

# Report on the status of African swine fever (ASF) laboratory diagnostic capacities in South-East Asia, China, Papua New Guinea and Timor Leste



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The participants of the third ASF Coordination meeting on 25 August 2021 provided significant inputs during the plenary discussion on the status of the laboratory diagnostic capacity, challenges faced by the member countries, and recommendations to enhance ASF diagnostic capacity and strengthen ASF control in the region.

# Acronyms

ASF: African Swine Fever **ASFV: African Swine Fever Virus** ACDP: Australian Centre for Disease Preparedness **BSL:** Biosafety level CAHEC: China Animal Health Epidemiology Centre DNA: Deoxyribonucleic acid ELISA: Enzyme-linked immunosorbent assay HAD: Haemadsorption FAO RAP: Food and Agriculture Organisation Regional Office for Asia and the Pacific **OIE: World Organisation for Animal Health** FAT: Fluorescent antibody test **IBT:** Immunoblotting test IFAT: Indirect fluorescent antibody test IPT: Indirect immunoperoxidase test LAMP : Loop mediated isothermal Amplification (test) MCs: Member countries PCR : Polymerase chain reaction PNG: Papua New Guinea **RL: Reference Laboratory** SOP: Standard Operating Procedures SGE-ASF: Standing Group of Experts on ASF for Asia and the Pacific

# **Executive Summary**

The laboratory diagnosis of ASF is an important component of ASF prevention and control. ASF cannot be diagnosed based on clinical signs alone because of its similarity with other haemorrhagic diseases such as Classical Swine Fever (CSF), Porcine Reproductive and Respiratory Sysndrome (PRRS), erysipelas etc. The rapid and reliable detection of ASF is essential for the implementation of strict sanitary and biosecurity control measures to prevent the spread of the disease.

The <u>third ASF coordination meeting</u> held on 25 August 2021, focused on ASF diagnosis was organized to provide platform to share experiences and challenges in ASF diagnosis, assess laboratory capacity and discuss measures to enhance laboratory diagnostic capacity and coordinate laboratory activities to strengthen ASF control in the region. An opportunity was taken to conduct survey during the third ASF Coordination meeting with an aim to understand the status of laboratory capacities and key challenges faced in ASF diagnosis.

The questionnaire was developed based on the recommendations of the first meeting of the Regional Laboratory Expert on ASF and other pig diseases in Asia and the Pacific on 24 June 2021 and further improved during the preparatory meeting of the Third ASF Coordination meeting among the Experts from ASF Regional Reference Laboratories (Australian Centre for Disease Preparedness and China Animal Health and Epidemiology Centre) and staffs of FAORAP and OIE (SRRSEA& RRAP) on 10 August 2021. Thirteen member countries in South-East Asia plus China, Papua New Guinea and Timor Leste completed the online survey questionnaire.

The preliminary findings of the Questionnaire survey were presented during the Third ASF Coordination meeting. This report presents the detailed analysis of the questionnaire survey response and outputs of the plenary discussion during the third ASF coordination meeting. Laboratory diagnostic procedures for ASF fall into two groups: detection of the virus and serology to detect immune response. Among the available tests, Real-Time Polymerase chain reaction (PCR) is the most popular one which is being used in 12 countries followed by Conventional PCR, Antigen ELISA and Field tests. For the detection of antibody against ASF virus, only Antibody ELISA is widely used by the member countries. Only five countries reported at least one test accredited to ISO/IEC 17025. The molecular and serological tests are carried out in BSL2 facilities by most of the countries and those countries carrying out the virus isolation works are performed only in BSL3 facilities.

Various quality control measures implemented in the laboratories by the member countries are listed in Section 3.2.2. The measures implemented by the ASF infected countries in dealing with surge of samples due to increased ASF outbreaks and the preparation by ASF free country for possible increase of sample testing in case of an ASF incursion in their countries is provided in Section 3.3.1 and Section 3.3.2, respectively. Only seven out of 13 countries have shipped ASF samples to overseas OIE Reference Laboratories, out of which three ccountries reported that the referring of samples to the overseas laboratories took very long time. The technical support received from the Reference and Leading Laboratories by member countries are reported in Section 3.4.1. The expected support from OIE, Partners and Reference laboratories are provided in Section 3.4.2.

Member countries reported various challenges encountered in diagnosis of ASF in Section 3.3.4 during the questionnaire survey and in Section 3.5.1 during the plenary session of the Third ASF Coordination meeting. Some of the recommendations by the member countries to address these challenges and to enhance ASF diagnostic capacity are provided in Section 3.4.2 - Technical support expected from OIE/ FAO,

Partners and Reference Laboratories; Section 3.4.3 - Training support on the laboratory diagnosis of ASF and Section 3.4.4 - Other comments to improve ASF diagnosis. The recommendations from RL and Partners during the plenary discussion of the third ASF coordination meeting is provided in Section 3.5.2. The summary of individual country reports based on the questionnaire response are presented in Annex <u>1</u>.

Through this survey, we understood the existing laboratory diagnostic capacities of the member countries and challenges faced by the MCs in ASF diagnosis. Number of recommendations to enhance ASF diagnosis in the region were provided by the MCs, RLs and Partners. The OIE and FAO should closely liaise with Reference Laboratories to come up with an action plan to support member countries to enhance ASF diagnostic capacity of the members and to strengthen ASF control in the region.

# 1. Introduction

African swine fever (ASF) is an infectious disease of domestic and wild pigs of all breeds and ages, caused by ASF virus (ASFV) which is the sole member of the family *Asfarviridae*. ASF epidemiology is complex with different epidemiological patterns of infection occurring in Africa, Europe and Asia. ASF occurs through transmission cycles involving domestic pigs, wild boars, wild African suids, and soft ticks'. Following the introduction and further spread of ASF in Asia and the Pacific, there has been increasing concern over this disease in Asia and the Pacific. The ASF situation is continuously evolving in the region and has also been impacted by COVID-19 and associated restrictive measures. With over 60% of the world's domestic pig population in Asia the impact of this disease in the region cannot be underestimated.

The laboratory diagnosis of ASF is an important component of ASF prevention and control. The rapid and reliable detection of ASF is essential for the implementation of strict sanitary and biosecurity control measures to prevent the spread of the disease. Laboratory diagnosis of ASF involves identification of animals that are, or have previously been, infected with ASFV. An appropriate diagnosis therefore involves the detection and identification of ASFV-specific antigens, DNA or antibodies, to obtain relevant information to support control and eradication programmes.

