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Small research and development activity (SRA)

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1 Acknowledgments

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2 Executive summary

Point-of-care tests (POCTs) allow for rapid diagnosis of infectious diseases in non-laboratory settings, and have the potential to significantly disrupt traditional animal health surveillance paradigms especially in Low- and Middle-Income Countries (LMICs), where maintenance of cold chain and transport of field-collected samples to the nearest capable laboratory is particularly challenging. Accurate and rapid field diagnosis of animal disease has many benefits including limiting further transmission of infectious pathogens that impact trade, protecting the health and well-being of animal populations that smallholders rely on for their livelihoods, and potentially containing epidemics. However, despite the abundance of POCTs on the commercial market and their clear benefits in remote and rural settings, the potential benefits of these tests in LMICs remain largely unrealised.

Many different POCT platforms exist, ranging from paper-based lateral flow assays (LFAs), simple chemical reaction-based dipsticks to portable isothermal nucleic acid detection systems (e.g. Loop Mediated Isothermal assays (LAMP), recombinase polymerase assays, portable PCR devices), handheld nanopore sequencing devices, wearable electronics sensors and more. In recent years there has been an exponential increase in the range and quantity of POCTs being developed and commercialised for human medical and veterinary diagnostics (1).

A literature review was undertaken to identify POCTs currently available for diagnosing infectious animal diseases and to identify facilitators and barriers to their use and uptake in LMICs (1). Questionnaires and structured interviews were conducted with POCT "manufacturers" (n=12) and with animal health staff in the Asia-Pacific region ("users") (n=18) to investigate their current knowledge, attitudes and practices on POCTs. Most respondents reported POCT usage for only a few of the ten OIE priority diseases that were selected for inclusion in the questionnaires (African Swine Fever, (ASF), Avian Influenza Virus (AIV), brucellosis, Classical Swine Fever (CSF), Foot and Mouth Disease (FMD), Hendra/Nipah, Infectious Bursal Disease (IBD), Newcastle Disease (ND), Peste des petits Ruminants (PPR), rabies). Of the manufacturers, the most commonly reported disease targets were ASF and AIV (both 60%), followed by FMD (50%), brucellosis (40%) and CSF (30%). Other POCTs are reportedly targeting bluetongue virus, Hendra/Nipah virus, IBD, ND and rabies (each reported by 20%) and many more including anthrax, *Mycoplasma bovis*, PPR, Porcine Reproductive and Respiratory Syndrome (PRRS), Porcine Epidemic Diarrhoea (PED) and Equine Influenza (EI). Most manufacturers reported that their POCTs were intended to be used by a variety of operators, predominantly private veterinarians (92%), government veterinarians and veterinary laboratories (67%), community animal health workers and/or animal technicians (56%), and farmers and/or animal owners (50%). Users rated rapidity and suitability for outdoors the most important factors, whereas manufacturers thought that ease of use/minimal training and minimal equipment are of greatest importance. A discord was observed between the user and manufacturer responses regarding factors leading to reduced POCT usage. Over 50% of users indicated that a lack of trust in the accuracy and difficulty in obtaining POCTs were the major factors. 75% of manufacturers thought a lack of policies and guidelines as the major factor for non-use, followed by distrust in POCT accuracy (67%) and that POCTs are too expensive (42%, compared with 11% of users).

Inadequate regulatory guidance and poor industry oversight has led to a proliferation of POCTs of varying quality and fitness for purpose released onto the market, presenting challenges to potential end users who are, by design, expected to have limited diagnostic experience. Accurate, independent test validation data for commercial POCTs are often lacking. There are also inconsistencies in the way POCTs are validated and lack of consistency in the information manufacturers provide, or at times do not provide, in the test kit technical data. The international community, led by the OIE, should be urged to adopt regulatory frameworks for the **standardised** test validation data published by POCT manufacturers.

There is an OIE registry for ‘fit for purpose’ diagnostic tests, including for POCTs, but this registry is heavily underutilised for various reasons. There are lengthy timelines for OIE to consider a manufacturers submission, to appoint experts to review the technical file and to approve its listing on the register. There is also a cost hurdle for the OIE submission and review. The underutilisation of the OIE test registry and the absence of other central peer reviewed registries makes it difficult for users to select ‘good’ POCTs.

Many users do not take into account these known, or unknown, limitations when using specific test kits, which can lead to inaccurate disease diagnoses or surveillance results if samples are not backed up with confirmatory tests in a laboratory. Government veterinary services especially in LMICs should be aware of the costs, impacts, cost-effectiveness and operational feasibility of incorporating POCTs into diagnostic workflows in their specific country context, and invest their resources accordingly.

3 Introduction

Despite considerable improvement over recent years in laboratory capacity in south and southeast Asia, disease surveillance in resource-limited settings has many challenges. Long distances, unreliable sample transport (e.g. poorly maintained roads and vehicles, fuel shortages, inadequate courier networks, seasonally inaccessible roads), difficulties maintaining cold chain, under-equipped laboratories, shortages of trained personnel and prohibitively expensive equipment operating costs contribute to long delays between sampling, diagnosis and follow-up, with sample turnaround times often taking days to weeks (2). Whilst laboratory obtained test results are generally collated for reporting to national veterinary authorities, the results of each individual farm or village investigation are often not reported back to the farmer or villagers leading to a lack of trust in the system. This diagnostic gap and subsequent disease under-reporting/misdiagnosis can allow animal pathogens to spread, with potentially serious and far-reaching outcomes.

Point-of-care tests (POCTs), which allow for rapid diagnosis of infectious diseases in non-laboratory settings, have the potential to significantly disrupt traditional animal health surveillance paradigms in LMICs, as they overcome many of the aforementioned challenges. The ability of POCTs to provide an immediate answer to the farmer empowers the service provider and builds trust in the system. Knowing the nature of the disease challenges whilst on-site provides confidence for the service provider to discuss appropriate control or treatment options with the farmer.

POCTs, also known as ‘rapid diagnostic tests’, ‘point of need tests’ and ‘near patient tests’, come in a range of different formats, and are currently being used by human, animal and plant industries for a range of applications worldwide. They are designed to be portable, user-friendly, simple to use, and usually have a turnaround time from sample to result in under an hour, allowing diagnosis and management decisions to be initiated within the same encounter. Many different POCT platforms and formats exist, from simple lateral flow assays (LFAs) to more complex portable polymerase chain reaction (PCR) devices and wearable biosensors, among others. Many POCTs utilise smartphones, Wi-Fi and/or Bluetooth connections to allow data transmission between remote field sites and central database (3, 4). POCTs typically require very small sample volumes, allowing minimally or non-invasive sampling methods such as capillary blood sampling or swabbing techniques that may increase testing compliance in some cultural settings where invasive sampling methods such as venepuncture are poorly tolerated.

Recent years have seen POCT development and commercialisation increase exponentially – a PubMed search for ‘point of care test’ returned over 16,900 results at the time of writing – and the trend is predicted to continue as methodologies are refined and new technologies emerge. But despite the excellent potential and logistical advantages being offered by POCTs, many are being released onto the market without any verifiable validation data being available about their diagnostic test performance. Many laboratories, health clinics and government services in LMICs are therefore purchasing these POCTs based on price alone (1). POCTs are likely to be operated within a higher range of variation than traditional laboratory tests, due to the varied competence and experience of operators, and more variable sample quality than might be submitted to diagnostic laboratories. Similarly, storage conditions and shelf life of kit reagents, and handling and maintenance of portable platforms may be subject to a higher variability than in an accredited laboratory. It is therefore important that the validation of POCTs is assessed under a range of realistic conditions. To assess and monitor reliability it is preferable that POCTs include internal quality controls to confirm the basic functioning of the device, for example a weak-positive sample (to confirm diagnostic sensitivity and avoid false-negative results), a negative control (to confirm that test reagents are not contaminated producing false-positive results), and for POCTs that detect nucleic acids, an “internal” test control to assess presence of matrix inhibitors, for example as described in the LAMP FMD validation (5). Given these additional issues that should be considered and

assessed by developers and users of POCTs it is perhaps not surprising that some recent publications have assessed different commercially-available POCTs and described wide differences in their diagnostic performance (6, 7, 8, 9, 10). Without transparent indications of a test's fitness for purpose, veterinary services in LMICs will struggle to correctly interpret the epidemiological implications of POCTs in different settings.

Despite the abundance of POCTs on the commercial market and clear role for POCTs in remote and rural settings, their true potential in LMICs is still far from being realised. This SRA sought to investigate the reasons for this underutilisation of POCTs for diagnosis of infectious and zoonotic animal diseases in LMICs through a comprehensive literature review and assessment of currently-available competing POCT platforms. Through engaging with key stakeholders and manufacturers to identify major facilitators and barriers to POCT use and uptake in LMICs, the findings from this SRA will ultimately provide technical guidance for POCT users in resource-limited settings.

4 Objective 1: Provide technical leadership and demonstrate and encourage strong test validation processes for the diagnostic POCTs that are being developed or currently being sold

4.1 Assist with validation of pan-FMD LAMP assay as example of 'best practice'

Agriculture Victoria, with assistance from this project, further developed, validated and published a reverse-transcription loop-mediated isothermal amplification foot-and-mouth disease virus (RT-LAMP-FMDV) test for use in the field (5), using a previously published LAMP primer set (11). An internal positive control was designed by ACDP and provided to Agriculture Victoria for use with the assay to mitigate any intrinsic interference from the unextracted field samples and avoid false negatives. Further modifications were included to improve the speed and operability of the test, for use by non-laboratory trained staff operating under field conditions, with shelf-stable reaction kits that require a minimum of liquid handling skills.

