Reviewer's report

Title: Importance of pre-analytical steps for transcriptome and RT-qPCR analyses in the context of the phase II randomised multicentre trial REMAGUS02 of neoadjuvant chemotherapy in breast cancer patients

Version: 1 Date: 29 October 2010

Reviewer: John Martens

Reviewer's report:

Quality control of gene expression analysis is essential, particularly if performed in multiple centers. This study tackles the problems that might arise. It is well worked out and nicely presented with sufficient detail. The authors conclude that more stringent criteria are need for integrating microRNA data compared to real time results.

Major Compulsory Revisions:
I think the authors need to include more details on how the gene expression datasets were merged. Were they united without normalisation of the separate datasets first. Furthermore, were any other algorithms used to better align the different datasets. There are several procedures to do this and it is not described whether or how this was done. Currently, the description of how that microarray datasets were merged is insufficient to judge the firm conclusion that more stringent criteria are need for integrating microarray data compared to real time results.

Minor Essential Revisions:
Inclusion of a REMARK-type of diagram would be helpful for the reader. What were exact inclusion criteria and what were the subsequent exclusion criteria (step by step) why from the initial included 340 biopsy-ed patients only 226 and 230 were in the end used in the microarray and real time studies that were presented.
What type of clustering was performed?
Have the authors tested for the robustness of clustering?
Table 3 and 4 are less essential and could be presented in the supplement.

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