



# **ZAMBIA'S INTEGRATED ANTIMICROBIAL RESISTANCE SURVEILLANCE FRAMEWORK**

**January 2020**



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CSE is grateful to the Swedish  
International Development  
Cooperation Agency (SIDA)  
for institutional support

The report presents Zambia's integrated antimicrobial resistance surveillance framework. It is an outcome of two workshops organized jointly by the national Antimicrobial Resistance Coordinating Committee (AMRCC) through the Zambia National Public Health Institute (ZNPHI) and the Centre for Science and Environment (CSE), India. ZNPHI and CSE would like to thank all experts who contributed to the development of this report. The list of experts is provided at the end of this report.

#### **About ZNPHI**

ZNPHI (<http://znphi.co.zm/>), a technical arm under the Ministry of Health, is a public health center of excellence that addresses all major public health concerns in Zambia. ZNPHI seeks to improve health of all Zambians through coordinating priority public health and health security activities and resources; leveraging strong partnerships at the international, national, and sub-national levels; generating and analyzing scientific evidence for advocacy, policies and programmes; and prioritizing public health functions. It serves as co-Secretariat to the national AMRCC with the Department of Veterinary Services under the Ministry of Fisheries and Livestock, and is responsible for coordinating the implementation of Zambia's Multi-sectoral National Action Plan on Antimicrobial Resistance.

#### **About CSE**

CSE ([www.cseindia.org](http://www.cseindia.org)), India is a non-profit public interest research and advocacy organization working on issues of public health, environment and development in India and global South. The Food Safety and Toxins team at CSE has been working to address the problem of antimicrobial resistance, particularly the animal and environmental aspects of it.

#### **A publication of AMRCC and CSE**

##### *Printed by*

Centre for Science and Environment  
41, Tughlakabad Institutional Area  
New Delhi 110 062  
Phone: + 91-11-40616000  
Fax: + 91-11-29955879  
E-mail: [cse@cseindia.org](mailto:cse@cseindia.org)  
Website: [www.cseindia.org](http://www.cseindia.org)

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## Foreword



**A**ntimicrobial Resistance (AMR) is a major global public health challenge of the 21st century. The lack of country specific data on the burden of AMR and factors driving its spread means that interventions are likely to be limited and even misdirected.

In line with the core objectives of the Global Action Plan (GAP) on AMR, in 2017 Zambia developed a multi-sectoral National Action Plan (NAP) on AMR, which sets out priority actions and strategies to address the factors influencing development and spread of AMR in the Zambian context. A key area recognized in Zambia's AMR NAP is the need to put in place mechanisms for the systematic and coordinated generation, collection and analysis of data on AMR.

The Zambia National Integrated Antimicrobial Resistance Surveillance Strategy (NIAMRSS) has, therefore, been developed to foster a coordinated approach to the collection of AMR data across all relevant sectors, namely human-health, animal health, agriculture and the environment. The NIAMRSS provides interventions and sets the framework for strengthening knowledge and the evidence base on AMR through surveillance and research.

To ensure focused and effective implementation as well as optimization of the limited available resources for maximum impact, the Zambian Ministry of Health, through the Zambia National Public Health Institute (ZNPFI), which serves as Secretariat to the national Antimicrobial Resistance Coordinating Committee (AMRCC), and the India-based Centre for Science and Environment (CSE) co-hosted a joint workshop in March 2019, one of whose objectives was to facilitate strengthening and prioritization of Zambia's multi-sectoral AMR NAP and NIAMRSS. The workshop incorporated expert participants from key sectors including human-health, animal health, environment, food, drug and agriculture. Workshop participants were drawn from several Zambian government departments, international AMR experts, and AMR focal points from select African countries.

This report sets out some key outputs from the workshop, particularly with respect to the strengthening and prioritization of Zambia's NIAMRSS. Implementation of interventions laid out in this document will inform research, policy and practice, including around the regulation and monitoring of import, distribution and end-user access to antimicrobials.



Dr Victor Mukonka  
Director - Zambia National Public Health Institute  
Chairperson - National Antimicrobial Resistance Coordinating Committee

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## Foreword



Antimicrobial resistance (AMR) is one of the biggest public health crises of our times. With the effectiveness of antibiotics in treating bacterial diseases dropping worryingly and steeply, and no breakthrough in developing new options in the last several decades, preserving existing antibiotics for future generations has become extremely critical. For this to happen, it is important to develop an adequate understanding of the extent of resistance in different

bacteria against several antibiotics. This can only be achieved through routine surveillance of AMR, specifically antibiotic resistance. But such surveillance will only serve its purpose if emphasis is put on resistance emanating from animal and environmental sectors. The information from surveillance in these sectors needs to be integrated with those from the human-health domains (which have been the focus of surveillance efforts till now). Such an integrated approach will help identify areas that need immediate or greater attention, apart from helping put together a complete picture of AMR.

The good news is that most countries have outlined extensive surveillance initiatives in their multi-year action plans to contain AMR. But implementation challenges persist, and include building the required understanding, consensus and capacity among multiple stakeholders, which needs considerable effort, time and resources. In addition, global guidance and country-level examples to adapt from are limited, particularly in case of environmental surveillance. These problems are amplified in low-and-middle income countries, which are going to be heavily impacted by AMR. The silver lining is that challenges and solutions are similar in different resource-constrained settings and there is huge merit in transfer of knowledge and learnings across borders. Keeping this in view, the Centre for Science and Environment, which is working on AMR containment in India, is working with the Ministry of Health, government of Zambia to implement its multi-sectoral action plan to contain AMR.

Based on inputs from stakeholders in Zambia and experts from Africa and other parts of the world, this report presents a detailed multi-year framework for integrated AMR surveillance across human, animal and environment sectors—perhaps a first of its kind for any country. Rooted in the ground realities of Zambia, the framework takes the country's national surveillance strategy a step forward and provides a phase-wise approach to gradually scale up data collection from multiple sources across ten provinces in Zambia. In addition to testing resistance in bacteria and identifying genetic markers, the framework offers an optimized sampling design for antibiotic residues in food from animals and in environment samples.

We are confident that our colleagues from the Zambia National Public Health Institute and the national Antimicrobial Resistance Coordinating Committee, who have successfully worked together to help develop this comprehensive framework, will find this report useful in their concerted efforts to contain AMR. We also hope that this framework helps other countries in Africa and beyond to fine-tune their surveillance initiatives and fight against AMR. We look forward to our continued collaboration in this critical area.

Sunita Narain  
Director General  
Centre for Science and Environment

Amit Khurana  
Director, Food Safety and Toxins  
Centre for Science and Environment

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## Abbreviations

AMR – Antimicrobial Resistance  
AMRCC – Antimicrobial Resistance Coordinating Committee  
AST – Antimicrobial Susceptibility Testing  
CIA – Critically Important Antimicrobial  
CLSI – Clinical and Laboratory Standards Institute  
CSE – Centre for Science and Environment  
CVRI – Central Veterinary Research Institute  
DHIS – District Health Information Software  
ESBL – Extended Spectrum Beta Lactamase  
ETP – Effluent Treatment Plant  
EUCAST – European Committee on Antimicrobial Susceptibility Testing  
FAO – Food and Agriculture Organization of the United Nations  
FDCL – Food and Drug Control Laboratory  
GLASS – Global Antimicrobial Resistance Surveillance System  
HPLC – High Performance Liquid Chromatography  
ID – Infectious Disease  
LIMS – Laboratory Information Management System  
LMIC – Low- and Middle-Income Country  
MFL – Ministry of Fisheries and Livestock  
NAP – National Action Plan  
NIAMRSS – National Integrated Antimicrobial Resistance Surveillance Strategy  
NISIR – National Institute of Scientific and Industrial Research  
OIE – World Organisation for Animal Health  
QMS – Quality Management System  
SDG – Sustainable Development Goal  
STP – Sewage Treatment Plant  
UNEP – United Nations Environment Programme  
UNZA – University of Zambia  
WHO – World Health Organization  
ZARI – Zambia Agriculture Research Institute  
ZBS – Zambia Bureau of Standards  
ZEMA – Zambia Environmental Management Agency  
ZNPFI – Zambia National Public Health Institute



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## 1. Introduction

Antimicrobial resistance (AMR), particularly antibiotic resistance, is recognized as a global public health threat, causing grave health problems and putting a severe economic burden on people and nations. AMR can also negatively impact food safety, nutrition security, livelihood and the attainment of Sustainable Development Goals (SDGs). Antibiotic use and misuse in humans, animals—particularly in the food-animal sector—and crops are known causes of rising AMR. Now, the environment is also recognized to play a key role in the emergence and spread of AMR. A growing concern is the waste from factories, healthcare settings, farms and community settings, which could contain antibiotics, resistant bacteria or genes that confer resistance to antibiotics. AMR is a ‘One Health’ issue that needs to be addressed through improved policy and practice across diverse sectors including human-health, animal and crop production, and environment.

The tripartite of the World Health Organization (WHO), Food and Agriculture Organization of the United Nations (FAO) and World Organisation for Animal Health (OIE) has been working towards AMR containment.<sup>1,2&3</sup> Recently, the United Nations Environment Programme (UNEP) has also been roped in to address the environmental aspects of AMR.

Integrated and multi-sectoral surveillance of AMR is vital to gather evidence for necessary action.<sup>4</sup> But there is limited laboratory capacity and understanding to integrate surveillance in different sectors. Globally, many reports provide insight into different aspects of surveillance—WHO’s *Global Antimicrobial Resistance Surveillance System (GLASS)*, *Guidance on Integrated Surveillance of Antimicrobial Resistance in Food-borne Bacteria*<sup>5</sup> and *Report on Surveillance of Antibiotic Consumption*;<sup>6</sup> OIE’s survey on use of antimicrobial agents in animals in 2018,<sup>7</sup> and Chapters in the Terrestrial Animal Health Code on AMR.<sup>8</sup> However, there is limited reflection through one guiding report on surveillance across all relevant sectors, which includes environmental AMR surveillance in particular.

Over the last few years, many countries have also developed plans for multi-sectoral AMR surveillance in their National Action Plans (NAPs). So far, the focus has been on surveillance in human-health sector while some countries are planning to conduct routine AMR surveillance in animal sectors. A few countries are also focusing on environmental AMR. Developed countries such as Canada, Denmark, England, Japan, Netherlands, Sweden and the United States already have surveillance programmes in the food-animal sector. AMR data in the environment sector relies heavily on research studies. The implementation of surveillance programmes in low- and middle-income countries (LMICs) is particularly challenging due to constraints in resources and capacities, competing priorities, and limited focus on waste and environment. Such countries would, therefore, need more assistance. On the positive side, global understanding on the need for enhanced AMR surveillance is evolving and opportunities of cross learning between nations are increasing.

The Zambia National Public Health Institute (ZNPHE) and the Centre for Science and Environment (CSE), as part of an existing collaboration to support implementation of Zambia’s NAP-AMR<sup>9</sup>, jointly organized a three-day workshop on Integrated Surveillance Framework for Antimicrobial Resistance in March 2019 in Lusaka, Zambia. Experts referred to the existing Zambia National Integrated Antimicrobial Resistance Surveillance Strategy (NIAMRSS) developed by the Zambian Antimicrobial Resistance Coordinating Committee (AMRCC), and

deliberated on the development of an integrated AMR surveillance framework for the country. The surveillance of AMR for food-animal sector was further finalized at the Expert Meeting on Implementation of Zambia's Multi-sectoral National Action Plan on AMR, which was organized jointly by ZNPHI and CSE in August 2019.

This report provides a framework to conduct AMR surveillance in an integrated manner, keeping in mind the capacities in Zambia. The framework aims to support the implementation of the surveillance component of Zambia's multi-sectoral NAP on AMR in the short- and long-term.

Other countries will also be able to draw from this framework and design their respective integrated surveillance frameworks for effective monitoring of AMR and implementation of their NAPs.

## 2. Approach to the development of Zambia's integrated antimicrobial resistance surveillance framework

To develop the framework, the expert group identified a set of thematic areas under surveillance, which are:

- Surveillance of antibiotic resistance in human-health sector
- Surveillance of antibiotic resistance and antibiotic residues in food-animal sector
- Surveillance of antibiotic resistance and antibiotic residues in the environment

The expert group also agreed upon a phased approach for surveillance that would allow progress in a step-by-step manner. The timeframe considered is as follows:

- Phase 1 (zero–three years; short-term): Surveillance activity that would be initiated in the first three years
- Phase 2 (four–five years; medium-term): Surveillance activity that would be initiated after three years
- Phase 3 (greater than five years; long-term): Surveillance activity that would be initiated after five years. No higher limit was set for this phase

Across all the phases, experts collectively finalized key elements of surveillance such as bacteria, antibiotics and sampling strategy in detail. The framework also highlights surveillance of resistance conferring genes, along with suggestions on laboratory networks and training requirements for effective surveillance. It aims to synergize existing laboratory capacities in the country, and supplement them with necessary systems and tools to facilitate AMR surveillance.

The focus of this framework is on surveillance of antibiotic resistance in bacteria. Wherever needed, the scope has been widened to include surveillance of antibiotic resistance in fungi or parasites. Additional surveillance components introduced in Phase 2 and Phase 3 are indicated in **blue** and **green** text respectively.

The framework does not include the surveillance of antibiotic use in human-health and food-animal sector. These should be considered in future editions of the framework.

### 3. Zambia's integrated surveillance framework

#### 3.1 Surveillance of antibiotic resistance in human-health sector

The following section provides a framework for surveillance of antibiotic resistance in human-health sector. The framework categorizes key elements of surveillance into different phases. It specifies key bacteria, antibiotics and genes for surveillance and also outlines sample type, sampling geography, size and frequency. Surveillance efforts are focused on hospital and community settings for better comparison and understanding of AMR. A stepwise approach to expand the surveillance, beginning with three provinces in Phase 1 to all provinces in Phase 3 is suggested. The framework also highlights the need for laboratory strengthening and necessary capacity building to facilitate routine surveillance in Zambia.

**Table 1: Framework for surveillance of antibiotic resistance in human-health sector**

	Phase 1 (0–3 years)	Phase 2 (4–5 years)	Phase 3 (> 5 years)
<b>Sample type(s)</b>	<ul style="list-style-type: none"> <li>Blood</li> <li>Urine</li> <li>Faeces</li> <li>Cerebrospinal fluid</li> <li>Samples from skin and soft tissue infections</li> </ul>	<ul style="list-style-type: none"> <li>Blood</li> <li>Urine</li> <li>Faeces</li> <li>Cerebrospinal fluid</li> <li>Samples from skin and soft tissue infections</li> <li>Urethral and cervical swabs</li> </ul>	<ul style="list-style-type: none"> <li>Blood</li> <li>Urine</li> <li>Faeces</li> <li>Cerebrospinal fluid</li> <li>Samples from skin and soft tissue infections</li> <li>Urethral and cervical swabs</li> </ul>
<b>Bacteria for AST</b>	<p><b>Blood</b></p> <ul style="list-style-type: none"> <li><i>Escherichia coli</i></li> <li><i>Staphylococcus aureus</i></li> <li><i>Klebsiella pneumoniae</i></li> <li><i>Streptococcus pneumoniae</i></li> <li><i>Salmonella</i> spp.</li> <li><i>Acinetobacter baumannii</i></li> <li><i>Pseudomonas aeruginosa</i></li> </ul> <p><b>Urine</b></p> <ul style="list-style-type: none"> <li><i>Escherichia coli</i></li> <li><i>Klebsiella pneumoniae</i></li> </ul> <p><b>Faeces</b></p> <ul style="list-style-type: none"> <li><i>Salmonella</i> spp.</li> <li><i>Shigella</i> spp.</li> <li><i>Vibrio cholerae</i></li> </ul> <p><b>Cerebrospinal fluid</b></p> <ul style="list-style-type: none"> <li><i>Streptococcus pneumoniae</i></li> <li><i>Haemophilus influenzae</i></li> </ul>	<p><b>Blood</b></p> <ul style="list-style-type: none"> <li><i>Escherichia coli</i></li> <li><i>Staphylococcus aureus</i></li> <li><i>Klebsiella pneumoniae</i></li> <li><i>Streptococcus pneumoniae</i></li> <li><i>Salmonella</i> spp.</li> <li><i>Acinetobacter baumannii</i></li> <li><i>Pseudomonas aeruginosa</i></li> </ul> <p><b>Urine</b></p> <ul style="list-style-type: none"> <li><i>Escherichia coli</i></li> <li><i>Klebsiella pneumoniae</i></li> </ul> <p><b>Faeces</b></p> <ul style="list-style-type: none"> <li><i>Salmonella</i> spp.</li> <li><i>Shigella</i> spp.</li> <li><i>Vibrio cholerae</i></li> </ul> <p><b>Cerebrospinal fluid</b></p> <ul style="list-style-type: none"> <li><i>Streptococcus pneumoniae</i></li> <li><i>Haemophilus influenzae</i></li> </ul> <p><b>Urethral and cervical swabs</b></p> <ul style="list-style-type: none"> <li><i>Neisseria gonorrhoeae</i></li> </ul>	<p><b>Blood</b></p> <ul style="list-style-type: none"> <li><i>Escherichia coli</i></li> <li><i>Staphylococcus aureus</i></li> <li><i>Klebsiella pneumoniae</i></li> <li><i>Streptococcus pneumoniae</i></li> <li><i>Salmonella</i> spp.</li> <li><i>Acinetobacter baumannii</i></li> <li><i>Pseudomonas aeruginosa</i></li> <li><i>Candida auris (fungus)</i></li> </ul> <p><b>Urine</b></p> <ul style="list-style-type: none"> <li><i>Escherichia coli</i></li> <li><i>Klebsiella pneumoniae</i></li> <li><i>Enterobacter</i> spp.</li> </ul> <p><b>Faeces</b></p> <ul style="list-style-type: none"> <li><i>Salmonella</i> spp.</li> <li><i>Shigella</i> spp.</li> <li><i>Vibrio cholerae</i></li> <li><i>Campylobacter jejuni</i></li> </ul> <p><b>Cerebrospinal fluid</b></p> <ul style="list-style-type: none"> <li><i>Streptococcus pneumoniae</i></li> <li><i>Haemophilus influenzae</i></li> <li><i>Cryptococcus neoformans (fungus)</i></li> </ul> <p><b>Urethral and cervical swabs</b></p> <ul style="list-style-type: none"> <li><i>Neisseria gonorrhoeae</i></li> </ul>

<p><b>Antibiotics for AST</b></p>	<p><i>Escherichia coli</i>*: Ampicillin, ceftriaxone, cefotaxime, ceftazidime, cefepime, colistin, ertapenem, imipenem, and trimethoprim/sulfamethoxazole</p> <p><i>Staphylococcus aureus</i>: Cefoxitin, vancomycin, cefoxidine, ciprofloxacin, clindamycin, erythromycin, gentamicin, and trimethoprim/sulfamethoxazole</p> <p><i>Klebsiella pneumoniae</i>: Ceftriaxone, cefotaxime, ceftazidime, cefepime, ciprofloxacin, colistin, ertapenem, imipenem, levofloxacin, and trimethoprim/sulfamethoxazole</p> <p><i>Streptococcus pneumoniae</i>: Ceftriaxone, cefotaxime, meropenem, oxacillin, penicillin G and trimethoprim/sulfamethoxazole</p> <p><i>Salmonella</i> spp.: Ampicillin, azithromycin, ceftriaxone, cefotaxime, ciprofloxacin, colistin and imipenem</p> <p><i>Acinetobacter baumannii</i>: Amikacin, ceftriaxone, cefotaxime, ceftazidime, cefepime, colistin, ertapenem, gentamicin, imipenem, minocycline and tigecycline</p> <p><i>Pseudomonas aeruginosa</i>: Cefepime, ceftazidime, ciprofloxacin, gentamicin, piperacillin, piperacillin/tazobactam, ticarcillin and ticarcillin/tazobactam</p> <p><i>Candida auris</i>: Amphotericin B (antifungal) and fluconazole (antifungal)</p> <p><i>Enterobacter</i> spp.: Ampicillin, amoxicillin, amoxicillin/clavulanic acid, ampicillin/salbutam, cefazolin, cefepime, ceftriaxone, cefotaxime, cefuroxime, imipenem, gentamicin, ciprofloxacin, levofloxacin, trimethoprim/sulfamethoxazole, chloramphenicol and nitofurantoin</p> <p><i>Shigella</i> spp.: Ampicillin, azithromycin, ceftriaxone, cefotaxime, ciprofloxacin, imipenem and nalidixic acid</p> <p><i>Vibrio cholerae</i>: Ampicillin, azithromycin, ciprofloxacin, doxycycline, tetracycline and trimethoprim/sulfamethoxazole</p> <p><i>Campylobacter jejuni</i>: Azithromycin, ampicillin, ciprofloxacin, erythromycin and tetracycline</p> <p><i>Haemophilus influenzae</i>: Ceftriaxone, cefotaxime and penicillin G</p> <p><i>Cryptococcus neoformans</i>: Amphotericin B (antifungal), fluconazole (antifungal) and 5-flucytosine (antifungal)</p> <p><i>Neisseria gonorrhoeae</i>: Azithromycin, ciprofloxacin, ceftriaxone, gentamicin and streptomycin</p>		
<p><b>Genetic markers</b></p>	<p>ESBL genes (blaCTX-M, blaSHV and blaTEM)</p>	<p>ESBL genes (blaCTX-M, blaSHV and blaTEM)</p> <p>Carbapenemase-encoding genes (KPC, NDM-1 and IMP-type)</p> <p>mecA and mecC</p>	<p>ESBL genes (blaCTX-M, blaSHV and blaTEM)</p> <p>Carbapenemase-encoding genes (KPC, NDM-1 and IMP-type)</p> <p>mecA and mecC</p>
<p><b>Provinces</b></p>	<ol style="list-style-type: none"> <li>1. Copperbelt</li> <li>2. Lusaka</li> <li>3. Muchinga</li> <li>4. Southern</li> </ol>	<ol style="list-style-type: none"> <li>1. Copperbelt</li> <li>2. Eastern</li> <li>3. Luapula</li> <li>4. Lusaka</li> <li>5. Muchinga</li> <li>6. Southern</li> <li>7. Western</li> </ol>	<ol style="list-style-type: none"> <li>1. Central</li> <li>2. Copperbelt</li> <li>3. Eastern</li> <li>4. Luapula</li> <li>5. Lusaka</li> <li>6. Muchinga</li> <li>7. Northern</li> <li>8. North Western</li> <li>9. Southern</li> <li>10. Western</li> </ol>
<p><b>Sampling sites</b></p>	<p>Hospitals</p>	<p>Hospitals and clinics</p>	<p>Hospitals and clinics</p>

\*Surveillance for ESBL *Escherichia coli* is recommended in two steps. First screening with cefotaxime if presence of ESBL *Escherichia coli* is suspected, followed by validation with additional cephalosporins

<b>Number of sampling sites per province</b>	One hospital 1. Ndola Teaching Hospital (Copperbelt) 2. University Teaching Hospital (Lusaka) 3. Chilonga Mission Hospital (Muchinga) 4. Livingstone Central Hospital (Southern Province)	One hospital 1. Ndola Teaching Hospital (Copperbelt) 2. University Teaching Hospital (Lusaka), and 2 other peri-urban hospitals in Lusaka 3. Chilonga Mission Hospital (Muchinga) 4. Livingstone Central Hospital (Southern Province) 5. Lewanika General Hospital (Western Province) 6. Chipata Central Hospital (Eastern Province) 7. Mansa General Hospital (Luapula)  5-10 clinics per province (for outpatient community samples)	One hospital 1. Ndola Teaching Hospital (Copperbelt) 2. University Teaching Hospital (Lusaka), and two other peri-urban hospitals in Lusaka 3. Chilonga Mission Hospital (Muchinga) 4. Livingstone Central Hospital (Southern Province) 5. Lewanika General Hospital (Western Province) 6. Chipata Central Hospital (Eastern Province) 7. Mansa General Hospital (Luapula) 8. Kabwe General hospital (Central Province) 9. Solwezi General Hospital (North Western Province) 10. Kasama General Hospital (Northern Province)  5-10 clinics per province (for outpatient community samples)
<b>Total number of samples per province per year</b>	As per routine sampling protocols		
<b>Frequency of sampling</b>	Once per week (as per standard routine sampling protocols)		
<b>AST interpretation method</b>	CLSI	CLSI**	CLSI**
<b>Number of laboratories/networks</b>	In Phase 1, only one laboratory per district, which has the capacity for conducting AMR surveillance. This is to be expanded in Phases 2 and 3 based on ongoing capacity building and laboratory strengthening efforts		
<b>Training required</b>	Training on ID, AST, GLASS and WHO-NET	Training on ID, AST, GLASS and WHO-NET Training on QMS	Training on ID, AST, GLASS and WHO-NET Training on QMS
<b>Stakeholder responsible</b>	ZNPHI and coordinating hospitals in respective provinces		

\*\*Presently CLSI is being followed. The feasibility of EUCAST to be adopted could be explored further

Note: Surveillance components introduced in Phase 2 are marked blue; surveillance components introduced in Phase 3 are marked green

### 3.2 Surveillance of antibiotic resistance and antibiotic residues in food-animal sector

The following sections provide a detailed framework for surveillance of antibiotic resistance and antibiotic residues in food-animal sector. Since cattle and chicken are the two main food-animal species produced in Zambia, a surveillance framework for them has been developed. Surveillance in other food-animals (e.g., pigs and fishes) as well as in correspondingly derived food-animal products (e.g., pork and fish meat) may be developed based on the proposed framework. Across all frameworks, routine surveillance expands from key food-animal producing and consuming provinces in Phase 1 to all provinces in Phase 3.

#### 3.2.1 Antibiotic resistance in cattle for meat

*Table 2: Framework for surveillance of antibiotic resistance in cattle for meat* represents a framework for monitoring of antibiotic resistance in cattle for meat. The framework gives a step-by-step approach to surveillance across the three phases. It prioritizes key bacteria for monitoring, specifies a detailed sampling strategy (for e.g., geography, location, size and frequency) and identifies the required laboratory support. In addition, it also emphasizes on the need for identification of genetic resistance markers.

Only carcass swab samples are recommended for collection from abattoirs, meat processing plants and butcheries. The frequency of sampling is to be increased from once a year in Phase 1 to quarterly per year in Phase 3. Antibiotics to be considered for this surveillance cater largely to those used during animal rearing or those against which resistance has been detected in the animal.

**Table 2: Framework for surveillance of antibiotic resistance in cattle for meat**

	Phase 1 (0–3 years)	Phase 2 (4–5 years)	Phase 3 (> 5 years)
<b>Sample type</b>	• Carcass swab	• Carcass swab	• Carcass swab
<b>Bacteria for AST</b>	<ul style="list-style-type: none"> <li>• <i>Salmonella</i> spp.</li> <li>• <i>Escherichia coli</i> (commensal and pathogenic)</li> <li>• <i>Enterococcus faecalis</i></li> </ul>	<ul style="list-style-type: none"> <li>• <i>Salmonella</i> spp.</li> <li>• <i>Escherichia coli</i> (commensal and pathogenic)</li> <li>• <i>Enterococcus faecalis</i></li> <li>• <i>Campylobacter</i> spp.</li> <li>• <i>Staphylococcus aureus</i></li> </ul>	<ul style="list-style-type: none"> <li>• <i>Salmonella</i> spp.</li> <li>• <i>Escherichia coli</i> (commensal and pathogenic)</li> <li>• <i>Enterococcus faecalis</i></li> <li>• <i>Campylobacter</i> spp.</li> <li>• <i>Staphylococcus aureus</i></li> </ul>
<b>Antibiotics for AST</b>	<ul style="list-style-type: none"> <li>• <i>Salmonella</i> spp. and <i>Escherichia coli</i>*: Ampicillin, cefataxime, cefpodoxime, ceftriaxone, ceftazidime, ciprofloxacin, colistin, gentamicin, imipenem, neomycin, streptomycin, tetracycline and trimethoprim/sulfamethoxazole</li> <li>• <i>Enterococcus faecalis</i>: Ampicillin, enrofloxacin, erythromycin, gentamicin, tetracycline, tylosin and vancomycin</li> <li>• <i>Campylobacter</i> spp.: Ampicillin, ciprofloxacin, erythromycin and tetracycline</li> <li>• <i>Staphylococcus aureus</i>: Amoxicillin, cefoxitin, erythromycin, gentamicin, lincomycin, methicillin, penicillin, tetracycline, trimethoprim/sulfamethoxazole and vancomycin</li> </ul>		
<b>Genetic markers</b>		mecA, mecC and ESBL genes (blaCTX-M, blaSHV and blaTEM)	mecA, mecC and ESBL genes (blaCTX-M, blaSHV and blaTEM)
<b>Provinces</b>	<ol style="list-style-type: none"> <li>1. Central</li> <li>2. Lusaka</li> <li>3. Southern</li> </ol>	<ol style="list-style-type: none"> <li>1. Central</li> <li>2. Copperbelt</li> <li>3. Eastern</li> <li>4. Lusaka</li> <li>5. Southern</li> </ol>	<ol style="list-style-type: none"> <li>1. Central</li> <li>2. Copperbelt</li> <li>3. Eastern</li> <li>4. Luapula</li> <li>5. Lusaka</li> <li>6. Muchinga</li> <li>7. Northern</li> <li>8. North Western</li> <li>9. Southern</li> <li>10. Western</li> </ol>

\*Surveillance for ESBL *Escherichia coli* is recommended in two steps. First screening with cefotaxime if presence of ESBL *Escherichia coli* is suspected, followed by validation with additional cephalosporins

<b>Sampling sites</b>	Abattoirs and meat processing plants	Abattoirs, meat processing plants and <b>butcheries</b>	Abattoirs, meat processing plants and <b>butcheries</b>
<b>Number of sampling sites per province</b>	<ul style="list-style-type: none"> <li>Two–three big abattoirs</li> <li>Two–three big meat processing plants</li> </ul>	<ul style="list-style-type: none"> <li>Two–three big abattoirs</li> <li>Two–three big meat processing plants</li> <li><b>Two–three big local markets</b></li> </ul>	<ul style="list-style-type: none"> <li>Two–three big abattoirs</li> <li>Two–three big meat processing plants</li> <li>Two–three big local markets</li> </ul>
<b>Total number of samples per province per year</b>	100–200	100–200	100–200
<b>Frequency of sampling</b>	Annual	Bi-annual (100–200 divided into two halves)	Quarterly (100–200 divided into four quarters)
<b>AST interpretation method</b>	CLSI		
<b>Number of laboratories/networks</b>	At least five laboratories (one laboratory from CVRI, two laboratories from UNZA, one VETLAB and one regional laboratory in Southern province)	<b>At least six laboratories</b> (one laboratory from CVRI, two laboratories from UNZA, one VETLAB and one regional laboratory each in Southern and <b>Eastern provinces</b> )	<b>At least nine laboratories</b> (one laboratory from CVRI, two laboratories from UNZA, one VETLAB and <b>five regional laboratories</b> )
<b>Training required</b>	<p>AST, GLASS, LIMS, QMS, WHO-NET, sample collection, preparation and transportation, and result analysis (both qualitative and quantitative aspects)</p> <p>Two technicians to be trained per laboratory, through two trainings per year (one internal training and one training by external expert)</p>	<p>AST, GLASS, LIMS, QMS, WHO-NET, sample collection, preparation and transportation, and result analysis (both qualitative and quantitative aspects; <b>more focus on quantitative aspect</b>)</p> <p><b>Four technicians to be trained per laboratory, through four trainings per year (three internal trainings and one training by external expert)</b></p>	<p>AST, GLASS, LIMS, QMS, WHO-NET, sample collection, preparation and transportation, result analysis (qualitative and quantitative; more focus on quantitative aspect), and <b>genomics</b></p> <p>Four technicians to be trained per laboratory, through four trainings per year (three internal trainings and one training by external expert)</p>
<b>Stakeholder responsible</b>	Ministry of Fisheries and Livestock		

Note: Surveillance components introduced in Phase 2 are marked **blue**; surveillance components introduced in Phase 3 are marked **green**

### 3.2.2 Antibiotic residues in beef

A guiding framework on how to carry out routine surveillance of antibiotic residues in beef (meat from cattle) is shown in *Table 3: Framework for surveillance of antibiotic residues in beef*. It is largely similar to the antibiotic resistance surveillance framework in cattle for meat (as shown in *Table 2*) to facilitate achievement of both resistance and residue surveillance with minimum effort (for example, samples could be collected from the same geographic location). This will also enable a better comparison between antibiotic resistance and antibiotic residue trends in the samples tested.

The framework considers monitoring of only meat samples for the presence of residues of antibiotics which are used in practice. Apart from abattoirs, meat processing plants and butcheries, the framework refers to the sampling from retail shops in Phases 2 and 3. CHARM test and HPLC are recommended for detecting presence of antibiotic residues. While qualitative detection of antibiotics could be carried out using the CHARM test, quantification of antibiotics in meat samples could be carried out by HPLC.



**Table 3: Framework for surveillance of antibiotic residues in beef**

	Phase 1 (0–3 years)	Phase 2 (4–5 years)	Phase 3 (> 5 years)
<b>Sample type</b>	• Meat	• Meat	• Meat
<b>Antibiotics for residue monitoring*</b>	Ampicillin, azithromycin, ceftiofur, ciprofloxacin, cloxacillin, colistin, enrofloxacin, erythromycin, gentamicin, neomycin, oxacillin, penicillin, streptomycin, sulfonamides (commonly used), tetracycline, tylosin and zinc bacitracin		
<b>Provinces</b>	1. Central 2. Lusaka 3. Southern	1. Central 2. Copperbelt 3. Eastern 4. Lusaka 5. Southern	1. Central 2. Copperbelt 3. Eastern 4. Luapula 5. Lusaka 6. Muchinga 7. Northern 8. North Western 9. Southern 10. Western
<b>Sampling sites</b>	Abattoirs and meat processing plants	Abattoirs, meat processing plants, butcheries and retail shops	Abattoirs, meat processing plants, butcheries and retail shops
<b>Number of sampling sites per province</b>	<ul style="list-style-type: none"> <li>Two-three big abattoirs</li> <li>Two-three big meat processing plants</li> </ul>	<ul style="list-style-type: none"> <li>Two-three big abattoirs</li> <li>Two-three big meat processing plants</li> <li>Two-three big local markets</li> </ul>	<ul style="list-style-type: none"> <li>Two-three big abattoirs</li> <li>Two-three big meat processing plants</li> <li>Two-three big local markets</li> </ul>
<b>Total number of samples per province per year</b>	100–200		
<b>Frequency of sampling</b>	Annual	Bi-annual (100–200 divided into two halves)	Quarterly (100–200 divided into four quarters)
<b>Analytical method</b>	CHARM test and HPLC Qualitative analysis to be done first by CHARM, followed by quantitative analysis by HPLC only in samples where residues are detected		
<b>Number of laboratories and laboratory networks**</b>	Three laboratories (one laboratory from CVRI and two laboratories from UNZA)		
<b>Training required</b>	<p>Sample collection, preparation, analysis and interpretation of results (qualitative and quantitative aspects)</p> <p>Two technicians to be trained per laboratory, through two trainings per year (one internal training and one training by external expert)</p>	<p>Sample collection, preparation, analysis and interpretation of results (qualitative and quantitative aspects; more focus on the quantitative aspect)</p> <p>Four technicians to be trained per laboratory, through four trainings per year (three internal trainings and one training by external expert)</p>	<p>Sample collection, preparation, analysis, interpretation of results (qualitative and quantitative; more focus on the quantitative aspect), and equipment operation</p> <p>Four technicians to be trained per laboratory, through four trainings per year (three internal trainings and one training by external expert)</p>
<b>Stakeholder responsible</b>	Ministry of Fisheries and Livestock		

\*List of antibiotics could be updated or revised from time to time, if necessary

\*\*Minimum number represented, subject to increase over time with increasing capacity and resources

Note: Surveillance components introduced in Phase 2 are marked blue; surveillance components introduced in Phase 3 are marked green

### 3.2.3 Antibiotic resistance in broiler and layer poultry

The framework for routine antibiotic resistance surveillance in broiler and layer poultry is shown in *Table 4: Framework for surveillance of antibiotic resistance in broiler poultry* and *Table 5: Framework for surveillance of antibiotic resistance in layer poultry* respectively. Multiple sample types such as air-sac swabs, faecal swabs and faecal samples are recommended for surveillance, which can be expanded to include bone marrow swabs after Phase 1. Additionally, eggs are important samples to be tested in all phases. Similar sampling sites are suggested for broilers and layers such as farms and live bird markets. Sampling from other sites such as hatcheries, veterinary clinics, slaughter houses or meat processing plants could be included and continued in Phases 2 and 3 in case useful results are obtained in Phase 1. Laboratory networks have been proposed for surveillance across all phases, which include the nodal laboratory CVRI, and its regional laboratories, UNZA and private laboratories. These are suggested as minimum number of laboratories required for surveillance, and could be increased gradually with more capacity and resources.

**Table 4: Framework for surveillance of antibiotic resistance in broiler poultry**

	Phase 1 (0–3 years)	Phase 2 (4–5 years)	Phase 3 (> 5 years)
<b>Sample type(s)</b>	<ul style="list-style-type: none"> <li>Air-sac swab</li> <li>Faecal swab</li> <li>Faecal sample</li> </ul>	<ul style="list-style-type: none"> <li>Air-sac swab</li> <li>Faecal swab</li> <li>Faecal sample</li> <li>Bone marrow swab</li> </ul>	<ul style="list-style-type: none"> <li>Air-sac swab</li> <li>Faecal swab</li> <li>Faecal sample</li> <li>Bone marrow swab</li> </ul>
<b>Bacteria for AST</b>	<ul style="list-style-type: none"> <li><i>Escherichia coli</i></li> <li><i>Salmonella</i> spp.</li> <li><i>Coccidia</i> spp. (parasite)</li> </ul>	<ul style="list-style-type: none"> <li><i>Escherichia coli</i></li> <li><i>Salmonella</i> spp.</li> <li><i>Coccidia</i> spp. (parasite)</li> <li><i>Campylobacter</i> spp.</li> <li><i>Enterococcus</i> spp.</li> <li><i>Clostridium</i> spp.</li> </ul>	<ul style="list-style-type: none"> <li><i>Escherichia coli</i></li> <li><i>Salmonella</i> spp.</li> <li><i>Coccidia</i> spp. (parasite)</li> <li><i>Campylobacter</i> spp.</li> <li><i>Enterococcus</i> spp.</li> <li><i>Clostridium</i> spp.</li> <li><i>Listeria</i> spp.</li> </ul>
<b>Antibiotics for AST</b>	<ul style="list-style-type: none"> <li><i>Escherichia coli</i>* and <i>Salmonella</i> spp.: Ampicillin, cefataxime, cefpodoxime, ceftriaxone, ceftazidime, ciprofloxacin, colistin, gentamicin, imipenem, neomycin, streptomycin, tetracycline, trimethoprim/sulfamethoxazole and zinc bacitracin</li> <li><i>Coccidia</i> spp.: Amprolium, salinomycin and sulphonamides (commonly used)</li> <li><i>Campylobacter</i> spp.: Ampicillin, ciprofloxacin, erythromycin and tetracycline</li> <li><i>Enterococcus</i> spp.: Ampicillin, enrofloxacin, erythromycin, gentamicin, tetracycline, tylosin and vancomycin</li> <li><i>Clostridium</i> spp.: Ampicillin, ciprofloxacin, tetracycline and vancomycin</li> <li><i>Listeria</i> spp.: Ampicillin, enrofloxacin, erythromycin, gentamicin, tetracycline, tylosin and vancomycin</li> </ul>		
<b>Genetic markers**</b>	ESBL genes (blaCTX-M)	ESBL genes (blaCTX-M)	ESBL genes (blaCTX-M) mcr, qnr and tet genes
<b>Provinces</b>	<ol style="list-style-type: none"> <li>Central</li> <li>Copperbelt</li> <li>Lusaka</li> <li>Southern</li> </ol>	<ol style="list-style-type: none"> <li>Central</li> <li>Copperbelt</li> <li>Eastern</li> <li>Lusaka</li> <li>Southern</li> </ol>	<ol style="list-style-type: none"> <li>Central</li> <li>Copperbelt</li> <li>Eastern</li> <li>Luapula</li> <li>Lusaka</li> <li>Muchinga</li> <li>Northern</li> <li>North Western</li> <li>Southern</li> <li>Western</li> </ol>

\*Investigate ESBL *Escherichia coli*

\*\* Monitoring as part of research and not routine surveillance

<b>Sampling sites</b>	Farms (small-scale, medium, commercial and breeder), and markets (live) Sampling from sites to be included and continued in Phases 2 and 3 in case of useful results: <ul style="list-style-type: none"> <li>• Markets (open, retail)</li> <li>• Slaughter houses</li> <li>• Meat processing plants</li> <li>• Hatcheries</li> <li>• Veterinary clinics</li> </ul>	Farms (small-scale, medium, commercial and breeder), and markets (live)	Farms (small-scale, medium, commercial and breeder), and markets (live)
<b>Number of sampling sites per province</b>	At least two sites (for each type of sampling site)	Minimum of two sites (for each type of sampling site; <b>should be expanded from Phase 1</b> )	Minimum of two sites (for each type of sampling site; <b>should be expanded from Phase 2</b> )
<b>Total number of samples per province per year</b>	200	<b>300</b> <b>200 for new province</b>	300 200 for new province
<b>Frequency of sampling</b>	Annual	Bi-annual (total samples divided into two halves)	Quarterly (total samples divided into four quarters)
<b>AST interpretation method</b>	CLSI		
<b>Number of laboratories/networks</b>	At least five laboratories (one laboratory from CVRI, two laboratories from UNZA, one VETLAB and one regional laboratory in Southern province)	<b>At least six laboratories</b> (one laboratory from CVRI, two laboratories from UNZA, one VETLAB and one regional laboratory each in Southern and <b>Eastern provinces</b> )	<b>At least nine laboratories</b> (one laboratory from CVRI, two laboratories from UNZA, one VETLAB and <b>five regional laboratories</b> )
<b>Training required</b>	AST, LIMS, QMS, sample collection, preparation and transportation and result analysis (both qualitative and quantitative aspects)  Two technicians to be trained per laboratory, through two trainings per year (one internal training and one training by external expert)	AST, LIMS, QMS, sample collection, preparation and transportation and result analysis (both qualitative and quantitative aspects; <b>more focus on quantitative aspect</b> )  <b>Four technicians to be trained per laboratory, through four trainings per year (three internal trainings and one training by external expert)</b>	AST, LIMS, QMS, sample collection, preparation and transportation, result analysis (qualitative and quantitative; more focus on quantitative aspect), and <b>genomics</b>  Four technicians to be trained per laboratory, through four trainings per year (three internal trainings and one training by external expert)
<b>Stakeholder responsible</b>	Ministry of Fisheries and Livestock		

Note: Surveillance components introduced in Phase 2 are marked **blue**; surveillance components introduced in Phase 3 are marked **green**

**Table 5: Framework for surveillance of antibiotic resistance in layer poultry**

	Phase 1 (0–3 years)	Phase 2 (4–5 years)	Phase 3 (> 5 years)
<b>Sample type(s)</b>	<ul style="list-style-type: none"> <li>Air-sac swab</li> <li>Faecal swab</li> <li>Faecal sample</li> <li>Egg</li> </ul>	<ul style="list-style-type: none"> <li>Air-sac swab</li> <li>Faecal swab</li> <li>Faecal sample</li> <li>Egg</li> <li>Bone marrow swab (to be considered if analysis from egg is not conclusive)</li> </ul>	<ul style="list-style-type: none"> <li>Air-sac swab</li> <li>Faecal swab</li> <li>Faecal sample</li> <li>Egg</li> <li>Bone marrow swab (to be considered if analysis from egg is not conclusive)</li> </ul>
<b>Bacteria for AST</b>	<ul style="list-style-type: none"> <li><i>Escherichia coli</i></li> <li><i>Salmonella</i> spp.</li> <li><i>Staphylococcus</i> spp.</li> </ul>	<ul style="list-style-type: none"> <li><i>Escherichia coli</i></li> <li><i>Salmonella</i> spp.</li> <li><i>Staphylococcus</i> spp.</li> <li><i>Campylobacter</i> spp.</li> <li><i>Enterococcus</i> spp.</li> <li><i>Clostridium</i> spp.</li> </ul>	<ul style="list-style-type: none"> <li><i>Escherichia coli</i></li> <li><i>Salmonella</i> spp.</li> <li><i>Staphylococcus</i> spp.</li> <li><i>Campylobacter</i> spp.</li> <li><i>Enterococcus</i> spp.</li> <li><i>Clostridium</i> spp.</li> <li><i>Listeria</i> spp.</li> </ul>
<b>Antibiotics for AST</b>	<ul style="list-style-type: none"> <li><i>Escherichia coli</i>* and <i>Salmonella</i> spp.: Ampicillin, cefataxime, cefpodoxime, ceftriaxone, ceftazidime, ciprofloxacin, colistin, gentamicin, imipenem, neomycin, streptomycin, tetracycline, trimethoprim/sulfamethoxazole and zinc bacitracin**</li> <li><i>Staphylococcus</i> spp.: Ampicillin, cefataxime, cefpodoxime, ceftriaxone, ceftazidime, ciprofloxacin, colistin, gentamicin, imipenem, neomycin, oxacillin, streptomycin, tetracycline, trimethoprim/sulfamethoxazole and zinc bacitracin**</li> <li><i>Campylobacter</i> spp.: Ampicillin, ciprofloxacin, erythromycin and tetracycline</li> <li><i>Enterococcus</i> spp.: Ampicillin, enrofloxacin, erythromycin, gentamicin, tetracycline, tylosin and vancomycin</li> <li><i>Clostridium</i> spp.: Ampicillin, ciprofloxacin, tetracycline and vancomycin</li> <li><i>Listeria</i> spp.: Ampicillin, gentamicin, enrofloxacin, erythromycin, tetracycline, tylosin and vancomycin</li> </ul>		
<b>Genetic markers***</b>	ESBL genes (blaCTX-M)	ESBL genes (blaCTX-M)	ESBL genes (blaCTX-M) mcr, qnr and tet genes
<b>Provinces</b>	<ol style="list-style-type: none"> <li>Central</li> <li>Copperbelt</li> <li>Lusaka</li> <li>Southern</li> </ol>	<ol style="list-style-type: none"> <li>Central</li> <li>Copperbelt</li> <li>Eastern</li> <li>Lusaka</li> <li>Southern</li> </ol>	<ol style="list-style-type: none"> <li>Central</li> <li>Copperbelt</li> <li>Eastern</li> <li>Luapula</li> <li>Lusaka</li> <li>Muchinga</li> <li>Northern</li> <li>North Western</li> <li>Southern</li> <li>Western</li> </ol>
<b>Sampling sites</b>	<p>Farms (small-scale, medium, commercial and breeder), and markets (live)</p> <p>Sampling from sites to be included and continued in Phases 2 and 3 in case of useful results:</p> <ul style="list-style-type: none"> <li>Markets (open and retail)</li> <li>Hatcheries</li> <li>Veterinary clinics</li> </ul>	<p>Farms (small-scale, medium, commercial and breeder), and markets (live)</p>	<p>Farms (small-scale, medium, commercial and breeder), and markets (live)</p>

\* Investigate ESBL *Escherichia coli*

\*\* Zinc bacitracin is not used in layer poultry, however it will be useful to check in phase 1 to understand extent of misuse

\*\*\* Monitoring as part of research and not routine surveillance

<b>Number of sampling sites per province</b>	At least two sites (for each type of sampling site)	Minimum of two sites (for each type of sampling site; <b>should be expanded from Phase 1</b> )	Minimum of two sites (for each type of sampling site; <b>should be expanded from Phase 2</b> )
<b>Total number of samples per province per year</b>	250	<b>350</b> 250 for new province	350 250 for new province
<b>Frequency of sampling</b>	Annual	Bi-annual (total samples divided into two halves)	Quarterly (total samples divided into four quarters)
<b>AST interpretation method</b>	CLSI		
<b>Number of laboratories/networks</b>	At least five laboratories (one laboratory from CVRI, two laboratories from UNZA, one VETLAB and one regional laboratory in Southern province)	<b>At least six laboratories</b> (one laboratory from CVRI, two laboratories from UNZA, one VETLAB and one regional laboratory each in Southern and <b>Eastern provinces</b> )	<b>At least nine laboratories</b> (one laboratory from CVRI, two laboratories from UNZA, one VETLAB and <b>five regional laboratories</b> )
<b>Training required</b>	AST, LIMS, QMS, sample collection, preparation and transportation and result analysis (both qualitative and quantitative aspects)  Two technicians to be trained per laboratory, through two trainings per year (one internal training and one training by external expert)	AST, LIMS, QMS, sample collection, preparation and transportation and result analysis (both qualitative and quantitative aspects; <b>more focus on the quantitative aspect</b> )  <b>Four technicians to be trained per laboratory, through four trainings per year (three internal trainings and one training by external expert)</b>	AST, LIMS, QMS, sample collection, preparation and transportation, result analysis (both qualitative and quantitative aspects; more focus on the quantitative aspect), and <b>genomics</b>  Four technicians to be trained per laboratory, through four trainings per year (three internal trainings and one training by external expert)
<b>Stakeholder responsible</b>	Ministry of Fisheries and Livestock		

Note: Surveillance components introduced in Phase 2 are marked **blue**; surveillance components introduced in Phase 3 are marked **green**

### 3.2.4 Antibiotic residues in broiler and layer poultry

*Table 6: Framework for surveillance of antibiotic residues in broiler poultry* and *Table 7: Framework for surveillance of antibiotic residues in layer poultry* provide guiding frameworks on routine surveillance of antibiotic residues in broiler and layer poultry respectively. The recommended samples for residue testing include kidney and liver in Phases 1 and 2 and breast muscle in Phase 3. Egg has been identified as an additional sample type in case of layers. These samples are proposed to be collected from retail markets, in addition to farms, processing plants (for broilers) and hatcheries (for layers). As in the case of beef, CHARM test and HPLC would be used as analytical methods. Laboratories of the CVRI and the UNZA have been suggested for surveillance of antibiotic residues in samples from broiler and layer poultry.

**Table 6: Framework for surveillance of antibiotic residues in broiler poultry**

	Phase I (0–3 years)	Phase 2 (4–5 years)	Phase 3 (> 5 years)
<b>Sample type(s)</b>	<ul style="list-style-type: none"> <li>• Kidney</li> <li>• Liver</li> <li>• Breast muscle</li> </ul>	<ul style="list-style-type: none"> <li>• Kidney</li> <li>• Liver</li> <li>• Breast muscle</li> </ul>	<ul style="list-style-type: none"> <li>• Kidney</li> <li>• Liver</li> <li>• Breast muscle</li> </ul>
<b>Antibiotics for residue monitoring*</b>	Amoxicillin, avilamycin, colistin, doxycycline, enrofloxacin, flavomycin, furazolidone, gentamicin, neomycin, olaquinox, sulphonamides (commonly used), sulfadiazine, tetracycline, trimethoprim/sulphonamide, tylosin, virginiamycin and zinc bacitracin		
<b>Provinces</b>	<ol style="list-style-type: none"> <li>1. Central</li> <li>2. Copperbelt</li> <li>3. Lusaka</li> <li>4. Southern</li> </ol>	<ol style="list-style-type: none"> <li>1. Central</li> <li>2. Copperbelt</li> <li>3. Eastern</li> <li>4. Lusaka</li> <li>5. Southern</li> </ol>	<ol style="list-style-type: none"> <li>1. Central</li> <li>2. Copperbelt</li> <li>3. Eastern</li> <li>4. Luapula</li> <li>5. Lusaka</li> <li>6. Muchinga</li> <li>7. North Western</li> <li>8. Northern</li> <li>9. Southern</li> <li>10. Western</li> </ol>
<b>Sampling sites</b>	Farms (small-scale, medium, commercial and breeder), processing plants and retail markets		
<b>Number of sampling sites per province</b>	At least two sites (for each type of sampling site)	Minimum of two sites (for each type of sampling site; <b>should be expanded from Phase 1</b> )	Minimum of two sites (for each type of sampling site; <b>should be expanded from Phase 2</b> )
<b>Total number of samples per province per year</b>	50 per sampling site	50 per sampling site	50 per sampling site (subject to revision)
<b>Frequency of sampling</b>	Annual	Bi-annual (total samples divided into two halves)	Quarterly (total samples divided into four quarters)
<b>Analytical method</b>	CHARM, HPLC Qualitative analysis to be done first by CHARM, followed by quantitative analysis by HPLC only in samples where residues are detected		
<b>Number of laboratories/networks**</b>	Three laboratories (one laboratory from CVRI and two laboratories from UNZA)		
<b>Training required</b>	<p>Sample collection, preparation, analysis and interpretation of results (qualitative and quantitative aspects)</p> <p>Two technicians to be trained per laboratory, through two trainings per year (one internal training and one training by external expert)</p>	<p>Sample collection, preparation, analysis and interpretation of results (qualitative and quantitative aspects; <b>more focus on the quantitative aspect</b>)</p> <p><b>Four technicians to be trained per laboratory, through four trainings per year (three internal trainings and one training by external expert)</b></p>	<p>Sample collection, preparation, analysis and interpretation of results (qualitative and quantitative; more focus on the quantitative aspect)</p> <p>Four technicians to be trained per laboratory, through four trainings per year (three internal trainings and one training by external expert)</p>
<b>Stakeholder responsible</b>	Ministry of Fisheries and Livestock		

\*List of antibiotics could be updated or revised from time to time, if necessary

\*\*Minimum number represented, subject to increase over time with increasing capacity and resources

Note: Surveillance components introduced in Phase 2 are marked **blue**; surveillance components introduced in Phase 3 are marked **green**

**Table 7: Framework for surveillance of antibiotic residues in layer poultry**

	Phase 1 (0–3 years)	Phase 2 (4–5 years)	Phase 3 (> 5 years)
<b>Sample type(s)</b>	<ul style="list-style-type: none"> <li>• Kidney</li> <li>• Liver</li> <li>• Egg</li> </ul>	<ul style="list-style-type: none"> <li>• Kidney</li> <li>• Liver</li> <li>• Egg</li> <li>• <b>Breast muscle</b></li> </ul>	<ul style="list-style-type: none"> <li>• Kidney</li> <li>• Liver</li> <li>• Egg</li> <li>• Breast muscle</li> </ul>
<b>Antibiotics for residue monitoring*</b>	Amoxicillin, avilamycin, colistin, doxycycline, enrofloxacin, flavomycin, gentamicin, neomycin, olaquinox, sulphonamides (commonly used), sulfadiazine, tetracycline, trimethoprim/sulphonamide, tylosin, virginiamycin and zinc bacitracin**		
<b>Provinces</b>	<ol style="list-style-type: none"> <li>1. Central</li> <li>2. Copperbelt</li> <li>3. Lusaka</li> <li>4. Southern</li> </ol>	<ol style="list-style-type: none"> <li>1. Central Province</li> <li>2. Copperbelt</li> <li>3. <b>Eastern</b></li> <li>4. Lusaka</li> <li>5. Southern</li> </ol>	<ol style="list-style-type: none"> <li>1. Central</li> <li>2. Copperbelt</li> <li>3. Eastern</li> <li>4. <b>Luapula</b></li> <li>5. Lusaka</li> <li>6. <b>Muchinga</b></li> <li>7. <b>North Western</b></li> <li>8. <b>Northern</b></li> <li>9. Southern</li> <li>10. <b>Western</b></li> </ol>
<b>Sampling sites</b>	Farms (small-scale, medium, commercial and breeder), hatcheries and retail markets		
<b>Number of sampling sites per province</b>	At least two per sampling site	Minimum of two per site (should be expanded from Phase 1)	Minimum of two per site (should be expanded from Phase 2)
<b>Total number of samples per province per year</b>	50 per sampling site	50 per sampling site	50 per sampling site (subject to revision)
<b>Frequency of sampling</b>	Annual	Bi-annual (total samples divided into two halves)	Quarterly (total samples divided into four quarters)
<b>Analytical method</b>	CHARM, HPLC Qualitative analysis to be done first by CHARM, followed by quantitative analysis by HPLC only in samples where residues are detected		
<b>Number of laboratories/networks***</b>	Three laboratories (one laboratory from CVRI and two laboratories from UNZA)		
<b>Training required</b>	<p>Sample collection, preparation, analysis and interpretation of results (qualitative and quantitative)</p> <p>Two technicians to be trained per laboratory, through two trainings per year (one internal training and one training by external expert)</p>	<p>Sample collection, preparation, analysis and interpretation of results (qualitative and quantitative with <b>more focus on the quantitative aspect</b>)</p> <p><b>Four technicians to be trained per laboratory, through four trainings per year (three internal trainings and one training by external expert)</b></p>	<p>Sample collection, preparation, analysis and interpretation of results (qualitative and quantitative with more focus on the quantitative aspect)</p> <p>Four technicians to be trained per laboratory, through four trainings per year (three internal trainings and one training by external expert)</p>
<b>Stakeholder responsible</b>	Ministry of Fisheries and Livestock		

\*List of antibiotics could be updated or revised from time to time, if necessary

\*\* Zinc bacitracin is not used in layer poultry, however it will be useful to check in Phase 1 to understand extent of misuse

\*\*\*Minimum number represented, subject to increase over time with increasing capacity and resources

Note: Surveillance components introduced in Phase 2 are marked **blue**; surveillance components introduced in Phase 3 are marked **green**

### 3.3 Surveillance of antibiotic resistance and antibiotic residues in environment

#### 3.3.1 Antibiotic resistance in environment

The framework, as provided in *Table 8: Framework for surveillance of antibiotic resistance in environment*, describes surveillance in waste from point sources (e.g., farms, factories, community and healthcare settings) as well as in samples which act as sinks of waste from point sources such as rivers and lakes. Key elements for carrying out environmental antibiotic resistance surveillance such as bacteria, antibiotics, genes, sampling strategy, laboratory support and training requirements are identified in the framework. The aim is to monitor a Gram positive and a Gram negative bacterium as a common indicator bacterium across all sectors, followed by surveillance of key bacteria specific to a particular sector such as *Salmonella* spp. in food-animal sector and *Vibrio* spp. in aquaculture sector. Antibiotics for AST in such bacteria will depend on various factors such as the type of antibiotics used in food-animal production or consumed in community; resistance trends in human-health, animal, aquaculture or crop sectors; and WHO categorization of critically important antimicrobials (CIAs).<sup>10</sup> Surveillance is phased and progressive in nature.

Given that the Zambia Environmental Management Agency (ZEMA), the key stakeholder for surveillance in environment, will need time and resources to build necessary capacity for antibiotic resistance surveillance in environmental samples, it is proposed that ZEMA is initially supported by additional stakeholders such as ZNPFI, MFL, ZARI, FDCL, ZBS and NISIR for capacity and resources.

**Table 8: Framework for surveillance of antibiotic resistance in environment**

	Phase 1 (0–3 years)	Phase 2 (4–5 years)	Phase 3 (> 5 years)
<b>Sample sites and types</b>	<ul style="list-style-type: none"> <li>Healthcare settings (human and veterinary): Sewage and effluent</li> <li>Farms (poultry, cattle, pig, fish): Effluent, farm litter/manure, drinking water (for animals) and pond water/sediment (for fish farms)</li> <li>Crop farms: Soils, including those where animal farm manure is applied</li> <li>Factory (feed mills, slaughter houses, processing plants, pharmaceutical units and ETPs): Sewage and effluent</li> <li>Community settings (STPs and drinking water treatment plants): Effluent (inlet, mid-point, outlet) and drinking water</li> <li>Others (open wells, rivers, lakes, drug disposal sites): Groundwater, river/lakes, surface water, river sediments and soil</li> </ul>		
<b>Bacteria for AST</b>	Across all sectors <i>Escherichia coli</i> * <i>Enterococcus</i> spp.  Human-health sector <i>Klebsiella pneumoniae</i>  Food-animal sector <i>Salmonella</i> spp. <i>Escherichia coli</i>  Crop sector <i>Aspergillus</i> spp. (fungus)	Across all sectors <i>Escherichia coli</i> * <i>Enterococcus</i> spp.  Human-health sector <i>Klebsiella pneumoniae</i>  Food-animal sector <i>Salmonella</i> spp. <i>Escherichia coli</i>  Crop sector <i>Aspergillus</i> spp. (fungus)  Aquaculture sector <i>Aeromonas</i> spp.	Across all sectors <i>Escherichia coli</i> * <i>Enterococcus</i> spp.  Human-health sector <i>Klebsiella pneumoniae</i> <i>Staphylococcus aureus</i>  Food-animal sector <i>Salmonella</i> spp. <i>Escherichia coli</i> <i>Campylobacter</i> spp.  Crop sector <i>Aspergillus</i> spp. (fungus) <i>Penicillium</i> spp. (fungus) <i>Fusarium</i> spp. (fungus)  Aquaculture sector <i>Aeromonas</i> spp. <i>Vibrio</i> spp.

\*Including ESBL *Escherichia coli*



<b>Antibiotics for AST</b>	<ul style="list-style-type: none"> <li><i>Escherichia coli</i>: Amoxicillin, cefotaxime, ciprofloxacin, colistin, imipenem, tetracycline and trimethoprim/sulfamethoxazole</li> <li><i>Enterococcus</i> spp.: Ampicillin, chloramphenicol, ciprofloxacin, erythromycin, levofloxacin, nitrofurantoin, penicillin, tetracycline and vancomycin</li> <li>For sector-specific bacteria: To be based on antibiotics used, resistance trends in humans/animals/aquaculture/crops and WHO categorization of CIAs</li> </ul>		
<b>Genetic markers**</b>		ESBL genes (blaCTX-M, blaSHV, blaTEM, etc.)	ESBL genes (blaCTX-M, blaSHV, blaTEM, etc.)
<b>Provinces</b>	<p>Hospitals (preferably the biggest):</p> <ol style="list-style-type: none"> <li>One private hospital in Lusaka</li> <li>One provisional hospital in three–four provinces each</li> <li>One veterinary hospital in three–four provinces each</li> </ol>	<p>Hospitals (preferably the biggest):</p> <ol style="list-style-type: none"> <li>One private hospital in Lusaka, Copperbelt, North Western and Southern provinces</li> <li>One provisional hospital in three–four provinces</li> <li>One veterinary hospital in three–four provinces</li> </ol>	<p>Hospitals (preferably the biggest). In addition to Phases 1 and 2:</p> <ol style="list-style-type: none"> <li>One private hospital in the remaining provinces</li> <li>One provisional hospital in the remaining provinces</li> <li>One veterinary hospital in the remaining provinces</li> </ol>
	<p>Farms:</p> <ul style="list-style-type: none"> <li>Central: Poultry, pig, fish, cattle and crop farms</li> <li>Copperbelt: Poultry, pig, fish and cattle</li> <li>Eastern: Poultry, pig, fish and cattle</li> <li>Luapula: Poultry, pig, fish and cattle</li> <li>Lusaka: Poultry, pig, fish and cattle</li> <li>Muchinga: Poultry, pig, fish and cattle</li> <li>Northern: Poultry, pig, fish and cattle</li> <li>North Western: Poultry, pig, fish and cattle</li> <li>Southern: Poultry, pig, fish, cattle and crop farms</li> <li>Western: Poultry, pig, fish and cattle</li> </ul> <p>(Few provinces to begin with for crop farms, followed by expansion in later phases)</p>		
	<p>Factories (feed mills, slaughter houses, abattoirs, pharmaceutical units, processing plants and ETPs): Based on concentration of establishments in a region/province or volume of waste generated</p>		
	<p>STPs: At least one in all provinces</p>		
	<p>Rivers: Kafue river, Luangwa river and Zambezi river</p>		
<b>Number of sampling sites per province</b>	<ul style="list-style-type: none"> <li>Healthcare settings: At least one hospital per province<sup>#</sup></li> <li>Farms: At least one–two farms per district, rotated year-wise</li> <li>Factories: At least one–two factory settings per province</li> <li>Community settings: Five–six per district, rotated year-wise</li> <li>Others: One–two sites per district, rotated year-wise</li> </ul> <p>(Number of sample sites could be increased with time in Phases 2 and 3)</p>		
<b>Total number of samples per province per year</b>	As per standardized national sampling protocols of ZEMA		
<b>Sampling frequency</b>	Bi-annual		
<b>AST interpretation method</b>	CLSI <sup>^</sup>		
<b>Number of laboratories/networks</b>	<ul style="list-style-type: none"> <li>Existing network of laboratories in human-health or animal sector plus additional laboratories</li> <li>ZEMA to be initially supported by laboratory capacity and resources of additional stakeholders such as ZNPHI, MFL, ZARI, FDCL, ZBS and NISIR, till necessary capacity for surveillance of AMR in environment is developed by ZEMA</li> </ul>		
<b>Training required</b>	Training on AST, GLASS, WHO-NET	Training on AST, GLASS, WHO-NET Training on QMS	Training on AST, GLASS, WHO-NET Training on QMS
<b>Stakeholder responsible</b>	Zambia Environmental Management Agency		

\*\*Metagenomic studies and whole genome sequencing (on a subset of isolates) could be considered in long term

<sup>#</sup>Hospitals being sampled for human-health AMR surveillance could be considered

<sup>^</sup> Presently CLSI is being followed. The feasibility of EUCAST to be adopted could be explored further

Note: Surveillance components introduced in Phase 2 are marked blue; surveillance components introduced in Phase 3 are marked green

### 3.3.2 Antibiotic residues in environment

Table 9: Framework for surveillance of antibiotic residues in environment provides framework for routine surveillance of antibiotic residues in environmental samples.

**Table 9: Framework for surveillance of antibiotic residues in environment**

	Phase 1 (0–3 years)	Phase 2 (4–5 years)	Phase 3 (> 5 years)
<b>Sample sites and types</b>	<ul style="list-style-type: none"> <li>Healthcare settings (human and veterinary): Sewage and effluent</li> <li>Factories (Feed mills and pharmaceutical units): Sewage and effluent</li> </ul>	<ul style="list-style-type: none"> <li>Healthcare settings (human and veterinary): Sewage and effluent</li> <li>Factories (Feed mills and pharmaceutical units): Sewage and effluent</li> </ul>	<ul style="list-style-type: none"> <li>Healthcare settings (human and veterinary): Sewage and effluent</li> <li>Factories (Feed mills and pharmaceutical units): Sewage and effluent</li> <li>Farms (poultry, cattle, pig and fish): Farm effluent and soil</li> </ul>
<b>Antibiotics for residue monitoring</b>	To be based on antibiotics used, resistance trends in humans, animals, aquaculture, crops and WHO categorization of CIAs		
<b>Provinces</b>	Hospitals (preferably the biggest) <ol style="list-style-type: none"> <li>One private hospital in Lusaka</li> <li>One provisional hospital each in three provinces</li> <li>One veterinary hospital each in three provinces</li> </ol>	Hospitals (preferably the biggest) <ol style="list-style-type: none"> <li>One private hospital in Lusaka and three more provinces</li> <li>One provisional hospital each in six provinces (including those in Phase 1)</li> <li>One veterinary hospital each in six provinces (including those in Phase 1)</li> </ol>	Hospitals (preferably the biggest). In addition to Phases 1 and 2: <ol style="list-style-type: none"> <li>One private hospital in the remaining provinces</li> <li>One provisional hospital in the remaining provinces</li> <li>One veterinary hospital in the remaining provinces</li> </ol>
	Pharmaceutical units <ol style="list-style-type: none"> <li>One unit in Lusaka</li> </ol>	Pharmaceutical units <ol style="list-style-type: none"> <li>Three units in Lusaka</li> <li>One unit in Copperbelt</li> <li>One unit in Central Province</li> </ol>	Pharmaceutical units <ol style="list-style-type: none"> <li>In addition to Phases 1 and 2, the remaining units in Zambia</li> </ol>
			Farms (poultry, cattle, pig, fish) <ol style="list-style-type: none"> <li>Central</li> <li>Copperbelt</li> <li>Eastern</li> <li>Lusaka</li> <li>Luapula</li> <li>Muchinga</li> <li>Northern</li> <li>North Western</li> <li>Southern</li> <li>Western</li> </ol>
	Feed mills: Based on concentration of establishments in a region/province or volume of waste generated		
<b>Number of sampling sites per province</b>	Healthcare settings: At least one hospital per province <sup>#</sup> Factories: At least one–two factory settings per province Farms: At least one–two farms per district, rotated year-wise		
<b>Total number of samples per province per year</b>	As per standardized national sampling protocols of ZEMA		
<b>Sampling frequency</b>	Bi-annual		
<b>Analytical method</b>	CHARM, HPLC		
<b>Number of laboratories/networks</b>	<ul style="list-style-type: none"> <li>Existing network of laboratories in human-health or animal sector plus additional laboratories</li> <li>ZEMA to be initially supported by laboratory capacity and resources of additional stakeholders such as ZNPHI, MFL, ZARI, FDCL, ZBS, NISIR, till necessary capacity for surveillance of AMR in environment is developed by ZEMA</li> </ul>		
<b>Training required</b>	Sample management and preparation	Sample management and preparation	Sample management and preparation Equipment operation
<b>Stakeholder responsible</b>	Zambia Environmental Management Agency		

<sup>#</sup>Hospitals being sampled for human-health AMR surveillance could be considered

Note: Surveillance components introduced in Phase 2 are marked blue; surveillance components introduced in Phase 3 are marked green

### **3.4 Data analysis and reporting**

Across human-health and food-animal sectors and the larger environment, surveillance related information needs to be appropriately recorded. A quarterly online system for reporting of surveillance data should be developed. Across all sectors, data from sentinel sites at the district or province level should be provided to a centralized database housed at a designated location. Designated reference laboratories across each sector may compare trends of antibiotic resistance or antibiotic residue surveillance. An annual report integrating analyzed surveillance data should be made available in the public domain. There should also be quarterly reporting through appropriate softwares such as WHO-NET and District Health Information Software (DHIS 2).

## List of expert contributors

### Workshop on Integrated Surveillance Framework for Antimicrobial Resistance (March 2019)

#### Experts from Zambia

- Amon Siame, Biomedical Scientist, Chilonga Mission General Hospital
- Chanda Chikwanda, Epidemiologist, Zambia National Public Health Institute
- Chewe Orbrie, Field Epidemiology Training Resident, Zambia National Public Health Institute
- Chileshe Lukwesa-Musyani, AMR Focal Point (Human-health), Lusaka District Health Office
- Christopher P Siame, Regional Laboratory, Ministry of Fisheries and Livestock
- Daniel Ndambasia, Registration Officer-Veterinary Medicines, Zambia Medicines Regulatory Authority
- Danny Sinyinza, Department of Fisheries, Ministry of Fishery and Livestock
- Doreen M Chomba, Principal Agricultural Research Officer/Plant Health Inspector, Plant Quarantine and Phytosanitary Service, Zambia Agriculture Research Institute
- Elebert Mtonga, Senior Analyst, Food and Drugs Control Laboratory
- Francis Chimpangu, AMR Focal Point, Food and Agriculture Organization of the United Nations, Country Office
- Joseph Kasongo, Head, Ndola Teaching Hospital Laboratory
- Kaunda Yamba, University Teaching Hospital, Ministry of Health
- Kellyson Mache, Ministry of Fisheries and Livestock
- Kenny Chisha, Regional Laboratory, Ministry of Fisheries and Livestock
- Kunda Musonda, Head-Laboratory Systems and Networks, Zambia National Public Health Institute
- Mabvuto Phiri, Operations Manager, Central Laboratory, Centre for Infectious Disease Research in Zambia
- Maxwell Nkoya, Director-Planning, Information and Research, Zambia Environmental Management Agency
- Mazyanga L Mazaba Liwewe, Head - Communication, Information and Research, Zambia National Public Health Institute
- Munkombwe Zuma, Director, Medicines Control, Zambia Medicines Regulatory Authority
- Mwiche Chiluba, Head, Emergency Unit, Ndola Teaching Hospital
- Ntombi Mudenda, President, Veterinary Association of Zambia
- Otridah Kapona, Laboratory Scientist and AMR National Focal Point and Coordinator, Laboratory Systems and Networks, Zambia National Public Health Institute
- Paul Fandamu, Deputy Director, Department of Veterinary Services, Ministry of Fisheries and Livestock
- Ranjit Warriar, Director, Biomedical Research, Centre for Infectious Disease Research in Zambia
- Ricky Chazya, National Livestock Epidemiology and Information Centre, Department of Veterinary Services, Ministry of Fisheries and Livestock
- Rodwell Chandipo, Principal Environmental Inspector, Natural Resources Management Unit, Zambia Environmental Management Agency
- Samuel Yingst, Chief, Laboratory Infrastructure and Support, Centers for Disease Control and Prevention
- Sly M Phiri, Research Officer, Department of Fisheries, Ministry of Fisheries and Livestock

- Victor Mukonka, Director, Zambia National Public Health Institute and Chair, Antimicrobial Resistance Coordinating Committee
- Victor Chishimba, Zambia Community Health Initiative

### **International experts**

- Amit Khurana, Programme Director, Food Safety and Toxins, Centre for Science and Environment, India
- Anuj Sharma, Technical Officer-Antimicrobial Resistance; Health Laboratories, World Health Organization Country Office for India, New Delhi, India
- Bashiru Boi Kikimoto, Head, Public Health and Food Safety Division Veterinary Services Directorate, Ministry of Food and Agriculture, Ghana
- Chandra Bhushan, Deputy Director General, Centre for Science and Environment, India
- Dennis Byarugaba, Professor of Microbiology, Faculty of Veterinary Medicine, Makerere University, Uganda
- Divya Khatter, Programme Officer, Food Safety and Toxins, Centre for Science and Environment, India
- Emmanuel Kabali, Regional AMR Project Coordinator, Food and Agriculture Organization Southern Africa Sub-regional Office, Zimbabwe
- Flemming Bager, Head, Department of Risk Assessment and Nutrition, National Food Institute, Technical University of Denmark, Denmark
- John Stelling, Co-Director, World Health Organization Collaborating Centre for Surveillance of Antimicrobial Resistance; Brigham and Women's Hospital, Microbiology Laboratory, Boston, Massachusetts, United States of America
- Joshua Obasanya, Head, Prevention and Programmes Coordination Department, Nigeria Centre for Disease Control, Federal Ministry of Health, Nigeria
- Karl Pedersen, Head of Section, Department of Animal Health and Antimicrobial Strategies, National Veterinary Institute, Sweden
- Mirfin Mpundu, Executive Director EPN and Head React Africa-Kenya
- Rajeshwari Sinha, Deputy Programme Manager, Food Safety and Toxins, Centre for Science and Environment, India
- Samuel Kariuki, Deputy Director, Research and Development, Kenya Medical Research Institute, Kenya

### **High level leadership**

- Chitalu Chilufya, Honorable Minister, Ministry of Health, Zambia, MP, MCC
- Ngulkham Jathom Gangte, High Commissioner, High Commission of India, Zambia
- Kennedy Malama, Permanent Secretary, Technical Services, Ministry of Health, Zambia

### **ZNPHI cooperating partners**

- Nathan Bakyaite, World Health Organization Country Representative, Zambia Country Office
- George Okechi, Food and Agriculture Organization of the United Nations Country Representative, Zambia Country Office

### **Expert Meeting on Implementation of Zambia's Multi-sectoral National Action Plan on AMR (August 2019)**

- Alfred Mangani, Surveillance Officer – Drug Information, Zambia Medicines Regulatory Authority
- Amit Khurana, Programme Director, Food Safety and Toxins, Centre for Science and Environment, India
- Chanda Mwamba, Inspector – Good Distribution Practice, Zambia Medicines Regulatory Authority
- Chileshe Lukwesa-Musyani, AMR Focal Point (Human-health), Lusaka District Health Office
- Clive Simwanza, Acting Senior Veterinary Officer, Department of Veterinary Services, Ministry of Fisheries and Livestock
- Daniel Ndambasia, Registration Officer-Veterinary Medicines, Zambia Medicines Regulatory Authority
- Divya Khatter, Programme Officer, Food Safety and Toxins, Centre for Science and Environment, India
- Elijah Munyama, Dairy Development Officer, Dairy Association of Zambia
- Francis Chimpangu, AMR Focal Point, Food and Agriculture Organization of the United Nations, Country Office
- Fusya Goma, AMR Focal Point (Animal Health), Ministry of Fisheries and Livestock
- Geoffrey Mainda, Veterinary Public Health Officer, Veterinary Services-Public Health Unit, Ministry of Fisheries and Livestock
- Geoffrey M Muuka, Microbiologist, Department of Veterinary Services, Ministry of Fisheries and Livestock
- Godfrey Chinyama, Senior Analyst, Ministry of Water Development Sanitation and Environment Protection
- Gregory Mululuma, Principal Veterinary Officer-Legislation Department of Veterinary Services, Ministry of Fisheries and Livestock
- Kaunda Kaunda, TB Laboratory Manager, Centre for Infectious Diseases Research
- Kaunda Yamba, University Teaching Hospital, Ministry of Health
- Kenneth Kapolowe, Senior Registrar, Internal Medicine, University Teaching Hospital, Ministry of Health
- Maxwell Nkoya, Director-Planning, Information and Research, Zambia Environmental Management Agency
- Menard Makungu, Breeder Production Manager, Tiger Animal Feeds
- Mooya Nzila, Plant Health Inspector, Plant Quarantine and Phytosanitary Service, Zambia Agriculture Research Institute, Ministry of Agriculture
- Mudenda Bernard Hang'ombe, Microbiology Unit, School of Veterinary Medicine, University of Zambia
- Munkombwe Zuma, Director-Medicines Control, Zambia Medicines Regulatory Authority
- Mwansa Songe, Central Veterinary Research Institute, Ministry of Fisheries and Livestock
- Ntombi Mudenda, President, Veterinary Association of Zambia
- Otridah Kapona, Laboratory Scientist and AMR National Focal Point and Coordinator, Laboratory Systems and Networks, Zambia National Public Health Institute
- Paul Zulu, Infectious Diseases Specialist, Ministry of Health

- Philippa Hamakasu, Environmental Research Officer, Ministry of Water Development Sanitation and Environment Protection
- Rajeshwari Sinha, Deputy Programme Manager, Food Safety and Toxins, Centre for Science and Environment, India
- Ranjit Warriar, Director-Biomedical Research, Centre for Infectious Disease Research in Zambia
- Ravindra Salagame, Veterinarian and Country Representative, Lusaka Agrovet
- Ricky Chazya, National Livestock Epidemiology and Information Centre, Department of Veterinary Services, Ministry of Fisheries and Livestock
- Rodwell Chandipo, Principal Environmental Inspector, Natural Resources Management Unit, Zambia Environmental Management Agency
- Rowena Blanco, Nutritionist, Tiger Animal Feeds
- Samuel Yingst, Chief, Laboratory Infrastructure and Support, Centers for Disease Control and Prevention
- Sly M Phiri, Research Officer, Department of Fisheries, Ministry of Fisheries and Livestock
- Sumbukeni Kowa, Head, Food and Drugs Control Laboratory, Ministry of Health
- Tipezenji Sakala, Registration Officer – Veterinary Medicines, Zambia Medicines Regulatory Authority
- Vigirio Mutemwa, Senior Veterinarian, Livestock Services Cooperative Society

### High-level leadership

- Kennedy Malama, Permanent Secretary-Technical Services, Ministry of Health, Zambia (represented by Andrew Silumesii, Director, Public Health, Ministry of Health)
- Benson Mwenya, Permanent Secretary, Ministry of Fisheries and Livestock (represented by Francis M. Mulenga, Deputy Director, Veterinary Services, Department of Veterinary Services, Ministry of Fisheries and Livestock)
- Victor Mukonka, Director, Zambia National Public Health Institute, Ministry of Health (represented by Nathan Kapata, Head—Epidermic Preparedness and Response, Zambia National Public Health Institute)

### Coordination of workshops

#### ZNPHI

- Otridah Kapona, Laboratory Scientist and AMR National Focal Point and Coordinator, Laboratory Systems and Networks, Zambia National Public Health Institute (both workshops)
- Kunda Musonda, Head-Laboratory Systems and Networks, Zambia National Public Health Institute
- Mpanga Kasonde, Antimicrobial Resistance Coordinating Committee Secretariat, Zambia National Public Health Institute

#### CSE

- Rajeshwari Sinha, Deputy Programme Manager, Food Safety and Toxins, Centre for Science and Environment, India (both workshops)
- Divya Khatter, Programme Officer, Food Safety and Toxins, Centre for Science and Environment, India (both workshops)

## Report writing

- Amit Khurana, Programme Director, Food Safety and Toxins, Centre for Science and Environment, India
- Rajeshwari Sinha, Deputy Programme Manager, Food Safety and Toxins, Centre for Science and Environment, India
- Divya Khatter, Programme Officer, Food Safety and Toxins, Centre for Science and Environment, India

With inputs from experts and key Zambian contributors:

- Chileshe Lukwesa-Musyani, AMR Focal Point (Human-health), Lusaka District Health Office
- Doreen M Chomba, Principal Agricultural Research Officer/Plant Health Inspector, Plant Quarantine and Phytosanitary Service, Zambia Agriculture Research Institute
- Fusya Goma, AMR Focal Point (Animal-health), Ministry of Fisheries and Livestock
- Geoffrey Mainda, Veterinary Public Health Officer, Veterinary Services-Public Health Unit, Ministry of Fisheries and Livestock
- Otridah Kapona, Laboratory Scientist and AMR National Focal Point and Coordinator, Laboratory Systems and Networks, Zambia National Public Health Institute
- Rodwell Chandipo, Principal Environmental Inspector, Natural Resources Management Unit, Zambia Environmental Management Agency



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**This report provides a framework to conduct AMR surveillance in an integrated manner, keeping in mind the capacities in Zambia. The framework aims to support the implementation of the surveillance component of Zambia's multi-sectoral National Action Plan on AMR.**

**A publication of AMRCC and CSE**



**Centre for Science and Environment**

41, Tughlakabad Institutional Area

New Delhi 110 062 Phone: + 91-11-40616000

Fax: + 91-11-29955879 E-mail: [cse@cseindia.org](mailto:cse@cseindia.org)

Website: [www.cseindia.org](http://www.cseindia.org)