Oxidative stress in the pathogenesis of thoracic aortic aneurysm: Protective role of statin and angiotensin II type 1 receptor blocker

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Abstract

Objective: The pathogenesis of thoracic aortic aneurysms (TAA) is still unclear. A recent investigation indicated that angiotensin II, a potent activator of NADH/NADPH oxidase, plays an important role in aneurysmal formation. We investigated the potential role of p22phox-based NADH/NADPH oxidase in the pathogenesis of TAA.

Methods: Human thoracic aneurysmal (n=40) and non-aneurysmal (control, n=39) aortic sections were examined, and the localization of p22phox, an essential component of the oxidase, and its expression differences were investigated by immunohistochemistry and Western blot. In situ reactive oxygen species (ROS) generation was examined by the dihydroethidium method, and the impact of medical treatment on p22phox expression was investigated by multiple regression analysis.

Results: In situ production of ROS and the expression of p22phox increased markedly in TAA throughout the wall, and Western blot confirmed the enhanced expression of p22phox. The expression was more intense in the regions where monocytes/macrophages accumulated. In these inflammatory regions, numerous chymase-positive mast cells and angiotensin converting enzyme-positive macrophages were present. Their localization closely overlapped the in situ activity of matrix metalloproteinase and the expression of p22phox. Multiple regression analysis revealed that medical treatment with statin and angiotensin II type 1 receptor blocker (ARB) suppressed p22phox expression in TAA.

Conclusion: Our findings indicate the role of p22phox-based NADH/NADPH oxidase and the local renin–angiotensin system in the pathogenesis of TAA. Statin and ARB might have inhibitory effects on the formation of aneurysms via the suppression of NADH/NADPH oxidase.

Keywords: Atherosclerosis; Histo(patho)logy; Renin angiotensin system; Statins; NADPH oxidase

1. Introduction

Aortic aneurysm is a chronic inflammatory disease that involves extensive extracellular matrix degradation and is characterized by localized weakening and dilatation of the aortic wall. The destruction of the tunica media and its...
elastin tissue is a histological feature of aortic aneurysms. Although numerous studies have been conducted to clarify the mechanism and pathogenesis of the disease, the precise mechanisms remain to be determined; however, several potential mechanisms have been proposed, including hemodynamic force, transmural inflammation, and destructive remodeling of the extracellular matrix [1–4]. An imbalance between matrix metalloproteinases (MMPs) and their inhibitors is crucial in the remodeling of the aortic wall, and the activities of these factors are more enhanced by inflammatory cell infiltrates [5–8]. Reactive oxygen species (ROS) activate MMPs [9], suggesting a possible role of oxidative stress in the pathogenesis of aortic aneurysms.

Oxidative stress induced by ROS plays an important role in the pathogenesis of a variety of cardiovascular diseases, including atherosclerosis, hypertensive vascular disease, and coronary artery disease [10,11]. The redox state is finely tuned to preserve cellular homeostasis through the expression of oxidant and antioxidant enzymes. The mechanisms of ROS production in non-phagocytic cells are not fully understood; however, it has become clear that NADH/NADPH oxidase plays a prominent role as the source of ROS [12]. On the other hand, angiotensin II (Ang II) exerts numerous biological effects on the cardiovascular system, including vascular hypertrophy, cell proliferation, migration, induction of cytokines, production of extracellular matrix, and activation of macrophages, and it activates NADH/NADPH oxidase in cultured vascular smooth muscle cells (SMCs) [13]. Furthermore, Ang II increases mRNA levels of p22phox and p67phox in vivo and in vitro [14]. Thus, Ang II is not only a potent stimulator of NADH/NADPH oxidase, but also an inducer of its components. The administration of Ang II induces aortic aneurysms in apoE-knockout mice [15], suggesting involvement of the renin–angiotensin system in the pathogenesis of the disease. Therefore, we hypothesized that NADH/NADPH oxidase plays a role in the pathogenesis of aortic aneurysms via oxidative stress. To prove this hypothesis, we investigated the localization of p22phox and its expression differences in aneurysmal and non-aneurysmal aortae and in situ generation of ROS. Furthermore, the influence of medical treatment on aortic p22phox expression was examined.

