UNIVERSITY OF ZAMBIA
BIOMEDICAL RESEARCH ETHICS COMMITTEE

Telephone: 260-1-256067
Telegrams: UNZA, LUSAKA
Telex: UNZALU ZA 44370
Fax: + 260-1-250753

Ridgeway Campus
P.O. Box 50110
Lusaka, Zambia
E-mail: unzarec@unza.zm

Federal Assurance No. FWA00000338 NHRAR-REC No 2021-05-0002 IRB00001131 of IORG0000774

16th March, 2026

Your REF. No. 7400-2025

Mr. Michelo Simuyandi,
CIDRZ,
PO Box 34681,
Lusaka.

Dear Mr. Simuyandi,

RE: REQUEST FOR APPROVAL FOR PROTOCOL AMENDMENT – "SPATIAL TRANSCRIPTOMIC PROFILING OF MUCOSAL IMMUNE RESPONSES TO ORAL ROTAVIRUS VACCINATION IN ZAMBIAN ADULTS WITH ENVIRONMENTAL ENTEROPATHY: A PILOT FEASIBILITY STUDY (REF. NO. 7400-2025)

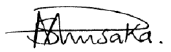
We acknowledge receipt of the amendment request to the above study.

The amendments were **reviewed** and **approved** as follows:

a. A total sample size of 46 participants will be recruited and allocated to three groups to optimise time-point capture while minimising procedures. Six (6) additional participants were enrolled during an initial feasibility phase of the study. These participants met all eligibility criteria and underwent study procedures in accordance with the approved protocol. They are now formally incorporated into the total study sample.
b. Study Design and Section 8: Study Procedures and Visit Schedule:
Participants will receive two (2) oral doses of Rotarix administered on Day 1 following completion of screening and baseline eligibility confirmation on Day 0, followed by a 2-hour observation period.
c. Amendment to Section 8: Timing of Vaccination:
Vaccination will occur one (1) day after completion of screening and baseline procedures to allow adequate time for the participant's gut mucosa to recover following baseline endoscopic biopsy procedures prior to vaccine administration.

d. Following vaccination, stool samples will be collected within a window of Day 1 to Day 5 post-vaccination to capture early viral shedding kinetics.
e. Amendment to Section 8: Blood Collection Volume Up to 40 mL of peripheral blood will be collected at the specified study time points for immunological analyses, including PBMC isolation and downstream cellular assays.

Yours sincerely,



Prof. Sody Mweetwa Munsaka, BSc., MSc., PhD
CHAIRPERSON
Tel: +260977925304

Temporal and Spatial Immune Profiling of Oral Rotavirus Vaccine Responses in Zambian Adults with Environmental Enteropathy

Protocol Version: 2.0
Protocol Date: 21 February 2026
Study Sites: St Augustine Clinic; CIDRZ Laboratories; Lusaka, Zambia

1. Synopsis

Study Type	Prospective, time-series pilot study (feasibility + discovery).
Principal Investigator (PI)	Michelo Simuyandi (CIDRZ)
Co-Investigators	Prof Zaza Ndhlovu
	Prof Paul Kelly
	Dr Monde Muyoyeta
Study Population	46 healthy adult participants (≥18 years) residing in Lusaka, Zambia.
Key Assay	**(i)** Spatial transcriptomics (ST) applied to duodenal biopsy tissue (FFPE and fresh-frozen, for complementary assays) **(ii)** Flow cytometry: for immediate and memory immune responses **(iii)** ELISA: for rotavirus specific serum IgA and IgG
Primary Objective	Demonstrate technical and operational feasibility of performing ST on small intestinal biopsies collected serially and generating analysable spatial data in a low-resource setting.
Secondary Objectives	Characterize spatial transcriptional dynamics of mucosal immune responses at early (Day 2), intermediate (Day 10) and (Day 28) time-points; identify spatial niches and cell states associated with reduced response in participants with markers of Environmental Enteropathy (EE).
Total Duration per Participant	Up to 28 days.
Total Study Duration	12 months (approvals, recruitment, sample processing, sequencing, analysis).

2. Background and Rationale

Environmental Enteropathy (EE) is a chronic inflammatory disorder of the gut prevalent in low- and middle-income countries (LMICs)[1]. It is characterized by villous blunting, crypt hyperplasia, and chronic inflammation[2,3]. This syndrome is strongly implicated in impaired nutrient absorption, growth faltering, and reduced efficacy of oral vaccines such as Rotarix®[3,4]. Despite robust global efforts to improve vaccine efficacy, the precise mucosal immune defects at the tissue level that drive this failure remain poorly understood, especially in the context of chronic EE[5].

The intestinal mucosa represents the largest immune interface of the human body and is constantly exposed to commensal microbes, dietary antigens, and potential pathogens. The mucosal immune system balances tolerance and defense through specialized epithelial and immune cell networks[6]. In healthy gut tissue, innate immune cells such as macrophages, dendritic cells, and innate

lymphoid cells (ILCs) maintain homeostasis, while adaptive lymphocytes, including IgA-secreting plasma cells, Th17 cells, and regulatory T cells (Tregs) mediate targeted immune responses[7].

Treg cells, characterized by (i) expression of the transcription factor Foxp3, (ii) $CD25^{hi}$ and (iii) $CD127^{lo}$, are induced both centrally in the thymus and peripherally in the gut mucosa. Within the intestine, their induction is driven by microbial metabolites (e.g., short-chain fatty acids such as butyrate)[8], retinoic acid, and antigen presentation by tolerogenic dendritic cells[9]. These signals promote the differentiation of naïve $CD4^+$ T cells into peripherally induced Tregs (pTregs), which maintain mucosal tolerance and limit excessive inflammation. This local induction of Tregs is essential for preventing tissue injury and sustaining epithelial integrity[7]. Thus, the disruption of this balance as seen in EE can result in persistent immune activation, villous atrophy, and impaired barrier repair, leading to microbial translocation and systemic inflammation which are associated with poor vaccine immune response.

Spatial transcriptomics (ST) offers a transformative approach to study these defects. Unlike bulk or single-cell RNA sequencing, ST integrates quantitative gene expression data with the histological architecture of the tissue, enabling discovery of discrete immune niches, epithelial–immune interactions, and spatial gradients of response that bulk approaches cannot resolve[10]. Such understanding is now critical to explain the defects in vaccine response which are such a problem in sub-Saharan Africa.

Key knowledge gaps this study will address:

- Feasibility: Technical and logistical challenges in generating high-quality ST data from small, serial duodenal biopsies in a Zambian clinical setting (a critical step for future larger trials).
- Mechanistic Insight: Defining the precise temporal and spatial organization of the mucosal immune response to an oral vaccine (Rotarix®) in adults with prevalent markers of EE.
- Biomarker Discovery: Identifying novel spatial biomarkers (e.g., specific gene expression domains or cell-cell interaction hotspots) that correlate with diminished serologic/mucosal immune responses and with fecal biomarkers of EE (e.g., MPO, calprotectin).

Study rationale

EE causes epithelial injury and chronic mucosal inflammation in the small intestine, creating altered tissue microenvironments that change immune cell localization and function[11]. Single-cell and spatial profiling studies show altered cellular composition and transcriptional states in distinct intestinal micro-domains, consistent with spatially compartmentalized immune outcomes[12]. EE is associated with expansion of intestinal regulatory T cell (Treg) populations that suppress local $CD4^+$ T cell responses and can reduce oral vaccine immunogenicity[13] . Mechanistic studies indicate that intestinal immune “microniches” enforce local Treg function, and disruption of epithelial integrity or inflammation can modify niche composition and immune compartmentalization[14]. Systemic nutritional status modulates epithelial repair, barrier function, and mucosal immune competence; nutrient deficiency can reproduce EE-like epithelial injury, linking nutrition to the timing and magnitude of mucosal immune responses[15] .

The use of healthy adult volunteers minimizes the clinical risk associated with serial endoscopy in a pediatric population while allowing us to study the underlying EE-associated pathology and immune response relevant to the oral vaccine immune failure challenge.

3. Objectives

Primary Objective

1. To demonstrate feasibility (clinical, logistic, and technical) of collecting serial duodenal biopsies and generating high-quality spatial transcriptomics data (as measured by predefined QC metrics) in a Zambian clinical-research setting.

Secondary Objectives

1. To describe spatial gene-expression changes in duodenal tissue across (i) Baseline, (ii) Day 3, (iii) Day 10 and (iv) Day 28 following a double dose of oral Rotarix.
2. To identify spatially localized immune cell populations and pathways that are associated with mucosal immunologic readouts and clinical biomarkers of EE.
3. To compare ST-derived cell-type maps with histology and targeted immunostaining for validation.

