Reviewer's report

Title: Use of a recombinant S. typhimurium strain expressing C-Raf for protection against C-Raf induced lung adenoma in mice

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Reviewer: Carlos Alberto A Guzman

Reviewer's report:

General
In the manuscript submitted by Gentschev et al. the authors described the generation of attenuated Salmonella vaccine carrier strains expressing C-Raf. Then, they evaluated the immunogenicity of the resulting vaccine prototypes and their capacity to prevent tumour growth and take using two different murine experimental models.

The experimental approach was appropriate. The obtained results have been clearly presented to the reader. It is expected that the knowledge emerging from this work will facilitate the development of novel immune therapeutic and/or prophylactic interventions against cancer. However, there are still several points requiring attention.

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Major Compulsory Revisions (that the author must respond to before a decision on publication can be reached)

Specific comments

1) The authors stated (page 5) that animals immunized by oral route received 1-2 boosts by i.v. route. It is not clear the rational for coupling both immunization strategies. In addition, in the rest of the manuscript they are just referring to oral vaccination, thereby misleading the reader (i.e., it is extremely different to discuss plain mucosal vaccination or a prime boost protocol). In a similar manner, it is not clear why they are using two different protocols for intranasal vaccination (e.g., different dosages and number of doses).

2) The authors proposed oral and intranasal vaccination protocols. However, they are not showing results for both approaches for all tested parameters in parallel. Thus, it is impossible to judge, which protocol works better or why a particular protocol was chosen. I suggest performing a side-by-side comparison throughout the manuscript.

3) The authors are showing Western blots for the analysis of the humoral immune responses stimulated by the candidates. This is inappropriate. Quantitative determinations should be provided (e.g. endpoint titration). It is also important to analyse the IgG isotypes, as well as to perform kinetic studies for both immunization protocols (i.n. and oral). Another important issue is that the authors mentioned that antibodies were detected in 20% of the animals. Considering that the size of the groups seems to be 5, that means that only 1 out of 5 animals scored positive (i.e., is this result meaningful?).

4) Do the authors have any experimental evidence concerning the role of antibodies in the observed protection (e.g. passive transfer)?

5) Fig .4. Results are only shown for one immunization route. What was the outcome of the other
group(s)? The same is valid for Fig. 6 (see also comment 2).

6) The authors mentioned background problems, which prevented them to perform immune monitoring based on ICS. Have they tried to perform classical CTL tests (in the text something is mentioned in this direction, but it is not clear if they refer to ICS or CTL tests).

7) The resolution/quality of Fig. 1 needs to be consistently improved.

8) The authors should use the proper bacterial nomenclature throughout the manuscript, namely Salmonella enterica serovar Typhimurium.

9) The age of the animals should be incorporated in the description of the immunization protocols (page 5).

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

Discretionary Revisions (which the author can choose to ignore)

What next?: Unable to decide on acceptance or rejection until the authors have responded to the major compulsory revisions

Level of interest: An article of importance in its field

Quality of written English: Acceptable

Statistical review: No

Declaration of competing interests:

none