2. Methods

2.1. Human tissue

Thoracic aneurysmal aortic sections were obtained from patients undergoing aneurysm repair surgery (n=40). Control (non-dilated) aortic specimens were obtained from patch-sections of the ascending aorta at coronary artery bypass graft surgery (CABG) or aortic valve replacement surgery (n=39) (Table 1). Since the structure of the aorta is not uniform and the physical stress is different along its length, these specimens were used as control in our study. In the operation room the tissues were rapidly immersed in physiological saline on ice and washed. For immunohistochemistry and fluorescent microtopography examinations, they were embedded in OCT compound and snap frozen in liquid nitrogen and stored at −80°C until use. For protein extraction, they were rapidly washed and immersed in liquid nitrogen. All tissues were taken in accordance with a protocol approved by the Kobe University of Medicine Human Research Committee, and informed consent was obtained from all patients.

2.2. Immunohistochemistry

Immunohistochemistry was carried out as previously described [16]. In brief, serial cryostat sections (6 μm) were fixed with 100% acetone at −20°C for 10 min, blocked with carrier protein (Dako LSAB kit™, Dako A/S) for 30 min at room temperature, and then incubated with a primary antibody overnight at 4°C. The primary antibodies were rabbit polyclonal anti-human p22phox antibody against the synthetic peptide corresponding to the

### Table 1

<table>
<thead>
<tr>
<th>Clinical characteristics of patients</th>
<th>Control (n=39)</th>
<th>TAA (n=40)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, yr</td>
<td>69.9±1.1</td>
<td>71.7±1.5</td>
</tr>
<tr>
<td>Sex, %male</td>
<td>72</td>
<td>70</td>
</tr>
<tr>
<td>Size, cm</td>
<td>&lt;3.0</td>
<td>6.0±0.2</td>
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<tr>
<td>C-reactive protein, mg/dl</td>
<td>0.84±0.23</td>
<td>0.99±0.35</td>
</tr>
<tr>
<td>Systolic pressure, mmHg</td>
<td>127±2.9</td>
<td>128±1.7</td>
</tr>
<tr>
<td>Diastolic pressure, mmHg</td>
<td>71±1.8</td>
<td>72±1.4</td>
</tr>
<tr>
<td>Risk factors</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hypertension, n (%)</td>
<td>33 (85)</td>
<td>33 (83)</td>
</tr>
<tr>
<td>Diabetes, n (%)</td>
<td>22 (56)</td>
<td>11 (28)*</td>
</tr>
<tr>
<td>Hyperlipidemia, n (%)</td>
<td>17 (44)</td>
<td>16 (40)</td>
</tr>
<tr>
<td>Smoking, n (%)</td>
<td>21 (54)</td>
<td>23 (58)</td>
</tr>
<tr>
<td>Medical treatment</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ca antagonist, n (%)</td>
<td>17 (44)</td>
<td>26 (65)</td>
</tr>
<tr>
<td>ACE-inhibitor, n (%)</td>
<td>10 (26)</td>
<td>13 (33)</td>
</tr>
<tr>
<td>ARB, n (%)</td>
<td>5 (13)</td>
<td>11 (28)</td>
</tr>
<tr>
<td>β-blocker, n (%)</td>
<td>10 (26)</td>
<td>12 (30)</td>
</tr>
<tr>
<td>α-blocker, n (%)</td>
<td>7 (18)</td>
<td>5 (16)</td>
</tr>
<tr>
<td>Statin, n (%)</td>
<td>8 (20)</td>
<td>11 (28)</td>
</tr>
</tbody>
</table>