Exploratory Objectives

1. Explore associations between baseline mucosal transcriptional signatures and magnitude or duration of systemic or mucosal antibody responses.
2. Assess feasibility of shipment and cross-platform ST comparisons.

4. Study Design

This is an observational pilot study using a time-series sampling design. Participants will receive a double oral Rotarix dose at baseline. Serial duodenal biopsies will be collected from predefined subgroups to capture early, mid, and late mucosal responses while minimizing per-participant invasive procedures (the sparse sampling design allows construction of profiles of responses over 4 time points while no individual participant undergoes more than 2 endoscopies). The study design avoids confounding from placebo-related procedural differences and allows for robust within-subject longitudinal comparisons.

5. Study Population and Eligibility Criteria

Inclusion Criteria

- Age ≥ 18 years and ≤ 50 years.
- Able and willing to provide written informed consent.
- Reside within Lusaka district and available for scheduled follow-up.
- Willing to undergo two endoscopy procedures with serial sample collection.

Exclusion Criteria

- Pregnant or breastfeeding.
- Active GI disease requiring treatment (e.g., active peptic ulcer disease, gastrointestinal bleeding).
- Known severe immunodeficiency (HIV with CD4 < 200 cells/mm³, current cancer chemotherapy, systemic corticosteroids equivalent to >20 mg/day prednisolone for >2 weeks).
- Severe comorbidities rendering endoscopy unsafe (e.g., severe cardiopulmonary disease, uncontrolled hypertension, oropharyngeal abnormalities).
- Use of anticoagulants where suspension is unsafe or unwillingness to withhold anticoagulation when clinically indicated.
- Receipt of any live vaccine within 30 days prior to enrolment or planned live vaccine within 30 days after Rotarix administration.

- Any condition judged by the investigator to compromise participant safety or data integrity.
- Participants who had a recent diarrhoea episode, or who have taken NSAID drugs or antibiotics, will be eligible for re-assessment after a month without this disqualifier.

Pregnancy testing and counselling: Women of reproductive potential will require a negative urine pregnancy test within 24 hours prior to endoscopy and vaccination and will be counselled to avoid pregnancy during the first 30 days post-vaccination.

6. Sample Size and Allocation

A total sample size of 46 participants, will be recruited, with six participants being allocated to a preliminary feasibility cohort, and allocated to three groups to optimize time-point capture while minimizing procedures:

Group	N	Endoscopy Visits (Biopsies & ST)	Non-Invasive Visits (Blood/Stool)	Key Timepoints Captured
A	12	Baseline (Day 0) and Day 3 post-vaccination (Endoscopy & Biopsy)	Baseline and Day 3 blood, stool sampling (Day3 ±2, Day 5±2, Day 10±2; early kinetics	Early kinetics
B	12	Baseline (Day 0) and Day 10 post-vaccination (Endoscopy & Biopsy)	Baseline and Day 10 blood, stool sampling Day 5±2; Day10 ±2; intermediate kinetics	Mid kinetics
C	16	Baseline (Day 0) and Day 28 post-vaccination (Endoscopy & Biopsy)	Baseline and Day 28 blood, stool sampling (Day 3±2, Day 5±2, Day 10±2; late kinetics	Serology/shedding kinetics only

Justification: This is a pilot feasibility study. The sample sizes are chosen to test logistical pipelines and generate robust pilot data across multiple timepoints, providing sufficient statistical estimates for future, powered trials.

7. Recruitment, Screening and Consent

- **Recruitment:** Potential participants will be identified via outpatient clinics, community outreach, and research registries following community engagement meetings with local leaders. Information materials will be utilized.
- **Screening Assessments:**
    - Medical history, physical exam, and vital signs.
    - Standard baseline blood tests (FBC) where indicated.
    - Rapid HIV testing with pre/post-test counselling.
    - Urine pregnancy test for women of childbearing potential.
    - ECG if clinically indicated.
    - Baseline stool sample for rotavirus PCR and fecal biomarker testing.
- **Consent Process:** A detailed, two-stage consent process will be utilized. Consent forms (in English, Bemba, and Nyanja) will explicitly cover endoscopy risks and sedation, genetic/transcriptomic analyses, possible de-identified data sharing, international shipment options, and the right to withdraw without prejudice.
- **Compensation:** The participants receive K200 per study visit as reimbursement for time

and transport. Meals will be provided on endoscopy days, and transport can be arranged if needed.

