

## Peer Review File

Article information: <https://dx.doi.org/10.21037/jtd-24-206>

### Reviewer A

Comment 1: The authors should be congratulated for their efforts in proving the validity of their decision regarding emergent operation for type A acute aortic dissection. The paper is well-written and logical. They assert that the choice between supra-aortic replacement or aortic root replacement procedures depends on the enlargement of the STJ or the presence of aortic regurgitation. My understanding is that histopathology during emergent surgery does not influence their decision in such situations, as surgeons must determine the procedure to save the patient's life within strict time constraints without waiting for histopathology results. Please explain the authors' perspective on future decisions about the procedure.

Reply 1: Thank you for the kind and thoughtful words. We completely agree with the Reviewer A that during emergent surgery, histopathology does not influence decision-making. Instead, our study shows that data obtained from histopathology of the diseased aorta increases understanding of type A acute aortic dissection and its consequences while some parts of the aorta remains unoperated after surgery. We envisage that histopathology of the aorta tissue resected from the ascending aorta may add to follow-up of the patient after initial surgery for type A acute aortic dissection. Increased degeneration of the diseased aorta upon histopathology may alert awareness of the association of the ascending portion with the extent of aorta disease involvement. This study enhances awareness of type A acute aortic dissection and aorta wall histopathology. In future, investigating aortic wall degeneration preoperatively may guide surgical decision-making and follow-up of the patients, as also commented by Reviewer B; these preoperative means are yet to be explored, however.

Changes in the text 1: We added an additional sentence in Conclusions, line 324, text in red: “Understanding the features of the dissected aorta may facilitate follow-up of the patients in the future”.

### Reviewer B

This study explores the relationship between ascending aortic wall degeneration and the extent of aortic root disease in patients undergoing surgery for ATAAD. Analyzing 141 patients, it compares those who had aortic root replacement with those undergoing root-sparing surgery. Histopathological analysis revealed that mucoid extracellular matrix accumulation—a marker of degeneration—was more pronounced in patients requiring aortic root replacement. Despite similar survival rates between groups, the findings underscore the significance of aortic wall histology in surgical decision-making, suggesting that degeneration extent can guide the choice between root replacement and sparing strategies in ATAAD treatment. I would like to congratulate the authors on this interesting manuscript. The topic is relevant. I have the following comments and questions:

---

Comment 1: The study “investigated whether the presence of ascending aortic wall degeneration in ATAAD patients would differ in patients with proximal extension of surgery encompassing the

aortic root compared to patients with aortic root-sparing surgery”. I am not certain about the clinical question asked in the study. What is the mechanistic plausibility to consider a difference in the aortic wall degeneration between these 2 procedures.

Reply 1: As rightfully hinted by the Reviewer, surgical decision-making to include aortic root replacement is a multifaced process during ATAAD. This is a hypothesis-based observational study; aortic root replacement patients were considered to have extended aortic dissection, and aortic wall histopathology was compared with patients either with or without aortic root replacement. The presence of ascending aortic wall degeneration was increased in patients with a surgical decision to include replacement of the aortic root suggesting that local degree of aortic wall degeneration is associated with extension of the disease as well. It remains to be investigated whether the ascending aorta wall degeneration is associated with increased aorta diameter that further distends the sinotubular junction and the aorta root diameter.

Changes in the text 1: Please observe Reply 1 for Reviewer A. We added an additional sentence in Conclusions, line 324, text in red: “Understanding the features of the dissected aorta may facilitate follow-up of the patients in the future”.

---

Comment 2: Relevant to above, the decision to perform root replacement or sparing surgery is multifaceted, influenced by factors beyond histopathological findings, including patient-specific anatomical and clinical considerations. The study may not fully account for these complexities, potentially oversimplifying the decision-making process.

Reply 2: As rightfully stated by the Reviewer, we completely agree that surgical decision-making to include aortic root replacement is a multifaced process during ATAAD. Nevertheless, those patients with increased ascending aorta wall degeneration had more often a surgical decision of having aortic root replacement. Our emergent surgical decision-making to replace the aorta root during ATAAD is based on many factors including patient characteristics and clinical considerations, but without endangering the emergent clinical set-up and always taking care of not increasing the complexity of surgery. We envisage that many aortic root surgeries are offered to patients only if necessary.

Interestingly, according to the adjusted multivariable binary logistic regression analysis, the extent of mucoid extracellular matrix accumulation (odds ratio [OR]: 9,981, 95% confidence interval [CI]: 1.832–54.363,  $P = 0.008$ ) was a significant factor related to patients with aortic root replacement, when confounders such as presence and location of intimal tear, aortic valve regurgitation, age, male sex, presence of bicuspid aortic valve and aortic diameter were taken into account.

Changes in the text 2: Please see Table 1 and 2 and Results, 4th paragraph, text in red color, Statistical analysis, and Discussion, paragraph 6, lines 294-297. Please see Limitations of the study, first sentence.