Clinical characteristics of patients: control and aneurysm groups. There was no significant difference between the control and aneurysm groups regarding with age, sex serum C-reactive protein level, and major risk factors except for diabetes. TAA=thoracic aortic aneurysms. ACE-inhibitor=angiotensin converting enzyme inhibitor, ARB=angiotensin II type I receptor blocker. Results are expressed as mean±SEM. * P<0.05 versus control.
carboxyterminal (residues 175 to 194), mouse monoclonal anti-human angiotensin converting enzyme (ACE) antibody, and mouse monoclonal anti-human chymase antibody. The sections were washed with Tris-based buffer, incubated with biotinylated goat anti-rabbit immunoglobulins (Dako), washed again in Tris-based buffer, and finally incubated with streptavidin horseradish peroxidase conjugate (Dako LSAB kit™, Dako). For negative control, the primary antibody was replaced with rabbit nonspecific immunoglobulin. To identify the type of p22phox-expressing cells, cellular markers in the tissues were analyzed by immunohistochemistry with monoclonal antibodies. The cell-specific antibodies were mouse monoclonal anti-human CD68 antibody (clone KP-1, Dako) for macrophages and mouse monoclonal anti-human α-actin antibody (clone 1A4, Dako) for SMCs.

2.3. Quantitative analysis of p22phox in immunohistochemistry

Six cross-sections were obtained from another two portions of each aneurysmal wall, stained and examined. The sections were blindly graded by three independent senior pathologists. All aortic sections were also digitized by a digital camera, and examined in detail regarding the tunica media. The surface area occupied by p22phox was outlined by computer-aided planimetry with the use of image analysis software (Macscope, Mitani Co). The positive immunoreactivity areas were measured automatically by using color-scale detection with a fixed threshold. The relative expression of p22phox was expressed as the ratio of positive area to total area of the tunica media in each section.

2.4. Western blot analysis

A portion of the aortic tissues was homogenized with a buffer containing protease inhibitors (50 mmol/1 Tris/HCl, pH 7.4, 1 mmol/l EDTA, 500 μmol/l phenylmethylsulfonyl fluoride (PMSF), 2 μmol/l leupeptin, and 10 μg/ml aprotinin), centrifuged, and the supernatant collected. Equal amounts of protein (50 μg protein per lane), estimated by the method of Bradford (Bio-Rad), were in situ Zymo-Film (Wako Pure Chemical Industries, Ltd), electrophoretically transferred onto a polyvinylidene difluoride membrane (Milli-pore™). The membrane was blocked with the buffer (10 mmol/l Tris/HCl, pH 7.4, 100 mmol/l NaCl, and 0.1% Tween-20™) containing 5% skim milk at room temperature for 1 h and incubated with anti-human p22phox antibody at 4°C overnight, then incubated with horseradish peroxidase-labeled donkey anti-rabbit immunoglobulin antibodies (Amersham) at room temperature for 1 h. The signals were detected by the enhanced chemiluminescence method (Amersham). Analysis of densitometry was carried out with NIH Image software, version 1.62.

2.5. Evaluation of ROS in aorta by dihydroethidium and lucigenin

Dihydroethidium oxidative fluorescence dye was used to evaluate the in situ production of ROS, as previously described [17]. Unfixed frozen samples cut into 20 μm thick sections were placed on glass slides, augmented with dihydroethidium (10 μmol/l), and then capped with coverslips. The slides were incubated in a light-protected humidified chamber at 37°C for 30 min. The dihydroethidium image was obtained by a laser scanning confocal imaging system (MRC-1024) equipped with a 585-nm long-pass filter.

