Reviewer's report

Title: Growth Inhibition of Anaplastic Thyroid Cancer by Opioid Growth Factor (OGF)

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Reviewer: jim yeung

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Major Compulsory Revisions:

1. Only KAT18 is anaplastic thyroid cancer: Rebecca E. Schwegge, et al. have just published a paper in JCEM (epub ahead of print, 2008) titled, “DNA profiling analysis of 40 human thyroid cancer cell lines reveals cross-contamination resulting in cell line redundancy and misidentification”. This landmark paper has shaken up the field in thyroid cancer research. They found that many of these human cell lines were widely used in the thyroid cancer field for the past twenty years and are not only redundant, but not of thyroid origin. This negates the relevance of many past publications in the thyroid cancer field including, unfortunately, some of my own papers. According to Schwegge et al., only the KAT18 cell line used by the authors of the current paper under review is anaplastic thyroid cancer, and the other 2 cell lines (KAT-4 and DRO) are not. Therefore, the data using KAT-4 and DRO cells in this paper need to be deleted, and the experiments need to be repeated with a couple of confirmed anaplastic thyroid cancer cell lines reported by Schwegge et al.

2. A “me-too” story for anaplastic thyroid cancer: This research group has been working on the impact of OGF and the role of OGFr in cancer progression for the past 12 years. They have published many interesting papers, but this current manuscript is basically a repeat of past experiments in different cell lines with similar findings. This manuscript did not offer any novel data nor advances in mechanistic insights into how the OGFr suppresses cancer cell proliferation. In past papers from this group, they have demonstrated suppression of cell cycling via p16INK4A and/or p21WAF1 in different cell lines. The connections between OGFr and these cell cycle regulators are still missing. Is p53 involved? What happens when p53 is mutated or deleted? Adding some new mechanistic data will perhaps rescue the significance of this manuscript.

3. “Tachyphylaxis?”: Figure 2B shows the growth curves of KAT18 cells in the absence or presence of OGF. I think that the shape of the growth curve in the continued presence of OGF can be explained by an initial 24 hours of growth suppression followed by an escape from the inhibitory action of OGF with the cells growing at the same rate as the untreated cells (i.e., the slope of the experimental growth curve after 48 hours parallels to that of the control growth curve). This raises the question whether the continued presence of OGF can downregulate the expression of OGFr and/or other post-receptor signaling
molecules leading to tachyphylaxis. For instance, the “binding capacity, but not affinity, of [3H-Met5]enkephalin was reduced by 58% of control levels in tumor tissue from mice of the OGF group” (Cancer Lett. 1997 Jan 30;112(2):167-75). This scenario is also consistent with previous findings from these authors using mouse xenograft models showing that intermittent intraperitoneal administration of OGF is more effective than continuous infusion via mini-osmotic pumps or intratumoral injection (Int J Oncol. 2004 Jan;24(1):227-32). Perhaps the intermittent rise and fall below a certain concentration of OGF to which the cancer cells are exposed is required to avoid tachyphylaxis, and this issue will have important implications on the best way to pharmaceutically administer OGF especially that OGF is going into phase I/II clinical trials. If tachyphylaxis cannot be overcomed, then OGF is useless as a cancer therapeutic agent.

Minor Essential Revisions:

1. Page 12: A growth rate of 7083 cells/hour is about 10% higher than 6375 cells/hour. It is not sure that this difference is statistically significantly different. The rate measurements need to be repeated a couple more times to see if the difference is statistically significant.

2. Figure 4 A & B: It would be more convincing to show the protein level of OGFr after siRNA treatment to assess the degree of suppression of the target protein. Although new protein synthesis is suppressed when the mRNA is knocked down, the protein level and presumably the function of the protein may not drop rapidly depending on the degradation

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.