Comment 3: Histopathological findings are the main focuses of the study, and they are obviously influenced by the status of the aorta, which may include presence of aortic dissection on the specimen, distance from the primary tear, presence of clot or flow in the false lumen, precise

location of the aorta, timing from onset to surgery, preexisting aneurysmal change, presence and degree of aortic regurgitation, as well as characteristics of the patients. Please provide as much data as able. Additional analyses adjusting for these relevant confounders would be interesting.

Reply 3: We fully agree with the Reviewer that additional analyses adjusting for all relevant details pertinent to ATAAD enhances interest. All patients were operated on emergent basis in less than 2 weeks of the onset of symptoms (Results, 1st paragraph), irrespective of the presence of clot or false lumen flow. Preexisting aneurysmal change was confirmed in 17 patients. Presence of aortic regurgitation was reported whenever it was moderate to severe according to a semiquantitative clinical grade of 2 or 3 out of 3, when 0 meant no regurgitation. Characteristic of the patients (Table 1) reflect the heterogeneity of patient presentation.

As advised by the reviewer, we provide additional data on entry tears and aortic diameters after studying retrospectively the patient charts and computer tomography images. We performed binary logistic regression models to further analyze clinically relevant confounders associated with aortic root involvement. According to binary logistic regression after adjusting for presence and location of entry tear, aortic valve regurgitation, age, male sex, presence of bicuspid aortic valve and aortic diameter, increased extent of mucoïd extracellular matrix accumulation remained an independent variable for need of aortic root replacement.

Changes in the text 3: Please see Table 1 and 2 and Results, 2<sup>nd</sup> paragraph, text in red color. In addition, the following text was added to Statistical analysis: Multivariable binary logistic regression analysis was performed to analyze the association of extent of mucoïd extracellular matrix accumulation adjusted for clinically relevant confounders with aortic root involvement. These confounders included presence and location of entry tear, aortic valve regurgitation, age, male sex, presence of bicuspid aortic valve and aortic diameter. Please see Results, paragraph Roc analysis and multivariable binary logistic regression analysis: According to the adjusted multivariable binary logistic regression analysis, the extent of mucoïd extracellular matrix accumulation (odds ratio [OR]: 9,981, 95% confidence interval [CI]: 1.832–54.363,  $P = 0.008$ ) was a significant factor related to patients with aortic root replacement.

Comment 4: While the study highlights the role of mucoïd extracellular matrix accumulation, it may oversimplify the complex nature of aortic wall degeneration. A more detailed analysis encompassing a wider range of histopathological features could provide a richer understanding of disease mechanisms. Please comment.

Reply 4: Please see Reply 3 above. Mucoïd extracellular matrix accumulation is one of the histological main parameters that describe the degree of aorta wall degeneration. A more detailed analysis encompassing a wider range of histopathological features from those recommended by Haluska et al remains beyond the scope of this study. However, it is interesting that with the use of a simple histopathological description of the aortic wall, one may observe different patterns of ATAAD extension. We have now, included Multivariable binary logistic regression analysis adjusted for clinically relevant confounders with aortic root involvement and ROC analysis to show that the predictive value of the extent of mucoïd extracellular matrix accumulation, together with clinically relevant variables, such as presence and location of intimal tear, aortic

valve regurgitation, age, male sex, presence of bicuspid aortic valve and aortic diameter, to identify patients with aortic root replacement was significant.

Changes in the text 4: Please see changes in the text 3 above.

Comment 5: The comparative analyses were done without adjusting for confounders. I suggest modifying it.

Reply 5: As stated above, this is an excellent suggestion, please see Comment 3 and Reply 3 above. According to the adjusted multivariable binary logistic regression analysis, the extent of mucoid extracellular matrix accumulation (odds ratio [OR]: 9.981, 95% confidence interval [CI]: 1.832–54.363,  $P = 0.008$ ) was a significant factor related to patients with aortic root replacement.

Changes in the text 5: Please see changes 3

### **Reviewer C**

This article investigates the association between histological degeneration of the ascending aortic wall and proximal extension to the aortic root in the ATAAD. This research is of mild to moderate interest and scientific importance. My questions and suggestions are listed below.

#### **Major**

Comment 1: First, I am wondering about your thoughts on aortic wall degeneration and the extent of dissection. Do you think that in the case of less degenerated ascending aorta, there is no extension of degeneration in the aortic root, and thus the dissection does not involve the aortic root? In other words, the dissection does not extend to the aortic wall without degeneration. Or, in the case of less degeneration in the ascending aorta, the less dissection extends, and, dissection does not necessarily extend to degenerated aorta. Depending on the context, I think you have the former idea. Let me know your thoughts.

