Reviewer's report

Title: Slug Down-Regulation by RNA Interference Inhibits Invasion Growth in Human Esophageal Squamous Cell Carcinoma

Version: 9 Date: 29 December 2010

Reviewer: Ian Beales

Reviewer's report:

The paper has been revised and the data which duplicates that in the authors' similar paper have been removed and the current manuscript is more concise and has greater scientific thrust. However this allowed several issues to become apparent. The paper is scientifically valid but let down by the presentation.

Major compulsory revisions:

1. Overall the quality of the written English seems worse, there are many mistakes in the use of grammar, tense and punctuation and all these need correcting. There are too many to individually list but the absence of spaces after commas and periods makes reading the paper particularly difficult.
2. The results section of the abstract is particularly poorly written.
3. The last sentence on page 1 of the introduction is also very poorly written.
4. Please cite the orginal source of the cell lines used and reference this.
5. The immunohistochemistry section of the methods reads like a set of instructions and not a methods section.
6. Although the data pertaining to the experiments already published in the oesophageal adenocarcinoma cell have been omitted from the current manuscript: the fact that the biological pattern is very similar makes the current data somewhat less unique and this should be pointed out in the discussion.
7. Figures 2 and 3 seem to be transposed and both the text and figure legends are incorrect.
8. The Y-axis in figure 3 is very skewed and I would suggest that a less biased and more representive scale is used (eg 0.4 - 0.8).
9. The fact that slug siRNA induces significant apoptosis (figure 1) but this is not evident in any significant change in total viable cell numbers (figure 2) remains puzzling. The authors explanation based on the methodology used, is rather simplistic and this part of the paper definitely needs further experiments to validate the results. I would strongly suggest that the experiments in figures 1 and 2 are repeated using a positive control to induce apoptosis, any apoptosis inducing agent should be sufficient (such as cisplatin or camptothecin). This would give a relative marker of how much apoptosis is induced by slug siRNA and also provide an internal control to show the reduction in viable cell numbers.
10. Figure 2 (or the MTT assay) also needs to show the MTT data for the time 0
point and there should be a significant rise in MTT over 3 days in the controls and mock transfectants and this does not seem to be the case, which either suggests that the assay is not working appropriately or that the cells are not actually proliferating?

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

**Quality of written English:** Not suitable for publication unless extensively edited

**Statistical review:** Yes, and I have assessed the statistics in my report.

**Declaration of competing interests:**

'I declare that I have no competing interests'