Author's response to reviews

Title: Identification of a candidate prognostic gene signature by transcriptome analysis of matched pre- and post-treatment prostatic biopsies from patients with advanced prostate cancer

Authors:

Prabhakar Rajan (p.rajan@beatson.gla.ac.uk)
Jacqueline Stockley (jacqueline.stockley@glasgow.ac.uk)
Ian M Sudbery (ian.sudbery@dpag.ox.ac.uk)
Janis T Fleming (j.fleming@beatson.gla.ac.uk)
Ann Hedley (a.hedley@beatson.gla.ac.uk)
Gabriela Kalna (g.kalna@beatson.gla.ac.uk)
David Sims (david.sims@dpag.ox.ac.uk)
Chris P Ponting (chris.ponting@dpag.ox.ac.uk)
Andreas Heger (andreas.heger@dpag.ox.ac.uk)
Craig N Robson (c.n.robson@ncl.ac.uk)
Rhona M McMenemin (rhona.mcmenemin@nuth.nhs.uk)
Ian D Pedley (ian.pedley@nuth.nhs.uk)
Hing Y Leung (h.leung@beatson.gla.ac.uk)

Version: 4
Date: 28 November 2014

Author's response to reviews: see over
28 November 2014

Dr Dafne Solera
Executive Editor, BMC Cancer

Dear Dr Solera,

Re: Manuscript ID: 1069870281140402 - “Identification of a candidate prognostic gene signature by transcriptome analysis of matched pre- and post-treatment prostatic biopsies from patients with advanced prostate cancer”

Thank you for your e-mail regarding the above manuscript. We have specifically addressed your points and are pleased to re-submit our manuscript with the responses given below.

We look forward to hearing from you with the outcome of your editorial decision.

Yours sincerely,

Hing Y. Leung PhD FRCS (Urol.)
Professor of Urology and Surgical Oncology

Prabhakar Rajan PhD FRCS (Urol.)
CR-UK/RCSEng Clinician-Scientist Fellow

Professor Hing Y. Leung
Urology Research Group
Email: e.hall@beatson.gla.ac.uk

The Beatson Institute for Cancer Research, Garscube Estate, Switchback Road, Bearsden, G61 1BD, UK
Tel: +44(0)141 330 3658
Fax: +44(0)141 942 6521
www.beatson.gla.ac.uk
1. Reviewer 1 had asked why the number top-ranking genes selected for validation was 10 (and not 20 or any other number). That question remains unanswered. Even if the choice was based on an arbitrary convenience (manageable number) criterion, this must be stated, as it has an impact on the subsequent results.

Response: We have amended the manuscript text to clarify that the selection strategy was arbitrary as follows:

Page 10, Lines 233-5 Results: “The 10 top-ranking genes differentially-regulated by docetaxel plus ADT were arbitrarily selected (range of fold changes -9.96 to 9.86) for further downstream knowledge-based validation.”

2. With regard to Figure 1B, the authors claim that a log-log scale would artificially inflate the significance of the trend. Have they actually compared the p-values of Pearson’s correlation when these are computed from linear and log values? With N=4 (the number of values should also be included in the figure, together with r and p), one is tempted to think that is the outlier point (top right) that is artificially inflating correlation and improving significance. This needs to be clarified.

Response: We agree that the presence of the outlier makes the correlation appear stronger. We felt that by presenting on a linear scale, we could allow the reader to draw his or her own conclusions from these data. However, we now include an amended Figure 1B on a log-log scale (with r- and p-values). The correlation here is still very strong and is significant at a 5% level. We have included changes to the manuscript text as follows:

Page 10, Lines 196-8 Results “The levels of expression of KLK3, which encodes PSA (Prostate Specific Antigen), detected by RNA-Seq of the docetaxel plus ADT arm correlated as expected with serum PSA levels (r² = 0.927; p=0.037) (Figure 1B).”

Page 24, Lines 503-6 Figure Legends “(B) Log-log plot demonstrating correlation between KLK3 (encodes PSA) mRNA expression levels (X-axis) normalized by trimmed means of M-value (TMM) in normalized counts per million (ncpm) and serum PSA levels (ng/ml) (Y-axis) (r² = 0.927; p=0.037).”

Please also proofread labels and citations for figures and tables. For instance, in the Methods ? Bioinformatics section, Table S4 (instead of Table S1) is associated with read quality control (line 170).

Response: We have checked the labels and citations and believe they are now accurate.