Estimating the Burden of Typhoid: The Role of Novel Techniques

Michael Owusu, Sampson Twumasi-Ankrah, Michael Owusu-Ansah, Eric Darko, Christopher B. Uzzell, Jonathan Rigby, Catherine M. Troman, Nicolette A. Zhou, John Scott Meschke, Nicholas C. Grassly, Yaw Adu-Sarkodie, Ellis Owusu-Dabo

Overview of presentation

- Background
- Objectives
- Methods
- Results
- Conclusion

Global Estimates of Typhoid Burden

- Globally, 14·3 million (95% uncertainty interval [UI] 12·5–16·3) cases of typhoid and paratyphoid fevers occurred in 2017

- The highest disease burden regions are sub-Saharan Africa and South Asia.

- In Asia, a recent study conducted in Bangladesh, Nepal, and Pakistan reported high adjusted incidence of disease exceeding 100 per 100,000 person-years of observation (PYO) in all three countries.

Global Estimates of Typhoid Burden

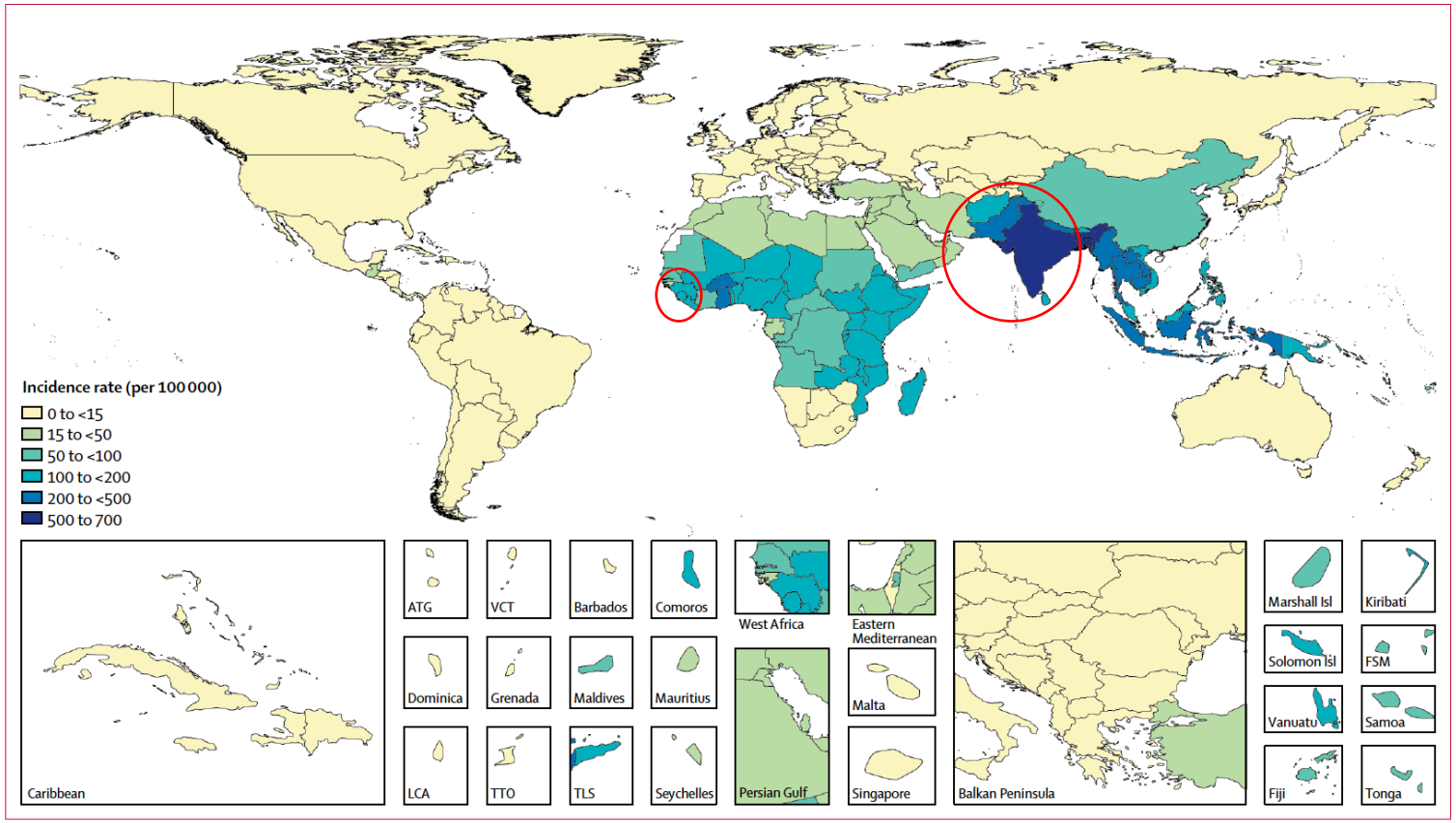


Figure 1: **Incidence rates (per 100 000) of typhoid and paratyphoid fevers, by country, in 2017**
Unfilled locations are those for which GBD does not produce estimates. The inset maps detail smaller locations. ATG=Antigua and Barbuda. FSM=Federated States of Micronesia. GBD=Global Burden of Diseases, Injuries, and Risk Factors Study. Isl=Islands. LCA=Saint Lucia. TLS=Timor-Leste. TTO=Trinidad and Tobago. VCT=Saint Vincent and the Grenadines.

