Links between malnutrition and intestinal “infections” in children in Lower Middle Income Countries

Mark Miller, MD

University of California, Berkeley on detail from US National Institutes of Health

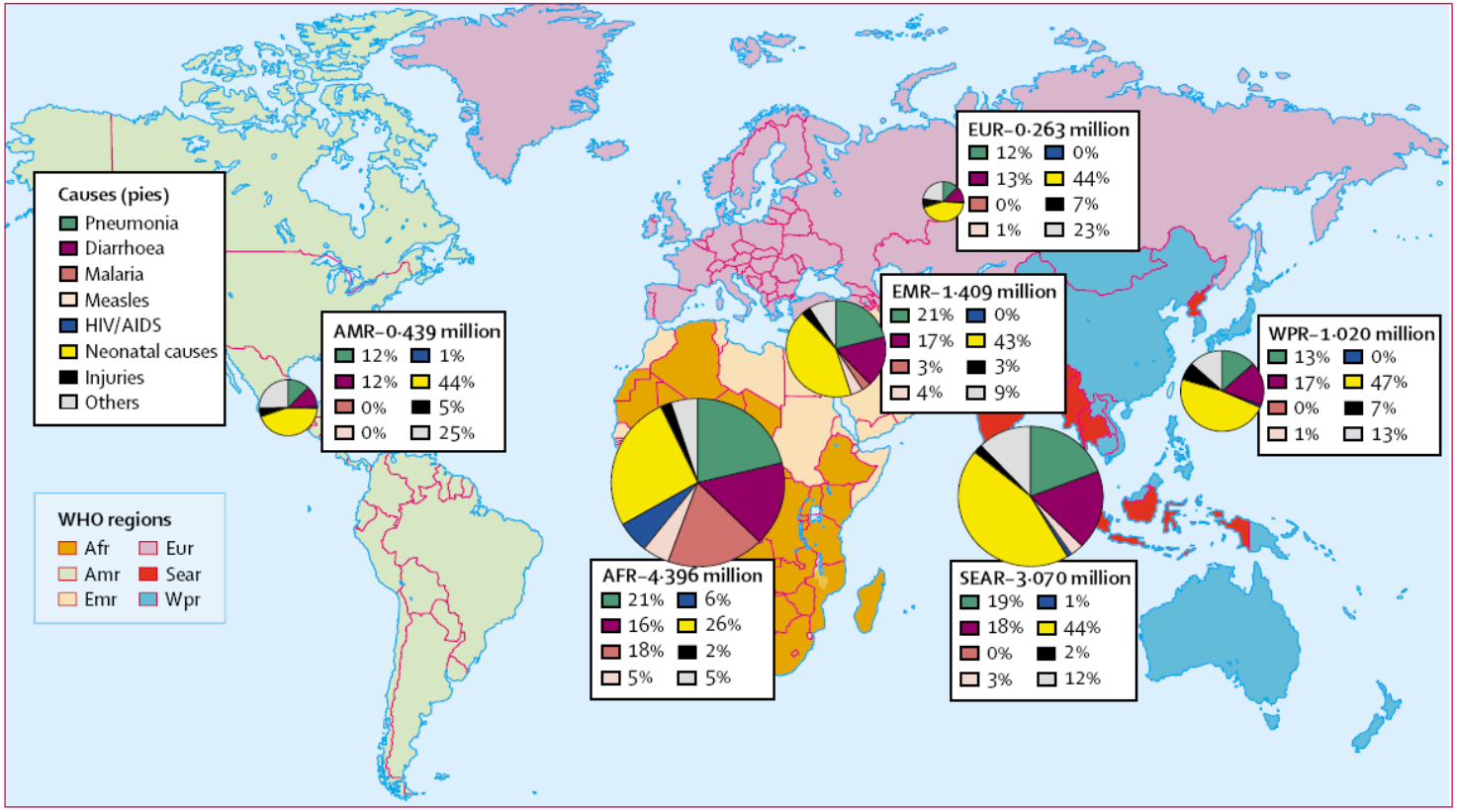
March 20-22, 2018

Veyrier-du-Lac

Child Heath Epidemiology Reference Group

WHO estimates of the causes of death in children

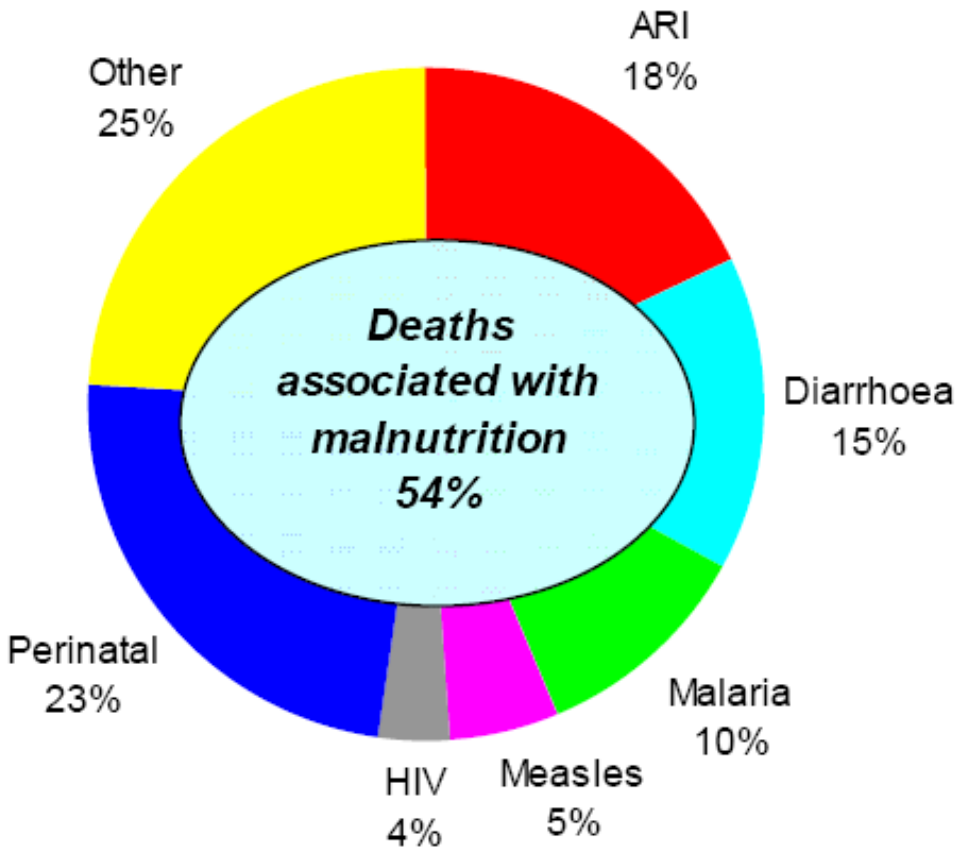
*Jennifer Bryce, Cynthia Boschi-Pinto, Kenji Shibuya, Robert E Black, and the WHO Child Health Epidemiology Reference Group**



Lancet 2003;361

Figure 3: **Number of deaths in children younger than age 5 years and their distribution by cause for the six WHO regions (yearly average for 2000–03)**
Size of circle represents number of deaths in region. Afr=Africa. Amr=Americas. Emr=Eastern Mediterranean. Eur=Europe. Sear=Southeast Asia. Wpr=Western Pacific.

Figure 1.2 Proportional mortality in children younger than five years old[a]



WHO (2004). *Inheriting the world. The atlas of children's health and the environment*. Geneva, World Health Organization.

[a] Source: WHO (2004).

Global Health Diagnostics Forum

Reducing stunting among children: the potential contribution of diagnostics

Authors: **Karen A. Ricci[1], Federico Girosi[1], Phillip I. Tarr[2], Yee-Wei Lim[1], Carl Mason[3], Mark Miller[4], James Hughes[5], Lorenz von Seidlein[6], Jan M. Agosti[7] & Richard L. Guerrant[8]**

PREFACE

Stunting is a major burden in developing countries, affecting ~147 million children. Repeated or prolonged episodes of diarrhoea during childhood increase the risk of stunting, which is believed to be associated with significant morbidity. Although the relationships between malnutrition, environment and diarrhoeal illnesses are complex, studies have suggested a connection between stunting and

diarrhoeal diseases posed special
Unlike the other diseases addres
forum, diarrhoea does not requi
determine its presence; rather, it is
of an infection that can be caused b
of different bacterial, viral and para
isms. Regardless of aetiology, dia
ease typically does not require
symptomatic interventions, and is
marily by oral rehydration thera

Nature. 2006 Nov 23;444 Suppl 1:29-38.

Systems Approach to Evaluate Health Outcomes Amenable to Public Health

What is the burden from diarrhea (enteric dysfunction?

Mortality? Morbidity?

Acute, Chronic?

Simple extrapolations?

What is the distribution change over time?
What we know?
What we know that we don't know?
What we don't know about what we don't know?

Evaluating a syndromic disease;
Qualitative and quantitative assessments regarding enteric disease and its impact on health and welfare

Review series introduction

Global health inequity: scientific challenges remain but can be solved

Carol A. Dahl and Tadataka Yamada

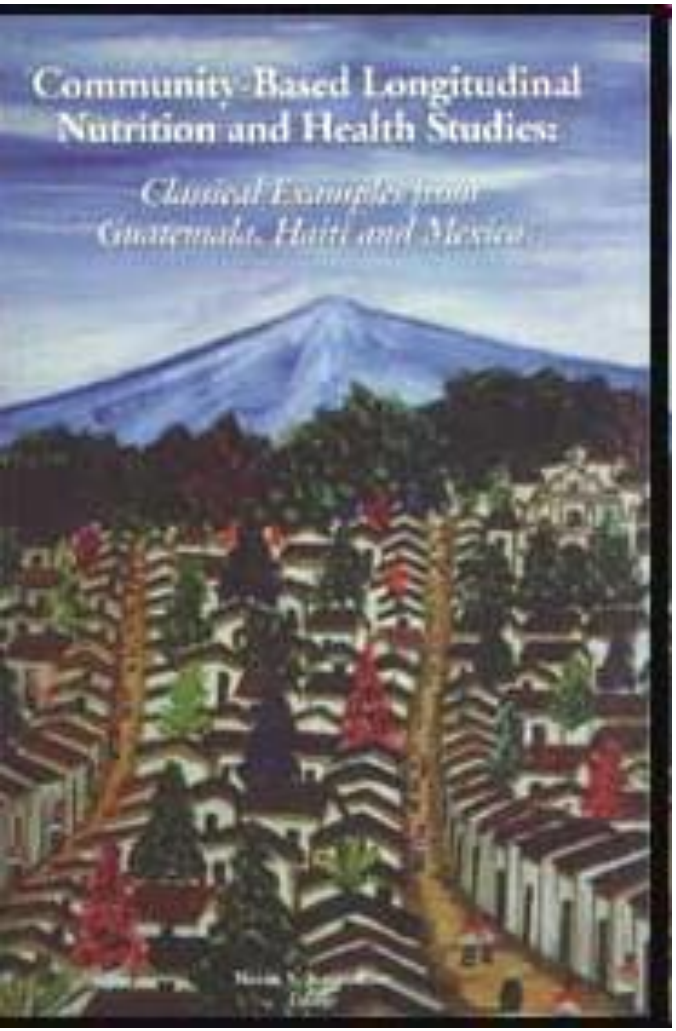
Enteric infections, diarrhea, and their impact on function and development

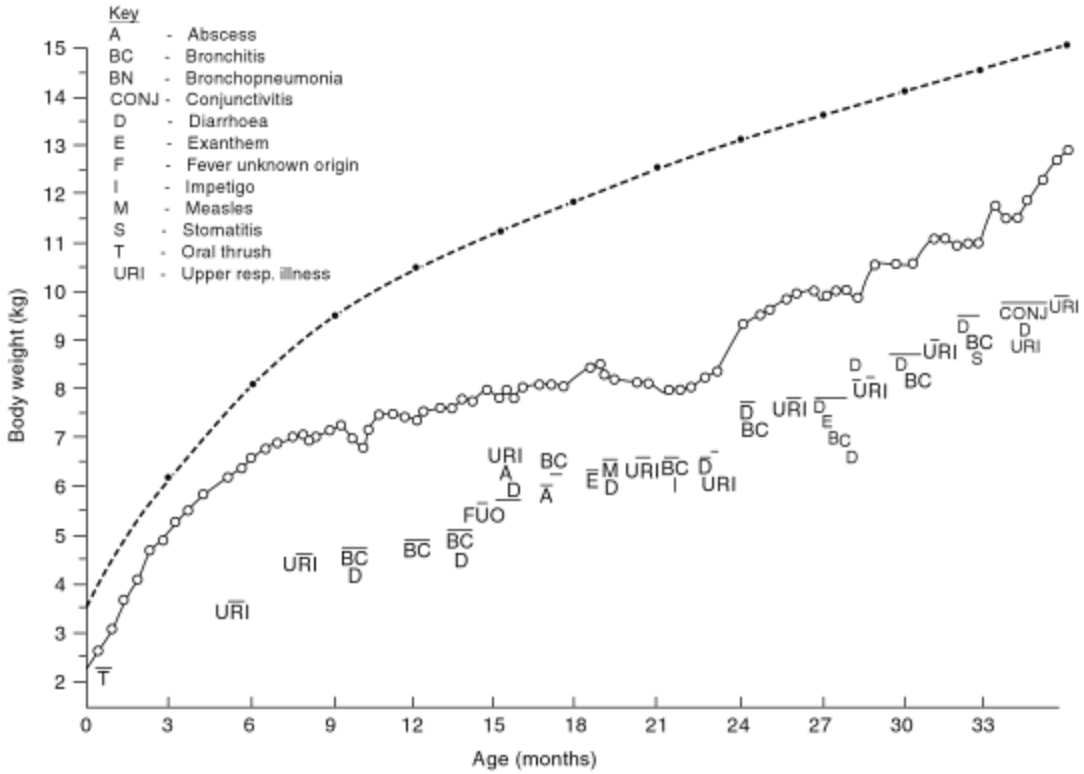
William A. Petri, Jr., Mark Miller, Henry J. Binder, Myron M. Levine, Rebecca Dillingham and Richard L. Guerrant

New challenges in studying nutrition-disease interactions in the developing world