The assay's performance was compared against an established laboratory-based real-time reverse transcriptase PCR (rRT-PCR) test targeting the 3D region of FMD virus (Tetracore Inc). A strategic validation approach was performed to assess the test's diagnostic sensitivity when used in the laboratory on extracted RNA samples of various FMD serotypes from Thailand, and when used in the field on unextracted samples collected from FMD-suspected animals (oral and foot lesion swabs) from Bhutan. Assessment of the test's diagnostic specificity was performed using abattoir samples derived from FMD free animals from Australia. LAMP FMD diagnostic kits containing lyopholised reagents were subsequently developed and have been released for sale by Geneworks Australia, which alleviate the need for cold chain storage and offer longer shelf life making the test more practical for both Australian and LMIC settings.

The statistical approach to validation was performed by both the Frequentist and the Bayesian Latent Class Methods, which both confirmed this new RT-LAMP-FMDV test as 'fit for purpose' as a herd diagnostic tool with diagnostic specificity >99% and sensitivity 79% (95% Bayesian credible interval: 65, 90%) on unextracted field samples (oral and foot lesion swabs). The results indicate that LAMP has the potential to complement current laboratory diagnostics, such as rRT-PCR, as a preliminary tool in the investigation of FMD.

4.2 Identify and assist with one or two POCTs undergoing validation in the region

Discussions and planning had commenced to assist laboratories in Cambodia and Vietnam in the validation of field tests for ASF diagnosis. These validations were to involve comparison of the Ingenasa lateral flow test, a LAMP based test using the Genie III platform, and laboratory-based results obtained from qPCR results using the King assay. However, just prior to the commencement of this work all international travel was stopped and ACDP was placed into a COVID-19 emergency response mode of operation with rosters to limit staff numbers on-site and with all non-core diagnostic and research work being deferred.

5 Objective 2: Identify market gaps/failures for animal disease POCTs and propose solutions to encourage the introduction of appropriately validated tests emerging from product development pipelines

5.1 Literature Review: Validation of animal disease POCTs and application in LMICs

An extensive literature review was conducted to investigate whether POCTs are currently being used in LMICs for diagnosis of infectious animal diseases, to determine characteristics of 'ideal' POCTs that would facilitate their use, and to identify any barriers to uptake in these settings.

This literature review used a hermeneutic approach that emphasised continuous engagement with and the gradual development of a large body of literature, to develop understanding and insights related to this broad and complex topic (12). The interpretation and critique that this narrative form of review would bring to this topic was preferred over a systematic approach more suited for addressing narrowly focussed research questions (13,14). We used an iterative search strategy of electronic databases, including PubMed, Web of Science and Google Scholar, using different combinations of the following words and phrases to identify relevant publications: POC [point-of-care], 'point of care tests', POCT, 'field test', 'rapid test', 'rapid diagnostic test', zoonoses, infectious, veterinary, livestock, animal, 'developing countries', 'low and middle income countries', socioeconomic, impacts, acceptability, barriers and innovations. We also searched reference lists from key reviews and articles to identify additional publications of interest.

We did not attempt a formal, comprehensive systematic review of the literature due to the breadth and complexity of the topic, and the large variety in the type of reference materials examined. Nevertheless, we screened articles based on titles, abstracts, and full texts, and purposively selected representative articles for inclusion in this review based on the following criteria:

i) Inclusion criteria: Any publications relating to the testing, validation, review and commentary of diagnostic POCTs for infectious animal diseases (including zoonoses) in LMICs, published in English, in any year through and including January 2020. We selected studies that were relevant under the following categories: 1) usage, including reviews, trials and comparative studies of diagnostic infectious animal disease POCTs in LMICs; 2) considerations for aspects of the 'ideal' POCT, with particular emphasis on applications in LMICs; 3) barriers to usage and uptake of infectious animal disease POCTs in LMICs.

ii) Exclusion criteria: Any publications involving non-diagnostic POCTs, or POCTs for diagnosis of non-infectious diseases in animals or of human-only diseases; publications relating to non-POCT animal diagnostic methods; any media in any languages other than English. Foreign language material was excluded because of the cost and time required for translation.

Where specific examples of publications regarding usage, implementation or impact of veterinary POCTs in LMICs were missing from the literature, we actively searched for relevant examples from the medical literature in order to provide a comparison for discussion. We also actively searched organisational websites including the World Organisation for Animal Health (OIE) and Food and Agriculture Organization of the United Nations (FAO) for information relating to POCTs for diagnosis of animal diseases. We also identified manufacturers of POCTs from key publications and documents, and

actively searched the internet for additional POCT manufacturers to identify those currently producing commercial diagnostic veterinary POCTs, and to obtain validation data and test kit inserts for infectious animal disease POCTs where available. The final bibliography included 567 documents (including journal articles, reports and guidelines) and 19 commercial POCT kit inserts from 11 manufacturers. This review entitled "The potential of diagnostic point-of-care tests (POCTs) for infectious and zoonotic animal diseases in developing countries: technical, regulatory and sociocultural considerations" has subsequently been published (1).

5.1.1 Key findings

Currently available diagnostic animal disease POCTs

Many POCTs are currently being used in LMICs for diagnosis of infectious animal diseases, particularly transboundary diseases that pose serious risk to human and/or animal health and trade such as FMD, highly pathogenic avian influenza (HPAI), rabies and ASF. While many published reports describe early development and evaluation of animal disease POCTs, and remark on their potential application to LMIC contexts, few actually take the validation beyond the laboratory and into the settings where they are predicted to have the biggest impact. However, some notable field validation studies do exist in the literature, for example portable PCR and LAMP assays for FMD detection in Africa and Asia (5, 8, 15, 16, 17 18); LFAs used for AIV screening of village chickens in Indonesia (6, 19, 20, 21, 22,), and PPR detection in small ruminants in Asia and Africa (23, 24, 25).

Field studies in eastern Africa used a commercially-available, pan-serotype-specific PCR-based assay for detection of FMD using lyophilised reagents and a portable, field-ready thermocycler, and obtained diagnostic accuracy comparable to that of an OIE-recommended laboratory-based test (8). The POCT was further able to reliably detect different serotypes of FMD viral material in a variety of samples taken from pre-clinical, clinical and clinically-recovered cattle, with results available in under 90 minutes (8). A LAMP assay has also been developed and validated for rapid field detection of FMD, specifically designed to optimise the speed and operability of the test by non-laboratory personnel on unextracted field samples (5). Loth et al (26) conducted field testing of two LFAs for detection of HPAI in oropharyngeal swabs taken from free-ranging village chickens in Indonesia, and PCR testing on replicate swabs confirmed diagnostic sensitivities and specificities of the POCTs as 69-71% and 98%, respectively. More recently, a Vietnamese pilot study reportedly took portable nucleic acid extraction and insulated isothermal PCR platforms into live bird markets to conduct rapid, on-the-spot testing of oropharyngeal swabs from poultry for detection of HPAI (27). A LFA for the detection of PPR virus in ocular and nasal swabs was trialled in field sites in Pakistan, Ethiopia, Ivory Coast and Uganda, and from the test results obtained within 15-30 minutes reported diagnostic sensitivity and specificity of 84% and 95%, respectively, compared to PCR (28). The authors of this study also reported feedback from the field trials as being uniformly positive, with the portability of the tests and ease of use particularly emphasised.

Whilst the above cited publications report on the technical performance of their respective POCTs in the field, the impacts of their usage for in-country disease control or surveillance and potential downstream benefits of rapid results for local communities were not readily available from the literature.

Considerations for the 'ideal' POCT

While POCTs are designed to be simple and easy to use, their underlying biochemical processes are nevertheless highly sophisticated, and results need to be interpreted with due consideration of the tests' fitness for purpose, including strengths, limitations, and applications in various settings (29).

Target product profiles (TPPs) outline the desired 'profile' or characteristics of a new product, and are used by organisations such as the OIE, United States Food and Drug Administration (FDA), and Drugs for Neglected Diseases Initiative as planning tools to identify key test criteria, guide test development, and set research and development (R&D) targets for funders and developers (30, 31). Ideally, TPPs for each novel POCT should be defined through several rounds of discussions with key stakeholders including disease experts, target users and manufacturers.

Once these characteristics have been defined to ensure fitness for purpose, other considerations should also be addressed. The 'ASSURED' criteria set out by the World Health Organisation (WHO) state the ideal characteristics for a field-ready diagnostic test as Affordable, Sensitive (few false negatives), Specific (few false positives), User friendly (able to be performed in a few steps with minimal training), Robust (no cold storage needed) and rapid (results available in under 30 minutes), Equipment-free, and Deliverable to those who need it (32). Social studies have indicated that shorter diagnostic turnaround times (up to 20 minutes), high diagnostic accuracy (sensitivity and specificity above 90%), low cost, and ease of use are particularly important factors for some public health workers (33 ,34). It should be emphasized that there can be no 'one size fits all' description of the ideal diagnostic POCT, as fitness for purpose will be determined by the needs, intentions and resources of the veterinary services in each distinct geopolitical setting.

