Reviewer's report

Title: N-nitroso-N-ethylurea Activates DNA Damage Surveillance Pathways and Induces Epithelial to Mesenchymal Transition in Mammalian Cells

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Reviewer: Huabing Li

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Major Compulsory Revisions

The manuscript by Bodakuntla et al. try to sort out the differences of two DNA repair activation pathways with NEU treatment on in vitro culture cells. The technique and results are clean, and the evidences is pretty convincing, while lack some quantification. I recommend publication of this manuscript on BMC cancer pending on the following concerns:

1. It is suggested to avoid to use two acronyms in the manuscript: MMR and DDR, which appear many times and are important for readers to understand/follow the paper.

2. The authors should quantify the western blot against the controls, to quantitatively and clearly show the increase of the phosphorylation of ATM, Chk2 and Chk1 in all the figures. The numbers may be denoted under each gel, or separately shown, whichever is more straightforward.

3. As for figure 1, while all the other assays were done after 2hrs treatment, the figure 1D &1E was done at 1hr time points, which just confuse the readers. Thus to be consistent, 2hr time point is required for figure 1D of pRPA immunostaining, as well as at the concentration of 0.3mM NEU treatment for both 1D & 1E.

4. As for figure 2, the Msh2 and Msh6 knockdown efficiency should be also quantified by qRT-PCR of their mRNA level. And the pChk2 looks like decreased after MSH6 kd as well as in figure 2C. So all the WB should be quantified.

5. For the important claim that ‘we could detect phosphorylation of ATM and Chk2 10 minutes after initial exposure to NEU while Chk1 phosphorylation was detected 30 minutes after drug damage (Figures 3A and 4A).’, the WB should also be quantified and calculated based on statistics to show agreement with Comet assay.

6. For figure 5, the concentration of NEU is very different from all previous assay, and the author need to explain why they chose those number, instead of 0.3 or 10mM, which are used in all previous figures. Another important point is that the authors need to quantify up to 100 cells to claim how many cells have the ‘EMT-like phenotypes’ after NEU treatment.

7. The discussion is really extensive and long, which is difficult for readers to finish. I would suggest the authors to make it more concise.
Level of interest: An article of outstanding merit and interest in its field

Quality of written English: Acceptable

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

Declaration of competing interests:
I declare that I have no competing interests