Proceedings Paper:
McKeegan, PJ orcid.org/0000-0002-5318-2317, Colucci, F, Picton, HM et al. (1 more author) (2018) The use of a novel microfluidic culture device and predictive metabolic profiling as a means to improve murine embryo developmental competence in vitro. In: AETE Proceedings 2018. 34th Scientific Meeting of the Association of Embryo Technology in Europe, 07 Sep 2018 - 08 Sep 2019, Nantes, France. AETE , p. 127.

The use of a novel microfluidic culture device and predictive metabolic profiling as a means to improve murine embryo developmental competence in vitro.

P.J. McKeegan[1], F. Colucci[2], H.M. Picton[1], V. Pensabene[2].

[1] Reproduction and Early Development Research Group, Discovery and Translational Science Department, Leeds Institute of Cardiovascular and Metabolic Medicine, School of Medicine, University of Leeds, UK, LS2 9JT.
[2]School of Electronic and Electrical Engineering, University of Leeds, Leeds, UK, LS2 9JT

Successful embryo development in vitro is directly dependent on the provision of an optimal culture environment that supports coordinated embryonic cell division, metabolism, and genetic and epigenetic development. A number of attempts have been made over recent years to use microfluidic devices in IVF as a means to control the culture environment and so improve embryo developmental competence (quality) in vitro. In this study we have designed, engineered and tested a novel microfluidic device for the in vitro production of murine embryos from the 1 cell zygote stage to the blastocyst stage. Soft lithography was used to prepare microfluidic devices in polydimethylsiloxane (PDMS). The microfluidic device consists of a 400 nL circular chamber (radius 750 mm) where 10 embryos can be loaded, kept in static culture for the full period of culture and visualized by optical and fluorescent microscopy. A series of 2 experiments were conducted to evaluate the efficacy of our microfluidic device for mouse embryo culture. Cryopreserved, IVF-derived, mouse embryos of strain C57BL/6N, provided by MRC Harwell, UK were cultured in KSOM media. Microfluidic culture was used in conjunction with non-invasive analysis of glucose (G), pyruvate (P) and lactate (L) metabolism in spent zygote culture media as a means to improve embryo quality. In both experimental series, data from microfluidic cultures were compared to equivalent end point analyses of control embryos grown in conventional microdrop cultures under oil.

Experiment 1: 2 cell embryos were thawed and cultured in groups of 8-10 in microfluidic devices (n=46) or 10µl control (n=32) drops for 3 days at 37°C under 5%CO_2/ 5%O_2/ N_2 balance. Embryos were removed to individual culture drops for 24h for analysis of energy substrate turnover using the method of Guerif et al. (PLOS ONE, 2013) followed by transfer to fibronectin-coated dishes for assessment of attachment and outgrowth according to the method of Hannan et al. (Endocrinology, 152 (12), p4948-4956, 2011). Blastocyst grade, hatching, attachment, outgrowth rates, and pyruvate and glucose consumption were assessed and were similar between device and control groups (P>0.05). However, lactate output was significantly reduced following device culture vs controls (4.1±0.8 vs 1.4±0.3 pmol/ embryo/ hr, P=<0.0001). GPL metabolism did not predict embryo attachment or outgrowth in either culture environment.

Experiment 2: 1 cell zygotes were cultured individually overnight for analysis of GPL metabolism and assigned to culture groups based on pyruvate consumption, with 1 device and 1 microdrop group per tertile per culture with 10 embryos per group (total n=60 device and n=60 control). Following group culture, individual blastocyst pyruvate consumption was reduced (5.4±2.2 vs 12±1.5 pmol/ embryo/ hr, P=<0.0001). Pyruvate consumption tertile was unaffected by device culture.

Device culture was non-toxic and did not affect embryo development. However, blastocyst pyruvate consumption and lactate output were reduced compared to controls. This may suggest microfluidic culture can be utilised to achieve a controlled, moderate metabolic phenotype, reducing variation between embryo metabolism.

Key words: Microfluidics, metabolism, embryo quality