Reply 1: As this observational study is retrospective, we may only show association of aortic wall degeneration with the surgical decision of encompassing the aortic root resection and replacement together with the ascending aorta replacement. Decision-making for aortic root replacement is obviously a multifaceted process; it is tempting to speculate that aortic wall degeneration reflects the weakness of the aorta that is susceptible to dilate, leading to the dilation of the sinotubular junction, and further weakness of the aorta root wall.

Changes in the text 1: Please see Limitations paragraph and text in red, and Reply 3, Reviewer B.

Comment 2: Abstract, conclusion: "Surgical assessment of disease extension in ATAAD justifies a limited resection strategy in many patients". This study only shows an association between the degree of mucoid extracellular matrix accumulation assessed postoperatively and the choice of surgical procedure, and did not compare individual assessment with intraoperative histologic assessment for decision-making. Therefore, such conclusions cannot be drawn. In addition, some papers suggest to avoid speculation in the abstract. Please pay attention to the author guidelines

of this journal. Similarly, lines 186 and 202, please reconsider the clinical significance of this study and draw the conclusions.

Reply 2: Thank you for this important specification. Please see previous Reply 2 above, and Reply 3, Reviewer B. We completely agree that we may only show association of aortic wall degeneration with the surgical decision of encompassing the aortic root resection and replacement together with the ascending aorta replacement.

Changes in the text 2: We revised the sentence as follows “The degree of mucoid extracellular matrix accumulation assessed postoperatively is associated with the choice of surgical procedure in many patients.” Abstract, conclusion, last sentence.

Comment 3: Line 47. “As the emergency ATAAD surgery strategy includes the evaluation of tissue destruction, the objective of this study was to evaluate the plausible association of aortic wall degeneration and the extent of proximal aortic disease” Why do the authors only hypothesize an association between the aortic wall degeneration and proximal extension of acute dissection, but not distal extension?

Reply 3: The definition of proximal extension of disease includes the aortic root that encompasses the aorta proximal to the very clear anatomical border of the sinotubular junction. According to previous studies, the root is often defining the extent of the aortic dissection (root phenotype). The distal extension and limits of the disease is more difficult to define, and the aortic tissue beyond the aortic arch may embryologically differ from the ascending aorta.

Changes in the text 3: We now include additional information on presence and location of intimal tears and the distal aortic extension (Table 2), and Results, paragraph 2.

Comment 4: Line 73. As this is one of the most important parts of the outcomes of interest, please provide more detailed information on Histology and immunohistochemistry and Quantification of Medial Degeneration.

How is each variable evaluated? For example, Hematoxylin and Eosin versus Verhoeff-van Gieson versus Alcian Blue and periodic acid-Schiff stains, and definition/criteria of severity. Who evaluated the histological findings?

Is it a blind fashion?

“Medial degeneration of the ascending aorta was assessed by quantifying 11 different variables that describe medial and adventitial damage (7,8). These included overall medial degeneration, mucoid extracellular matrix accumulation, elastic fiber loss/fragmentation, elastic fiber thinning, elastic fiber disorganization, smooth muscle cell nuclei loss, laminal medial collapse, smooth muscle cell disorganization, medial fibrosis, and adventitial fibrosis”. It appears that only 10 variables were included in the sentence. Also, Table 3 seems to include more variables without definition. It is very confusing.

Please provide the representative and comparative figures in Figure 1 in both groups, especially, indicating the difference in mucoid extracellular matrix accumulation.

Reply 4: The aortic specimens were assessed as a part of routine surgical pathology evaluation according to guidelines of The Society for Cardiovascular Pathology and The Association for

European Cardiovascular Pathology (7,8) by two experienced cardiovascular pathologists (IK, TP). At least 18 sections (six stained with hematoxylin-eosin, six stained with Verhoeff-van Gieson stain and six with Alcian Blue and periodic acid-Schiff) were evaluated in each case. The assessed features included overall medial degeneration, mucoid extracellular matrix accumulation, elastic fiber loss/fragmentation, elastic fiber thinning, elastic fiber disorganization, smooth muscle cell nuclei loss, laminal medial collapse, smooth muscle cell disorganization, medial fibrosis, vasa vasorum medial thickening, and adventitial fibrosis. All features were assessed for grade (absent, mild, moderate, and severe) and distribution (focal, multifocal, and extensive). The worst present grade/distribution was reported and semi-quantified on a scale of 0-3 (8).

Immunohistochemistry was used to characterize the inflammatory infiltrate (CD3 as marker for T-lymphocytes, CD68 as macrophage marker and CD20 as B-lymphocyte marker). Hematoxylin-eosin was used for overall evaluation, inflammation assessment and evaluation of smooth muscle cell nuclei. Verhoeff-van Gieson was applied in assessment of elastic fibers including laminal medial collapse. Alcian Blue and periodic acid-Schiff was used for mucoid extracellular matrix accumulation assessment and fibrosis evaluation. The presence of ascending aorta atherosclerosis was categorized as mild, moderate, severe, atherosclerosis with plaque disruption including surface thrombus, and calcific atherosclerosis was reported. The pattern of ascending aorta inflammation was defined as granulomatous including giant cells, lymphoplasmacytic, mixed inflammatory and suppurative.

