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A literature review of the use of environmental sampling in the surveillance of avian influenza viruses

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Abstract

This literature review provides an overview of use of environmental samples (ES) such as faeces, water, air, mud and swabs of surfaces in avian influenza (AI) surveillance programs, focussing on effectiveness, advantages and gaps in knowledge. ES have been used effectively for AI surveillance since the 1970s. Results from ES have enhanced understanding of the biology of AI viruses in wild birds and in markets, of links between human and avian influenza, provided early warning of viral incursions, allowed assessment of effectiveness of control and preventive measures, and assisted epidemiological studies in outbreaks, both avian and human. Variation exists in the methods and protocols used, and no internationally recognized guidelines exist on the use of ES and data management. Few studies have performed direct comparisons of ES versus live bird samples (LBS). Results reported so far demonstrate reliance on ES will not be sufficient to detect virus in all cases when it is present, especially when the prevalence of infection/contamination is low. Multiple sample types should be collected. In live bird markets, ES from processing/selling areas are more likely to test positive than samples from bird holding areas. When compared to LBS, ES is considered a cost-effective, simple, rapid, flexible, convenient and acceptable way of achieving surveillance objectives. As a non-invasive technique, it can minimize effects on animal welfare and trade in markets and reduce impacts on wild bird communities. Some limitations of environmental sampling methods have been identified, such as the loss of species-specific or information on the source of virus, and taxonomic-level analyses, unless additional methods are applied. Some studies employing ES have not provided detailed methods. In others, where ES and LBS are collected from the same site, positive results have not been assigned to specific sample types. These gaps should be remedied in future studies.

KEYWORDS

avian Influenza, environmental sampling, epidemiological monitoring, surveillance

1 | INTRODUCTION

Avian influenza viruses (AIV)[1] constitute a significant economic, animal and public health threat. Carefully planned and implemented epidemiological surveillance is a cornerstone of avian influenza preparedness and response, and also critical for the planning, implementation, and monitoring and evaluation of control and preventive programs.

Surveillance for AIV is recommended for all member countries of the World Organisation for Animal Health (OIE, 2018). However, considerable variation exists in the type and number of samples collected and the reasons for conducting tests. Among the samples collected are cloacal and tracheal/oropharyngeal swabs and blood (for serology) from live birds, swabs and/or organ samples from dead birds and various environmental samples (ES). ES include samples or swabs taken from faeces, mud, water, feeding-source, feathers, air and surfaces likely to be contaminated with virus such as cages in markets, chopping boards and defeathering machines.

Currently, avian influenza surveillance is carried out in diverse forms and to various extents across the globe, by a range of bodies including the governmental and inter-governmental organizations, non-governmental organizations (NGOs), research institutions, regional organizations and large-scale commercial farms (Pavade, Weber-Vintzel, Hamilton, Dehove, & Zepeda, 2009; Von Dobschuetz et al., 2015). Many of these authorities and bodies have incorporated ES as one of the elements of surveillance either in isolation or alongside other surveillance testing. ES provides a convenient approach—in addition to live and dead bird sampling—to assess complex AIV amplification sites such as wild bird habitats or live bird markets (LBMs) that gather a significant number of birds of different species from multiple origins. Increased use of ES has also been driven by the increasing knowledge on the role and importance of these sites in the amplification and the spread of AIV (Martin et al., 2011; Soares Magalhaes et al., 2010).

Overall, studies included in this review took place in different settings such as LBMs, poultry processing factories, poultry farms, backyard poultry pens and wild bird habitats, particularly those of the Orders Anseriformes (waterfowl such as ducks, geese and swans) and Charadriiformes (shorebirds, gulls and terns).

This paper discusses usage of ES and, where possible, its advantages and disadvantages compared to live bird samples (LBS), and also illustrates cases where ES have provided important information in understanding the biology and epidemiology of avian influenza, effectiveness of control and preventive measures, and providing early warning of possible outbreaks.

Finally, this work allows for identification of gaps in the current knowledge, thus providing the basis for further investigation.

2 | MATERIALS AND METHODS

2.1 | Literature search strategy

A systematic literature review protocol was developed in accordance with 'Preferred Reporting Items for Systematic literature reviews and Meta Analyses' (PRISMA) guidelines. We identified and reviewed publications involving the use of ES for AIV surveillance.

The database MEDLINE (PubMed) was searched for articles relevant to certain keywords. The search was conducted between the dates of 1 February 2019 and 10 June 2019 using the search strings: ("avian influenza" OR "zoonotic influenza") AND ("surveillance" OR "sampl*"). The search string was developed by identifying common Medical Subject Heading (MeSH) terms from previously identified relevant publications and combining them utilizing Boolean logic.

Citations were compiled, and duplicates were identified and removed using the citation software program EndNote X7 (Thomson Reuters).

2.2 | Selection criteria

The search strategy and inclusion criteria were aimed at identifying literature related to the use of ES in the epidemiological surveillance of avian influenza. Articles were limited to those in the English language. In cases where the abstract was available in English, this was reviewed. No restrictions were placed on the date of publication.

The titles, abstracts and keywords of all articles identified from the literature search were reviewed, and publications that did not involve AIV or did not state the use of ES methods were excluded. Otherwise, full texts were read, with focus on the methods section, to determine whether ES, or data from ES, were used.

