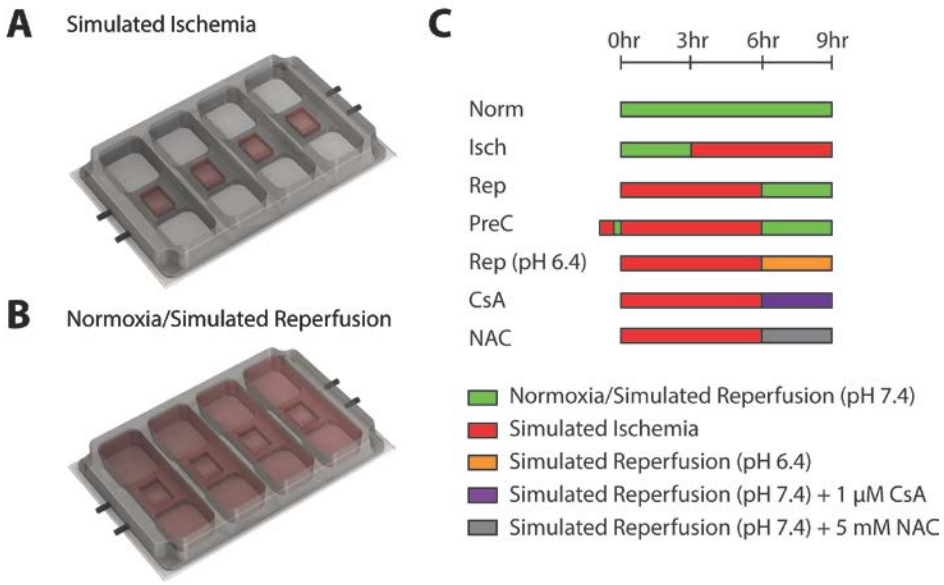

Supplementary FIG. 1. Experimental protocol for simulating ischemia-reperfusion injury and testing cardioprotective therapeutics. Schematic detailing the simulated ischemic and reperfusion state. (**A**) Media volume is restricted to just 120 µL of solution in the construct formation well during simulated ischemia to promote metabolic waste accumulation in the extracellular space. (**B**) Reperfusion is simulated by adding 2 mL of media to the large well to fully washout the accumulated waste, replenish nutrients, and reestablish extracellular pH and ionic balance. (**C**) Schematic detailing the experimental timeline of all the groups. Normoxic (control) constructs ("*Norm*") were maintained under simulated reperfusion conditions for the duration of the experiment, 9 hours. Constructs subjected to ischemia only ("*Isch*") were place in simulated ischemia for 6 hours. Constructs subjected to ischemia-reperfusion injury (IRI) ("*Rep*") were placed in simulated ischemia for 6 hours, and then under simulated reperfusion for 3 hours. Ischemia-preconditioned constructs ("*PreC*") were subjected to 30 minutes of simulated ischemia and 15 minutes of simulated reperfusion prior to simulated IRI. Reperfusion with acidic media ("*Rep (pH 6.4)*"), and cyclosporine A ("*CsA*") and N-acetyl-L-cysteine ("*NAC*") treatment were tested by modification of simulated reperfusion conditions.