Author's response to reviews

Title: Matrix Metalloproteinase-9 (MMP-9) polymorphisms in patients with cutaneous malignant melanoma

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Author's response to reviews: see over
Deborah Saltman, M.D., Ph.D.
Editorial Director of BMC Medical Genetics
BioMed Central Ltd.

Re: MS# 1902446735118839
Title: Matrix Metalloproteinase-9 (MMP-9) polymorphisms in patients with cutaneous malignant melanoma

January 23rd 2007

Dear Dr. Saltman:

We are resubmitting our manuscript (attached), after making changes in response to the comments made by the referees. Specifically, changes have been made and can be found in pages 3, 4, 5, 7, 9, 11, 12, 13, 14, 17. In addition we have added names under the acknowledgments in page 19.

Reviewer 1.

1. The reviewer suggested adding a table including the overall SNP frequencies for each of the variants. In order to provide the reader with an easier access, we have taken former Tables 2a and 2b from the Supplemental data and inserted the ‘New’ Tables 2 & 3 (“Frequency of the different SNP genotypes and alleles in the population studied” and “Microsatellite (-)131 (CA)n allele frequencies”) in the main file.

2. The reviewer suggested the inclusion of haplotype analysis following the single polymorphisms analyses. We agree that this can provide additional information on the role of MMP-9 in melanoma. In response, we’ve conducted haplotype analysis and in this revised version of the manuscript we have included the corresponding statistical methods (page 9) and results (page 13). The haplotype analysis did not show any significant association between haplotypes and the variables of interest, therefore no further discussion has been added under the Discussion section.

3. It has been suggested to combine data from the small tables in a single table. We have condensed the significant findings from former Tables 2, 3 and 4 into a “New Table 4” entitled “Statistically significant associations between the different SNPs and clinico-pathological variables”.

Reviewer 2.

1. The reviewer pointed out the omitted word “Matrix” when describing what the abbreviation MMP stands for. We have made this correction in page 4 and throughout the manuscript.

2. The reviewer noted that on page 4: “the indicated literature references 6 and 7 are not about MMP-9 activity, but instead about MMP-9 levels”. We agree with the reviewer with the fact that activity is an important measure. To clarify further the text, we have added “expression and MMP-9 activity” in page 4. We consider that the references 6 and 7 are appropriate to support this knowledge. Specifically, Zhao et al 2001 uses gelatin zymography for determination of collagenase activity (pages S53-S54). Similarly, Shellman et al (2006) describes the choice of the method to determine activity on page 208.

3. We are thankful for the reviewer comment “It is relevant to address the finding of immunohistopathological analysis in association with different types of growth modes in melanoma (van den Oord JJ, Paemen L, Opdenakker G, deWolf-Peeters C. Expression of gelatinase B and the
extracellular matrix metalloproteinase inducer EMMPRIN in benign and malignant pigment cell lesions of the skin. Am J Pathol. 1997 Sep;151(3):665-70”]. In response, we have added in the Introduction on page 4, the following sentence: “In primary melanomas, MMP-9 is variably expressed in radial but not in the vertical growth phase and the de novo expression seems associated with early invasion” followed by the suggested citation (van den Oord et al 1997).

4. As per suggestions of the reviewer, we have added the literature reference (Van den Steen et al 2002) and the description of the known polymorphisms in the Introduction Section (page 5, third paragraph). In addition, the referee has suggested that we replace gelatinase b with gelatinase B (capital letter) throughout the manuscript. We have instead changed gelatinase b to MMP-9 since MMP-9 and gelatinase B are synonyms and ‘gelatinase b’ appeared only once in our original text. This will make it easier to the reader.

5. As per the reviewer’s suggestion we have replaced the word “contradicting” with “divergent” in the Discussion section (page 14, second paragraph).

6. The reviewer noted that “PGBD-like and kringle-like are terminologies not applicable to MMPs”. We have made this correction in the Methods on page 7 and replaced the sentence “…3) the effect on conserved or specificity residues of four superfamilies of MMP-9 domains (catalytic, hemopexin-like, PGBD-like, kringle-like) by multiple sequence analysis“ with the following sentence: “the effect on conserved or specificity residues of four superfamilies of MMP-9 domains (MMP N-terminal domain, catalytic domain, fibronectin type II domain, hemopexin domain) by multiple sequence analysis.” We have also added a citation to support the methodology (Gough et al 2001). PGBD-like and kringle-like actually refer to superfamilies of protein domains which include the MMP N-terminal domain, catalytic domain, fibronectin type II module (as per the database ‘Superfamily-HMM library and genome assignments server’ (http://supfam.org/SUPERFAMILY/), however we still agree with the reviewer; MMP N-terminal domain, catalytic domain, fibronectin type II domain, and hemopexin domain –are more appropriate terms.

7. We have replaced Esteve with Estève in former reference 31 (reference 37 in the present revision). We would like to note that the last name for this author is listed as Esteve in PubMed.

Tables:
Please note that while the original version of the submitted manuscript contained: 4 tables in the Main File (1,2,3,4) and 5 Supplementary Tables (1,2a, 2b, 3, 4, 5), this revised version contains: 4 tables (Main File) and 4 Supplementary Tables.

Citations:
The total number of citations (39) in this revised version of the manuscript is due to the additional [haplotype] analysis performed, to a previously omitted citation in the methods section (Methods, Selection of SNPs, page 7), and to referee’s suggestions.

We are grateful to the referees’ comments and we hope that this current version of the manuscript meets the criteria for publication.

Respectfully,

Irene Orlow, Ph.D.