

# Canine Intestinal Organoids in a Dual-Chamber Permeable Support System

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## Materials

| Name                                 | Company             | Catalog Number | Comments             |
|--------------------------------------|---------------------|----------------|----------------------|
| <b>Organoid media</b>                |                     |                |                      |
| ROCK inhibitor (Y-27632)             | EMD Millipore Corp. | SCM 075        |                      |
| [Leu <sup>15</sup> ]-Gastrin I human | Sigma               | G9145-.5MG     |                      |
| A-83-01                              | PeproTech           | 9094360        |                      |
| Advanced DMEM/F12                    | Gibco               | 12634-010      |                      |
| B27 supplement                       | Gibco               | 17504-044      |                      |
| FBS                                  | Corning             | 35-010-CV      |                      |
| Glutamax                             | Gibco               | 35050-061      | glutamine substitute |
| HEPES                                | VWR Life Science    | J848-500ML     |                      |
| Human R-Spondin-1                    | PeproTech           | 120-38-500UG   |                      |
| Murine EGF                           | PeproTech           | 315-09-1MG     |                      |
| Murine Noggin                        | PeproTech           | 250-38-250UG   |                      |
| Murine Wnt-3a                        | PeproTech           | 315-20-10UG    |                      |
| N2 supplement                        | Gibco               | 17502-048      |                      |
| N-Acetyl-L-cysteine                  | Sigma               | A9165-25G      |                      |
| Nicotinamide                         | Sigma               | N0636-100G     |                      |
| Primocin                             | InvivoGen           | ant-pm-1       |                      |
| SB202190 (P38 inhibitor)             | Sigma               | S7067-25MG     |                      |
| Stemolecule CHIR99021 (GSK3β)        | Reprocell           | 04-0004-base   |                      |
| TMS (trimethoprim sulfate)           | Sigma               | T7883-5G       |                      |
| <b>Reagents</b>                      |                     |                |                      |
| Acetic Acid, Glacial                 | Fisher Chemical     | A38-500        |                      |

|   |                   |              |                               |
|---|-------------------|--------------|-------------------------------|
| alpha-D(+)-Glucose, 99+%, anhydrous   | Acros Organics    | 170080010    |                               |
| Cell Recovery Solution  | Corning           | 354253       |                               |
| Collagen I, Rat Tail 3 mg/mL  | Gibco             | A10483-01    |                               |
| FITC-CM-Dextran   | Millipore Sigma   | 68059-1G     |                               |
| Formaldehyde (37%)  | Fisher Chemical   | F79P-4       |                               |
| Glutaraldehyde solution   | Sigma             | G5882        |                               |
| HBSS (1x)   | Gibco             | 14025-076    |                               |
| Matrigel Matrix For Organoid Culture  | Corning           | 356255       | Extracellular Membrane Matrix |
| Paraformaldehyde, 97%   | Alfa Aesar        | A11313       |                               |
| PBS, 1x (Phosphate-Buffered Saline)   | Corning           | 21-040-CM    |                               |
| TrypLE Express  | Gibco             | 12604-021    | Trypsin-like Protease         |
| <b>Materials and Equipment</b>  |                   |              |                               |
| 15 mL Centrifuge Tube   | Corning           | 430766       |                               |
| 9" Pasteur Pipets   | Fisherbrand       | 13-678-6B    |                               |
| Corning Transwell 6.5 mm Polyester Membrane Inserts Preloaded in 24-Well Culture Plates, Pore Size: 0.4 µm, Sterile | Corning           | 3470         | Permeable Support             |
| Millicell ERS (Probes)  | Millipore Sigma   | MERSSTX01    |                               |
| Millicell ERS-2 Voltohmmeter  | Millipore Sigma   | MERS00002    |                               |
| Panasonic incubator   | Panasonic         | MCO-170ML-PA |                               |
| Parafilm M Wrapping Film  | Bemis Company Inc | PM996/EMD    | Flexible Laboratory Film      |
| Tissue Culture Plate 24 wells   | Fisherbrand       | FB012929     |                               |