Furthermore, superoxide production from aortic tissue was measured with the use of lucigenin chemiluminescence according to a method modified from that of Münzel et al. [18]. Briefly, specimens of thoracic aorta were incubated with 10 mM diethyldithiocarbamate (Sigma), an inhibitor of copper zinc superoxide dismutase, for 30 min in Hepes-buffered physiological saline solution and then gently transferred to vials containing 5 μmol/l lucigenin (Sigma); this concentration of lucigenin accurately reflects levels of ambient superoxide and is not subject to the artifactual production of superoxide observed with higher concentrations of the agent [19,20]. The light reaction between superoxide and lucigenin was detected using a chemiluminescence reader (BLR-201, Aloka). The chemiluminescence signal was expressed as average counts per minute for 15-min periods per mg dry tissue. Superoxide levels were also measured in the presence of 10 μM diphenylene iodium (DPI), a selective inhibitor of flavin-containing enzyme including NADH/NADPH oxidase.

2.6. In situ zymography

To examine the interaction of p22phox and the activities of MMPs, in situ zymography was carried out with MMP in situ Zymo-Film (Wako Pure Chemical Industries, Ltd), composed of a 7 μm thick layer of special gelatin on a polyester. Unfixed frozen samples were cut into 5 μm thick sections, placed on the films and incubated at 37°C in a humidified chamber for 24 h. The films were then dried at room temperature for 30 min, stained with Biebrich Scarlet Stain Solution for 4 min, washed, and then counter-stained with Mayer’s hematoxylin solution. The films were thus stained, leaving light colored the areas where gelatin was digested with proteases. Furthermore, to
distinguish MMP activity from other protease activities, we also used MMP-PT in situ Zymo-Film (Wako Pure Chemical Industries, Ltd), pre-coated with gelatin and 1,10-phenanthroline as an MMP inhibitor.

2.7. Statistics

Results are expressed as the mean±S.E.M. \( P<0.05 \) was considered statistically significant. For comparison of clinical and immunohistochemical data, the unpaired Student’s \( t \)-test was used for continuous data, and the Mann–Whitney \( U \)-test was used for nonparametric continuous data. For comparison of continuous data within the four groups (control-advanced, control-early, TAA-early, TAA-advanced), one-way analysis of variance and a post hoc multiple comparisons test (Fisher’s) were used. Univariate and multivariate regression analyses were performed to discern which medical treatment (Ca antagonist, Ang II type 1 receptor blocker, ACE inhibitor, \( \beta \)-blocker, or \( \alpha \)-blocker) was associated with a variety of \( p22^{\text{phox}} \) expression in the aneurysmal aortae. Simple linear regression analysis was performed to study the relation between continuous variables.

3. Results

3.1. Clinical characteristics of patients and histological findings

The clinical profiles of patients are shown in Table 1. All sections were examined by hematoxylin–eosin staining. Intimal thickening was observed in all specimens of control and aneurysmal aortae. In the segments of the aneurysmal aortae, additional pathological changes, including thinning of the media followed by loss of SMCs, and intimal disorganization, were observed. The majority of intima and media of aneurysmal aortae were hypocellular, and these regions consisted of dense connective tissue and a few SMCs. In some aneurysmal aortae, however, dramatic changes were observed in the cellular composition of the aortic wall associated with massive infiltration of mononuclear cells in the adventitia and the external layer of the media.

3.2. Enhanced generation of ROS in TAA

In situ generation of ROS was examined by fluorescence microtopography with dihydroethidium. Weak ROS signals were detected in the endothelium, intima, media, and adventitia of control aortae (Fig. 1A). In contrast, intense production of ROS was observed in all layers of TAA. The

Fig. 1. In situ detection of superoxide in the control and aneurysmal aortae. Fluorescence of confocal microscopic sections of aorta labeled with the oxidative dye dihydroethidium (red fluorescence when oxidized to ethidium bromide by superoxide). (A) Weak superoxide signals were detected in the endothelium, intima, and media of the control aortae. (B, C) Intense production of superoxide was observed in all layers of the aneurysmal aortae. The generation of superoxide was markedly increased in hypercellular regions containing inflammatory cells (arrows). i, intima; m, media; a, adventitia; TAA, thoracic aortic aneurysm. Bars: 200 \( \mu \text{m} \).