Following the literature review, expert opinion was sought on historical information and surveillance activities that had not been detected or selected via the literature search. Relevant information from additional papers, reports and grey literature, as identified by subject matter experts, was extracted and included in the analysis.

2.3 | Data extraction and analysis

Data were extracted from the selected articles and compiled. A template was created using Microsoft Excel(R) v. 15.26 (Microsoft Incorporation), and a standardized set of data on the context, objectives, methods and findings of the study were recorded from each of the articles. Any comments made on the advantages, disadvantages, cost-effectiveness, sensitivity, positive-predictive value, representativeness, simplicity, acceptability or flexibility of ES were also recorded and compiled. Descriptive analyses of search outputs were performed using Stata(R) v. MP (StataCorp).

A list of the references that were assessed but not cited in this paper is provided in Appendix 1.

3 | RESULTS AND DISCUSSION

A total of 2,935 publications were identified through the literature search, whose titles and abstracts were screened for inclusion. Of these, 338 articles from peer-reviewed journals were subjected to

full-text review for inclusion (17 were excluded due to language constraints, that is non-English publications). Full text review of articles was required, as the sampling method used to obtain the AIV was often not explicitly described in the abstract. Of these 338 articles, 175 articles met the criteria for selection and had data extracted. An additional 54 articles that were not initially captured were included based on expert advice.

3.1 | History of ES for AIV surveillance

ES have been used as part of avian influenza surveillance systems since the 1970s when water and faecal samples from wild bird and domestic duck habitats were cultured to detect influenza viruses. These early 'One Health' studies were conducted to unravel the biology of influenza viruses and confirmed links between human and avian strains (Hinshaw, Webster, & Turner, 1979; Markwell & Shortridge, 1982; Sims & Peiris, 2013). Drinking water samples were used in surveillance programmes for avian influenza in turkey flocks in Minnesota, United States (US) during the 1980s as an alternative to live bird testing (Halvorson et al., 2003). Air samples and other ES were also collected from farms in the 1983–1984 highly pathogenic avian influenza (HPAI) outbreak in Pennsylvania, US to understand aspects of the epidemiology of the disease, including the duration of persistence of virus in various sites on affected farms, and the possibility of airborne spread (Brugh & Johnson, 2003).

Lake water samples were collected and tested in Eastern Europe for AIV, with negative results (Tůmová, 2003). Environmental sampling in markets was first used in the late 1980s in the United States and demonstrated the role that LBMs (as managed at that time) played in viral replication and persistence (Senne, Pearson, & Panigrahy, 2003). Market studies have continued in the United States including collection of ES. Results have been used to assess the effectiveness of preventive measures and to identify novel strains of virus (Bulaga, Garber, Senne, Myers, Good, Wainwright, & Suarez, 2003).

3.2 | Emergence of Gs/GD/96-lineage H5Nx HPAI viruses—a catalyst for increased surveillance and use of ES

In 1997, when HPAI viruses of the H5N1 subtype emerged as a cause of severe disease in both chickens and humans in Hong Kong Special Administrative Region (SAR), swabs of faeces were among the samples collected to detect AIV (Shortridge, 1999). This study, conducted in LBMs just prior to market depopulation, not only considered H5N1 subtype viruses that were present in >21% of chicken samples but also other subtypes. The study identified other AIV that may have been the donors of internal genes for the HK/97 H5N1 virus, including H9N2 viruses and H6N1 viruses (Guan, Shortridge, Krauss, & Webster, 1999; Hoffmann et al., 2000). The results provided additional justification for the decision (already made) to depopulate all LBMs (excluding the song bird market) and close them for seven weeks. When trade in live poultry resumed in Hong Kong SAR, LBM surveillance was introduced as a way of determining whether preventive measures implemented were working as expected. The main samples collected were swabs from fresh faecal samples on trays located under cages of birds. ES identified the first (and subsequent) incursions of Gs/GD/96-lineage viruses to the separate waterfowl market that was established in Hong Kong SAR in 1998 (Cauthen, Swayne, Schultz-Cherry, Perdue, & Suarez, 2000; Webster et al., 2002). Examination of viruses from these samples demonstrated that multiple new H5N1 genotypes had emerged and that domestic ducks were infected with these viruses; the original 1997 viruses from Hong Kong SAR were poorly adapted to ducks, based on experimental studies (Perkins & Swayne, 2002). Studies have continued since then in LBMs in Hong Kong SAR. These studies identified increases in prevalence of infection in markets in 2001 that preceded increased mortality in these markets (followed by depopulation) and the detection of antigenic variant H5N1 strain in 2008 that also resulted in partial depopulation.

ES have also been collected in LBMs in other parts of China, Cambodia, Bangladesh, Vietnam and Indonesia (Bui et al., 2019; Horm, Deboosere, Gutierrez, Vialette, & Buchy, 2011; Indriani et al., 2010; Rimi et al., 2019; Smith et al., 2006). Studies in these places have provided information on the range of virus subtypes present and in many cases the high levels of contamination within markets. Collection of ES in LBMs in China increased with the emergence of zoonotic AIV of the H7N9 subtype that caused more than 1,000 human cases of disease with most cases linked to contaminated LBMs. Following the introduction of influenza H5/H7 virus vaccination, surveillance results from LBMs using ES demonstrated the marked reduction of virus prevalence (Wu, Ke, et al., 2019; Wu, Lau, et al., 2019). Studies using ES also demonstrated that market rest periods reduced rates of virus detection but virus returned when birds were re-introduced to the markets (Yuan et al., 2015).

Results from tests on ES from LBMs collected in Bangladesh demonstrated the high prevalence of infection (Rimi et al., 2019) including Gs/GD/96 viruses. Viruses from these studies have been characterized and demonstrate that a wide array of subtypes is present in markets.

ES have been collected from a range of sites in Cambodia and have demonstrated the presence of virus in pond water and mud (Vong, Ly, Mardy, Holl, & Buchy, 2008) which appears to be a risk factor for human infection for those who swim in ponds potentially contaminated by faeces from domestic ducks (Ly et al., 2016). High rates of infection have been detected in a range of sample types in LBMs in Cambodia (Horm et al., 2016), and by conducting surveillance using the same techniques over several years, it was possible to demonstrate increases in prevalence in both ES (carcass wash water) and LBS (Horwood et al., 2018) in 2015 compared to 2013. Groundwater samples were collected in the United States during the 2015 H5N8 HPAI outbreak, and influenza virus RNA was detected (Hubbard, Kolpin, Fields, Hladik, & Iwanowicz, 2017).

Wild bird testing has been undertaken in a number of countries since the initial studies by Hinshaw et al in 1979 (Hinshaw et al., 1979). It increased once it became evident from 2003 onwards that Gs/GD/96-lineage H5Nx HPAI viruses could be carried over relatively long distances by migratory birds. Studies using ES, in particular fresh faeces, have been used in a number of countries including Australia, where the majority of samples tested are ES (Grillo et al., 2015). Surveillance in Mongolia has used samples from live birds and faeces. Sick and dead birds were preferred samples for detection of highly pathogenic H5 viruses (Gilbert et al., 2012).

At present, some countries/regions still focus on live bird testing whereas others have switched to ES, especially faecal samples.

Several other examples highlight the effectiveness of ES used either on its own (for low pathogenic AIV (LPAIV)) in wild bird surveillance (Baek et al., 2010; Barbara et al., 2017; Perez-Ramirez, Gerrikagoitia, Barral, & Hofle, 2010; Piaggio et al., 2012), or as a component (for HPAIV) (Bevins et al., 2014; Haynes et al., 2009; Khan et al., 2018) of large-scale risk-based surveillance systems in both high- (Deliberto et al., 2009; Grillo et al., 2015; Piaggio et al., 2012; Preskenis, Ladman, & Gelb, 2017) and low-resourced (Gaidet et al., 2007; Gerloff et al., 2014; Khan et al., 2018; Ofula et al., 2013; Tun Win et al., 2017) settings, where broad-scale surveillance proves logistically challenging (Grillo et al., 2015).

The information presented above demonstrates that ES have been used for over 50 years in avian influenza surveillance programmes and played an important role in assessing and guiding preventive and control programmes. ES have been used successfully for a number of purposes including the following:

- As a signal/trigger for action including early warning of presence of AIV of significance (covering threats to both animal and human health)
- Evaluation of effectiveness of avian influenza control and preventive measures
- Characterizing the ecology and epidemiology of AIV (including risk factors)
- Describing the geographical distribution of AIV (and relative prevalence of subtypes) in selected epidemiological units or locations at a given point in time
- Monitoring AIV trends (spatiotemporal distribution, genetic diversity, etc.) over time
- Characterizing circulating strains of AIV antigenically and genetically, including detection of novel AIV subtypes, antigenic variants, assessment of virulence and presence of molecular markers typical of mammalian adaptation
- Investigating avian influenza outbreak sources and potential routes of transmission.

Secondary purposes have included avian influenza diagnostic protocol optimization and validation. AIV isolated from ES have been used in pathogenicity studies in birds and mammals.

3.3 | Study types and settings

Two broad categories of study types were identified. The first collected ES as part of a, sometimes limited-term,research-oriented epidemiological study or activity. The second utilized ES data from ongoing surveillance systems, which typically involved governments or inter-governmental organizations, sometimes in partnership with university or research institutions. Multiple examples of such regional, national and subnational surveillance systems were identified, and several papers utilized subsets of data from the same surveillance system. For example, multiple studies utilized data from the United States Department of Agriculture (USDA)-Animal and Plant Health Inspection Service (APHIS)-Wildlife Services' National Wildlife Research Center (NWRC) which operated from 1 April 2006 to 31 March 2011,[2] and employed ES of faeces in wild bird habitats as a component of its surveillance for highly pathogenic AIV in the continental United States of America, American Samoa, the Marshall Islands, Guam, Canada and Mexico (Bevins et al., 2014; Piaggio et al., 2012).

3.4 | Study Protocols

While the determination of the ideal methods and protocols for ES are context- and region-dependent, and fall outside the scope of this review, major highlights regarding the methods identified in the literature are briefly summarized.