The Standing Group of Experts on ASF for Asia Pacific (SGE-ASF Asia Pacific) identified several priority topics related to ASF. Laboratory diagnostic capacity development was one of the identified priorities. Much work has been done across the South-East Asia region to build capacity over the past years, however there is still a need to further enhance the laboratory and the field diagnostic capacity, harmonize laboratory diagnostic techniques, facilitate the sharing of information and coordinate laboratory activities amongst national laboratories, Reference Laboratories and laboratory experts to strengthen ASF control.

The <u>third ASF coordination meeting</u> focused on ASF diagnosis was organized to provide platform to share experiences and challenges in ASF diagnosis, assess laboratory capacity and discuss measures to enhance laboratory diagnostic capacity and coordinate laboratory activities to strengthen ASF control in the region. An opportunity was taken to conduct survey during the third ASF Coordination meeting with an aim to understand the status of laboratory capacities and key challenges faced in ASF diagnosis.

The preliminary findings of the Questionnaire survey to review the status of ASF laboratory diagnostic capacities in South-East Asia plus China, Papua New Guinea and Timor Leste were presented during the Third ASF Coordination meeting. This report presents the detailed analysis of the questionnaire survey response and outputs of the plenary discussion during the third ASF coordination meeting.

The objective of this cross sectional survey were:

- to understand the existing laboratory activities and diagnostic tests used by the countries for ASF diagnosis;
- to understand the challenges faced by countries in ASF diagnosis;
- to understand the areas of support required by the member countries to enhance ASF diagnosis from OIE Reference laboratories, OIE/FAO and other partners;
- to seek recommendations to enhance diagnostic capacity of the members and to strengthen ASF control in the region.

# 2. Study methodology

# 2.1 Questionnaire design and administration

The questionnaire was developed based on the recommendations of the first meeting of the Regional Laboratory Expert on ASF and other pig diseases in Asia and the Pacific on 24 June 2021 and further improved during the preparatory meeting of the Third ASF Coordination meeting among the Experts from ASF Regional Reference Laboratories (Australian Centre for Disease Preparedness and China Animal Health and Epidemiology Centre) and staff of FAORAP and OIE (SRRSEA& RRAP) on 10 August 2021.The questions were broadly grouped into types of tests currently used for ASF diagnosis by the member countries, available biosafety and quality control measures, key challenges faced, and technical support received by the member countries (MCs) from the Reference Laboratories (RL) and expected technical support from OIE, FAO and RLs.

The finalized questionnaire was than incorporated in Microsoft Form (MS Form) and the link to the questionnaire in MS Form was shared to the Member countries. Thirteen member countries (Brunei, Cambodia, China, Indonesia, Laos, Malaysia, Myanmar, Papua New Guinea, Philippines, Singapore, Timor Leste, Thailand and Vietnam), completed the online survey questionnaire. The relevant official involved in coordinating the ASF laboratory activities at the national level or laboratory expert involved in ASF diagnosis or the OIE National Focal Point for Veterinary Laboratories completed questionnaire virtually.

# 2.2 Plenary session:

The plenary discussion during the Third ASF Coordination meeting on 25 August 2021 focused on the Challenges in ASF diagnosis in member countries, initiatives to enhance laboratory diagnostic capacity and ideas to strengthen collaboration and coordination between ASF Reference and National Laboratories. In the first round of panel session three member countries (Malaysia, Philippines and Vietnam), provided insights on the challenges faced in ASF diagnosis. This was followed by some response and further insights by the Experts and participants from Reference Laboratories and partners.

# 2.3 Data Analysis

Descriptive analysis on the information received from 13 member countries were performed. We report the analysis of situation and status of ASF diagnostic capacity at the regional level. Response from individual countries are included as annex in this report.

# 3. Results

# 3.1 Diagnostic tests

# 3.1.1 Diagnostic tests for agent identification/ detection of virus

Laboratory diagnostic procedures for ASF fall into two groups: detection of the virus and serology. For detection of virus, around seven types of tests are currently being used by the Member countries. Among the available tests, Real-Time Polymerase chain reaction (PCR) is the most popular, which is being used in 12 countries, followed by Conventional PCR, Antigen ELISA and Field tests (Figure 1). Virus sequencing is performed by five countries and only four countries carry out virus isolation.

While ranking tests based on usage and frequency, Real-time PCR ranked number one followed by Conventional PCR (Figure 2).



Figure 1: Types of diagnostic tests used by the member countries for the detection of virus.



Figure 2 : Ranking of tests as per usage and frequency of use by the member countries.

Of the five countries using Antigen ELISA test, four countries reported using Commercial ELISA (INgezim and Ingenesa), and one country is using in-house ELISA. One country reported using Loop mediated isothermal Amplifications test (LAMP) test for detecting ASFV DNA. Of the five countries using sequencing, the following combinations of platforms and technologies are used:

- one country is using Sanger sequencing and Next generation sequencing (Illumina)
- one country is using Sanger and next generation sequencing (Illumina, Nanopore)one country is using first generation high-throughput sequencing)
- one country is using Sanger sequencing and next generation sequencing (platform not specified)
- one country is using next generation sequencing (platform not specified).

Of the six countries using field tests, the type of device/ platform used are: (i) Lateral Flow Device (Rapid Test Kits) for both ASF antigen and antibody detection, including the Ingenasa INgezim ASFV CROM antigen rapid test; (ii) Portable PCR, including POCKIT Insulated Isothermal PCR and TACO mini automatic nucleic acid extraction system, Genereach PCR, and Shweitzer Biotech Co. (SBC) convective PCR (cPCR); (iii) Isothermal assays, including LAMP and recombinase polymerase amplification);

Numbers of real-time PCR tests conducted by the national laboratories varies to a great extent, such as 100 tests a year, 3 to 5 test per day, to 100 - 200 tests per day, to 500 tests per day.

Six member countries extract DNA from field samples manually, five countries extract using automatic nucleic acid extraction machine and two countries extract both manually and by using automatic extraction machine.

# 3.1.2 Diagnostic tests for detection of antibody

Although five diagnostic tests are used for detection of antibody against ASF virus, only Antibody ELISA is widely used by 11 member countries (Figure 3) and also ranked number 1 in terms of its usage and frequency of its use (Figure 4). All the countries reported using commercial ELISA kits (2 countries – ID.Vet and Ingenasa, 4 countries – ID.Vet, 2 countries – Ingenasa, 1 country – Qingdao Lijian Bio Tech., Co. Ltd and two countries did not mention the manufacturer).



Figure 3 : Types of diagnostic tests used by the member countries for the detection of antibodies.