Fig. 2. Expression of NADH/NADPH component \( p22^{\text{phox}} \) was increased in TAA compared with the control. Serial cryostat sections (6 \( \mu \text{m} \)) were incubated with anti-human \( p22^{\text{phox}} \) antibody overnight at 4\(^\circ\)C. In control aortae (A, B, C), weak but positive immunoreactivity of \( p22^{\text{phox}} \) was observed mainly in the adventitia and neointima, but scarcely in the media. In the aneurysmal aortae (D, E, F), the immunoreactivity of \( p22^{\text{phox}} \) was more intense than that in the control aortae. (F) In the hypercellular regions, the expression of \( p22^{\text{phox}} \) was especially marked. (E) \( p22^{\text{phox}} \) was also detectable in the hypocellular regions of the media. (B, C, E, F) Higher magnification boxed areas in panels (A) and (D), respectively. Bars: 200 \( \mu \text{m} \) (A, D) and 50 \( \mu \text{m} \) (B, C, E, F).
3.3. Expression of NADH/NADPH oxidase p22\textsuperscript{phox} in TAA

In control aortae, weak but positive immunoreactivity of p22\textsuperscript{phox} was observed mainly in the adventitia and neointima, but scarcely in the media (Fig. 2A–C). It was more intense in TAA than in the control aortae (Fig. 2D–F). In the hypercellular regions, the expression was especially marked (Fig. 2F). Even in the hypocellular portions of TAA, positive immunoreactivity was detectable in SMCs (Fig. 2E). When the primary antibody was replaced with rabbit non-specific immunogloblin, no reactivity was detected.

To investigate the influence of the degree of atherosclerosis, we compared the expression of p22\textsuperscript{phox} in aortic specimens having a similar degree of atherosclerosis. According to the American Heart Association (AHA) classification [21], all specimens were divided into early (class I–III) and advanced stage (IV–VI) atherosclerosis. As shown in Table 2, the degree of atherosclerosis influenced the expression of p22\textsuperscript{phox}. However, in the early and advanced stages, expression in the aneurysmal aorta was much higher compared with the non-aneurysmal aorta.

Western blot analysis confirmed an approximately 5.5-fold enhanced expression of p22\textsuperscript{phox} in aneurysmal aortae compared with the control (Fig. 3A and B).

To identify the types of p22\textsuperscript{phox}-expressing cells, immunohistochemistry of serial sections and double labeling immunofluorescence with cell type specific antibodies were carried out. Most of the p22\textsuperscript{phox}-expressing cells in the atheromatous neointima were positive for CD68, and some were positive for \(\alpha\)-actin. Mononuclear inflammatory cells infiltrated in the media and adventitia also expressed p22\textsuperscript{phox}, and those cells were mainly positive for CD68 and CD3.

3.4. Relation between NADH/NADPH oxidase p22\textsuperscript{phox} and the renin–angiotensin system

To examine the relationship between the renin–angiotensin system and p22\textsuperscript{phox} expression, immunohisto-
Fig. 5. Chymase-positive cells were increased in the aneurysmal aortae. The number of chymase-positive cells in the aneurysmal aorta was significantly higher than that in the control (A, B). The arrows show chymase-positive cells. Chymase-positive mast cells did not express p22phox (C, D, E), but the localization was closely associated with p22phox. m, media; a, adventitia. Mann–Whitney U-test: *P<0.05 versus control.

3.5. Association between MMP activity and p22phox-expressing cells

The activity of MMPs is an important determinant of the integrity of the extracellular matrix and stiffness of the aortic wall, and ROS activate MMPs [9]. Therefore, the relation of NADH/NADPH oxidase p22phox to the activity of MMPs in TAA was examined. In situ zymography demonstrated that the activity of MMPs was markedly increased in the aneurysmal aortae compared with the control (Fig. 6). Interestingly, the immunohistochemical analysis of serial sections demonstrated that the activity of MMPs almost overlapped the immunoreactivity of p22phox.

