



Better lives through livestock

Wastewater Surveillance

Pandemic Preparedness

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Viral Pathogen Genome Sequencing and Bioinformatics
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Background

- Wastewater Surveillance (Wastewater-Based Epidemiology (WBE))
 - **Global Grand Challenges**
 - Climate change/Global population growth and, transboundary movement/increased wildlife/animal-human interactions induce:
 - Re-emergence and emergence of pathogenic infections
 - Surveillance is therefore crucial for early detection, inform prevention and prompt response to future pandemics.
 - Global Antimicrobial Resistance (AMR) crisis need robust tool for tracking and reporting
 - **WBE**-measure, or estimate the occurrence, spread, or resurgence of disease in a community (population).
 - Prevalence of pathogens circulating within a community (overall health of a community)
 - Detect drug use, or other biological or chemical indicators in a population
 - WBE a powerful and cost-effective tool for tackling AMR (less costly compared to clinical surveillance/diagnosis),
 - WBE surveillance of pathogens, zoonoses, in wastewater from populations, livestock farms, wet markets and their surrounding areas is of utmost importance.

Background

Pandemic Preparedness in Africa

Grand Challenges

- The COVID-19 pandemic
 - coordination and collaboration on surveillance and epidemic intelligence challenges
 - » poor and led to ineffective and inefficient decision-making.
- Countries at different stages of building their data collection, analysis, and broader epidemic intelligence capabilities.
- Countries generate data but potential for meaningful analysis remains limited as the data are collected in ad hoc ways, with differing standards that are not sufficiently connected to answer complex questions.
- channels of coordination for countries to learn from each other and produce global models, or share national insights limited.
- Development of public health surveillance and intelligence systems on account of limited capacities and scarce resources.
- *The sharing of data can facilitate timely response, robust research, and overall, better-informed policy outcomes.*

Background

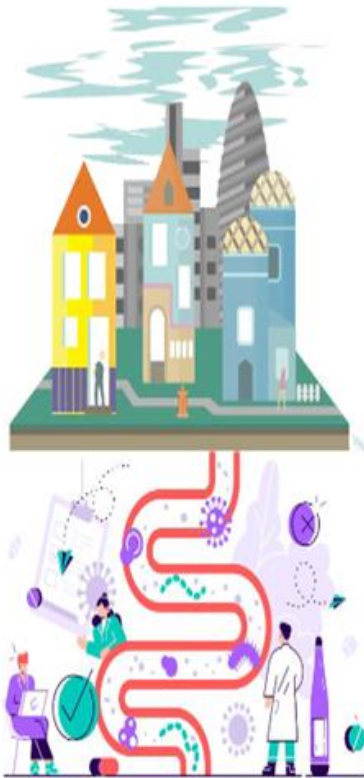
Pandemic Preparedness in Africa

Preparedness and Resilience for Emerging Threats (PRET) initiative (WHO), Africa CDC (PGI)



WBE Strategy and One Health

Gut of a City



Populations (Pool of Guts)



Strategy



Wastewater/Sewage/Sludge

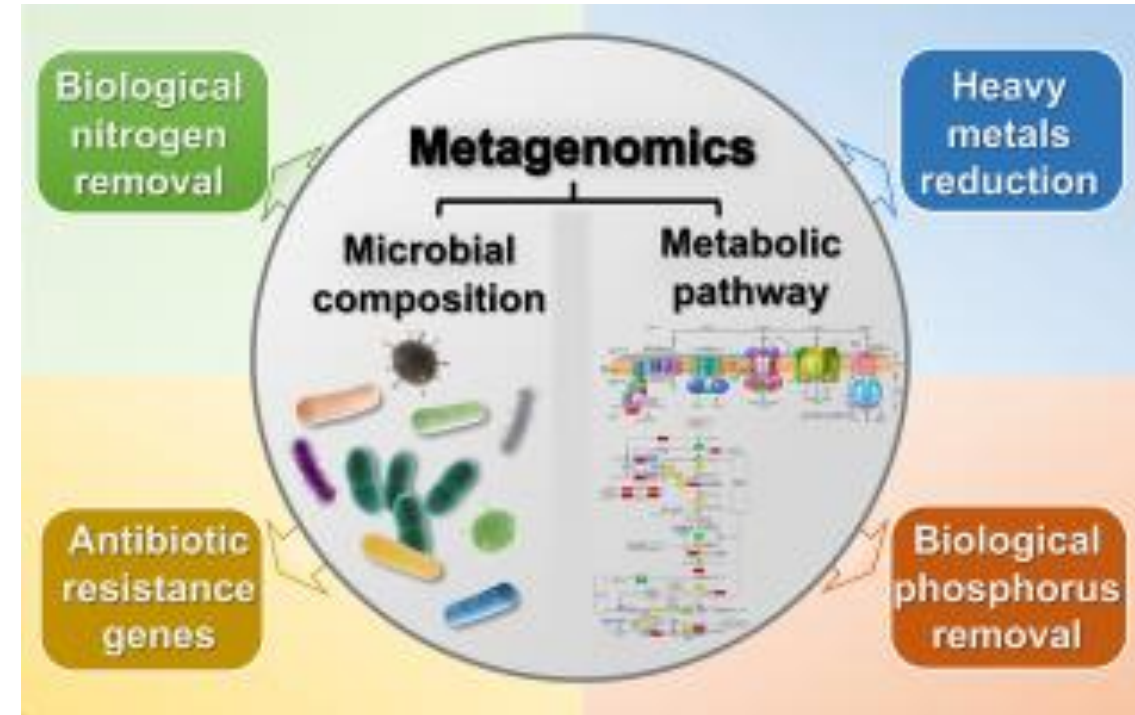
- Is an indicator of consumption, exposure and health status of a population

Contains

- Excretion from human/stool/feces/pathogen/drugs (Human)
- Waste from Slaughterhouses (Animal)
- Environmental and agricultural input (Environment)

Extracting Wastewater Surveillance Insight

- Next Generation Sequencing
 - Apply metagenomics sequencing
 - Estimation of total genetic material in a microbiome
 - Analysis of the microbial and the genetic diversity,
 - metabolic processes
 - Microbiome `surveillance in Wastewater
- Elucidate microbiome composition
 - Identify pathogenic species abundance/diversity
 - PAOs/GAOs/Denitrification species and diversity
- Characterize resistome
 - Antibiotic resistance bacteria
 - Antibiotic resistance genes/associated class of antibiotics/pathogens
- Relevance of findings to public health interventions
 - Early warning system/diagnosis/inform strategic response/action



<https://doi.org/10.1016/j.scitotenv.2021.150737>.

Metagenomics and amplified based sequencing

	Low-throughput amplified sequencing	Metagenomics sequencing
Target segment	16S rRNA gene fragment	Non-targeted, random access to gene fragments in the environment
Function	The species composition and diversity of microbes	The composition, diversity, genome structure, and potential functions of microbial communities
Cost	Lower	Expensive
Data analysis	Simpler analysis of 16S rRNA gene fragment analysis	The analysis is difficult, requires bioinformatics knowledge and an excellent server
Advantages	Simple, fast, and universal	Obtain isolate difficult species metagenome-assembled genome, discover novel functional genes/ enzymes
Disadvantages	PCR is biased, single function	Difficult to assemble, high computational cost, and difficult to find unexpressed genes, complex information analysis

Antibiotic Resistance in Wastewater

- Wastewater plants are recipient sites for resistance genes
- A site for ARGs integration and transfer between microbes (bacteria) (Transformation)
 - Mobile genetic elements (plasmids/phages)
 - producing multiple resistance genes in a bacterium accelerating their spread in the environment
- Thus wastewater metagenomics can help detect and analyze functional gene fragments and provide insight into has formation and distribution of ARGs in wastewater

GENOMIC SURVEILLANCE TO CONTROL PATHOGEN INFECTIONS IN AFRICA (One Health work package)



Herd health

Food safety and AMR

Zoonoses and EID

Vaccines and diagnostics

GENOMICS

One Health component

2023

Specific Objectives and activities

3.1.1: **To develop wastewater-based pathogen genomic epidemiology and surveillance systems**

- ❖ Use metagenomic approaches to analyse wastewater samples
- ❖ Samples collected from epidemiological zones within two cities in Kenya
- ❖ Detect the presence pathogens using genomics
- ❖ Perform genomic characterization of clinically relevant microbes detected in the wastewater
- ❖ Profile antimicrobial genes associated with detected pathogens in wastewater

