Probiotic, Anti-Inflammatory Hydrogels for Ulcerative Colitis Treatment

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Ulcerative Colitis (UC)

Part of IBD (Inflammatory Bowel Disease) with UC and Crohn's Disease

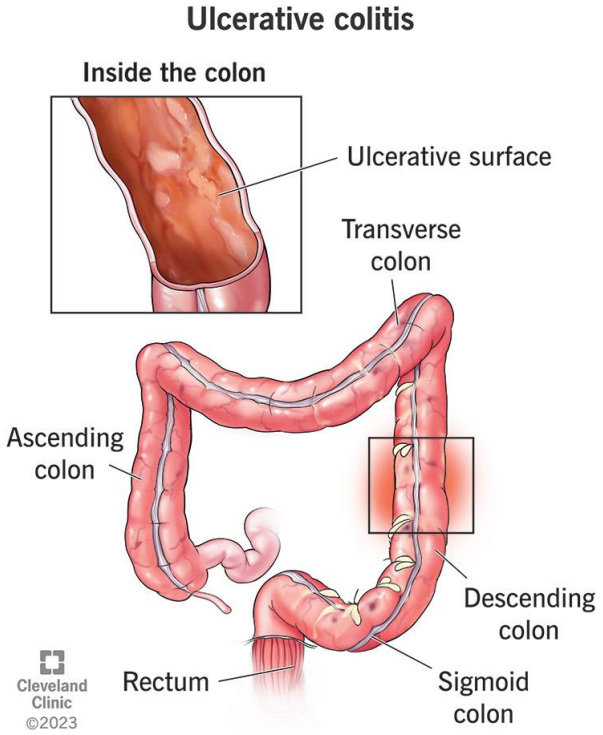
900,000 Americans live with UC

Immune System response

- Overactive WBCs detect FBs and enter the gut
- Sends inflammation to the colon

Symptoms: stomach pain and discomfort, frequent bathroom visits, blood or mucus in stool, fatigue, sudden weight loss

High risk of Colon Cancer - many colonoscopies needed



Cleveland Clinic, 2023

Current Therapies

Unknown causes - reactive care

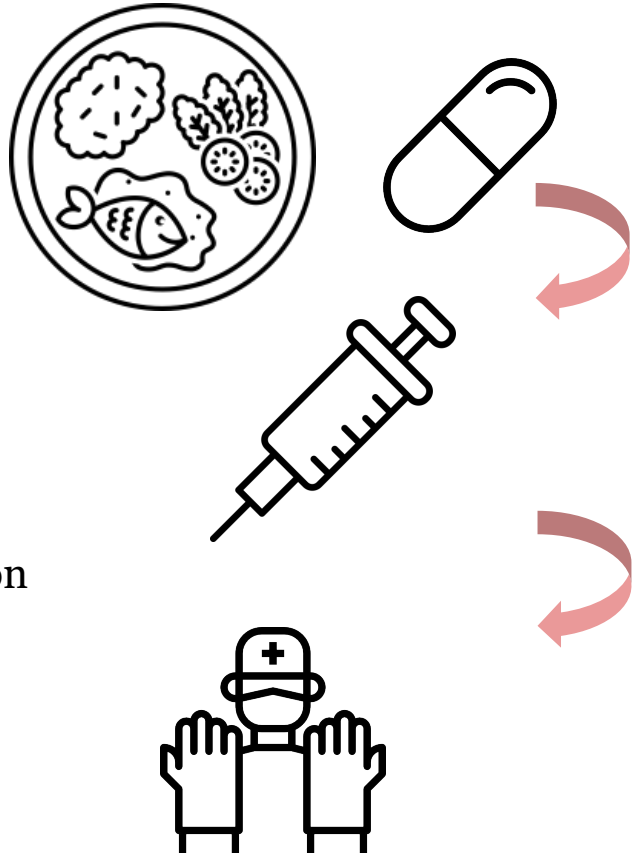
Many patients change diet and lifestyle + medication

Medication - moderate to severe cases

- Oral medication (immunosuppressive steroids)
  - Prednisone - many side effects
- Biologic Therapy - Entyvio, Humera
  - More than half of patients to not achieve remission

Surgery - serious complications, 30%

- Proctocolectomy - removal of colon
  - Ileal pouch
  - Ileostomy - outside pouch



Problem

Over 50% of patients are not in remission

30% of patients go through surgery

There are no preventative therapies for UC

Immunosuppressive steroids induce side effects

High risk of colon cancer - must get frequent colonoscopies

Proposed Biomaterial Treatment

Sprayable bacterial hydrogel infused with anti-inflammatory medicine



Wyss Institute, 2019

Bacteria: Bioengineered E. coli

- Genetically engineered to produce anti-inflammatory cytokine
- Naturally resides in intestines, non pathogenic
- Self-regenerating: longer term treatment, one-time procedure

Interleukin-10: Anti-inflammatory cytokine

- Inhibits antigen presentation and pro-inflammatory cytokine release
- IBD responds well to high dose IV IL-10, cannot be administered orally because stomach acid destroys it

Hypothesis

Our bioengineered bacterial hydrogel will reduce inflammation in ulcerative colitis by delivering IL-10 to regulate immune response at the affected area while being biocompatible and maintaining strong adhesion to the intestinal mucus layer.

Material Properties Assay - Porosity and Diffusivity Analysis

How does the hydrogel network structure affect drug delivery?

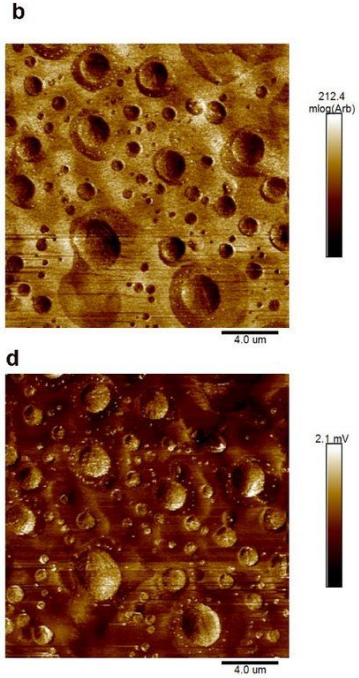
Porosity characterization

- Compare imaging via Atomic Force Microscopy (AFM) & Stimulated Emission Depletion (STED) Microscopy
  - **AFM**: Native conditions, but only images surface + no osmotic swelling
  - **STED**: 3D, but imaged at ambient humidity
  - No **Cryo-SEM**. Tedious, frozen, in vacuum, depth resolution issues
- Image Processing to determine average pore diameter, size distribution, and void fraction

Diffusivity assay

- Hydrogel exposed to IL-10 (or model protein), measure/graph [] vs. time, calculate D_{eff}

Control: Just mucus exposed to IL-10. Test different crosslink densities and distributions! Compare to standard curve for IL-10.



Erkoc, 2017

Biocompatibility Assay - Direct Contact Cytotoxicity

Cytotoxicity Assay

In accordance with ISO 10993-5, we will conduct a **direct contact test** (above cells) to evaluate cytotoxic response of human mucosal cells to bacterial hydrogel infused with interleukin-10 (BHI)

Controls

Cells: NCM460 - Normal Human Colon Mucosal Epithelial Cells at ~80% confluency

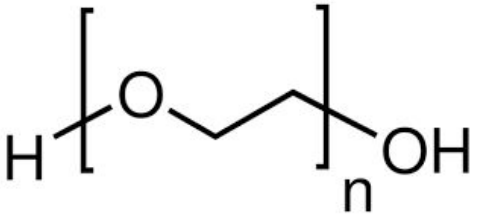
Medium: M3:10F medium (INCELL) + 10% fetal bovine serum + 1% penicillin-streptomycin

