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aquaya

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Identifying and Interrupting Fecal-Oral Pathways in Young Children:

A QUICK GUIDE TO FORMATIVE RESEARCH, MONITORING, AND EVALUATION

TABLE OF CONTENTS

FIGURES

TABLES

LIST OF ACRONYMS

CBT – Compartment Bag Test
EED – Environmental Enteric Dysfunction
IP – Implementing Partner
IPTT – Indicator Performance Tracking Table
IRB – Institutional Review Board
IYC – Infants and Young Children
LMIC – Low and Middle Income Country
MPN – Most Probable Number
qPCR – Quantitative Polymerase Chain
U2 – under the age of two
WASHPaLS – Water, Sanitation, and Hygiene Partnerships and Learning for Sustainability

PROBLEM

Malnutrition is a factor in 45% of deaths of children under five, and Africa experiences the second-highest rate of acute malnutrition. In 2019, 21 percent or 144 million of the world's children under age five globally experienced stunting, with the vast majority living in low- and middle-income countries (LMIC).[1] Current research has linked stunting in the first 1,000 days of life to increased susceptibility to infections, lower economic productivity in adulthood, cognitive deficits, and diminished educational attainment, as well as increased mortality.[2] Parallel exposure to inadequate diet and poor water, poor care practices, and repeated infection caused by inadequate sanitation and hygiene conditions put infants and young children (IYC) at greater risk of stunting and undernutrition in the earliest stages of life.[3]

Enteric infections have also been linked to persistent acute malnutrition. Chronic exposure to enteric pathogens can contribute to acute malnutrition by reducing the ability of the gut to absorb nutrients. Malnutrition in turn can worsen frequency and severity of diarrhea.[4] Prior work suggests that the first two years of life are vital to long-term child nutrition, cognitive development, and later health and educational outcomes.

Global evidence indicates that while the overall childhood mortality rate attributable to diarrheal disease is decreasing, there is little effect on the total number of episodes, pointing towards other potential pathways towards stunting beyond diarrhea.[5] Over the past decade, environmental enteric dysfunction (EED), a condition characterized by inflammation of the small intestinal lining that inhibits permeability and nutrient absorption, has been identified as a potential major mediating pathway linking poor WASH conditions and chronic undernutrition.[6] Evidence indicates that assaults on the gut, linked with ingestion by IYC of animal and human feces, changes the shape and function of the gut, weakening ability to absorb nutrients, and making IYC more susceptible to enteric infection. EED is also thought to explain why current nutrition interventions, implemented in isolation from others focused on preventing child exposure to feces, only have modest effects on stunting.

For the past several decades, potential pathways of fecal pathogen transmission have been understood through the framework of the "F-diagram" which linked ingestion of contaminated fluids, fingers, flies, fields (floors, earth, and dirt), fomites (surfaces), and food with sub-standard IYC growth and development outcomes. However, the traditional F-diagram overlooks other risk factors, such as exposure to animal feces, often facing children in developing contexts. Infants and children living in low and middle income countries (LMICs) often live closely with their animals and sometimes share sleeping quarters, which can increase risk of contamination through multiple transmission pathways inside of homes. Children's behaviors, such as eating dirt (geophagy) or human/animal feces through mouthing behaviors, are often ignored when considering risk factors associated and potential

1 WHO. Available online at: https://www.who.int/gho/child-malnutrition/en/

2 De Sanctis V, Soliman A, Alaaraj N, Ahmed S, Alyafei F, Hamed N. Early and Long-term Consequences of Nutritional Stunting: From Childhood to Adulthood. Acta Biomed. 2021 Feb 16;92(1):e2021168. doi: 10.23750/abm.v92i1.11346. PMID: 33682846; PMCID: PMC7975963

3 Wolf J., Prüss-Ustün A., Cumming O., Bartram J., Bonjour S., Cairncross S., Clasen T., Colford J.M., Jr., Curtis V., De France J., et al. Systematic review: Assessing the impact of drinking water and sanitation on diarrhoeal disease in low-and middle-income settings: Systematic review and meta-regression. Trop. Med. Int. Health. 2014;19:928–942. doi: 10.1111/tmi.12331

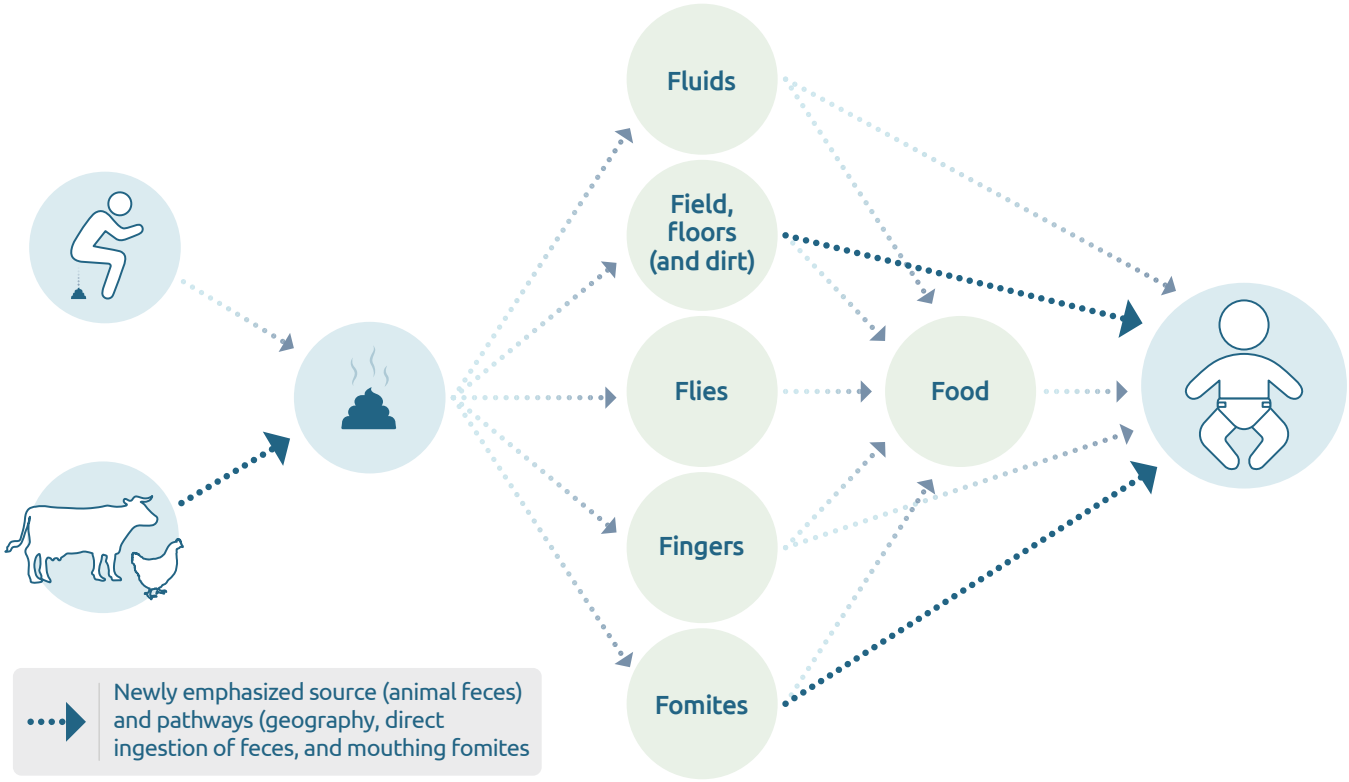
4 Jones KD, Thitiri J, Ngari M, Berkley JA. Childhood Malnutrition: Toward an Understanding of Infections, Inflammation, and Antimicrobials. Food and Nutrition Bulletin. 2014;35(2_suppl1):S64-S70. doi:10.1177/15648265140352S110

5 Black R, Fontaine O, Lamberti L, Bhan M, Huicho L, El Arifeen S, Masanja H, Walker CF, Mengestu TK, Pearson L, Young M, Orobaton N, Chu Y, Jackson B, Bateman M, Walker N, Merson M. Drivers of the reduction in childhood diarrhea mortality 1980-2015 and interventions to eliminate preventable diarrhea deaths by 2030. J Glob Health. 2019 Dec;9(2):020801. doi: 10.7189/jogh.09.020801. PMID: 31673345; PMCID: PMC6815873.

6 Budge S, Parker AH, Hutchings PT, Garbutt C. Environmental enteric dysfunction and child stunting. Nutr Rev. 2019 Apr 1;77(4):240-253. doi: 10.1093/nutrit/nuy068. PMID: 30753710; PMCID: PMC6394759.

interventions. Young children are also often given contaminated water, and food that is not properly reheated or fed using dirty utensils or caregiver's hands. To account for these additional pathways, the USAID/WASH PaLS project published a modified F-diagram in 2018, shown in Figure 1:

FIGURE 1: MODIFIED F-DIAGRAM THAT HIGHLIGHTS GEOPHAGY AND DIRECT FECES INGESTION BY INFANTS AND YOUNG CHILDREN[7]



Few available studies studies (listed in Table 1) have focused on observing children's behavior, and most of them focused on mouthing behaviors only.

TABLE 1: PREVIOUS STUDIES EXAMINING BEHAVIORS AMONG CHILDREN UNDER THE AGE OF TWO IN LOW AND MIDDLE INCOME COUNTRIES

COUNTRY	OBSERVATIONS	AUTHORS
Zimbabwe	Formative Research on Hygiene Behaviors and Geophagy among Infants and Young Children and Implications of Exposure to Fecal Bacteria	Ngure et al., 2013
Ghana	Characterization and quantification of children's (0-5 years old) behavior, including environment (household/nursery), compartment and activity	Teunis et al., 2016
Bangladesh	Quantification of hand- and object-mouthing frequency of children aged 3-18 months	Kwong et al., 2016
DRC	Assessment of children's (0-5 years old) mouthing behaviors	George et al., 2021

7 USAID, 2018. Toward a Hygienic Environment for Infants and Young Children: A Review of the Literature. Washington, D.C., USAID Water, Sanitation, and Hygiene Partnerships and Sustainability (WASHPaLS) Project

Understanding zoonotic (transmission from animals to humans) transmission is critical because without rigorous assessment it remains difficult to implement intervention strategies to prevent the spread of enteropathogens (organisms that causes disease of the intestinal tract). Interventions specifically targeting human-animal interaction, such as use of chicken corralling and improved floor material (enabling washing of poultry feces from inside the household), may be more effective strategies to reduce child exposure to enteric infections and reduce child stunting.[8]

PURPOSE OF THE QUICK GUIDE

This quick guide presents planning considerations and a menu of data collection methods to help BHA Resilience Food Security Activity implementing partners meet three objectives:

1. Determine whether your program needs to conduct mixed-method formative research to identify and prioritize fecal-oral pathways for infants and young children in your implementation geography, or if you can rely on existing evidence.
2. Provide guidance on how to design formative research activities to identify and prioritize the ingestion pathways of fecal pathogens (both human and animal) for IYC. This objective sets the stage for prioritizing the pathways and potential interventions implementing partners will deploy during their award life.
3. Regardless of whether your program may need to conduct formative research on this topic, this quick guide provides useful guidance on which data collection methods may be appropriate to incorporate in a program Indicator Performance Tracking Table (IPTT) to measure the effectiveness of WASH interventions on interrupting targeted fecal-oral contamination pathways in children under the age of 2 years old.

