Author's response to reviews

**Title:** Myocardial topical negative pressure of -25 mmHg increases myocardial microvascular blood flow

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**Author's response to reviews:** see over
We have consulted a statistical advisor at our hospital, and unfortunately he disagrees about using a repeated measurement test. I assume my explanation about how the tests were performed have been inadequate. I will again try to explain how the tests were done. Below you can see in which order the test were performed in the six different animals.

As you can see the tests were performed in a randomized order and baseline was restored between the different tests. Ischemia is unfortunately impossible to be performed before normal circumstances. According to our statistical adviser, the statistical method preferred would be a two tailed t-test, which is the statistical method used in the article. Do you still believe repeated measures of analysis would be performed? If you feel differently please let me know and I will change it according to your expectations.

Question number 2

Regarding the number of animals, I assume you would like us to specify a power calculation as an objective reason for the present study. Unfortunately, to be able to perform a power calculation, one must have information about the expected or eventual differences between the pressure level groups, which we did not have in the present study. Without that information a power calculation is impossible to be performed, according to our statistical adviser. In the present study no previous data regarding the effects of -25 mmHg on normal and ischemic myocardium exists, since this is a totally new method regarding blood flow changes in the myocardium. According to our statistical adviser, our study has to be considered as a primary indicating study, in order to find out if there is a difference or not between the two different pressure levels of groups. This study, however, may therefore be considered as an indicator for further studies. I hope you will find this information satisfying, and I apologise that this question was not appropriate answered in my previous letter.