Reviewer: Janet Powell
1. General comment: The principal merits of publishing this large body of work is the to prevent others from undertaking similarly futile studies. The results should be presented, but the conclusions sharply moderated.
RESPONSE: We have modified the title, abstract, results, discussion and conclusions to reflect this request.

2. Provide power calculations for numbers of patients and controls needed to detect a 10% (or 7.5%) change in allele frequency for HLA-DQA1*01102 and consider the results in the light of this power calculation.
RESPONSE: We have included results on power calculations in the second last paragraph of the results section and made a comment about the results related to this.

3. This reviewer concludes that there is little evidence to support association between the HLA loci studied and AAA, but in Belgian males there is a weak evidence that the HLA-DQA1 locus harbors a risk factor for AAA-associated conditions (and this may be hypertension, coronary heart disease etc). Why this should be so in Belgium but not in Canada is far from clear.
RESPONSE: We have added sentences at several locations in the manuscript to clarify that the allele (DQA1*0102), giving rise to the association in the Belgian males was also present at increased frequency in all the other case samples (Belgian females, Canadian males and Canadian females). The conclusion that there is evidence for a potential association among Belgian males is therefore true. The conclusion that there is evidence for a lack of association in the other sample groups is incorrect. Rather, the data are inconclusive due to lack of power in these smaller sample groups. The reviewer is correct in that the potential association may be with confounders (AAA-associated conditions) although the frequency of these conditions would have to be high among the AAA cases given the relatively low power to detect association with the primary phenotype. For the most significant findings, we have also added the p-values corrected for multiple testing using the Sidak function.

4. The confounders for the study, such as presence of hypertension etc. deserve be the amplified. After all several lines are used for the validity of spouse controls, when this would lead to a control group biased for female gender. The possible inappropriateness of the control group could lead to false conclusions.
RESPONSE: Unfortunately, such parameters were not collected from the control population.

Reviewer: Christian Gatting
1. The patient cohort is not well defined. Is further information on the patient characteristics available, especially on further AAA risk factors? This should be presented in a table including a statistical analysis for each parameter in the patient cohorts. This information might be helpful for elucidating the reasons for the occurrence of the statistical association of HLA-DQA and AAA in Belgian males and not in Canadian patients. Have the authors looked at the potential occurrence of Marfan or Turner syndrome in the cohorts?
RESPONSE: We do not have sufficient data on the other clinical parameters in the AAA population to be able to prepare such a table. We have described in the Methods section how many of them were operated, how many had family history, how many were females and what was the ethnic distribution. The controls were mostly population controls and no additional information was collected from them.

We have clarified that the association was detected in the Belgian male sample due to the sample size and consequent greater power to detect it. We have also clarified that the DQA1*0102 allele was also present at higher frequency in the other case groups, suggesting that all case groups had increased risk due to the same locus. The other sample groups did not reach the threshold for statistical significance due to smaller sample size and lower power.

We carefully excluded from the study all patients who had the Marfan syndrome. Although it is possible that Turner syndrome patients may have AAA, the literature indicates an increased prevalence of aortic root dilatation (8-42%) and dissection (see Elsheikh et al. Endocine Reviews 23:120-140, 2002). The prevalence of Turner syndrome is approximately 1:2,500 live female births. Our sample contained relatively few females; even with a substantial enrichment due to selection of AAA cases (assuming that Turner syndrome patients have an increased prevalence of AAA) the number of Turner cases in our sample will remain very small.

2. A calculation of the relative risk/odds ratio for the risk allele should be performed.
   Furthermore, the power of the study cohort has to be calculated and discussed.
   RESPONSE: We have added OR and 95% CI in the second last paragraph of the results. We have also provided the results of the power calculations and commented on the findings related to these in the second last paragraph of the results.

3. A description of the accuracy of the genotyping methods and the whole-genome amplification should be added or the corresponding references may be cited if the methods were performed without any modifications.
   RESPONSE: We have added a comment about this in the methods section. Quality control included genotyping samples whose alleles are known. We have validated the PEP protocol extensively and published the results in a previous publication which was cited in the original manuscript (Kuivaniemi et al. 2002).