Andrew M. Prentice, M. Eric Gershwin, Ulrich E. Schaible, Gerald T. Keusch, Cesar G. Victora and Jeffrey I. Gordon

Classic Studies



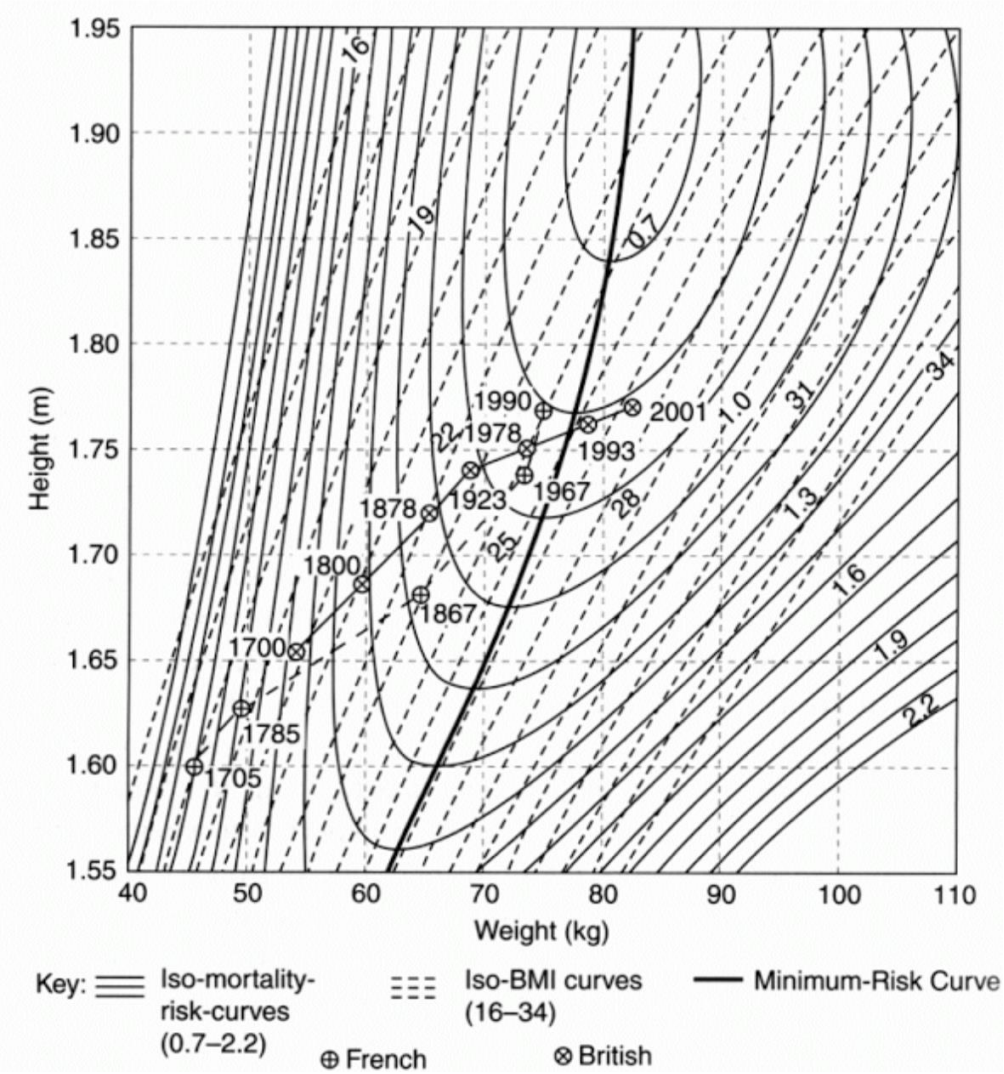


Longitudinal community-based studies of host, agent, and environmental factors responsible for disease

Studies are difficult and costly

Five classical nutrition oriented field studies, 1 Mexico, 3 Guatemala, 1 Haiti

Robert Fogel, Nobel Prize Economist BMI and Life Expectancy across time



Fogel R. The Escape from Hunger and Premature Death, 1700-2100, 2004

Public health significance of growth

- 26% of children worldwide are stunted
- Stunting is an underlying cause of ~17% of child mortality
- Long-term associations:
  - Decreased economic activity?
  - Impaired cognitive development?
  - Chronic disease at older ages?

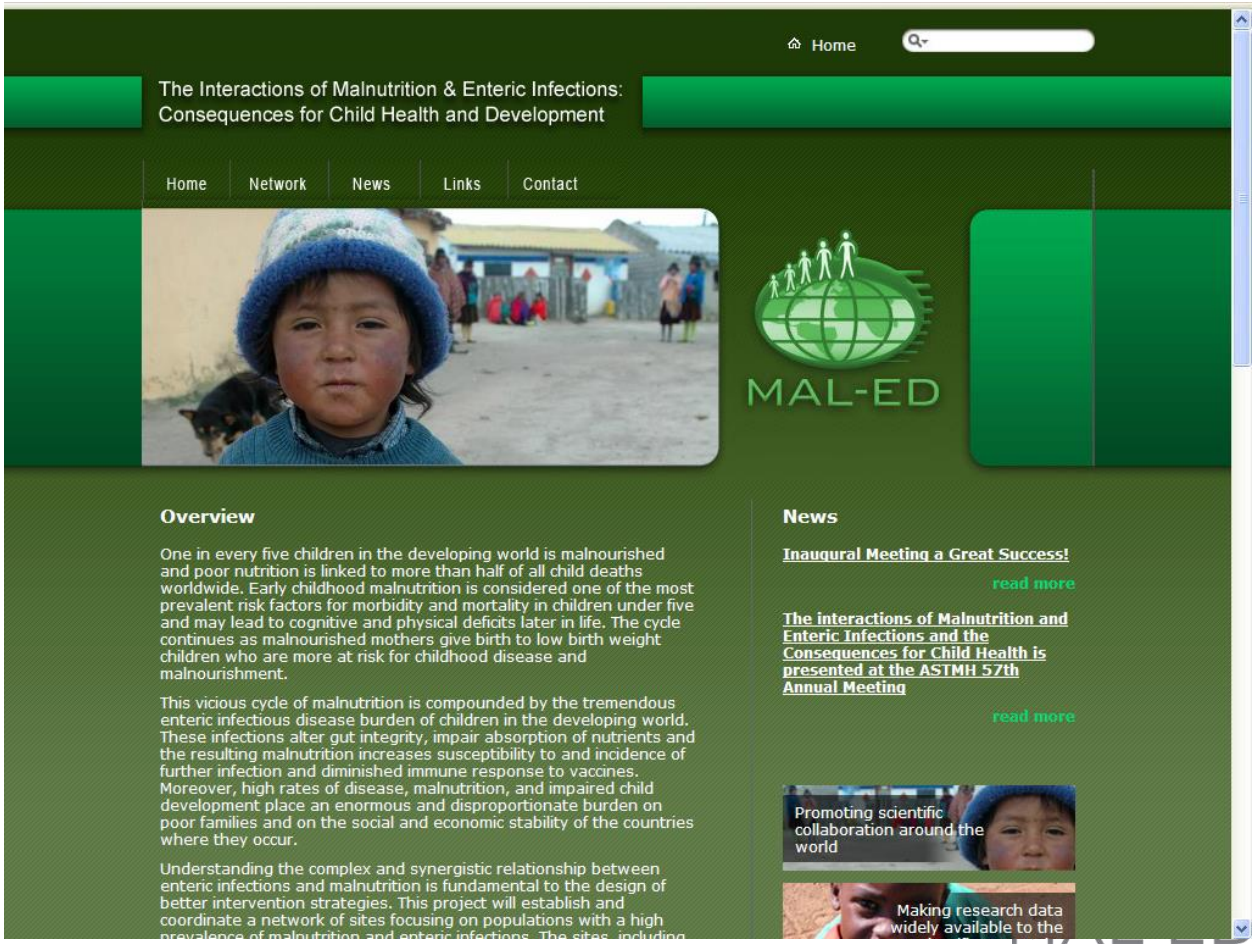
Maternal and Child Nutrition 1

Maternal and child undernutrition and overweight in low-income and middle-income countries

Robert E Black, Cesar G Victora, Susan P Walker, Zulfiqar A Bhutta*, Parul Christian*, Mercedes de Onis*, Majid Ezzati*, Sally Grantham-McGregor*, Joanne Katz*, Reynaldo Martorell*, Ricardo Uauy*, and the Maternal and Child Nutrition Study Group†

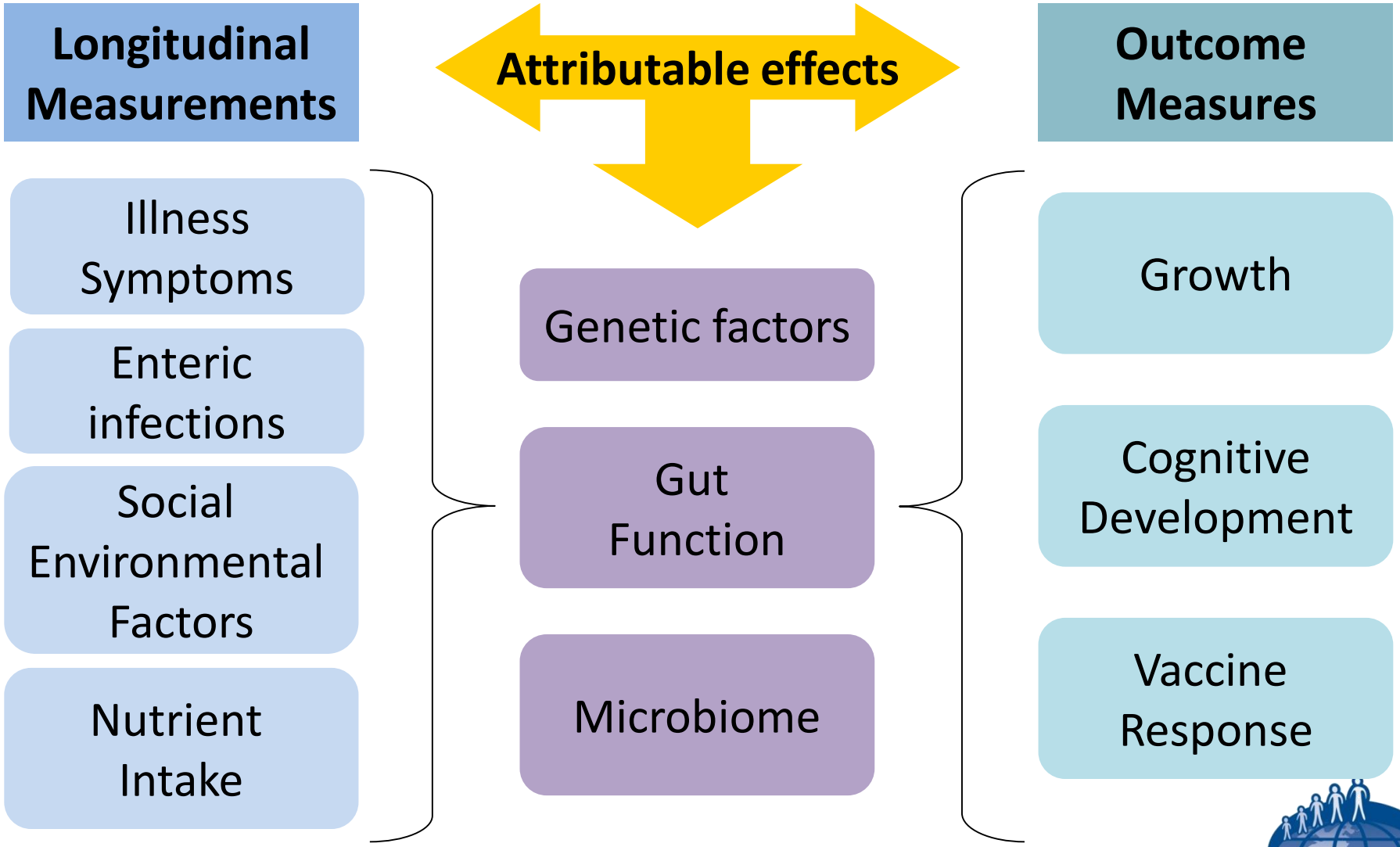
The MAL-ED Study

Platform to decipher relationships among enteric infection, gut physiology and malnutrition and their effects on child growth, development and vaccine response



