Food and Agriculture Organization of the United Nations

Meeting report

Regional consultation on environmental surveillance for zoonotic influenza in Asia

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14-16 November 2023

Bangkok, Thailand

Contents

Acknowledgements

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Abbreviations and acronyms

AI	Avian influenza
AIV	Avian influenza virus
COVID-19	Coronavirus disease 2019
ECTAD	Emergency centre for transboundary animal diseases
ENVT	*École Nationale Vétérinaire de Toulouse*
ES	Environmental sampling
FAO	Food and Agriculture Organization of the United Nations
HPAI	High pathogenicity avian influenza
IPC	*Institut Pasteur du Cambodge*
LBM	Live bird market
LPAI	Low pathogenicity avian influenza
PCR	Polymerase chain reaction
RAP	Regional Office for Asia and the Pacific
SOP	Standard operating procedure
USAID	United States Agency for International Development
WOAH	World Organisation for Animal Health
WGS	Whole genome sequencing

Executive summary

Background

In recent years, high pathogenicity avian influenza (HPAI) spread globally, causing significant losses in poultry and wild birds, particularly in regions where the disease was previously absent. The re-emerging HPAI H5N1 subclade 2.3.4.4b, originating in Asia, reached Europe, Africa and the Americas. In addition, mammalian infections with avian influenza viruses (AIVs) have risen globally, affecting mink in Spain, seals in the United States of America, sea lions in Peru and Chile, and domestic cats in Poland and Republic of Korea. Human cases of avian influenza, including A(H5), A(H5N1), A(H5N6), A(H10N3), A(H3N2) and A(H9N2), have also been reported in the Americas and Asia between 2022-2023. The Food and Agriculture Organization of the United Nations (FAO) Emergency Centre for Transboundary Animal Diseases (ECTAD) has conducted consultations to address challenges posed by emerging zoonotic influenza, emphasizing the need for international guidance on environmental sampling for avian influenza. The global spread of HPAI, coupled with increasing mammalian and human cases, warrants close attention and rapid detection of zoonotic influenza in animals and humans including the use of innovative approaches such as environmental sampling. To respond to these challenges, FAO ECTAD in the Regional Office for Asia in the Pacific (RAP) organized a consultation of experts to discuss environmental sampling (ES) for the detection of zoonotic influenzas in Asia, specifically highlighting its benefits, challenges and operational uses for countries.

Objectives of the meeting

The objectives were to:

1. Update current information on epidemiology, genotypic, and antigenic aspects of AIVs viruses in Asia;
2. Identify advantages/disadvantages of environmental surveillance for influenza viruses and other priority pathogens based on specific field experiences;
3. Define strategies to operationalize environmental sampling for influenza, including use of systematic approaches, cost-benefits and data analysis for governments' decision-making; and
4. Review FAO's Environmental Sampling Guidance for Avian nfluenza (AI).

Meeting format, venue and participants

The consultation was held from 14 to 16 November 2023 at Athenee hotel in Bangkok, with the opportunity to join virtually. Ten experts attended in person, four of which travelled to Bangkok from Australia, India and Lao People's Democratic Republic, while 15 participants attended online.

Programme of the consultation

The meeting was planned as a series of five interactive sessions to meet the objectives outlined above:

1. **Session 1: Introduction and setting the scene.** introductory presentation on meeting objectives, expected results and programme were provided to participants, in addition to update on global

overview of AI situation in Asia, most notably related to recent updates on H5 2.3.4.4b and human cases in the region.

2. **Session 2: Learning from the field.** Panellists shared their experiences and lessons learned in implementing environmental surveillance for influenza viruses at country-levels, specifically focusing on the advantages and disadvantages of the approach. Following the panellists' presentations, a facilitated exercise with participants further defined benefits and barriers to environmental sampling, including elements of sensitivity, specificity, costs-benefits, acceptability, multi-pathogen approach and use of data for decision-making.
3. **Session 3: Overcoming barriers, leveraging advantages, taking action.** Using the outputs of the previous sessions, participants will identify strategies to enhance the operationalization of environmental sampling for influenza viruses and other priority pathogens, based on specific surveillance objectives, and linking with recommended actions for governments to take based on surveillance results. Steps to operationalize environmental sampling for AI at country levels were also discussed.
4. **Session 4: Feedback on FAO's Environmental Sampling Guidance.** Based on the outputs of the preceding discussions, experts provided input on the current draft of FAO's Guidance on Environmental Sampling for AI. It was proposed to adapt this current draft into a series of guidelines, focusing on specific aspects of ES sampling, including: 1) general considerations, 2) surface sampling, 3) air sampling, 4) water sampling, etc.
5. **Session 5: Conclusions and ways forward.** A final discussion with participants was conducted to summarize the outputs of the meeting and next steps.

Outputs and next steps

Participants discussed the main surveillance objectives for which ES sampling can be used, consisting of monitoring of pathogen presence and trends. This can be utilised to inform risk-based approaches, detect incursion of pathogen early on, or inform disease control interventions. On the other hand, ES may not be as adapted for traceback during outbreak investigations, determining epidemiological prevalence/incidence, or understanding factors linked to individual animals.

A diagnostic flowchart was developed, ranging from approaches using polymerase chain reaction (PCR) assays can be used for targeted testing, to metagenomic assays for more broad, multi-pathogen testing.

During the concluding session, experts discussed the next steps to continue supporting countries in operationalising ES approaches to surveillance. The following steps were identified:

1. **Reviewing of existing evidence and practices** – specifically linked to sensitivity/specificity of ES, cost-benefit, comparative studies of different approaches.
2. **Piloting ES approaches at country level** – alongside relevant partners (e.g. government, private sector, etc.) to improve acceptance of practice.
3. **Provide situation-specific guidance** – adapted to countries contexts and using a stepwise approach based on capacities. This can be supported by country-level consultations bringing together experts as well as policy-makers.

Background and objectives

Background

In recent years, HPAI has swept across the globe, with several incursions into new geographic regions such as the Americas. This has led to significant losses in poultry and wild birds, especially in those areas where animals were naïve to the disease. This expansion has largely been driven by the re-emerging HPAI H5N1 subclade 2.3.4.4b, which originated in Asia before spreading to Europe, Africa and finally the Americas.

In Asia, several endemic HPAI subtypes circulate in both poultry and wild birds, including HPAI H5N6, H5N8, H9N2 and others. There is evidence however, that H5N1 subclade 2.3.4.4b is returning to the region, introduced into East Asia by wild birds migrating from the Americas using the West Pacific or East Asian migration routes. This puts domestic and wild bird populations in the region at risk, due to the potential introduction of antigenically different HPAI subtypes with genes acquired in Europe or North America.

In addition, a rise in mammalian species infected with HPAI has been recorded globally including outbreaks in farmed mink in Spain, seals in the United States of America, and sea lions in Peru and Chile, and more recently in domestic cats in Poland and the Republic of Korea (WHO, 2023; FAO, 2024).

During the period of 2022-2023, AI has also been detected in humans, including cases of influenza A(H5) in the Americas in the United States of America (April 2022), Ecuador (January 2023) and Chile (March 2023). Several cases have been noted in Asia (WHO, 2024), most notably:

- A(H5): in Viet Nam (October 2022) and Cambodia (two cases in February 2023)
- A(H5N1): in Cambodia (October and November 2023)
- A(H5N6): in China (July 2023)
- A(H10N3): in China (June 2022)
- A(H3N2): in China (February 2023)
- A(H9N2): in China (June 2023)

Lastly, while AI remains the focus of international attention due to its zoonotic risk and impact on wild and domestic birds, little is known about the prevalence and epidemiology of non-avian influenzas (i.e. swine) in the region. There is evidence that influenza viruses from non-avian species can be a source of infection to humans, as seen with the elevated seroprevalence to Eurasian avian-like (EA) H1N1 in swine workers in China (Sun *et al.*, 2020).

The global spread of HPAI coupled with the increase in mammalian and human cases is worrisome and close attention to this evolving situation is needed to rapidly detect unusual cases of zoonotic influenza in both animals and humans.

The FAO ECTAD office in RAP has conducted regular technical consultations with both human and animal influenza experts to address the challenges posed by emerging and re-emerging zoonotic influenza subtypes in the region, and guide future early warning/early response strategies. In February 2021, an initial online consultation identified gaps and areas to address related to non-avian influenzas in Asia. This was followed by an additional online meeting in the same year, where experts developed a framework for an "ideal early warning" system linking AI surveillance at the field with feedback mechanisms from regional and global networks to facilitate countries' response to HPAI threats. In 2022, experts met in person in Geelong, Australia to link specific components of this early warning framework with innovative approaches to facilitate surveillance data generation, analysis and dissemination. During this last meeting, participants identified the lack of international guidance for ES for AI, limiting its implementation at country level.

Building upon the outputs of the three previous consultations, FAO ECTAD RAP brought together experts once more to focus discussions on the use of environmental sampling for the detection of zoonotic influenzas in Asia, specifically highlighting its benefits, challenges, and operational uses for countries. Outputs of this consultation will directly inform the finalisation of a "Guidance on Environmental Surface Sampling for Avian Influenza Surveillance" under development by FAO headquarters, therefore directly addressing one of the needs identified in Geelong. Funding for this consultation was provided by the OSRO-GLO-302-USA project, financed by the United States Agency for International Development (USAID).

Objectives

The objectives of the consultation were to:

1. Update current information on epidemiology, genotypic, and antigenic aspects of AI viruses in Asia;
2. Identify advantages/disadvantages of environmental surveillance for influenza viruses and other priority pathogens based on specific field experiences;
5. Define strategies to operationalize environmental sampling for influenza, including use of systematic approaches, cost-benefits and data analysis for governments' decision-making;
6. Review FAO's Environmental Sampling Guidance for AI.

Date and venue

The consultation was planned for 14-16 November 2023, and invitations were sent in early October. Due to several competing missions and meetings around the same period, it was decided to plan this meeting in hybrid format, to allow the opportunity for participants to contribute to the consultation, or parts of it virtually, and activities were designed to facilitate both in-person and online contributions. In-person participants met at The Athenee Hotel in Bangkok, Room Atheneum, and a Zoom link was shared with all registered participants for the three days of the meeting.

Participants

A registration form was distributed to 20 prospective participants to express their interest in attending the consultation, either virtually or in-person. They were selected based on their expertise in AI epidemiology, virology or diagnostics in the Asian context, as well as from countries where environmental sampling for AI is conducted or may be conducted in the future. Fourteen responses were received from experts interested in attending.

Actual participants attending the event consisted of ten in-person experts, four of which travelled to Bangkok from Australia, India, Lao People's Democratic Republic arranged by FAO, and 15 participants who attended online (Fig. 1). A detailed participant list is available in Annex 1.

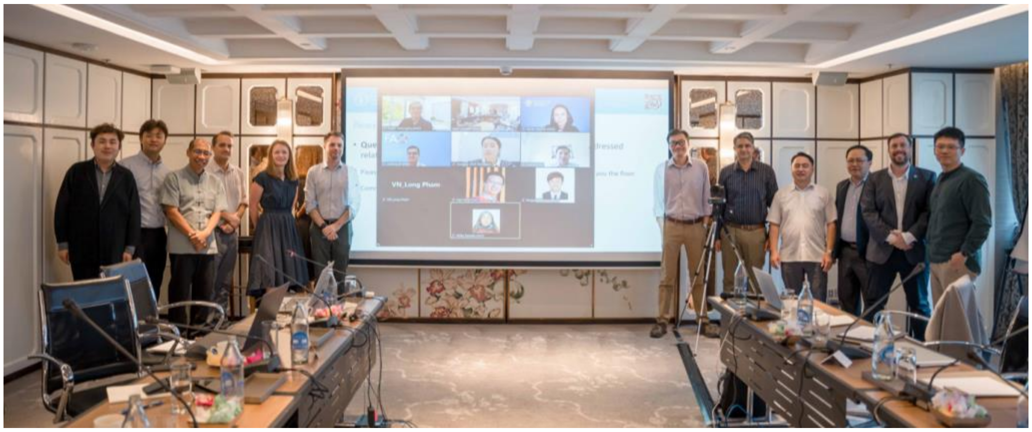

Figure 1. Participation of both in person-and online experts during the consultation. (© FAO)

Programme of the consultation

The meeting's programme was broken down into five specific sessions, building upon each other through facilitated activities to gather the experts' inputs and meet the meeting's objectives (Annex 2).

Session 1 – Introduction and setting the scene (14 November)

The initial session of the meeting consisted of presenting the meeting's objectives and introducing the participants, followed by presentations to update attendees on the current zoonotic influenza situation globally and in Asia.

Presentation: Update on zoonotic influenza situation (Gaël Lamielle, FAO ECTAD RAP Regional Surveillance Coordinator)

The presentation highlighted the shift in HPAI epidemiology at a global level, and the spread of H5 viruses around the world, including its introduction into naïve regions such as the Americas, largely facilitated by the dominance of H5 subclade 2.3.4.4b. In addition, recent years have shown that the virus has been increasingly detected in wild and domestic mammals including farmed minks, cats, marine mammals and more. The zoonotic potential of AI viruses also remains present and human infections with influenza A have occurred in Europe, the Americas and Asia.

In Asia, several subclades continue to circulate, including H5 2.3.4.4b, 2.3.2.1c and others – which can lead to reassortment and the appearance of viruses with new characteristics. Cases in humans have also been regularly reported, such as in Cambodia in February and October 2023. This constant presence of AI in the region and the threat of emerging subclades continues to threaten the health and livelihoods of communities in Asia and the Pacific.

(Link to slides available here)

Plenary discussion

Following the initial presentation, participants were invited to comment in plenary on any other important topics that should be addressed related to zoonotic influenzas in Asia. Forty minutes were dedicated to this discussion, which touched on the following topics.

1. *Surveillance and reporting of other AI subtypes, including low pathogenicity avian influenza (LPAI)*
    a. LPAI is still not reportable to the World Organisation for Animal Health (WOAH), therefore there is limited focus on these viruses at country-level.
    b. H9N2 has zoonotic potential, as seen in China: https://www.cidrap.umn.edu/avian-influenza-bird-flu/who-confirms-3-h9n2-avian-flu-cases-china-plus-2-h1n1v-infections, therefore, we should prioritize influenza surveillance based on the risk, rather than only H5 or H7.
    c. ES can support testing for other subtypes.
2. *Diagnostic algorithms and protocols*
    a. Current standard diagnostics for AI in Asia focus on a series of PCR assays to identify the subtype. As whole genome sequencing (WGS) technologies become more readily available, it would be possible to use PCR only to screen the sample (i.e. influenza A) followed by WGS, which is subtype-agnostic.
    b. This approach can also be useful for ES methodologies.
    c. There are some questions related to ES when samples are contaminated (e.g. mud, faeces) and whcih sample treatment techniques should be used – these protocols need to be clarified before moving to issues related to diagnostics.
3. *Spatiotemporal distribution of AI*
    a. Temporal analyses of AI in different regions can be useful to identify new trends and patterns in virus emergences, especially considering viral amplification in local domestic bird populations. These analyses can be easily generated using global disease databases such as Empres-i+.
    b. As HPAI has reached Antarctica (https://www.bas.ac.uk/media-post/first-confirmed-cases-of-avian-influenza-in-the-antarctic-region/), it is possible that the risk of virus introduction may come from the south – especially putting Australia at risk.
    c. Information from Antarctica is still new, and there are a lot of unknowns, some birds may help virus hop in stepwise manner rather than long distance migration.
4. *Wildlife/wild bird surveillance*
    a. Wildlife impact in Europe vs. Asia:
        i. In Europe/the United States of America, about 70 percent of notifications are related to wildlife and wild birds due to manageable control mechanisms in poultry.
        ii. The situation in Southeast Asia is unclear, with unknown impacts on wild birds. Possible factors include lack of reporting, investigation, or laboratory follow-up for wildlife mortality events.
    b. Dynamics of flyways and poultry practices in Southeast Asia:
        i. There is limited information on the different dynamics for wild bird flyways in Southeast Asia.

    ii. Poultry-raising practices in Southeast Asia are considered drivers for the emergence of avian influenza.

c. Challenges in surveillance in Lao People's Democratic Republic and Cambodia:
    i. Need for a flyway study before conducting surveillance in Lao People's Democratic Republic.
    ii. Active surveillance in Cambodia has low virus yield in wild birds; trapping may not provide an accurate picture.
d. Rethinking surveillance approaches:
    i. Risk-based surveillance is used for poultry; similar considerations are needed for wild birds.
    ii. Emphasis on sampling at interfaces between domestic and wild birds, such as live bird markets (LBMs) and water bodies.
    iii. Advocacy is needed for a targeted, risk-based surveillance approach, considering specific high-risk bird species.
e. Results from previous surveillance efforts:
    i. Previous wildlife surveillance efforts in 2005 and 2009 yielded limited results, with challenges in surveying live wild birds.
f. India's experience with AI surveillance:
    i. India's surveillance plan involved active and passive surveillance in wild birds and poultry and H5 avian influenza was detected in wild birds during an outbreak during late 2020 and early 2021.
    ii. Emphasizes the challenge of blanket surveillance and the importance of understanding the implications of positive findings.
g. Linking surveillance with response and responsibility:
    i. Consideration of whether wild birds fall under environmental surveillance.
    ii. There should be emphasis on linking surveillance with a response and identifying responsible departments with adequate laboratory networks.
h. Risk-based surveillance and syndromic events:
    i. Advocacy for risk-based surveillance and follow-up on syndromic events.
    ii. Challenges in determining responsibility and testing samples in Southeast Asia.
i. Examples of surveillance practices from other regions:
    i. Belgium imposes control measures based on surveillance results, acknowledging the risk.
    ii. Kenya conducts surveillance along the Rift Valley during migratory seasons.

In summary, the discussion emphasized the need for a targeted and risk-based approach to wild bird surveillance, considering regional dynamics and challenges in sample collection and testing.

