Author's response to reviews

Title: Variation in the CXCR1 gene (IL8RA) is not associated with susceptibility to chronic periodontitis

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Version: 2 Date: 29 September 2011

Author's response to reviews: see over
To the JNRBM Editorial Team

We greatly appreciate the Reviewer’s comments that have enabled us to develop a revised and improved manuscript. We have responded to all of these comments and we have provided point-by-point responses to the reviewer’s critiques in the end of this letter. Corrections made in the revised manuscript were highlighted using colored text.

We really appreciate your time and consideration.

Sincerely yours,
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Reviewer's report
Title: Variation in the CXCR1 gene (IL8RA) is not associated with susceptibility to chronic periodontitis
Version: 1 Date: 21 July 2011
Reviewer number: 1
Reviewer's report:

Major Compulsory Revision:
1. The authors show a lack of association between rs2234671 SNP and chronic periodontitis. However, the authors did not show if the sample used in this study present the statistical power to detect the association. Therefore, the authors should include in this paper a statistical power analysis in order to strength the conclusion of lack of association between rs2234671 SNP and chronic periodontitis. This analysis can be perform using GPOWER software, as decribed in “Faul, F., Erdfelder, E., Lang, A.G., and Buchner, A. (2007). G*Power 3: a flexible statistical power analysis program for the social, behavioral, and biomedical sciences. Behav Res Methods 39, 175-191.”

We thank the Reviewer for the suggestion of the software. Utilizing this software, we observed that our sample presented 93% of power to detect association. Therefore, we included this information in the Materials and Methods: “To verify statistical power of our sample, we used the G*POWER 3 software software (Faul, F., Erdfelder, E., Lang, A.G., and Buchner, A. (2007). G*Power 3: a flexible statistical power analysis program for the social, behavioral, and biomedical sciences. Behav Res Methods 39, 175-191)”

The odds ratio value used was the highest OR value obtained in the previous calculation. In this revised version of the manuscript we included in the analysis four other individuals in the control group. Therefore, the sample size increased to 395 individuals. Considering the analysis in the G*Power 3 software, in the Results section, we included: “The power calculations showed that the sample size of 395 individuals demonstrated a power of 93%. Therefore, the number of subjects enrolled in this study is large enough to detect association with an acceptable level of confidence”
Minor Essencial Revision:
1. In third paragraph within Findings section the authors preferred to use +860G>C notation, used in SNP500Cancer database. The authors should use the correct notation 860G>C (without the plus signal), according the following reference: “den Dunnen, J.T., and Antonarakis, S.E. (2000). Mutation nomenclature extensions and suggestions to describe complex mutations: a discussion. Hum Mutat 15, 7-12.”. The author also could use the notation based on SNP database (rs2234671) instead 860G>C notation, which avoids the problem regarding to the different positions for this SNP, pointed in this paragraph. In addition, the author should use only one notation, instead of all notations, as showed in last paragraph in Finding section: “…Ex2+860G>C (S276T) (rs2234671) SNP…”

The Reviewer’s corrections were made. The third paragraph was rewritten: “The 860G>C (S276T) SNP in CXCR1 gene was identified by comparison of multiple sequences deposited in the GenBank/EMBL data banks[11, 13]. These authors named this polymorphism differently: +2607 (position 6334 of sequence accession number L19592) in exon 2, and +827 (starting from the initiation of the ATG codon in exon 2 of L19592), respectively. The variant position 860G>C is based on the NM00634 exon 2 initiations, however, it is important to be clear that all these different positions at the CXCR1 gene refer to the same polymorphism (G>C), which results in a conservative amino acid substitution from serine to threonine at the 276 amino acid residue of the CXCR-1 (or IL8R-α) protein. Here, we preferred to use the reference sequence number (refSNP ID: rs2234671) in NCBI's Entrez system (www.ncbi.nlm.nih.gov/SNP).”

Level of interest: An article of importance in its field
Quality of written English: Acceptable
Statistical review: No, the manuscript does not need to be seen by a statistician.

Reviewer's report
Title: Variation in the CXCR1 gene (IL8RA) is not associated with susceptibility to chronic periodontitis
Version: 1 Date: 2 August 2011
Reviewer number: 2
Reviewer's report:
I have the following comments about the manuscript:
Major Compulsory Revisions
1. The authors describe chi-square was used to assess differences between cases and controls. Why not a multivariable analysis taking into consideration gender and smoking status Table 1 clearly show cases had more smokers than controls.

As suggested by the Reviewer we executed a multivariable analysis, as demonstrated in the revised manuscript.
2. A power calculation was not included to help the interpretation of the results. Is the study underpowered?

Since the Reviewer 1 also requested the power calculation and suggested specific software, after we utilized it, we observed that our sample showed 93% of power to detect association. Details are presented in the revised manuscript.

**Discretionary Revisions**

1. Brazilians are a tri-hybrid of White Europeans mostly of Portuguese origin, Africans, and Amerindians. How this variable was accounted for in the analysis? Data on this should have been presented and maybe included in the analysis.

As a matter of fact, we have the data of the skin color of the enrolled patients. However, in a previous study of our group (Anovazzi et al, 2010), a stressed recommendation was made by a Reviewer to exclude this stratification of our sample in order to avoid misleading interpretations with regards to “race”. Because of this, in the initial version of the manuscript, we opted for not to stratify our sample, since we only obtained skin color information of our sample, and skin color are not related with ancestry in the Brazilian population, according Pena et al.[39], Parra et al.[40] and Pimenta et al.[41]. It could be interesting to perform a genomic control analysis in further studies with our sample.

**Level of interest:** An article of limited interest  
**Quality of written English:** Acceptable  
**Statistical review:** No, the manuscript does not need to be seen by a statistician.

**Reviewer's report**  
**Title:** Variation in the CXCR1 gene (IL8RA) is not associated with susceptibility to chronic periodontitis  
**Version:** 1 Date: 6 August 2011  
**Reviewer number:** 3  
**Reviewer's report:**  
**Minor Essential Revisions**

The authors should clarify whether the lack of differences are really due to lack of genotype effects or to an interaction with the smoking status. This is because the % of smokers is statiscally significant when the patients and controls are compared (22% and 9%, respectively; P<0.05). Therefore, because smoking is a risk factor for periodontitis, it is possible that the disease is due to smoking or to an interaction with the smoking status. A multivariate analysis could let us know the final answer.

The Reviewer is correct. As suggested, we executed a multivariate analysis, as observed in the revised manuscript.

**Level of interest:** An article of importance in its field  
**Quality of written English:** Acceptable
Statistical review: No, the manuscript does not need to be seen by a statistician.

Reference:
Anovazzi G, Kim YJ, Viana AC, Curtis KM, Orrico SR, Cirelli JA, Scarel-Caminaga RM: