Additional comments on the timing used for exercise, rest and sedation:

The timing for the exercise, rest and sedation used in this study was determined from our pilot experiments carried out in rats. We noticed that not all rats exercised for the same amount of time and at a given speed reached exhaustion (a proxy for 100% VO2max), whereas others would become exhausted after a shorter period of exercise (not dissimilar to humans). We later confirmed these observations in mice. Thus, we chose to run the mice at speeds and times that exhausted each individual mouse rather than for a fixed time that would be too much for some and not enough for others (i.e. lead to a range of percent VO2max). This way each mouse reached an exercise endpoint at or near its VO2max. Using this protocol, we observed activation of AMPK and eNOS in the aorta of the rats in our pilot experiments and subsequently in the aorta of mice. The 10 minute rest period added for mice also was the result of our pilot experiments in rats. Post exercise, rats were given pentobarbital and on average required between 5-10 min to be sufficiently sedated for organ extraction. Thus, we added a 10 min rest period for mice to mimic this. After the 10 min rest the mice were placed in an induction chamber in which they were anesthetized by inhaled isoflurane. This generally sedated them within 5-10 seconds and was followed by immediate cervical dislocation and removal of organs.