Author's response to reviews

Title: Isolation and Genomic Analysis of Circulating Tumor Cells from Castration Resistant Metastatic Prostate Cancer

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Author's response to reviews: see over
Editorial Board
BMC Cancer

Dear Members of the Editorial Board:

We are submitting our revised manuscript by M.J.M. Magbanua, E.V. Sosa, J.H. Scott, J. Simko, C. Collins, D. Pinkel, C. Ryan and J.W. Park entitled:

**Isolation and Copy Number Analysis of Circulating Tumor Cells from Castration Resistant Metastatic Prostate Cancer**

that we wish to have considered for publication as a Technical Advance in *BMC Cancer*. We include in this submission our point-by-point response to the reviewer’s concerns.

If you have any questions, please do not hesitate to contact me at any of the following: Tel.: (415) 502-3844; Fax: (415) 353-9592; email: jpark@cc.ucsf.edu

Thank you very much.

John W. Park, M.D.
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Reviewer’s report
Title: Isolation and Genomic Analysis of Circulating Tumor Cells from Castration Resistant Metastatic Prostate Cancer
Version: 2 Date: 9 January 2012
Reviewer: Thomas Höfner

Reviewer’s report:
After second major revision of the manuscript there are still some details missing that should be included to strengthen the scientific quality of the work.

Minor compulsory revisions:
- As mentioned before the discussion should include a sentence about limitations that arise when using the manual scraping technique of adjacent tissue for the subsequent CGH comparison tumor versus CTCs. The sentence should include the limitation that the manual scraping technique could have potentially led to a bias of including benign tissue in the molecular comparison. Thereafter the authors should include their own sentence about the laser microdissection.

- We have added the following to the text (p. 14):

Finally, we performed manual microdissection of archival samples, which can potentially include adjacent benign tissue.

- The fact that FACS sorted CTCs could not be reanalyzed for their integrative purity and EpCAM+/CD45- consistency because of limited numbers is a clear limitation of the study and also has to be mentioned in the discussion. This should include the fact, that this could have potentially led to confounding results from molecular analyses of cells that were initially false positively sorted as EpCAM+/CD45- cells.

- We have added the following to the text (p. 12):

Due to the paucity of FACS sorted CTCs, re-analysis by FACS to confirm purity and the correct expression of biomarkers was not performed; hence, the isolation of falsely positive “EpCAM+/CD45-” cells cannot be ruled out. However, the resulting aCGH profiles showed unequivocal evidence of cancer-associated genomic alterations, and did not appear to be significantly dampened by false positive normal cells.