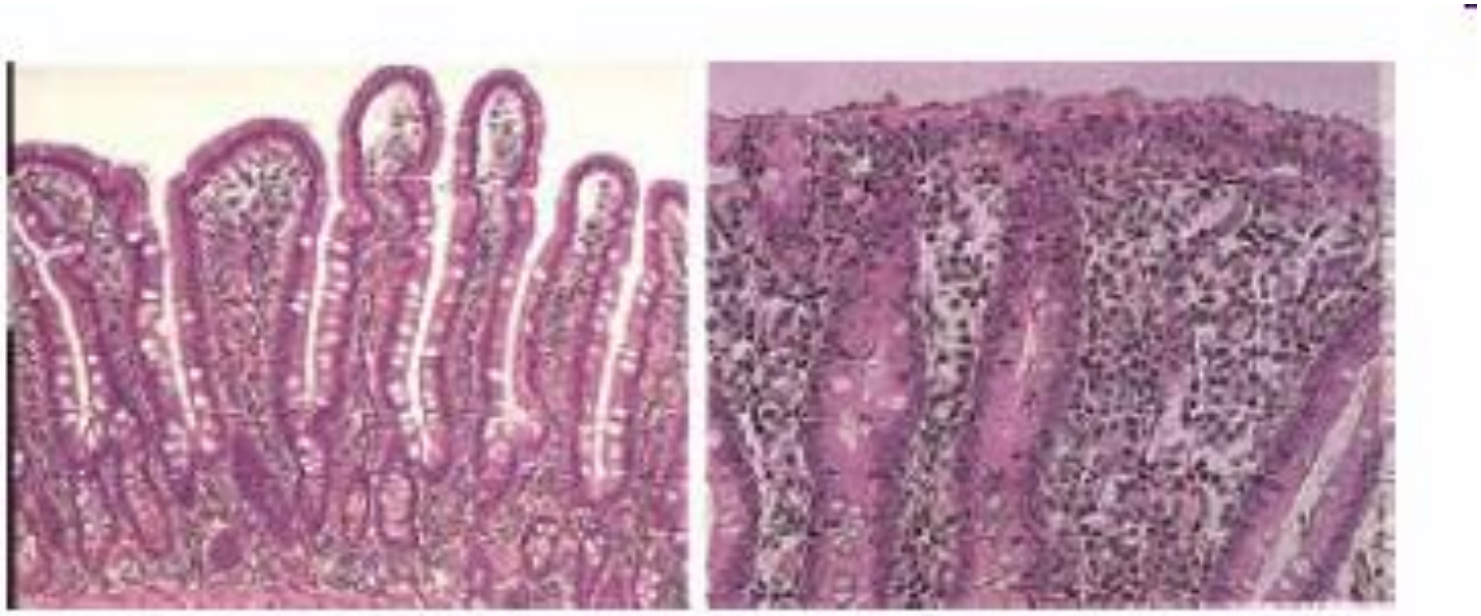

MAL-ED Network Field Sites
Iquitos, Peru
Fortaleza. Brazil*
Haydom, Tanzania
Limpopo, S. Africa
Bhaktapur, Nepal
Mirpur, Bangladesh*
Naushero Feroz, Pakistan
Vellore, India
Collaborating Institution
Longitudinal Cohort Site Institution
Case-Control Site
Suomi Finland
Sverige Sweden
Norge Norway
Россия Russia
Polska Poland
Deutschland Germany
Україна Ukraine
France
Italia Italy
España Spain
Türkiye Turkey
Kazakhstan
Mongolia
中国 China
Iraq
Iran
Afghanistan
Pakistan
India
Thailand
Saudi Arabia
Algeria
Libya
مصر Egypt
Mauritania
Mali
Niger
Chad
Sudan
Nigeria
Ethiopia
DR Congo
Tanzania
Angola
Namibia
Botswana
Madagascar
South Africa
Canada
United States
México
North Atlantic Ocean
Venezuela
Brasil Brazil
Perú
Bolivia
Chile
South Atlantic Ocean
Indian Ocean

Hypotheses/Research Questions



MAL-ED

Environmental Enteropathy



Explorations of Gastrointestinal Physiology in Environmental Milieu

The New York Times

Poor Sanitation in India May Afflict Well-Fed Children With Malnutrition

July 13, 2014

"The cause of many of our diseases is the condition of our lavatories and our bad habit of disposing of excreta anywhere and everywhere," Gandhi wrote in 1925.

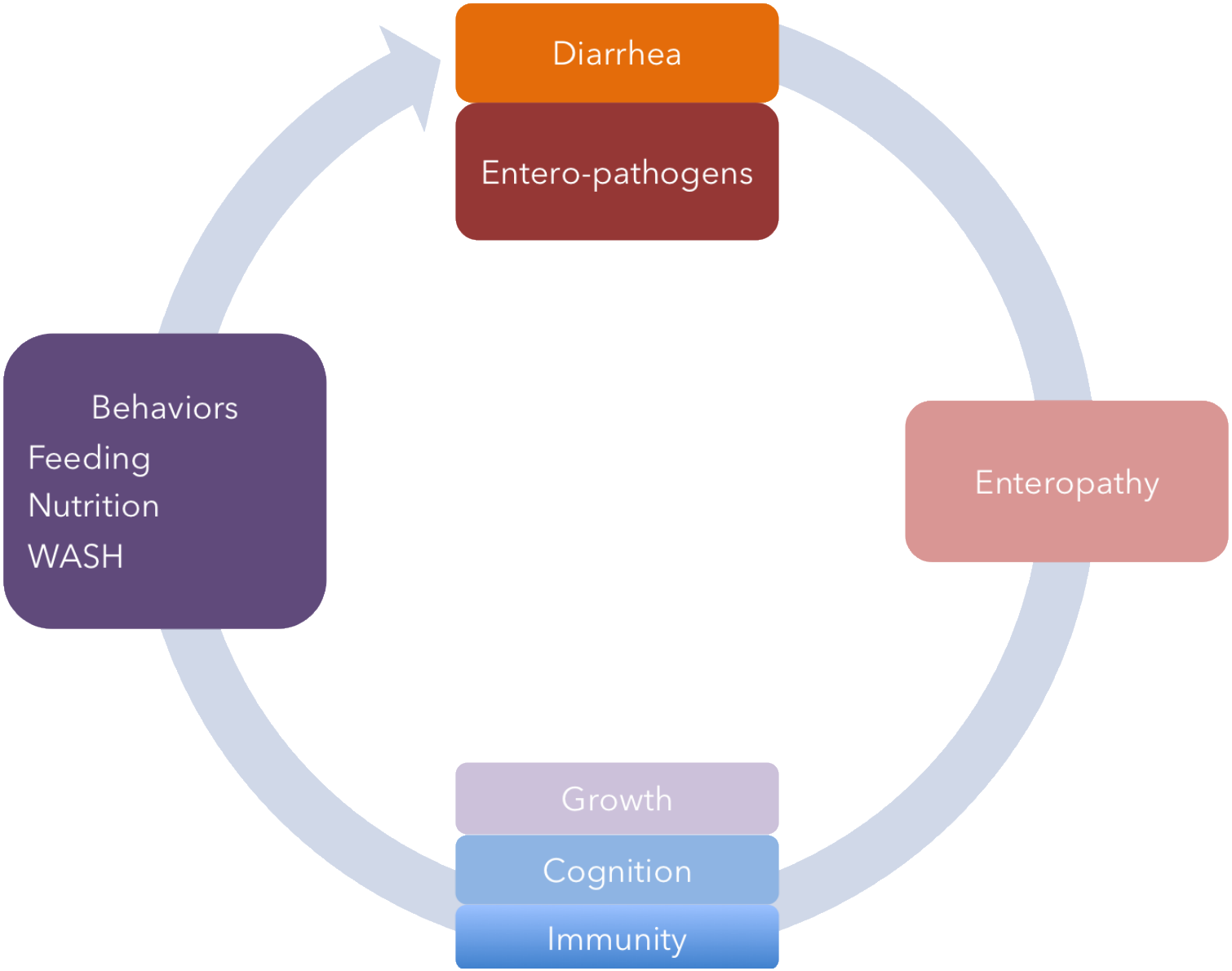
The Perils of Poor Sanitation in India

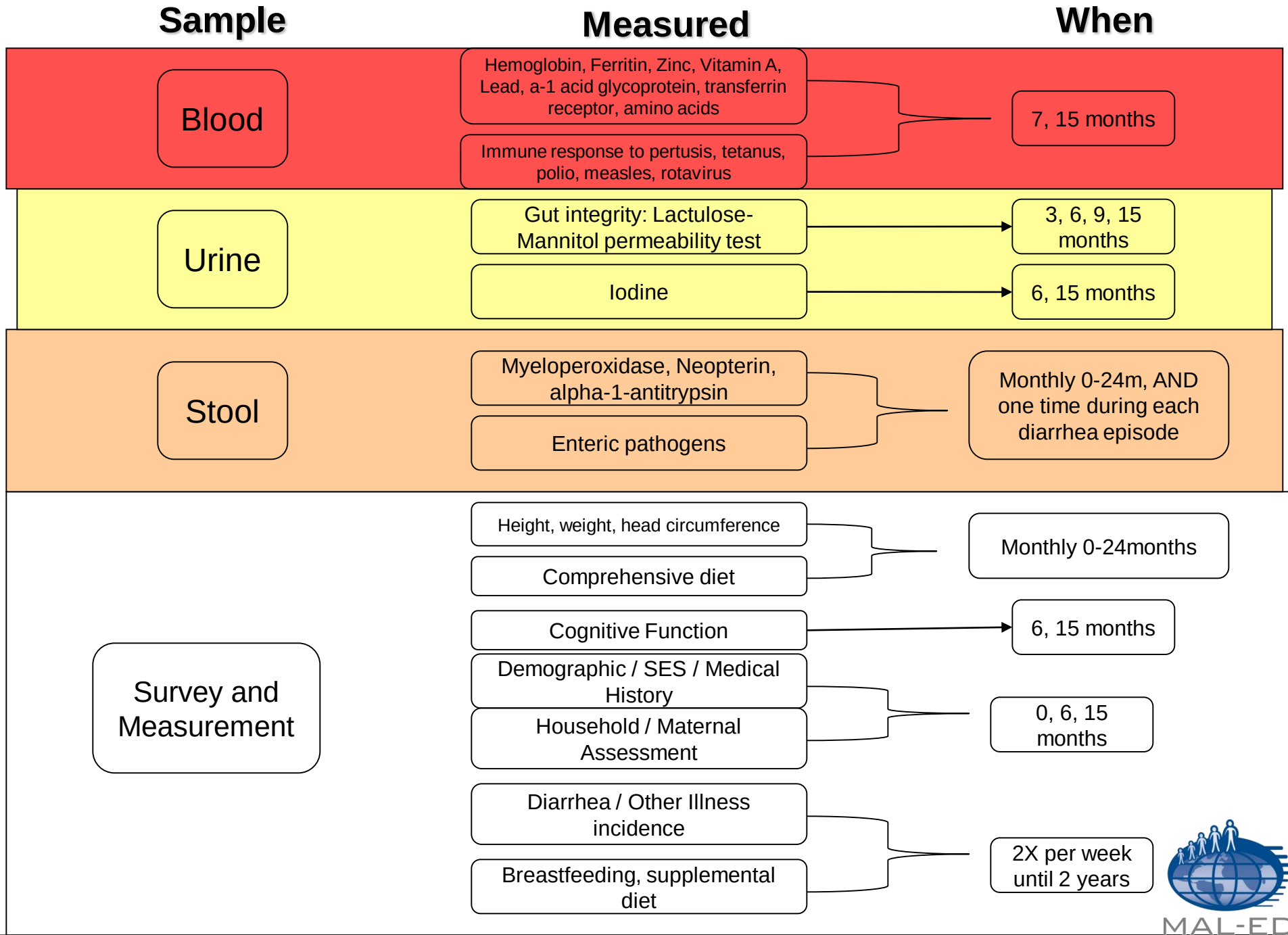
Hindu devotees bathed in the polluted waters of the Ganges River in Varanasi, India. Millions of pilgrims bathe along Varanasi's ancient riverfront, but a stream of human waste flows directly into the river just above the bathing ghats, steps leading down to the river.

Daniel Berehulak for The New York Times

"In the meantime, I think we can all agree that it's not a good idea to raise children surrounded by poop."

Cycle(?) of Malnutrition and Enteric Infection and Possible Interventions

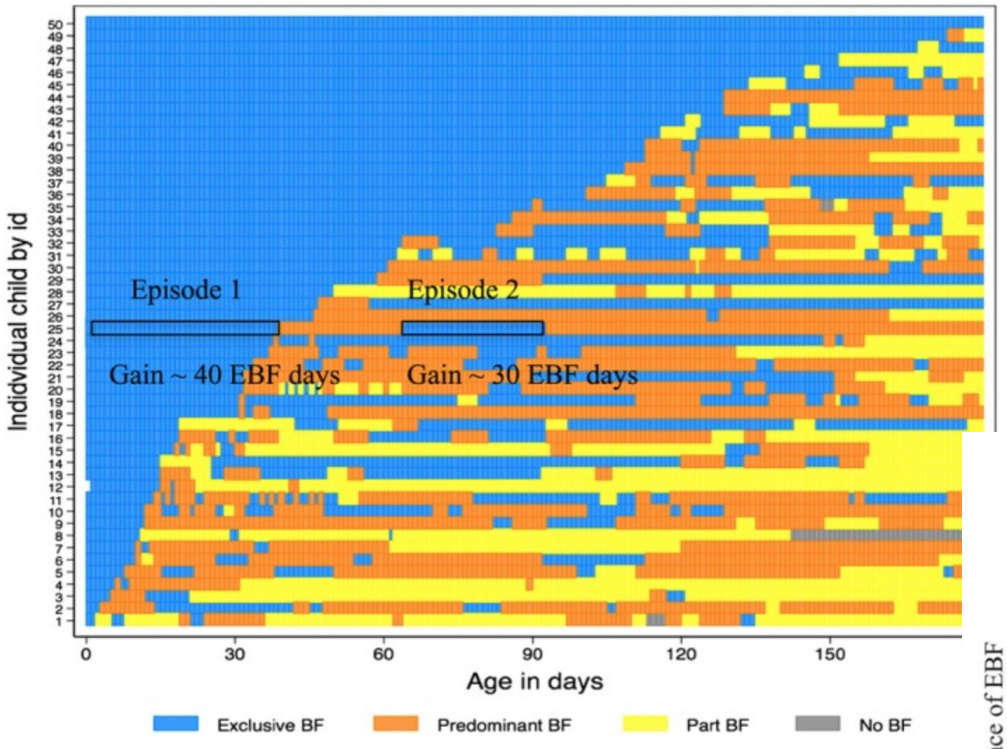


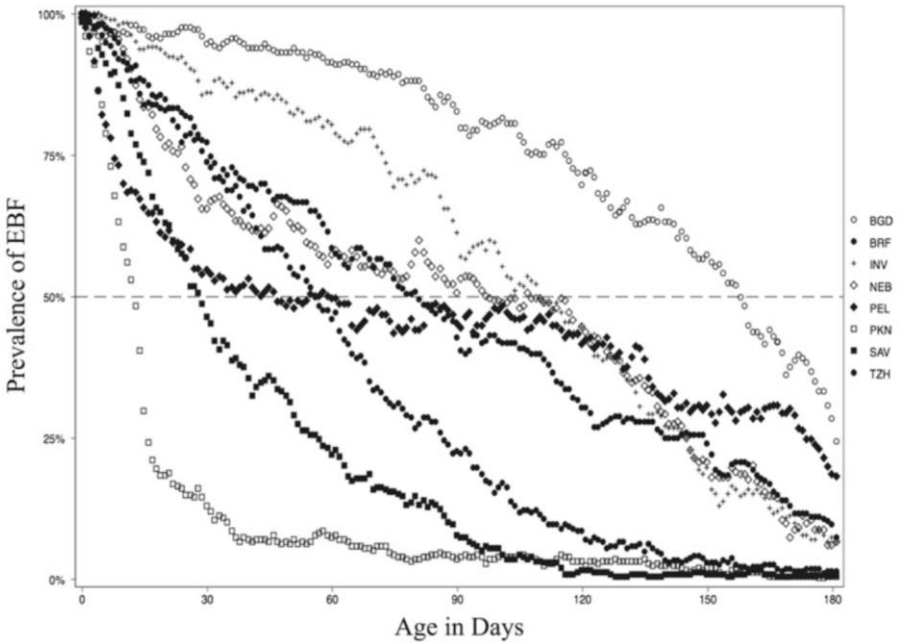

Sample
Measured
When
Blood
Hemoglobin, Ferritin, Zinc, Vitamin A, Lead, a-1 acid glycoprotein, transferrin receptor, amino acids
Immune response to pertusis, tetanus, polio, measles, rotavirus
7, 15 months
Urine
Gut integrity: Lactulose-Mannitol permeability test
3, 6, 9, 15 months
Iodine
6, 15 months
Stool
Myeloperoxidase, Neopterin, alpha-1-antitrypsin
Enteric pathogens
Monthly 0-24m, AND one time during each diarrhea episode
Survey and Measurement
Height, weight, head circumference
Comprehensive diet
Monthly 0-24months
Cognitive Function
6, 15 months
Demographic / SES / Medical History
Household / Maternal Assessment
0, 6, 15 months
Diarrhea / Other Illness incidence
Breastfeeding, supplemental diet
2X per week until 2 years
MAL-ED

