Ex vivo culture of gastric and intestinal stem cells as organoids

Commonly used acronym: *Gut organoids*

Created on: 12-08-2022 - Last modified on: 16-08-2022

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Organisation

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SCOPE OF THE METHOD

The Method relates to	Animal health
The Method is situated in	Basic Research, Translational - Applied Research
Type of method	In vitro - Ex vivo
Species from which cells/tissues/organs are derived	Mouse
Type of cells/tissues/organs	Gastric and intestinal stem cells

DESCRIPTION

Method keywords

intestine

development
cellular differentiation
gut organoids
epithelium
adult stem cells
foetal progenitors

Scientific area keywords

intestinal stem cells
gastric stem cells
intestinal development
Organoid biobank
organoid preservation
gut regeneration
cellular differentiation

Method description

The tri-dimensional (3D) culture protocols allow isolation and culture of gastric epithelial antral and intestinal stem cells to efficiently generate organoids that recapitulate the mature pyloric and intestinal epithelium *in vitro* and foetal epithelial progenitors of these tissues growing as immature undifferentiated spheroids. The *ex vivo* culture approach is suitable to study gastric and intestinal function in homeostasis as well as in disease and developmental aspects of the gut.

Lab equipment

- Biological safety cabinet,
- Cell culture incubator (37°C, 5% CO2),
- Inverted bright field microscope Binocular,
- Cold light source,
- Ultra-low temperature upright Freezer (for biobank storage),
- FACS for single cell isolation.

Method status

History of use
Internally validated

Published in peer reviewed journal

PROS, CONS & FUTURE POTENTIAL

Advantages

The organoid technology offers the possibility to grow indefinitely foetal progenitors and adult stem cells in culture. The cryo-preserved organoid samples (as a biobank) can be regrown upon thawing, keeping their original properties. This substantially reduces the use of individual animals (pre and post-natally). For adult-derived organoid cultures, all epithelial cell lineages are spontaneously differentiated from adult stem cells, in a proportion similar to that observed in the tissue of origin *in vivo* . Cellular imaging, genomic, transcriptomic and proteomic studies can be easily performed on organoids as well as Drug screening assays and gene editing. A similar protocol is reported for generation of human biobanks from biopsies.

Challenges

This 3R compliant method requires initial use of animals as starting material to generate the biobank. Moreover, this method only allows to investigate epithelium function and does not reproduce the complex *in vivo* environment of a tissue (which includes stromal mesenchymal, immune, nervous cells and systemic derived-factors). Therefore, this organoid technology cannot totally avoid animal use in research.

Modifications

Epithelial-derived organoids can be co-cultured with additional cell types to recomplexify the *in vivo* environment.

Future & Other applications

Patient-derived organoids for personalized medicine.

REFERENCES, ASSOCIATED DOCUMENTS AND OTHER INFORMATION

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Links

Ex vivo Culture of Fetal Mouse Gastric Epithelial Progenitors

Ex vivo Culture of Adult Mouse Antral Glands

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