Reviewer's report

Title: Multicentric and multifocal versus unifocal breast cancer: differences in the expression of E-cadherin suggest differences in tumor biology

Version: 1 Date: 8 March 2013

Reviewer: Ana Sofia Ribeiro

Reviewer's report:

The manuscript entitled “Multicentric and multifocal versus unifocal breast cancer: differences in the expression of E-cadherin suggest differences in tumor biology” by Weissenbacher T et al. evaluates the expression of MUC-1, β-catenin and E-cadherin in multicentric/multifocal breast cancer in comparison to unifocal disease. They aim to identify potential differences in the biology of these tumor types. The authors conclude that multicentric/multifocal and unifocal breast cancers with identical TNM-staging clearly differ in the expression level of E-cadherin.

Although the subject is interesting, and the question is clear, the paper is a bit confusing and the conclusions drawn from the results are a bit speculative. Therefore, a number of key issues should be addressed:

Major compulsory revisions:

1. The authors describe very well how they performed the matched pair analysis. Although the criteria is very clear (tumor size, grading and lymph node status) the authors should also match for HR status, as done by the authors in a previous work. Is there any specific reason for not having done that in this study as well? Also matched group should be analysed concerning

2. Hormone receptor status and for Her2 expression should also be evaluated in multicentric/multifocal and unifocal tumors, since they are key molecular markers, and should be presented in table 2.

3. This reviewer is concerned about the Immunohistochemistry classification of β-ctn. From the materials and methods part, seems that the authors only classified the overall expression of β-ctn. However, β-ctn localization is more important than its overall expression. The sentence in the discussion “Therefore, elevated β-ctn expression appears to be linked with worse outcome for the patients” is not true. Most of the data show that abnormal β-ctn expression (it does not mean that it is overexpressed) or mislocalization of the protein to the cytoplasm is important for tumor cell aggressiveness. Did the authors investigated that?

4. Muc1 results are confusing. The authors say that the expression pattern of MUC1 is related to tumor characteristics and clinical outcome, being its
intracellular expression associated with poor prognosis, whereas membrane expression correlated with better outcome. Like b-ctn, MUC1 localization should be discriminated. In this situation, do the authors still observe the same inverse correlation between MUC1 and grading? If indeed the mPANkoMab antibody recognizes a special tumor-associated MUC1 epitope, is this epitope correlated to better or worse prognosis?

5. In the end of the discussion, the authors state that: “In summary, we have confirmed and extended our earlier results which demonstrated that multicentric/multifocal tumors as compared to unifocal breast tumors correlate with a reduced survival and relapse-free interval”. This is an overstatement, since the authors do not show in this paper the results concerning survival and relapse of the patients as shown in their previous work. If possible, the authors could include those results, since it will strengthen their conclusions.

6. Graphs with IR-score for E-cadherin and grading should be shown in figure 1, since they are described in the results part.

Minor essential revisions:

1. Figure 2D and 2E – In the y axe of the graph is written IR-score for E-cadherin, however the p-values shown and the legends refer to MUC1. This should be corrected.

2. Table showing all the data in what concerns E-cadherin/bctn and MUC1 expression and patient characteristics. This is described in the results parts, however the results are not shown.

3. Some of the references are not correctly attributed. For example, in page 12 of the discussion part, the authors refer to reference 24 and 25, but this do not correspond to the described papers.

**Level of interest:** An article whose findings are important to those with closely related research interests

**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