Levels of Analysis

- Individual
- Site
- Entire Cohort

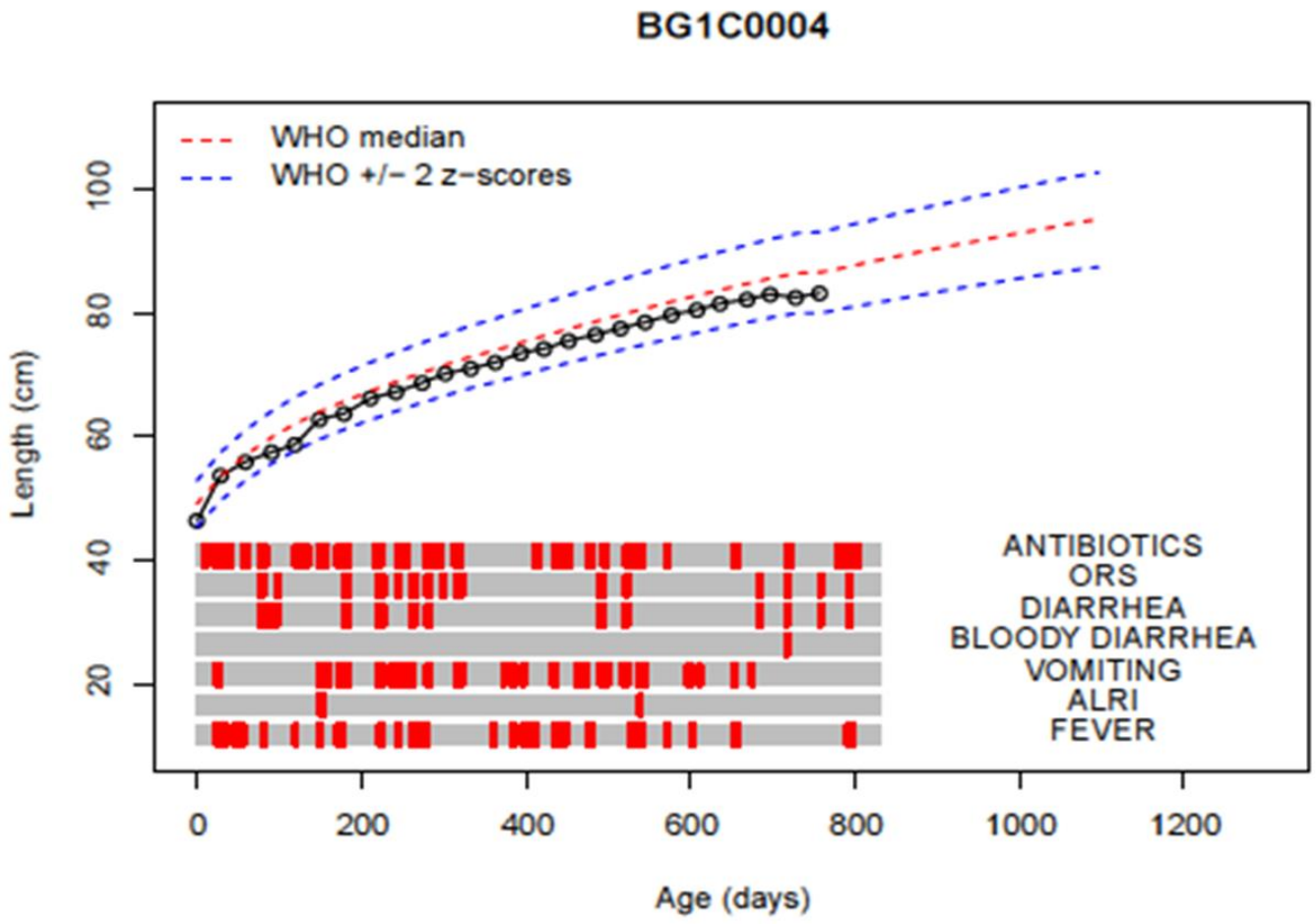
Exclusive Breast Feeding Practices by Day



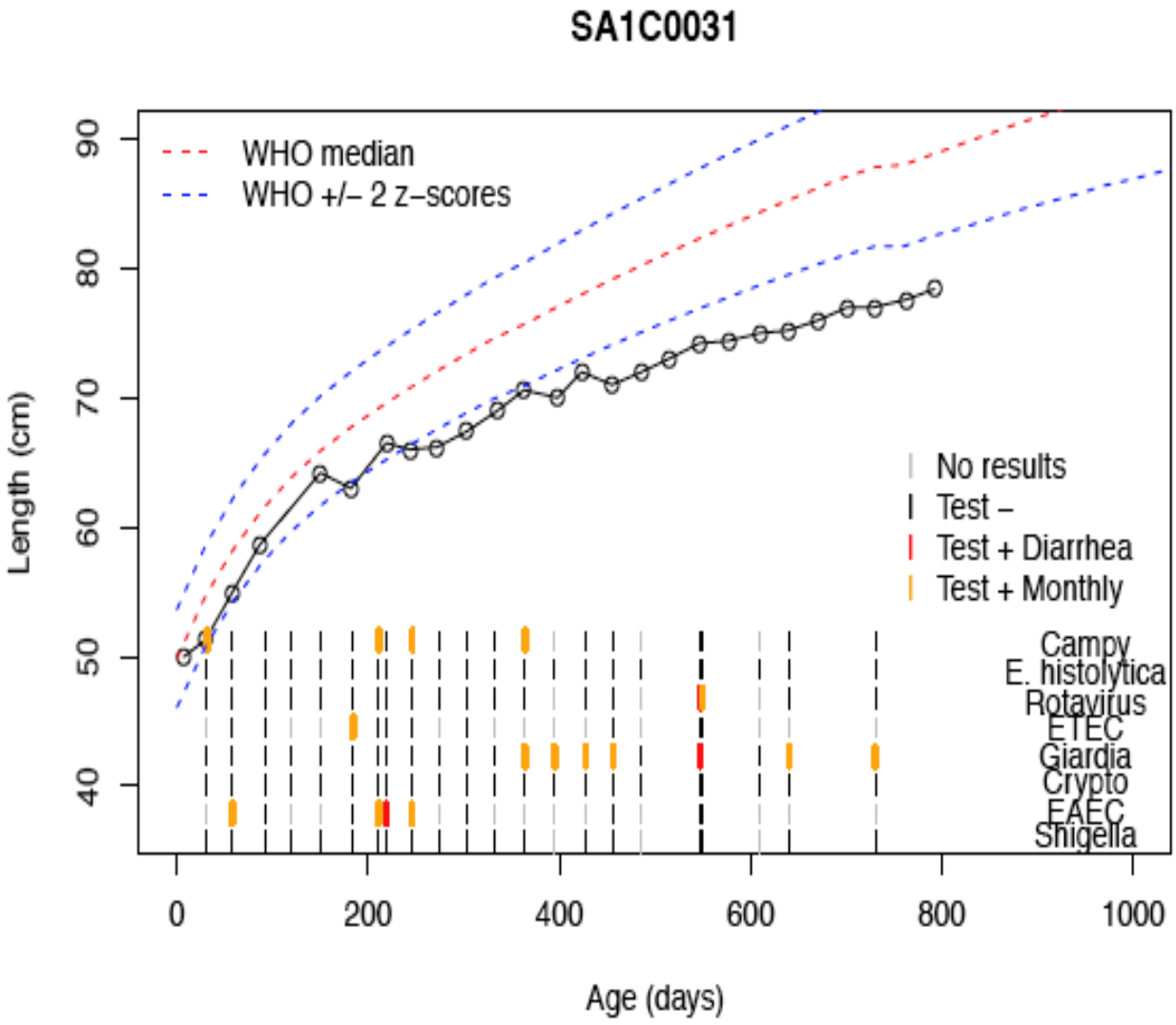


Ambikapathi R et al. Maternal & Child Nutrition (2016), 12, pp. 740–756

Growth trajectory and illness indicators of one MAL-ED child



Growth trajectory and pathogen isolation experience of one child

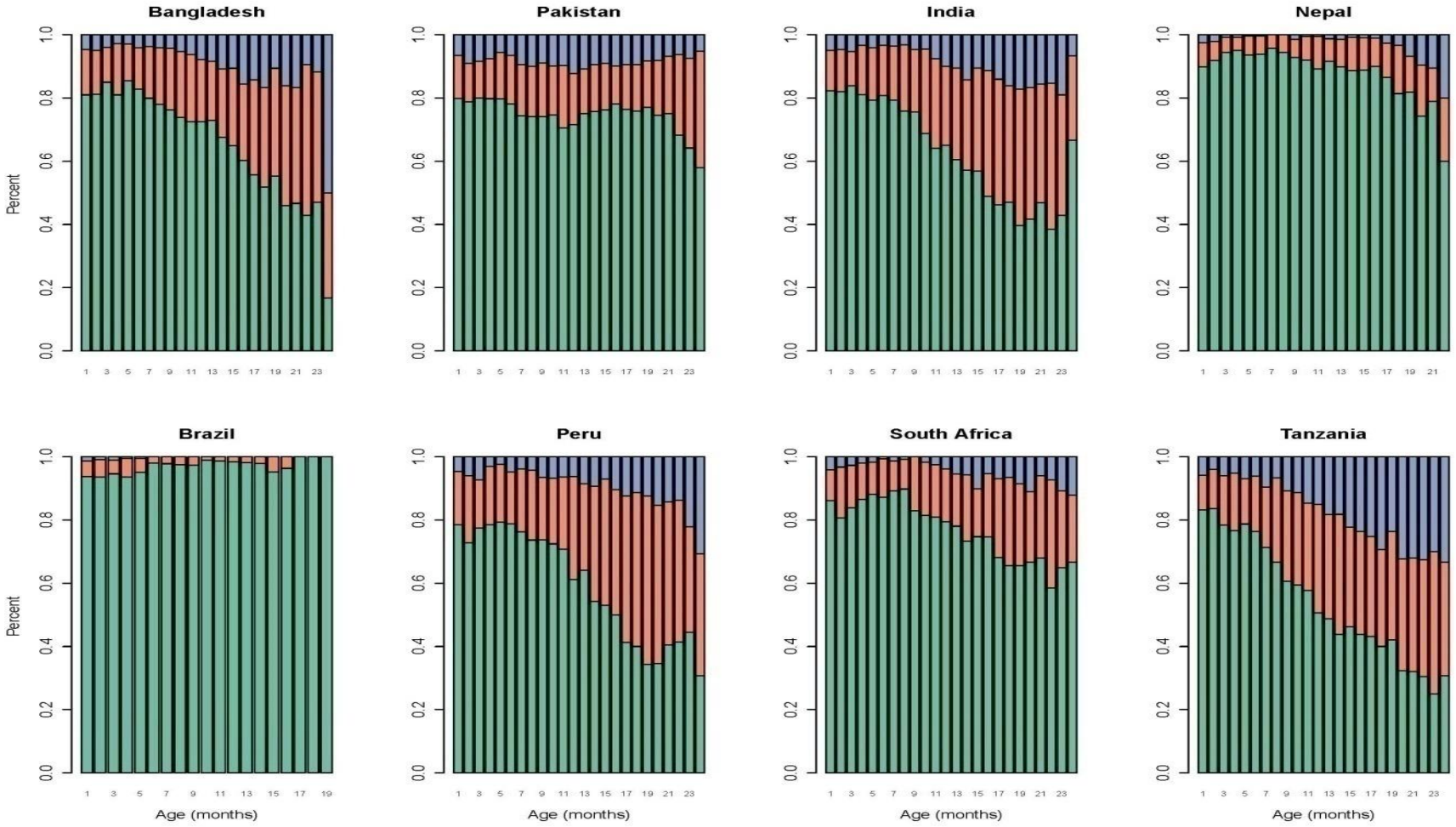


Individual Analysis
Pathogen Detection in one MAL-ED child

Stool Type	Age (months)	Pathogen(s) Isolated
Monthly	0.6	
Monthly	1.0	Aeromonas, Giardia , ETEC
Monthly	1.9	
Diarrhea	2.3	Campy
Diarrhea	2.9	EPEC, Giardia
Diarrhea	3.6	EPEC, Giardia (?)
Monthly	4.0	Aeromonas
Diarrhea	4.2	Campy, Giardia
Monthly	4.9	Giardia
Diarrhea	5.2	EAEC, EPEC, ETEC, Astrovirus, Giardia
Diarrhea	5.4	Astrovirus
Monthly	6.0	Aeromonas
Monthly	6.9	Giardia
Diarrhea	7.4	Crypto
Monthly	7.9	
Diarrhea	8.3	Aeromonas, Campy, Rotavirus
Diarrhea	8.6	Aeromonas, Campy
Monthly	9.0	Campy
Monthly	9.9	Campy, Giardia
Monthly	11.0	Aeromonas, Campy, EPEC, Giardia
Diarrhea	11.5	Campy (?)
Monthly	11.9	Campy

