lower IL-6

| Table 2: Correlations of all possible combinations of all the variables |
|------------------------|-----------------|-----------------|-----------------|
| CRP (mg/dl) | IL-6 (pg/ml) | CT/CM | CRP/IL-6 |
| CRP (mg/dl) | 1.0000 | 0.3780 | -0.0694 | 0.1074 |
| IL-6 (pg/ml) | 0.3780 | 1.0000 | -0.0070 | -0.2695 |
| CT/CM | -0.0694 | -0.0070 | 1.0000 | -0.2150 |
| CRP/IL-6 | 0.1074 | -0.2695 | -0.2150 | 1.0000 |

Figure 2: First (a), second (b) and third (c) measurement, correlations between the variables and the aneurysmal size.

Figure 3: Correlation between aneurysmal size and the ratio of CRP/IL-6.
Figure 4: Correlation between the ascending aorta aneurysm size and the level of the serum CRP (a), IL-6 (b) and the ratio CRP/IL-6 (c).

Figure 5: Correlation between the descending aorta aneurysm size and the level of the serum CRP (a), IL-6 (b) and the ratio CRP/IL-6 (c).
level values ($P = 0.216$ and $P = 0.141$, respectively). The mean values for serum CRP was 0.707 mg/dl for dyslipidemic vs 0.397 mg/dl for non-dyslipidemic patients and the mean values for serum IL-6 was 4.90 pg/ml for dyslipidemic vs 11.00 pg/ml for non-dyslipidemic patients (Fig. 6).

The association between hypertension, diabetes mellitus and sex and the aneurysmal size was non-significant ($P = 0.737$, $P = 0.139$ and $P = 0.461$, respectively).

**DISCUSSION**

Elevated serum CRP has been reported in patients with stenotic atherosclerotic disease and is associated with an increased risk of developing cardiovascular events [5, 6]. However, very little information exists about serum CRP and IL-6 in patients with (TAA) disease. A previous study by Powell et al. [7] showed that patients with symptomatic AAA had increased serum CRP compared with patients with obstructive disease (mean serum CRP levels 56 ± 10 mg/l). Also another study by Domanovits et al. [8] showed that patients with symptomatic and ruptured aneurysms had elevated serum CRP compared with patients with asymptomatic AAA, but failed to verify the elevated serum CRP in asymptomatic AAA. Finally, Vainas et al. [9] showed that in symptomatic AAA patients, serum hsCRP is associated with aneurysmal size (strongest association at levels of hsCRP >4.15 mg/l) and in some cases aneurysmal tissue is capable of producing CRP.

Concerning the role of IL-6 in the aneurysmal disease it has been shown by Jones et al. [10] that in AAA patients, the aortic aneurysm appears to be an important source of circulating IL-6 and the concentration is influenced by a genotype. Additionally, Dawson et al. [11] demonstrated that TAA is also a source of IL-6 in the circulation and lastly, Rohde et al. [12] showed that IL-6 values in AAA patients increased in a stepwise fashion among groups of aortic size and peaked in patients with aortic dilatation.

The extent of aortic aneurysm was examined in this analysis. From the literature, it is known that patients with AAA have a high frequency of thoracic aneurysms. This was studied by Larsson et al. [13]. They found that more than one-fourth of patients with AAA have a concomitant TAA and women are particularly affected. The majority of the patients suffered from aneurysm of the descending aorta or had both ascending and descending aortic aneurysms which shows an association between the presence of thoracic and AAs.

Another interesting study by Achneck et al. [14] shows that patients with ascending thoracic aneurysms (annuloaortic ectasia or type A dissection) are associated with decreased systemic atherosclerosis. This finding is consistent with related studies in the literature. A lower incidence of coronary disease was found among patients with thoracic aortic dilatation compared with those with the same pathology in the abdominal aorta [15, 16]. Furthermore, autopsied cases revealed a lower incidence of atherosclerosis in type A dissections compared with type B dissections [17], and in type A dissections compared with AAs [18]. However, the literature is deficient in systemic comparisons between the degree of atherosclerosis in patients with TAs and the general population. From the above, the role of atherosclerosis in the pathogenesis of the abdominal and descending aortic aneurysms can be assumed. Also the role of inflammation in the pathogenesis of atherosclerosis and the pathogenesis of the aneurysms of the abdominal aorta has been very well studied. It could be concluded that in this study patients who suffered from aneurysms of the descending aorta would have a distinct biomarker profile as a result of the greater involvement of inflammation in the pathogenesis compared with the patients with ascending aorta aneurysms, but this is not the case. There was no statistical difference between the mean values of the serum CRP and IL-6 levels of the patients suffering from ascending or descending aortic aneurysms. On the other hand, ascending aortic aneurysm patients had higher values with statistical difference of the ratio CRP/IL-6 compared with the patients having descending aortic aneurysms.