Presentation: Review of previous consultations (Gaël Lamielle)

A summary of previous expert consultations organized by FAO ECTAD RAP was provided to participants:

1. **Non-avian zoonotic influenza in Asia (January-February 2021) – online**

a. Objectives – to jointly identify:
    i. Stakeholders of non-avian influenza in Asia
    ii. Major gaps/needs to inform future activities
    iii. Priorities for regional coordination for surveillance and research

2. Toward mitigating pandemic influenza risk (December 2021) – online

a. Objectives:
    i. Discuss AIV surveillance activities, including lessons learned from previous surveillance and the current coronavirus disease 2019 (COVID-19) response to be used in AIV early warning.
    ii. Determine major gaps and needs of AIV surveillance, and how they can be addressed.
    iii. Discuss novel technologies and sampling strategies for improved surveillance.
    iv. Explore possible utilisation of AIV surveillance data, including the coordinated animal sector AIV vaccine composition discussion/forum.

3. Better detection, better response (November 2022) – in person in Geelong, Australia

a. Objectives:
    i. Review progress and challenges related to global AI early warning mechanisms & poultry vaccine composition meeting.
    ii. Discuss novel approaches that can support faster detection and response to AI threats, including advancements in next generation sequencing and bioinformatics.
    iii. Discuss progresses towards poultry vaccine composition meeting.

Summarized priorities from the previous consultations were then presented to the group:

1. Identification of risk factors for emergence and spread of zoonotic influenzas – multiple-species value chains, wild bird migration models, epidemiological role of different species including wildlife in zoonotic influenzas.

2. Providing global guidance, validation and best-practices for risk-based surveillance of zoonotic influenzas (including non-avian), including novel approaches.

3. Routine use at country level of innovative technologies and methodologies to improve surveillance, risk assessment, genetic data analysis and interpretation.

4. Enhancing information-sharing and feedback mechanisms within and between countries.

5. Establish a global platform to advise on vaccine composition for animal influenza vaccines.

6. Integration of big data and artificial intelligence into predictive and early warning systems.

(Link to slides available here)

Group activity 1: progresses and remaining gaps related to priorities from last consultations

In this first group exercises, participants were invited to review the priorities above, discuss whether progresses have been made in the past two years, and select which priority is most important to them to focus efforts on. These activities included feedback from both in-person and online participants through the Mentimeter platform.

1. Progresses

Participants were asked to select if they thought progress had been made addressing the six priorities listed above, selecting more than one option if appropriate. Results of this voting process are highlighted in Figure 2.

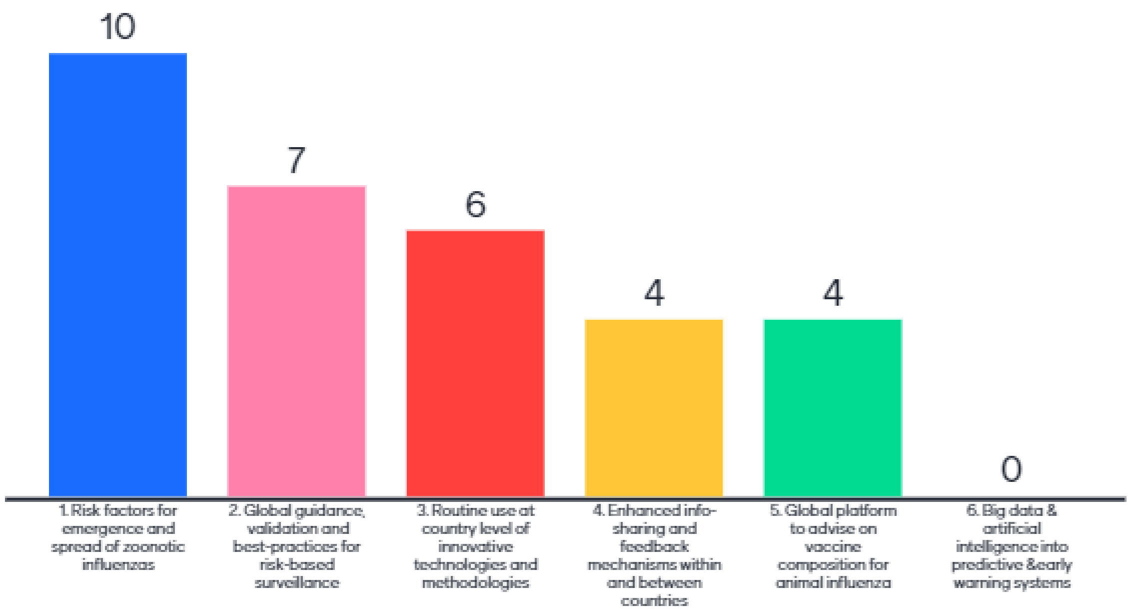

Figure 2. Priorities from previous consultations for which progress has been made in the past two years prior to the consultation. (© FAO)

2. Why some priorities have not been addressed?

Participants were invited to provide brief wording on why priorities may not have been addressed and results were generated in the form of a word cloud (Fig. 2), with ideas related to "Funding" and "Competing priorities" receiving the most input.

Figure 2. Reasons why priorities from previous consultations have not been addressed. (© FAO)

3. Which should be the top priority to address next year?

As a last part of this group exercise, participants were asked to select one priority that they think is most important to work on in the next year. An option for "Other" was incorporated (Fig. 3).