Changes in the text 4: Please see added text in the revised paragraphs for Histology and immunohistochemistry, and Quantification of Medial Degeneration.

Comment 5: Line 126. Did all patients undergo hemi-arch replacement, but not partial/total aortic arch replacement? In the Surgery subsection in the Methods section, you mentioned that “the aortic arch was resected completely or a hemiarch fashion depending on the involvement of aortic wall disease”. I think there is a contradiction here. In addition, the entry site and distal extension of the dissection are important factors in this topic and should be provided.

Reply 5: There were 38 patients with hemi-arch replacements and 12 total arch replacements. In 19 patients, the entry tear was situated in the aortic arch, while there were 73 patients with entry tears in the ascending aorta. There were 49 unreported or unknown sites of entry tears; many entry tear locations are difficult to confirm once the ascending aorta is transected during ATAAD. We have previously reported that the entry tear alone may not always determine the distal site of anastomose during surgery for ATAAD (Uimonen et al, *Annals Thor Surg*, 2022).

Changes in the text 5: Please see paragraph Operative technique and Table 2 for added information in red color, added new reference #18, and Limitations paragraph, text in red.

Comment 6: Line 136. Histological findings in six patients who required proximal root reoperation will be very informative. Any significant characteristics in the histological findings of those patients?

Reply 6: There were 5 patients with root-sparing surgery who required reoperation for proximal root replacement and one patient with a biological root replacement that required a reoperation for a second root replacement due to root pseudoaneurysm. The extent of elastic fiber fragmentation and/or loss was increased in the five patients needing reoperations after root-sparing surgery as compared to those without need of reoperations for the aortic root ( $2.40 \pm 0.55$  vs  $1.87 \pm 0.49$ ,  $P = 0.027$ ). This is concordant to our previous report on patients undergoing reoperations after surgery for ATAAD (Chen et al. J Cardiothor Surg 2024;19:80-89).

Changes in the text 6: Please see added text in red color, paragraph Aortic reoperations and survival, added new reference #17, and Introduction, 2<sup>nd</sup> paragraph, 1<sup>st</sup> sentence.

Comment 7: Line 153. “This study shows that decision-making regarding aortic root resection was consistent with the degree of ascending aortic wall degeneration investigated histologically after surgery”. I recommend that you propose an ROC curve for the association between surgical procedure and the degree of mucoid extracellular matrix accumulation.

Reply 7: We believe that the degree of a histological parameter is not a predictive variable for a surgical decision. The predictive value of the extent of mucoid extracellular matrix accumulation alone using a ROC curve did not bring forward the association between the decision-making for a surgical solution.

The predictive value of the extent of mucoid extracellular matrix accumulation alone to identify those patients that require aortic root surgery was not significant as assessed by ROC analysis (area under the curve [AUC] 0.556, Standard Error [SE] 0.066, 95% confidence interval [95% CI] 0.427-0.686,  $P = 0.395$ ). However, the predictive value of the extent of mucoid extracellular matrix accumulation together with clinically relevant variables, such as presence and location of intimal tear, aortic valve regurgitation, age, male sex, presence of bicuspid aortic valve and aortic diameter, to identify patients with aortic root replacement was significant as assessed by ROC analysis (AUC 0.783, SE 0.052, 95% CI 0.682-0.885,  $P < 0.001$ ).

Indeed, the association of the extent of mucoid extracellular matrix accumulation of the ascending aorta is significantly increased in those aortas that were surgically operated for both the ascending aorta and the aortic root as compared with a limited solution of aortic root-sparing surgery. According to the adjusted multivariable binary logistic regression analysis, the extent of mucoid extracellular matrix accumulation (odds ratio [OR]: 9.981, 95% confidence interval [CI]: 1.832–54.363,  $P = 0.008$ ) was a significant factor related to patients with aortic root replacement.

Changes in text 7: Please see the completely new paragraph Roc analysis and Discussion, paragraph #6, text in red.

Minor

Comment 8: Abstract, Background. The phrase “the extension of the aortic wall disease during ATAAD” is confusing. I suggest “extension to the aortic root of ATAAD” or “aortic root involvement”.

Reply 8: Done

Changes in the text 8: Please see corrected sentence, Abstract, line 18.

Comment 9: Abstract, Methods. Similarly, “aortic wall degeneration during ATAAD” that implies the wall degeneration is caused by ATAAD. Do you think that ATAAD causes aortic wall degeneration? I suggest “the degree of degeneration of the ascending aortic wall found in patients with ATAAD”.