8. Study Procedures and Visit Schedule

Visit	Timing	Procedures / Groups Involved	**Groups Involved**
Screening	Day −14 to −1	Eligibility, Labs (HIV, PBMCs,Viral load,CD4, Pregnancy), Consent Confirmation. All groups	All
Baseline (V1)	Day 0	Endoscopy & Biopsy, Sample Handling, Blood Draw (20-40 mL), Stool Collection	All
Follow-up V1b	Day 1	Rotarix Administration, 2-hour observation. All groups	All
Follow-up V2	Day 3 (±1 days)	Stool sampling (All groups) Endoscopy & Biopsy (Group A only), Blood Sampling Draw (20-40 mL), AE Review.	All (stool); A (endoscopy)
Follow-up V2b (stool only)	Day 5 (±1 day)	Stool sampling only	All
Follow-up V3	Day 10 (±2 days)	Stool sampling (All groups) Endoscopy & Biopsy (Group B only), Blood Draw (20-40 mL)/Stool Sampling, AE Review.	All (stool); B (endoscopy)
Follow-up V4	Day 28 (±3 days)	Endoscopy & Biopsy (Group C only), Blood Draw (20-40 mL)/Stool Sampling, AE Review.	C
Exit / Final	Day 28 (±3 days)	Clinical Review, Blood Draw (20-40 mL)/Stool Sampling, Exit Survey. All groups as applicable	All (as applicable)

9. Endoscopy and Biopsy Procedures

All endoscopy and Biopsy procedures including tissue processing will be done at the St Augustine Clinic Endoscopy Unit.

Sedation: Conscious sedation with midazolam and pethidine with dosage adjusted by clinical judgement; continuous monitoring (pulse oximetry, BP, heart rate).

Biopsy Technique: Upper GI endoscopy used to obtain 8 biopsies from the D2–D3 portion of the duodenum using standard forceps. Avoidance of actively bleeding or lesioned areas.

Immediate Handling: Biopsies are immediately allocated to pre-labelled specimen containers:

1. Formalin container for histology/FFPE (Priority sample) for ST (4 biopsies)
2. Single-Cell Dissociation of Gut Biopsy Tissue for Immune Cell Analysis (2 biopsies)
3. Cryovials on dry ice/liquid nitrogen for fresh-frozen ST (2 biopsies)

Post-procedure Care**:** Minimum 2-hour observation, discharge instructions, and provision of travel compensation.

10. Sample Processing, Storage and Shipping

All samples will be processed according to respective SOPs

Storage and Tracking:

- Samples stored in **−80°C freezers** with continuous temperature monitoring and alarm systems, backed by generators.
- **Chain-of-Custody:** Electronic sample tracking using LIMS/REDCap with barcoded labels, supplemented by a physical logbook.

Shipping: International transfer requires pre-approved MTAs (Material Transfer Agreements) and NHRA (National Health Research Authority) approvals. Samples are shipped on dry ice in validated shippers.

11. Laboratory Assays and Spatial Transcriptomics Workflow

Platform Selection
Platform selection for spatial transcriptomic analysis will be determined following the pilot assessment of tissue quality and available budget, ensuring that the chosen approach provides adequate spatial resolution and sensitivity for downstream immune profiling. Quality control thresholds established during the pilot phase will guide data inclusion, with minimum criteria such as enough detected genes per capture area, acceptable mapping rates, low mitochondrial gene fractions, and verified tissue integrity through histological review.

Laboratory Procedures
Laboratory procedures will include cryo-sectioning of gut biopsies, histological staining and imaging, spatial capture and library preparation according to the optimized protocol, sequencing, and primary data processing using validated pipelines.

Bioinformatics and Data analysis
Downstream bioinformatics analyses will encompass normalization, spot-level quality control, clustering, cell-type deconvolution based on established reference atlases, and spatial differential expression analysis. Validation of spatial expression patterns will be performed through immunohistochemistry or immunofluorescence on adjacent tissue sections.

Complementary Assays
Complementary assays will include bulk RNA sequencing from snap-frozen matched biopsies, single-cell dissociation of gut biopsy tissue for immune cell analysis, targeted protein validation using immune cell and epithelial markers, quantification of rotavirus-specific antibodies and viral load in plasma and stool, and fecal biomarker profiling for environmental enteric dysfunction.