Non-standardized surveillance has been described as a chronic challenge for global AIV surveillance in wild birds (Machalaba et al., 2015). Factors that need to be considered include the relative effectiveness of active versus passive surveillance, which in turn depends on the virulence of the virus and the susceptibility of the host, determination of sample size, species to be sampled, sites type to be sampled, location to be sampled, sampling frequency and seasonality, among others. These differ widely, and the absence of standardized protocols implies that data are not collected, and codified comparably between different countries and regions. Nevertheless, progress has been made in some countries and regions in establishing standardized protocols used by different groups (European Food Safety et al., 2019; United States Department of Agriculture, 2015; Wildlife Health Australia, 2018). However, even in the European Union (EU), the extent of surveillance varies considerably from country to country (Knight-Jones, Hauser, Matthes, & Stark, 2010). The purpose of surveillance and nature of the AIV can also determine the types of samples collected. For example, dead wild bird sampling has been demonstrated as being more sensitive for detection of HPAIV than other methods of sample collection from birds, depending on the mortality rate in affected birds (Knight-Jones et al., 2010).

The selection of the type of sample to be collected varied according to the sampling site. Within wild bird habitats, the majority of ES collected were fresh faecal material; however, the collection of water and, less commonly, mud/soil/sediment samples has been

described (Densmore et al., 2017). Aquatic plants have been sampled in experimental studies in household ponds frequented by domestic ducks (Horm, Gutierrez, Sorn, & Buchy, 2012; Vong et al., 2008).

Within the LBMs or poultry farms, multiple types of ES were collected from cages (Biswas et al., 2018; Chen et al., 2014; Horm et al., 2016; Indriani et al., 2010; Lopez et al., 2018; Yuan et al., 2015), floors (Biswas et al., 2018; Bulaga et al., 2003; Chen et al., 2014; Indriani et al., 2010), poultry drinking water sources (Chen et al., 2014; Horm et al., 2016; Indriani et al., 2010; Munoz-Aguayo et al., 2019; Yuan et al., 2015), feeding sources (Indriani et al., 2010; Lopez et al., 2018; Munoz-Aguayo et al., 2019), processing (Chen et al., 2014; Indriani et al., 2010; Lopez et al., 2018; Yuan et al., 2015), slaughtering and display surfaces (Biswas et al., 2018; Bulaga et al., 2003; Chen et al., 2014; Indriani et al., 2010; Yuan et al., 2015), carcass wash water (Biswas et al., 2018; Horm et al., 2016; Yuan et al., 2015), equipment, waste bins, water run-off and cleaning materials (Chen et al., 2014; Indriani et al., 2010). Several studies also described the collection of air samples (Bui et al., 2019; Kang, Chen, Bi, Chen, & Tan, 2016; Scoizec et al., 2018; Wei et al., 2018; Wu, Ke, et al., 2019; Wu, Lau, et al., 2019; Wu et al., 2018; Zeng et al., 2017; Zhou et al., 2016).

The sampling methods for poultry processing factories, poultry farms and backyard poultry pens were less frequently described in detail; however, multiple types of ES were collected including feathers, surface swabs of cages, barn floors and walls, feed troughs, egg belts and processing locations, areas with faecal contamination, pond water, mud and air samples (Horm et al., 2016; Munoz-Aguayo et al., 2019; Vong et al., 2008). As a general rule, sampling of birds, especially sick or dead birds, is the sample of choice for HPAIV on farms. Other samples are usually only collected to answer specific questions relevant to disease transmission pathways (e.g. air samples to ascertain the likelihood of airborne transmission between farms) and the epidemiology of the outbreak.

Sample testing methods and diagnostic approaches have changed over time reflecting developments in methods available for virus detection. Early studies up to the early 2000s using ES relied on culture in embryonated chicken eggs given there were few viable alternatives (e.g. Guan et al., 1999; Hinshaw et al., 1979). Virus culture is still used but is usually preceded by quantitative reverse transcription polymerase chain reaction (RT-PCR), followed by virus isolation and partial or whole genome sequencing where required. Studies using targeted resequencing and genomics have now been reported but additional work is needed to confirm the validity of this approach (Himsworth et al., 2019).

For some ES, the concentration of virus is low because of dilution by the sample matrix (e.g. lake water at wild bird congregation points). As a result, concentration of the virus is required prior to testing. Various methods have been used including filtration/elution, centrifugation (Munoz-Aguayo et al., 2019), polyethylene glycol concentration (Deboosere et al., 2011, 2012; Ronnqvist, Ziegler, von Bonsdorff, & Maunula, 2012) and erythrocyte absorption using chicken red blood cells (Horm, Gutierrez, Sorn, et al., 2012; Khalenkov, Laver, & Webster, 2008). In some studies in markets, no concentration of drinking water samples was applied but only a small volume of water (0.5 ml) was collected (Leung et al., 2007).

3.5 | Advantages of environmental sampling

Multiple advantages of ES have been put forward. ES was considered a cost-effective (Deliberto et al., 2009; Perez-Ramirez et al., 2010; Stallknecht et al., 2012), simple, rapid, flexible, convenient and acceptable way of achieving surveillance objectives (Indriani et al., 2010; Leung et al., 2007; McLean et al., 2007; Munoz-Aguayo et al., 2019; Onuma et al., 2017; Pannwitz, Wolf, & Harder, 2009). As a non-invasive technique, it was posited by authors to improve animal welfare (Bui et al., 2019; Lee et al., 2010) and to reduce impact on wild bird habitats and communities (Barbara et al., 2017; Pannwitz et al., 2009; Perez-Ramirez et al., 2010). It can also be representative of wider geographical areas compared to location-specific sampling sites for live birds.

Comparison of the sensitivity of ES and LBS (and dead bird sampling) for the surveillance of AIV was challenging for several reasons. Firstly, literature interpretation was limited, for example AIV detection rates could not always be used as a proxy for sensitivity because between studies comparability of true prevalence was poor, and settings, study designs and methodologies of studies represented different epidemiological units, with an ES representing single (fresh faecal sample) to multiple birds (faecal matter, surface swab or drinking water sample) while an LBS (oropharyngeal or cloacal) always represent a single bird.

Secondly, the sensitivity varied significantly with multiple factors, and appropriately stratified comparisons were identified as a major gap in the literature. These included ES type (e.g. air sample, fresh faecal swab, surface swab and poultry drinking water), geographical setting and its respective environmental factors (climate), detection method utilized (virus isolation versus molecular techniques), the pathogenicity of the virus (LPAI versus HPAI), and the virus–host interplay determining viral tissue predilection (i.e. gastrointestinal versus. respiratory tracts) and shedding (e.g. faecal, airborne). In this context, it is important to note that prevalence also typically varies with avian influenza subtype (Wang et al., 2017). ES was found to be comparable to that of LBS in several contexts (Grillo et al., 2015; Pannwitz et al., 2009; Vergne et al., 2019), while less sensitive in others (Bulaga et al., 2003; Mellor et al., 2018; Sonnberg et al., 2012; Tracey, 2010). Furthermore, the use of environmental faecal sampling in the surveillance of LPAIV is well supported by the literature (Baek et al., 2010; Barbara et al., 2017; Gaidet et al., 2007; Lebarbenchon et al., 2010; Perez-Ramirez et al., 2010). In particular, when fresh faecal samples were collected immediately, they were found to have similar (Perez-Ramirez et al., 2010) or higher detection rates than oropharyngeal swabs (Busquets et al., 2010) and cloacal swabs (Chen et al., 2014; Latorre-Margalef, Avril, Tolf, Olsen, & Waldenstrom, 2016). However, where faecal samples are not immediately obtained, they can potentially be rapidly diluted in the environment and are subjected to the various externalities of

physical, chemical and biological factors (e.g. salinity, dryness, ultraviolet radiation, pH, temperature, humidity and microbial flora) (Henaux, Samuel, Dusek, Fleskes, & Ip, 2012; Irwin et al., 2011; Keeler, Berghaus, & Stallknecht, 2012; Nielsen, Jensen, Stockmarr, & Jorgensen, 2013; Yamamoto, Nakamura, Yamada, & Mase, 2010); hence, the poorer viral detection rates found in some studies on ES compared with live bird cloacal samples (Sonnberg et al., 2012; Tracey, 2010). While there are examples of HPAI virus strains being sporadically isolated from faecal samples (Gerloff et al., 2014; Hiono et al., 2015; Jeong et al., 2014; Negovetich et al., 2011; Ozawa et al., 2019; Poen et al., 2016; Willeberg et al., 2010) during wild bird surveillance, these are often infrequent incidents, and environmental faecal sampling in isolation may have poor sensitivity as a method of HPAI virological surveillance (Latorre-Margalef et al., 2016; Poen et al., 2016). For certain AI viruses (e.g. H9N2 in LBMs), higher rates of detection have been found in drinking water samples compared to LBS cloacal and/or oropharyngeal (Kale, Mishra, & Pawar, 2013; Munoz-Aguayo et al., 2019) or faecal samples (Leung et al., 2007). However, observations in markets demonstrate that water supply to birds varies considerably from market to market, potentially impacting virus prevalence in water samples. In some situations, water is either not available to collect or it is not an appropriate sample. In some markets, birds have access to drinking water provided in troughs or bowls whereas some market stall operators supply birds directly with water from hoses connected to taps. Some do not supply water if the duration of stay is short. In other markets, birds are tied by the legs and cannot access water bowls or troughs even if these are provided (L. D. Sims unpublished).

Various approaches to improving the sensitivity of surveillance systems were discussed. Sensitivity of surveillance can be improved by optimizing laboratory algorithms, for example through the addition of molecular-based detection methods (Onuma et al., 2017), and utilizing appropriate sampling strategies (Vergne et al., 2019) which rely on risk-based surveillance approaches through consideration of environmental, climatic or socioeconomic factors (e.g. poultry density, wild bird demographics, human incidence, seasonal oscillation and cultural festivities) in order to target geographical areas and/or sampling sites at higher risk.

It is also possible to perform risk-based sampling through the selection of sites, such as slaughter zones (Chen et al., 2014; Indriani et al., 2010), and surfaces, such as sewage (Kang et al., 2015; Yuan et al., 2015; Zhang et al., 2015) and chopping boards (Kang et al., 2015; Wang et al., 2015; Yuan et al., 2015; Zhang et al., 2015). Previous studies have found prevalence to be especially high where heavy contamination occurs and on visibly dirty, moist, or difficult-to-clean surfaces (Indriani et al., 2010; Trock, Gaeta, Gonzalez, Pederson, & Senne, 2008), for example along the wall-floor junctions, cracks and holes. However, one of the disadvantages of using ES constitutes the possibility of detecting viruses which are not currently present in the birds at the time due to accumulation and persistence within the environment (Lang, Kelly, & Runstadler, 2008; Zhang et al., 2014).

Furthermore, sensitivity can be improved by utilizing appropriate sample collection and handling methods such as elution and concentration steps, for example filtration and erythrocyte agglutination (Khalenkov et al., 2008; Munoz-Aguayo et al., 2019; Ronnqvist et al., 2012).

3.6 | Limitations and weaknesses of environmental sampling

Various limitations of ES methods in wild bird surveillance were also identified in the literature. ES may potentially result in the loss of species-specific information on the virus shedding source, limiting its utility in taxonomic-level analyses (Barbara et al., 2017; Grillo et al., 2015; Pannwitz et al., 2009). However, this potential disadvantage has largely been overcome, with studies utilizing fresh faecal samples mitigating this through the use of mitochondrial DNA barcoding for host bird species identification (Ge, Chai, et al., 2017; Kang et al., 2010, 2011; Lee et al., 2010; Onuma et al., 2017), collecting samples from single species flocks (Ghersi et al., 2009; Kou et al., 2009; Pawar et al., 2012), through the observation and species identification of the individual host immediately prior to defecation and sample collection (Ge, Chai, et al., 2017; Haynes et al., 2009; Kang et al., 2010; Pannwitz et al., 2009; Ye et al., 2016) and/or by observation of faecal morphology (Ge, Chai, et al., 2017; Ge, Yao, et al., 2017; Hansbro et al., 2010; Pannwitz et al., 2009; Ye et al., 2016).

However, ES can entail the loss of individual information (with each environmental sample, other than faeces, likely representing multiple birds, and also allowing for the possibility that multiple positive environmental samples are from the same individual) as opposed to an oropharyngeal or cloacal swab which is definitive and represents a single bird (Barbara et al., 2017; Lee et al., 2010; Leung et al., 2007). In addition, because an association between the sample and individual birds cannot be established, ES might not allow for the simultaneous collection of relevant biological metadata, valuable for advanced epidemiological or risk factor analysis (e.g. host age, host condition, or other data retrieved from band recoveries), especially when dealing with wild birds. It also provides no information on the time of deposition of the virus, especially if using samples such as mud or sewage.

Techniques for the demonstration of viable virus through embryonated egg inoculation in certain ES (e.g. fresh faecal samples) are well-established, and many studies have successfully isolated virus from a range of ES (Barbara et al., 2017; Bui et al., 2019; Dong et al., 2017; Fujimoto et al., 2010; Ge, Yao, et al., 2017; Ghersi et al., 2009; Guan et al., 1999; Hiono et al., 2015; Jahangir et al., 2009; Kang et al., 2011; Lee et al., 2010; Li et al., 2019; Munoz-Aguayo et al., 2019; Negovetich et al., 2011; Onuma et al., 2017; Qi et al., 2018; Trock et al., 2008; Yuan et al., 2015). However, isolation rates have been variable and in some cases very low (Haynes et al., 2009; Horm, Gutierrez, Nicholls, & Buchy, 2012; Onuma et al., 2017), and sometimes unsuccessful (Kelvin et al., 2012; Vong et al., 2008). This may be due to the low true

prevalence of viable virus in the areas sampled, as supported by two studies conducted in migratory wild bird habitats, which found no difference in the virus isolation rate between environmental (faecal) and live bird (cloacal and oropharyngeal) samples over a five year surveillance period (Grillo et al., 2015). Alternatively, it may speak to the poorer suitability of viral culture techniques for ES (Kelvin et al., 2012; Vergne et al., 2019), as supported by other studies which found higher virus isolation rates among LBS (cloacal or oropharyngeal), compared to ES (Latorre-Margalef et al., 2016; Stallknecht et al., 2012; Tracey, 2010). Poor suitability of viral culture techniques may be a limiting factor especially in the case of aerosol sampling: while some studies had successes with virus isolation and phylogenetic analysis (Wu et al., 2017, 2018; Zeng et al., 2017; Zhou et al., 2016), and good correlation with subtypes detected in the environment (Zhou et al., 2016), others did not, even from RT-PCR-positive samples, possibly due to low survival and concentration of infectious AIV particles (Kang et al., 2016). One pilot study conducted at a LBM in Viet Nam compared air samples with paired oropharyngeal swabs (collected contemporaneously from the same location) and found strong, but not perfect, agreement between RT-PCR-positive air and swab samples. Virus recovery (1/30) was poorer from air samples compared to pooled oropharyngeal samples (25/116) (Bui et al., 2019). Another aerosol sampling study conducted in a LBM in China found viral recovery rates of 1/275 RT-PCR-positive samples (Zeng et al., 2017). Other studies have demonstrated reasonable correlation between air samples and other results especially when virus is present at high concentrations (Cheng et al., 2020).

However, even with RT-PCR, a study which compared AIV detection results from bioaerosol sampling across a number of contexts

TABLE 1 Summarized applicability of environmental sampling as compared to live bird sampling in the surveillance of avian influenza viruses

Environmental sampling	Live bird sampling (cloacal or oropharyngeal swab)
Flexible; easy to adapt sample size, timing, frequency, location, and can be applied across value and supply chains and in a number of contexts (Jennelle et al., 2016; Pannwitz et al., 2009)	Less flexible; contingent on presence and number of birds (live, hunter-killed birds, post-mortality events or otherwise) when wild bird sampling
Acceptable to traders and stall vendors in live bird markets (Indriani et al., 2010; Vergne et al., 2019; Zeynalova, Guliyev, Vatani, & Abbasov, 2015)	Potential for reduced willingness of persons in live bird markets (traders, stall vendors, etc.) to participate due to perceived disruption of business operations, trading and selling activities (Bui et al., 2019; Indriani et al., 2010)
Cost-effective; simple and rapid procedure, minimal training, equipment required, and easily scalable to increase sample size at minimal cost (Deliberto et al., 2009; Deliberto et al., 2009; Gaidet et al., 2007; Grillo et al., 2015; Grillo et al., 2015; Lebarbenchon et al., 2010; McLean et al., 2007; Onuma et al., 2017; Pannwitz et al., 2009; Pannwitz et al., 2009; Stallknecht et al., 2012; Tracey, 2010)	Additional financial, technical, and logistical implications associated with bird trapping or capture and invasive and labour- and time-intensive sample (oropharyngeal, cloacal, blood) collection (Pawar et al., 2012; Tracey, 2010)
Bird welfare; does not require trapping, capture or handling of birds (Bui et al., 2019; Zeynalova et al., 2015)	Requires stressful trapping or capture and handling of birds, invasive sampling procedures, disruption of wild bird communities (Pannwitz et al., 2009)
Safer; reduced potential for virus aerosolization) (Indriani et al., 2010; Zeynalova et al., 2015)	Risk of virus aerosolization and infection for sample collectors and bystanders (Indriani et al., 2010)
Loss of individual-level data (for example, host species, host age, host condition)	Allows for the collection of individual-level data, and corresponding epidemiological analyses
Markets: Smaller number of samples required to detect virus if sites recognized to be highly contaminated are sampled	Large number of samples required to detect virus if present at low prevalence.
Positive result may reflect infection in birds at an earlier time depending on type of sample collected	Represents situation in birds at the time of sample collection

along the poultry supply chain (e.g. farms, LBMs) found lower detection (9/338 or 2.7% (95% CI:0.9–4.3)) compared to other environmental samples (e.g. surfaces) (311/991 or 31.4% (95% CI:28.5–34.4)) or LBS (cloacal or oropharyngeal swabs) (104/442 or 23.5% (95% CI:19.6–27.5)) (Wu, Ke, et al., 2019; Wu, Lau, et al., 2019).

Thus, due to the low viral concentration in ES compared to LBS, inactivation of viral particles by environmental factors, and presence of bacterial contamination of samples (Khan et al., 2018), RT-PCR techniques may be more suited for ES compared with viral culture techniques; this has been demonstrated by studies which show virus isolation rates ranging from 0% (0/27) (Vong et al., 2008), 4.6% (13/280) (Indriani et al., 2010), 11% (10/90) (Horm, Sorn, Allal, & Buchy, 2013), 14% (47/327) (Terregino et al., 2007), 28% (214/759) (Piaggio et al., 2012), 43.4% (153/352) (Onuma et al., 2017) up to 66.67% (4/6) (Barbara et al., 2017) among RT-PCR-positive environmental samples. It is noteworthy that when ES with high virus content (CT values < 30) were cultured the rate of isolation was high (Horm et al., 2016).

Use of PCR alone does not differentiate between non-viable viral nucleic acid and infectious virus (Lang et al., 2008) and does not necessarily translate into quantitative risk of pathogen exposure or transmission (Wu, Ke, et al., 2019; Wu, Lau, et al., 2019). Nevertheless, markets with high rates of contamination with H7N9 virus were frequently associated with human infections. Furthermore, due to the segmented genome of AIV, it may be unable to provide information on the specific combination of gene segments and only provides information on a single gene sequence at a time (Lang et al., 2008), making reconstruction of parental gene constellations a challenge in mixed samples (Latorre-Margalef et al., 2016). Therefore, subtype or strain characterization in the event of several co-circulating subtypes or strains can be challenging.

Due to these limitations, ES are less suitable for certain surveillance objectives, including those requiring individual or host population information, the characterization of co-circulating subtype combination, trace-back of infected flocks, or estimation of prevalence, incidence or any absolute measures of AIV infection within a specific population.

An overall summary of the applicability of ES as compared to LBS in the surveillance of AIV is presented in Table 1.

3.7 | Gaps in Knowledge

Various gaps in current knowledge were identified. Only a small number of studies were designed for comparison with appropriately stratified variables. The literature on the sensitivity of ES techniques compared to LBS was limited, with estimates varying according to the actual prevalence (Indriani et al., 2010), subtype (Vergne et al., 2019), species, age, and density of birds sampled (Latorre-Margalef et al., 2016; Lickfett, Clark, Gehring, & Alm, 2018), factors affecting viral persistence such as temperature, pH and salinity (Lickfett et al., 2018), sample type (Latorre-Margalef et al., 2016; Vergne et al., 2019), collection (Indriani et al., 2010; Spackman, Pedersen, McKinley, & Gelb, 2013; Trock et al., 2008), sample handling including swab construction material, transport media, media volume, and pooling (Spackman et al., 2013), testing methods (Horm et al., 2013) and sampling strategy (Latorre-Margalef et al., 2016; Vergne et al., 2019).

Given the costs of maintaining surveillance programs, it is essential to establish best practices for field methodologies in order to provide a cost-effective approach yielding robust data for epidemiological interpretation. Currently, there is a lack of practical information within the literature for developing and implementing the optimal avian influenza surveillance programs, and detailed descriptions are lacking for various aspects of the protocols and methods, such as the sampling strategy, describing the methods for selecting an appropriate sample size, sample type, epidemiological unit selection, sampling frequency and pooling strategies to reach the desired sensitivity.

3.8 | Limitations of the study

While efforts have been made to conduct a comprehensive literature review on the use of ES for AIV, undertaking this work noted several limitations or biases. Firstly, studies included for review were limited to those indexed by the database used, and did not capture literature published in languages other than English, or unpublished data. A large body of field experience related to the logistics and challenges of implementing ES in the field which was not documented or made publically available, exists in grey literature or internal reports of government departments. This constraint was overcome in part by including papers identified by one author through his experiences with avian influenza over the past 25 years. In some cases, protocols used in national and regional surveillance programs published on government websites were included (EFSA et al., 2019; USDA, 2015; Grillo et al., 2015).

Secondly, few studies described the precise method of collection of ES. A number of papers did not specify whether the viruses isolated and then utilized for analyses were recovered from ES, or the simultaneously collected LBS (Barman et al., 2019; Kou et al., 2009; Yang et al., 2015). This limited our ability to determine to what extent the collection of environmental samples served in meeting the surveillance objectives.

Finally, the lack of comparability between studies and the lack of appropriately stratified results within studies limited the systematic evaluations of ES as an alternative to LBS. Nevertheless, there are still many situations where ES has provided valuable information.

4 | CONCLUSIONS

Surveillance for avian influenza viruses (AIV) is an important element of animal and public health programmes. Samples for surveillance have been taken directly from live birds (cloacal or oropharyngeal swabs and feathers for detection of virus or blood for serology), from dead birds (diseased or hunter-killed) or from the environment

in which the birds are located. Environmental samples (ES) have been collected at sites where wild birds congregate, in live poultry markets and, in some cases, poultry farms. ES have mainly been taken from faeces, water and surfaces contacted by birds. In wild birds, faecal samples have been used to understand the biology and distribution of influenza A viruses since the 1970s and some studies have shown ES to offer equivalent sensitivity to samples collected directly from birds without the cost and welfare issues associated with wild bird capture. The potential disadvantage of not being able to collect information on the bird of origin for samples has been largely overcome by observing birds during defaecation, through morphology of faeces or through use of DNA barcodes that allow host identification. Water samples have also been used to identify AIV at wild bird congregation points but require filtration or other methods to concentrate virus. ES have played an important role in detecting AIV in live poultry markets. Results of tests from ES have been used to assess the effectiveness of interventions in markets and for determining risk to public health (e.g. levels of contamination with or presence of H7N9 viruses). A range of ES from markets has been shown to be of benefit for detecting and characterizing AIV, including drinking water (with or without concentration) and other sites along the marketing and processing chain, including chopping boards and drains. Air samples have been deployed on farms and in markets to detect the presence of AIV in air and the size of particles associated with virus. Available results suggest they are less sensitive than samples from birds for detection of live virus. Global standardized sampling procedures for ES have not been developed and depend on the purpose of surveillance (e.g. determining whether virus is present or prevalence of infection). Many studies in which ES and samples from live birds were collected at the same time did not provide information on the source of positive samples. This prevents assessment of the overall sensitivity of methods used for virus detection in these studies. It is expected that surveillance systems for avian influenza in the future will continue to deploy a mix of sample types from live birds, dead birds and the environment (Rimi et al., 2019). Better information on the manner in which ES and other samples are collected and detailed results by sample type will allow better assessments of the relative merits and appropriate mix and number of ES.

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CONFLICT OF INTEREST

The authors whose names are listed immediately below certify that they have NO affiliations with or involvement in any organization or entity with any financial interest (such as honoraria; educational grants; participation in speakers' bureaus; membership, employment, consultancies, stock ownership or other equity interest; and expert testimony or patent-licensing arrangements), or non-financial interest (such as personal or professional relationships, affiliations, knowledge or beliefs) in the subject matter or materials discussed in this manuscript. Grace Hood, Xavier Roche, Aurélie Brioudes, Sophie von Dobschuetz, Folorunso Oludayo Fasina, Wantanee Kalpravidh, Yilma Makonnen, Juan Lubroth, Leslie Sims.

ETHICS APPROVAL

No ethical approval was required as this is a review article with no original research data.

DATA AVAILABILITY STATEMENT

Data sharing is not applicable to this article as no new data were created or analysed in this study.

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ENDNOTES

[1] In this document, we are using a broader definition of avian influenza than that used by OIE in the Terrestrial Animal Code (OIE 2018) to include influenza viruses of all subtypes detected in birds.

[2] This programme has continued in a modified form since 2011 See for example the HPAI plan prepared in 2015 https://www.aphis.usda.gov/animal_health/downloads/animal_diseases/ai/2015-hpai-surveillance-plan.pdf

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APPENDIX 1
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