Reviewer's report

Title: Resistance to growth inhibition by TGF-beta is associated with a partial loss of Smad signaling in the absence of alterations of Smad protein levels during in vitro progression of HPV16-immortalized human keratinocytes

Version: 1 Date: 20 April 2013

Reviewer: Ayse Batova

Reviewer's report:

The work presented is technically sound and appropriate for the goals of the study. In addition, the manuscript is well written. Thus, there are no major compulsory revisions or essential minor revisions. The following are discretionary revisions, only, to help improve the manuscript:

1. In relation to fig. 2C: the levels of SMAD 4 appear to be significantly lower in normal HKc compared to all the HKc/HPV16 lines tested. This is not discussed in the text. Some comment on this is warranted as there is a clear difference which should be pointed out.

2. In relation to fig. 6:, it would be helpful to know why the 6 h time point was chosen to compare levels of phosphoSmad2 in normal HKc and the different HKc/HPV16 lines. It appears that by this time point, levels of phosphoSmad2 have decreased substantially after peaking. Why not look at earlier time points?

3. In relation to figs 3 and 4.: It could be more clearly stated that while there exists a difference in the time course of nuclear accumulation of smad3 among normal HKc and HKc/DR, the levels of nuclear Smad 3 appear to be similar in both cell lines once they peak. Is this not true?

4. Out of curiosity, were levels of TGFBR I protein levels examined in the past in addition to the mRNA which was found to be decreased in HKc/DR?

5. Minor grammatical revision: Second paragraph under Results, in the comparison of smad 7 levels in HKc/HpV16 lines versus normal HKc; normal HKc is stated twice as if comparing to itself. This is repeated in the 5th paragraph of the discussion.

6. Minor grammatical revision: In the 5th line, 4rth paragraph under Discussion; stated "have showed" instead of have shown.

7. Include abbreviation SBE for Smad binding element in the introduction where it is first mentioned.

8. The abbreviation ALL refers to acute lymphoblastic leukemia which can either be of B-cell origin (B-ALL) or T-cell origin (T-ALL). These abbreviations should be used as appropriate in the 3rd paragraph under the Discussion.
9. Any theories on the authors’ part that may explain why HKc/DR are completely resistant to growth inhibition by TGF-B despite mostly intact Smad signalling?

**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.