

Cold storage of cardiac slices better preserve native characteristics of cardiac tissue than traditional culture methods

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Background: Cardiac slices are a representative experimental model for cardiovascular research, as they maintain the structure and function of the native tissue. Multiple slices can be produced from each heart or specimen from both animals and humans. Although experiments have traditionally used freshly prepared slices to avoid rapid tissue remodeling, prolonged preservation would improve reproducibility while reducing the number of animals needed in research.

Aims: To compare histological and electrophysiological remodeling of cardiac slices maintained in a transwell culture system or stored at 4°C.

Methods: Domestic pigs (n=3) were cardioplegically arrested under deep anesthesia and sacrificed following the guidelines from Directive 2021/63/EU and local ethics approval. 2-cm² transmural tissue blocks were collected from the ventricular wall and used to produce 300 µm-thick slices, which were maintained in transwell culture with supplemented MEM199 or stored at 4°C in Tyrode's solution. Slices were optically mapped immediately after being produced and 24, 48 and 72 hours after sacrifice. Action Potential Duration at 80% repolarization (APD80) and conduction velocity (CV) were measured at all the time points. Slices were fixed and stained with Masson's trichrome to assess histological remodeling.

Results: APD80 and CV were significantly ($p < 0.05$) altered in transwell-cultured slices (APD80 +37%, +109% and +45% and CV +0.58, -1.53, and -1.54 cm/s) but not in cold-stored slices (APD80 +5%, +1% and +0% and CV -0.2, +0.8, and +0.95 cm/s) at 24, 48 and 72 hours respectively. Increased fibrosis was observed after culture, with this increment being lower in the cold-stored slices. Cardiomyocyte striations were only visible in cold-stored slices.

Conclusions: Cardiac slices maintain the native electrophysiology and structure for up to 72h when stored at 4°C but not when maintained in transwell culture. These results support the suitability of cold storage to extend the use of slices during several days.