# 3.1.3 Laboratory tests that are accredited to ISO/IEC 17025

Five countries reported at least one test accredited to ISO/IEC 17025 (Figure 5). Real-time PCR tests were accredited to ISO/IEC 17025 by four countries, antibody ELISA by three countries and conventional PCR by one country.



Figure 5 : Number of countries with at least one ASF diagnostic test accredited to ISO/IEC 17025.

# 3.2 Biosafety and Quality control

# 3.2.1 Biocontainment classification of the laboratory to perform various tests

Table 1 shows the biocontainment classification of laboratories of the member countries at which routine diagnosis of ASF is performed. Seven countries reported carrying out molecular and serological tests in BSL2 facilities, while two countries reported conducting these tests in BSL2 plus facilities. Two countries reported conducting serological tests in Class II Biosafety Cabinet. Only five countries reported carrying out virus isolation work in BSL3 facilities.

Table 1: Biocontainment classification of laboratory to perform routine diagnosis of ASF by the member countries.

Test type	Biocontainment classification	Number of countries
Molecular tests	BSL2	7
Virus isolation	BSL3	5
Serological tests	BSL2	7
Molecular/ Serological tests	BSL2 plus	2
Serological tests	Class II Biosafety Cabinet	2

# 3.2.2 Implementation of quality control in the laboratory

The measures implemented by the member countries for the quality control in the laboratories is presented in Figure 6. Some of the key quality control measures are:

- Accreditation to ISO/IEC17025 or ISO:9001 and following its requirement;
- Internal control system which includes right from sampling, transport, storage to testing in the laboratories;
- Positive/negative extraction control and positive/negative reaction control for Real-time PCR; Inter-laboratory comparison (National Lab and Regional Labs);
- Proficiency test (National Lab and OIE reference laboratory);
- Regular maintenance of laboratory equipment through annual maintenance contract or as per the schedule required for the maintenance of specific machines;
- For the laboratory tests member countries refer OIE Terrestrial manual or SOPs for the specific tests;
- Implement intermediate checks for reagents and calibration of weighing scale and pipettes;
- Upgrade competency of laboratory staff by attending the technical course (such as specific task for virus isolation, handling suspected case of ASF);
- Using reference materials from the reference laboratories such as positive and negative control sera.





#### Figure 6: Types of quality control measures implemented by the member countries in the ASF laboratories.

# 3.3 Logistics and Challenges

# 3.3.1 Dealing with surge of sample due to increased ASF outbreaks

Member countries adopted following measures to deal with surge of samples due to increase number of outbreaks in their countries.

- Develop capacity of the provincial/ regional laboratories in performing and setting up the PCR laboratories through training;
- Develop capacity to test ASF samples with real-time PCR as this method can accommodate many samples at one time;
- Set up mobile laboratories with portable qPCR at the point of outbreak. Test only those samples that meet the case definition for suspected ASF;
- Accreditation of private and government laboratories to perform ASFV testing;
- Pooling of the samples that come from the same house/ shed;
- The first outbreak in each province was confirmed by the national reference laboratory, the subsequent outbreaks were confirmed by the provincial animal disease prevention and control center;
- Increased the number of human resources for laboratory work, implement shift system by rotating the staff (day and night shift) and overtime payment system;
- Implement inter laboratory partnership in case of shortage of kits or reagents with other ASF diagnostic laboratories;
- Intervention with modified reporting and testing procedures by the dedicated trained personnel;
- Government proclamation of a State of Emergency which will lead to budget allocation for ASF testing needs of the national and regional laboratories;
- Technical support including provision of diagnostics kits and consumables from FAO and US DTRA.

# 3.3.2 Preparation for testing of samples in case of ASF incursion in a ASF free countries

- Prepare Laboratory Contingency/ Preparedness Plan which should include sampling, transporting and testing of ASF suspected samples;
- Arrange logistics for transport of samples within country and its referral outside the country;
- Ensure availability of extraction kit, consumables, reagents, positive controls, PCR kits in case of surge of sample including availability of well-functioning equipment;
- Strengthen the laboratory diagnostic capacity for detection of ASF and emerging diseases including field tests;
- Source fund either through additional allocation using operational budget or special allocation using emergency budget;
- Participate in the ASF laboratory proficiency testing program either with Reference or Leading laboratories;
- Develop laboratory network partnership (authorised private or university laboratories) to deal with the surge of samples.

# 3.3.3 Experience in shipping samples to overseas OIE Reference Laboratories for ASF diagnosis

Seven countries reported successful experience in shipping samples to overseas OIE Reference Laboratories. Out of this, three countries reported that the referring of samples to the overseas laboratories took very long time. Six countries have not referred any samples to the overseas laboratories.



# Figure 7: Member countries experience in shipping samples to overseas OIE Reference Laboratories for ASF diagnosis

# 3.3.4 Main Challenges in the diagnosis of ASF and other pig diseases

The main challenges faced by the member countries in the diagnosis of ASF and other pig diseases is shown in Figure 8. The availability of reference materials (9 countries), availability of diagnostic kits and consumables (8 countries), lack of funds (8 countries), lack of human resources (6 countries) and lack of technical capacity (6 countries) are the most common challenges reported by the member countries.



Figure 8: Challenges faced by the member countries in the diagnosis of ASF and other pig diseases.

# 3.4 Technical support

# 3.4.1 Support received by Member Countries from Reference and Leading Laboratories

Twelve countries reported receiving technical support from the Reference and Leading Laboratories. The areas of support received by the member countries is presented in Figure 9. Nine countries reported receiving support on proficiency testing program, seven countries received training on ASF diagnosis and five countries received reference materials. Other support received are provision of SOPs, supply of laboratory consumables and exchange programmes among the laboratories.



Figure 9: Types of technical support received by member countries from the Reference and leading laboratories.

# 3.4.2 Technical support expected by the Member Countries from OIE/ FAO, Partners and Reference Laboratories:

Eleven countries expected support on ASF laboratory diagnosis from OIE, FAO, partners and Reference Laboratories. The types of technical support expected by the member countries are presented in Figure 10.

Some specific support expected by the member countries are:

- Support Proficiency Testing Program for ASF PCR, ELISA, and LAMP;
- Provision of reference materials (positive and negative controls for PCR, DNA from various genotypes of ASFV, positive/negative control antisera for serology);
- Provision of SOPs for virus isolation/HAD, immunoblotting test (IBT), immunoperoxidase test (IPT), immune-fluorescence assay (IFA), preparation of IQC materials for PCR, preparation of PT panels, biobanking or sample banking, validation/verification of diagnostic tests, including assistance in development of relevant SOPs;

- Supply of laboratory consumables such as PCR and extraction kits and related consumables including advice on the procurement of diagnostic kits, reagents and lab consumables.



