Reviewer's report

Title: Allelic variants of the transient receptor potential channel TRPV6 and risk of prostate cancer

Version: 2 Date: 28 August 2009

Reviewer: Halima Ouadid-Ahidouch

Reviewer's report:

The authors have previously demonstrated that TRPV6 is correlated with the aggressiveness of prostate cancer and more recently they isolated two allelic TRPV6 variants (TRPV6a, TRPV6b) from human placenta. In the current study, the authors studied the TRPV6a and TRPV6b allele frequencies in prostate cancer (PC) in Caucasians and correlated it with clinical parameters.

Although, the work done is important, there are some major issues that have to be dealt with.

Major points:

1. Why did you analyze the second polymorphic? please explain
2. As a control, you have used the salivary fluid from the healthy persons, why don’t you analyze the salivary of patients with PC instead of tumor tissues, especially that you are using a relatively large amount of cancerous tissue?
3. Are the patients only Germans or originating from other European countries? (see table, this study Europe)
4. The question you ask in this study concerns not only TRPV6a, but the potential correlation between TRPV6 allelic variants and prostate cancer (development and aggressiveness)? Rephrase your question
5. In the last paragraph of the results, you investigated the frequency of the expression of TRPV6 in advanced PC and you conclude that TRPV6 is expressed in all tumor samples. Unfortunately, you cannot make such conclusion since the number of PC samples used is very insufficient (4 pT3b and 1 pT2). If you want to keep this paragraph you should increase the number of samples and add the correspondent figure. Otherwise, you can remove this paragraph, since there is no link between the expression of TRPV6 and the aim of the article (TRPV6 allelic variants). Moreover, you had already published the expression of TRPV6 in PC.
6. The last paragraph of the discussion is confused. 1) How did you show that the 16 polymorphisms located within introns have the same frequencies as the 5 polymorphisms described in the study? 2) Figure 3 does neither show the polymorphisms of the adjacent genes nor their frequencies. Please clarify.

Minor points
1. Methods (paragraph: isolation of genomic DNA and RNA): line 5, 1 ml (1ml), line 6, 180 µl (180µl); 20 µl (20µl), line 7, 200 µl (200µl), line 8, 200 µl (200µl), line 16, 50 µl (50µl).

2. Paragraph Statistics? Lacking!

3. Results, p2, line 3, (sample 88, supp table 1 instead of table 1).

4. Fig .2 ABCDE: put them in order of their appearance in the text.

5. Change Fig. 2a (Fig.2 A), Fig. 2b (Fig. 2B)

6. In the sentence “Only from 99 of the 169 tissues samples …” change 169 by 142 tissues samples. In the legends of fig 2 D you mentioned 98 tissues samples vs. 99 in the text!

7. Change Fig.2 by Fig.2 D at the end of “However, the allele frequencies between these groups are similar (Fig. 2)”.

8. Added Fig. 2E at the end of the paragraph “on the Gleason score and TRPV6 allele frequency”.

9. Added Fig. 2C after you result on the incidence of tumor stage and TRPV6 allele frequency.

10. Discussion: line 6: change 169 by 142

11. Line 22, polymorphisms instead of polymorphism

12. Line 28, significantly instead of significant

13. Change 141 by 142 prostate cancer sample (line 30)

14. Conclusion: last paragraph: modulate your question about TRPV6a allele because of the low frequency of this allele.

15. Discussion: you make an association between the occurrences of genotype b/b and the distance from Africa. How can you explain that the Afro-Americans express the TRPV6b allele? It seems to be better ethnic than geographic.

16. Figures: Fig.2; Change B/B (b/b), A/B (a/b).

17. Legends to figures: Fig.2 replaces commas par points. Fig.2 (B) what does (n=230) means? Number of tissue samples? (169 + 141 PC = 310)

18. Table 1: you have 311 samples (169 + 142 PC).

**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.

**Declaration of competing interests:**

'I declare that I have no competing interests'