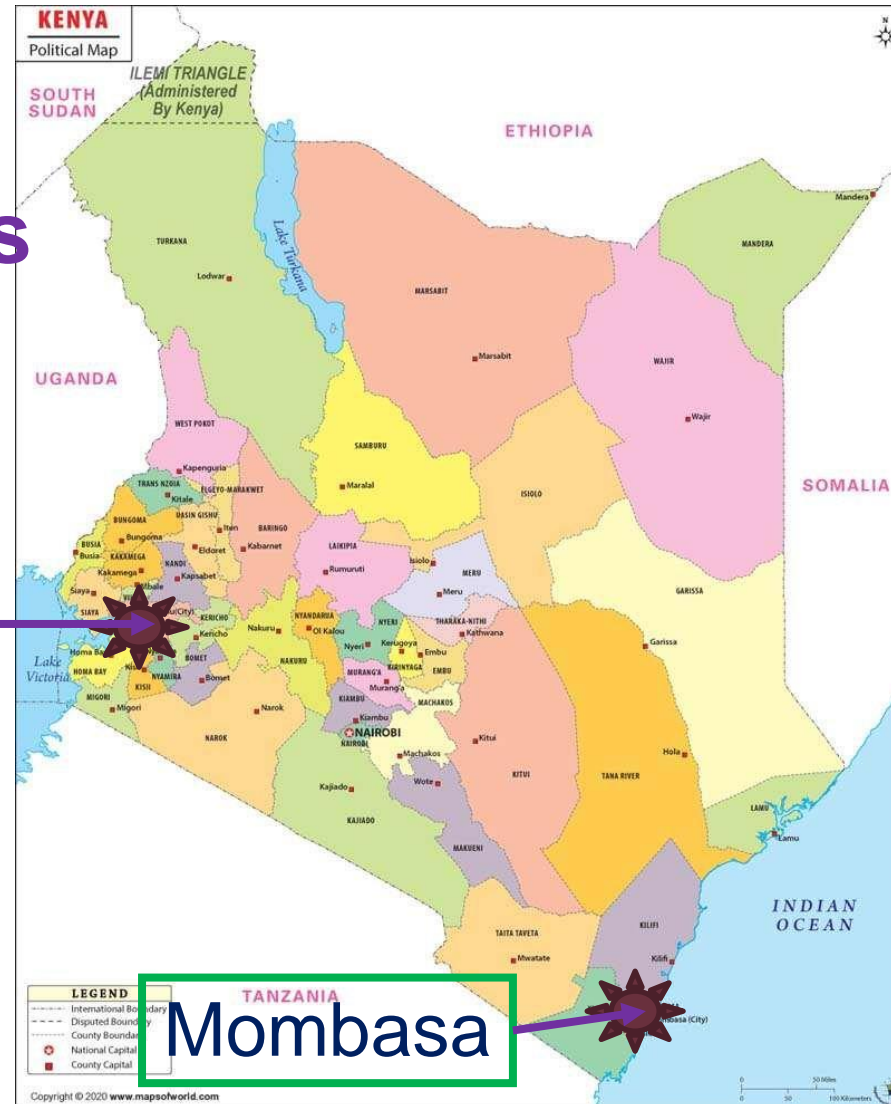


Study Sites

Wastewater study sites Kisumu and Mombasa

Kisumu

Mombasa



Wastewater Genomic epidemiology

Study design & sampling strategy

- ❖ Downstream VS Upstream sampling – upstream to represent localized catchments and downstream to represent composite catchment
- ❖ *Formal VS informal settlements* – representing levels of access to healthcare services, unique risk for AMR
- ❖ *Closed VS open sewer systems* – evaluate influence of environmental contamination
- ❖ Sewer inlet (upstream) and outlet (downstream) into the two water bodies, Lake Victoria and Indian Ocean
- ❖ *Longitudinal sampling – rainy VS dry seasons*



Grab-sampling



Auto-sampling

KISUMU

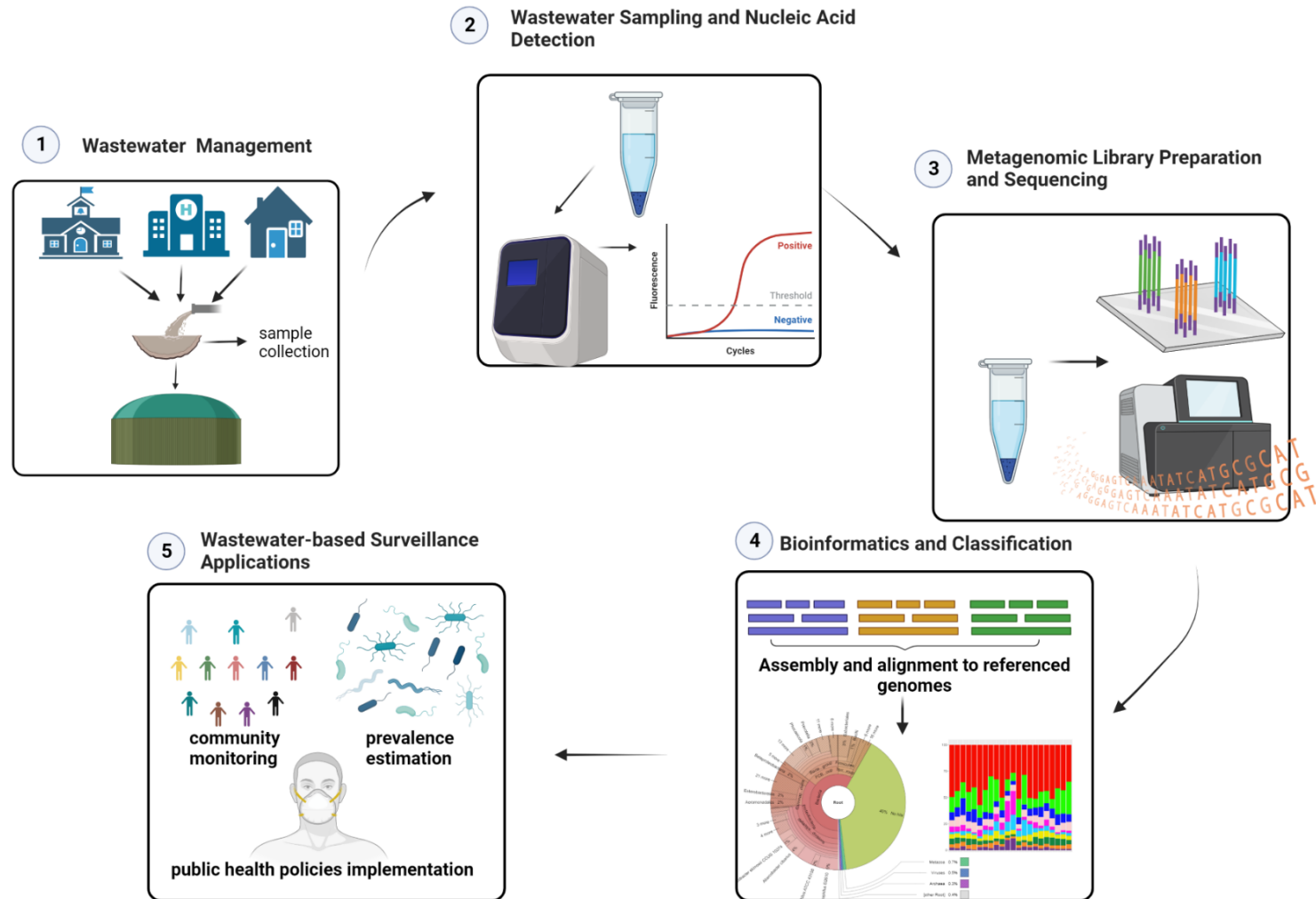
Month	Season	sites	Days/w eek	weeks	Total smples
Oct	Rainy	18	3	2	108
Nov	Rainy	18	3	2	108
Dec	Dry	18	3	2	108
Jan	Dry	18	3	2	108

MOMBASA

Month	Season	sites	Days/ week	weeks	Total smples
Oct	Rainy	12	3	2	72
Nov	Rainy	12	3	2	72
Dec	Dry	12	3	2	72
Jan	Dry	12	3	2	72

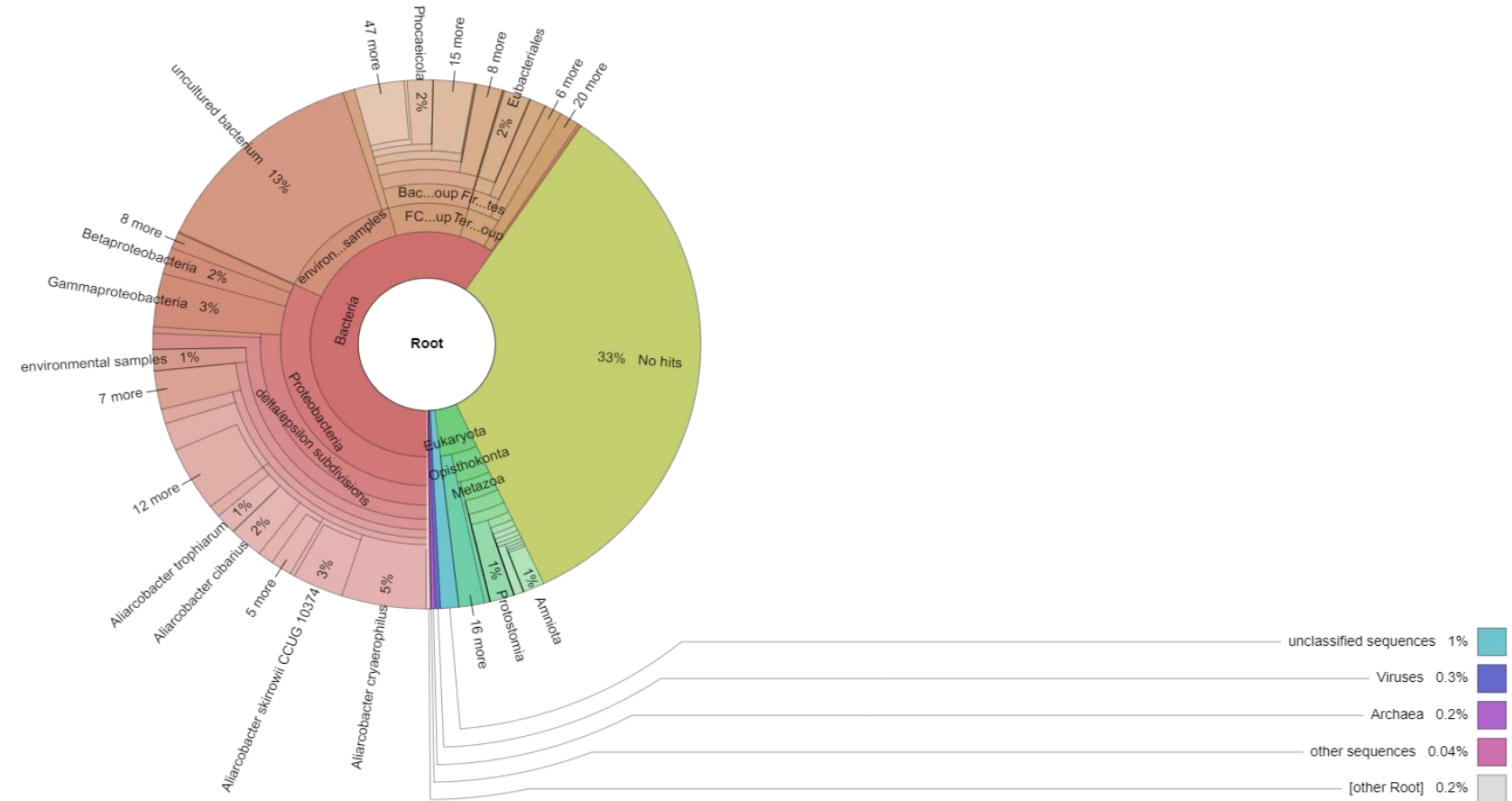
N=720 samples

Wastewater sample processing

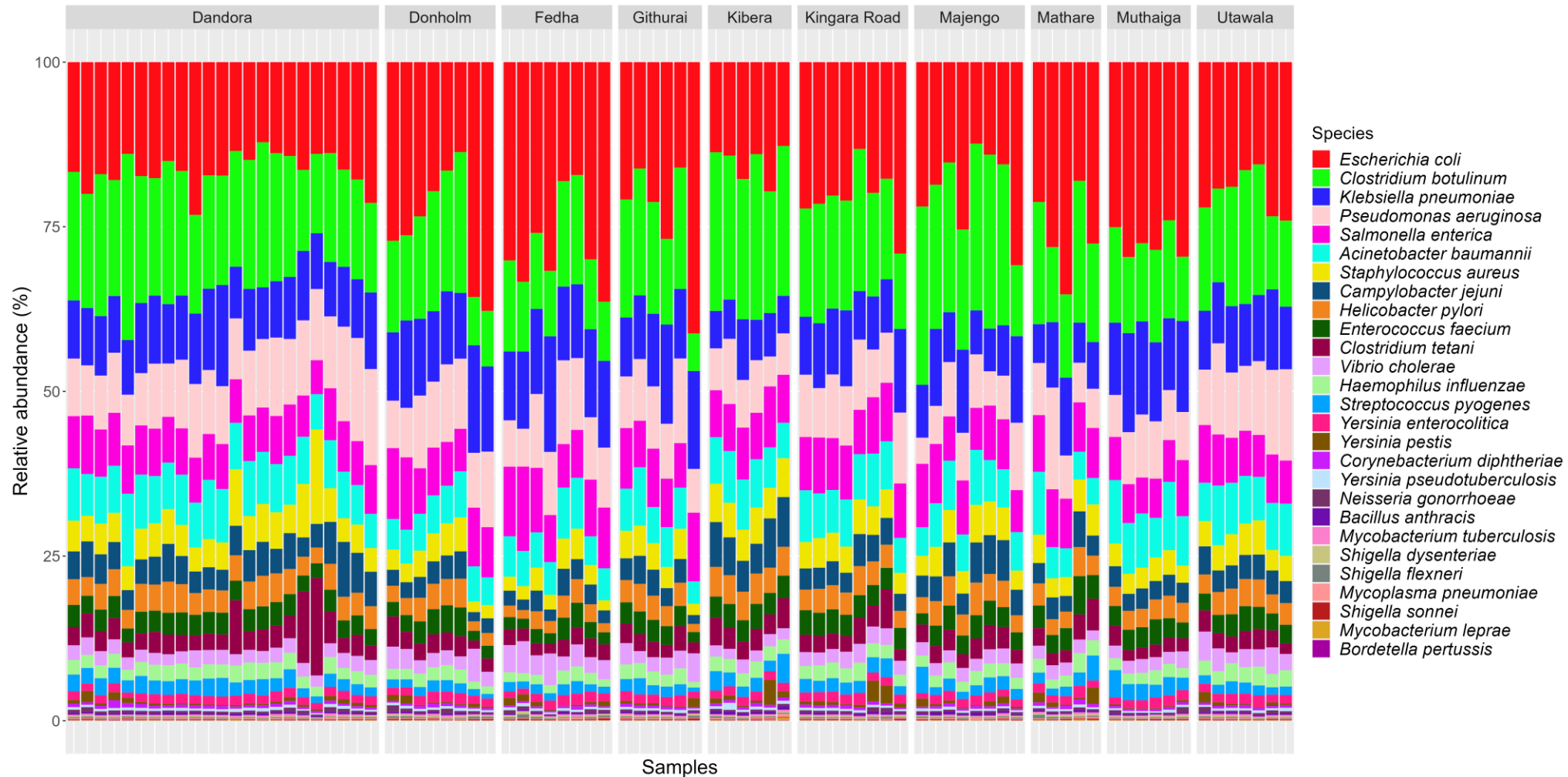


WBS process established at ILRI in the previous work being applied for sample processing.

Microbiome Classification and Composition



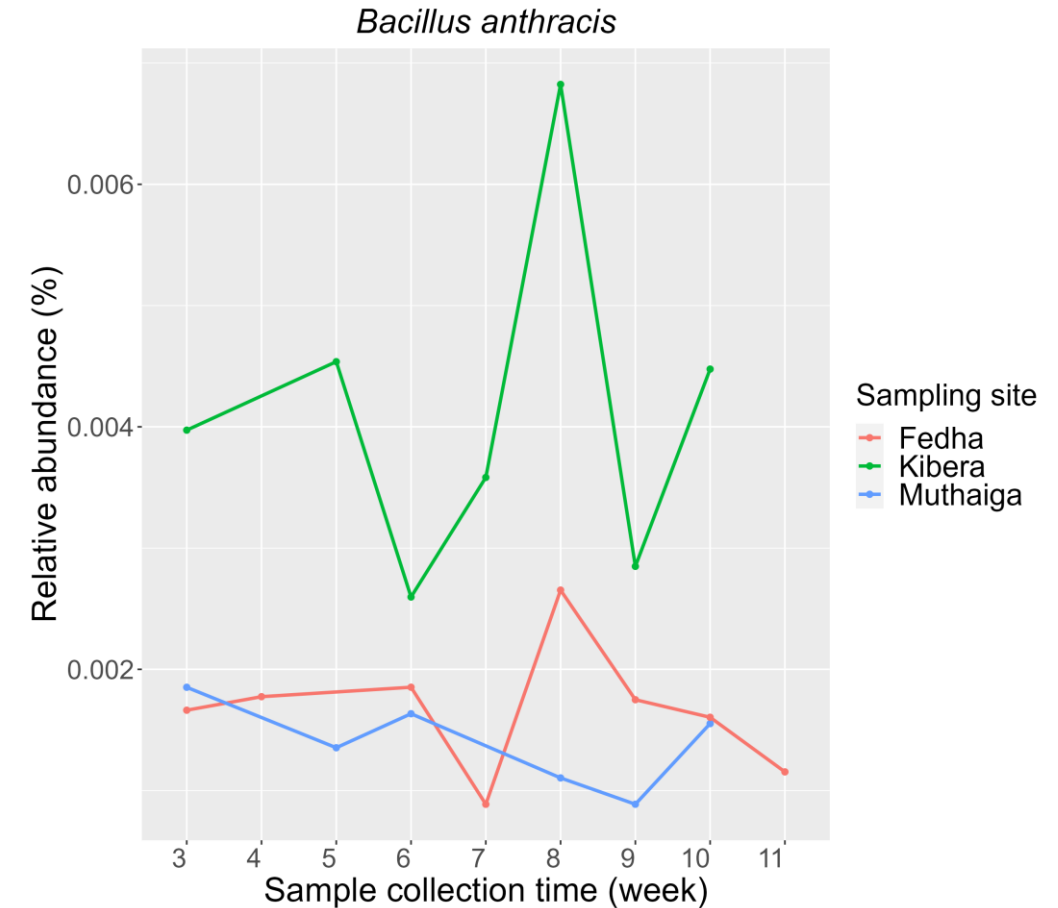
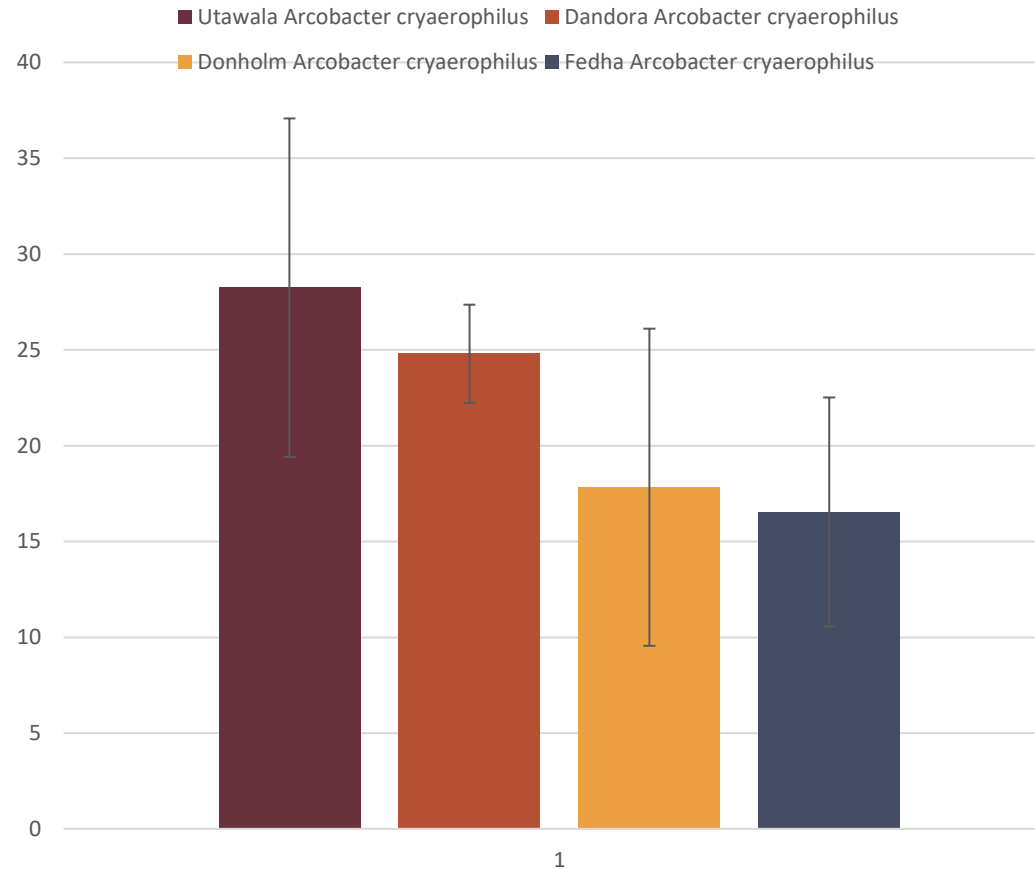
Bacterial Species Composition and Abundance



Bacterial Species Abundance/Public health Relevance

**Arcobacter cryaerophilus* most abundant, emerging human pathogen/express ARGs.

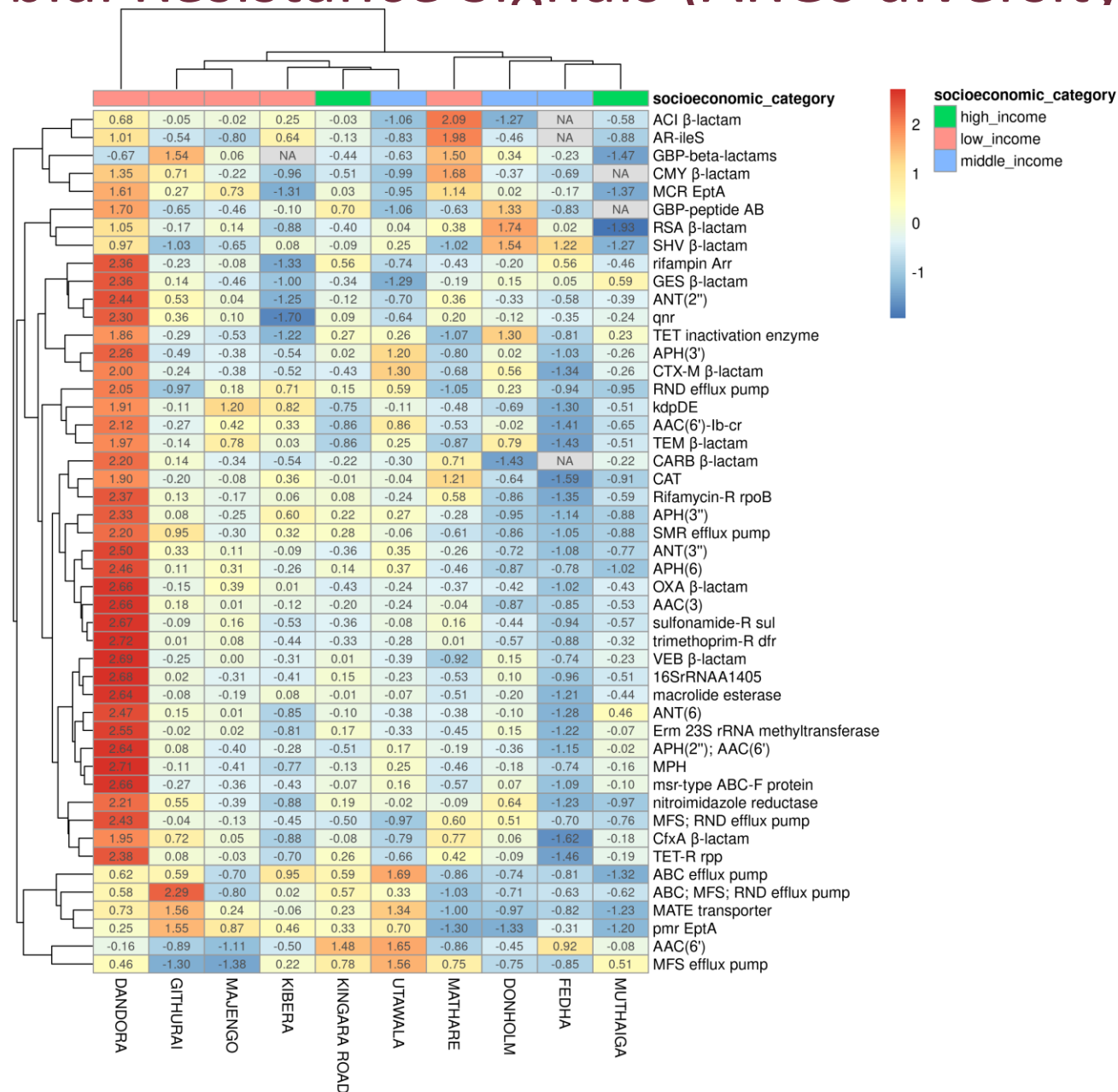
- abortion and enteritis in animals
- Diarrhea and bacteremia in adults and children



Pathogen abundance fluctuation monitored over time in different sites and peaks used to inform potential outbreaks and used to draw correlation with clinical data

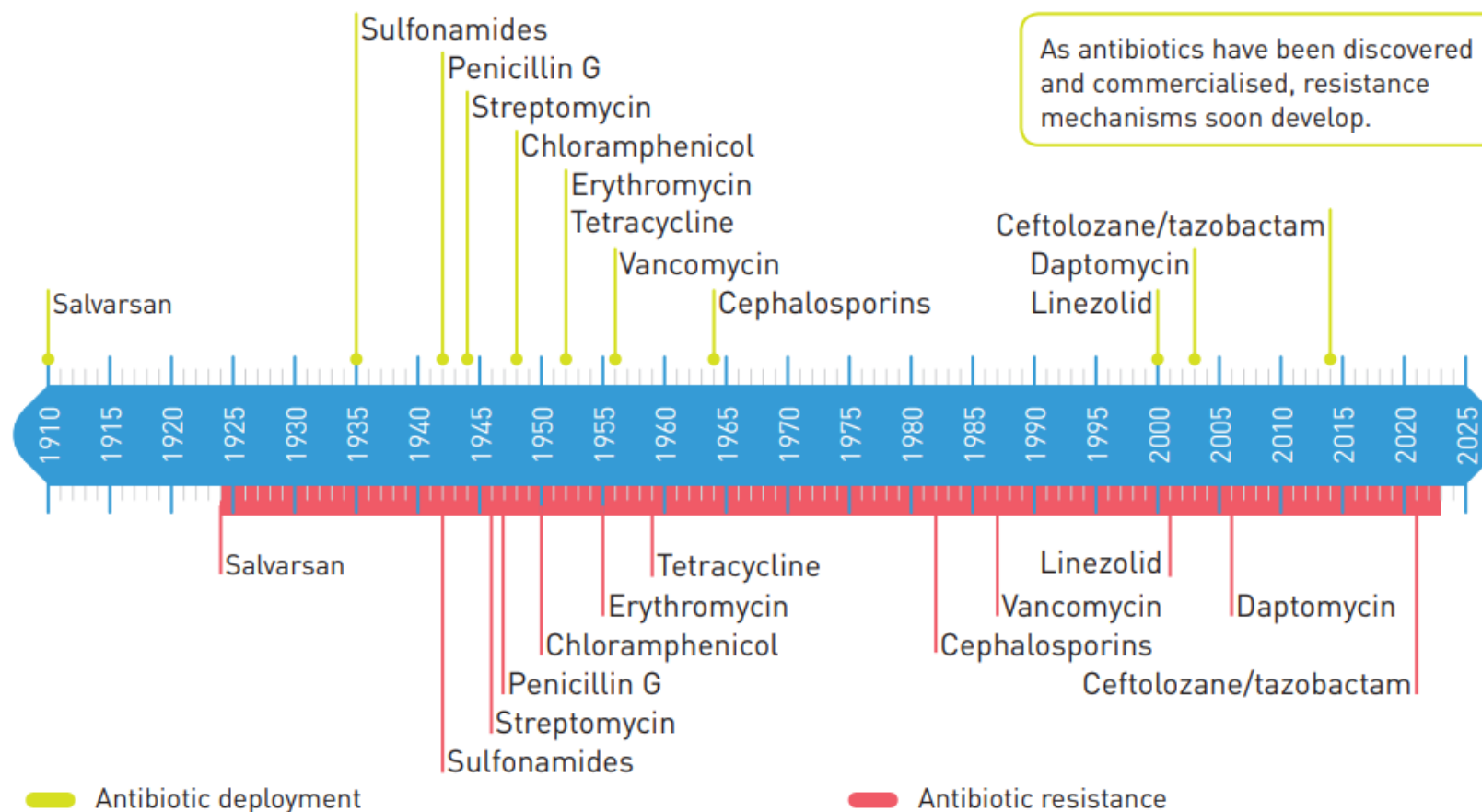
Antimicrobial Resistance Signals (ARGs diversity/abundance)

17



ARGs abundance based on social economics and site of sample collection

WBS for Tracking Antibiotic Resistance



Application of WBE (Utility)

Early detection
of outbreaks



Population-wide
surveillance

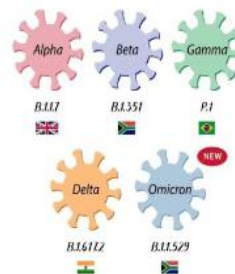


**Interaction with health authorities
and integration with clinical data is of pivotal importance**

Specific
Surveillances



- population parts
- transportation hubs
- transport vessels



Surveillance and
tracking variants of
concern



Acknowledgements



THANK YOU