Reviewer’s report

Title: Cytotoxicity of amide-linked local anesthetics on melanoma cells via inhibition of Ras and RhoA signaling independent of sodium channel blockade

Version: 0 Date: 18 Sep 2019

Reviewer: Weian Zeng

Reviewer's report:

This is a basic research that tried to address the potential effect of local anesthesia on cancer recurrence. I would like to congratulate the authors for the data presented and well-executed experiments. However, this investigation has several points to consider:

Major issues:

1. Demonstrate straight forward with pictures of migration and apoptosis in all Figures.

2. In this study, bupivacaine doesn't have the anti-melanoma activity. Is it because the concentration of bupivacaine is too low? Have you ever tried a higher concentration of bupivacaine?

3. Overexpressed Ras (Q61L) in A431 melanoma cells have to prove by q-PCR and WB.

4. As shown in Fig. 1, ropivacaine and lidocaine significantly inhibited both A375 and A431 cell migration, growth or survival in a concentration-dependent manner. Although the above effects of ropivacaine were more obvious, the researchers only verified the effects of ropivacaine on small GTPases, and did not verify whether lidocaine affected the expression of small GTPases.

5. In vitro, not in vivo. Further tests in vivo will make the results more convincing.

6. Ropivacaine may have a role in reducing Rac1 activity, even if it is weak (Fig. 3B). In subsequent studies, this role was completely ignored. Is it inappropriate?

Minor issues:

1. The first and third line on page 8 of the manuscript : (Fig. 2D to F) should be (Fig. 3D to F)

Are the methods appropriate and well described?
If not, please specify what is required in your comments to the authors.
Yes

**Does the work include the necessary controls?**
If not, please specify which controls are required in your comments to the authors.

Yes

**Are the conclusions drawn adequately supported by the data shown?**
If not, please explain in your comments to the authors.

No

**Are you able to assess any statistics in the manuscript or would you recommend an additional statistical review?**
If an additional statistical review is recommended, please specify what aspects require further assessment in your comments to the editors.

I recommend additional statistical review

**Quality of written English**
Please indicate the quality of language in the manuscript:

Needs some language corrections before being published

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