MAL-ED

Prevalence of stunting by severity by 0-24 months

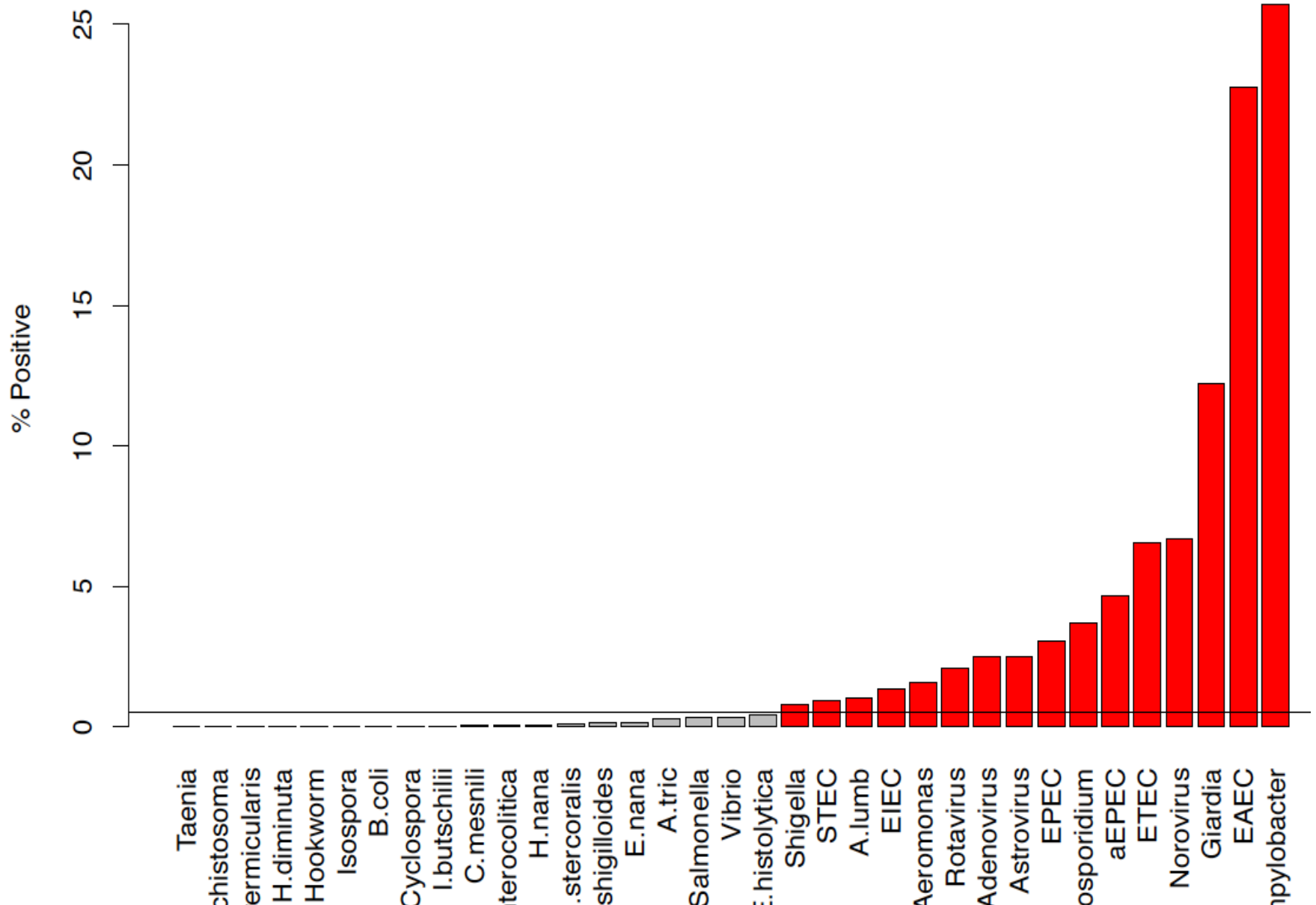


Z-scores **>2 No stunting**

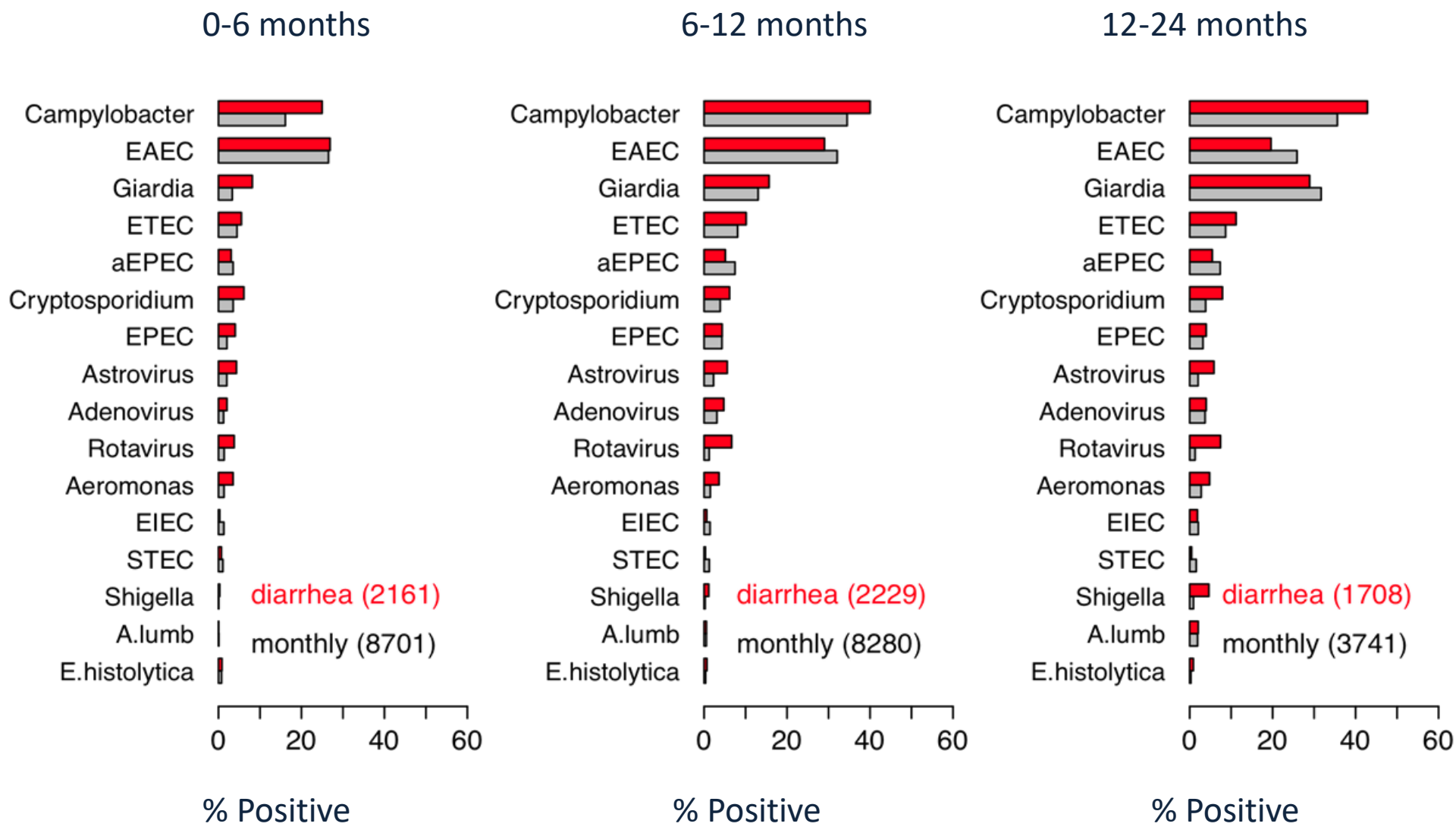
<2 to <3 Moderate stunting

<3 Severe

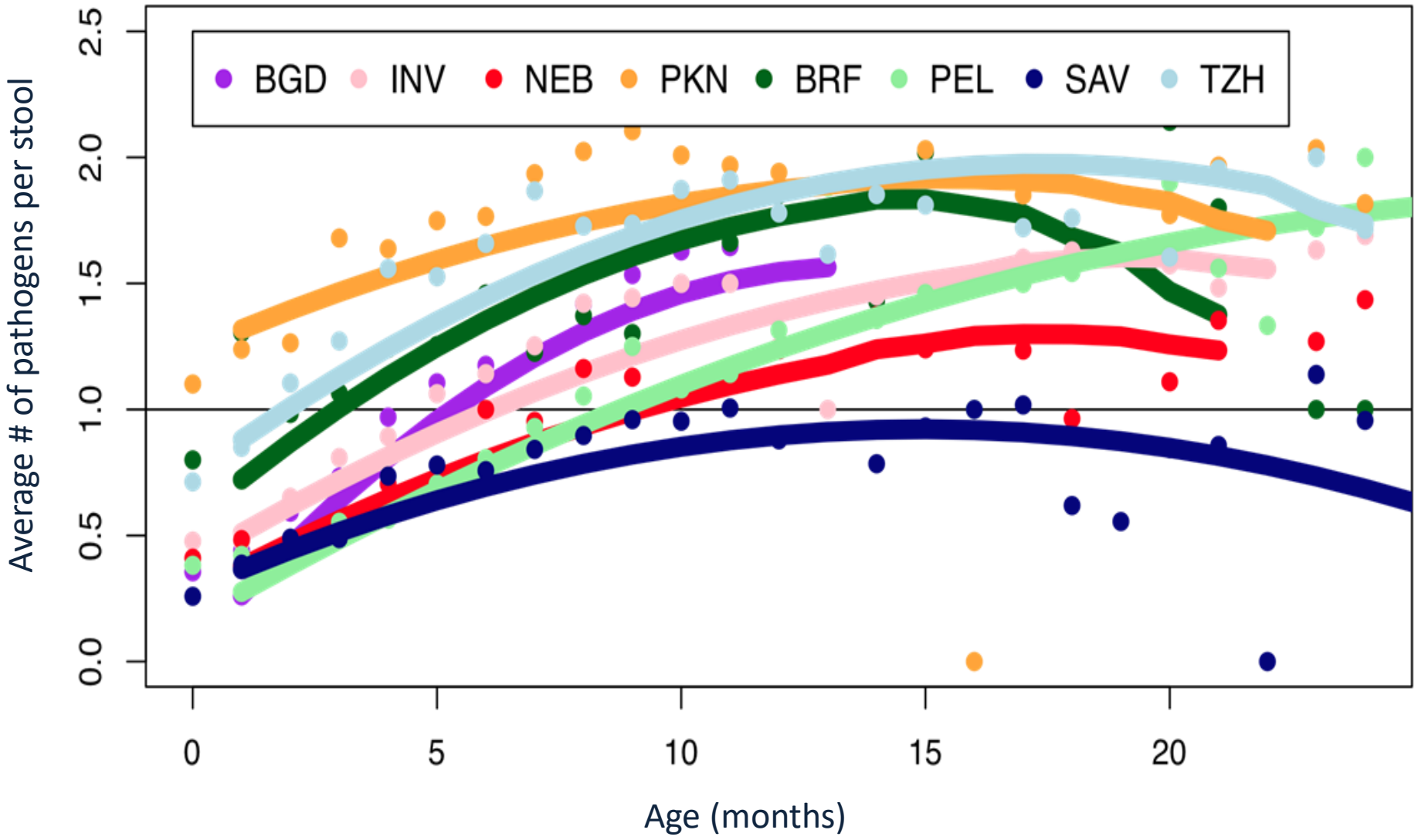
Stool Analysis Pooled Data



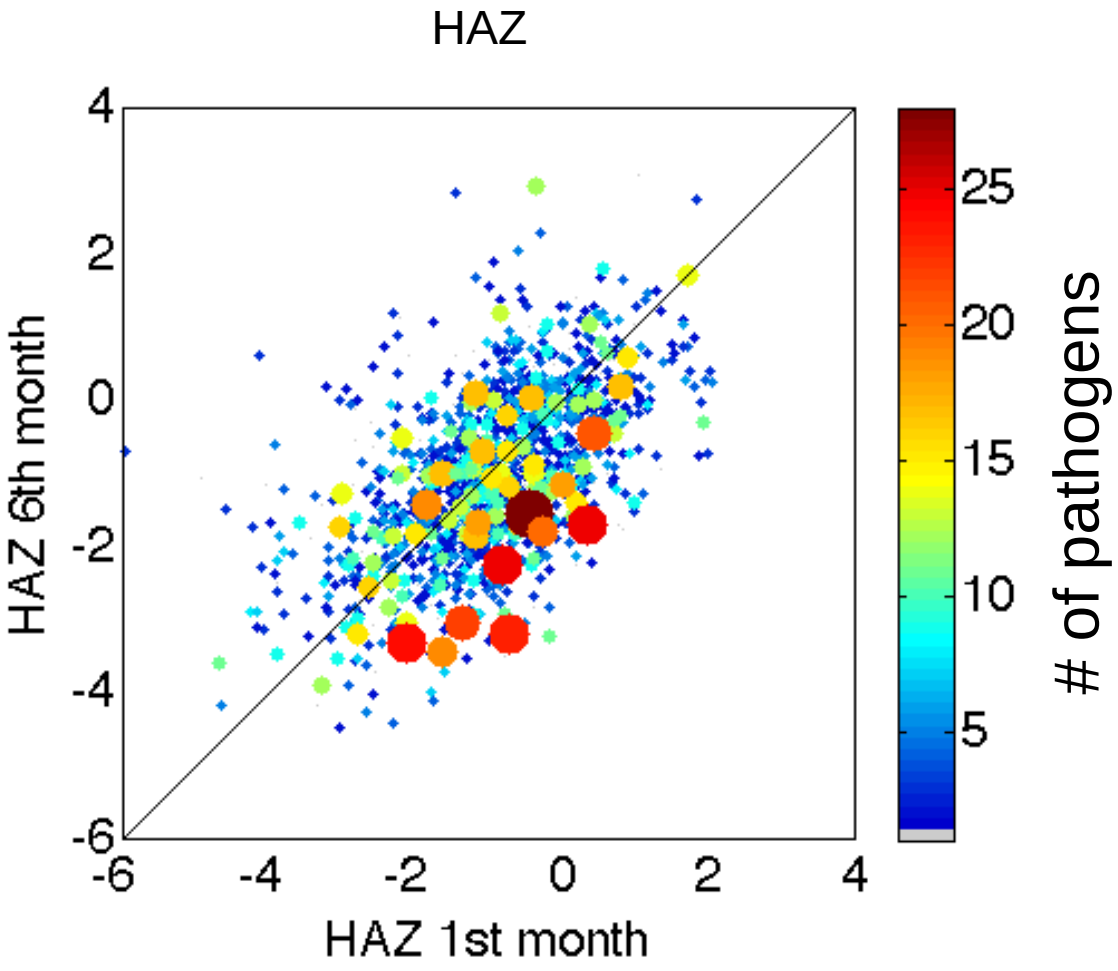
Pathogen: Diarrhea vs Monthly Stools



Pathogen Detection Monthly Stools by Age, Site

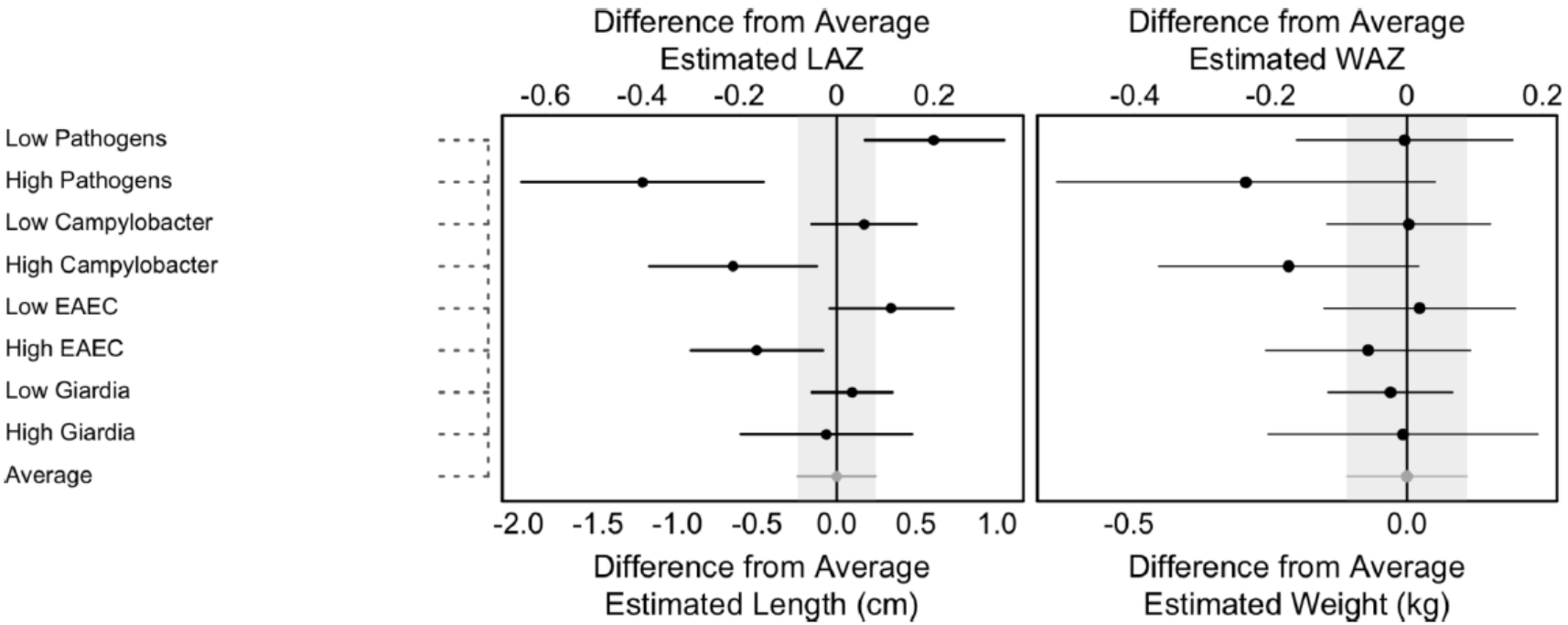


All MALED children, each a dot, complete data, # of pathogens in first 6 months



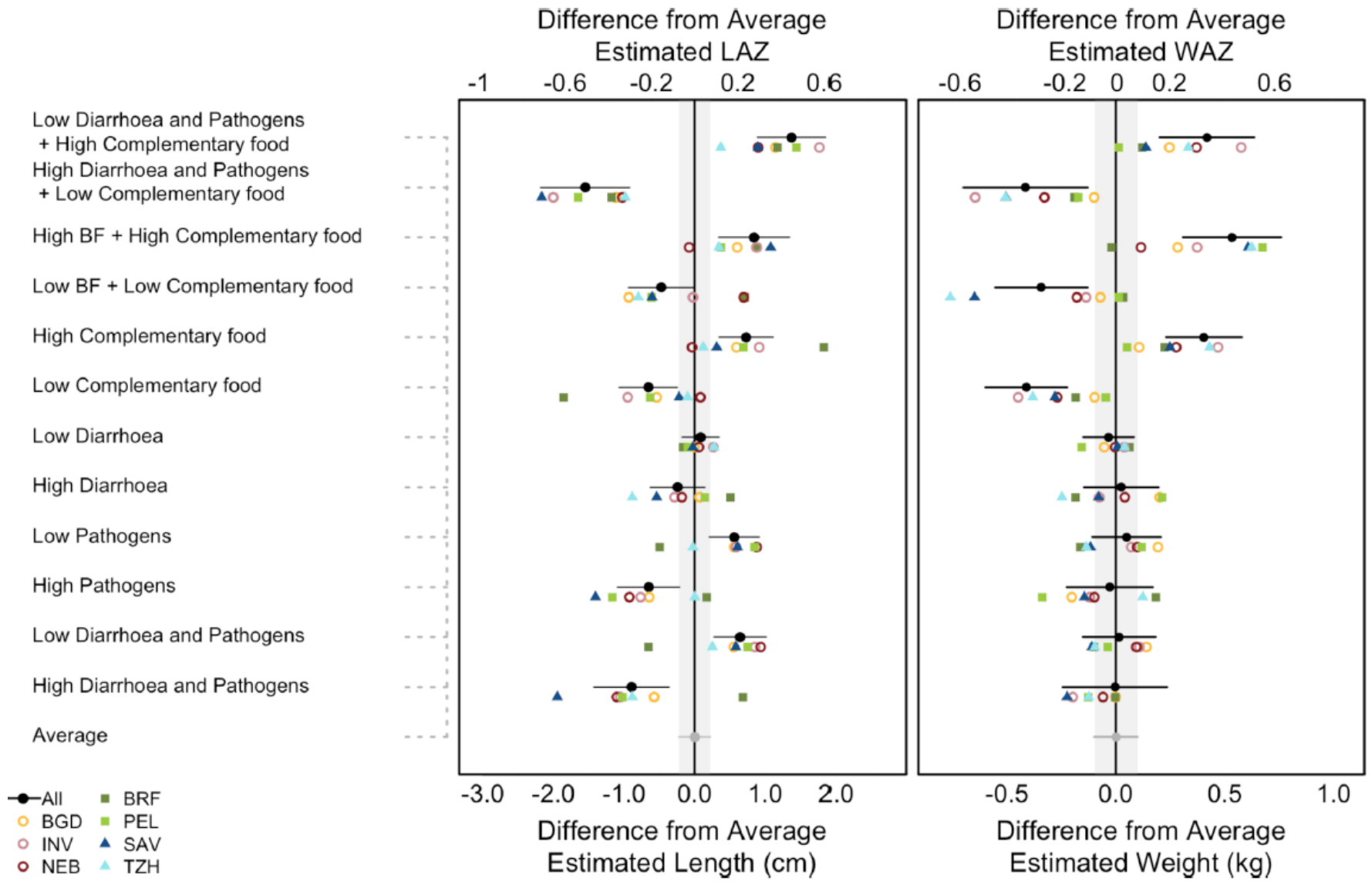
MAL-ED

Relationship between Growth and Specific Enteropathogens

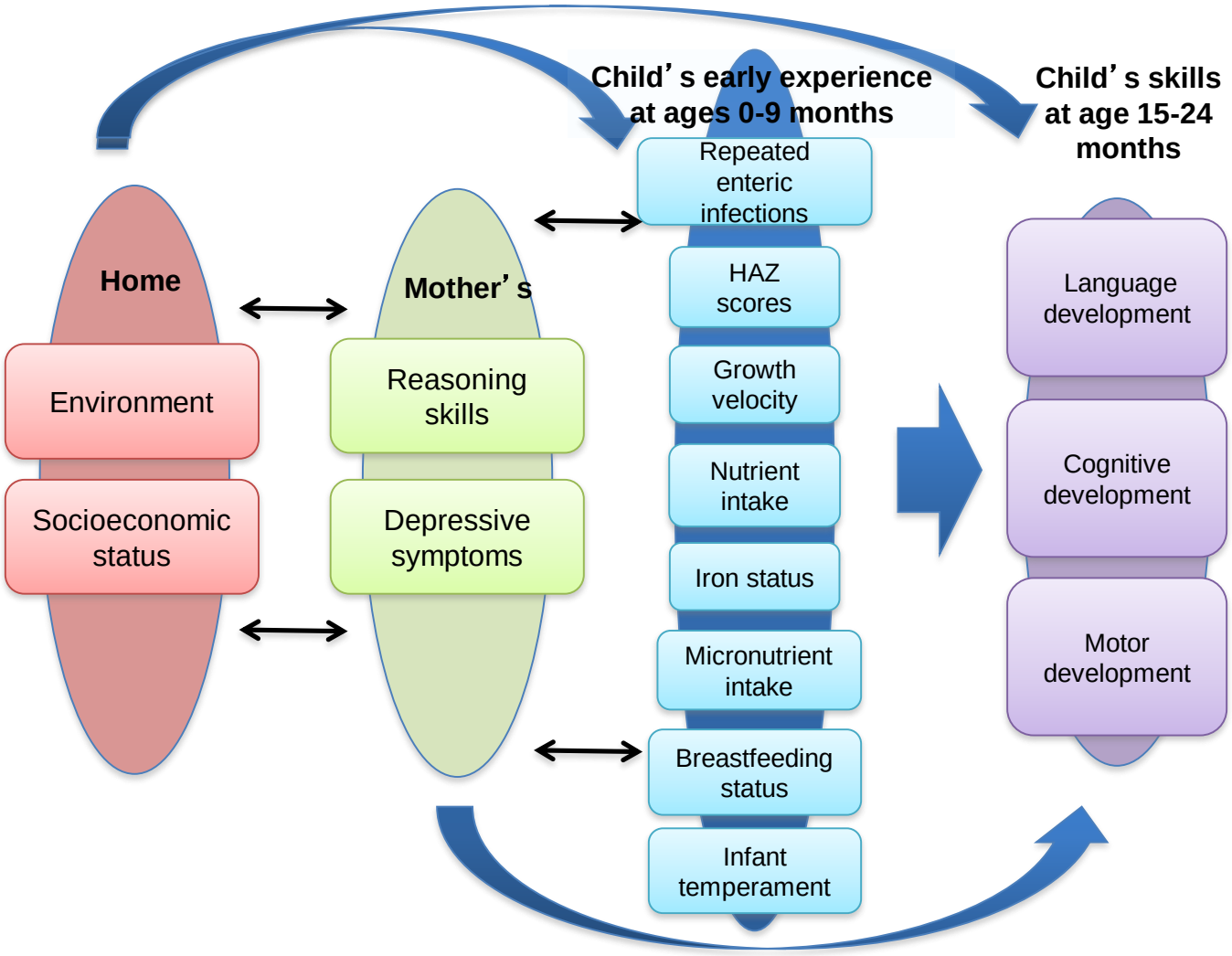


MAL-ED Network Investigators. BMJ Glob Health 2017;2:e000370

Relationship between Growth and Other Factors by Site



MAL-ED Network Investigators. BMJ Glob Health 2017;2:e000370


Child's early experience at ages 0-9 months
Child's skills at age 15-24 months
Home
Environment
Socioeconomic status
Mother's
Reasoning skills
Depressive symptoms
Repeated enteric infections
HAZ scores
Growth velocity
Nutrient intake
Iron status
Micronutrient intake
Breastfeeding status
Infant temperament
Language development
Cognitive development
Motor development

Major outcomes to date from MAL-ED:

<5% of the children at 6 of 8 sites are fed according to the WHO recommendation for exclusively breastfeeding to 6m

At several sites, it is common practice to expose neonates to solid foods.

The quality of the early complementary food diet is low in diversity, in access to vitamin A and iron source foods.

Major outcomes to date from MAL-ED:

~50% of children had a mean length-for-age Z-score <-1 during the first month of life

Despite intense follow, 23-70% were stunted; Z score <-2 at 24 months in 7 of the 8 sites; Brazil site, linear growth is normalized (WHO standard)

Major outcomes to date from MAL-ED:

- Differences in the patterns of diarrhea illness and specific pathogens
  - Common: rotavirus, ST-producing ETEC, Shigella, Cryptosporidium
  - Unexpected: campylobacter, astrovirus and norovirus

- High incidence of the carriage without diarrhea

- Diarrhea, does not appear to be associated with linear growth in children, but pathogen load does

- Nutritional factors may have synergistic effects.

