Human Small Intestinal Organoid-Derived Transwell Model for Studying Inflammatory Bowel Disease (IBD)

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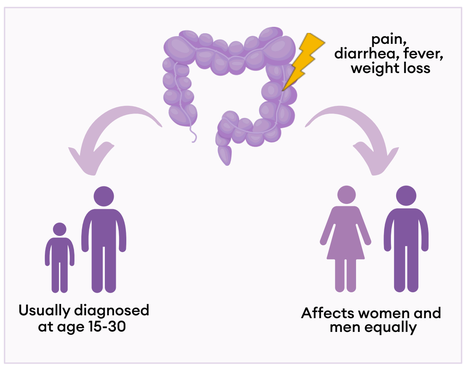
BACKGROUND

Inflammatory Bowel Disease (IBD) is a chronic inflammatory condition of the gastrointestinal (GI) tract characterized by disruption of the intestinal epithelial barrier, leading to persistent inflammation and tissue damage. Despite advances in understanding IBD, effective treatments remain limited, highlighting the need for physiologically relevant models to study disease mechanisms and evaluate therapeutics.

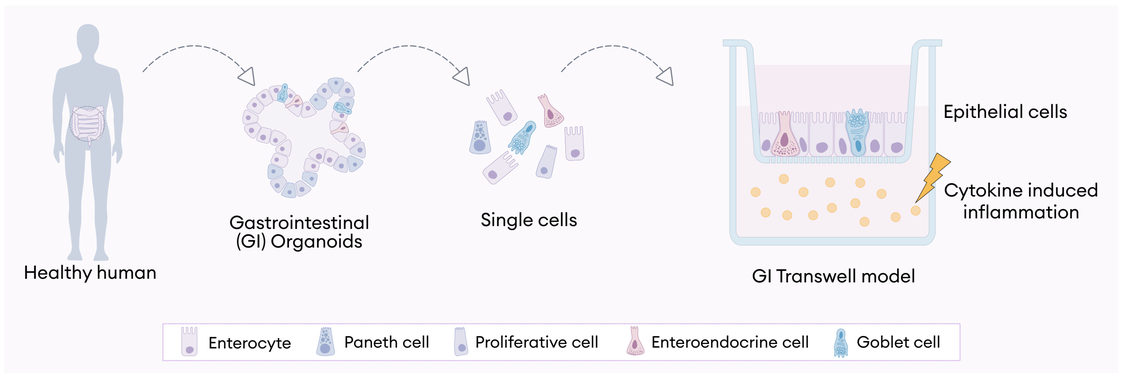
Organoid-derived Transwell systems replicate the cellular diversity and function of human gut epithelium, providing a promising platform for drug testing. We developed a transwell model using primary small intestinal organoids that form stable, functional barriers. To model IBD, we stimulated these epithelial monolayers with pro-inflammatory cytokines, which induced barrier disruption and triggered inflammatory responses.

This approach allows us to mimic key features of IBD pathology in a physiologically relevant, scalable system suitable for studying disease mechanisms and evaluating potential therapeutics.



METHODS

Human gastrointestinal (GI) organoids were dissociated into single cells and seeded onto conventional Transwells to form polarized monolayers. These monolayers establish tight junctions and distinct apical and basolateral compartments. The system enables assessment of drug absorption, toxicity, and therapeutic efficacy through various functional assays. Addition of pro-inflammatory cytokines allows *in vitro* modeling of inflammatory diseases such as IBD.



Readouts include:

- Transepithelial electrical resistance (TEER)
- Permeability assay (e.g. Lucifer Yellow)
- Cytokine secretion (e.g. IL-8)
- Gene expression profiling
- Viability assessment
- Imaging-based analyses of barrier integrity and cell morphology

Small intestinal organoid-derived transwell

PDO-derived monolayers recapitulate the cellular complexity of the small intestinal epithelium and support the study of cytokine-induced epithelial damage and therapeutic responses. When cultured on Transwells, these monolayers polarize, form tight junctions, and establish stable barrier integrity with distinct apical and basolateral compartments.

