RESPONSE TO EDITOR AND REVIEWER COMMENTS

R2

Manuscript ID: JoVE58554R1  
  
Title:***In vitro* method to control concentrations of halogenated gases in cultured alveolar epithelial cells**  
Authors: Raiko Blondonnet1,2, Bertille Paquette1,2, Damien Richard3, Rémi Bourg2,4, Géraldine Laplace2,4, Romain Segurel2,4, Henria Pouvelle2,4, Corinne Belville2, Loic Blanchon2, Thomas Godet1,2, Jean-Michel Constantin1,2, Jean-Etienne Bazin1,2, Vincent Sapin2, Matthieu Jabaudon1,2

We thank the Editor for their careful read and thoughtful comments on previous manuscript version. We have carefully taken their comments into consideration in preparing our revision, and we hope the manuscript has been improved. Please find below a point-by-point response to the comments and questions.

**Editorial comments:**

**C1. Some parts of the manuscript are still hard to understand (see e.g. notes in text); please proofread, ideally by a fluent English speaker.**

R1. As recommended, the manuscript had been proofread by an editing and proofreading service ([www.scribendi.com](http://www.scribendi.com)). We added the marked proofread version of the manuscript during the submission process.

**C2. Please revise 1.2.6 and 1.2.7 to avoid previously-published text.**

R2. We apologize for this mistake and the text about this 2 steps had been changed.

**C3. The highlighting is currently very scattered, and it’s hard to pick out a narrative for most of it-e.g., are you intending to film the construction of the chamber? How much of the GC/MS are you intending to show? Please redo highlighting to make this clearer.**

R3. We apologize if the narrative appears hard to understand. We did our best to highlight a new narrative and also to make it clearer.

If it could help the Editor, we took a video when we built the chamber. Of course we could give our movie to the Editor unless he prefers to shoot a new video of the construction of the chamber.

In order to bring more continuity and cohesion into the movie, we decided to removed all the part about GC/MS and to focus the video about the building of the chamber and it use with the anesthetic device.

**C4. 2.1: The dimensions here do not agree with the dimensions in Figure 1; also, this appears to describe a container with a volume of 9.3 (or 9 in Figure 1) L, well over 6.5 L. Lastly, please include the box in the Table of Materials.**

R4. We thank the Editor for this great comment. Indeed if we calculated the volume of the chamber using the dimensions provide in the manuscript we have 9.3 L. We checked again in the catalog of the provider (Please see the attached link - <https://www.curver.com/fra/wp-content/uploads/multisites/2018/01/curver-cata-france-2018.pdf> - *Page:* 106 *Reference:* 00012-416-00) and the container had a volume of 6.5 L. After contacted the provider, the volume was not of 9.3 L because the container was not a “perfect” rectangle but a rectangle with rounded corners. In order to avoid a misunderstanding, we added a sentence in the manuscript in step 2.1 to explain it to the readers.

We apologized for the figure 1 that it was a mistake. We changed the correct width (*i.e.*, 155mm) in figure 1.

The reference of the box had been added in the Table of Materials.

**C5. 4.1.1: Is there a particular procedure for aspirating media? Are you, e.g., opening the chamber, and for how long?**

R5. We thank the Editor for this comment. For aspirating media we used a multi-volume adjustable micropipette set to 1000 µL. We only opened very briefly the chamber to take the studied samples. After we immediately closed the lid of the container in order to keep the concentration of halogenated gases as stable as possible. As suggested, we added some details in the step 4.1.1 to improve the readability and the understanding.

**C6. Figure 3B: What is an experiment here? One well? Were these taken during the same exposure to agent?**

R6. For our experiment we analyzed 1 samples(*i.e.,* 1 well) at each time point during the same exposure to sevoflurane or isoflurane. We repeated the experiment five times. The figure 3B showed the variability between the different experiment. We added in sentence in the legend of the figure 3 to improve the understanding.