Additional considerations for POCTs that are to be used in LMICs include portability and suitability for being used outdoors, ease of sample acquisition and preparation, availability of consumables and reagents, storage and transport conditions for devices and reagents, viable options for device maintenance and requirements for quality control testing, which may include provision of positive and negative controls specific to the disease agent strains in that LMIC. Factors that would be expected to increase user uptake in LMICs include capability for multiplexing (allowing testing for multiple pathogens in parallel) and for differentiating infected from vaccinated animals.

5.1.2 Barriers for usage and uptake of animal disease POCTs in LMICs

Some key themes were identified from the literature that may be preventing maximal usage and uptake of diagnostic animal disease POCTs in LMICs, as presented in Table 1.

Table 1: Major barriers to usage and uptake of animal disease POCTs in LMICs

Theme	Considerations	Impacts in LMICs
Lack of validated and affordable diagnostic POCTs for infectious animal diseases	• Diagnostic POCTs for many important animal diseases are not commercially available at present (35) • Lack of buying power from LMICs for neglected animal disease POCTs (2) • Lack of funding and resources for POCT manufacturers in LMICs (31)	• Diagnostic POCTs for animal diseases are not selected for commercial development • Neglected animal diseases continue to be under-diagnosed and underreported back to local communities
Underutilised central register of approved POCTs for detection of infectious animal diseases and no regulations enforcing test validation	• The OIE 'register of approved diagnostic kits' only lists 2 POCTs (https://www.oie.int/scientific-expertise/registration-of-diagnostic-kits/the-register-of-diagnostic-kits/)	• POCT users are unable to reliably differentiate approved, fit for purpose POCTs from cheaper, unvalidated tests • Using inaccurate, unfit tests in LMICs wastes

Theme	Considerations	Impacts in LMICs
	• No other central register for approved veterinary diagnostic POCTs exists • Lack of regulations enforcing test validation for animal disease diagnostics – including POCTs means POCTs of varying quality are being marketed (35)	limited resources and may undermine confidence in POCTs • Using inaccurate, unfit POCTs in LMICs can cause significant over- and under-reporting of animal diseases, which lead to secondary impacts on human and animal health, and trade
Limitations to current POCT validation and regulatory processes	• Lack of consistency and transparency of validation data specific to POCTs (36) • Lack and limitations of field validation studies for infectious animal disease POCTs (1) • Technical difficulties and costly to conduct full validation for some infectious animal disease POCTs (1)	• Using inaccurate, unfit tests in LMICs wastes limited resources and may undermine confidence in POCTs • Using inaccurate, unfit tests in LMICs can cause significant over- and under-reporting of animal diseases, which can have important impacts on human and animal health, and trade
Socioeconomic considerations for animal disease POCTs at the community level	• Veterinary services must be capable of reporting and acting on POCT results (37) • Policies should be in place to ensure animal owners are suitably compensated for impacts of test positive animals to avoid unintended actions by local farming communities such as hiding or salvage selling (38) • Limited capacity building opportunities for POCT users (39) • Communities must be engaged and educated regarding POCT use, benefits and limitations (38)	• Animal owners are not motivated to spend money on diagnostics • Animal owners may be reluctant to test or report sick or disease animals, and may lead to unauthorised movement, hiding or salvage selling of these animals • Transmission of animal diseases may be exacerbated, and disease control efforts undermined

5.2 Engage with manufacturers of animal disease POCTs regarding test validation processes

Organisations that currently or have previously manufactured veterinary diagnostic POCTs (“manufacturers”) were identified during Google searches, and from publications examined during the literature review. The manufacturer questionnaire (Appendix 2) was

developed and emailed out to the manufacturer contact list in February 2020, with reminder emails sent in late February and June 2020.

In total, 39 invitations were sent to 38 organisations. Responses were received from 13 organisations (13/39 = 33% response rate), but one respondent did not consent to undertaking the questionnaire and was not included in the final dataset. Ultimately, 12 completed responses were received, making an overall participation rate of 31% (12/39).

5.2.1 Results of manufacturer questionnaire

Respondent and demographic data

Of the 12 completed responses almost all were from company product managers (6/12, 50%) or directors (5/12, 42%), with one response from a scientist (8%). Companies were located mainly in Europe (7/12, 58%), with two each based in Australia/Pacific and East/Southeast Asia, and one in North America (Table 2). Three quarters (9/12, 75%) of the companies employed less than 100 employees; two (17%) employed between 100 and 499, and one (8%) employed 1000 or more. Table 2 describes the range of POCTs produced by surveyed manufacturers and the disease targets.

Current and in-development manufacture of POCTs

LFAs were the most commonly manufactured POCT type, made by 58% (7/12) of respondents. Portable PCRs, LAMPs and primers/reagents are also commonly manufactured, each being reported by 33% (4/12) of respondents. Two of the respondents do not currently manufacture POCTs. Five respondents (42%) reported that their organisation currently manufactures only one type of POCT, four of which were LFAs. Two organisations reportedly manufacture four different POCT formats, both of which included LFAs, portable PCRs, LAMPs and primers/reagents. None of the respondents are currently manufacturing nanopore sequencing devices.

For the ten manufacturers currently making animal disease POCTs, these items made up an average of 45% of manufactured products, but ranged between 1 and 100%. The proportion of companies' incoming revenue that was from the sale of animal disease POCTs also ranged from 1 and 100%, but with average and median proportions of 40% and 25%, respectively.

Of the organisations currently manufacturing animal disease POCTs, the most commonly reported disease targets were ASF and AIV (both 6/10, 60%), followed by FMD (5/10, 50%), brucellosis (4/10, 40%) and CSF (3/10, 30%). Other POCTs are reportedly targeting bluetongue virus, Hendra/Nipah virus, IBD, ND and rabies (each reported by 2/10, 20%) and many more including anthrax, *Mycoplasma bovis*, PPR, PRRS, porcine epidemic diarrhoea and equine influenza (each reported by one manufacturer). POCTs targeting diseases of companion animals, including feline herpesvirus and parvovirus, were also reported by two manufacturers.

POCTs currently in development included a range of infectious and vector-borne companion, production and pest animal diseases, zoonotic diseases including COVID-19, and plant diseases.

Table 2: Surveyed manufacturer demographics, POCTs produced and disease targets

Manufacturer	Head Office Location	POCTs as % of products	POCTs as % of revenue	Type of POCT produced	Disease targets
1	Asia	85	85	LFA, Portable PCR	ASF, AI, Brucellosis, CSF, FMD, IBD, rabies, MERS,
2	Asia	0	0	Not applicable	Not applicable
3	Australia/ Pacific	50	35	LAMP, general diagnostic reagents	FMD, ASF, AI, virulent footrot, Hendra/Nipah
4	Australia/ Pacific	0	0	Not applicable	Not applicable
5	Europe	10	16	LFA	ASF, CSF, BTV, M bovis
6	Europe	1	1	Portable PCR, general diagnostic reagents	ASF, AI, FMD
7	Europe	100	100	LAMP	Theleria equi, Babesia caballi, Anaplasma borrelia, leptospirosis, EI, EHV1 & 4, Strep. equi, feline herpes, parvovirus and feline calici virus
8	Europe	10	25	LFA	Calf scours, inflammatory markers
9	Europe	25	25	LFA	Diarrhoea of production animals
10	Europe	50	85	LFA	AI, brucellosis and infectious diseases of companion animals
11	Europe	96	44	LFA, Portable PCR, LAMP, general diagnostic reagents	ASF, AI, brucellosis, FMD, IBD, NDV, rabies, salmonella, pasteurella, enzootic bovine leucosis, coronavirus, E. coli K99, BTV, coccidiosis
12	North America	20	25	LFA, Portable PCR, LAMP, general diagnostic reagents	ASF, AI, brucellosis, CSF, FMD, Hendra/Nipah, PPR, Anthrax, PRRS, PED Swine SADS coronavirus

Target users and target markets of manufactured POCTs

Most manufacturers reported that their POCTs were intended to be used by a variety of operators, predominantly private veterinarians (11/12, 92%), government veterinarians and veterinary laboratories (each 8/12, 67%), community animal health workers and/or animal technicians (7/12, 56%), and farmers and/or animal owners (6/12, 50%). Only one manufacturer (based in Europe and targeting production animal diarrhoea disease (1/12, 8%) reported a single target user of their animal disease POCTs, which was private veterinarians; all others reportedly targeted two or more different types of POCT users.

Of the ten organisations that are currently manufacturing POCTs, all are reportedly exporting between 30 and 100%, with average and median proportions of 70% and 81% POCTs being exported, respectively. The major target markets are Europe (5/10, 50%) and East and Southeast Asia (4/10, 40%).

POCT R&D

The major motivation for commencing research and development (R&D) into a new animal disease POCT was to fill an assessed commercial opportunity based on the company's recognised gap or need in the market (10/12 manufacturers, 83%). Other reasons included to fulfill a commercial contract (4/12, 33%), in response to an external funding call (3/12, 25%) and to compete with existing products (2/12, 17%).

The funding source for POCT R&D was usually internal company revenue (10/12, 83%), followed by commercial contracts (6/12, 50%), government (4/12, 33%), NGOs, (2/12, 17%) and private donors (1/12, 8%). Most (7/11, 64%) of organisations reportedly obtained funding from more than one source.

The overall R&D process for a new animal disease POCT reportedly took up to two years, with 5 respondents (42%) reporting completion in under 6 months, 1 respondent reporting completion between 7 and 12 months, and 5 respondents reporting completion between 1 and 2 years. Overall costs of POCT R&D reportedly ranged between $30 USD - $235,000 USD, with average and median costs of $114,500 USD and $88,500 USD, respectively.

5.3 Interview veterinary services personnel in Asia-Pacific region regarding animal disease POCT knowledge, attitudes and practices

Interviews were planned to investigate current knowledge, attitudes and practices of animal health personnel using diagnostic animal disease POCTs in the Asia-Pacific region ("users"). A participant information sheet and user questionnaire template were developed (Appendix 3) and received CSIRO ethics approval (approval number 175/19) for participant interviews to be conducted between 1 January 2020 and 30 June 2020. A contact list of key veterinary and laboratory organisations and personnel was developed, leveraging working relationships wherever possible, and invitations for in-country interviews were emailed in early 2020. Interviews began in January 2020, with one interview conducted in Indonesia and four in Vietnam. However, with COVID restricting most international and regional travel during 2020, a decision was made in March 2020 to convert the questionnaire to an online form that could be emailed to participants (Appendix 4), who were also offered the option of being interviewed via phone or video call. Reminder emails were sent out monthly until 22 June 2020.

5.3.1 Results of user questionnaires

Respondent and demographic data

Overall, 48 potential participants were contacted from 9 countries, with completed responses received from 18 participants in 8 countries (Table 3).

Table 3: User questionnaire survey invitations and participants.

Country	**Invitations sent**	**Completed responses– face-to-face or via WebEx**	**Completed responses– online**	**Response rate**
Cambodia	7	1	1	28.6%
Lao PDR	3	0	1	33.3%
Thailand	6	0	0	0.0%
Vietnam	12	4	0	33.3%
Indonesia	5	2	2	80.0%
Myanmar	8	0	3	37.5%
Philippines	1	0	1	10.00%
Bhutan	1	0	1	100.0%
Australia & Pacific*	5	0	2	40.0%
TOTALS	48	7	11	37.5%

*Australian-based consultants who work across multiple locations, including Timor-Leste and Papua New Guinea.

Of the 18 respondents to the user questionnaire, 11 (61%) were male. A broad range of age groups, experience, education, roles and geographical location were represented (Table 4)

Table 4: Demographic data of respondents (user questionnaire)

	Number of respondents	Percentage of respondents
Gender		
Male	11	61.1%
Female	7	38.9%
Age group		
26-35 years	4	22.2%
36-45 years	5	27.8%
46-55 years	4	22.2%
56 years and older	4	22.2%
No data	1	5.6%
Current role		
Veterinarian	14	77.8%
Veterinary technician	1	5.6%
Manager	1	5.6%
Senior researcher	1	5.6%
Consultant	1	5.6%
Current employer		
Government veterinary services- national	4	22.2%
Animal health laboratories – national	1	5.6%
Animal health laboratories – state/regional	4	22.2%
Laboratories – other	2	11.1%
International non-profit organisations	2	11.1%
Academia / research	2	11.1%
Consultancy / other	3	16.7%
Length of time in role		
Less than 1 year	2	11.1%
Between 1 and 5 years	7	38.9%
Between 6 and 10 years	5	27.8%
More than 10 years	4	22.2%
Highest education		
Diploma / certificate	1	5.6%
Bachelor degree	2	11.1%
Master degree	8	44.4%
Doctoral degree	7	38.9%
Country/ies of work*		
Indonesia	6	33.3%
Vietnam	4	22.2%
Cambodia	3	16.7%
Myanmar	3	16.7%
Bhutan	1	5.6%
Philippines	1	5.6%
Timor-Leste	2	11.1%
Lao PDR	1	5.6%
Papua New Guinea	1	5.6%

*Australian-based consultants work across multiple locations.

Current diagnostic practices

Laboratory testing

Most users reported using multiple diagnostic methods for diagnosing animal diseases and were on average ‘quite satisfied’ (average score 2.93 out of 5) with current diagnostic methods. Over 70% of respondents reportedly collect and send samples to veterinary laboratories for diagnosis of animal diseases. In most cases, costs of laboratory testing are covered by the government, with no charges to animal owners, except in Myanmar and Cambodia where costs of between $10–58 USD are reportedly being charged to owners. Average turnaround times from sampling to results typically ranged between one day and one week, with the exception of one month for FMD samples being sent from

Vietnam to the world FMD refence lab [Pirbright Institute, United Kingdom] (Table 5). National sample transport networks were reported to be 'effective' in Bhutan, the Philippines and Myanmar, 'limited' in Timor-Leste, 'ineffective' in Cambodia and variable in Vietnam – one Vietnamese respondent each rated the national network for transport of AIV samples as 'effective' and 'ineffective', and the FMD sample network was rated as 'effective' for transport to the regional Vietnamese laboratory, but 'ineffective' for transport to the world reference laboratory in the UK.

Table 5: Current laboratory diagnostic data for selected animal diseases

Disease	Average turnaround time for results	How much do owners pay for test? ($ USD)	Do you start treatments while waiting for results?	Is there an effective sample network in place?
ASF & CSF	1 day (Cam, Mym) 1-2 days (Bht, Viet) 5 days (Php) 1 week (TL)	$0 (Bht, Php, TL, Viet) $11 (Mym) $58 (Cam)	No (all)	Yes (Bht, Php, Mym) Limited (TL) No (Cam)
AIV	1 day (Cam, Mym) 1-2 days (Bht, Viet) 3 days (Indo) 5 days (Php)	$0 (Bht, Indo, Php, TL, Viet) $11 (Mym) $58 (Cam)	Yes (Mym) Depends on local government policy (Indo) No (Others)	Yes (Bht, Mym, Php, Viet1) Limited (TL) No (Cam, Indo, Lao, Viet2)
Brucellosis	1 day (Cam) 1-2 days (Bht) 5 days (Php) 1 week (TL)	$0 (Bht, Php, TL) $17 (Cam)	No (all)	Yes (Bht, Php) Limited (TL) No (Cam)
FMD	1-2 days (Bht, Cam, Viet- to regional lab) 5 days (Php) 1 week (TL) 1 month (Viet- to world reference lab)	$0 (Bht, Php, TL) $16.50 for ELISA (Cam) $58 for PCR (Cam)	No (all)	Yes (Bht, Php, Viet- to regional lab) Limited (TL) No (Viet- to world ref lab)
IBD	1-2 days (Bht) 5 days (Php)	$0 (Bht & Php)	No (Bht & Php)	Yes (Bht & Php)
ND	1 day (Cam) 1-2 days (Bht) 5 days (Php) 1 week (TL)	$0 (Bht, Php, TL) $58 (Cam)	No (all)	Yes (Bht, Php) Limited (TL) No (Cam)
PPR	1-2 days (only Bht)	$0 (only Bht)	No (only Bht)	Yes (only Bht)
Rabies	1 day (Php) 1-2 days (Bht, Viet) 1 week (TL)	$0 (all)	No (all)	Yes (Bht, Viet) Limited (TL)

ASF= African swine fever; CSF= classical swine fever; AIV= avian influenza virus; FMD= foot and mouth disease; IBD= infectious bursal disease; ND= Newcastle disease; PPR= peste des petits ruminants; PRRS= porcine reproductive and respiratory disease; JE= Japanese encephalitis.
Bht= Bhutan; Cam= Cambodia; Indo= Indonesia; Lao= Lao PDR; Mym= Myanmar; Php= Philippines; TL= Timor-Leste; Viet= Vietnam

The most selected barrier to laboratory diagnosis was unaffordability for owners (39% of respondents), with seven respondents from six countries selecting this option, even though lab tests were reportedly free of charge for animal owners in three of those countries (Indonesia, Lao PDR and Philippines). Other commonly reported barriers included long turnaround times for results (33% of respondents), and that owners did not want laboratory results (28% of respondents).

POCTs

Most respondents (15/18, 83.3%) had used or were familiar with LFAs, followed by portable PCR devices (10/18, 55.6%) and LAMP (5/18, 27.8%). No users were reportedly familiar with nanopore based sequencing devices. On average, respondents reported that they currently found POCTs 'quite useful' (average score 2.92 out of 5). 7 of the 18 respondents were lab based and found POCTs somewhat useful (average 2.7 score out of 5), compared with an average score of 3.1 (quite useful) for non-laboratory based users.

A wide range of POCT formats are reportedly being used in the region for many infectious animal diseases. For ASF for example, POCT types currently in use include LFAs developed by Ingenasa, Bionote/Anigen and Shenzhen Lvshiyuan Biotechnology Co. Ltd.; POCKIT iiPCR by GeneReach; and LAMP, developed by OptiGene. For detection of AIV, LFAs are also being utilised in the region, and some POCTs including Ingenasa's IndiField's Point of Need PCR system and other portable PCR assays are also being validated at present in Cambodia. Other respondents reported using POCTs for detection of brucellosis, FMD, IBD, ND and rabies.

