Reviewer's report

Title: The Effects of Stress on Myocardial Apoptosis in myocardial infarction

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Reviewer: Guy ROUSSEAU

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The aim of this paper is to confirm whether apoptosis vulnerability markers are present in the myocardium of rats after MI in a MDD model.

The originality of these data is that the authors divided the rats after MI, presenting or not, behavioural signs of depression and compared the results against a well-known model of depression.

However I still have many concerns about different elements of this study.

(MAJOR)
- Did some rats in the depression group present data in the behavioural tests that are similar to the MI group. In other words, did all rats in the depression perform similarly in the behavioural tests?
- In the sucrose consumption test, it is important to indicate the number of total ml of liquid taken for each group. Otherwise the difference in the sucrose consumption could be due to other parameters.
- If I understand correctly, the measurements of caspase-3 or Bax-Bcl-2 were performed on tissues that were previously used in the determination of infarct size (TTC staining). Do you know if TTC influences the results?
- More importantly, how can you claim that apoptosis is important whereas the only apoptotic marker presenting a difference is the Bax/Bcl2 ratio. Is it possible that Bax/Bcl-2 did not only reflect apoptosis but other mechanisms?

(MINOR)
Abstract:

Evan Blue staining is used to determine the area at risk and not to measure infarct size.

Background section:

-It is difficult to follow this section. The link between depression, MI and apoptosis is difficult to understand.
- The description of caspase-3 in this section is very weak compared to Bax/Bcl2.
- Humans are not used in ref 23.
- I don’t believe that the study was designed to clarify the molecular mechanisms but instead to document the apoptosis in the myocardium in a post-MI
Methods:
- A student's t test could not be performed in this kind of experiments. You should perform an ANOVA followed by post-hoc tests such as Bonferonni.
- How did you divide the rats between MI and post-MI depression groups. What are the criteria.

Results:
- The size of area at risk (expressed as % of left ventricle) is not clearly described.
- Similarly, I believe that infarct size is expressed as % of area at risk. This is not clear.

Discussion:
- The pertinence to discuss about extrinsic and intrinsic pathways is questionable. Caspase-3 is an effector enzyme of apoptosis and can be activated by both pathways whereas bax/Bcl-2 is involved mainly in the intrinsic pathway.
- In page 15, the authors indicate that caspase-3 may not be activated during post-MI time-course [21]... in this reference the authors studied the hippocampus and not the myocardium. However the argument to explain the low activity of caspase-3 is very weak. Is it possible that the caspase-3 activity is low because the apoptotic process in the myocardium is finished after 2 weeks post-MI?
- The section on limitations is not convincing.

There are many mistakes throughout the text and in the references.

**Quality of written English:** Not suitable for publication unless extensively edited

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

**Declaration of competing interests:**

I declare that I have no competing interests.