At the outset of program design for nutrition-sensitive WASH activities, PRO-WASH encourages BHA Implementing Partners (IPs) to conduct the following assessment steps in sequence:

STEP 1 Review existing global, national, and sub-national research to determine the type of knowledge available and remaining information gaps.

PRO-WASH encourages IPs to first conduct a desk mapping of existing global, national, and sub-national research to assess the state of evidence and to identify relevant research on the context of interest. This exercise will help determine whether undertaking formative research in your context is necessary and which questions you need to answer. Recent global literature reviews on this topic have been completed and can serve as a starting point for IPs in completing a review of existing evidence. These documents, along with supplemental evidence published afterwards, are listed in Table 2. However, research in this space is dynamic and partners should confirm whether more recently completed research has been conducted. If your team determines that sufficient evidence exists to begin designing and testing solutions, skip to Step 3.

TABLE 2: EXISTING EVIDENCE ON FECAL-ORAL PATHWAYS FOR CHILDREN UNDER THE AGE OF TWO (AS OF NOV. 2022)

PUBLICATION YEAR	RESOURCE
2018	**Toward a Hygienic Environment for Infants and Young Children: A Review of the Literature.** Washington, D.C., USAID Water, Sanitation, and Hygiene Partnerships and Learning for Sustainability (WASHPaLS) Project.
2022	**Toward a hygienic environment for infants and young children: Limiting early exposures to support long-term health and well-being.** Washington, D.C., USAID Water, Sanitation, and Hygiene Partnerships and Learning for Sustainability (WASHPaLS) Project.
2022	Innovations for Poverty Action–led research study underway: Pathogen Pathways Study for Children Under Two Years in Turkana South and Samburu North Sub-counties of Kenya.
2022	Aquaya-led research study underway: Pathogen Pathways Study for Children Under Two Years in Southeastern Madagascar.

8 Pickering, A. J. et al. The WASH Benefits and SHINE trials: interpretation of WASH intervention effects on linear growth and diarrhoea. Lancet Glob Health 7, e1139–e1146 (2019).

STEP 2 Design and conduct formative research (quantitative and qualitative) as needed to identify and prioritize fecal-oral contamination pathways for infants and young children

If insufficient context-specific evidence exists to help your team identify and prioritize the fecal-oral contamination pathways for children under the age of two (U2), consider designing a mixed methods formative research activity using **Annex A: A Menu of Data Collection Methods** to inform program strategies and activities. PRO-WASH has also included links to study protocols and tools from two recently completed (2022) pathogen pathway studies in **Kenya** and Madagascar for reference. When selecting the appropriate combination of methods for formative research, keep in mind the following considerations:

a. **Evidence availability and gaps:** What information does your project lack in order to identify and prioritize fecal-oral contamination pathways and/or develop effective and appropriate solutions to test and scale (should be informed by Step 1)?

b. **Funding:** Does your project have enough funding to dedicate to a pathogen transmission pathways study? A full mixed methods study using all the data collection methods is outlined in **Annex A**. If all the methods described in **Annex A** were applied within a formative research activity, the cost would likely be US$250,000-350,000. Costs can be reduced if IPs (and any research partners) determine that not all methods are necessary to answer the identified learning question(s).

c. **Time:** In order to conduct a full mixed methods research study like those listed under Table 1, IPs should plan on a timeline of 12-20 months between identification of research partner(s) and obtaining results for use in program design. PRO-WASH estimates the following timeline for specific study phases outlined in **Table 3**:

TABLE 3: STUDY ACTIVITIES AND ESTIMATED TIMELINES

ACTIVITY AND NOTES	TIMEFRAME
Development of study concept note (or RFA) and recruitment of Principle Investigators and co-investigators.	2-3 months
Protocol and tool development and IRB approvals. International Institutional Review Board (IRB) approvals can typically be obtained within a few months of submission. However, local IRBs are highly variable. PRO-WASH recommends consulting with local stakeholders early to determine the appropriate IRB authority for this type of research and to establish anticipated timelines and processes for approvals.	2-5 months
Data collection.	2-3 months
Shipping lab samples internationally and completing data analysis (if applicable) *[note: PRO-WASH strongly recommends exhausting all local options before considering international sample shipment].*	3-4 months
Data analysis.	2-3 months
Summarization of findings/publications.	1-2 months

d. **Access to laboratory equipment:** For any selected laboratory analyses, IPs and their research partners should conduct extensive scoping activities to determine the capacity of local laboratories and the availability of those laboratory staff and equipment. Any consumable items needed for analysis should be ordered prior to starting data collection activities to avoid delays in data analysis. PRO-WASH also recommends relying on domestic supply purchases to the extent possible to avoid potential delays associated with importation and customs.

e. **International shipment of biological samples and/or molecular laboratory supplies:** PRO-WASH strongly recommends close examination of the requirements, processes, and timelines associated with the following logistical considerations in order to determine whether domestic or international laboratory analysis is most practical. Specific considerations are outlined in **Table 4**:

TABLE 4: CONSIDERATIONS FOR DETERMINING LOCATION OF LABORATORY ANALYSIS (IF APPLICABLE)

LOCATION OF LAB ANALYSIS	METHODOLOGY, TIMELINE, AND OPERATIONAL CONSIDERATIONS
Domestic	Importation of molecular laboratory supplies, which in certain contexts can take months to clear customs and be ready for use.
International	Local and international ethics and permit requirements for export and import are understood and regularly reviewed to maintain an up-to-date understanding of requirements. In certain countries, requirements and processes are ever-changing. There are also increasingly complicated permit requirements across countries for exporting and importing environmental and biological samples.
	Early and extensive scoping activities to identify primary and secondary dry ice courier services and verify a common understanding of all import and export requirements and approvals.

f. **Consider complementary research/monitoring efforts:** PRO-WASH recognizes that research and learning agendas among BHA RFSA IPs are robust. IPs should consider how other research and learning activities may enhance the findings from any planned WASH/nutrition activities (and vice versa). PRO-WASH also recognizes that other relevant WASH, nutrition, and health data is often collected by IPs through standard BHA indicators as relevant to each project and can be used to supplement methods outlined in this quick-guide to provide a more complete understanding of children's environmental risks and potential solutions.

STEP 3 Incorporate quantitative, qualitative, and/or laboratory methods within your M&E plan to reliably measure progress and evaluate intervention activities

While many of the data collection methods outlined in **Annex A** could ideally be used to monitor and evaluate the effectiveness of nutrition-sensitive WASH interventions, some can also be time and resource intensive. Therefore, in Table 5 PRO-WASH has outlined methods that should be considered as routine monitoring and/or evaluation methods for BHA IPs. Recommended indicators for monitoring and evaluation within the Table 5 are based on PRO-WASH's assessment of cost and feasibility.

Annex A includes illustrative monitoring indicators that IPs may consider if planning to incorporate the relevant method into program M&E plans – though many of these indicators will require further contextualization. IPs should only include suggested indicators within M&E plans if the indicators are directly related to RFSA activities and fulfill one or more of the following functions:

- To assess if progress is made in achieving expected results
- To identify bottlenecks/challenges in implementation
- To highlight whether there are any unintended effects (positive or negative) from a particular RFSA activity or strategy

PRO-WASH recognizes that IPs are implementing RFSAs across a wide variety of contexts and use a variety of intervention packages. When using this quick-guide to inform M&E plans, IPs are encouraged to consider the circumstances under which attribution or causal pathways can be identified to ensure that data collected can be used to evaluate specific activities or strategies. Where possible, PRO-WASH also encourages RFSA teams to suggest inclusion of these indicators within baseline, midline, and endline data collection protocols.

Project teams can also refer to the protocols and tools used for recent pathogen pathways research studies in **Kenya** and Madagascar to inform method and tool development.

TABLE 5: SUGGESTED DATA COLLECTION METHODS FOR MONITORING AND EVALUATION ACTIVITIES

DATA COLLECTION METHODS	RELEVANT FORMATIVE RESEARCH METHOD	RELEVANT MONITORING & EVALUATION METHOD
Quantitative Survey Methods		
Cross-sectional household environmental and WASH service observations	Yes	Yes
Qualitative Methods		
Caregiver Interviews (15-45 min)	Yes	Yes
Observational Methods		
Household spot observations (5-15 min)	Yes	Yes
Structured observations of children (1-4 hours)	Yes	No
Environmental and Feces Sampling for Laboratory Analysis		
Enumerating E.coli from Environmental compartments only*	Yes	Yes
Enteric pathogen profiles in soil, food, drinking water, hand rinse, animal fecal samples, and infant/ child fecal samples	Yes	Evaluation only

*Environmental sampling should take place from compartments (sample types) listed in **Annex B**.

In addition to these data collection methods, it is assumed that BHA IPs are collecting child nutritional status data (e.g. wasting and stunting prevalence) at baseline, endline, and potentially, during routine monitoring to complement the data collected through methods listed in this quick-guide.

As noted under Step 2, PRO-WASH recognizes that BHA IP research and learning agendas are robust. IPs should consider how other project-led monitoring and evaluation activities may enhance the findings from any planned WASH/nutrition activities (and vice versa). For example, it would be ideal to enroll children with project-collected anthropometric data into any nutrition-sensitive M&E activities in order to layer anthropometric data, pathogen pathway data, and intervention data to better assess over time how health outcomes change among specific children who received WASH interventions. PRO-WASH also recognizes that other relevant WASH, nutrition, and health data should be collected by IPs through standard BHA indicators as relevant to each project.

ANNEX A:
Menu of Data Collection Methods for Nutrition-sensitive WASH Formative Research, Monitoring, and Evaluation Activities

There are various data collection methods that provide different, yet complimentary information to inform program design and monitoring and evaluation frameworks. Ideally, a combination of the methods described below would be used in formative research, monitoring, and evaluation activities as part of BHA-funded programs to accurately identify and prioritize fecal-oral contamination pathways for U2 children, while also monitoring and evaluating the effectiveness of any interventions meant to disrupt prioritized pathways. Note that implementing multiple methods together may be most efficient (e.g., caregiver surveys followed immediately by household observations and collection of environmental samples), as the same households can provide data of various types, enabling some level of triangulation and complementarity.

TABLE 6: DATA COLLECTION METHOD 1

DATA COLLECTION METHOD 1: CROSS-SECTIONAL HOUSEHOLD ENVIRONMENTAL AND WASH SERVICE OBSERVATIONS	
METHOD DESCRIPTION	**STRUCTURED QUANTITATIVE SURVEY**
GUIDANCE OF FORMATIVE RESEARCH ACTIVITIES	
Method purpose during formative research	Assess a variety of environmental conditions at one point in time to understand child living conditions and potential environmental risks (e.g. household WASH service level and type, housing and environmental conditions, prevalence and type of animals, etc.)
Sample size guidance	Variable based on context and resources available General Guidance: *Minimum:* 50-100 households *Ideal:* representative sample of study population
Data analysis timeline required	2-3 weeks
Timeline, staffing, and budget assumptions	3-4 surveys per enumerator/day
Technical expertise, facilities, or equipment required	• Can be conducted by trained enumerators (project staff or research specialists) • Limited subject matter expertise required (can be provided in training)
Advantages	• Provides a snapshot of environmental conditions in which children reside and variations among households, communities and geographies (depending on sample size) and potential fecal-oral risks • If implemented routinely (annually, seasonally, post-shock), this method provides insight on the variation in WASH service access and environmental conditions • Low cost • Relatively easy to implement using trained enumerators or program staff • Likely streamlined IRB approvals due to non-human subjects research or minimal-risk IRB determinations

DATA COLLECTION METHOD 1: CROSS-SECTIONAL HOUSEHOLD ENVIRONMENTAL AND WASH SERVICE OBSERVATIONS	
METHOD DESCRIPTION	**STRUCTURED QUANTITATIVE SURVEY**
Disadvantages/limitations	• Unable to identify specific fecal-oral pathways or prevalent pathogens
GUIDANCE FOR ROUTINE MONITORING AND EVALUATION ACTIVITIES	
Method is appropriate for routine monitoring	Yes
Method purpose during monitoring and evaluation of program activities	Monitor changes in household and community environmental conditions and WASH access over time, seasons, and after significant environmental changes or shocks (dependent on frequency and timing of data collection)
Sample size guidance	Sample size will vary by project size, scope, and context. Refer to BHA guidance on Monitoring, Evaluation, and Reporting for sample size calculations within for data collection based on the indicators being measured and the sampling frame
Suggested data collection frequency (for routine monitoring)	• Baseline (accounting for seasonal variations) • Annually or at midline as time, staffing resources allow (accounting for seasonal variations) • Endline (accounting for seasonal variations)
Illustrative M&E indicator(s)	• % of households with at least a basic water service (refer to BHA indicator PIRS) • % of households with at least a basic sanitation service (refer to BHA indicator PIRS) • % of households with a basic hygiene service (refer to BHA indicator PIRS) • % of households with improved (impermeable, cleanable) flooring • % of household covered by improved flooring (e.g. certain rooms, entire indoor space, etc.) • % change in household compliance with improved/safer animal management practices promoted by the project (e.g. corralling of livestock) • % change in household compliance with any project-promoted solutions for separating children from animals, soil, or other contaminants (e.g. limiting domestic animal access to household interior) • % of households with domestic animals (dogs, cats, livestock such as poultry, cows, or pigs)

TABLE 7: DATA COLLECTION METHOD 2

DATA COLLECTION METHOD 2: CAREGIVER INTERVIEWS	
METHOD DESCRIPTION	**QUANTITATIVE STRUCTURED QUANTITATIVE SURVEYS WITH CAREGIVERS**
GUIDANCE OF FORMATIVE RESEARCH ACTIVITIES	
Method purpose during formative research	Provides caregiver-reported insight into common household and community WASH, nutrition, and childcare practices that may influence the specific fecal-oral contamination risks for children
Sample size guidance	*Minimum:* Variable based on what is being measured, variation in that outcome, and diversity of study population *Ideal:* representative sample of study population
Data analysis timeline required	2-3 weeks
Timeline, staffing, and budget assumptions	3-6 surveys per enumerator/day depending on the remoteness of the villages and the length of the survey
Technical expertise, facilities, or equipment required	• Can be conducted by trained enumerators (project staff or research specialists) • Limited subject matter expertise required (can be provided in training) • Phone or tablet to record answers (plus data collection app)
Advantages	• Provides in-depth information on caregiver practices, perspectives, challenges, as well as variations between households/geographies (depending on sample size). Also provides contextualized insights into social and cultural norms and taboos
Disadvantages/limitations	• Data is self-reported and not verifiable (though observational data can provide some triangulation) • Likely requires higher level IRB approvals • Lack of statistical representation of target population
GUIDANCE FOR ROUTINE MONITORING AND EVALUATION ACTIVITIES	
Method is appropriate for routine monitoring	Yes
Method purpose during monitoring and evaluation of program activities	Provides insight into changes in household and community WASH, nutrition, and childcare practices
Sample size guidance	Sample size will vary by project size, scope and context. Refer to BHA guidance on Monitoring, Evaluation and Reporting for sample size calculations within for data collection based on the indicators being measured and the sampling frame
Suggested data collection frequency (for routine monitoring)	• Baseline (accounting for seasonal variations) • Annually or at midline as time, staffing resources allow (accounting for seasonal variations) • Endline (accounting for seasonal variations)
Illustrative M&E indicator(s)	• Variable depending on content of interview guide and IP priorities

TABLE 8: DATA COLLECTION METHOD 3

DATA COLLECTION METHOD 3: HOUSEHOLD SPOT OBSERVATIONS (5-15 MIN)	
METHOD DESCRIPTION	**RAPID HOUSEHOLD OBSERVATIONS**
GUIDANCE OF FORMATIVE RESEARCH ACTIVITIES	
Method purpose during monitoring and evaluation of program activities	Provides a quick view of environmental conditions to understand child living conditions and variations among households, communities and geographies
Sample size guidance	*Minimum:* Variable based on what is being measured, variation in that outcome and diversity of study population *Ideal:* representative sample of study population
Data analysis timeline required	• 2-3 weeks if combined with caregiver interview • 15 minutes per one spot observation
Timeline, staffing, and budget assumptions	• 3-5 household spot observations per day per enumerator (if combined with caregiver interview) • 5-8 spot observations per day per enumerator (if not combined with anything else)
Technical expertise, facilities, or equipment required	• Can be conducted by trained enumerators (project staff or research specialists) • Phone or tablet to record observations (plus data collection app)
Advantages	• Provides some detailed, verifiable data on child environment, access to WASH infrastructures, and housing • Easy and quick to implement
Disadvantages/limitations	Does not provide in-depth information on caregiver practices or on child behavior
GUIDANCE FOR ROUTINE MONITORING AND EVALUATION ACTIVITIES	
Method is appropriate for routine monitoring	Yes
Method purpose during monitoring and evaluation of program activities	Provides insight into changes in household and community WASH, nutrition, and childcare practices
Sample size guidance	Sample size will vary by project size, scope, and context. Refer to BHA guidance on Monitoring, Evaluation, and Reporting for sample size calculations within for data collection based on the indicators being measured and the sampling frame
Suggested data collection frequency (for routine monitoring)	• Baseline (accounting for seasonal variations) • Annually or at midline as time, staffing resources allow (accounting for seasonal variations) • Endline (accounting for seasonal variations)
Illustrative M&E indicator(s)	• % caregivers complying with project promoted behaviors, including (but not limited to): » drinking water storage and treatment » use of basic sanitation facilities » handwashing with soap at promoted times » safe animal management practices » animal/children separation solutions » safe food hygiene practices

TABLE 9: DATA COLLECTION METHOD 4

DATA COLLECTION METHOD 4: IN-DEPTH, STRUCTURED OBSERVATIONS OF CHILDREN (3-6 HOURS)	
METHOD DESCRIPTION	**QUANTITATIVE OBSERVATIONS TO CHARACTERIZE CHILD BEHAVIOR AND CHILD INTERACTIONS WITH ITS ENVIRONMENT**
GUIDANCE OF FORMATIVE RESEARCH ACTIVITIES	
Method purpose during formative research	• Identify specific fecal-oral pathways for each child observed • Capture information on hand-to-object contacts and record every object-to-mouth and hand-to-mouth contact of the target child • Record information on each behavior, defined as hand or mouth contact with one or more of the following: own or mother's hand/skin, soil, feces, animals, cloth, toys, and food, including food waste • Frequency of fecal-oral exposures by type for each child observed • Record child-specific information, such as mobility and sleeping times, as well as caregiver practices (e.g. food preparation, cleaning, where child spends time, etc.) • Attempt to account for variations in time of day (capture meal time, sleep time, awake time)
Sample size guidance	Variable based on context and resources available. General Guidance: *Minimum:* 15-20 for each age group (may be highly dependent on available time and resources due to time intensity) *Ideal: r*epresentative sample of study population
Suggested data collection frequency (for routine monitoring)	• Baseline (accounting for seasonal variations) • Annually or at midline as time, staffing resources allow (accounting for seasonal variations) • Endline (accounting for seasonal variations)
Data analysis timeline required	• 2-3 weeks of data analysis • One hour per observation to process written data, if not collected electronically
Timeline, staffing, and budget assumptions	• One child observed per day per enumerator
Technical expertise, facilities, or equipment required	• Can be conducted by trained enumerators (project staff or research specialists) • Limited subject matter expertise required (can be provided in training) • Phone or tablet to record observations (plus data collection app) • To reduce data collection quality concerns, it is suggested to: » Pair enumerators (at least during piloting/training) and ask them to conduct structured observations in parallel and compare data to understand discrepancies. » Spot check of 10-15% of total sample size to ensure enumerator compliance is advised
Advantages	• Provides detailed, verifiable data on caregiver and child behaviors, presence of specific fecal-oral contamination risks and frequency of fecal-oral exposures • Minimizes, recall and information biases because the biological samples analysis provides additional quantitative results on the fecal-oral exposures

DATA COLLECTION METHOD 4: IN-DEPTH, STRUCTURED OBSERVATIONS OF CHILDREN (3-6 HOURS)	
METHOD DESCRIPTION	**QUANTITATIVE OBSERVATIONS TO CHARACTERIZE CHILD BEHAVIOR AND CHILD INTERACTIONS WITH ITS ENVIRONMENT**
Disadvantages/limitations	• Data collection and analysis is time intensive, often resulting in lower sample sizes and lower study • Likely requires IRB approvals (though likely a minimal risk determination) • Statistical representation of study population is not feasible • Requires qualitative interview skills and subject matter expertise • May result in higher "opt-out" rates among potential participating caregivers • Operational planning to plan observations to account for variation in daily routines or time of day and to meet children where they are (home, agricultural fields, etc)
GUIDANCE FOR ROUTINE MONITORING AND EVALUATION ACTIVITIES	
Method is appropriate for routine monitoring	Not recommended due to high cost in time, resources, funding