Reply 9: Agree. The finding is only an association and not a cause for ATAAD. Please see Comment 13.

Changes in the text 9: We corrected the sentence as requested, line 21.

Comment 10: Abstract, Methods. What are the 42 degenerative, atherosclerotic, and inflammatory histological variables? I cannot find them in the Methods section of the main text.

Reply 10: We apologize for this misnomer. There were 11 different variables that describe medial and adventitial damage. In addition, atherosclerosis and inflammation were separately evaluated.

Changes in the text 10: Please see the corrected sentence, line 24.

Comment 11: Abstract, Results. “A little more than” is not scientific. Just “39% of the patients” is enough. In addition, one patient seemed to have undergone the David procedure, which does not involve aortic valve prosthesis. “Aortic root replacement together with the ascending aortic replacement was performed in 39% of the patients. (n=55)” is sufficient.

Reply 11: Thank you for the correction.

Changes in the text 11: We revised as instructed: “Aortic root replacement together with the ascending aortic replacement was performed in 39% of the patients. (n=55)”, line 29.

Comment 12: Abstract, Results. Please indicate the severity of aortic regurgitation included. Mild or more?

Reply 12: Aortic valve regurgitation included moderate and severe aortic valve regurgitation.

Changes in text 12: We added “moderate to severe aortic valve regurgitation”, line 31.

Comment 13: Abstract, Conclusions. “Histopathology of the ascending aorta during ATAAD reveals distinctive aortic wall degeneration in patients with aortic root involvement vs not despite patient characteristics”. Patient characteristics between groups were not reported in the Result section of the abstract. Please provide a brief description of the patient characteristics between groups if you include this sentence in the Conclusions section.

Reply 13: Thank you for the comment.



Changes in the text 13: We revised the sentence as follows: “Histopathology of the ascending aorta during ATAAD reveals distinctive aortic wall degeneration in patients with aortic root involvement vs not.”, line 38.

Comment 14: Line 37. I disagree with the authors’ thoughts “the extent of diseased tissue involvement during ATAAD is believed to depend on tissue degeneration”. It also depends on a variety of factors, such as hemodynamics and location of the entry. I suggest “the extent of ATAAD is believed to depend in part on tissue degeneration of the aortic wall”.

Reply 14: We completely agree.

Changes in the text 14: Please see corrected sentence, line 55.

Comment 15: Line 41. Please cite at least one previously reported reference.

Reply 15: Done.

Changes in the text 15: We added the following new reference #7 (Chen T et al, J Cardiothor Surg 2024;19:80-88), line 59

Comment 16: 9) Line 43. “Aortic root replacement”, not “aortic replacement”?

Reply 16: Agree, aortic root replacement.

Changes in the text 16: We added the word “root”, line 62.

Comment 17: 10) Line 74. What does “intact” mean? The aorta should be dissected.

Reply 17: Agree and corrected.

Changes in the text 17: We added the following correction: “A minimum of six pieces of resected ascending aorta including all three aortic wall layers, i.e., the intima, media and the adventitia, were embedded...”, line 103.

Comment 18: Line 92. Spell out SD here, because this is the first time the word appears.

Reply 18: Done

Changes in the text 18: We added the complete word “standard deviation”, line 140.

Comment 19: 12) Line 116. Where do you evaluate the aortic diameter? Maximum in the ascending aorta? Please specify. Again, please indicate the severity of aortic valve regurgitation included here.

Reply 19: The diameter of the aorta was not always reported for the patients in the acute situation of ATAAD. We have now retrospectively calculated the largest measured diameter of the aorta

during ATAAD from CT images. The largest diameter of the ascending aorta was measured retrospectively from CT images. The diameter is 49 (SD 9.8) in patients with aortic root-sparing and 51 (8.1) in patients with aortic root replacement ( $P = 0.168$ , missing cases = 3). The severity of aortic valve regurgitation includes moderate to severe regurgitation graded as 2-3 out of 3, please see Reply 3, Reviewer B.

Changes in the text 19: Please see added sentence in Ethical Statement and Study Design, line 84. The following sentence is also added: “The severity of aortic valve regurgitation includes moderate to severe regurgitation graded as 2-3 out of 3”, line 85 and lines 181-184.

Comment 20: Line 170. Please provide references to support this rationale.

Reply 20: Done

Changes in the text 20: We added the following references: Osada et al, *Interact Cardiovasc Thorac Surg* 2018;27:277-83, reference #15; Leone et al, *J Thorac Cardiovasc Surg* 2018;156:1776-85 (reference #16); Buja et al. *The Texas Heart Institute Journal* • 2024, Vol. 51, No. 1, reference #17. Please see line 274.

Comment 21: Line 180. Unfortunately, I do not understand the meaning of these sentences.

Reply 21: Thank you for the remark.

Changes in the text 21: We deleted the unclear sentences. We added the following “Marfan syndrome may not have been diagnosed in some patients prior to ATAAD. We also speculate that many patients with Marfan syndrome may have had aortic surgery before developing ATAAD.”, line 287-289.

Comment 22: Table 1. When p value = 1, please consider mentioning  $> 0.99$ . Many variables in each group lack percentage. Prior or previous CABG/AVR is common, instead of earlier CABG/AVR. Similarly, CAD (coronary artery disease) instead of MCC.

Reply 22: Done.

Changes in the text 22: Please see corrected Table 1.

Comment 23: Table 2. Please provide information of dissection (distal extension, entry site, etc.) and reconstruction of the distal aorta. What does “prosthesis” mean?

Reply 23: We have included the requested information on the manuscript.

Changes in the text 23: Please see corrected Table 2. Instead of “prosthesis”, we now say “supracoronary aortic replacement”.

Comment 24: Table 3. There are a lot of variables needing explanation in the Methods section.

Reply 24: We explain the variables in the Methods section now. Please see Reply 4, Reviewer C.

Changes in the text 24: Please see Methods section.

Comment 25: Figure 2. Please indicate the number of patients at risk.

Reply 25: Done.

Changes in the text 25: Please see corrected Figure 2.

## **Reviewer D**

In this submission, the authors aimed at investigating whether ascending aortic wall degeneration during ATAAD is associated with the extension of aortic root disease. In this study, 141 consecutive patients were evaluated. Patients undergoing aortic root replacement were compared with those with supracoronary reconstruction of the ascending aorta during a mean 4.9-year follow-up. One third of patients received an aortic valve prosthesis together with replacement of the aortic root and ascending aorta (n=55, 39.0%). Mean age was 65 years. Eighty-five percent of patients with aortic root replacement had aortic valve regurgitation. The degree of mucoid extracellular matrix accumulation was more prominent in patients with aortic root replacement compared to patients with root-sparing surgery. During follow-up, there were 52 deaths. Authors conclude by saying that histopathology of the ascending aorta during ATAAD reveals distinctive aortic wall degeneration in patients with aortic root involvement. Surgical assessment of the extension of the disease during ATAAD justifies a limited resection strategy in many patients.

Some comments.

Comment 1: The submission needs some language edition.

Reply 1: We have now re-revised the manuscript extensively also for language.

Changes in the text 1: please see the manuscript, text in red.

Comment 2: Do not start paragraphs/sentences with abbreviations or numbers.

Reply 2: Thank you for the comment.

Changes in the text 2: Please see line 82.

Comment 3: Abstract. Do not identify the Institution in the abstract. This is done in the title page.

Reply 3: done.

Changes in the text 3: Please see “XXX Hospital”, line 23.

Comment 4: One can read “...Most of the patients with aortic root-sparing surgery included a supracoronary tube prosthesis (89.5%), while nine patients also had aortic valve replacement...”.

This is confusing as root-sparing should ideally not include valve replacement. This may be controversial but the root is a unit including the sinuses of Valsalva, annulus, valve and aortic tissue until STJ. Authors may argue that this is what it is and what, generally speaking, the community accepts. And this is also mentioned in the results section. In the way it is phrased, this may sound somewhat confusing for a part of the readership. Elaborate here and in the results section for better understanding.

Reply 4: We fully agree the reviewer's concern.

Changes in the text 4: We added "with/without aortic valve surgery" in the Abstract, Statistical analysis, line 163. Please see also Table 2 and Introduction, lines 62, 73. paragraph Surgery, line 93. Results, line 173.

Comment 5: Methods. The first subheading of this section must be "Study design", where authors confirm for the readership the type of study they present. This is not done here. "Study protocol" is not "study design". The readership has an unclear idea of the type of study.

Reply 5: We have revised the manuscript accordingly.

Changes in the text 5: Please see added "Study Design", line 76.

Comment 6: Authors confirm "...The ascending aortic wall resection was obtained from 141 consecutive patients who underwent ATAAD surgery and processed for histology. All patients experienced the onset of symptoms leading to surgery for ATAAD in less than 2 weeks...". This is not methods, this is results as data (N=141) are included. Data are not methods. Data are results. Shift this accordingly to the results section.

Reply 6: Thank you for the specification.

Changes in the text 6: Please see the transferred sentences, lines 169-171.

Comment 7: Authors state "...Similarly, the aortic arch was resected completely or in a hemiarch fashion depending on the involvement of aortic wall disease...". Authors confirm what is known, that the extent of resection is organized around the entry point, proximal or distal. The "discretion of the surgeon" is fine but usually the discretion goes around where the entry point is located and how is the assessed damage, proximal or distal, intraoperatively.

Reply 7: We agree with the reviewer, thank you for the comment. On the other hand, we have previously reported that the entry tear alone may not always determine the distal site of anastomose during surgery for ATAAD (Uimonen et al, Annals Thor Surg).

