Reviewer's report

Title: Cell type-specific anti-cancer properties of valproic acid: a unique role of Erk1/2 signaling at the level of Raf

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Reviewer: André Fedier

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The paper covers in principle an interesting issue on the (pleiotropic) effects of HDAC inhibitors regarding their anticancer activity as well as their role in regulating essential processed in every cell.

Major Compulsory Revisions:

A: The paper must be re-written; In the "Background" section, the last chapter should be omitted and replaced by a clearcut statement of the aim of study, the key questions addressed in the study, and perhaps a concluding sentence giving the main outcome. The "Results" subsections are long and packed with too many details that might be placed in the "Discussion" section. They should be written more concise and better structured. Sometimes it is very hard for the reader to see what the authors want to tell us. The same applies to the "Discussion" section. The authors need to discuss their findings relating them to other studies rather than just repeating the statements from the "Results" section. The "conclusion" section should contain 2 or 3 sentences stating on what's new and in what way this study expands on pre-existing knowledge in the filed.

The figure presentation is clear and self-comprehensive in most cases.

In general, the paper is very dense on information and sometimes the data presentation and interpretation in the text is confusing. Moreover, a clearcut study aim is missing as is the presentation of the key questions that are to be addresses in this study. The authors jet "investigate" or "determine effects of VPA on xxx and yyy. They should be more explicit on this, because it helps the reader to understand why this study was performed. To me it looks like the study does not provide any new information; this is authors seem to "confirm" in 10 (!) cell lines what is already known (e.g. it is widely known that HDAC-inhibitors have pleiotropic effects and that some effects are unrelated to the inhibition of HDAC). The responses to drug and stimuli can be cell-type-specific is known and may be expected.

B: There are some specific points I want to address to:

1. Did the authors determine the proliferation rates of the cell lines? some differential effects observed in the study may be caused by differences in the proliferation rate.
2. Did the authors measure the activity of HDACs (AssayKits are available) before and after VPA treatment? The acetylation of just H3 may not be sufficient
to conclude on HDAC activity inhibition.

3. Likewise the term ERK activity is misleading here. Phosphorylation of ERK is measured and not its kinase activity.

4. The term growth is misleading: growth means gain in weight or volume; better is proliferation (=gain in cell number).

5. The authors use several terms to express ability to migrate (e.g. motility, cell speed, cell displacement, motile behavior (motility is not necessarily migration).

6. Did the authors check whether apoptosis is responsible for some effects? 3mM VPA is quite a high concentration (why is 3 mM chosen?; why 48hrs?)

7. Statistics: why is SEM used instead of SD? In particular, in Figure 4, the authors should indicate the number of cells (=n) assessed. I have some difficulties to see extreme statistical significance (0.001) with U87MG and PC12-E2 if n would have been 100 or more and SEM is calculated. A biologically significant effect is only seen with N2a and BT4Cn, but not with U87MG and PC12-E2 (speed reduction about 12% with U87MG or 20% with PC12-E2).

8. How come that the authors examined 10 cell lines (to find that most effects are cell-type specific and that "changes in ERK are inhibited, activated or unaffected")? It seems a lot of work for not so many "positive" conclusions and new findings in the field.

9. Did the authors consider to test for p21 upregulation by VPA; in particular to compare the growth inhibition seen with the 9 cell lines with the growth stimulation of U78MG. The p21-promoter is directly responsive to HDAC-inhibitors and is rapidly upregulated by them. What happens to p21 in U78MG?

**Level of interest:** An article of limited interest

**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.