Study Protocol: Single-Arm Validation Study of AI-Driven Personalized Microbiome Modulation for Inflammatory Bowel Disease (IBD)

Title

Evaluating the Efficacy of a Hyper-Personalized Microbiome-Based Treatment for Inflammatory Bowel Disease Using AI-Driven Modulation: An Internal Validation Study

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Background and Rationale

Inflammatory Bowel Disease (IBD) presents a substantial clinical and economic burden, with annual treatment costs exceeding $300 billion. Despite significant medical advancements, IBD—encompassing Ulcerative Colitis (UC), Crohn's Disease, and other severe chronic digestive conditions—remains inadequately managed by conventional treatments. Standard therapies generally target immune suppression but often fail to address a crucial contributor to IBD pathogenesis: gut dysbiosis, or microbial imbalance. Recent studies indicate that personalized microbiome modulation may offer a new, impactful treatment pathway for patients unresponsive to conventional therapies.

The present study, conducted pro-bono by NostraBiome, intends to validate a novel treatment approach combining AI-powered microbiome analysis with precision interventions targeting pathogenic gut bacteria and supporting beneficial species. This AI-driven approach, developed by NostraBiome,draws on the principle that hyper-personalization may dramatically improve therapeutic outcomes, enhancing existing treatments and providing an alternative pathway for patients with treatment-resistant IBD.

This internal validation study will assess whether microbiome modulation, powered by NostraBiome's proprietary AI algorithm, leads to clinical improvements in stool patterns, symptom severity, inflammatory markers, and quality of life over three months. Additionally, the study aims to build internal confidence among NostraBiome's founding team in the efficacy and safety of this innovative therapeutic direction.

Study Objectives

1. **Primary Objective**: Evaluate the efficacy of personalized microbiome modulation in reducing stool frequency, improving stool consistency, and reducing the presence of blood in stool among IBD patients.
2. **Secondary Objectives**:

    - Determine changes in inflammatory markers, nutrient deficiencies, and overall quality of life.
    - Establish baseline data for potential improvements in existing IBD treatments through microbiome modulation.
3. **Exploratory Objective**: Establish preliminary evidence supporting the potential of AI-driven microbiome modulation to enhance existing treatment protocols for IBD.

Study Design and Schema

Study Type

- **Design**: Single-arm, open-label study
- **Duration**: 3 months

Study Schema

1. **Enrollment**: Participants recruited through a combination of online advertisements (Google AdWords,) and direct outreach in IBD support groups.
2. **Baseline Assessments**: Initial microbiome profiling, disease history intake, and baseline laboratory testing.
3. **Intervention Period**: Monthly microbiome profiling with AI-driven adjustments to individualized treatment protocols.
4. **Follow-up and Final Assessment**: Comprehensive end-of-study evaluation, including clinical markers and microbiome composition.

Participant Selection Criteria

- **Inclusion Criteria**:
    - Diagnosed with UC, Crohn's Disease, or another severe chronic digestive condition for >3 years.
    - Documented failure with at least three previous IBD treatments.
    - Currently on or previously attempted biologic or immunosuppressive therapy.
    - Representing diverse age groups (26-55 years), gender (62% male, 38% female), and ethnic backgrounds.
    - Consent to participate in a three-month study and commitment to follow prescribed intervention protocols.
- **Exclusion Criteria**:
    - Sensitivities or allergies to any recommended treatment components (e.g., antibiotics, probiotics).
    - Recent use of antibiotics within one month prior to study initiation.

Risk Mitigation

- All treatment recommendations and tools utilized are regulatory approved.
- A licensed physician reviewed and validated each treatment protocol to ensure safety.
- Blood tests were conducted, and participants' microbiome profiles were monitored for dysbiosis and changes.

- Allergy testing was performed for all participants with antibiotic sensitivities; three participants were excluded based on allergy preliminary results.

Intervention Protocol

Personalized Microbiome Modulation

Participants underwent a hyper-personalized treatment plan aimed at modulating their microbiome composition through AI-powered recommendations.