Changes in the text: We deleted the word "Similarly" from the sentence, line 96.

Comment 8: Results. One can read "...The mean aortic diameter was 54 mm (SD 11), but was available in only 26 patients before ATAAD surgery...". This is also confusing. How did these patients have no estimates of aortic diameter before surgery? As per authors' description, all patients should have a CT scan performed before the operation as indicated and performed.

Therefore, if there is a CT scan, there should be estimates on aortic size, meaning diameter, length of the aorta, etc. Elaborate.

Reply 8: The diameter of the aorta was not always reported for the patients in the acute situation of ATAAD. We have now retrospectively calculated the largest measured diameter of the aorta during ATAAD from CT images. The largest diameter of the ascending aorta was measured retrospectively from CT images. The diameter is 49 (SD 9.8) in patients with aortic root-sparing and 51 (8.1) in patients with aortic root replacement ( $P = 0.168$ , missing cases = 3).

Changes in the text 1: Please see added text in Methods, 1st paragraph: “The largest diameter of the ascending aorta was measured retrospectively from CT images.” Table 1 and Results, 1st paragraph:” The diameter is 49 (SD 9.8) in patients with aortic root-sparing and 51 (8.1) in patients with aortic root replacement ( $P = 0.168$ , missing cases = 3).”

Comment 9: What does “...There were two patients with early hemostasis...” mean?

Reply 9: These included re-sternotomies due to immediate postoperative bleeding, unrelated to resection of the aorta *per se*.

Changes in the text 9: We deleted the unclear sentence, please see line 202.

Comment 10: “one fasciotomy of the distal extremities...” is not aortic surgical technique. What does it mean here in this context? Were fasciotomies performed in the same emergency operation? Or were these fasciotomies performed in the postoperative period. The statement is rather confusing. Elaborate.

Reply 10: fasciotomies were done immediately after surgery for ATAAD.

Changes in the text: please see above, line 202.

Comment 11: Which were the 4 “reconstructions of the aortic arch” performed during the follow-up?

Reply 11: There were only 4 reoperations for the aortic arch that initially had root-sparing surgery.

Changes in the text 11: We added the following wording “...that initially had root-sparing aortic surgery.”, line 235.

Comment 12: How were the follow-up reoperations on the aorta allocated according to the original operation? In fact, authors clearly state “...Aortic reoperations included six operations of the aortic root, of which three patients also had reconstruction of the distal aorta. 15 patients underwent reoperation of the distal aorta encompassing six endografts of the descending aorta, five frozen elephant trunk prostheses, and four reconstructions of the aortic arch...”. This is fine; however, there is no clue to understand in which type of patients were these reoperations

performed, namely if the valve-sparing had more reoperations than the root replacements. Elaborate.

Reply 12: There were five patients with aortic root replacement (9.1%) and 16 patients with aortic root-sparing surgery that were reoperated (18.6%). Five patients with root-sparing surgery required reoperation for proximal root replacement and one patient with a biological root replacement required a reoperation for a second root replacement due to root pseudoaneurysm. Otherwise, 15 patients underwent reoperation of the distal aorta, as mentioned above.

Changes in the text 12: Please see lines 225-230.

Comment 13: Which were the causes of death at 30-days and during the follow-up? This is not mentioned and it is relevant information.

Reply 13: Unfortunately, we have no data on postmortem studies for the time being.

Changes in the text 13: We added this important information in Limitations, line 315.

Comment 14: Were there postmortem studies? If yes, authors should confirm the autopsy findings. All this is relevant information to understand why these patients died. It is important to know as the perioperative mortality was 22% (31/141) and the follow-up mortality was 58% (52/110). Were the deaths related to the aorta or to other causes?

Reply 14: Please see above. Again, the reviewer's point is valid. Unfortunately, we have no data on postmortem studies for the time being.

Changes in the text 14: We have now stated the following in paragraph Limitations "Postmortem investigation data is not available.", line 315.

Comment 15: Why patients with aortic tissue disorder (Marfan, etc.) did undergo root-sparing surgery? It is known for ages that these patients have much higher risk of developing aortic problems in the non-operated segments. Any specific reason?

Reply 15: Pragmatically, we have often experienced that Marfan syndrome was not preoperatively diagnosed in the patients. We speculate that patients with known Marfan syndrome have preventive aorta surgery before developing ATAAD.

Changes in the text 15: The following sentence is added in Discussion "Marfan syndrome may not have been diagnosed in some patients prior to ATAAD. We also speculate that many patients with Marfan syndrome may had had aorta surgery before developing ATAAD.", lines 289-293.

Comment 16: Discussion. In the limitations, authors should add that the aortic diameter (as stated by the authors) was not known before the operation.