Most respondents reported POCT usage for only a few of the ten OIE priority diseases that were selected for inclusion in the questionnaires (ASF, AIV, brucellosis, CSF, FMD, Hendra/Nipah, IBD, ND, PPR, rabies), while some also reported POCT use for additional animal diseases, including PRRS and pan-coronavirus and COVID-19 animal surveillance using a portable PCR device (Cambodia), caprine mastitis and porcine epidemic diarrhoea (Philippines), swine influenza (Vietnam) and for diseases of companion animals including canine parvovirus, distemper, *Ehrlichia* spp (Indonesia, Bhutan and the Philippines).

Two responses regarding current POCT usage were of particular note: in Bhutan, current and frequent POCT usage was reported for seven of the ten listed diseases and four additional canine diseases, and it was stated that for the additional three listed diseases (ASF, CSF, Hendra/Nipah), POCTs would be used if they were available due to their convenience of obtaining a test result whilst in the field. Also of note was the response from Lao PDR which stated that LFAs for several diseases (AIV, FMD, ND and rabies) had been given to their national laboratory; the respondent reported that the kits were developed by the Shenzhen Lvshiyuan Biotechnology Co. Ltd. company based in China. As these kits were provided in large quantities we conducted an online search to obtain the test kit literature, however, we were met with difficulties; the company's website (lsybt.com) was repeatedly blocked by the server due to security issues, and the only other discoverable listing for the products were on online marketplaces such as Alibaba.com. Here, the product listing included the test procedure and some information regarding reagent storage conditions, but provides no information regarding the test's validation or verification processes, nor any data regarding the test's diagnostic accuracy such as sensitivity or specificity values. For other POCTs a search of the manufacturers websites generally do not describe the tests performance as this material is often only supplied in the technical brochure that follows a purchase of the test.

Veterinarians, veterinary technicians and laboratory staff were the most commonly reported operators and interpreters of POCTs. POCT results were typically available within 10–20 minutes but ranged up to 1 day for LFAs, and were within 60–90 minutes for portable PCR devices. Most respondents reported that costs of POCTs were borne by governments and no charges were issued to animal owners, except for some cases – for example, animal owners are reportedly paying $11-15 USD for AIV, ND and rabies testing by LFA in Myanmar, and between $20-40 USD for AIV testing by portable PCR in Cambodia.

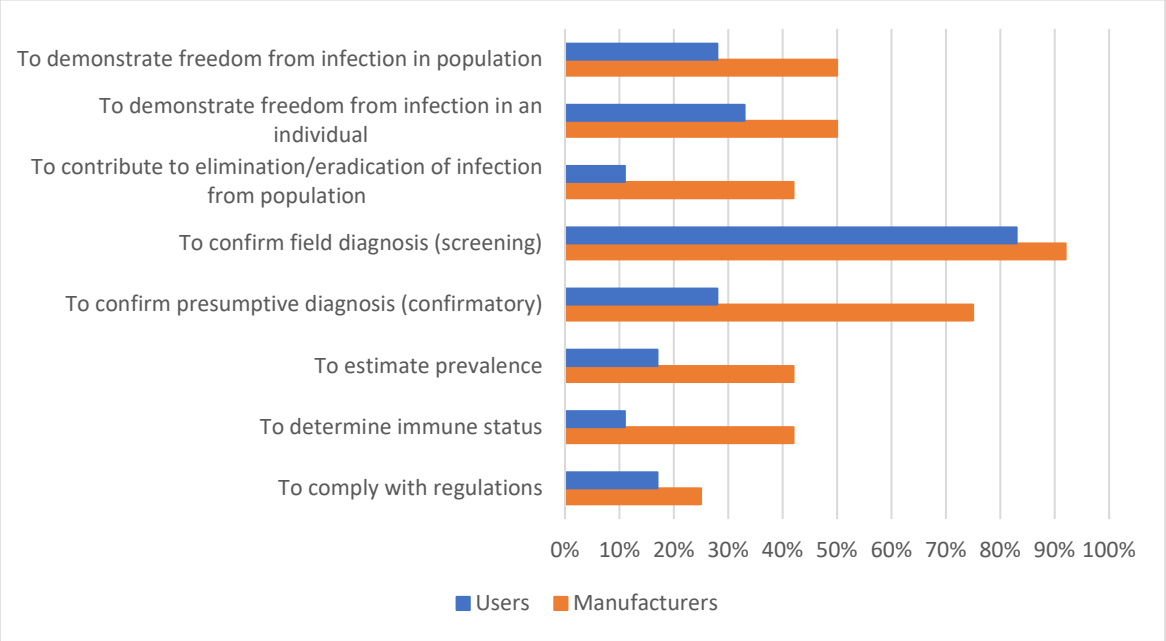
5.4 Comparison of user and manufacturer survey responses, including areas of agreement and disconnect

A number of questions in the surveys were posed to both users and manufacturers in order to investigate potential areas of accord and disconnect.

5.4.1 Purposes and factors for (non) use of animal disease POCTs

The most common purpose for both the manufacture and use of animal disease POCTs was for confirmation of field diagnosis (screening), with 92% of manufacturers and 83% of users selecting this option (see Figure 1). Manufacturers were more likely to select additional options than users: six of the seven remaining options were selected by at least 40% of manufacturers, while only three of the remaining options were selected by more than 25% of users.

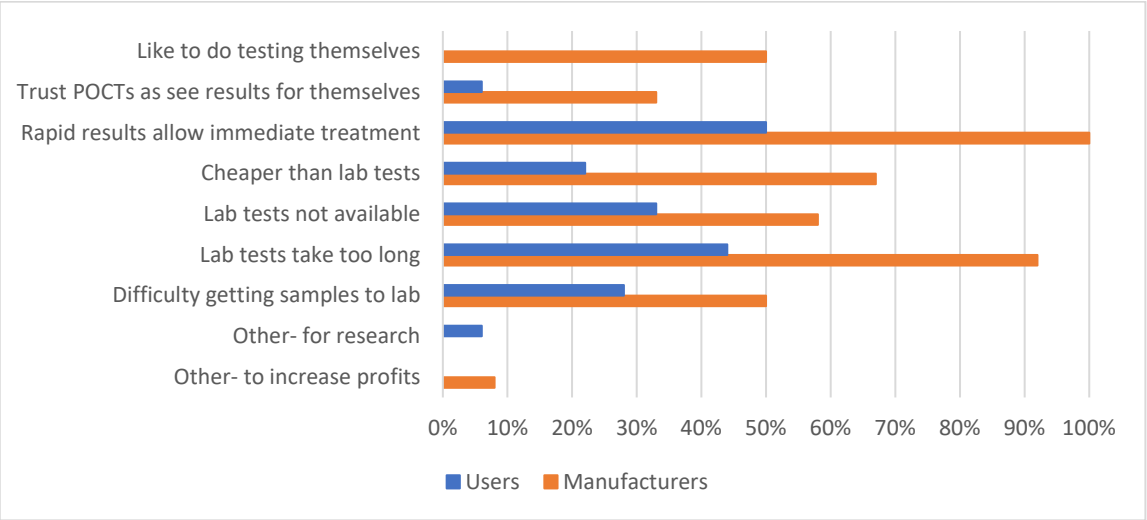
Figure 1: Major stated purpose for manufacture and use of animal disease POCTs.



Both sets of respondents were asked to select the most important factors that affected whether users would and would not purchase/use animal disease POCTs. The two most important actual (users) and predicted (manufacturers) factor for using POCTs was that the rapid results allow diagnosis and immediate implementation of treatment, and that laboratory tests take too long to be useful, although both options were selected by larger proportions of manufacturers compared than of users (see Figure 2). Other factors commonly selected by users included that laboratory tests are not always available (33%), and that it is too difficult to send samples to the laboratory (28%).

Manufacturers were again far more likely to select the remaining options than users, including 50% of manufacturers selecting the option that users like to do the testing themselves, compared to 0% of users.

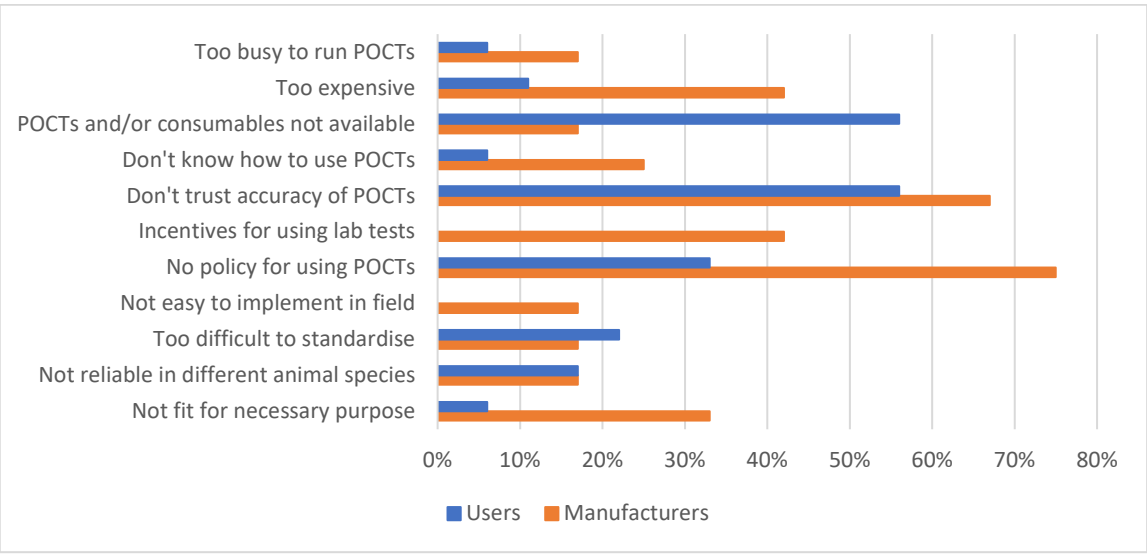
Figure 2: Most important reasons for buying/using animal disease POCTs: actual (users) vs predicted (manufacturers)



Regarding the factors that users do not use POCTs, a greater discord was observed between the user and manufacturer responses (see Figure 3). Over 50% of users indicated that a lack of trust in the accuracy of POCTs and difficulty in obtaining POCTs and/or their reagents were the major factors for not using POCTs. Other important factors included a lack of policy recommendations or guidelines for using POCTs (33%) and difficulty standardising POCTs (22%).

