Reviewer's report

**Title:** Effects of carvedilol treatment on cardiac cAMP response element binding protein expression and phosphorylation in acute coxsackievirus B3-induced myocarditis

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**Reviewer:** Rudin Pistulli

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In this study, Ge Li-Sha et al. report on their analysis of the effect of carvedilol on the CREB protein in a mouse model of viral myocarditis. While the effects of betablockers on murine myocarditis, including carvedilol, have been described from several authors, this work adds new insights to the intracellular mechanisms of their actions.

There are some concerns that must be addressed from the authors:

**Major concerns:**

1. The authors have chosen TUNEL staining for their analyses of apoptosis. The quantification of the apoptotic cells is described in the methods section as the percentage of total cells counted in randomly selected fields by microscope.
   a. How did the authors count “total cells” in the selected fields?
   b. How were these “fields” defined in size?
   c. The microscopic photographs in figure 5 do not show “brown” DAB+ staining, as described in methods (!). They resemble rather fluorescence microscopy images of surely something else.

   The authors must therefore re-describe their characterization of apoptosis by a clearer quantification method and appropriate example photographs. As TUNEL staining cannot differentiate between cellular apoptosis and tissue necrosis, I would also suggest a second more specific staining method for apoptotic cells.

**Minor concerns:**

2. The reported heart rate of the control balb/c mice is too low (around 450 b/min, instead of the normally reported 600 b/min). Can the authors please mention this and the possible reasons such as stress-induced bradycardia or the limitations due to the tail-cuff method?

3. The authors characterized the severity of myocarditis merely through HE and fibrosis staining. The lack of any echocardiographic data should be at least mentioned as a study limitation.

4. Recent publications on the cellular mechanisms in murine myocarditis are missing.
Level of interest: An article of importance in its field

Quality of written English: Needs some language corrections before being published

Statistical review: No, the manuscript does not need to be seen by a statistician.

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