The global burden of typhoid and paratyphoid fevers: a systematic analysis for the Global Burden of Disease Study 2017

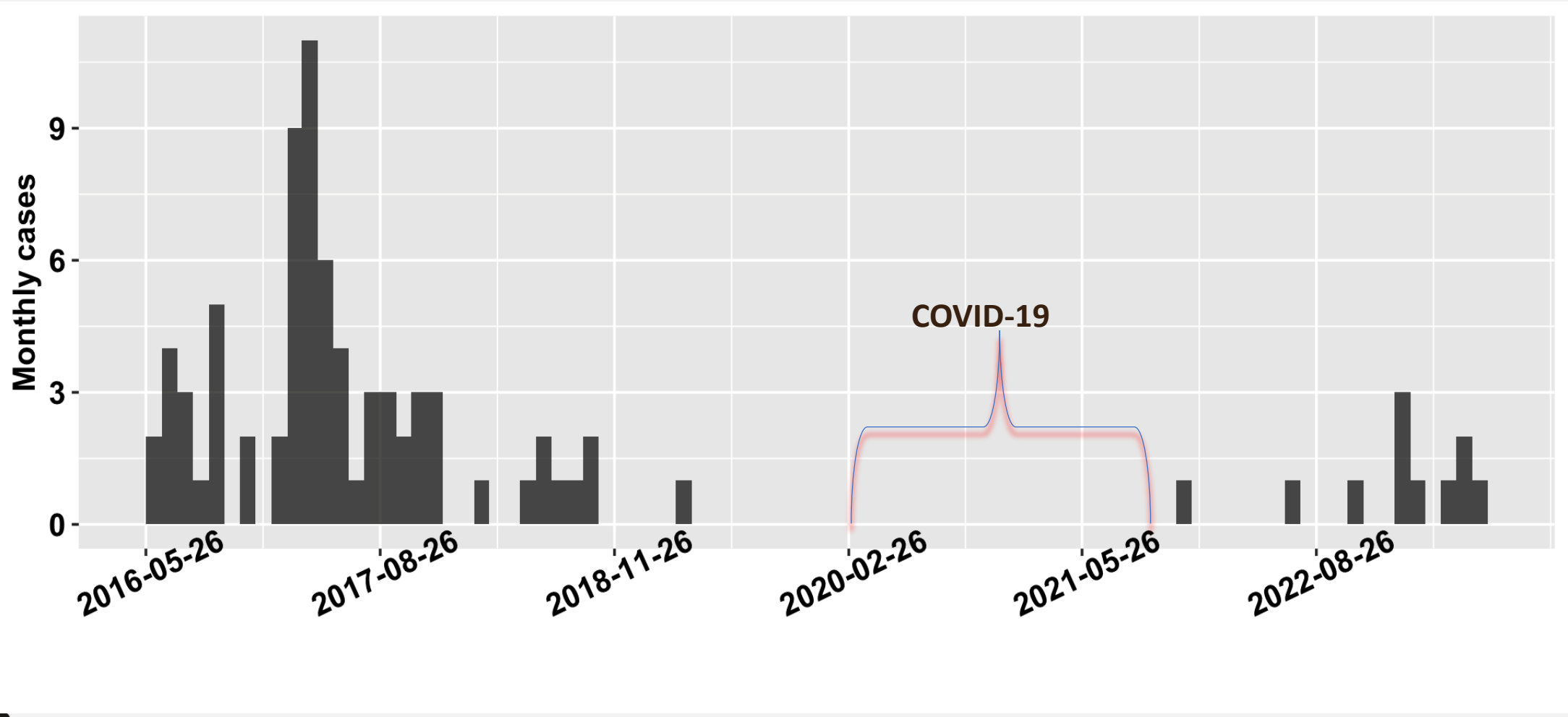
*GBD 2017 Typhoid and Paratyphoid Collaborators**

Lancet Infect Dis 2019; 19: 369–81

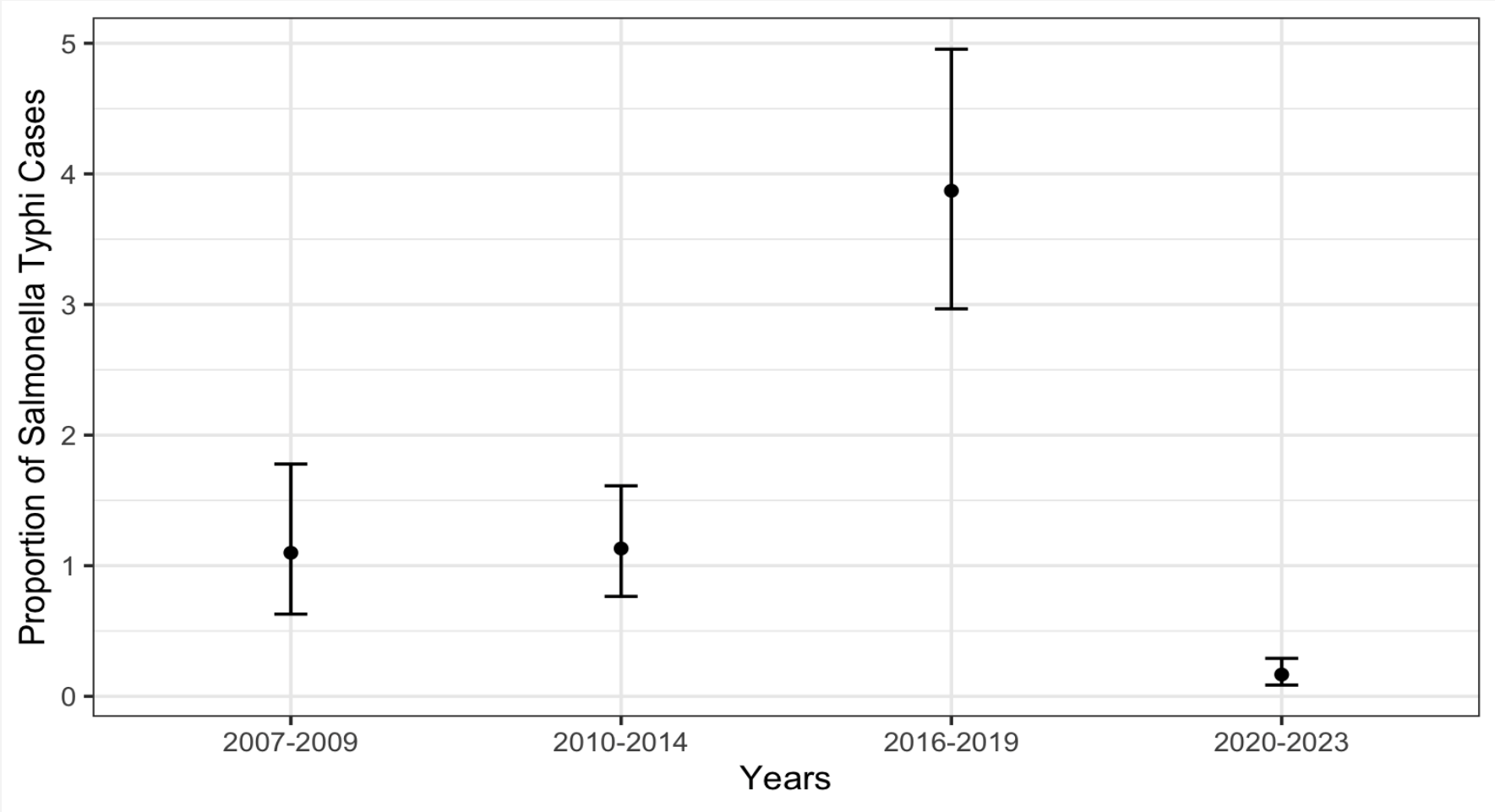
WHO THRESHOLD FOR TYPHOID
100 cases per 100,000 PYO