# Figure 10: Types of technical support expected by the MC from OIE, FAO, Partners and Reference Laboratories.

# 3.4.3 Training support on the laboratory diagnosis of ASF

Twelve member countries expect laboratory hands-on training and eight member countries expected to receive on-line training course on ASF diagnosis (Figure 11).

The following training topics were identified by the member countries. The proposed topics varied among the countries depending on the existing diagnostic capacity and their plans to enhance capacity.

- Specimen collection, packaging, transport and submission, including biosafety and biosecurity for the field veterinarian and animal health staff;
- Diagnostic tests:
  - Real-time and conventional PCR
  - Serological tests such as IBT, IPT and IFA;
  - Virus isolation/ HAD, cell culture, virus characterization, sequencing; bioinformatics, including phylogenetic analysis
  - o Troubleshooting
- Quality assurance and Bio risk management;
- In depth analysis and interpretation of PCR results, including setting threshold, baseline, use of Shewart Chart, setting CT value cut-offs, etc;
- Validation /verification of diagnostic tests; and
- Evaluating efficiency of PCR/isothermal PCR assays.



Figure 11 : Types of laboratory training expected by the MCs.

# 3.4.4 Other comments to improve ASF diagnosis

Member countries provided the following additional comments to improve ASF diagnosis:

- Strengthening validation of onsite field tests under COVID-19 restrictions;
- Policy and legislation for mandatory reporting of suspected ASF by farmers and animal health professionals;
- Build and equip new PC2 laboratory facility to accommodate the storage, testing and disposal of porcine specimen waste and lab generated waste in accordance with minimum international standards;
- Technical support (such as continuous training either online or hands on, sharing of tests methods) and procurement of equipment such as thermocycler PCR machine, automatic nucleic acid extraction machine, refrigerated centrifuge, deep freezer, shaker etc;
- Early detection through disease recognition in the field, collection of appropriate samples and use of appropriate and validated diagnostic test/kits are needed to improve diagnosis. Training the field personnel and field vets on the disease recognition and correct interpretation of lab results will also help improve diagnosis;
- Improve the ease of use of ASF diagnosis;
- Reduce the cost of ASF diagnosis;
- Explore development of DIVA (Differentiating Infected from Vaccinated Animals) tests for ASF since ASF vaccines development is in advance stage.

# 3.5 Outputs of the Plenary Discussion

# 3.5.1 Challenges and issues faced by the member countries in ASF diagnosis

Three countries (Malaysia, Philippines and Vietnam) participated as Panellists in the round table discussions and provided highlights of the challenges faced during the initial incursion of ASF, at the time of COVID restrictions and in the era of new developments of ASF vaccine. Main challenges are summarized below:

- Difficulties in collecting and transporting specimen to laboratories in time due to movement restrictions;
- Difficulties in proper storage of samples;
- Unavailability of enough PCR machines hindered timely diagnosis and sustainability of reagents;
- Lack of skills in advance laboratory diagnosis such as virus isolation and sequencing;
- Low sensitivity and specificity of the currently available field tests;
- Since the development of ASF vaccines are making good progress, need of a diagnostic test to differentiate the vaccine strain and ASF virus field strain;
- Concerns of risks of environmental contamination through use of live attenuated vaccines were raised.

# 3.5.2 Recommendations from the Reference Laboratories and Partners

The ACDP proposed means to strengthen collaboration and support from OIE Reference Laboratories to national laboratories which could be done by way of inclusion of laboratories working on ASF diagnosis in networks with reference laboratories through which proficiency testing, provision of reagents, SOPs and trainings could be possible.

CAHEC further added that support could now be provided virtually to demonstrate most laboratory procedures. They advised members to upgrade laboratories to the ISO 17075 level for assured quality control, for which support can be provided also.

Additionally, MCs were advised to be prepared for the vaccine usage in individual countries. MCs should be aware of the type of vaccine candidates used and prepare for establishing tests for differentiation of vaccine and field strains. Apart from that, importance of sharing of epidemiological information at the national and regional levels were stressed upon.

Partners like FAO added that most training was moved to a virtual platform due to the sanitary situation, but the preparations were underway to provide hands-on trainings once the sanitary situation improves. Direct support to targeted countries in terms of providing reagents through available projects will also be implemented and they are considering to also providing validated field test kits for early diagnosis, especially in remote areas where access to specialized laboratory tests is difficult.

Deliberations were also made on how OIE and ACDP can expand on the training materials from the laboratory twinning project, to be made available for all laboratories in the region (regional e-learning platform), and for OIE to contemplate on creating a platform for repository of SOPs for sharing in the region.

# 4. Discussion and Conclusion

In the absence of approved vaccines, the rapid and reliable detection of ASF is essential to implement ASF control measures. ASF cannot be diagnosed based on clinical signs alone because of its similarity with other haemorrhagic diseases such as Classical Swine Fever (CSF), Porcine Reproductive and Respiratory Syndrome (PRRS), erysipelas etc. Laboratory testing is therefore essential, using virus or antibody detection methods. In accordance with Chapter 3.9. of the OIE Terrestrial Manual, the recommended laboratory tests for agent identification are conventional PCR, Real-time PCR, Antigen ELISA and FAT. Of these, real-time PCR test is the most popular one and is used by 12 member countries. This may be due to the high sensitivity and specificity of the real-time PCR test, convenience in testing high numbers of samples at a time and shorter time it takes to compete the tests. Besides the OIE recommended tests, the field tests such as lateral flow devices were used by six countries as the screening tests when ASF is suspected. The OIE recommended tests for laboratory antibody detection are ELISA, IPT, IFAT and IBT. The commonly used tests for antibody detection is ELISA, which is used by 11 member countries. ELISA tests are commercially available. Other tests recommended by the OIE are being used by only few countries due to the comparatively higher costs and time it takes to complete the tests and obtain the results. Moreover, the use of antibody detection tests are limited to the back yard farms or in wild pigs as the pigs in the commercial farms are usually culled once the ASF is detected. The selection of different types of tests for virus detection and antibody detection in different scenarios is being provided by the OIE Refence Laboratories. National laboratories should refer and apply the laboratory diagnostic algorithm for ASF prepared by the ASF RL.

Only five countries reported accrediting the test to ISO/IEC 17025. This includes real-time and conventional PCR and ELISA. Accreditation of tests to ISO/IEC 17025 standards enhances the reliability of test results generated by the laboratories. Therefore, member countries should work towards accrediting their tests and laboratories by the relevant laboratory accreditation bodies.

Although ASF virus do not have a human health risk, it is highly infectious to the pig population and any mishandling of samples contaminated with ASF virus will potentially lead to further spread of ASF amongst the pig population. Member countries also raised concerns on the use of live attenuated ASF vaccines by some countries, which could lead to environmental contamination and subsequent infection of the pig population. Therefore, ASFV should be handled with an appropriate level of bio-containment, determined by risk analysis in accordance with <u>Chapter 1.1.4</u> Biosafety and biosecurity: Standard for managing biological risk in the veterinary laboratory and animal facilities, Manual of Diagnostic Tests and Vaccines for Terrestrial Animals 2021.

Various quality control measures implemented in the laboratories by the member countries are listed in Section <u>3.2.2.</u> The measures implemented by the ASF infected countries in dealing with surge of samples due to increased ASF outbreaks and the preparation by ASF free country for possible increase of sample testing in case of an ASF incursion in their countries is provided in Section <u>3.3.1</u> and Section <u>3.3.2</u>, respectively. Member countries can learn from each other's experiences and adapt the best practices to enhance the quality control in the laboratories, to deal with surge of samples due to increased ASF outbreaks in infected countries and due to incursion of ASF in the free countries.

Only seven out of 13 countries have shipped ASF samples to overseas OIE Reference Laboratories, out of which three ccountries reported that the referring of samples to the overseas laboratories took very

long time. Since the referring of samples is essential to characterize and understand the strain of circulating ASF virus, the capacity of the national laboratories to refer the samples to the Reference Laboratories should be developed.

The technical support received from the Reference and Leading Laboratories by member countries are reported in Section <u>3.4.1</u>. The expected support from OIE, Partners and Reference laboratories are provided in Section <u>3.4.2</u>. Some members countries had been receiving technical support from RL such as training, proficiency testing, reference material, SOPs and supply of consumables (Figure 9). More member countries are expecting similar support in these areas (Figure 10), which could be enhanced through strengthening the collaboration and coordination between the RL and NL.

Member countries reported various challenges encountered in diagnosis of ASF in Section <u>3.3.4</u> during the questionnaire survey and in Section <u>3.5.1</u> during the plenary session of the Third ASF Coordination meeting. Some of the recommendations by the member countries to address these challenges and to enhance ASF diagnostic capacity are provided in Sections <u>3.4.2</u>; <u>3.4.3</u>; <u>3.4.4</u>; and <u>3.5.2</u> of this report.

Key recommendations to the member countries, Reference Laboratories and Partners to enhance ASF diagnostic capacities and ASF control in the region are:

# **Recommendations to the Member countries**

- National laboratories should refer and apply the <u>laboratory diagnostic algorithm</u> for ASF prepared by the ASF RL.
- Since it is important to characterize and understand the strain of circulating ASF virus, national laboratories should develop capacities to refer the samples to the Reference Laboratories.
- Member countries to upgrade laboratories to the ISO 17075 level quality assurance, and enhance the reliability of test results generated by the laboratories.
- Since the development of ASF vaccines are under progress, MCs should be prepared for vaccine usage in their respective countries, aware of the type of vaccine candidates used and prepare for establishing tests for differentiation of vaccine and field strains.
- Develop measures to implement systematic quality control system in the laboratory and to deal with surge of samples during the ASF outbreaks.

#### **Recommendations to OIE, FAO and ASF Reference Laboratories**

- Inclusion of national laboratories working on ASF diagnosis in networks with reference laboratories through which the training and technical support could be facilitated.
- Provide hands-on or online training on ASF diagnosis to the National Laboratories. Priority topics identified and proposed by member countries is in Section <u>3.4.3</u> of this report.
- Provide technical support to the national laboratories including Proficiency testing, provision of reference materials, SOPs and laboratory consumables (Section <u>3.4.2</u>).
- FAO and other Partners to provide direct support to targeted countries in terms of providing reagents through available projects and also providing validated field test kits for early diagnosis, especially in remote areas where access to specialized laboratory tests is difficult.

- Since the development of ASF vaccines are making good progress, there is a need for diagnostic tests and SOPs to differentiate vaccine and field strains;
- Expand on the training materials from the laboratory twinning project (Philippines and Pirbright), to be made available for all laboratories in the region (regional e-learning platform); and
- The OIE and FAO should closely liaise with Reference Laboratories to come up with an action plan to support member countries to enhance laboratory diagnostic capacity of the members and to strengthen ASF control in the region.
- OIE to organise follow-up meeting on ASF diagnosis at the appropriate time in consultation with ASF RL, development partners and MCs.

-

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# Annexes

# Annex 1: Country Reports – response to questionnaire

Brunei Darussalam

# **BACKGROUND DETAILS**

Department: Department of Agriculture and Agrifood

Name of the National Laboratory: Veterinary Laboratory Services

Contact Person: Hamsiah Mohd Saat

**Position: Section Head** 

Email: hamsiah.saat@agriculture.gov.bn

# Laboratories involved in diagnosis of ASF at the national and provincial level:

Veterinary Laboratory Services under Department of Agriculture and Agrifood. Currently, unable to do diagnosis of ASF

# National Laboratory accredited to Biosafety level 3 (BSL-3): No

# **DIAGNOSTIC TESTS**

# Diagnostic tests for agent identification/ detection of virus

Unable to diagnose ASF diagnosis. For other diseases:

- Conventional PCR
- Real-time PCR
- Antigen ELISA (Indicate in-house or commercial/ brand)

Brunei is free from ASF with no pig industries. Although tests are available, Brunei reported not having a diagnostic capabilities for ASF.

# Extraction of DNA from field samples

- Extract manually
- Extract using automatic nucleic acid extraction machine

# Diagnostic tests for (ASF serological tests) detection of immune response

- Antibody ELISA (Indicate inhouse or commercial/ brand)

# **BIOSAFETY AND QUALITY CONTROL**

#### Biocontainment classification of the laboratory to perform routine diagnosis for ASF

Molecular	BSL2
Virus isolation/FAT/Ag ELISA	BSL2
Serology	BSL2

# Quality control in the Laboratories

- Follow ISO/IEC17025 requirement.

# LOGISTICS AND CHALLENGES

# Preparation for possible increase of sample testing in case of an ASF incursion

- We are in the process of developing test procedure for ASF diagnosis.
- Importation of pork is only allowed from ASF free countries.

# Experience in shipping samples to overseas OIE Reference Laboratories for ASF

- No

# Main challenges in the diagnosis of ASF and other pig diseases

- Availability of reference materials (antisera against different strains)
- Lack of technical capacity

# **TECHNICAL SUPPORT**

# Technical support/ training received from Reference Laboratories/ Leading Laboratories

- No

# Expected technical support from OIE/ FAO/ Partners and Reference Laboratories

- Provision of reference materials
- Procurement of diagnostic kits, reagents and lab consumables
- SOPs

In addition to technical support for ASF diagnosis, Brunei expect support for other diseases such as: PT for Rabies and PPR; Reference materials for Rabies and PPR; and SOPs/ recommended test for those diseases not present in Brunei.

# Expected training on laboratory diagnosis of ASF from OIE/ FAO partners and Reference Laboratories

Currently, Brunei is only confident to carry out the molecular diagnosis of H5N1. They will highly benefit from the training of their laboratory staff on ASF diagnosis through laboratory hands-on training and training course.

# Cambodia

# **BACKGROUND DETAILS**

Department: General Directorate of Animal Health and Production (GDAHP).

Name of the National Laboratory: National Animal Health and Production Research Institute (NAHPRI).

Contact Person: Dr Sothrya Tum

**Position: Director** 

Email: sothryatum@gmail.com

#### Laboratories involved in diagnosis of ASF at the national and provincial level

- National Animal Health and Production Research Institute (NAHPRI)

National Laboratory is accredited to Biosafety level 3 (BSL-3): No

#### **DIAGNOSTIC TESTS**

#### Diagnostic tests for agent identification/ detection of virus

- Real-time PCR
- Field tests

Real time PCR test is the most popular and commonly used tests followed by field test.

Approximate number of ASFV Real-time PCR performed per day: 0 – 25.

# Extraction of DNA from field samples (for other diseases): Extract manually.

# Diagnostic tests for (ASF serological tests) detection of immune response

- Antibody ELISA - Commercial test kit

Laboratory tests that are accredited to ISO/IEC 17025: None.

#### **BIOSAFETY AND QUALITY CONTROL**

#### Biocontainment classification of the laboratory to perform routine diagnosis of ASF

Molecular	-
Virus isolation/FAT/Ag ELISA	-
Serology	-

#### Quality control (QC) implemented in Laboratories

- Follow SOPs

#### LOGISTICS AND CHALLENGES

#### Dealing with surge of samples due to increased ASF outbreaks

Following measures were undertaken to deal with the surge of samples due to increasing ASF outbreaks.

- Overtime work.

# Experience in shipping samples to overseas OIE Reference Laboratories for ASF

- No

# Main challenges in the diagnosis of ASF and other pig diseases

- Lack of fund
- Availability of diagnostic kits and consumables

# **TECHNICAL SUPPORT**

# Laboratory technical support/ training received from Reference Laboratories

Cambodia received technical support and training from the OIE Reference Laboratories and Partners which include:

- FAO proficiency testing program; FAO Lab-Epi Exchange Program between Cambodia and Vietnam.

# Technical support expected from OIE/ FAO partners and Reference Laboratories on diagnosis of ASF and other pig diseases

- Proficiency testing (PT) Routine PT programme for regional TADs and emerging diseases.
- Provision of reference material

# Expected training on laboratory diagnosis of ASF from OIE/ FAO/ Partners and Reference Laboratories

- Philippines expect support on the training of their staff through laboratory hands-on training or via on-line training course.

# Other comments to improve ASF diagnosis

- Reagents support.

# China

### **BACKGROUND DETAILS**

Department: China Animal Health and Epidemiology Centre (CAHEC), Ministry of Agriculture and Rural Affairs (MARA).

Name of the National Laboratory: National ASF Reference Laboratory.

Contact Person: Dr Wu Xiaodong.

**Position: Director** 

Email: wuxiaodong@cahec.ch

#### Laboratories involved in diagnosis of ASF at the national and provincial level:

- National ASF reference laboratory
- Provincial Animal Disease Prevention and Control Centers

# National Laboratory is accredited to Biosafety level 3 (BSL-3): Yes

#### **DIAGNOSTIC TESTS**

#### Diagnostic tests for agent identification/ detection of virus

- Real-time PCR
- Conventional PCR
- Virus isolation/ HAD test )Haemadsorption)
- Sequencing First generation sequencing, High-throughput sequencing.
- Antigen test in-house
- Fluorescent antibody test (FAT)
- Field test Real-time PCR, Multiple brands Loop-Mediated Isothermal Amplification Technique, Multiple brands Recombinase Aided Amplification Technique, Multiple brands.

Real-time PCR test is the most popular and common tests used followed conventional PCR, virus isolation, sequencing and Antigen ELISA.

Approximate number of ASFV Real-time PCR performed per day: About 100 tests per day.

**Extraction of DNA from field samples (for other diseases):** Extract using automatic nucleic acid extraction machine.

#### Diagnostic tests for (ASF serological tests) detection of immune response

- Antibody ELISA ID Screen<sup>®</sup> African Swine Fever Indirect (ID-VET) and INgezim PPA COMPAC (EUROFINS)
- Immunoblotting test (IBT)
- Immunoperoxidase test (IPT)
- Indirect Fluorescent antibody test (IFAT)

- Lateral Flow device (antibody)

Antibody ELISA is the most common test used followed by IFAT, LFD, IBT and IPT.

Laboratory tests that are accredited to ISO/IEC 17025: Real-time PCR and Antibody ELISA.

# **BIOSAFETY AND QUALITY CONTROL**

#### Biocontainment classification of the laboratory to perform routine diagnosis of ASF

Molecular tests/ handling of suspected	BSL3
samples	
Virus isolation	BSL3
Serology	BSL2

# Quality control (QC) implemented in Laboratories

QC is implemented in the laboratory by:

- Follow ISO/IEC 17025

#### LOGISTICS AND CHALLENGES

#### Dealing with surge of samples due to increased ASF outbreaks

- The first outbreak in each province was confirmed by the National ReferenceLaboratory, the subsequent outbreaks were confirmed by the Provincial Animal Disease Prevention and Control Center.

# Experience in shipping samples to overseas OIE Reference Laboratories for ASF

- No

# Main challenges in the diagnosis of ASF and other pig diseases

- Availability of reference materials (antisera against different ASF strains)

# **TECHNICAL SUPPORT**

#### Laboratory technical support/ training received from Reference Laboratories

China received technical support and training from the Reference Laboratories which include:

- Laboratory training: EU ASF Reference Laboratory CISA-INIA, Madrid, Spain;
- 2010 laboratory training: OIE Reference laboratory for ASF, Madrid, Spain;
- 2015 reference materials: EU ASF Reference Laboratory CISA-INIA, Madrid, Spain, 2015
- Laboratory training; Proficiency testing; Reference materials; Diagnostic reagents and lab consumables; and SOPs.