12. Data Management and Confidentiality

Database: DHIS2 database implementation with role-based access for CRFs and sample logs.
Identifier Management: Participants are assigned a numeric study ID; a linkage file is stored separately under double encryption.
Confidentiality: Pseudonymization and secure storage; all public data release will be de-identified (gene expression matrices) and governed by Data Transfer Agreements or /Materials Transfer Agreements.

13. Statistical Analysis Plan

Histology and Region of Interest Identification: Spatial transcriptomic (ST) data will be analysed following rigorous quality control and region of interest (ROI) identification, guided by hematoxylin and eosin (H&E) histology to ensure accurate mapping of tissue architecture and cellular context.

Feasibility Metrics: Primary feasibility metrics will include the proportion of biopsies meeting defined QC thresholds, the time from biopsy collection to freezing, and the percentage of participants completing all scheduled visits. Descriptive statistics will summarize participant demographics and feasibility outcomes.
Statistical and Spatial Analyses: Temporal changes in gene expression within individuals will be analysed using linear mixed-effects models to account for repeated measures. Spatially aware differential expression analyses will be applied to identify genes and pathways exhibiting spatial regulation within tissue sections.

Correlation and Adjustment for Confounders: Correlation analyses, using Spearman rank tests, will assess relationships between spatial module scores and serologic or stool-based immune outcomes, with multivariate regression models adjusting for potential confounders such as age, sex, and baseline environmental enteric dysfunction biomarkers.

Multiple Testing Correction: Multiple testing correction will be performed using the Benjamini–Hochberg or other false discovery rate (FDR) methods to control for false positives in downstream analyses.

14. Safety Monitoring, Adverse Event Reporting and Management

- AE Reporting: All Adverse Events (AEs) will be documented in the CRF. Serious Adverse Events (SAEs) will be reported to the PI within 24 hours, and to UNZABREC and NHRA per regulatory timelines.
- Data Safety Monitoring: An independent Study Monitoring Committee (SMC), composed of independent experts, will review safety data at pre-defined intervals (after the first 10 procedures, mid-recruitment, and completion) and ad-hoc if SAEs occur.
- Stopping Rules: Immediate suspension and SMC review will be triggered by any procedure-related SAE resulting in death or permanent disability attributable to study procedures.

15. Ethical Considerations

- **Ethics Submissions:** Complete protocol, ICFs, community engagement plan, and SOPs will be submitted to UNZA BREC and NHRA.
- **Informed Consent:** ICFs will use clear language, include optional consent for international shipment or future unspecified research, and detail compensation.
- **Incidental Findings:** Clinically actionable incidental findings will be reviewed by a clinician and communicated to participants, followed by appropriate referral.

16. Quality Assurance and Monitoring

- **Training:** All staff will receive GCP, endoscopy safety, sample handling, and data entry training.
- **SOPs:** Detailed SOPs will be attached for every critical step (Appendices C, D, E).
- **Monitoring:** Routine monitoring visits and corrective action plans for deviations will be performed.

17. Timeline and Milestones

Timeframe	Key Milestones and Deliverables
Months 0–2	Final SOPs, staff training, UNZABREC & NHRA submission and approval.
Month 3	Site readiness, pilot tissue processing run (n=2 donor samples) to test cold chain.
Months 4–9	Recruitment and sample collection (rolling enrolment).
Months 5–10	ST library preparation, sequencing, and preliminary analyses (interim reports).
Month 10–12	Final analyses, report writing, community dissemination, and data deposition.

19. Budget Summary

Category	Cost (ZMW)	Narrative / Justification
Personnel	490,240	Supports key study personnel responsible for project coordination and execution, including proportional effort for: PI (15%), two laboratory technologists (20% each), and partial support for an administrative officer and grants officer (5% each). Ensures efficient implementation, regulatory compliance, and fiscal oversight.
Travel	215,175	Covers round-trip travel, per diem, and accommodation for study staff traveling to the Africa Health Research Institute (AHRI) for protocol harmonization, hands-on laboratory training, and collaborative meetings to strengthen implementation quality.
Other Direct Costs	673,954	Supports procurement of essential study inputs including vaccines, laboratory reagents, sample packaging and shipment, personal protective equipment, cold-chain consumables, and ethics review fees necessary for safe, compliant, and scientifically rigorous research.
Subawards	906,000	Sub-contract to TROPGAN for clinical activities including gut biopsy collection, clinical coordination, on-site personnel support, supplies, and logistics required for high-quality clinical data acquisition aligned with protocol requirements.
Total (ZMW)	2,285,369	

20. Dissemination Policy and Authorship

Publications will follow ICMJE authorship criteria. Processed, de-identified data will be deposited in public repositories after publication, respecting consent and ethics approvals. Local dissemination will occur via community meetings and a policy brief to the Ministry of Health.