3.6. Medical treatment with statin and angiotensin receptor antagonist decreased oxidative stress in TAA

Of 40 patients with TAA, 36 had received one or several anti-hypertensive drugs, and 11 had received statin therapy before operation (Table 1). First, univariate regression analysis was performed to discern which medical treatment was associated with a variety of p22 expression in the aneurysmal aortae. In the univariate model, there was an

<table>
<thead>
<tr>
<th>Independent factors</th>
<th>β</th>
<th>P value</th>
</tr>
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<tbody>
<tr>
<td>Ca antagonist</td>
<td>−3.274</td>
<td>0.1974</td>
</tr>
<tr>
<td>ARB</td>
<td>−7.993</td>
<td>0.0056</td>
</tr>
<tr>
<td>ACE-inhibitor</td>
<td>−0.743</td>
<td>0.7443</td>
</tr>
<tr>
<td>β-blocker</td>
<td>−4.712</td>
<td>0.0681</td>
</tr>
<tr>
<td>α-blocker</td>
<td>1.813</td>
<td>0.6017</td>
</tr>
<tr>
<td>Stain</td>
<td>−5.609</td>
<td>0.0297</td>
</tr>
</tbody>
</table>

r²=0.450, β=coefficient values in multiple regression analysis. Multiple regression analysis was carried out to evaluate the relative impact of medication on p22 phox expression. Medical treatment with statin or ARB suppressed the expression of p22 phox in the aneurysmal aorta.
inverse association between p22phox expression and the following medications, β-blocker (r = -6.18, P = 0.0211), Ang II type 1 receptor blocker (ARB) (r = -7.28, P = 0.0096) and statin (r = -7.09, P = 0.0093). On the other hand, ACE inhibitor (r = -0.32, P = 0.9035), α-blocker (r = -3.41, P = 0.3723), and Ca antagonist (r = -0.89, P = 0.7429) had no significant influence on the expression of p22phox. Next, we also performed multivariate regression analysis to evaluate the relative impact of medication on p22phox expression. As shown in Table 3, ARB (r = -7.993, P = 0.0056) and statin (r = -5.609, P = 0.0297) significantly suppressed the expression of p22phox in the aortic wall, but β-blocker (r = -4.712, P = 0.0681) did not reach significance (Table 3).

4. Discussion

In the present investigation, we demonstrated that the generation of ROS in the aneurysmal aorta was markedly increased compared with the control. The enhancement of ROS was associated with the expression of NADH/NADPH oxidase p22phox. Although the degree of atherosclerosis had an influence on the expression of p22phox, its expression in the aneurysmal aorta was much higher than that in the non-aneurysmal aorta in the early as well as the advanced stage. These increases in ROS and the expression of p22phox were especially pronounced in the hypercellular region of the aneurysms. Chymase-positive mast cells did not express NADH/NADPH oxidase p22phox; however, a close association of the immunoreactivity of p22phox and ACE or chymase was observed, indicating an interaction between p22phox-based NADH/NADPH oxidase and the renin–angiotensin system. Furthermore, in situ zymography demonstrated that the activity of MMPs in the aneurysmal aortae overlapped this oxidase.

Multiple interacting factors are probably involved in the pathogenesis and mechanisms of aortic aneurysmal formation such as hemodynamic forces, transmural inflammation, apoptosis, and destructive remodeling of the extracellular matrix [1–4]. MMPs are important enzymes responsible for extracellular matrix degradation by proteases derived from inflammatory cells infiltrating aortic walls [5–8]. In the present study, the production of ROS and the expression of p22phox increased in the aneurysmal aortae, and the in situ activity of MMPs clearly overlapped. It is interesting to speculate that ROS derived from p22phox-based NADH/NADPH oxidase can activate MMPs, and this activation may, in turn, contribute to destructive remodeling. On the other hand, recent investigations indicate that ROS are crucial pro-apoptotic factors. Therefore, ROS generated by p22phox-based NADH/NADPH oxidase might activate aortic MMP as well as promote apoptosis, with these changes contributing to the formation of aneurysms. However, we have to admit that our results do not prove a causal relation. It is important to determine the effects of antioxidative agents on the reversibility or prevention of aneurysmal formation, and the establishment of a cause–effect relation will be a major focus of our future research.