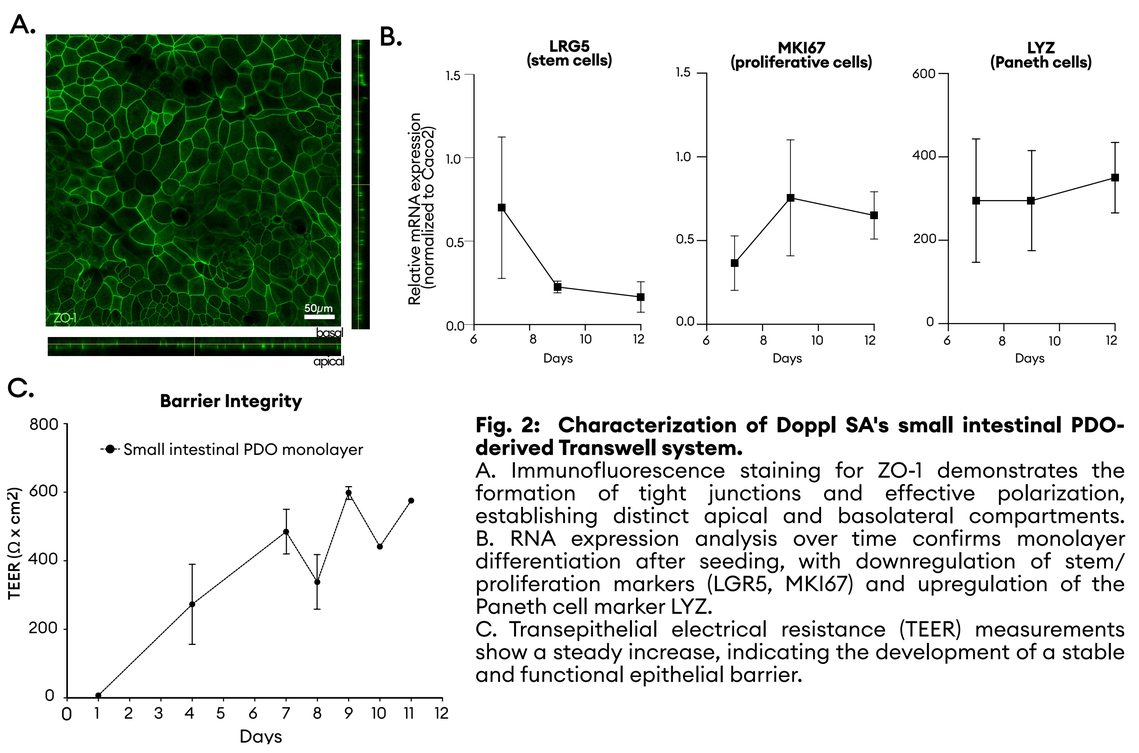


Fig. 2: Characterization of Doppl SA's small intestinal PDO-derived Transwell system.
A. Immunofluorescence staining for ZO-1 demonstrates the formation of tight junctions and effective polarization, establishing distinct apical and basolateral compartments.
B. RNA expression analysis over time confirms monolayer differentiation after seeding, with downregulation of stem/ proliferation markers (LGR5, MKI67) and upregulation of the Paneth cell marker LYZ.
C. Transepithelial electrical resistance (TEER) measurements show a steady increase, indicating the development of a stable and functional epithelial barrier.

RESULTS

Gastrointestinal (GI) organoids

Doppl SA's biobank includes patient-derived organoids (PDOs) from multiple regions of the gastrointestinal tract, as well as intestinal organoids from animal models such as mouse, dog, and other test species. PDOs are characterized by immunofluorescence and RNA-seq to confirm expression of key gastrointestinal cell-type markers at both the protein and transcriptomic levels.

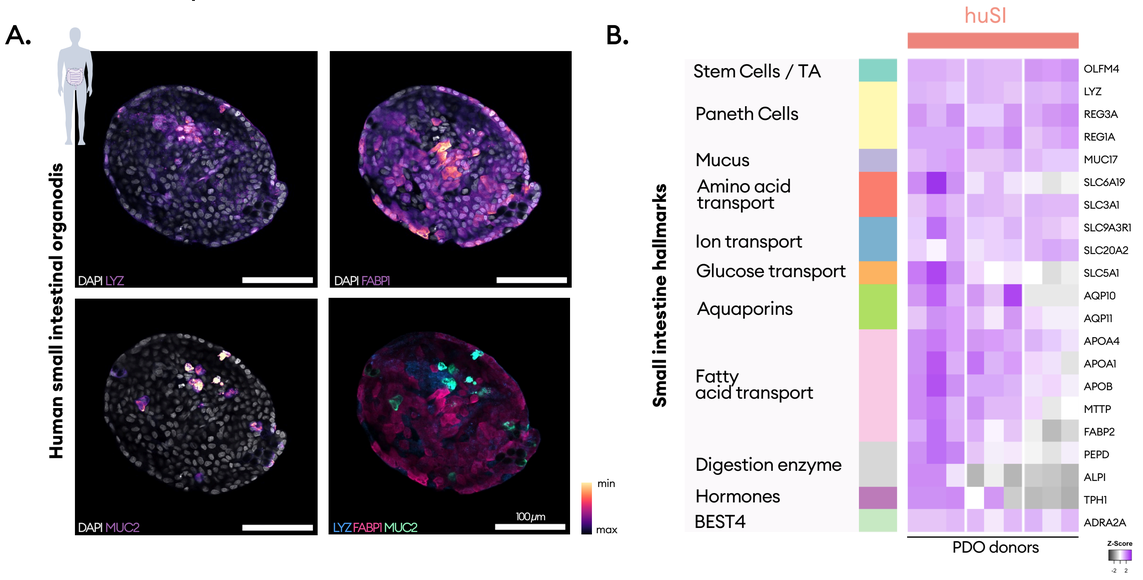


Fig.1: Characterization of Doppl SA's small intestinal organoids.
A. Immunofluorescence staining shows expression of Paneth cell marker (LYZ), enterocyte marker (FABP1), and goblet cell marker (MUC2).**B.** RNA-seq analysis confirms consistent expression of key small intestinal markers across multiple PDO donors.