Bangladesh,
Nepal, and
Pakistan
Ghana
Burkina Faso

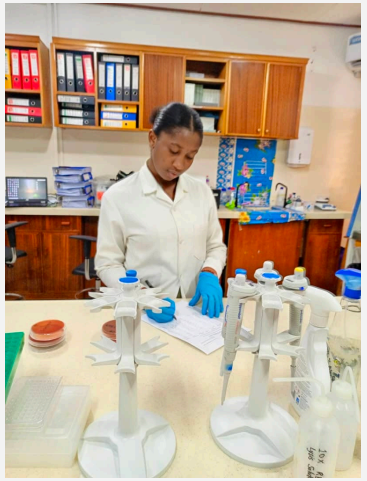
Estimates of typhoid cases in Ghana

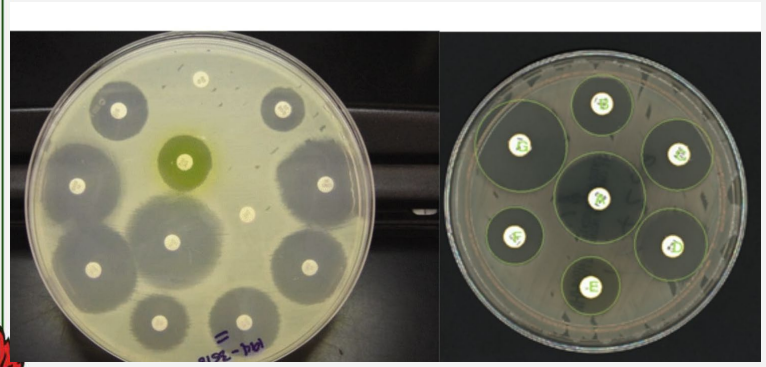


Trend of typhoid cases

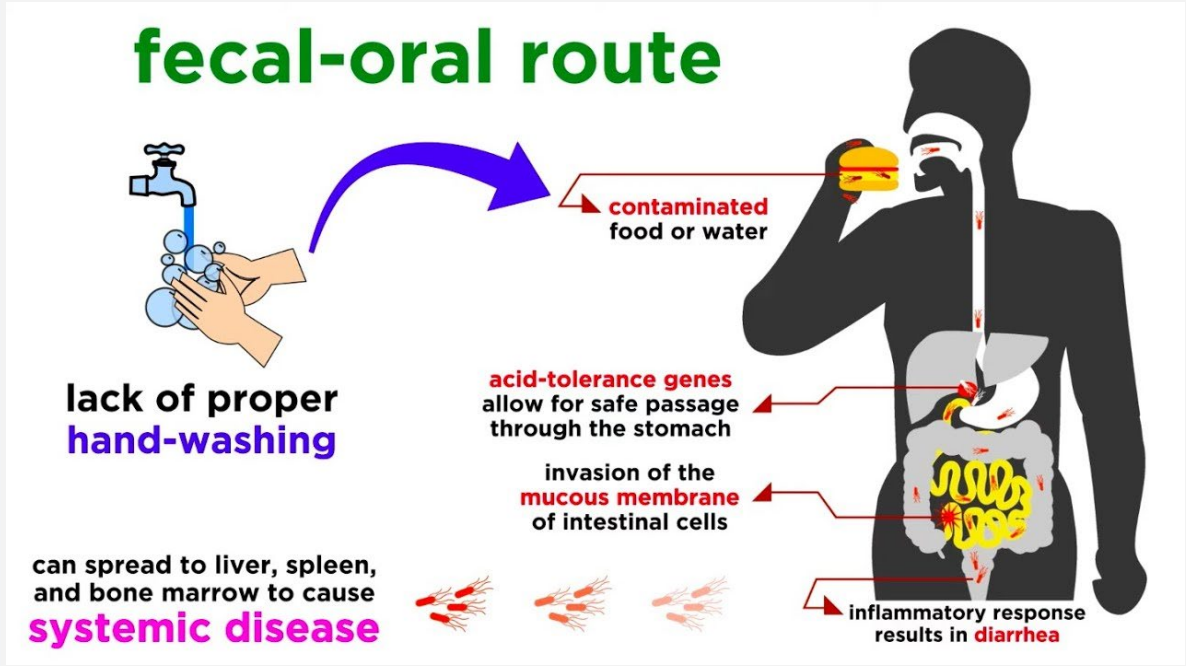


Standard methods for typhoid surveillance





The need for additional tools for S.Typhi detection



Prof Davis, lecture series, 2020

The Journal of Infectious Diseases
MAJOR ARTICLE

Environmental Surveillance for *Salmonella* Typhi and its Association With Typhoid Fever Incidence in India and Malawi

Christopher B. Uzzell,[1,a] Dilip Abraham,[2,a] Jonathan Rigby,[3,a] Catherine M. Troman,[1,a] Satheesh Nair,[4] Nicola Elviss,[5] Lalithambigai Kathiresan,[6] Rajan Srinivasan,[6] Veeraraghavan Balaji,[7] Nicolette A. Zhou,[8] John Scott Meschke,[8,©] Jacob John,[6] Gagandeep Kang,[2,©] Nicholas Feasey,[3,9] Venkata Raghava Mohan,[6] and Nicholas C. Grassly[1,©]

Project objectives

- Implement typhoid ES with partners at sites conducting blood-culture surveillance over a 12-month period

- Measure ES site characteristics and investigation of their association with detection of S. Typhi and human-restricted or other control organisms that indicate faecal contamination

- Investigate whether S. Typhi load and genetic diversity correlate with disease incidence rates in the local population

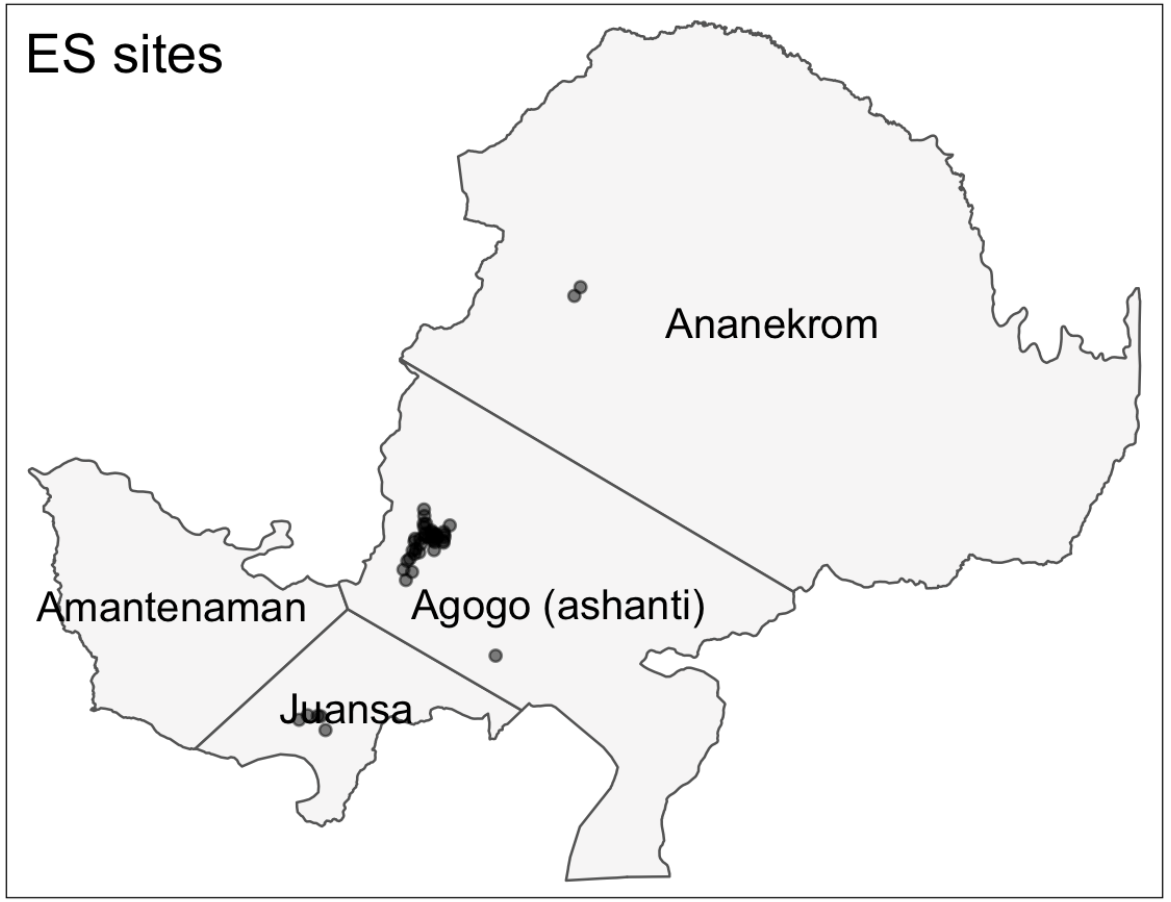
Study site

- Studies on malaria
- Studies on Typhoid
- Fever without source

Catchment population
140,000

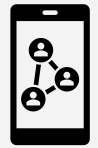
ES sites

	Number of Sites
AGOGO	37
ANANEKROM	2
DOMEABRA	2
HWIDIEM	1
JUANSA	3
MAGYEDA	1
Sum	46



Study procedures

- **43 sites mapped out at the study site**
- **Monthly sampling (12 months)**



ES site selection, characterization & validation

↓

sample collection (grab or trap samples)

↓

sample processing & DNA extraction

↓

qPCR for *Salmonella* and *S*. Typhi targets (ttr, staG, tviB)

Typhoid ES methods: https://www.medrxiv.org/content/10.1101/2021.05.21.21257547v1
Protocols and community at: https://www.protocols.io/workspaces/typhoides

Target genes for *Salmonella* Typhi detection

A	B
Primer/Probe Name	**Sequence (5' -3')**
staG_F	CGC GAA GTC AGA GTC GAC ATA G
staG_R	AAG ACC TCA ACG CCG ATC AC
staG_P	[TAMRA] - CA TTT GTT CTG GAG CAG GCT GAC GG - [BHQ2]
ttr_F	CTC ACC AGG AGA TTA CAA CAT GG
ttr_R	AGC TCA GAC CAA AAG TGA CCA TC
ttr_P	[FAM] - CA CCG ACG GCG AGA CCG ACT TT - [BHQ1]
tviB_F	TGT GGT AAA GGA ACT CGG TAA A
tviB_R	GAC TTC CGA TAC CG GAT AAT G
tviB_P	[JOE] - TG GAT GCC GAA GAG GTA AGA CGA GA - [BHQ1]
HF183_F	ATC ATG AGT TCA CAT GTC CG
HF183_R	CTT CCT CTC AGA ACC CCT ATC C
HF183_P	[FAM] - CT AAT GGA ACG CAT CCC - [BHQ1]

Detections of all targets (staG, ttr, tvib) is considered positive

Field Training and community engagements

Training in protocol development

Field engagements in study implementation

Training by ICL and others

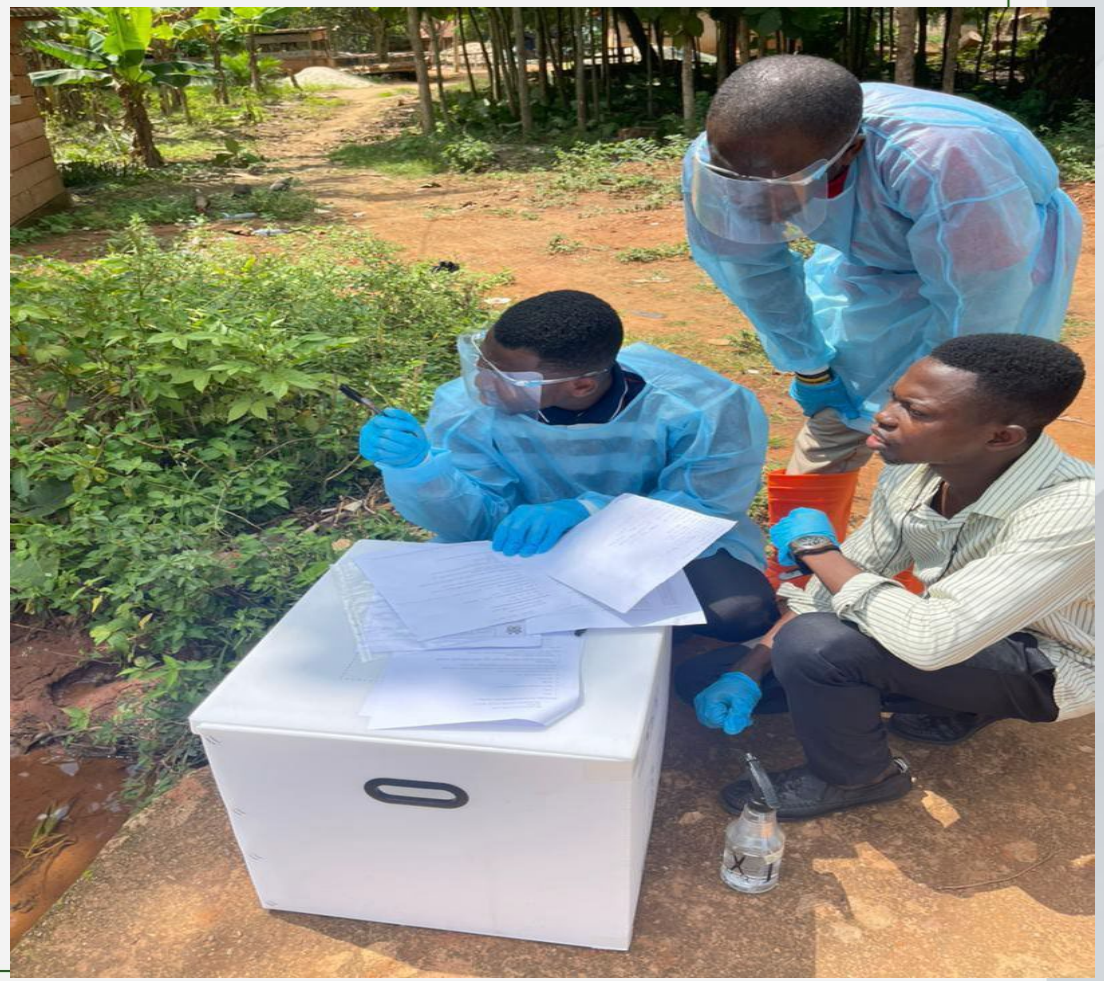
Field training

Moore swabs preparation

Validation of sites

Deployment of Moore swabs/Grab samples

Cold chain for sample transport



Data collection

❖ **Questionnaire for environmental site sample collection**

- Date &time
- Site ID
- GPSaddress
- Flow speed
- Depth of wastewater
- Width of wastewater
- MS ID, deployment duration,
- Water quality measurements (Temp, pH, DO, EC, TDS, ORP, SAL, SSG)

Data capture

- Paper and tablets were used in capturing data
- Data were entered in REDCap
- Field and lab data were cleaned merged

Laboratory Processing and Testing

Grab

1L No enrichment

↓

Filter (0.45uM membrane)

↓

Elute by massaging in 10m RL

↓

Centrifuge and store pellets at -20

Moore Swab

450 ml pre-enrich in UPE

↓

Incubate overnight at 37^0C

↓

Pipette 2 aliquotes of 20ml

↓

Filter (0.45uM membrane)

↓

Cut filters and place in powerbead tube

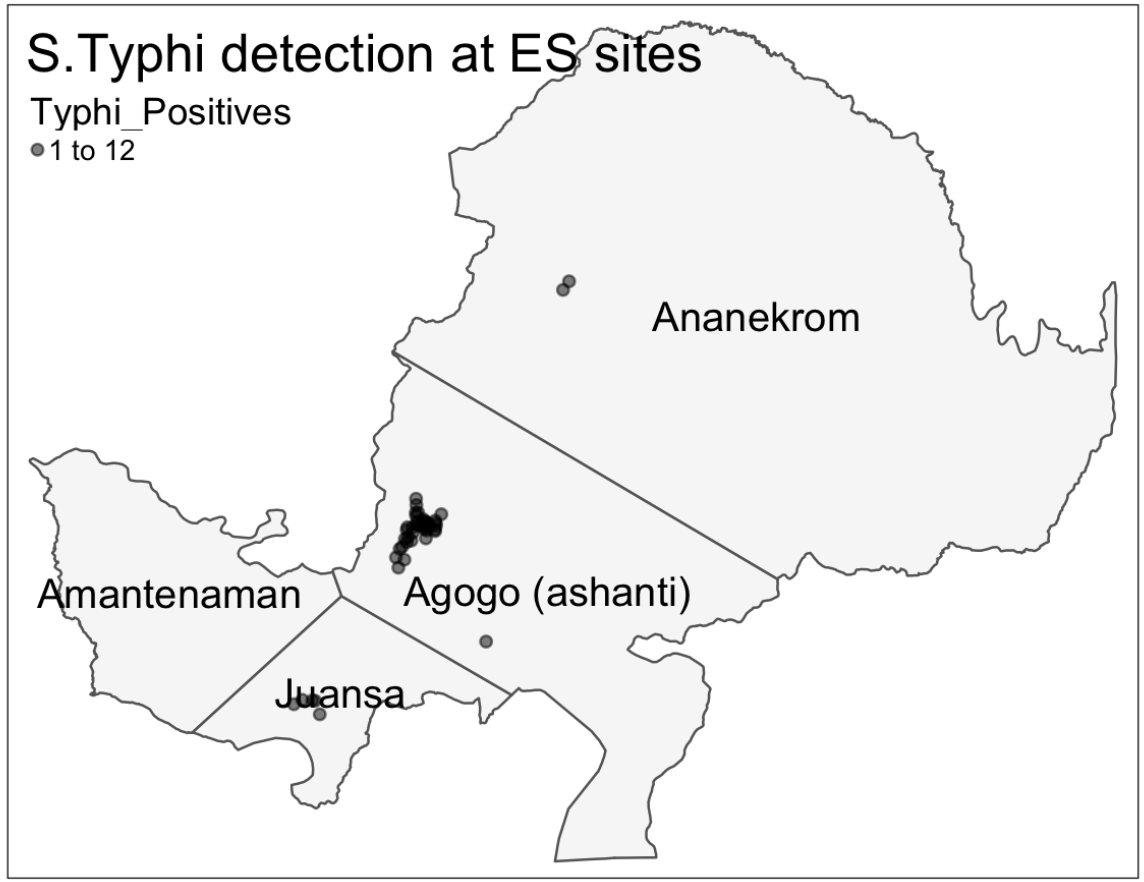
↓

Centrifuge and store pellets at -20^0C

Detection of S.Typhi at ES sites

Detection of *Salmonella* Typhi

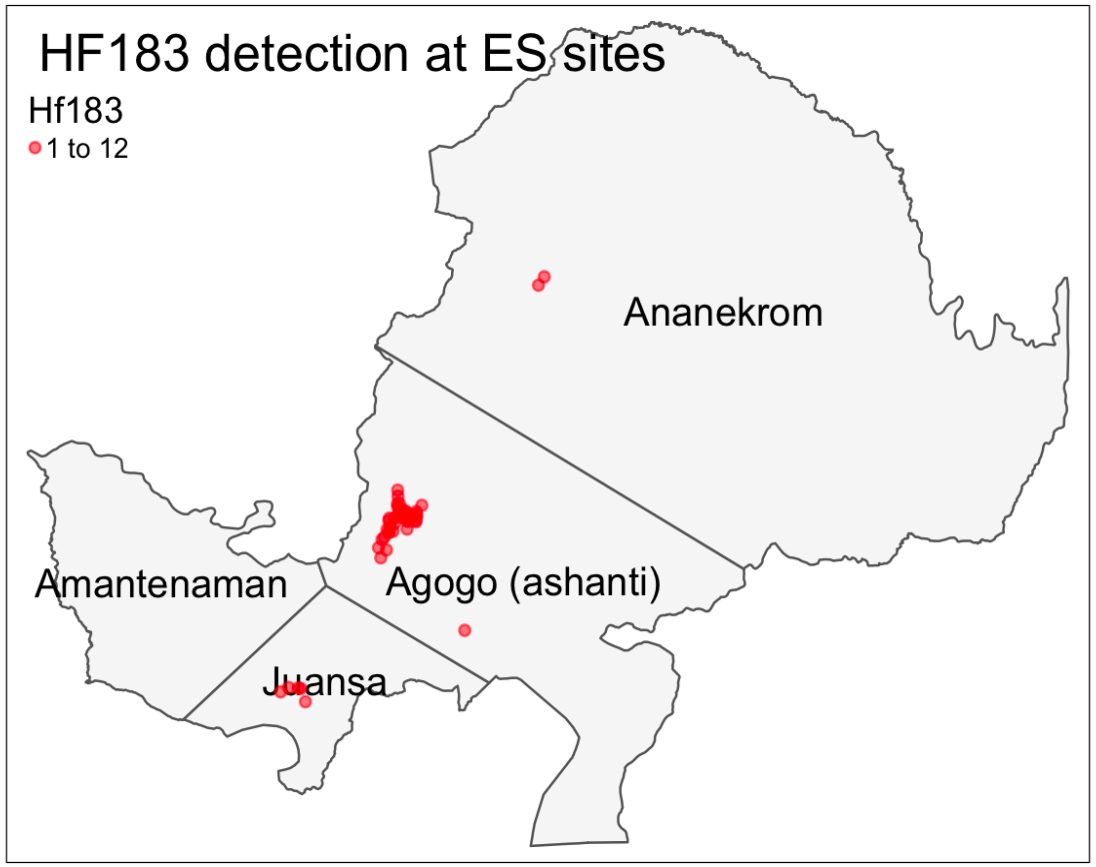
	Typhoid Positives	Total
AGOGO	201 (31.7%)	633
ANANEKROM	2 (11.1%)	18
DOMEABRA	3 (15.7%)	19
HWIDIEM	8 (44%)	18
JUANSA	16 (30%)	53
MAGYEDA	0 (0)	14
Sum	**230**	**755**



