## IntestiCult<sup>™</sup> Organoid Growth Medium (Human)

Efficient Establishment and Maintenance of Intestinal Organoids

### **Intestinal Epithelial Organoids**

The ability to grow intestinal stem cells as organoid cultures has provided researchers with a highly physiologically relevant model system for a wide range of applications. Organoids enable the propagation and maintenance of intestinal stem cells, as well as differentiated cell types, in long-term culture. When grown in organoid culture, intestinal stem cells self-organize into threedimensional (3D) structures containing a single layer of polarized epithelial cells surrounding a functional central lumen, preserving the key cell types and structures of the adult intestinal epithelium. The preservation of these features provides a highly organotypic model system complementary to existing 2D cell culture and in vivo animal models, in some cases reducing or eliminating the need for experimentation in animal models.

The continued adoption of organoid technologies has led to the development of new tools and applications. Characterization of organoid culture systems has enabled further research into the stem cell niche and the factors required to maintain stem cells in vitro. Additionally, functional assays are being developed to inform decisions in precision medicine or to screen drug response in the development of new treatments. Organoids have been cultured from tumor biopsies allowing patient-specific insights into treatment outcomes and creating living bio-banks for the continued study of disease mechanisms. The implementation of organoids has provided a range of tools and techniques allowing for transduction, transfection, and genome editing using CRISPR/ Cas9. Co-culture of intestinal organoids with gut microbes is allowing detailed study of the interactions between microbes and the host epithelium giving insights to pathogenic mechanisms. Organoids have also been grown incorporating other tissues, such as neurons or mesenchyme, providing a model of the complex interactions and signaling events that take place in vivo.



#### Advantages of IntestiCult<sup>™</sup> Organoid Growth Medium (Human):

**COMPLETE.** A complete medium not requiring additional cytokines or growth factors.

**RELEVANT.** Enables easy culture and expansion of intestinal stem cells and differentiated cell types.

**ROBUST.** Consistent generation of intestinal organoids in less than one week.

**EASY-TO-USE.** Simple format and easy-to-follow, optimized protocol.

As new tools continue to be developed for the manipulation of organoid cultures, they are increasingly being viewed as a highly relevant model system.

#### IntestiCult<sup>™</sup> Organoid Growth Medium (Human)

**IntestiCult™ Organoid Growth Medium (Human)** is a complete cell culture medium for the efficient establishment and long term maintenance of human intestinal organoids from primary colonic samples or previously frozen colonic organoids. Organoids are grown from isolated intestinal crypts and can be observed in culture in less than one week, requiring passaging every 7 to 14 days. Organoids grown in IntestiCult™ are suitable for expansion as well as long term maintenance of intestinal stem cell populations allowing for a stable supply of organoids for downstream applications.



Figure 1. Human colonic organoids grown in IntestiCult<sup>™</sup> Organoid Growth Medium (Human)



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#### **Organoid Growth Efficiency**



**Figure 3.** Primary culture of organoids grown in IntestiCult<sup>™</sup> Organoid Growth Medium (Human) as imaged by light microscopy after (A) 2 days, (B) 6 days, (C) 8 days, and (D) 10 days. (E) Organoids grow efficiently over multiple passages

A 48.8  $\pm$  4.5% growth percentage was observed for the first passage, increasing to 85.4  $\pm$  1.4% and 96.5  $\pm$  3.9% for passages 2 and 3 respectively (n=3-4).

#### **Product Information**

PRODUCT	QUANTITY	CATALOG #
IntestiCult™ Organoid Growth Medium	100 mL	06010
Gentle Cell Dissociation Reagent	100 mL	07174
Corning <sup>®</sup> Matrigel <sup>®</sup> Matrix, Growth Factor Reduced (GFR), Phenol Red-free	10 mL	(Corning®) 3526
Costar <sup>®</sup> 24-Well Clear TC-Treated Multiple Well Plates	100 Plates	38017

#### **Protein Localization**



Figure 4. Organoids grown in IntestiCult™ Organoid Growth Medium (Human) display markers of intestinal epithelial cells

Immunofluorescence of intestinal organoids showing (A) DAPI, (B) EPCAM, and (C) Ki67. (D) A merged image demonstrating the position of actively proliferating progenitor cells (Ki67\*).

#### Forskolin Swelling Assay



Figure 5. Forskolin-induced swelling of organoids. Organoids were treated with (A) 5  $\mu$ M Forskolin or (B) with DMSO. Organoid area was measured at 0 minutes and 120 minutes

Forskolin-treated organoids increased 33.5  $\pm$  3.8% in size compared to 7.5  $\pm$  0.8% for control organoids (n=3).



#### WEBINAR

Modeling Human Gastrointestinal Development and Disease Using Pluripotent Stem Cells Scan the QR code or visit: www.stemcell.com/organoids\_webinar

WALLCHART

SnapShot: Growing Organoids from Stem Cells Scan the QR code or visit:

www.stemcell.com/growingorganoids\_wallchart

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