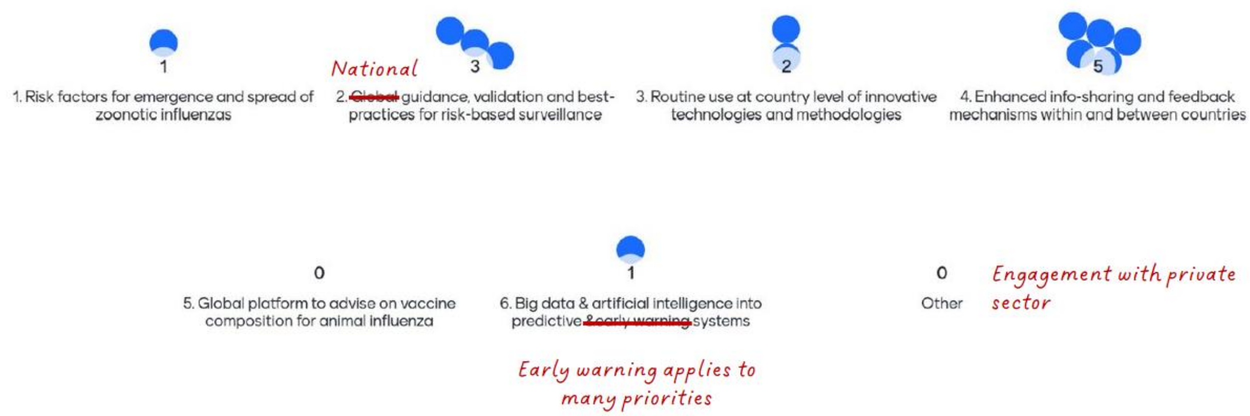

Figure 3. Top priority to address within the next year, and feedback from participants (red). (© FAO)

Though no other priority was noted at the time of voting, follow-up discussion indicated the need to engage with the private sector for early warning of avian influenza. In addition, participants suggested to focus on "national" instead of "global" guidance for priority 3 and opted to remove the term "early warning" from priority 6, as this concept is cross-cutting.

Additional discussion points proposed a paradigm shift related to testing samples collected from the environment. Specifically, participants suggested introducing simplified field sampling processes, acknowledging existing technology barriers, particularly governmental restrictions on cloud-based data uploads. Information sharing concerns centred on incentivizing farmer reporting, addressing legal frameworks, and finding a balance between transparency and privacy. Despite resistance from the private sector due to commercial reasons, there was a recognition that advocacy and inclusion efforts may encourage sharing, exemplified by positive experiences in Indonesia with both the private sector and academia participating in data-sharing initiatives.

Session 2 – Learning from the field (14 November)

Following a lunch break, a round table of discussions with both online and in-person participants was conducted, so that they can share experiences with environmental sampling for surveillance of zoonotic influenza in their countries. Summaries from the discussion with each contributor are provided below.

Cambodia

AI surveillance in Cambodia is conducted through sampling at LBMs, where 20 ducks and 20 chickens throughout the year, and specifically one week before, during, and one week after festivals. Individual samples are collected, with subsequent pooling in the lab. In addition, a pilot project is conducted, in collaboration between FAO and *Institut Pasteur du Cambodge* (IPC) to validate different environment sampling and diagnostic methods, exploring sampling options such as wastewater and air samples. Using diagnostics such as Aerocollect, Thermofisher, multiplex, singleplex and nanopore techniques, demonstrate superior results compared to traditional approaches individual animal sampling. These novel approaches present the opportunity for pan-viral diagnostics, extending beyond avian influenza.

École Nationale Vétérinaire de Toulouse (ENVT)

The team from ENVT conducted an analysis to assess the effectiveness of ES compared to sampling ducks and chickens in Viet Nam. The study utilized data from 2019 to 2022, focusing on obtaining informative results. The data were subjected to modelling to estimate sensitivity, specificity, and the prevalence of AIV subtypes. The findings indicated that duck samples outperformed environmental samples, which, in turn, outperformed chicken samples for all tested A(H5) subtypes. A key question raised was how to ensure the quality of sampling, with the acknowledgment that collecting faecal and environmental samples is a straightforward method, minimizing potential issues in the sampling process.

(Link to slides available here)

India

In India, environmental sampling for AI is performed following a routine action plan. Samples taken for testing include cage swabs/knife swabs, droppings in the cages and wastewater from live bird markets, drinking water (various points of water source/drinkers etc.), droppings in the cages and waste water from organized and backyard farms/units, water from waterbodies nearby the farm (water along with the sediments collected at the banks from various points) are also collected.

Indonesia

In Indonesia, the sampling strategy involves collecting 5-6 swabs from various sources, including and faeces, with potential variations in sensitivity. LBMs are identified as the most crucial source for virus samples, with the ability to extract the virus from environmental samples; remarkably, 60 percent of PCR-positive samples can be cultured. However, the challenge lies in traceability within LBMs, making it impossible to determine the origin of the virus. To identify the virus's source, one must investigate farms rather than relying on traceability within the market.

Viet Nam

Surveillance efforts have been conducted using government protocols, focusing on high-risk provinces with previous outbreaks, targeting ducks, chickens, and environmental samples for H7 and H9 using penside PCR along the border. The primary goal is to monitor the circulating avian influenza (AI) virus, enabling early detection of new subtypes. Additionally, collected virus samples are used for vaccine efficacy testing, informing recommendations on vaccines. This approach benefits provincial authorities by facilitating planning and budgeting for control and response efforts, covering 26 provinces. The surveillance objectives emphasize the supplementary role of environmental sampling to ensure comprehensive coverage, although results mainly stem from chicken or duck samples. Decision-makers act based on the obtained results, with a primary utility in formulating recommendations for vaccine development, including isolate and vaccine testing. Notably, despite ongoing penside PCR activities, the first detection of H5N8 occurred 200 km from the border and not within the market.

Lao People's Democratic Republic

Following FAO guidelines, surveillance in Lao People's Democratic Republic consists of the collection of 30 oropharyngeal samples from both chickens and ducks, along with 5 environmental samples. Upon detecting positive results, the information is relayed to the Department of Communicable Diseases for further investigation into traders to identify potential issues. The animal sector then implements control measures. Notably, in LBMs, where birds are housed together, it becomes challenging to discern the origin of sampled faeces, leading to a lack of traceability in the environmental sampling process.

Other comments

Concluding points were made during the discussion, specifically:

- Sampling can be done from underneath cages so that the species can be noted. In addition, genetic materials in samples can be used to identify species environmental samples also though it is not that specific.
- Air sampling locations should be based on high to low risk – slaughtering/defeathering area highest amount of virus, then where birds are kept, then sales areas. Ongoing projects are currently validating which is the best size and position for testing so that standard operating procedures (SOPs)/guidance documents can be developed.

Group activity 2: successes from environmental sampling

Based on the previous round-table discussion, participants were invited to provide their thoughts on successes from environmental sampling. Online contributors used Mural, while in-person participants provided their input on flip charts (Fig. 4).

Figure 4. In-person and online input during Activities 2 and 3. (© FAO/Gaël Lamielle)

A summary of major successes identified during the activity is included below:

- **Efficiency** – faster, easier, lower cost, less workload, requires minimal training, accumulation of viruses.
- **Acceptability** – animal welfare, less invasive, acceptable for traders, less workload for field staff and requires minimal training, biosafety for sampling personnel, anonymity.
- **Effectiveness** – virus isolation/sequencing, comparable with live animal sampling, relatively good sensitivity, more representative of what is in market/farm than if sampling animals themselves, comprehensiveness.
- **Opportunities** – can use for continuous sampling, can be used for high-risk areas, can be used for free-ranging birds or wild birds, can use for AMR etc.

Group activity 3: challenges of environmental sampling

Challenges were also identified using the same format as the previous exercise, as summarized below:

- **Validity** – sensitivity and specificity, may be more difficult to isolate and sequence viruses, risk of dilution, inhibitors, contamination, validation, agnostic approaches may compromise Se, lacks gold standard/standardization.
- **Practicality** – requires purchase of air sampling machines, potentially loud and disruptive if noisy, expensive sampling kit, needs field staff training.
- **Consistency** – reduced consistency between LBMs or between samplers etc., large heterogeneity in sampling methods without explicit instructions, SOPs, changing environment.

- **Traceability** – trace-back ability of positive samples lacking.

- **Utilization of results** – decision in case of positives, interpretation of results, no baseline data, acceptability by trade partner/international community, may not be able to link with host data.

Conclusions of day 1 and additional discussion points

Prior to closing of day 1, a few final comments were provided by the participants:

- Considerations regarding data sharing involved assessing the benefits for data sources, such as livestock owners, and exploring the incentives for sharing "good" information. Questions arose about the quantity of data to share, with a need to define the minimum required information.
- Challenges stemmed from a lack of shared goals between public and private stakeholders regarding information sharing.
- The delineation of responsibilities between those in charge of wildlife at the country level and those overseeing diagnostic labs became a crucial aspect.
- There was also a recognition of the potential threat of highly pathogenic avian influenza (HPAI) spreading within the Antarctic region, particularly from the south.
- To enhance efficiency, there was a proposal for an improved diagnostic algorithm, suggesting a streamlined process from flu A testing directly to sequencing.

Session 3 – Overcoming barriers, leveraging advantages, taking action (15 November 2023)

The second day of the meeting was dedicated to group activities, which started following a brief review of the previous day's outputs and introduction of the agenda for day 2.

Group activity 4: defining the objectives

Using the Mural platform, online and in-person participants discussed which surveillance objectives could be met by ES, as well as which objectives cannot be supported by ES. Results of the activity are summarized below (Fig. 5).

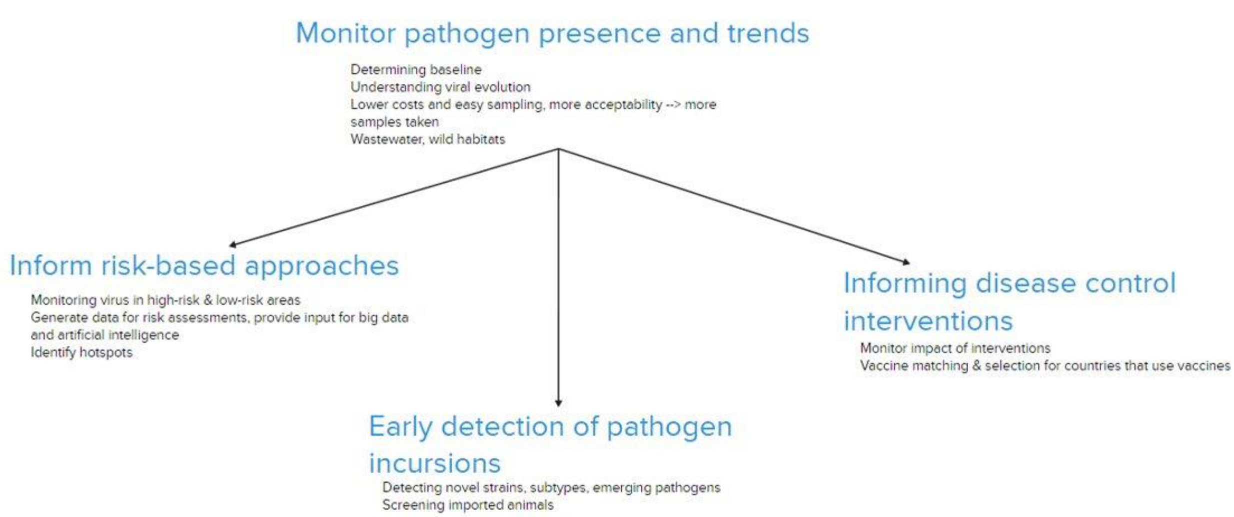

Figure 5. Surveillance objectives for zoonotic influenzas that can be supported by environmental sampling. (© FAO)

In addition, the experts identified some objectives that cannot be supported by environmental sampling – including:

- Outbreak investigation, specifically traceback (i.e. source of pathogen) – some assumptions can be made based on local knowledge of context, especially value chains;
- Determining epidemiological prevalence and incidence of infection in animals;
- Identifying full genome directly from environmental sample, especially when sample have mixed results detected; and
- Individual animal factors, including confirmation of infection, linking presence in environment with severity of disease, co-infection, absence of infection in animals.

Group activities 5-7: determining approaches to implement ES

Initially, a series of activities were planned to identify approaches to implement ES based on objectives defined in group activity 4. These approaches focused on:

1. **Where** - Which sites should be sampled to meet the objective? (e.g. LBMs, farms, wild areas, etc.)
2. **What** - What areas should be sampled (cutting boards, intake, etc.)? Which samples to take? (e.g. feathers, water, faeces, etc.)
3. **How** – Approaches to maximize the sensitivity of pathogen detection (e.g. pooling samples, mixed live/env sampling, etc.)
4. **Diagnostics** – What assays exist that can support this approach?
5. **Considerations** – what can support roll out of these diagnostic tools at country level?

During the exercise, however, it was clear that these elements were not specific to each objective identified and an overarching approach to these activities was chosen.

Table 1. Areas to sample and samples to take to implement ES

Which areas to sample?	Which samples to take?

• Areas should be based on risk assessments linked to country-specific objectives
• Value chain nodes
  ○ Live animal markets
  ○ Trucks, transport
  ○ Farms
  ○ Slaughter facilities
• Wildlife
  ○ Wildlife habitats, migratory sites
	• Surface samples – trucks/transport, cages, cutting board & utensil, fresh faeces, feathers, carcasses, bedding (e.g. farms)
	• Water – wastewater, standing water
	• Air – ventilation systems in markets, farms
	• Other – animal feed, rope sampling (swine/mammals)

○ Interface between domestic/wild – poultry open grazing areas ○ Hunter collector sites ○ Captive wildlife farms, breeding farms, zoo • Border posts – especially for early detection of pathogen incursions	• Specific sampling surfaces – e.g. tarps under cages/specific habitat to collect samples

Several diagnostics tools are available to countries to support testing of environmental samples, and the group classified them based on their scope (from specific to general) and including specific considerations for implementation of the techniques (Fig. 6.)

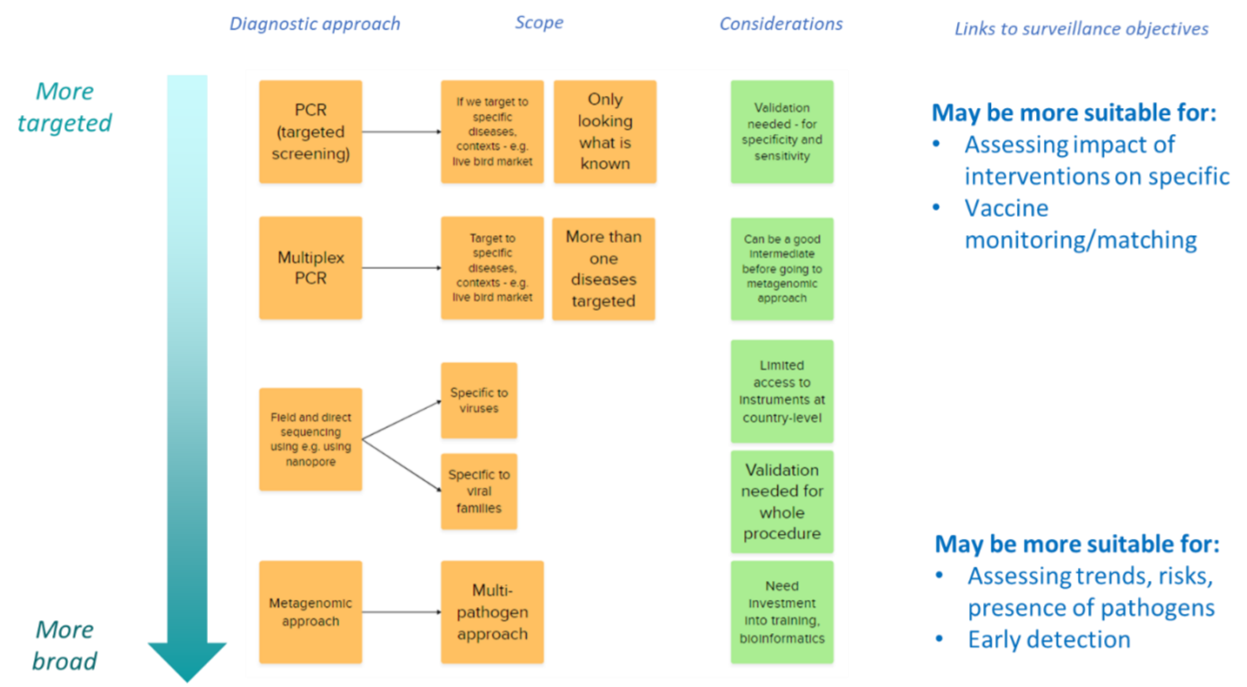

Figure 6. Diagnostic approaches to support testing of environmental samples. (© FAO)

Group activity 8: other strategies to improve ownership at country-level

A final facilitated activity for the day included bringing together all the discussions from the preceding sessions and identify some strategies to improve uptake of ES approaches at country-level. The group identified the following:

1. Reviewing existing evidence and practices
    a. Scientific evidence on ES approaches, including sensitivity/specificity, economic considerations
    b. Pros- and cons- of different approaches and diagnostic methods
    c. Countries practices of ES and sharing experiences/lessons learned

2. Generate more evidence
    a. Pilot ES approaches with government alongside traditional methods to improve acceptance, experience and uptake
        i. Partner with private sector
        ii. Stepwise approach for country ownership
    b. Prioritize what to test for and what not to test for, with feasibility in mind – e.g.:
        i. Endemic, priority reportable diseases, etc.
        ii. Diseases at risk of introduction/emergence
        iii. Other – human/zoonotic diseases, bioterrorism, etc.
    c. Conduct studies on cost effectiveness of ES approaches and diagnostic tools
3. Provide guidance to countries to support implementation of ES
    a. Develop a stepwise approach – from easy to complex
    b. Provide situation-specific scenarios, including stakeholders to notify – e.g. wildlife, markets, intersectoral partners
        i. Consider multi-disease approach
        ii. Incorporate mandates of different institutions and funding across all sectors (One Health)

Session 4 – Feedback on FAO's Draft Environmental Sampling Guidance (16 November 2023)

FAO is a draft guiding document to support countries in implementing ES approaches for surveillance of avian influenza. The objectives of this document are to:

1. Provide guidance for the use of ES collection for AI surveillance and may be adapted to countries' national contexts.
2. Specifically support veterinary and animal health authorities in planning, implementing or improving their active surveillance for AI using ES.

During the development process, the scope of the guidance became more specific to surface sampling, especially in the context of LBMs.

An initial draft provided by FAO headquarter team was circulated to FAO ECTAD RAP and some FAO ECTAD country offices with environmental surveillance experience.

This first round of feedback included the following points:

- Include a section on analysis and interpretation – extrapolation of results and conclusions that can be drawn from positive sample results, especially for endemic viruses
- Recommended actions based on the findings (e.g. scenarios)
- Decision tree to support choosing between ES vs traditional sampling methods
- Annex with example of SOPs with list of supplies needed in the field

Following an initial presentation introducing the proposed guidance to the participants, a facilitated discussion was conducted to get additional recommendations from the experts to ensure the guidance developed is relevant to country contexts (Table 2).

Table 2. Experts' feedback on FAO's Environmental Surveillance Guidance

<table>
<tr><th>Topic</th><th>Feedback</th></tr>
<tr><td>Scope</td><td>Some countries are already conducting surface surveillance so this would be useful to update country protocols, but definitely need to include water and air, up to FAO whether it is in one volume or three.
Propose to change the guidance for zoonotic influenza, not just for avian influenza, depending on surveillance objectives.
Should incorporate air and water into the guidance. The environmental sector could use guidelines, which can be more specific for wildlife etc. rather than focus on LBM.
One potential objective for use of ES is to enhance surveillance during outbreaks, specifically in buffer zones.</td></tr>
<tr><td>Structure</td><td>Propose to divide this guidance into a technical series of documents. Can have an initial guidance with overview of the series and then release volumes gradually.
Different volumes in the series can target specific aspects of ES, including:
Sampling and storage, transport
Surface sampling
Air sampling
Water sampling
Etc.
Different guidelines within the series can target specific audience, e.g. sampling storage and transport for field actors, diagnostics for laboratory personnel and actions to take for decision-makers.</td></tr>
<tr><td>Content</td><td>Clarify the pros and cons of each surveillance approach, including between different environmental and animal sampling at the beginning of the guidelines so that countries can decide which ES to do.
Protocols for water and soil will be different, different types of water – sewer, wastewater, drinking water. This can be addressed in specific follow-up guidance detailing these approaches.
Transport of samples – needs review by countries to ensure is it applicable to national context, also need to add sections on prevention from contamination.
Need to review the table on number of pools required to provide 95 percent confidence in detecting AIV, as this table seems to have been generated</td></tr>
</table>

	based on sampling of individual animals. Are these calculations relevant to ES? • Use Bayesian analysis to determine whether ES is comparable in sensitivity. • Can pool samples from same location but different surfaces. • Need to review PPE: won't wear PPE in markets, need to be aware of other pathogens, may not apply to other places in wildlife, need a risk-based approach. • Consider asking farmers to take samples to increase sample numbers.

Session 5 – Conclusions and ways forward (16 November 2024)

In the final session of the consultation, experts discussed the next steps to continue supporting countries in operationalising ES approaches to surveillance. Several strategies have been listed already in the preceding sessions, especially under Group Activity 8 above which identified the following steps:

3. **Reviewing of existing evidence and practices** – specifically linked to sensitivity/specificity of ES, cost-benefit, comparative studies of different approaches.
4. **Piloting ES approaches at country level** – alongside relevant partners (e.g. government, private sector, etc.) to improve acceptance of practice.
5. **Provide situation-specific guidance** – adapted to countries contexts and using a stepwise approach based on capacities. This can be supported by country-level consultations bringing together experts as well as policy-makers.

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Annex 1 – Participant list

No.	Name	Country	Organization	Gender (M/F)	Participation			Participation
					14-Nov	15-Nov	16-Nov	
1	Akiko Kamata	Italy	FAO HQ	F	X	X	X	Online
2	Angélique Angot	Italy	FAO HQ	F	X			Online
3	Artem Metlin	Italy	FAO HQ	M	X			Online
4	Bill Davis	Thailand	US CDC	M	X		X	In-person
5	Brandon Hayes	France	*École Nationale Vétérinaire de Toulouse*	M	X			Online
6	Dung Le	Viet Nam	FAO ECTAD Viet Nam	F	X	X		Online
7	Filip Claes	Thailand	FAO ECTAD RAP	M	X	X	X	In-person
8	Frank Wong	Australia	ACDP	M	X	X	X	In-person
9	Gaël Lamielle	Thailand	FAO ECTAD RAP	M	X	X	X	In-person
10	Hao Tang	Thailand	FAO ECTAD RAP	M	X	X	X	In-person
11	Karoon Chanachai	Thailand	USAID - RDMA	M	X	X	X	In-person
12	Long Pham	Viet Nam	Department of Animal Health	M	X			Online
13	Luuk Schoonman	Indonesia	FAO ECTAD Indonesia	M	X			Online
14	Makara Hak	Cambodia	FAO ECTAD Cambodia	M	X			Online
15	Manoj Kumar	India	ICAR-National Institute of High Security Animal Diseases	M	X	X	X	In-person
16	Martha Montgomery	Thailand	US CDC	F	X	X	X	In-person
17	Mugyeom Moon	Thailand	FAO ECTAD RAP	M	X	X	X	In-person
18	Pawin Padungtod	Viet Nam	FAO ECTAD Viet Nam	M	X	X		Online
19	Phouvong Phommachanh	Lao People's Democratic Republic	Department of Livestock and Fisheries	M	X	X	X	In-person
20	Rindu Putri	Thailand	FAO ECTAD RAP	F	X			Online
21	Sarah Van Dyk	Australia	FAO ECTAD RAP	F	X	X	X	Online
22	Sideth Dith	Cambodia	FAO ECTAD Cambodia	M	X			Online
23	Sokhim Ol	Cambodia	FAO ECTAD Cambodia	M	X			Online

24	Soubanh Silithammavong	Lao People's Democratic Republic	FAO ECTAD Lao People's Democratic Republic	M	X	X	X	In-person
25	Timothée Vergne	France	*École Nationale Vétérinaire de Toulouse*	M	X			Online
26	Thipphasone Vixaysouk	Lao People's Democratic Republic	USAID	M			X	Online