Reviewer's report

Title: aB-crystallin is a marker of aggressive biologic behavior in patients with breast cancer but not a prognostic or predictive marker for taxane based therapy: A combined analysis of two randomized studies

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Reviewer: Gary Tse

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This is an interesting study on aB crystallin expression in a large cohort of breast cancers, and the authors found association of aB crystallin with other adverse biomarkers expression and specific molecular subtypes. Most of these results were previously reported in the literature, and if there were any new findings, there had not been highlighted in the manuscript. Also, the patient groups were selective for the clinical trials and the results may not be applicable to all breast cancers patients.

There are specific comments that need to be addressed, listed as follows:
1. In general, new findings are absent in the current study. Some important details were absent in the paper and make it difficult to analyze. Nevertheless this study still has the relative advantage of a large cohort of breast cancer.
2. What was the rationale of assessing aB crystalline and paclitaxel treatment? Was aB crystalline known to affect tumor response to chemotherapy agents? Also, though treatment information for the cohorts has been reported elsewhere, the current study is focus on specific treatment response and so detailed information on treatment and patient selection is important.
3. Why the tumor grade was not assessed from the TMA slides but rather copied from the medical charts?
4. Though previously described, brief description for TMA construction (area selection, number, size of cores and how it is scored: average of score of cores/highest score from the core?) should be included. This is relevant as the staining of aB crystalline was focal, so there needs to be adequate representation of the TMA.
5. Vimentin and CK8/18 were used as control for tissue immune-reactivity. Tissue negative for above two antibodies were excluded. Any supportive evidence for using them as control? In particular, vimentin expression rate in breast cancer is low.
6. What was the definition of high Ki67?
7. The definitions of BCP_I and BCP_II appear to overlap. What is the significant to distinguish BCP_I and _II? The number of TNBC cases should be equaled to BCP_II plus unclassified cases. However, it did not add up. Please clarify.
8. IHC evaluation - suggest to list out the positive criteria briefly in a table for
9. For assessment of αB crystalline, it was mentioned that the staining was evaluated for percentage and intensity. But in the final ‘grading’ of αB crystalline staining, only % was used to define weakly positive or strongly positive. Has the intensity been taken into account? Also, it is described to be assessed as negative, weakly positive and strongly positive. Then a definition of negative and positive staining of 30% cutoff was also mentioned. It is confusing. 15% staining was scored as weakly positive or negative? This requires clarification.

10. Was HER2 FISH done for all cases or only 2+ cases?

11. Only 127 cases were with DNA for BRCA1 mutation analysis. Is there any difference with these cases compared to the overall cohort?

12. αB crystalline was evaluated as an ordinal scale. Significance of chi-square test may only indicate a differential distribution. It would be better to perform also a correlative analysis.

13. In the results, were there any discrepancies in αB crystalline staining in the cores of the same case?

14. In results the authors stated ‘αB crystalline expression was significantly associated with ER, PgR numeric’ please state whether these associations were positive or negative.

15. In discussion, the authors noted significant differences in the overall percentage of αB crystalline expression in breast cancers in their current series and some other series in the literature (up to 88%). As the difference was very significant, could the authors postulate the underlying reasons (rather than saying very loosely ‘differences in methodology and heterogeneity of staining’ were there any detailed analysis as to the possible reason)?

16. The discussion was well written, but again please highlight the significance of the current result and any new findings.