Ang II exerts a wide variety of biological effects on vascular cells, including proliferation, migration, vascular hypertrophy, vascular remodeling, and atherogenesis. The local formation of Ang II is mediated by ACE, or by a number of serine proteinases such as kallikrein, cathepsin G, and chymase. A recent investigation by Ihara et al. indicated that the enhanced formation of Ang II in atherosclerotic lesions was mediated by the action of chymase [22]. Furthermore, they demonstrated that ACE immunoreactivity was seen in endothelial cells of the vasa vasorum in the adventitia as well as in macrophages of the neointimal lesions of aneurysmal aortae, and that chymase immunoreactivity appeared in mast cells in aneurysmal aortae but not in normal or atherosclerotic aortae [22]. Furthermore, Leskinnen et al. reported that mast cells induced apoptosis in cultured SMCs [23], suggesting that mast cells might participate in the apoptotic regulation of SMCs in atherosclerotic and aneurysmal formation. Our findings are consistent with theirs, and the present investigation was extended to the association with oxidative stress. Interestingly, ACE-positive macrophages were positive for p22phox; however, chymase-positive mast cells did not express p22phox, and ACE-positive endothelium of microvessels did not express p22phox. Although overlapping was not complete, its local association was clearly observed.

In the present investigation, we found that the number of diabetic patients in the aneurysm group was less than that in the control group. Diabetes has, in general, a deleterious influence on the pathogenesis of vascular disease. However, many previous epidemiological investigations indicated that diabetes was a negative risk factor for aortic aneurysm, although the precise mechanisms remain undetermined [24,25]. Our results confirmed these previous results. The presence of diabetes might influence the results; however, there was no correlation of these variables, indicating that the diabetic state had no effect in the present study.

Currently, the management of small thoracic aneurysms (ascending <5.5 cm, descending <6.5 cm) and small abdominal aneurysms (<5.5 cm) is controversial, because the risk of their rupture within a short period of time is low [26–29]. Were oral medications that inhibited aneurysm expansion with a good safety profile available, pharmacological treatment of small aneurysms would be possible. Various drugs, including anti-inflammatory ones such as glucocorticoids, leukocyte-depleting (anti-CD18) antibody and indomethacin, or MMP inhibitors, have been tested experimentally for the treatment of aortic aneurysms [30–
In the present study, we revealed that medical treatment with ARB, but not with ACE inhibitors, significantly suppressed the expression of p22^phox^ in the aortic wall. These findings suggest that the Ang II type 1 receptor-mediated pathway plays a potent role in the generation of ROS in the aortic wall, and that non-ACE serine proteinases such as chymase are likely involved in this process. Also, statin therapy suppressed p22^phox^ protein expression in the aortic wall. This observation is consistent with Wassmann's result that statin suppresses the expression of NADH/NAPD oxidase in cultured vascular cells [33]. Thus, statin and ARB might directly suppress the expression of aortic NADH/NAPD oxidase. In univariate regression analysis, there was an inverse association between p22phox and β-blocker. In our investigation, 13 patients were treated with β-blockers, and two of these patients were administered with carvedilol, which has antioxidative properties. The direct effects of this drug on aneurysmal formation deserve further investigation.

In conclusion, p22^phox^-based NADH/NAPD oxidase and the local renin–angiotensin system play a notable role in the pathogenesis of thoracic aortic aneurysms. Statin and ARB might have inhibitory effects on aneurysmal formation via the suppression of NADH/NAPD oxidase.

References