4. The authors mention in the study limitations (p.13) that they have not adjusted for multiple testing. However, the adjustment for multiple testing is necessary in order to avoid false-positive associations which only occur by chance due to the multiple testing of the same cohort. These analyses should be added.
   RESPONSE: We have added a comment about the multiple testing in the second last paragraph of the results section. For the most significant findings, we have also added the p-values corrected for multiple testing using the Sidak function.

5. P.6, 2nd paragraph, lines 3 ff.: adjust font size
   RESPONSE: We thank the reviewer for pointing out this problem and have corrected it.

6. p.11, 2nd paragraph, line 10: delete blank (TIMP1)
   RESPONSE: We thank the reviewer for pointing out this problem and have corrected it.
7. p.14, line 3, insert blank humans [43]

**RESPONSE:** We thank the reviewer for pointing out this problem and have corrected it.

**Reviewer: Matthew Bown**

1. There is no attempt at a sample size calculations in the study design. In the final table the authors give a list of the previous studies that have investigated HLA loci in relation to AAA. The figures from these studies could have been used to determine a rough sample size despite being based on differing populations. A larger study such as this should have been aimed at ‘cleaning up’ the previous literature and powered as such, taking into account the necessity of multiple testing.

**RESPONSE:** We have provided the results of the power calculations and commented on the findings related to these in the second last paragraph of the results. We also wish to point out that all previous studies were so small that using them as guides as to what sample size to use would not be a good approach. In addition, the statistical methods used in these other studies were different from ours. Also, the HLA-DQA locus has not been studied previously for AAA.

2. Given the large number of hypothesis tests, although it is acknowledged no corrections made for multiple hypothesis testing and this should be done.

**RESPONSE:** We have added a comment about the multiple testing in the second last paragraph of the results section. For the most significant findings, we have also added the p-values corrected for multiple testing using the Sidak function.

3. The number of spousal controls in each of the study populations is not given (Canadian vs. Belgian).

**RESPONSE:** We have added this information in the methods section.

4. No demographics or phenotypic details about the study populations is given and would be useful (smoking history, AAA sizes etc.).

**RESPONSE:** The reviewer raises an important point. Unfortunately these types of data were not collected consistently and are therefore not available.

5. Whether the controls were screened for AAA should be stated.

**RESPONSE:** Some of the spousal controls did have an abdominal ultrasonography examination. The population controls did not have an ultrasonography examination. Based on population prevalence for this age group, it is expected that about 5% of the controls could have an AAA, but since this would not lead to false positives, but rather false negative results, we do not consider it a major problem in the design.

6. The authors have split their analyses by the two study populations on the basis that the total sample deviated from Hardy-Weinberg equilibrium. Whilst this appears to be valid as a separate analysis why have the overall study group results not been reported. Are the authors suggesting that the aetiology of AAA is different in Belgian and Canadian Caucasians? If so this should be covered in the discussion in detail. Does this suggest that
the marker found in the Belgian group is of relatively little significance in the pathogenesis of AAA since it is not consistent across different Caucasian populations?

**RESPONSE:** The distributions of the allele frequencies between the Belgian and Canadian control groups were compared and significant differences between the two control groups for all of the HLA loci were found. The p–values were: 0.062 (empirical: 0.056) for DQA; 0 (empirical: 0.0001) for DQB, 0.008 (empirical: .0046) for DRB, and 0 (empirical: 0.0001) for DRB3-5.

We have added statements in several locations to indicate that the allele (DQA1*0102) giving rise to the association in the Belgian males was also present at increased frequency in all the other case samples (Belgian females, Canadian males and Canadian females). The conclusion that there is evidence for a potential association among Belgian males is therefore true. The conclusion that there is evidence for a lack of association in the other sample groups is incorrect. Rather, the data are inconclusive due to lack of power in these smaller sample groups.

7. There are 15 authors – is each’s inclusion justified?

**RESPONSE:** This is a multicenter study where a large number of clinical and basic scientists were involved including vascular surgeons, study nurses, statistical geneticists, experts on HLA genotyping methods and molecular biologists. As requested by BMC Medical Genetics the contribution of each author is detailed at the end of the manuscript. Each author had a significant contribution to the study.