Culture Vessels - Glass Petri Dishes

Incubated in 37℃ and 5% CO2

Evaluate cell viability every 24 hours over 3 days

	Material	Expected Result
Test	BHI	TBD
Positive Control	PEG infused with Doxorubicin	Near 0% cell viability
Negative Control	PEG	100% cell viability
Blank	None	No cells, N/A

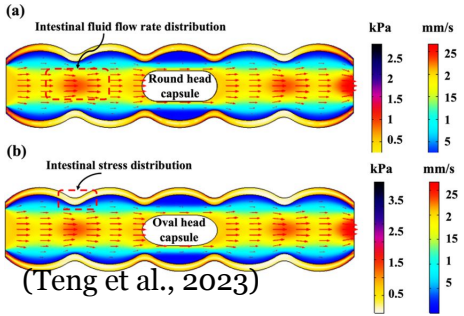




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Benchtop Assay - Mucus Adhesion and Retention Assay

Determine whether the hydrogel can withstand intestinal conditions to enable effective drug delivery



(Teng et al., 2023)

Method

- Coat surface with simulated intestinal mucus
- Apply bacterial hydrogel
- Exposure to fluid shear stress to mimic GI fluid conditions
- Measure adhesion strength and retention time
- Test how mucus (control) vs hydrogel on mucus behaves under shear stress

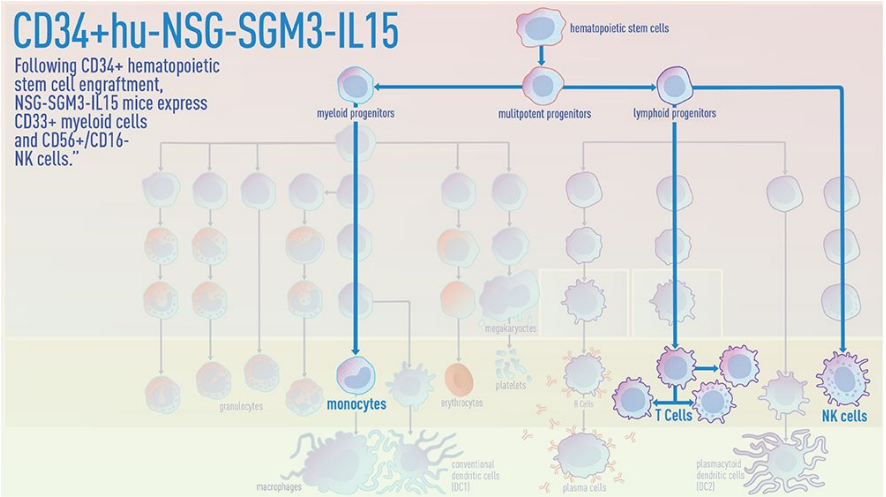
Expected results

- Strong adhesion to mucus layer
- Sustained retention under flow conditions
- Minimal degradation before therapeutic action

Proposed Animal Model - CD34+ NSG-SGM3 mouse models

What is a humanized mouse model?

- NSG mice are highly immunodeficient mouse models that lack native T-cells, B-cells, and natural killer (NK) cells
- Human IL-3, SCF, and GM-CSF genes (all cytokines) are inserted into the mouse genome
- Mice are irradiated to clear bone marrow of hematopoietic stem and progenitor cells (HSPCs)
- CD34+ HSPCs injections that differentiate into human monocytes, T-Cells, and NK cells
- Why? Mouse immune response and inflammation highly mimics human immune response (ISO 10993-6 & 11)
- Limitation: Hydrogel injections rather than endoscopic spray.



CD34+ Humanized NSG-SGM3-IL15. (2026). Jax.org. https://www.jax.org/jax-mice-and-services/preclinical-research-services/humanized-mice/cd34/nsg-sgm3-il15

- Epithelial colitis will be induced via oral administration of Dextran Sulfate Sodium (DSS), which will cause acute or chronic inflammation and mimic ulcerative colitis

Thank You!

Questions?

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