Reviewer's report

Title: CA1 Contributes to Microcalcification and Tumourigenesis in Breast Cancer

Version: 4 Date: 10 June 2015

Reviewer: Rajini Rao

Reviewer's report:

The revised manuscript addresses some of the criticisms raised in the former review. The quality of the figures has improved, the introduction and discussion have been edited to include more relevant details. The efficacy of siRNA knockdown of CA1 in MCF7 cells has slightly improved, although the reduction in protein appears to be the same (about 40% knockdown in both original and revised figures).

Major Compulsory Revisions:

The deficiency in linking CA1 to microcalcification in 4T1 cells has not been addressed at all. The authors claim that they treated 4T1 cells with acetozolomide, a carbonic anhydrase inhibitor, and detected “significantly decreased calcification”. Where are these data? If they have the results, why do they not show them especially since the previous review specifically suggested the experiment? Why did they not knockdown CA1 with siRNA in 4T1 cells as suggested in my previous review, or address this issue? This should be technically feasible, yet the authors ignore this request.

I accept their explanation that they were unable to monitor calcification in MCF7 cells. The point is that calcification in 4T1 cells (Fig 4-5) has already been well characterized in the literature, and up regulation of bone matrix proteins in OC medium (Fig 6) has been previously reported by Professor Morgan’s group. While the new findings in this study that CA1 is induced in the calcification medium (Fig 7) are interesting, the functional link between carbonic anhydrase and microcalcification, if any, is missing. Therefore, Figs 4-7 are not clearly linked to the remainder of the paper, which characterizes expression of CA1 in breast cancer.

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.