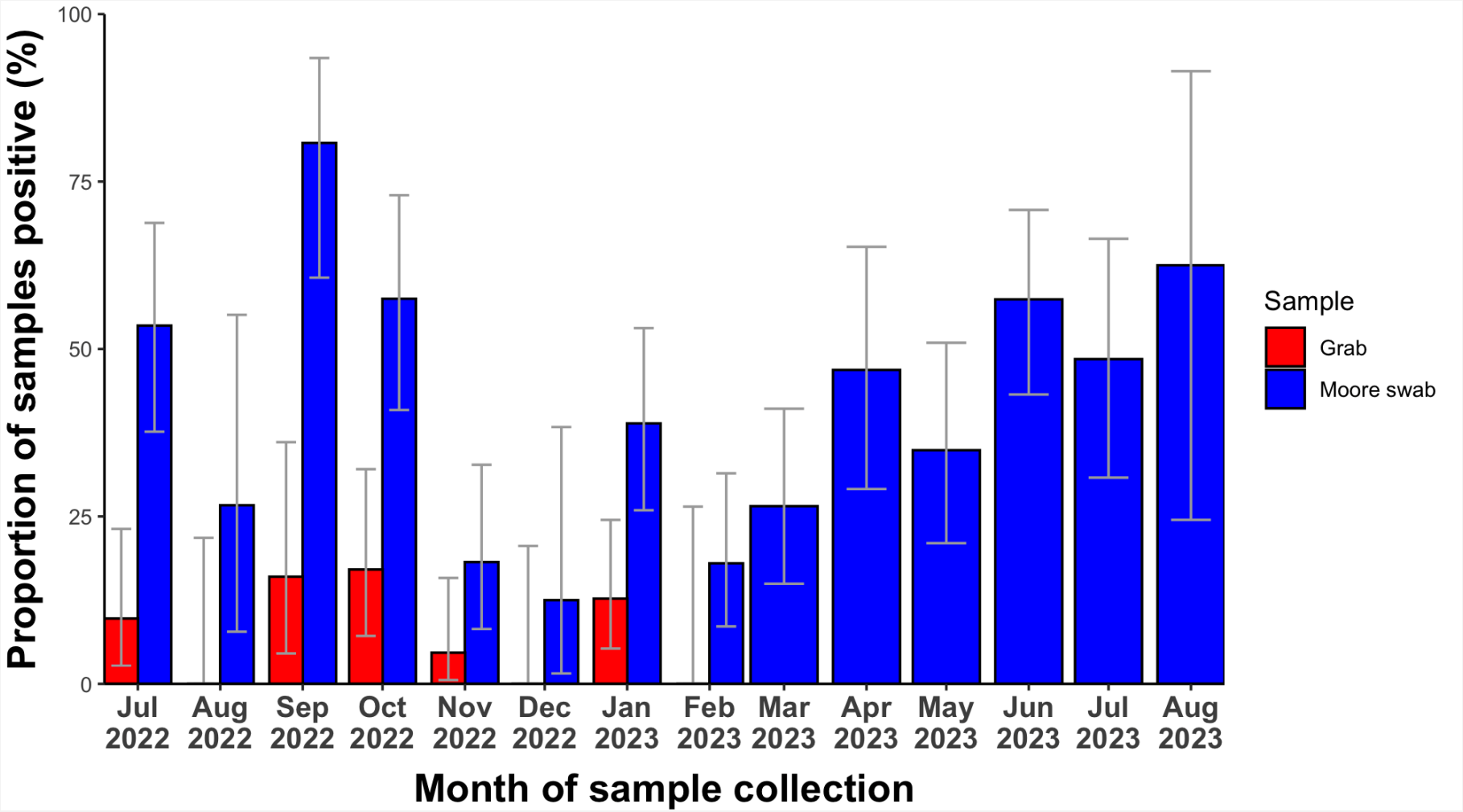
Detection of faecal contamination marker at ES sites