Reply 16: This is an important comment as all patients did undergo emergency CT prior to surgery. In practice, at emergency surgery for ATAAD, only 54 patients (10.6%) had their aortic

diameter reported prior to surgery. We have now retrospectively calculated the largest measured diameter of the aorta during ATAAD from CT images. The largest diameter of the ascending aorta was measured retrospectively from CT images. The diameter is 49 (SD 9.8) in patients with aortic root-sparing and 51 (8.1) in patients with aortic root replacement ( $P = 0.168$ , missing cases = 3).

Changes in the text 16: Please see added text in Methods, 1st paragraph: “The largest diameter of the ascending aorta was measured retrospectively from CT images.” Table 1 and Results, 1st paragraph:” The diameter is 49 (SD 9.8) in patients with aortic root-sparing and 51 (8.1) in patients with aortic root replacement ( $P = 0.168$ , missing cases = 3).”

Comment 17: Tables. In table 1 replace MCC by a more frequently used English term.

Reply 17: Done.

Changes in the text 17: Please see corrected Table 1. Instead of “MCC” we now say “CAD” for coronary artery disease.

## **Reviewer E**

Comment 1: Please provide de location and technique of root diameter measurement. it makes little sense that the diameter of the root of the bentall group is 50 and the diameter of the root sparing 55.

Even if this value is not significant this is irritating since one would expect a greater root diameter in the bentall group (were the measurement performed in a multiplanar reconstruction?)

Reply 1: In this study, we have not measured the root separately, unfortunately. We have now retrospectively calculated the largest measured diameter of the aorta during ATAAD from CT images. The largest diameter of the ascending aorta was measured retrospectively from CT images. The diameter is 49 (SD 9.8) in patients with aortic root-sparing and 51 (8.1) in patients with aortic root replacement ( $P = 0.168$ , missing cases = 3).

Changes in the text 1: Please see added text in Methods, 1st paragraph: “The largest diameter of the ascending aorta was measured retrospectively from CT images.” Table 1 and Results, 1st paragraph:” The diameter is 49 (SD 9.8) in patients with aortic root-sparing and 51 (8.1) in patients with aortic root replacement ( $P = 0.168$ , missing cases = 3).”

Comment 2. line 147 The decision to replace the aortic root is also based on its diameter! Please discuss this as well. Do you see an increase in the histological degeneration that is proportional to the diameter?

Reply 2: Unfortunately, in this study, we have not measured the root separately. This is an important comment, and we wish to take this into account in our subsequent study and measure categorically different parts of the aorta separately. However, according to the adjusted multivariable binary logistic regression analysis, the extent of mucoid extracellular matrix

accumulation (odds ratio [OR]: 9,981, 95% confidence interval [CI]: 1.832–54.363,  $P = 0.008$ ) was a significant factor related to patients with aortic root replacement, even when confounders such as the extent of mucoid extracellular matrix accumulation, presence and location of intimal tear, aortic valve regurgitation, age, male sex, presence of bicuspid aortic valve and aortic diameter were taken into account.

Changes in the text 2: Please see Table 1 and 2 and Results, 4th paragraph, text in red color, Statistical analysis, and Discussion, paragraph 6, lines 294-300.

Comment 3: You need to discuss the fact that few surgeon will tend to replace the root in younger patient even if the usual "indications" are not met. So you may underestimate the degree of degeneration in the Bentall group since "maybe" not all of these patients had a strong indication to perform a bentall procedure.

Reply 3: Indeed, we are aware that there is a possibility to underestimate or overestimate the degree of aorta degeneration in any patients during surgery for ATAAD. The main task is to save the patient, and the extent of surgery is decided upon surgery; histology for the time being, is only available after the procedure. However, we are fascinated to learn that decision-making at ATAAD surgery for encompassing aorta root replacement seems, nevertheless, associated with aortic wall degeneration. Please see above; age has now been taken into account in the adjusted multivariable binary logistic regression analysis.

Changes in the text 3: Please see above for Reply 2 and Changes in the text 2.

## **Reviewer F**

Comment 1: I would like to congratulate the authors on their manuscript. Although I lack to see the translational value of this knowledge for emergency surgery, it is useful to be aware of. Extent of corrective surgery remains at the discretion of the surgeon at the time. It may be useful to incorporate the histology findings in a more intense follow up of 'high-risk' feature patients (for the discussion).

Reply 1: We thank the reviewer for the kind comment. Understanding the features of the dissected aorta leads to further information facilitating follow-up of the patients; we have recently studied patients with ATAAD and reoperations during follow-up (Chen et al, J Cardiothor Surg 2024;19:80-88). Aortic wall medial degeneration, specifically increased mucoid extracellular matrix accumulation and elastic fiber degeneration, may add to the susceptibility of aortic tissue frailty that require additional attention after surgery for ATAAD.

Changes in the text 1: Please see Conclusions, text in red.