Most manufacturers (75%) selected a lack of policies and guidelines as the major factor for non-use of POCTs, followed by distrust in POCT accuracy (67%) and that POCTs are too expensive (42%, compared to 11% of users). Unavailability of POCTs and their reagents was only selected by 17% of manufacturers. Manufacturers also believed that incentives for using laboratory tests (42%) and difficulty implementing POCTs in the field (17%) play a large role in non-use of POCTs, while these options were not selected by any users.

Figure 3: Most important reasons for not buying/using POCTs: actual (users) vs predicted (manufacturers).

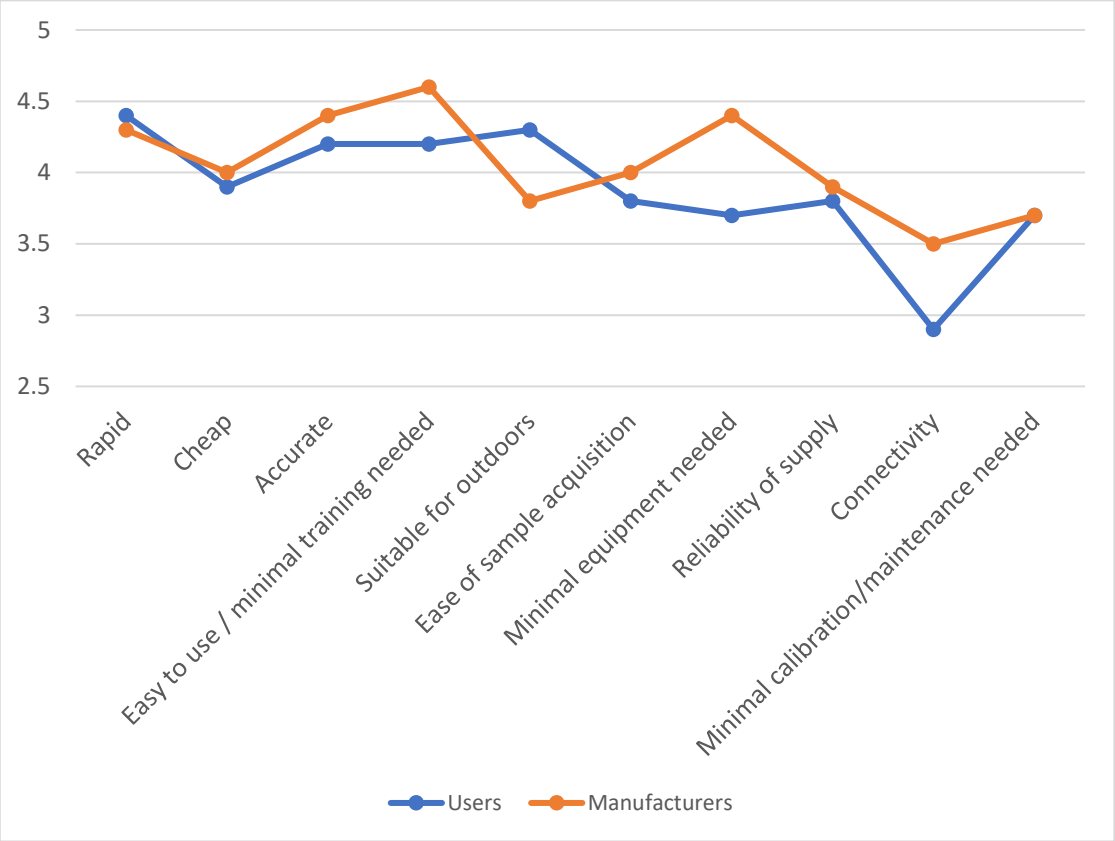


Participants were asked to rate the actual (users) and perceived (manufacturers) importance of various factors that influence decisions to buy or use animal disease

POCTs on a scale from 1 ('not at all important') to 5 ('extremely important'). Responses were averaged for each category and are presented in Figure 4. All average scores ranged between 2.9 and 4.6.

Rapidity of results was rated highest by users (average score of 4.4). Manufacturers rated ease of use/minimal training needed the most important factor (average score of 4.6), with accuracy and minimal equipment equal second (average score 4.4). compared to 4.2 from users. Suitability for outdoors e.g. portable, waterproof, battery-powered was rated second highest for users (average score 4.3) but third last for manufacturers (average score 3.8). Connectivity was rated lowest by both users (2.9) and manufacturers (3.5).

Figure 4: Rated importance of POCT factors: actual (users) vs perceived (manufacturers).



5.4.2 Values for the ideal POCT

Participants were asked to define the terms 'rapid', 'cheap' and 'accurate', by providing ideal values. The ideal turnaround time for animal disease POCTs was overwhelmingly within 30 minutes or less, as answered by 64% of users and 75% of manufacturers. Approximately 20% of both sets of respondents also selected within one to three hours.

Ideal running costs per test were not as synchronised between the two groups. Almost three-quarters of users (70%) stated ideal running costs of $10 USD or less, compared to 59% of manufacturers. Costs of between $10 and $20 USD were suggested by 33% of manufacturers, compared to 10% of users. Interestingly, 20% of users suggested costs of $20-50 USD, compared to 8% of manufacturers.

Responses for ideal diagnostic sensitivity and specificity values are presented in Table 6. The most commonly stated ideal diagnostic sensitivity value was '90% or above', stated by 54% of users and 50% of manufacturers. The remaining user responses were reasonably well spread from '70% and above' (8%), to '100%' (15%), whereas one-quarter of manufacturers stated '95% or above' and none stated '70% or above'. In total,

diagnostic sensitivity values of 90% or above were stated by 77% of respondents and 92% of manufacturers.

Diagnostic specificity values were more disparate within and across both groups. The highest proportion of users selected '90% or above' (25%) but selected four of the other five categories in equal numbers (17% of users). One third of the manufacturers each selected '90% or above' and '95% or above', and a quarter selected '100%'. Overall, 75% of users stated ideal diagnostic specificity values of 90% or above, compared to 92% of manufacturers.

Overall, respondents from both groups rated ideal diagnostic specificity values higher than for diagnostic sensitivity, with 50% of users and 58% of respondents stating ideal DSp values 95% or higher. For DSe, only 23% of users and 42% of respondents stated ideal values of 95% or higher.

Table 6: Ideal diagnostic sensitivity and specificity values for animal disease POCTs as stated by users and manufacturers

	Ideal DSe		Ideal DSp	
Ideal diagnostic value	**Users**	**Manufacturers**	**Users**	**Manufacturers**
70% or above	8%	0%	8%	0%
80% or above	15%	8%	17%	8%
90% or above	54%	50%	25%	33%
95% or above	8%	8%	17%	33%
99% or above	0%	25%	17%	0%
100%	15%	8%	17%	25%
Overall diagnostic values of 90% or higher	77%	92%	75%	92%
Overall diagnostic values of 95% or higher	23%	42%	50%	58%

DSe= diagnostic sensitivity; DSp= diagnostic specificity

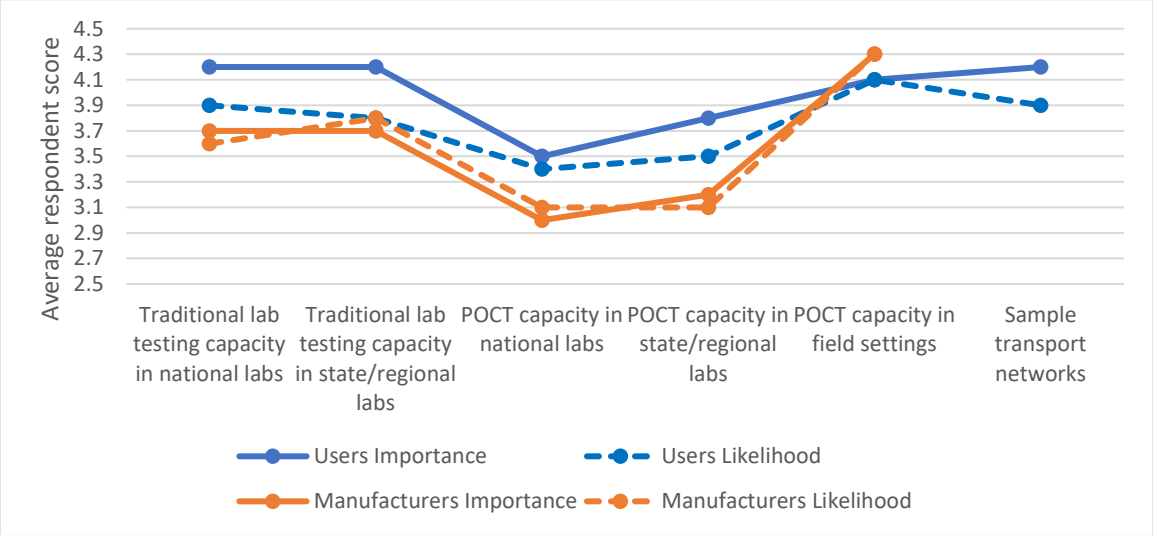
5.4.3 Future planning

Respondents were asked to consider the likelihood and importance of government (users) and organisational (manufacturers) investment in improving laboratory and POCT testing capacity in different settings over the next five years, by rating each category on a scale of 1 ('not at all likely/important') to 5 ('extremely likely/important'). Users were also asked to consider the likelihood and importance of government investment in improving sample transport networks. Average scores are presented in Figure 5.