Major outcomes to date from MAL-ED:

Gut permeability, immunology and physiology demonstrated that lactulose-mannitol the “gold standard” of gut permeability was not consistently associated with growth faltering

Other measures of gut function with 3 other biomarkers, alpha-1- antitrypsin, neopterin, and myeloperoxidase were associated with growth faltering

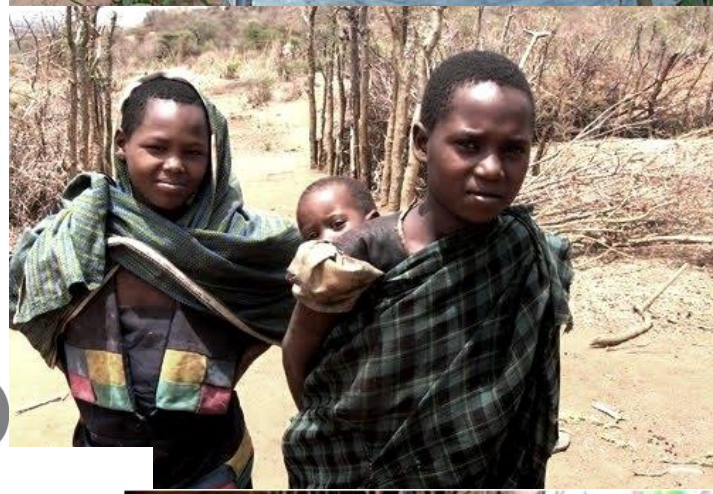
Kosek M et al. Am J Trop Med Hyg. 2013 Feb 6; 88(2): 390–396

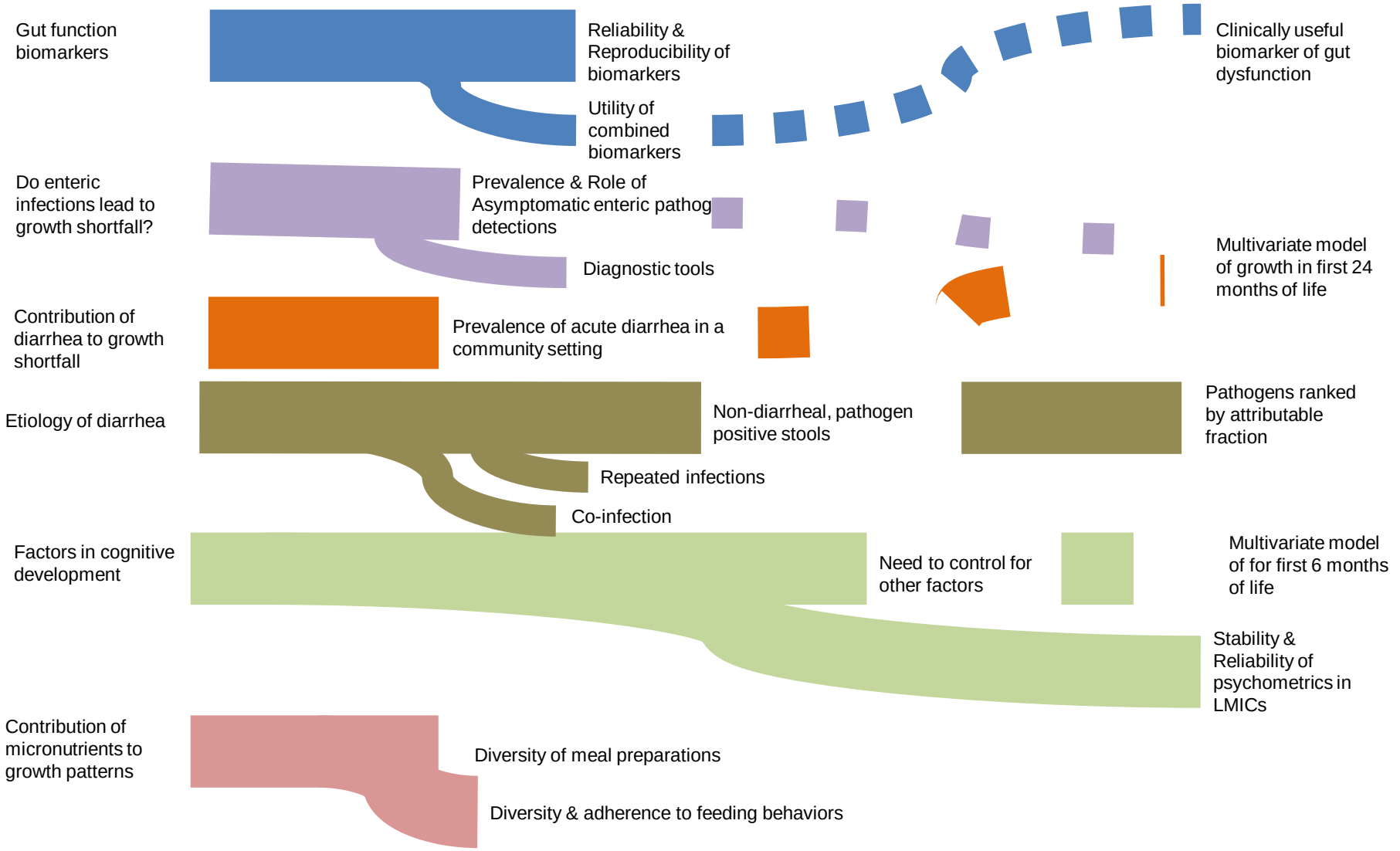
Summary

- MAL-ED study designed to advance biomedical knowledge for public health action/intervention related to nutrition
  - Elucidate attributable causes of under <5 morbidity
  - Define further gaps of knowledge
- Develop datasets that could be utilized for additional add-on future studies
- Increase capacity and development of field sites in resource poor settings for harmonious multi-discipline studies of <5 morbidity
- Develop tools to evaluate disease burden distributions and therefore interventions for public health policy




MAL-ED



Aim **Learning through Experience** **Progress** **Target Product**

dotted, on-going

solid, completed

Objectives

Introduce the purpose of the study SEP general framework and timelines and objectives of health outcomes

introduction to the study participants in the sites and network

understand the strengths and limitations of the data to form conclusions SEP review the processes of quality assurance quality control

demonstrate the ability to make conclusions and studies based on the individual the site in the overall network type data

provided a description of all the sites and descriptive review of the data

demonstrates improve preliminary analyses that have been conducted in the field of microbiology diarrhea growth gut physiology.

present also data in the fields of cognitive development as well as the vaccine response

Overview of primary/secondary endpoints
central hypotheses (re: growth, cognitive development, and immune response);
use of a harmonized protocol
analytical methods (single site, pooled, multisite comparative);
point out differences between GEMS and MAL-ED companion projects

Summary

- Neonatal and maternal factors were found to play a more influential role than postnatal
- factors during early childhood, and their contributions remained significant throughout
- the first 24 months

- · Postnatal exposures, including a higher burden of non-diarrheal enteropathogens, lower socioeconomic status, and lower protein content of the diet became increasingly
- important contributors with age relative to neonatal and maternal factors.

- · Maternal interventions, especially during pregnancy, are likely to have intergenerational effects and a lasting impact on birthweight and child growth outcomes.
- Neonatal and maternal factors were early determinants of lower length-for-age, and their
- contribution remained important throughout the first 24 months of life, whereas the average
- number of enteropathogens in non-diarrheal stools, socioeconomic status, and dietary
- intake became increasingly important contributors by 24 months relative to neonatal and
- maternal factors.

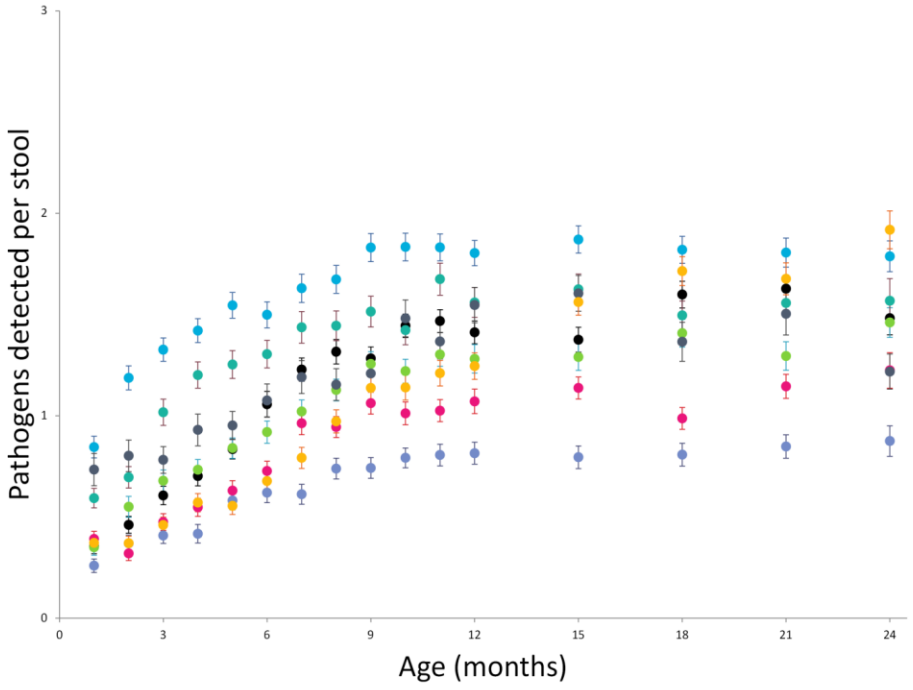
NIH National Institutes of Health
Turning Discovery Into Health

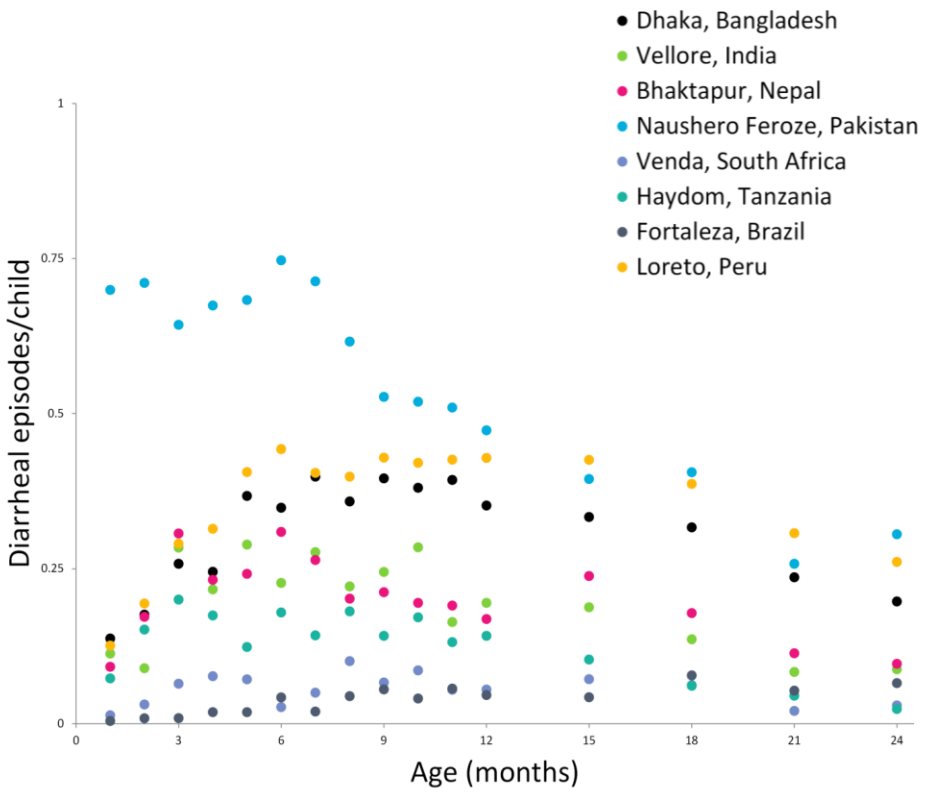
FOGARTY INTERNATIONAL CENTER

MAL-ED cohort completeness of surveillance and testing

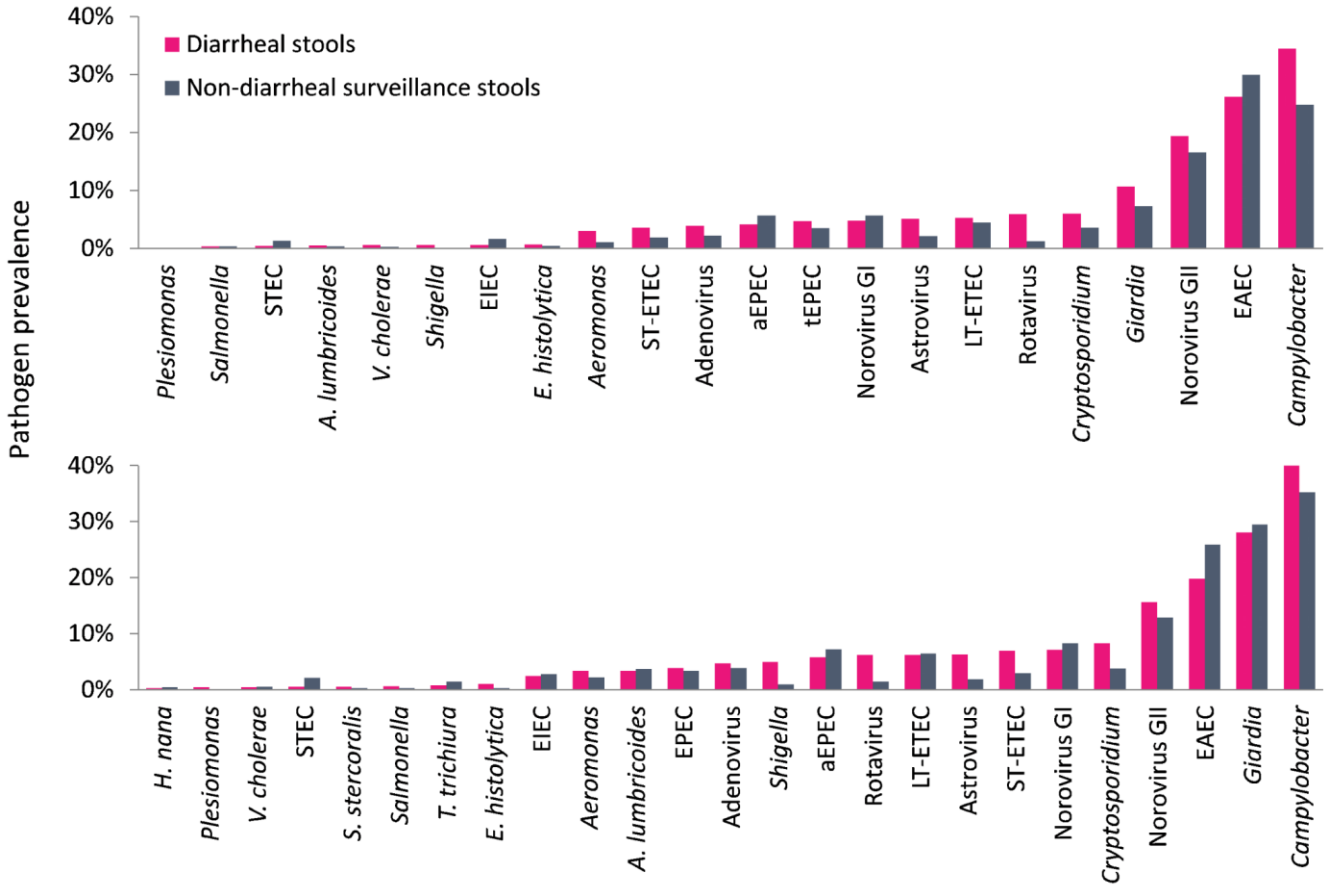
Site	Children enrolled	Diarrhea episodes collected	Diarrheal stools completely tested (%)	Surveillance stools collected	Monthly stools completely tested (%)
BGD	265	1591	1526 (95.9)	2937	2910 (99.1)
INV	251	749	698 (93.2)	3215	3181 (98.9)
NEB	240	976	925 (94.8)	3105	3071 (98.9)
PKN	277	2272	1836 (80.8)	2820	2777 (98.5)
SAV	314	200	157 (78.5)	3720	3617 (97.2)
TZH	262	206	171 (83.0)	3295	3252 (98.7)
BRF	233	129	117 (90.7)	2519	2425 (96.3)
PEL	303	2047	1888 (92.2)	3185	3077 (96.6)
TOTAL	**2145**	**8170**	**7318 (89.6)**	**24796**	**24310 (98.0)**