1. **Initial Microbiome Profiling**:
    - Collected stool samples using commercially available 16S rRNA sequencing kits.
    - Analyzed baseline microbial diversity and identified pathogenic bacteria.
2. **Lifestyle and Health Assessment**:
    - Comprehensive intake of dietary habits, stress factors, and personal IBD history.
    - Detailed recording of disease triggers and previous treatment history.
3. **AI-Driven Treatment Customization**:
    - The AI algorithm identified pathogenic bacterial signatures unique to each participant and suggested modulation strategies, which included:
        - **Dietary Recommendations**: Targeted dietary changes to support microbial diversity.
        - **Prebiotic and Probiotic Supplementation**: Specific formulations based on microbiome profiles.
        - **Nutritional Adjustments**: Addressing vitamin and mineral deficiencies (notably Vitamins B, D, iron, calcium, and zinc).
        - **Antimcrobial against**

          Monthly adjustments to treatment plans based on updated microbiome profiles and clinical response.

4. **Continuation of Standard Therapy**:
    - Participants on biologic or immunosuppressive therapies maintained their regular regimens to evaluate any synergistic effects with microbiome modulation.

Data Collection Points

Timepoint	Baseline	Month 1	Month 2	Month 3 (End)
Clinical Symptoms (Stool Frequency, Consistency)	✔	✔	✔	✔
Stool Diaries	Daily	Daily	Daily	Daily
Microbiome Profiling (16S rRNA)	✔	✔	✔	✔
Inflammatory Markers (CRP, Fecal Calprotectin)	✔	✔	✔	✔

Nutritional Status (Vitamins B, D, Iron)	✔	—	—	✔
QoL and Symptom Scores	✔	✔	✔	✔

Outcome Measures

Primary Clinical Outcome Measures

1. **Stool Frequency and Consistency**:
    - Daily self-reports logged via a mobile app; consistency evaluated monthly using the Bristol Stool Scale.
2. **Blood Presence in Stool**:
    - Self-reported and verified through fecal tests.

Secondary Outcome Measures

1. **Microbiome Composition and Diversity**:
    - Monthly 16S rRNA sequencing to measure microbial diversity and track changes in pathogenic and beneficial bacterial populations.
2. **Inflammatory Markers**:
    - Blood CRP levels and fecal calprotectin measured monthly to assess systemic and gut-specific inflammation.
3. **Vitamin and Mineral Levels**:
    - Baseline and end-of-study testing for Vitamin B, Vitamin D, iron, calcium, and zinc.
4. **Quality of Life (QoL)**:
    - Monthly QoL assessment using the Short Inflammatory Bowel Disease Questionnaire (SIBDQ).
5. **Bloating, Gas, and Abdominal Pain Scores**:
    - Daily self-reported scores on abdominal symptoms logged via mobile app.

Exploratory Measures

- **Libido and Sexual Health**: Self-reported monthly to explore any secondary effects of microbiome modulation on general well-being.

Statistical Analysis

Primary Analysis

- **Changes in Stool Frequency, Consistency, and Presence of Blood**: Paired t-tests or Wilcoxon signed-rank tests for within-group comparisons from baseline to the three-month endpoint. Significance level set at $p < 0.05$.

Secondary Analysis

- **Microbiome Composition**: Within-subject analyses to evaluate shifts in bacterial diversity, with clustering methods to assess changes in pathogenic and beneficial species.
- **Nutritional and Inflammatory Markers**: Repeated measures ANOVA or equivalent non-parametric tests to track changes in vitamin deficiencies, CRP, and fecal calprotectin over time.
- **Quality of Life and Symptom Scores**: Descriptive and trend analyses using linear mixed models to evaluate symptom trajectory.

Data Management and Missing Data Handling

- A digital platform will capture via questionnaire the inputs from the users. Missing data will be imputed using multiple imputation methods, with sensitivity analyses conducted to verify data robustness.

Ethical Considerations

- **Confidentiality**: Patient data is anonymized and coded.
- **Informed Consent**: Participants were informed about study objectives, risks, and benefits before enrollment.
- **Ethics Approval**: Conducted in alignment with the Declaration of Helsinki
- **Safety Oversight**: Monthly physician reviews and continuous monitoring for any adverse effects.

Expected Outcomes

This study anticipates that AI-driven microbiome modulation will demonstrate clinical improvements in stool control, inflammation reduction, symptom severity, and quality of life in IBD patients. Furthermore, these findings may substantiate the effectiveness of hyper-personalized treatment approaches as viable adjuncts to standard IBD therapies, highlighting NostraBiome's potential as a pioneer in personalized microbiome-based medicine.