TABLEAU 10: MÉTHODE DE COLLECTE DES DONNÉES 5

DATA COLLECTION METHOD 4: IN-DEPTH, STRUCTURED OBSERVATIONS OF CHILDREN (3-6 HOURS)	
METHOD DESCRIPTION	**LABORATORY TESTING IDEXX MOST PROBABLE NUMBER (MPN) METHOD WITH COLILERT-18 MEDIA TO DETECT E. COLI AND FECAL COLIFORM OR COMPARTMENT BAG TEST (CBT) METHOD FOR LOW RESOURCE SETTINGS**
ORIENTATIONS POUR LES ACTIVITÉS DE RECHERCHE FORMATIVE	
Method purpose during formative research	Using *E. coli* as a proxy indicator, describe fecal contamination levels across different exposure pathways
Sample size guidance	Variable based on context and resources available. General Guidance: *Minimum:* sampling a minimum of 100 households *Ideal:* representative sample of study population
Data analysis timeline required	• Laboratory analysis: same duration as the sample collection (samples have to be processed a few hours after collection, and then read 24-48 hours after processing and incubation) • 2-3 weeks of data analysis
Timeline, staffing and budget assumptions	• CBT method: US$760 for 100 kits (plus incubator US$500-2,000) • IDEXX: US$700for 100 Kits (plus incubator US$500-2,000+ sealer US$5,000-7,000) • One lab analyst for 20-40 samples per day (though dependent on pre-processing steps for various sample types)
Technical expertise, facilities, or equipment required	• Can be conducted by trained enumerators (project staff or research specialists) • Lab analyst required
Advantages	• Provides information on fecal contamination in different environmental compartments • Laboratory testing can be done in most LMIC settings
Disadvantages/limitations	• Data collection and analysis is time intensive, often resulting in lower sample sizes and lower study • Sample size may be more limited by availability of lab/incubator space, experienced lab personnel, and budget for consumables • Likely requires higher level IRB approvals • Lack of statistical representation of target population • Requires laboratory skills and subject matter expertise • For remote areas, need to set up a field lab
GUIDANCE FOR ROUTINE MONITORING AND EVALUATION ACTIVITIES	
Method is appropriate for routine monitoring	Yes, but expensive. Consider sampling drinking water and food only for routine monitoring
Method purpose during monitoring and evaluation of program activities	• Using *E. coli* as a proxy indicator, assess whether promoted environmental and behavioral interventions result in lower prevalence and/or quantity of *E. coli* exposure for children through prioritized pathways • Using *E. coli* as a proxy indicator, compare the contribution of different exposure pathways to negative health outcomes. *(note: in order to make this comparison, health data for outcomes of interest, e.g. diarrhea, must be collected among sampled children and with sufficient statistical power to make comparative statements)*

DATA COLLECTION METHOD 4: IN-DEPTH, STRUCTURED OBSERVATIONS OF CHILDREN (3-6 HOURS)	
METHOD DESCRIPTION	**LABORATORY TESTING IDEXX MOST PROBABLE NUMBER (MPN) METHOD WITH COLILERT-18 MEDIA TO DETECT E. COLI AND FECAL COLIFORM OR COMPARTMENT BAG TEST (CBT) METHOD FOR LOW RESOURCE SETTINGS**
Sample size guidance	Sample size will vary by project size, scope and context. Refer to BHA guidance on Monitoring, Evaluation, and Reporting for sample size calculations within for data collection based on the indicators being measured and the sampling frame
Suggested data collection frequency (for routine monitoring)	• Seasonally • Before/after shock
Illustrative M&E indicator(s)	• Average concentration of E.coli in collected samples (disaggregated by compartment type)

TABLE 11: DATA COLLECTION METHOD 6

DATA COLLECTION METHOD 6: ENTERIC PATHOGEN PROFILES IN SOIL, FOOD, HANDS, DRINKING WATER, AND FECAL SAMPLES	
METHOD DESCRIPTION	**QUANTITATIVE POLYMERASE CHAIN REACTION (QPCR)**
GUIDANCE FOR FORMATIVE RESEARCH ACTIVITIES	
Method purpose during formative research	Assess enteric pathogen presence and measure concentration to characterize the health risks with different transmission pathways
Sample size guidance	Variable based on context and resources available. General Guidance: *Minimum:* 50 households *Ideal:* representative sample of study population
Data analysis timeline required	• 6-8 weeks for laboratory analysis of all samples (each sample must also be processed for storage within a few hours of collection) • 2-3 weeks for data analysis
Timeline, staffing, and budget assumptions	• TaqMan Array Card method (detection of multiple pathogens at once): approx. US$200-500/sample for a selection of dozens of pathogens • Standard qPCR: US$20-50/sample per pathogen • For international shipment on dry ice: US$5,000-10,000
Technical expertise, facilities, or equipment required	• Can be sampled by trained enumerators (project staff or research specialists) • Limited subject matter expertise required for sampling (can be provided in training) • Analyses need to be done in a laboratory with expertise conducting qPCR
Advantages	• Provides information on the presence and concentration of different pathogens in different environmental compartments
Disadvantages/limitations	• Data collection and analysis is time intensive, often resulting in lower sample sizes and lower study • Likely requires higher level IRB approvals (especially when including human fecal samples) • Lack of statistical representation of target population • Requires laboratory with the capacity of conducting qPCR • Storage and transport: samples have to be frozen after collection and stay frozen until analysis • A continuous supply of electricity is required to process and store samples
GUIDANCE FOR ROUTINE MONITORING AND EVALUATION ACTIVITIES	
Method is appropriate for routine monitoring	Yes, but recommended for evaluation only (due to the high cost of analysis and intensive logistics)
Method purpose during monitoring and evaluation of program activities	• Monitor and assess impact of program or intervention on pathogen burden in young children, or in environmental samples