IBD and inflammatory response

To model IBD, a pro-inflammatory cytokine mix was applied to the apical side of the PDO-derived Transwell monolayers. This treatment induced key features of epithelial inflammation, enabling the assessment of cytokine-induced barrier disruption and cellular responses in a controlled, patient-specific *in vitro* system.

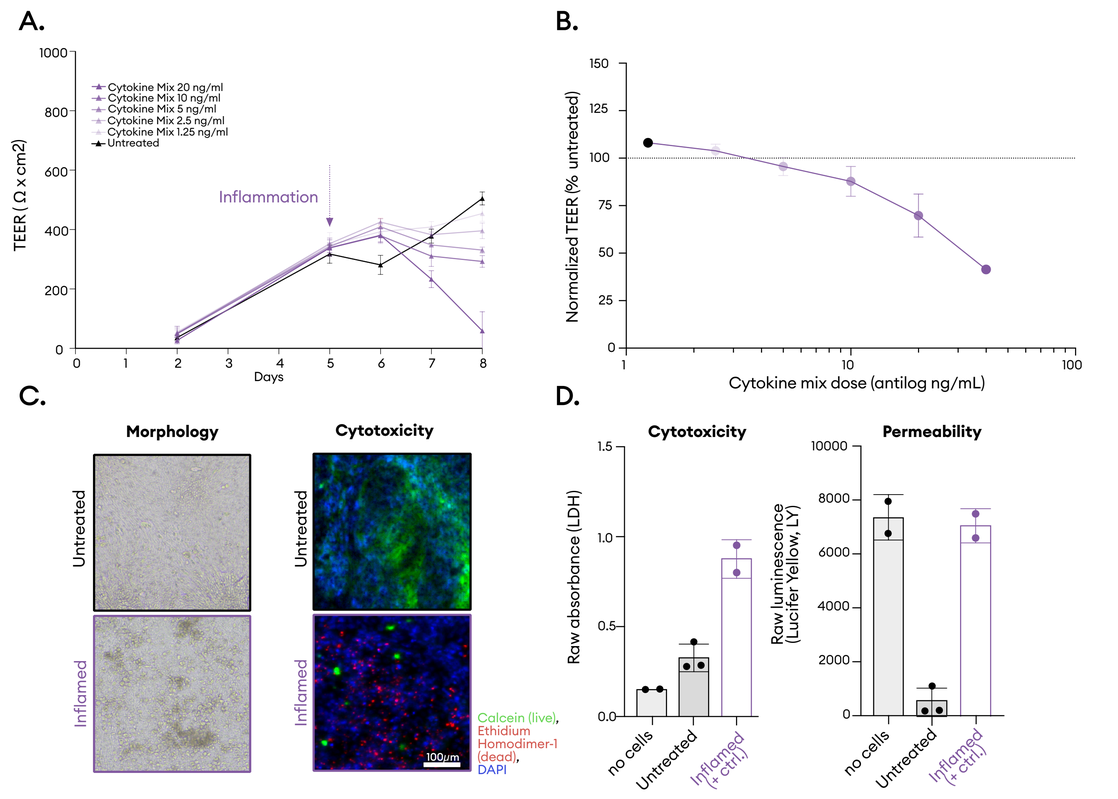


Fig. 3: Effects of cytokine exposure on small intestinal Transwell cultures.
A, B. Barrier integrity, assessed by transepithelial electrical resistance (TEER), decreases in a dose-dependent manner following cytokine treatment. **C.** Representative bright-field images show morphological alterations in cultures exposed to cytokines compared to untreated controls at the experimental endpoint. **D.** Cytotoxicity analysis indicates increased cell damage in response to cytokine exposure. Permeability, measured using the Lucifer Yellow assay, is elevated in cytokine-treated cultures, reflecting compromised barrier function.

SUMMARY

- GI organoid-derived Transwell model mimics gut epithelial structure and function: Polarized, functional monolayers with tight junctions
- Pro-inflammatory cytokine exposure induces key IBD-like phenotypes, including barrier disruption and increased permeability.
- Supports robust functional assays (TEER, permeability, cytokine profiling) for disease modeling and therapeutic screening.

Learn more about Doppl SA, contact us at info@doppl.ch

ACKNOWLEDGMENTS

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