All average scores ranged between 3 and 4.3, with a narrower range for users (range of 0.8) than for manufacturers (range of 1.3). Users consistently rated the importance of government investment in all six categories higher than the likelihood, except for 'POCT capacity in field settings' which they scored equally likely and important (4.1). Improving traditional laboratory testing capacity in both national and regional settings, and improving sample transport networks, were rated of equal highest importance to users (average score 4.2).

Manufacturers tended to rate both the importance and likelihood of their organisational investment in improving the various categories lower than the users. The one exception was improving POCT capacity in field settings, which they equally rated as both more important and more likely (average score 4.3) than did users. Increasing capacity of traditional laboratory testing in both national and regional settings were rated higher than for POCTs in those settings.

Figure 5: Perceptions of importance and likelihood of investment in improving diagnostic capacity over the next five years



5.4.4 Additional comments provided by respondents

The final question of both surveys invited respondents to provide additional comments relating to animal disease diagnosis. More users (10/12, 83%) provided additional comments than manufacturers (4/12, 33%). Responses from both groups were combined into Table 7 according to theme.

Table 7: Additional considerations raised by survey respondents.

Theme	Sub-theme	Comments	Respondent type*
Technical	Test accuracy	• POCT accuracy should have good correlation with laboratory tests • If a POCT returns a positive result, users should be able to have confidence in the results without needing laboratory confirmation	U
	Test validation	• Test validation needs to be verified before implementing	U
	Quality assurance and quality control	• Discussions are needed regarding quality assurance and quality control of POCTs	U
	Other	• Dried-down (lyophilised) consumables are very important to avoid need for cold-chain	M
Education	Baseline research	• A technical review is needed to demonstrate the current situation of what POCTs are out there, which ones are being used, what the validation processes are currently like and how they can be improved	U

Theme	Sub-theme	Comments	Respondent type*
	Social sciences	• Need to capture practitioners' real limitations to using more POCTs	M
	Engagement and awareness	• Need to educate and engage users and policy makers to increase uptake of POCTs	U
		• Currently, capacity building tends to just involve buying 'stuff' for users rather than understanding what their needs are and what would be most useful	U
	Training	• Some couriers refuse to accept diagnostic animal disease samples for transport to laboratories because they are afraid of infection, don't know how to package the samples correctly or what the requirements are	U
		• Field teams need to be appropriately trained, including in areas of biosafety, use of PPE, risk communication	U, M
Economic	Data analysis	• Governments would be interested in cost-benefit analyses to compare POCTs with traditional lab tests	U
		• Need economic analysis of impacts of false positive and false negative POCT results, especially for high-impact diseases	M
	Affordability	• POCTs should be affordable to all users, even those in least developed countries	U
	Funding	• Sourcing funding for POCT research and development is difficult	M
Policies and politics	Variability of government support	• Government buy-in is key	U
		• Support for POCT policy has to come from highest levels of government, so when governments change, support for various projects and areas can suddenly be withdrawn	U
	Favouritism	• Some international organisations and governments heavily promote the use of one particular POCT	U
	International regulation and approval	• POCTs should be recommended/recognized by the OIE or an OIE reference laboratory	U
		• Without OIE endorsement, a POCT is unlikely to get government approval for its use	U
		• A working group is needed to define standards for POCT validation based on OIE/FAO guidelines, and to publish a list of approved POCTs and data	U

Theme	Sub-theme	Comments	Respondent type*
		about their diagnostic accuracy and fitness for different purposes, to assist and empower users	

* U= user; M= manufacturer

6 Objective 3: Work with stakeholders (industry, government and international agencies) to develop a guidance document on the use and best practice for POC diagnostics for animal disease investigation and surveillance

The results obtained from the user and manufacturer questionnaires identified many issues and constraints currently existing with POCTs. Even though the overall response rate to the questionnaires was low there was generally a consistency of findings. It is proposed that the results will be presented at a future ASEAN Regional Laboratory Technical Advisory Group (LabTAG) meeting, when such face to face meetings can next be scheduled. These meetings are historically supported by FAO and OIE and discussions with FAO Bangkok had tentatively planned that an additional session on POCTs would be incorporated into the next workshop so that POCT manufacturers could be invited to attend that session.

ACDP is a partner in the OIE Collaborating Centre for Diagnostic Test Validation and the OIE have requested ACDP to prepare an article for a special edition of the OIE Scientific and Technical Review on “Test Validation Science”. An article for this special edition has been prepared and accepted for publication; entitled “Perspectives and Challenges in Validating New Diagnostic Technologies (41). The article contains a section on the specific challenges in validating POCTs and includes a POCT test validation flow chart (Figure 6) developed by Dr Emma Hobbs (as adapted from the OIE test validation flowchart). ACDP is also developing a proposal for an example-based validation template for POCTs which will be submitted to the Australian Subcommittee on Animal Health Standards (SCAHLS).

6.1 Regional workshop with key stakeholders for idea sharing and recommendations for technical guidance document

As described above this Regional workshop was originally planned for mid to late 2020 to be an additional workshop session coinciding with the next planned meeting of the FAO-OIE supported ASEAN Regional Laboratory Technical Advisory Group (LabTAG), which is then immediately followed by the ASEAN Laboratory Directors Forum. However, the impacts of COVID travel restrictions and the increasing health emergency in many of the countries in the region has meant that only a web based meeting of the ASEAN Laboratory Directors Forum will take place in March 2021, and the LabTAG meeting has been deferred. John Allen and Axel Colling will proceed with technically supporting and leading a discussion on POCTs at a future ASEAN Regional LabTAG workshop when the opportunity presents.

7 Conclusions and recommendations

7.1 Conclusions

POCTs have the potential to significantly disrupt traditional laboratory-based diagnostic pathways, especially in remote decentralised settings where sample referral networks and adequately equipped laboratories are particularly lacking. Diagnostic medical and veterinary POCTs are being used in LMICs and show clear benefits for disease diagnosis and surveillance, particularly when supported by policy makers. However, despite the abundance of POCTs on the commercial market and their clear benefits in remote and rural settings, the potential benefits of these tests in LMICs remain largely unrealised.

Inadequate regulatory guidance and poor industry oversight has led to a proliferation of POCTs of varying quality and fitness for purpose released onto the market, presenting challenges to potential end users who are, by design, expected to have limited diagnostic experience. Accurate, independent test validation data for commercial POCTs are often incomplete or absent. Even after robust initial validation testing, POCTs that demonstrate excellent diagnostic performance in the laboratory can show markedly lower accuracy under field conditions, for a variety of reasons. Similarly, incorporation of POCTs in diagnostic pathways and disease testing algorithms that are successful in one setting may not have similar uptake in others due to the varying and complex interplays between political, sociocultural and geographic factors, among others. Government veterinary services in LMICs should be aware of the costs, impacts, cost-effectiveness and operational feasibility of incorporating POCTs into diagnostic workflows in their specific country context and invest their resources accordingly.

This SRA sought to investigate the reasons for underutilisation of POCTs through a literature review, and questionnaires and interviews with key stakeholders and manufacturers of POCTs. The onset of the COVID 19 pandemic meant that a series of planned face to face structured interviews could not proceed. The questionnaire was then re-formatted to an on-line format with offer of follow-up interview by phone or video link. The growing COVID 19 pandemic no doubt contributed to a reduction in the response rate that would have been achieved with face to face interviews. Twelve of 38 manufacturers that were approached completed the interviews, and of the 48 key stakeholders that were approached, 18 responded and agreed to web based interviews.

Despite the lower than anticipated response rate it is clear from the literature review and the questionnaires and interviews that there is an increasing offering and usage of POCTs globally and specifically in the region. It is also clear that the range of diseases covered by manufactured POCTs will always be narrower and will lag in development compared with the range of published qPCR test methodologies. Accordingly, there are some important animal diseases that don't have current commercially available POCTs. Low purchasing power in LMICs means that these specific diagnostic gaps may continue as manufacturers of POCTs will focus on diseases and areas where they will get a return on their R&D and marketing investments.