One important finding of this study consistent with previous studies discussed above which examine the relationship between the abdominal aortic aneurysms and the levels of serum CRP and IL-6, is the correlation which is found between the descending aortic aneurysm diameter and the level of the serum CRP, IL-6 and the ratio CRP/IL-6.
Another issue to discuss is whether there were any different biomarker profiles between the different aortic pathologies. There are different pathophysiological mechanisms that contribute to the development of the aneurysms of the ascending aorta. In this study, in 20 patients, the aneurysms of the thoracic aorta were secondary to atheroma and only one patient had an aneurysm secondary to type B dissection. In our study, there were no distinct biomarker profiles between the patients with aneurysm secondary to atheroma and the patient with aneurysm secondary to type B dissection, and it may be explained by the fact that there were not enough patients with aneurysms secondary to type B dissection in the sample in order to have reliable results. Further studies with large populations of patients, particularly to examine the different biomarker profiles in each aortic pathology independently, should follow in order to have a better picture of this issue. One study that contributed to this problem was done by Sakamura et al. [19]. They showed that a higher peak CRP value is a strong predictor of adverse long-term events in patients with type B acute aortic dissections.

The role of various matrix metalloproteinase in the pathogenesis of the TAAs has been very well established and studied [20], but in this particular study, they were not considered in its design.

In this study, the mean serum CRP and IL-6 were above the range for supposedly healthy individuals. Contrary to some of the above-mentioned studies, serum CRP and IL-6 in these asymptomatic TAA patients (both ascending and descending aneurysms) did not correlate with the aneurysm dimension. Analysing particularly the patients having descending aortic aneurysms, there was a strong correlation between the level of the serum CRP, IL-6 and the ratio CRP/IL-6 (higher increase of CRP ratio and smaller increase if IL-6 ratio). These findings are consistent with other studies that analyse AAs. On the other hand, CRP/IL-6 ratio values above 0.8 show a strong linear correlation with aneurysm size where any increase in TAA diameter is associated with an increase of the CRP/IL-6 ratio value. A possible explanation is that in this group of patients, with the increase of the aneurysmal size the ratio of the CRP becomes higher and the increase ratio of IL-6 becomes smaller. It is early to establish the clinical significance of this cut-off value of 0.8.

Patients of the age group of 69–80 years and smokers have larger TAA dimensions where dyslipidemic patients have smaller aneurysm dimensions. The latter may be attributed to the use of statins by these patients. It appears that the statin exposure slows down the aneurysm expansion and affects the aneurysm progression; however, this finding needs further studies to be verified. Recently, a study by Jovin et al. [21] showed that the intake of statins was associated with an improvement in long-term outcomes in this cohort of patients with TAAs and this was driven mainly by a reduction in aneurysm repair. Also, the aneurysm growth rate difference was significant over time (0.09 vs 0.12 cm/year in patients on and off statins).

This report shows that serum CRP, IL-6 levels and the ratio CRP/IL-6 are associated with ascending aortic aneurysmal dimensions. Also, values of the ratio CRP/IL-6 above 0.8 are associated with aneurysmal dimensions for both ascending and descending aortic aneurysms.

In conclusion this study has a few limitations. The biomarker profiles of the patients were not analysed according to each aortic pathology that causes aneurysms. Further studies with a larger group of patients and longer follow-up are needed to confirm the results of this study and to evaluate the usefulness of serum CRP and IL-6 as markers of the disease progression.

**Conflict of interest:** none declared.

**REFERENCES**


