Reviewer’s report

Title: Validation of HER2 testing with core needle biopsy specimens from primary breast cancers in terms of interobserver reproducibility and concordance with surgically resected specimens

Version: 1 Date: 4 June 2010

Reviewer: Sophia Apple

Reviewer’s report:

Minor Essential Revisions:
Authors studied 100 core needle biopsies and some surgical specimens from 3 different institutions for interobserver reproducibility and comparison on Her-2/neu by IHC and FISH tests. They found that CNB was more reliable with high k statistic, contrasts to their speculated belief stated in the Introduction on page 5.

Materials and Methods:
1. FISH Her 2 test using PathVysion is a FDA test and they require 4 um thickness (not 5 um)
2. Authors mentioned that they examined 100 paired samples for CNB and surgicals but the FISH test states 100 CNB and 27 surgicals. This is not “paired” for at least FISH test.
3. Reading under the IHC methods, it is not clear whether the authors did perform 100 CNB and 100 paired surgical IHC test for Her2.
4. For 27 surgical FISH test, did you do 2+ reflex tests? If not why? IHC 2+ Her2 is reflex to FISH test by CAP 2007 guidelines (pertaining to institution B in particular).
5. Page 5, when a tumor was judged as equivocal, was it 40 additional nuclei in another tumor area or the same area on invasion were studied?

Results:
1. Pages 10-11: one case had 3+ Her2 on CNB and 0 on surgical (case 54). Please provide reasons, not just Fig 2A.
2. Page 11: Define what you mean by “the borderline nature of the tumor”.
3. Did you encounter polysomy 17 on FISH test?
4. Page 11: In case 92, did you perform FISH on invasive carcinoma only? Please mention this fact whether FISH was included in DCIS or not. (was there 40 nuclei to study for invasion only?)

Abstract:
1. It reads as though it is not mandatory to study Her-2 on CNB. It is mandatory
to study Her-2 on breast cancers and either CNB or surgical specimen can be used. Please reword this section.

2. Surgicals: 80% k of 0.77 for 3x3 and 92% k of 0.83 for 2 x 2 (not 3x3)

3. CNB IHC was 76% K of 71 for 3x3 and 90% k of .80 for 2x2. So, the final conclusion in page 16 is inaccurate when the authors mentioned “CNB specimens are most reliable…”

Additional comments:

1. It would be helpful to include a specific % of positive cells used as 1+ 2+ and 3+ although you mentioned that the authors followed CAP/ASCO 2007 guidelines.

2. Include article(s) that a prolonged formalin fixation could lose FISH amplification and/or yield to unsuccessful test (in pertaining to institution A problem).

3. Include number of years of experiences of pathologists in all 3 institutions to achieve high (substantial) k value.

4. Ideally, FISH should have been done in all 100 paired cases with CNB.

Level of interest: An article of importance in its field

Quality of written English: Acceptable

Statistical review: Yes, but I do not feel adequately qualified to assess the statistics.

Declaration of competing interests:

I declare that I have no competing interests’ below.