21. References

1 Regassa R, Tamiru D, Duguma M, Belachew T. Environmental enteropathy and its association with water sanitation and hygiene in slum areas of Jimma Town Ethiopia. *PLoS One* 2023; **18**: e0286866.

2 Hossain MS, Khodeza Nahar Begum SM, Rahman MM, *et al.* Environmental enteric dysfunction and small intestinal histomorphology of stunted children in Bangladesh. *PLoS Negl Trop Dis* 2023; **17**: e0010472.

3 Naylor C, Lu M, Haque R, *et al.* Environmental Enteropathy, Oral Vaccine Failure and Growth Faltering in Infants in Bangladesh. *EBioMedicine* 2015; **2**. DOI:10.1016/j.ebiom.2015.09.036.

4 Church JA, Rukobo S, Govha M, *et al.* Associations between biomarkers of environmental enteric dysfunction and oral rotavirus vaccine immunogenicity in rural Zimbabwean infants. *EClinicalMedicine* 2021; **41**. DOI:10.1016/j.eclinm.2021.101173.

5 Kang G. Success from the South: the rotavirus vaccine story and its lessons. *The Lancet* 2024; **403**: 111–6.

6 Mowat AM, Agace WW. Regional specialization within the intestinal immune system. *Nature Reviews Immunology 2014 14:10* 2014; **14**: 667–85.

7 Atarashi K, Tanoue T, Shima T, *et al.* Induction of colonic regulatory T cells by indigenous Clostridium species. *Science* 2011; **331**: 337–41.

8 Furusawa Y, Obata Y, Fukuda S, *et al.* Commensal microbe-derived butyrate induces the differentiation of colonic regulatory T cells. *Nature 2013 504:7480* 2013; **504**: 446–50.

9 Tan J, Taitz J, Sun SM, Langford L, Ni D, Macia L. Your Regulatory T Cells Are What You Eat: How Diet and Gut Microbiota Affect Regulatory T Cell Development. *Front Nutr* 2022; **9**. DOI:10.3389/FNUT.2022.878382.

10 Li H, Wu H, Ma W, *et al.* Spatial transcriptomics: a bibliometric analysis with large language model on English literatures. *Brief Bioinform* 2025; **26**. DOI:10.1093/BIB/BBAF553.

11 Marie C, Ali A, Chandwe K, Petri WA, Kelly P. Pathophysiology of environmental enteric dysfunction and its impact on oral vaccine efficacy. *Mucosal Immunol* 2018; **11**: 1290–8.

12 Kummerlowe C, Mwakamui S, Hughes TK, *et al.* Single-cell profiling of environmental enteropathy reveals signatures of epithelial remodeling and immune activation. *Sci Transl Med* 2022; **14**: eabi8633.

13 Bhattacharjee A, Burr AHP, Overacre-Delgoffe AE, *et al.* Environmental enteric dysfunction induces regulatory T cells that inhibit local CD4+ T cell responses and impair oral vaccine efficacy. *Immunity* 2021; **54**: 1745-1757.e7.

14 Gu Y, Bartolomé-Casado R, Xu C, *et al.* Immune microniches shape intestinal Treg function. *Nature 2024 628:8009* 2024; **628**: 854–62.

15 Bein A, Fadel CW, Swenor B, *et al.* Nutritional deficiency in an intestine-on-a-chip recapitulates injury hallmarks associated with environmental enteric dysfunction. *Nat Biomed Eng* 2022; **6**: 1236.

Temporal and Spatial Immune Profiling of Oral Rotavirus Vaccine Responses in Zambian Adults with Environmental Enteropathy

Version: 2.0| Date: 21st January 2026
Sponsor: CIDRZ | Principal Investigator: Mr. Michelo Simuyandi
Study Sites: UTH Endoscopy Unit, St Augustine Clinic, CIDRZ Laboratories, Lusaka.

Introduction

We are researchers from the Centre for Infectious Disease Research in Zambia (CIDRZ), working with doctors and scientists from the Tropical Gastroenterology and Nutrition Group (TROPGAN) at St Augustine Clinic. We would like to invite you to take part in a research study about how the gut responds to an oral vaccine. Participation is voluntary, and if you choose not to take part, your medical care will not be affected.

Why we are doing this research

In Zambia and similar settings, adults often have Environmental Enteropathy (EE), a mild gut inflammation due to repeated infections. This may affect how the body absorbs nutrients and responds to oral vaccines like Rotarix. We aim to understand how the intestinal immune system responds after taking Rotarix by studying gene activity in small tissue samples from the intestine.

What taking part will involve

If you agree to participate, you will be followed for about 28 days and attend up to four clinic visits at St Augustine Clinic. You will undergo screening, vaccination, sample collection (blood, stool, and tissue biopsies), and follow-up visits at either Days 3, 10, or 28.

About the Endoscopy

An endoscopy allows doctors to look at your upper intestine using a thin, flexible camera. It is done under light sedation so that you remain relaxed. Small tissue biopsies will be taken; you will not feel this. Afterward, you will rest for at least two hours and can go home once you are fully awake.

Risks and Discomforts

Endoscopy is generally safe, but there are small risks such as mild throat discomfort, bleeding, or sedation-related drowsiness. You may also experience slight bruising after blood draws. Serious complications are rare, and medical care will be available if needed.

Benefits of Taking Part

You may not directly benefit from this study, but you will receive a free medical check-up and HIV counseling and testing. The information from this study will help improve oral vaccine performance in Zambia and other countries.

Compensation

You will receive K200 per study visit as reimbursement for time and transport. Meals will be provided on endoscopy days, and transport can be arranged if needed.

Confidentiality and Data Protection

All information collected about you will be kept strictly confidential. You will be assigned a study number, and your name will not appear on any laboratory samples or reports. Only authorized study staff will have access to identifiable data.

Storage and Shipment of Samples

Some of your samples will be collected and stored at TROPGAN and CIDRZ in secure, access-controlled freezers for a period of **up to ten (10) years.** The samples may be sent abroad for specialized analysis following approval from the National Health Research Authority (NHRA). Any leftover samples will either be stored for future approved research or destroyed in accordance with institutional and regulatory guidelines.

Voluntary Participation and Right to Withdraw

Participation is voluntary. You may withdraw from the study at any time without giving a reason and without affecting your healthcare.

Contacts for Questions or Concerns

For any questions about this study, please contact:
Mr. Michelo Simuyandi
Centre for Infectious Disease Research in Zambia (CIDRZ)
Stand 378A , Main Street, Ibex,
P.O. Box 34681 Lusaka,
Reception: +260 211 242 257 – 63

For concerns about your rights as a participant, contact:

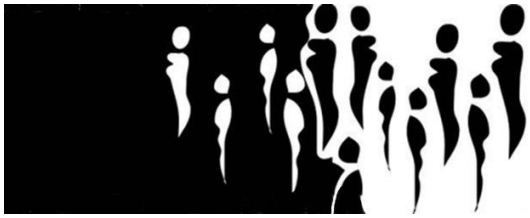
University of Zambia Biomedical Research Ethics Committee
(UNZABREC) Ridgeway Campus, P.O. Box 50110, Lusaka, Zambia
Tel: +260 211 256067
Email: unzarec@unza.zm

Consent Record

Study Title:

Temporal and Spatial Immune Profiling of Oral Rotavirus Vaccine Responses in Zambian Adults with Environmental Enteropathy.

Ethics Committee Reference: _____



Participant's Statement

I have read (or have had read to me) the information above (version 1.0, dated 13th November 2025). I understand what the study involves, and I agree to take part. I understand that I may withdraw at any time without affecting my healthcare.

Participant's Name: _____

Signature: _____Date: _____
Thumbprint: _____Date: _____

Witness (if applicable):_____Date: _____

Investigator's Statement:

I confirm that I have carefully explained the nature and purpose of the study to the participant. I believe that they have understood the information and have freely given informed consent.

Investigator's Name: _____

Signature: _____Date: _____