Reviewer’s report

Title: Overexpression of CDC2/CyclinB1 in gliomas, and CDC2 depletion inhibits proliferation of human glioma cells in vitro and in vivo

Version: 3  Date: 5 September 2007

Reviewer: Michael Berens

Reviewer’s report:

General
This manuscript examined a possible molecular mechanism of CDC2/CyclinB1 on human glioma progression. CDC2 initiates protein kinase activity by complexing with cyclin A, cyclin B and p13suc1. It is known as an active sub-unit of the M-phase promoting factor (MPF) and the M-phase specific histone H1 kinase. This study investigates malignant progression associated with gene expression profiles in human gliomas. Tissue microarrays including grades 1-4 using clinical glioma samples were utilized to determine prognostic power of the CEC2/Cyclin B1 expression in different grades of glioma. Furthermore, to identify the roles of CDC2/Cyclin B1 in human malignant gliomas, CDC2 expression was knocked down in SHG-44 and U251 cell lines by retroviral vectors-expressed short hairpin RNAs. The concept of this study could be very useful and informative for patients in the clinic.

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Major Compulsory Revisions (that the author must respond to before a decision on publication can be reached)
<< none >>

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Minor Essential Revisions (such as missing labels on figures, or the wrong use of a term, which the author can be trusted to correct)

1. Abstract:
a. Designate sub-sections of the text using colons; alternatively, combine sections into one paragraph.
b. Methods section: “Real-time PCR” should not be capitalized
c. Methods section: 4th line down should be “level was subjected to..”
d. Methods section: last sentence, Weight should not be capitalized.
e. Results section: 5th line down, in vitro should be italicized.
f. Results section: Last sentence, “…increased survival ____ times compared to control.” How many times?
g. Conclusions: 3rd line down, in vitro and in vivo should be italicized.

2. Background:
a. 6th line down, “Increasing results demonstrates…” is not a complete sentence.
b. 1st line last paragraph, “…progression associated with gene expression…”
c. 1st sentence last paragraph, take out the word brain for it is understood with glioma.
d. 2nd sentence last paragraph, last word of this sentence, “grades” should be plural.
e. Last sentence last paragraph, “…overexpression of CDC2…”, overexpression of should be eliminated. This wording is confusing.

3. Materials and Methods:
a. Section 4, “Construction of tissue…”. 3rd line down, “All this patients didn’t…” should read “All of these patients did not…”.

4. Results:
a. “Expression of CDC2…” title should all be in italics.
b. First line of first section, “Expression of CDC2 in primer…”, primer should read primary.
c. 2nd line of first section, “…first and second recurrence tumor respectively.” ‘respectively’ should be included.
d. Second section, 3rd line down, “…showed appreciable strong staining…”. Should be appreciably.
e. Second section, 9th line down, When describing figure 4, the manuscript should describe what panels A-R portray. The Figure legend is insufficient to convey the results from the panels in Figure 4. Also, there is no “Table 1” present that coincides with these results.
f. Third section, there is no mention of results for U251 cells at 48-96 hrs. If this is due to death at the prior time point, should a different cell line have been chosen?
g. Third section, unless phenotypic changes occurred, there is no need to show cell images (figure 5), but rather just the cell growth curve (fig. 6) or potentially a death assay (i.e. apoptosis).
h. 4th section, last line of first paragraph, what figure is the real-time PCR results referring to? If not in the manuscript, please state “data not shown”.
i. 4th section, last line, “Recombinant C3…” it is indistinguishable to see the differences between the effectiveness of C1 vs. C3.
j. 6th section, “Therapeutic potential…” 3rd line down, “Virus titre (spelled titer)...RCFU…” Please indicate what the acronym RCFU represents.
k. 6th section, 5th line down, Figure 10 results are said to be significant, therefore it would be good to possibly make a table indicating the size and weight of the tumors along with p-values.
l. 6th section, 6th line down, “Tissue microarrays indicated that the percentage…” Please indicate what these percentages are.
5. Discussion:
   a. 3rd paragraph, for in vivo experiments, were mice ever treated with current
      CDC2 inhibitors to run along side with C3 to make a true comparison of the
      treatments?
   b. It was stated that this viral treatment was unable to infect all tumor cells when
      injected directly. What effects would this CDC2 inhibitor (C3) have on patients if it
      cannot be injected directly into the tumor?

6. Figures:
   a. Would be easier to read if there was one figure per page.
   b. Figure 2 is never cited or mentioned in the text.
   c. Figure 3 needs to be flipped horizontally so that the image can be read from
      left to right.
   d. Figure 5 legend, “… proliferation in human glioma…” human is misspelled.
   e. Figure 7 needs to be labeled A-C; panels A and B need to be flipped
      horizontally to read left to right; panel C needs to have the lanes labeled and also
      formatted to grayscale.
   f. Figure 7 legend, panel C mentioned the “same membrane”. What is this
      referring to?
   g. Figure 8 figure legend needs to be more specific about each point shown, not
      just the overall figure in general.
   h. Figure 9, label the % apoptosis within the quadrant of interest. The quantitative
      message of the figure is poorly depicted.
   i. Figure 10, Are these images the same size or is one expanded? Why are there
      two separate images?

Discretionary Revisions (which the author can choose to ignore)
<< none >>

What next?: Accept after minor essential revisions

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

Quality of written English: Needs some language corrections before being
published

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