Pathogen detection and diarrheal episodes per child by age





Pathogen testing results by year of life



Year 1

Year 2

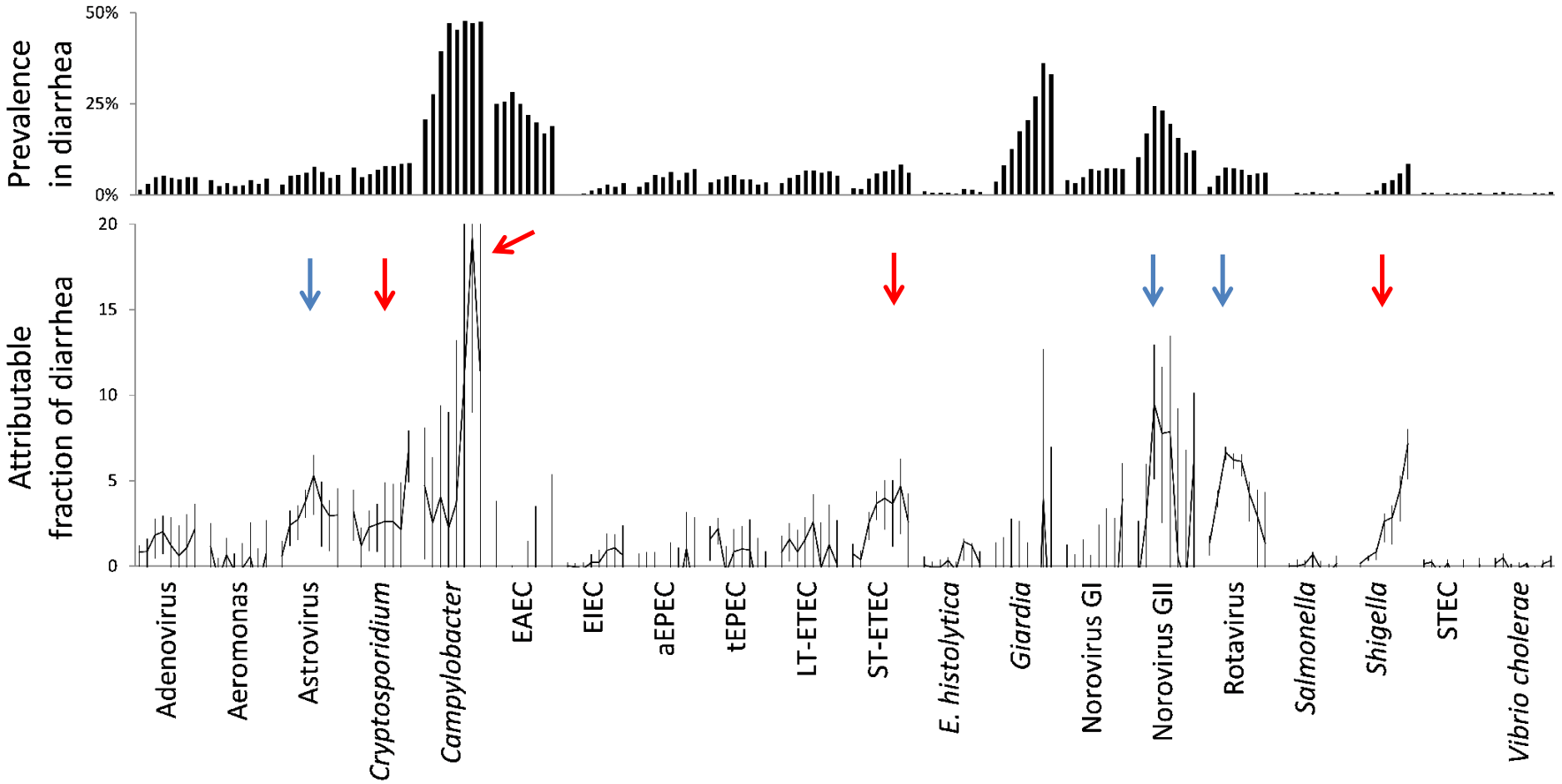
Adjusted AF of diarrhea for the **first** year of life

	BGD	INV	NEB	PKN	SAV	TZH	BRF	PEL	Overall
0-1 years									
# diarrheal	819	419	524	1230	84	145	38	1021	4280
# non-diarrheal	2194	2252	2264	1902	2665	2391	1747	2354	17769
Norovirus GII			8.4			8.2		5.1	5.2
Rotavirus	9.6	6.0	6.6	3.2		9.5		1.0	4.8
Campylobacter					16.9		30.9	5.6	3.5
Astrovirus	2.0	4.2		2.2				3.6	2.7
Cryptosporidium				3.6		6.3	5.5	2.6	2.0
ST-ETEC	4.7	1.7	2.0	1.2	3.3				1.9
Adenovirus		2.7	2.3	1.1				1.5	1.6
tEPEC	2.2								1.3
LT-ETEC	2.0						16.9		1.3
Shigella			0.7	0.9					0.4

Adjusted AF of diarrhea for the **second** year of life

	BGD	INV	NEB	PKN	SAV	TZH	BRF	PEL	Overall
1-2 years									
# diarrheal	707	279	401	606	73	26	79	867	3038
# non-diarrheal	716	929	807	875	952	861	678	723	6541
Campylobacter			8.8					9.9	7.9
Norovirus GII			11.2		19.2			11.7	5.4
Rotavirus	6.0	4.8	8.7	2.2		14.3	4.3	2.9	4.9
Astrovirus	2.6	3.1	4.6			9.7	4.7	7.4	4.2
Shigella	1.5	9.4	6.8	5.1			3.7	2.1	4.0
ST-ETEC	8.0	5.4	4.6			9.1		2.0	3.9
Crypto	2.5	6.9	3.2	5.5		13			3.8
LT-ETEC	2.4					16.1			1.2
Adenovirus		3.6	3.9				3.8		0.9
EIEC			1.2						0.8
E. histolytica		0.7		0.8					0.7
Salmonella		0.7	0.5					0.5	0.3
Norovirus GI			1.0						
Aeromonas								1.0	
Plesiomonas		0.7							
STEC								0.2	

Prevalence and adjusted AF of diarrhea for 3-month intervals



Adjusted AF for specific diarrheal syndromes (1-2 years)

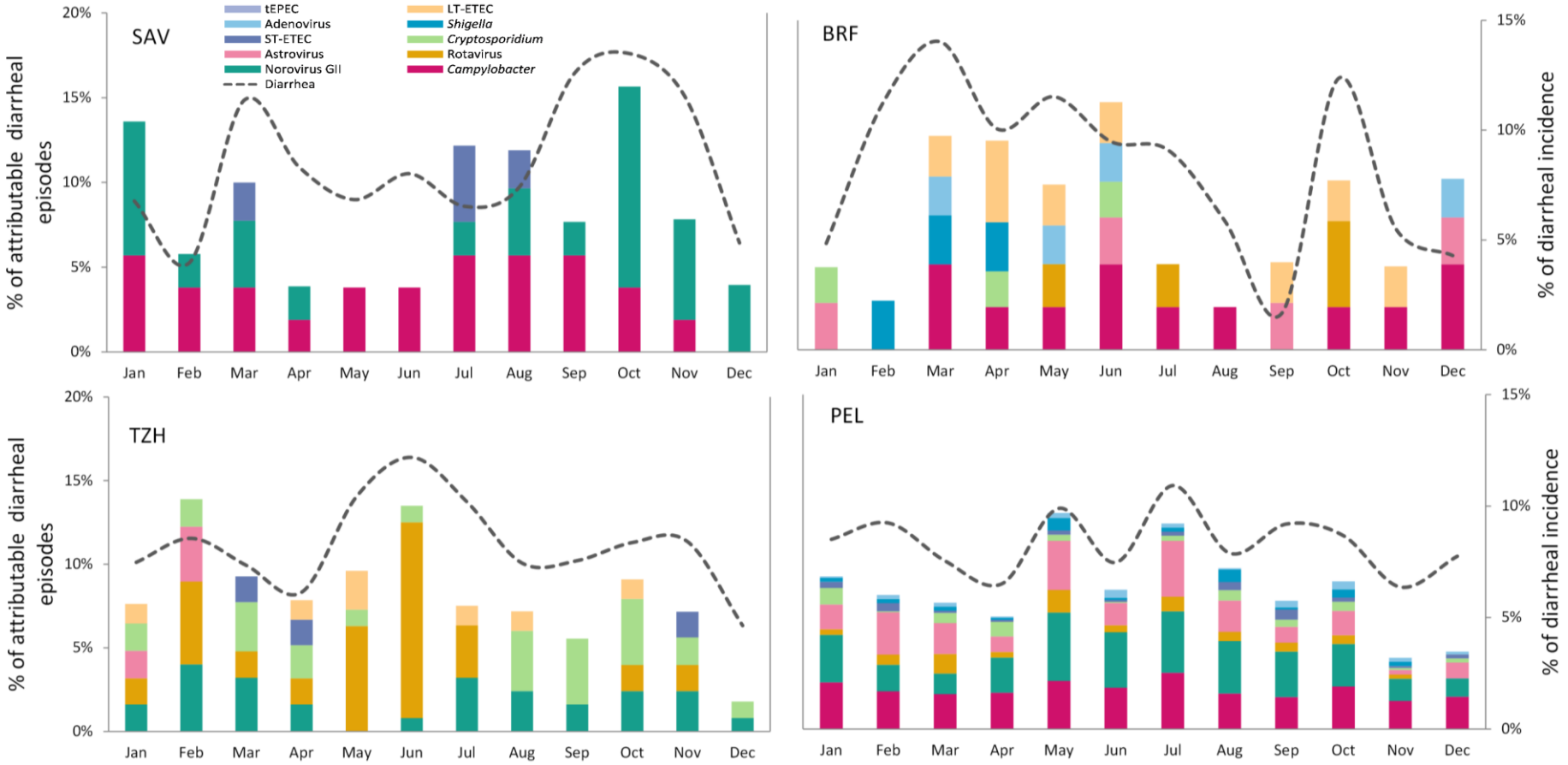
	Acute (<7 days)	Prolonged (≥7 days)	Mild (score 1-3)	Moderate (score 4-6)	Severe (score >6)	Blood in stool	Associated fever	Associated vomiting	Overall
1-2 years of age									
# diarrheal stools	2568	470	1553	1104	381	159	142	698	3038
% of diarrhea	84.5	15.5	51.1	36.3	12.5	5.2	4.7	23.0	N/A
Campylobacter	8.9		9.7	8.3					7.9
Norovirus GII	5.1	6.9	4.5	6.2	6.9			8.9	5.4
Rotavirus	5.2	2.9	3.8	5.1	7.9		4.9	10.0	4.9
Astrovirus	4.5	2.3	4.1	4.7	2.8		5.4	4.5	4.2
Shigella	3.4	7.0	2.7	5.1	5.7	17.0	6.9	3.1	4.0
ST-ETEC	3.6	5.5	3.4	3.9	5.8		3.6	5.5	3.9
Cryptosporidium	3.4	6.1	3.0	4.5	3.2			3.8	3.8
LT-ETEC	1.3			1.5			5.0	2.2	1.2
Adenovirus	1.0		0.8	1.9				1.9	0.9
EIEC	0.8		0.9	1.2		5.0			0.8
E. histolytica	0.7		1.1						0.7
Salmonella	0.4		0.4				1.8		0.3
Aeromonas						3.3			
Plesiomonas						1.2			

Adjusted AFs associated with hospitalization, dehydration or dysentery

	0-1 years of age		1-2 years of age
# stools	587		443
% of all diarrhea	13.7		14.6
Rotavirus	10.6	*Shigella*	10.0
Norovirus GII	8.1	Rotavirus	5.7
Campylobacter	7.9	ST-ETEC	5.1
Cryptosporidium	4.1	*Cryptosporidium*	3.5
Astrovirus	3.5	EIEC	2.5
Adenovirus	2.5	Astrovirus	2.3
LT-ETEC	1.8		
ST-ETEC	1.5		
Shigella	1.3		

Seasonal pathogen-specific attributability of diarrhea

Seasonal pathogen-specific attributability of diarrhea



Conclusions

Detection of enteropathogens in the absence of diarrheal symptoms was common

There was substantial heterogeneity in pathogen-specific burdens of diarrhea, with important determinants including age, geography, season, and symptomatology

We observed an unexpectedly high burden of disease due to *Campylobacter*, norovirus GII, and astrovirus

A preliminary association with linear growth shortfalls is seen for *Campylobacter*, EAEC, *Giardia*, and *Cryptosporidium*