Distribution of Typhoid Cases

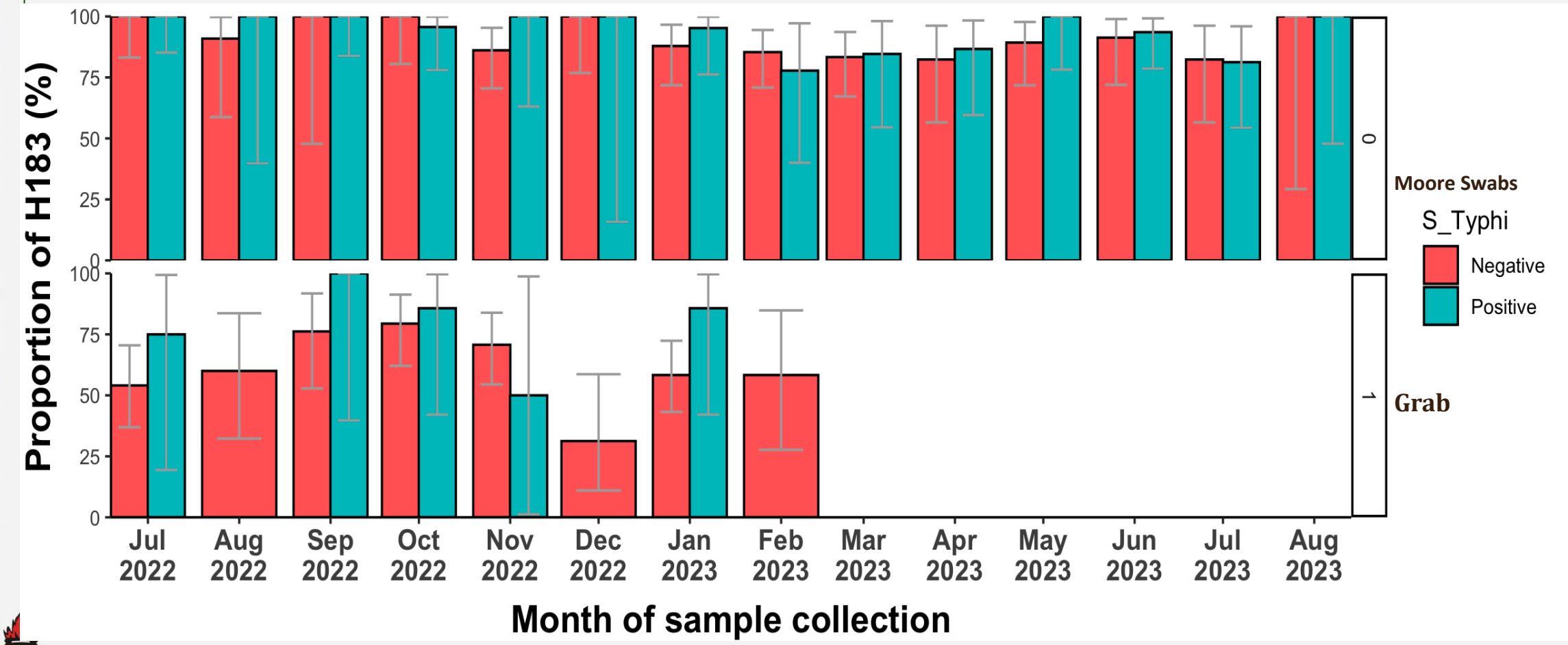
	HF183 Positives	Total
AGOGO	526 (83.1%)	633
ANANEKROM	13 (72.2%)	18
DOMEABRA	17 (89.4%)	19
HWIDIEM	11 (61.1%)	18
JUANSA	43 (81.1%)	53
MAGYEDA	12 (14.3%)	14
Sum	**622**	**755**



Salmonella Typhi detections in Grab and Moore swabs



Faecal contamination and S.Typhi detections



Predictors of Salmonella Typhi detections in wastewater

Parameters	Odds Ratio	P-value	Lower 95% CI	Upper 95%CI
Method [Moore Swab]	4.43	<0.0001	2.59	7.58
HF183 [Positive]	3.94	0.007	1.47	10.60
pH [High]	1.97	0.006	1.22	3.19
Dissolved_oxygen	1.45	0.109	0.92	2.30
Electrical_conductivity	0.84	0.359	0.58	1.22
Salinity	1.41	0.048	1.00	1.97
Seawater_specific_gravity	1.01	0.905	0.83	1.23

Conclusion

- Moore swab samples have better yield for *Salmonella* Typhi detection compared to Grab samples
- pH level and level of faecal contamination is associated with S.Typhi detection in environmental samples. However more analysis is needed to provide additional evidence
- While we have observed high number of S.Typhi in the environment, the incidence of blood culture Salmonella detections is low. We plan to sequence the environmental S.Typhi detected in order to further confirm this.

Thanks to the study team and collaborators

Acknowledgement

Imperial College London, UK
Chris Uzzell, Catherine Troman, Sam Horsfield, Alex Shaw, Nick Grassly

LSTM, Malawi
Nick Feasey, Jon Rigby

Kwame Nkrumah University of Science & Technology, Ghana
Michael Owusu, Michael Owusu-Ansah, Sampson, Twumasi-Ankrah, Eric Darko, Yaw Adu-Sarkodie, Ellis Owusu-Dabo
** All field and data team

University of Washington
Scott Meschke, Nicolette Zhou, Jeff Shirai

University of Ibadan, Nigeria
Aderemi Kehinde and Team

Collaborators:
Public Health England
Satheesh Nair, Nicola Elviss

International Vaccine Initiative (IVI)/TyFIVE team, Fiji
Florian Marks, Raphael Zellweger

Funding and guidance:

EDD team at BMGF
Supriya Kumar, Simon Harris, Duncan Steele

THANK YOU

Kwame Nkrumah University of Science and Technology, Kumasi | Leaders In Change

Visit us at www.knust.edu.gh

uro@knust.edu.gh | Follow KNUST on: