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**Second Consultation Meeting on Regional AMR Monitoring and Surveillance Guidelines Volume 3:
“Monitoring and surveillance of AMR in aquaculture”**
22, 23, 24, 25, 26 June 2020
(Daily 13:00 -15:00 BKK Time | 14:00 – 16:00 Singapore Time)

SUMMARY AND CONCLUSIONS

1. The Second Regional Consultation of the AMR aquaculture surveillance guideline aimed to: (1) review and provide further technical inputs to the current draft Regional AMR Monitoring and Surveillance Guidelines Volume 3 (*Monitoring and surveillance of AMR in aquaculture*), (2) discuss and deliberate on the approaches being taken as outlined in the guideline; and (3) plan the ways forward towards the finalization and future practical utility of the guideline.
2. The virtual meeting was attended by a total of 55 national, regional, and international representatives and experts on AMR and/or aquaculture, as well as observers, from: Viet Nam, Thailand, India, Indonesia, Singapore, two FAO AMR Reference Centres (Chulalongkorn University based in Thailand and Centre for Environment, Fisheries and Agriculture Science based in the United Kingdom), Clinical and Laboratory Standards Institute (CLSI), World Organisation for Animal Health (OIE), WorldFish, Network of Aquaculture Centres in the Asia-Pacific (NACA), Ireland University, Nitte University, Utrecht University, US Agency for International Development (USAID), FAO Viet Nam, FAO Regional Office for Asia and the Pacific (RAP), and FAO Headquarters.
3. The meeting was opened by Dr Kelvin Lim (AVS/NParks), Dr Kachen Wongsathapornchai (FAO) and Dr Daniel Schar (USAID), who all highlighted the significance of the work at hand given the importance of aquaculture in the region, and the continuing challenge that AMR continue to pose despite the global disruption that COVID-19 pandemic has brought. They also acknowledged the organizers and support received from various experts. The opening was followed by a brief introduction to the meeting objectives by Dr Mary Joy Gordoncillo (FAO RAP), including the overarching background on the series of regional antimicrobial resistance monitoring and surveillance guidelines.
4. The first session was led by Dr Kelvin Lim (AVS/NParks) who provided an overview of Chapter 1: Overview of the Guidelines. The discussions on various sections of the Chapter highlighted the need to include clear definitions of terminologies at the outset, the need to emphasize its aptness for the region, as well as the needed for clarity on the scope of the guideline.

- **Scope of the guideline.** The meeting agreed that the scope of the guideline should clearly indicate who the target audience of this guideline is, what aquaculture species are targeted, and also focus on bacterial pathogens of aquatic species, rather than those that may impact food safety or due to environmental contamination. AMR surveillance for aquatic environment will be addressed under Regional AMR Monitoring and Surveillance Guideline Volume 4 of the series (Monitoring and surveillance of AMR in animal environment).
5. Chapter 2 on Sampling for AMR monitoring and surveillance, led by Dr Mary Joy Gordoncillo (FAO RAP), covered various topics related to prioritizing the target population and bacterial pathogens, and sampling approaches. The following comprehensive summaries are as follows:
- **For inclusion in the chapter:** The chapter should include (1) clear definitions of key terminologies, (2) an assessment prior to commencing AMR surveillance design and implementation in aquaculture, (3) data sources, (4) stakeholder mapping and consultation process, (5) linkage and building interconnections between aquatic disease surveillance and AMR surveillance.
 - **Guidance on prioritizing the target population.** Anchored on the objectives set for the AMR surveillance by the country, the basis for prioritization of target population will rely on national (and international) data sources and can be any of the following (a) based on economic value; (b) based on production data; (c) based on antimicrobial use data; (d) based on human consumption; or (e) as aligned with other national AMR surveillance initiatives. Table 1 summarizing this needs to be further improved to convey the importance of linking objectives with each of these options.
 - **Guidance on prioritizing target bacterial pathogens.** A provisional list of priority bacterial pathogen species to be monitored in the region was introduced and discussed. Participants agreed on the three prioritized bacterial pathogens shown in Table 2 of the draft. It was suggested that the list of other common pathogens in the region will be taken from the compilation provided by FAO HQ, particularly the major bacterial pathogens of aquatic species from tropical waters. For each of these, a menu of references for bacterial isolation, identification, and antimicrobial susceptibility testing (AST) will be provided in the guideline. Priority will be given to international standards, and where these do not exist, guidelines and referenced methodologies that have been shown to work in the region will be listed. Countries and experts also highlighted that important pathogenic bacteria need to be decided by each country or area depending on its importance and country's capacity for AMR surveillance.
 - **Surveillance approaches.** Given that the agreed emphasis will now be for bacterial pathogens from aquatic animals, sampling methodologies will be focused on diseased/infected aquatic species. This also implies: (1) "cycle" will be used rather than "chain"; (2) farms rather than processing plants will be the target sites for sample collection; (3) the sample size should be sufficient to address the surveillance objectives; (4) inclusion of both passive and active surveillance with consideration of: affected life

stages, prevailing conditions (temperature), frequency of case submissions particularly from small farms, active aqua operation during that period, permissive conditions, and country's capacity and resources; and the understanding that (5) sampling from diseased animals will select for higher resistance levels due to likely exposure to treatment prior to sampling.

6. Discussions on Chapters 3 (Sample collection and transport) and 4 (Laboratory Methods) was led by Dr Biyun Ching of NParks Singapore, with presentations from Dr He Sheng Neo, Veterinarian from Vet Public Health of Singapore and Dr Biyun Ching, respectively. The following summary key points were captured:

- **Sample collection and transport.** Collected samples should be transported at low temperature without contact with ice. Additional methodologies for sampling and transportation with consideration to molecular diagnosis and metagenomics analysis should also be included in the chapter.
- **Sample processing and bacterial isolation.** Countries should have determined their specific objectives for AMR surveillance, target populations, target bacterial pathogens, and their laboratories' available testing capabilities and resources, before designing an appropriate sample processing and bacterial isolation/identification protocol suitable for their needs. In view of the fact that the existing format of the protocols currently presented in (e.g. flowcharts) are (1) inadequate in addressing the differing needs of each country, and (2) may be misinterpreted as a mandate to follow the stated protocols, these sections should be re-organized. Suggestions to improve the sections include (i) providing a list of guiding questions/considerations to help in designing the protocol, (ii) references for commonly used methodologies, as well as (iii) a list of contacts of experts available to advise the laboratories in designing a suitable protocol. Some common needs/concerns can also be consolidated from the countries and appropriate references to methodologies/guidelines provided to address those issues (e.g. need for non-molecular techniques for bacterial identification, non-selective agars for isolation, number of colonies to be tested for AST)
- **Antimicrobial susceptibility testing (AST).** While additional clinical breakpoints may be developed in the future, there is a need to rely on currently available epidemiological cut-off values (ECOFFs/ECVs). It is critical to carry out testing under standardized conditions with QC, whenever available, that will allow for additional ECOFFs/ECVs to be established. Participants also agreed that: (1) disk diffusion, which is more economically feasible, should be included in the methodologies; MIC testing may be ideal, but optional methodology, as carrying this out will depend on the currently available laboratory capacity and resources; (2) laboratories need to follow standard procedures published by CLSI and guidelines such as EUCAST's while conducting AST. Participants discussed CLSI standards and incubation parameters in generating ECVs and QC data for listed prioritized bacterial pathogens. There is a need to decide whether countries should perform AST only at $\leq 28^{\circ}\text{C}$ generating results relevant to animal health (and establishing local ECOFFs/ECVs when internationally-validated ECOFFs/ECVs are unavailable), or to perform AST at 35°C as more ECOFFs/ECVs are already available.

However, interpretations using clinical breakpoints developed in humans may not be very predictive of clinical outcome in aquatic species.

- **Organizing a network of AMR surveillance laboratories for aquaculture in Asia.** One clear limitation recognized is the lack of standardized AST methods, QC, and interpretive criteria (ECOFFs/ECVs, clinical breakpoints) for testing many of the aquatic bacterial pathogens. This is particularly important given that this guideline will place emphasis on linking AMR surveillance with ultimately improving antimicrobial use in aquatic species, as well as improving production. Recognizing the value of an internationally harmonized AST data set, until additional international standards or guidelines are written, it was proposed by FAO RAP to establish a network of aquaculture AMR testing laboratories in Asia. This guideline will be instrumental in harmonizing and standardizing methodologies in view of this important cooperation. FAO RAP will take the coordination role to initiate this regional collaborative work.
7. Chapter 5 discussion was led by Kar Hui Ong, who opened the session with a brief presentation on data collection, compilation, and interpretation. The starting point of the discussions were the comments received in advance. The group recommended the following:
- **Minimum dataset as the regional template.** The participants proposed to add to the minimum dataset (originally created during the 1st consultation) the following information: raw data (e.g., diameter of zone of inhibition in disk diffusion), and GPS coordinates of sample collection points.
 - **Revision of the tables for data summary.** The tables for data summary was largely drawn from Guideline #1 and may not be best suitable for the anticipated use in aquaculture. Individual tables for each bacterial species was proposed. Adaption of the Regional Template developed originally for livestock was also proposed in anticipation of data analysis using the WHONET platform in the future. Countries were requested to share their current templates to ensure integration of their data types into the regional template.
 - **Alignment of the regional template with WHONET configurations.** As FAO RAP is currently working with WHONET to improve AMR data management in food and agriculture, the regional template will thus be designed to align with WHONET configurations to allow data following the template to be exported to WHONET platform with ease via baclink.
8. Additional sections to the guideline were also proposed and supported by the participants:
- A chapter to cover aspects on operationalizing AMR surveillance in aquaculture will be added (Chapter 6: Operationalization). This additional chapter will cover planning, consultation process, incentivization, setting out roles and responsibilities, costing, essential regulatory frameworks, addressing potential biases, and other matters pertaining to operationalizing AMR surveillance in aquaculture will be included.

- An introductory section/preface will also be included to provide a more replete overview of the broader scope of AMR surveillance in aquatic animal species. This will also include a clear emphasis on the specific scope that the guideline covers.
 - An additional annex to include list of relevant experts, resources, and available technical support to the countries will be added to help countries find support as they commence AMR surveillance in aquaculture.
9. As discussions evolved in the last four days, it became clear that providing guidance to aquaculture AMR surveillance is complex given the (1) variety of aquatic animal species; (2) variety of aquatic bacterial pathogens; (3) exposure of aquatic animals to an environmental continuum which implies AMR incidence cannot be attributed to actual antimicrobial use alone for commensal bacteria; (4) varying approaches depending on the purpose of AMR surveillance in aquatic animals – whether for protecting human health, or for protecting animal health and production. There is therefore a need for the guideline to be more specific; and (5) current gaps in standard methods and QC for findings to be meaningful for treatment guidance. Given these, the group agreed that:
- The guideline will particularly limit its scope to cover AMR surveillance in bacterial pathogens in aquaculture species, with the data to be generated intended for use in improving antimicrobial use in aquaculture.
 - Because there are still various gaps on available international standards, the region will initially build an internationally harmonized AST data set using standardized methods and QC to contribute to the establishment of ECOFFs/ECVs by the appropriate international standard-setting body (CLSI). Laboratories will be encouraged to work closely with country representatives and/or members of the CLSI Aquaculture Working Group on AST to help in this process.
 - Countries who may be interested to initiate AMR surveillance in aquaculture for the same purpose may refer to CLSI standards which will allow them to be regionally harmonized, and therefore can opt to contribute their data to the regional pool of information that will then serve as the data set to establish new ECOFFs/ECVs.
 - The AMR surveillance initiatives will be closely linked with disease surveillance and will therefore also optimize the opportunities for building capacities and stakeholder engagement.
10. To close the four-day virtual sessions, Angela Li (Director/Research & Risk Assessment of Singapore) and Dr Melba Reantaso (Fishery Resources Officer, FAO Headquarters, Rome, Italy) provided closing remarks. Both acknowledged the rich contributions from, and strong engagement of, participants and experts from across the world.

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