Reviewer’s report

Title: Estrogen receptor (ER) signaling regulates the expression of the breast tumor kinase (BRK) in breast cancer cells

Version: 1 Date: 20 Aug 2018

Reviewer: Guy Leclercq

Reviewer's report:

Overall, objective outlined in the abstract of the manuscript are satisfied, taken into account that most of the reported data refer to ERα as specified in this abstract. Even if ERβ transfections within ER-negative cells may enhance BRK level (Fig. 5 D), one may consider that most observations reported in the manuscript relate to ERα or eventually to an ERα/ERβ ratio favorable to ERα, ERβ presenting an antagonistic activity against the ERα action. The very significant BRK expression in BRCA vs normal mammary tissue supports this primacy of ERα (Suppl. Table 1; normal tissue express mainly ERβ rather than ERα). Hence, ERα rather than ER frequently written in the manuscript would better fit.

On the other hand, studies conducted on breast cancer cell lines clearly show that BRK expression relates to the level of ERα subjected to activation. Tamoxifen known to provoke accumulation of ERα in an inactive form fails to enhance BRK expression. Hence, presence of ERα is required but not sufficient for such an enhancement. Note in this context that the very significant correlation between REα and BRK in breast cancer cell lines (p11; line 1/2) would most probably not similarly hold if the analysis had been restricted to ERα-positive cells. Some ERα-positive cells display indeed relatively low amounts of BRK. (Fig 5 B vs C). Reason for such a property would probably be related to a basal growth condition which may limit BRK expression. Test with E2 stimulated cells would logically validate this view (as well as other ERα activators).

In this context of ERα activation, one may wonder upon a possible contribution of BRK in a rapid phosphorylation of the membrane bound form of ERα required for activations of signal transduction pathways as well as subsequent ERE-dependent and independent transcriptions (see p14). Even if BRK-shRNA fail to affect ERα levels, would such receptors still able to generates such responses. Assessment of this question seems quite easy. It wound not necessary be included within this study but proposed for further investigations.

Other findings of the study were not addressed here: they are sufficiently commented and do not suffer of criticisms.

In brief, this study is original, well conducted and interesting.
Complementary remark: Text p9/fig 3, bottom of the page should be simplified. It looks to be an uncorrectly modified form of a previous manuscript (Fig 3 A/B; Fig 4 not existing)

**Are the methods appropriate and well described?**
If not, please specify what is required in your comments to the authors.

Yes

**Does the work include the necessary controls?**
If not, please specify which controls are required in your comments to the authors.

Yes

**Are the conclusions drawn adequately supported by the data shown?**
If not, please explain in your comments to the authors.

Yes

**Are you able to assess any statistics in the manuscript or would you recommend an additional statistical review?**
If an additional statistical review is recommended, please specify what aspects require further assessment in your comments to the editors.

I recommend additional statistical review

**Quality of written English**
Please indicate the quality of language in the manuscript:

Acceptable

**Declaration of competing interests**
Please complete a declaration of competing interests, considering the following questions:

1. Have you in the past five years received reimbursements, fees, funding, or salary from an organisation that may in any way gain or lose financially from the publication of this manuscript, either now or in the future?

2. Do you hold any stocks or shares in an organisation that may in any way gain or lose financially from the publication of this manuscript, either now or in the future?

3. Do you hold or are you currently applying for any patents relating to the content of the manuscript?

4. Have you received reimbursements, fees, funding, or salary from an organization that holds or has applied for patents relating to the content of the manuscript?
5. Do you have any other financial competing interests?

6. Do you have any non-financial competing interests in relation to this paper?

If you can answer no to all of the above, write 'I declare that I have no competing interests' below. If your reply is yes to any, please give details below.

I declare that I have no competing interests

I agree to the open peer review policy of the journal. I understand that my name will be included on my report to the authors and, if the manuscript is accepted for publication, my named report including any attachments I upload will be posted on the website along with the authors' responses. I agree for my report to be made available under an Open Access Creative Commons CC-BY license (http://creativecommons.org/licenses/by/4.0/). I understand that any comments which I do not wish to be included in my named report can be included as confidential comments to the editors, which will not be published.

I agree to the open peer review policy of the journal