DATA COLLECTION METHOD 6: ENTERIC PATHOGEN PROFILES IN SOIL, FOOD, HANDS, DRINKING WATER, AND FECAL SAMPLES	
METHOD DESCRIPTION	**QUANTITATIVE POLYMERASE CHAIN REACTION (QPCR)**
Sample Size guidance	Sample size will vary by project size, scope, and context. Refer to BHA guidance on Monitoring, Evaluation, and Reporting for sample size calculations within for data collection based on the indicators being measured and the sampling frame
Suggested data collection frequency (for routine monitoring)	• Baseline • Endline • If the budget allows for it, consider sampling seasonally (rainy versus dry season) or sampling before/after shock
Illustrative M&E indicator(s)	• Average pathogen load identified in environmental and stool samples (disaggregated by compartment type) (note: combine with any available anthropometric data available for same individuals) • Number of pathogens detected per sample (and average number per sample among sampled households) • Prevalence of specific pathogens of interest by pathway

ANNEX B:
Suggested Compartments (sample types) to Consider for Environmental Sampling as Contextually Relevant

DATA COLLECTION COMPARTMENTS	*E. COLI* SAMPLING	ENTERIC PATHOGEN SAMPLING	DATA COLLECTION ANALYSIS CONSIDERATIONS
Child (and potentially caregiver) stool sampling	No	Yes	Depending on the cultural context, organizing community (or influential leaders) meetings to explain the purpose of the study and ask for volunteers might increase the number of participants
Animal feces sampling	No	Yes	If recently excreted animal feces are present within the house or compound, prioritize those for collection
Drinking water samples	Yes	Yes	Collection at Point of Use, but consider collecting at Point of Collection as well to assess the level of recontamination within the house
Child and caregiver hand rinse samples	Yes	Yes	*Consider rinsing both hands with equal timings (e.g if 15 seconds for the left, also put 15 seconds for the right hand). Sterility of the water is key to avoid cross contamination*
Soil samples	Yes	Yes	Should be representative of the area where children are playing
Food samples	Yes	Yes	Collection of recently prepared food and stored food to assess the level of contamination during storage
Infant formula	Yes	No	
Toys or other fomites	Yes	No	Objects may be formal toys or other objects the child most commonly plays with or places in mouth
Bottle-top rinses (if applicable)	Yes	No	

ANNEX C:
Compendium of Relevant Resources & Publications

1. Budge S, Parker AH, Hutchings PT, Garbutt C. Environmental enteric dysfunction and child stunting. Nutr Rev. 2019 Apr 1;77(4):240-253. doi: 10.1093/nutrit/nuy068. PMID: 30753710; PMCID: PMC6394759.
2. Robb K, Null C, Teunis P, Yakubu H, Armah G, Moe CL (2017). Assessment of Fecal Exposure Pathways in Low-Income Urban Neighborhoods in Accra, Ghana: Rationale, Design, Methods, and Key Findings of the SaniPath Study. Am J Trop Med Hyg. 2017 Oct;97(4):1020-1032. doi: 10.4269/ajtmh.16-0508. Epub 2017 Jul 19. PMID: 28722599; PMCID: PMC5637580.
3. USAID (2020) USAID Technical Brief. WASH and its Links to Nutrition. **https://www.globalwaters.org/sites/default/files/usaid_wash_nutrition_tech_brief_3.pdf**
4. USAID. (2022). Toward a hygienic environment for infants and young children: Limiting early exposures to support long-term health and well-being. Washington, DC, USAID Water, Sanitation, and Hygiene Partnerships and Learning for Sustainability (WASHPaLS) Project
5. USAID, 2018. Toward a Hygienic Environment for Infants and Young Children: A Review of the Literature. Washington, DC., USAID Water, Sanitation, and Hygiene Partnerships and Learning for Sustainability (WASHPaLS) Project.
6. USAID (2019). WASH and Its Links to Nutrition. WASH tech series. Available from: **https://www.globalwaters.org/resources/assets/usaid-water-and-development-technical-series-wash-and-its-links-nutrition**
7. TetraTech & FHI360. Improving Hygienic Environments for Children. UNC Water & Health Conference. Available from: **https://waterinstitute.unc.edu/knowledge/2020-w-h-conference-improving-hygienic-environments-for-children**
8. WASH Benefits Study resource center. Available from: **http://www.washbenefits.net/**
9. World Bank (2019). Nutrition-Sensitive Water Supply, Sanitation, and Hygiene. Water Global Practice. Guidance Note. **https://documents1.worldbank.org/curated/en/598771553098171805/pdf/135459-WP-P166089-PUBLIC.pdf**
10. Zvitambo Institute for Maternal and Health Research. SHINE Trial resource center. Available from: **https://www.zvitambo.com/shine-trial/**

ABOUT PRO-WASH

PRO-WASH (Practices, Research and Operations in Water, Sanitation and Hygiene) is an initiative funded by USAID's Bureau for Humanitarian Assistance (BHA) and led by Save the Children. PRO-WASH aims to improve the quality of activities, strengthen the capacity and skills of BHA implementing partners in WASH, and improve the level of knowledge and practices around WASH.

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