There is an OIE registry for 'fit for purpose' diagnostic tests, including for POCTs, but this registry is heavily underutilised for various reasons. There are lengthy timelines for OIE to consider a manufacturers submission, to appoint experts to review the technical file and to approve its listing on the register. There is also a cost hurdle of around $4,500 for the OIE submission and review. The underutilisation of the OIE test registry and the absence of other central peer reviewed registries makes it difficult for users to select 'good' POCTs. Lack of regulations on manufacturers mean that POCTs of varying quality are allowed to be marketed and sold. There are also inconsistencies in the way POCTs are validated and lack of consistency in the information manufacturers provide or at times do not provide in the test kit technical data. Many users do not take into account these known, or unknown, limitations when using specific test kits, which can lead to inaccurate disease

diagnoses or surveillance results if samples are not backed up with confirmatory tests in a laboratory.

Users predominantly use POCTs for detection of field cases with putative symptoms (screening) where greater amount of the target analyte is usually present, whereas manufacturers are producing POCTs for a range of different purposes, including surveillance where larger numbers of kits are sold but where testing of asymptomatic cases is occurring. These different uses require different test performance thresholds in terms of DSe and DSp. Most users and manufacturers stated ideal diagnostic sensitivity values of 90% or greater, and of 95% or higher for diagnostic specificity. Users rate rapidity and suitability for outdoors the most important factors, whereas manufacturers think ease of use/minimal training and minimal equipment are of greatest importance.

7.2 Recommendations

1. To avoid a proliferation of POCTs of varying quality and fitness for purpose released onto the market, the international community, led by the OIE, should be urged to adopt regulatory frameworks for the standardised test validation data published by POCT manufactures in kit literature.

2. It is important that developers and users of diagnostic tests understand the principles and practices of test validation. This is even more important for manufacturers of POCTs where the intended user in the field is even more reliant on the manufactures claims regarding the test. It is recommended that the following POCT extended validation pathway is adopted by POCTs manufacturers when developing and validating new kits prior to their market release (Figure 6).

3. International agencies such as the FAO and OIE, and POCT manufacturers should coordinate and encourage standardised technical guidance and training for evaluating POCT characteristics based on manufacturer information and available literature, conducting in-house field validation and verification of diagnostic POCTS, and correctly interpreting and applying POCT results according to the relevant clinical setting and intended diagnostic purpose.

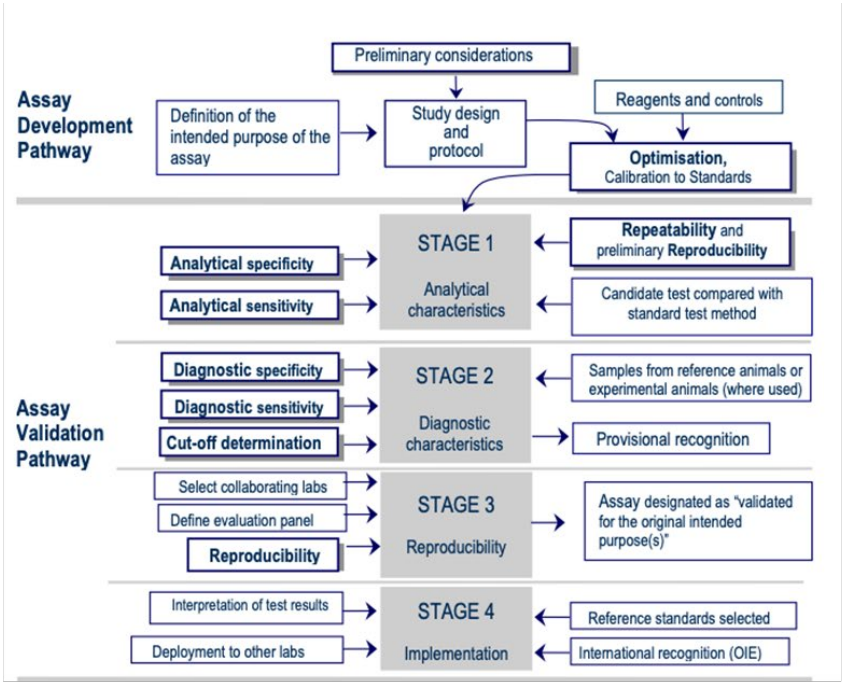
4. Sociological research and user needs analyses are required to understand the drivers of animal disease transmission in LMICs, to investigate local knowledge, attitudes and practices relating to animal disease diagnosis and surveillance, to identify barriers and facilitators to POCTs use in regional contexts and to develop strategies for implementing POCTs in the most impactful way to benefit animals, people, and communities.

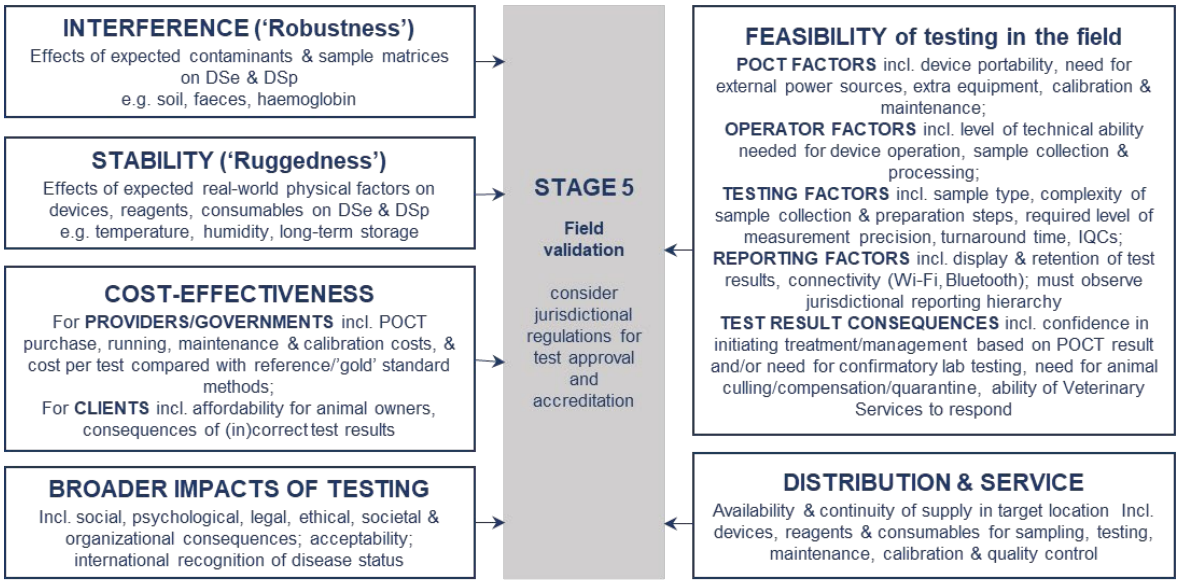
5. Even with reliable access to properly validated, fit for purpose POCTs, for diseases of high importance there will still be a need for confirmatory laboratory diagnosis and pathogen typing in LMICs for the foreseeable future. Therefore, support should also be maintained for initiatives that strengthen national veterinary and laboratory capacity, such as the Performance of Veterinary Services and laboratory twinning programs supported by the OIE, and conduct of harmonised laboratory technical assessments through the Laboratory Mapping Tool supported by the FAO.

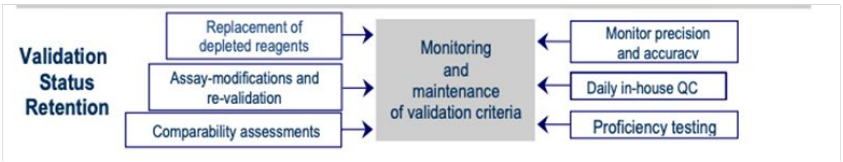
6. Advances in, and increasing accessibility to technology should also be utilised, e.g. using drones to assist with sample transport to laboratories from remote field locations (40). and dried tube (lyopholised) temperature stable reference samples for external quality assurance through proficiency testing of POCTs and laboratory

tests should be a regular practice wherever possible to ensure on-going test accuracy.

Figure 6. The extended point-of-care test development and validation pathway, with additional field validation stage, adapted from the World Organisation for Animal Health Validation Pathway (provided by Halpin et al. (41).







8 References

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8.2 List of acronyms

AI	avian influenza
AIV	avian influenza virus
ASF	African swine fever
ASe	analytical sensitivity
ASp	analytical specificity
BTV	blue tongue virus
CSF	classical swine fever
DSe	diagnostic sensitivity
DSp	diagnostic specificity
EHV1 & 4	equine herpes virus 1 and 4 `
EI	equine influenza
FAO	Food and Agriculture Organization of the United Nations
FMD	foot and mouth disease
IBD	infectious bursal disease
Incl.	including
IQC	internal quality control
LAMP	loop-mediated isothermal assay
LFA	lateral flow assay
LMIC	low- and middle-income country
MERS	Middle East respiratory syndrome
ND	Newcastle disease
OIE:	World Organisation for Animal Health
PED	porcine epidemic diarrhea
(ii)PCR	(insulated isothermal) polymerase chain reaction
POCT	point-of-care test
PPR	peste des petits ruminants
QC:	quality control
PRRS	porcine reproductive and respiratory syndrome
R&D	research and development
SCAHLS	Sub-Committee on Animal Health Laboratory Standards (Australia)
Swine SADS	swine severe acute diarrhoea syndrome coronavirus
TPP	target product profile
RPA	recombinase polymerase assay
USD	United States of America dollars