# Technical support expected from OIE/ FAO partners and Reference Laboratories

- Provision of reference materials: Nucleic acids of various genotypes of ASFV.

# Expected training on laboratory diagnosis of ASF from OIE/ FAO/ Partners and Reference Laboratories

China expects support on the training of their staff through laboratory hands-on training and online training course.

# Other comments to improve ASF diagnosis

- Improve the ease of use of ASF diagnosis;
- Reduce the economic cost of ASF diagnosis.

# Indonesia

# **BACKGROUND DETAILS**

Department: Directorate General of Livestock and Animal health Services (DGLAHS).

Name of the National Laboratory: Animal Health Laboratory Network.

Contact Person: Dr Syafrison Idris

Position: Senior Veterinary Officer, Contact Person for OIE NFP for Veterinary Laboratory

Email: syafrison.idris@yahoo.com

#### Laboratories involved in diagnosis of ASF at the national and provincial level:

- Research Institute for Veterinary Science (BBLitvet);
- National Research Laboratory for research and for confirmation Disease Investigation Center, Medan;
- National Reference Laboratory for confirmation Disease Investigation Center Bukittinggi;
- Regional Laboratory for confirmation Disease Investigation Center Lampung;
- Regional Laboratory for confirmation Disease Investigation Center Subang;
- Regional Laboratory for provisional diagnosis and refer samples to national reference laboratory Disease Investigation Center Wates;
- Regional Laboratory for confirmation Disease Investigation Center Banjarbaru;
- Regional Laboratory for confirmation Disease Investigation Center Denpasar;
- Regional Laboratory for provisional diagnosis and refer samples to national reference laboratory Disease Investigation Center Maro;
- Regional Laboratory for provisional diagnosis and refer samples to national reference laboratory to National ASF reference laboratory.

#### National Laboratory is accredited to Biosafety level 3 (BSL-3): Yes

# **DIAGNOSTIC TESTS**

#### Diagnostic tests for agent identification/ detection of virus

- Real-time PCR
- Conventional PCR
- Virus isolation/ HAD test )Haemadsorption)
- Sequencing NGS
- Antigen test Commercial test. ID Vet
- Fluorescent antibody test (FAT)

Real-time PCR test is the most popular and common tests used followed by Antigen ELISA, conventional PCR, FAT, virus isolation and sequencing.

Approximate number of ASFV Real-time PCR performed per day: No information.

**Extraction of DNA from field samples (for other diseases):** Extract using automatic nucleic acid extraction machine.

# Diagnostic tests for (ASF serological tests) detection of immune response

- Antibody ELISA ID Screen<sup>®</sup> African Swine Fever Indirect (ID-VET) and INgezim PPA COMPAC (EUROFINS)
- Indirect Fluorescent antibody test (IFAT)

Antibody ELISA is the most common test used followed by IFAT.

Laboratory tests that are accredited to ISO/IEC 17025: RT-qPCR.

# **BIOSAFETY AND QUALITY CONTROL**

# Biocontainment classification of the laboratory to perform routine diagnosis of ASF

Molecular tests	BSL2
Virus isolation	BSL3
Serology	BSL2

# Quality control (QC) implemented in Laboratories

QC is implemented in the laboratory by:

- All laboratories involved in ASF diagnosis implemented ISO 17025 and accredited for range of test;
- Implemented harmonised national ASF diagnosis protocol;
- Interlaboratory comparative test.

# LOGISTICS AND CHALLENGES

# Dealing with surge of samples due to increased ASF outbreaks

- Involvement of Animal Health Laboratory Network.

# Experience in shipping samples to overseas OIE Reference Laboratories for ASF

- No

# Main challenges in the diagnosis of ASF and other pig diseases

- Lack of fund
- Availability of reference materials (antisera against different ASF strains)
- Poor reporting and submission of samples

# **TECHNICAL SUPPORT**

# Laboratory technical support/ training received from Reference Laboratories

Indonesia received technical support and training from the Reference Laboratories which include:

- Laboratory training, proficiency testing, reference material from Australia Animal Health Laboratory with support of FAO-RAP

# Technical support expected from OIE/ FAO partners and Reference Laboratories

- Proficiency test for RT-qPCR, support national reference laboratory to provide interlaboratory proficiency test according to ISO 17043
- Provision of reference materials
- Provision of reference materials
- SOPs

# Expected training on laboratory diagnosis of ASF from OIE/ FAO/ Partners and Reference Laboratories

Indonesia expects support on the training of their staff through laboratory hands-on training which include:

- RT-qPCR including its quality assurance and bio-risk management;
- Specimen collection and handling for frontliner veterinarian.

# Other comments to improve ASF diagnosis

- None.

# Lao PDR

#### **BACKGROUND DETAILS**

Department: Department of Livestock and Fisheries (DLF).

Name of the National Laboratory: National Animal Health Laboratory (NAHL).

Contact Person: Dr Chattouphone Keokhamphet

Position: Head of LIMS

Email: keokhamphet.w@hotmail.com

#### Laboratories involved in diagnosis of ASF at the national and provincial level:

- In Laos, there is only NAHL that has the capacity for ASF testing. All ASF cases in the country are confirmed by this laboratory.
- Provincial labs work as stations for sample collection and sample storage.

#### National Laboratory is accredited to Biosafety level 3 (BSL-3): No

#### **DIAGNOSTIC TESTS**

#### Diagnostic tests for agent identification/ detection of virus

- Real-time PCR

Real-timePCR test is the only test used for ASF diagnosis in Laos.

**Approximate number of ASFV Real-time PCR performed per day:** About 50 samples for various diseases.

#### Extraction of DNA from field samples (for other diseases): Extract manually.

# Diagnostic tests for (ASF serological tests) detection of immune response

- Antibody ELISA - ID Vet

Laboratory tests that are accredited to ISO/IEC 17025: None.

#### **BIOSAFETY AND QUALITY CONTROL**

#### Biocontainment classification of the laboratory to perform routine diagnosis of ASF

Molecular tests/ handling of suspected	Safety Cabinets class 2B
samples	
Virus isolation	NA
Serology	Safety Cabinets class 2B

# Quality control (QC) implemented in Laboratories

QC is implemented in the laboratory by:

- Using reference controls, test record sheets and Proficiency Testing.

# LOGISTICS AND CHALLENGES

# Dealing with surge of samples due to increased ASF outbreaks

- Pool the samples in the case the samples come from the same house/ shed.

# Experience in shipping samples to overseas OIE Reference Laboratories for ASF

- Yes

# Main challenges in the diagnosis of ASF and other pig diseases

- Lack of human resources
- Lack of fund
- Availability of diagnostic kits, reagents and consumables

# **TECHNICAL SUPPORT**

# Laboratory technical support/ training received from Reference Laboratories

Loas received technical support and training from the Reference Laboratories which include:

- Laboratory training;
- PT panel;
- Technical Exchange Programme with AAHL (Australia) and NCVD (Vietnam)

# Technical support expected from OIE/ FAO partners and Reference Laboratories

- Proficiency testing Real-time PCR
- Provision of reference materials: Reference Control for tests.
- Provision of diagnostic kits, reagents and lab consumables RT PCR kit (Ag path), extraction kit (Qiagen), related consumables

# Expected training on laboratory diagnosis of ASF from OIE/ FAO/ Partners and Reference Laboratories

Lao PDR expects support on the training of their staff through laboratory hands-on training. The priority topics identified for the training are virus characterization and phylogenetic analysis.

# Other comments to improve ASF diagnosis

- None.

# Malaysia

### **BACKGROUND DETAILS**

Department: Department of Veterinary Services (DVS).

Name of the National Laboratory: Veterinary Research Institute (VRI).

Contact Person: Dr Faizah Hanim binti Mohd Saeid

Position: Veterinary Officer/ Director

Email: faizah@dvs.gov.my / faizah9764@gmail.com

#### Laboratories involved in diagnosis of ASF at the national and provincial level:

- Veterinary Research Institute, Ipoh (National Laboratory)
- Central Region Veterinary Laboratory.
- Southern Region Veterinary Laboratory.
- Northern Region Veterinary Laboratory.
- Sabah Veterinary Diagnostic Laboratory.
- Sarawak Veterinary Diagnostic Laboratory.

# National Laboratory is accredited to Biosafety level 3 (BSL-3): Yes

#### **DIAGNOSTIC TESTS**

#### Diagnostic tests for agent identification/ detection of virus

- Real-time PCR
- Conventional PCR

Real-time PCR test is the most common test used followed by the Conventional PCR.

Approximate number of ASFV Real-time PCR performed per day: About 60 tests.

Extraction of DNA from field samples (for other diseases): Extract manually.

#### Diagnostic tests for (ASF serological tests) detection of immune response

- None

Laboratory tests that are accredited to ISO/IEC 17025: None.

#### **BIOSAFETY AND QUALITY CONTROL**

#### Biocontainment classification of the laboratory to perform routine diagnosis of ASF

Molecular tests/ handling of suspected	BSL2
samples	
Virus isolation	NA
Serology	BSL2

# Quality control (QC) implemented in Laboratories

QC is implemented in the laboratory by:

- For machine and equipment, we are implementing consistent maintenance according to the schedule, either annually, every 2 years or once in 5 years.
- For test methods, we are referring to OIE Terrestrial Animal health Manual.
- Intermediate checks for reagents, weighing scale and pipettors.
- For personnel or lab technicians, we encourage them to upgrade their competency by attending the technical course (such as specific task for virus isolation, handling suspected case of ASF )

# LOGISTICS AND CHALLENGES

# Dealing with surge of samples due to increased ASF outbreaks

- As infected country, we use Real-time PCR as this method can accommodate many samples at one time;
- Implement rotation of work internally (own laboratory);
- Interlaboratory partnership in case of shortage of kit or reagent in other ASF diagnostic laboratories.

# Experience in shipping samples to overseas OIE Reference Laboratories for ASF

- No

### Main challenges in the diagnosis of ASF and other pig diseases

- Lack of human resources
- Lack of fund
- Availability of diagnostic kits, reagents and consumables
- Availability of reference materials
- Lack of technical capacity

# **TECHNICAL SUPPORT**

# Laboratory technical support/ training received from Reference Laboratories

Malaysia received technical support and training from the Reference Laboratories which include:

- OIE Webinar on ASF;
- Proficiency testing of ASF by Australian Centre for Disease Preparedness, annually. (February 2021).

# Technical support expected from OIE/ FAO partners and Reference Laboratories

- Proficiency testing for swine diseases (ASF, CSF, PRRS and Aujesky's disease)

# Expected training on laboratory diagnosis of ASF from OIE/ FAO/ Partners and Reference Laboratories

Malaysia expects support on the training of their staff through on-line training course. The priority topics identified for the training are molecular characterization of the virus, bioinformatics and sequencing.

# Other comments to improve ASF diagnosis

- Require technical support (such as continuous training either online or hands on, sharing of tests methods);
- procurement of equipment such as thermocycler PCR machine, refrigerated centrifuge, deep freezer, shaker.

# Myanmar

### **BACKGROUND DETAILS**

Department: Livestock Breeding and Veterinary Department (LBVD).

Name of the National Laboratory: Yangon Veterinary Diagnostic Laboratory.

Contact Person: Dr Win Myint

Position: Head/ Deputy Director

Email: wynnm1991@gmail.com

#### Laboratories involved in diagnosis of ASF at the national and provincial level:

- Yangon Veterinary Diagnostic Laboratory
- Mandalay Veterinary Diagnostic Laboratory

#### National Laboratory is accredited to Biosafety level 3 (BSL-3): No

#### **DIAGNOSTIC TESTS**

#### Diagnostic tests for agent identification/ detection of virus

- Real-time PCR
- Field test

Real-time PCR test is the most common test used followed by the field test.

Approximate number of ASFV Real-time PCR performed per day: No information.

Extraction of DNA from field samples (for other diseases): Extract manually.

#### Diagnostic tests for (ASF serological tests) detection of immune response

- Antibody ELISA – Commercial test kit (Id Vet)

Laboratory tests that are accredited to ISO/IEC 17025:2017 : None.

#### **BIOSAFETY AND QUALITY CONTROL**

#### Biocontainment classification of the laboratory to perform routine diagnosis of ASF

Molecular tests/ handling of suspected	BSL2
samples	
Virus isolation	NA
Serology	BSL2

# Quality control (QC) implemented in Laboratories

QC is implemented in the laboratory by:

- Follow ISO/IEC 17025 :2017

# LOGISTICS AND CHALLENGES

#### Dealing with surge of samples due to increased ASF outbreaks

- Use Real-time PCR as this method can accommodate many samples at a time;
- Implement rotation of work internally (own laboratory);
- Interlaboratory partnership in case of shortage of kit or reagent in other ASF diagnostic laboratories.

### Experience in shipping samples to overseas OIE Reference Laboratories for ASF

- Yes

#### Main challenges in the diagnosis of ASF and other pig diseases

- Lack of human resources
- Lack of fund
- Availability of diagnostic kits, reagents and consumables
- Lack of technical capacity
- Poor laboratory infrastructure/ limited space.
- Poor reporting and submission of samples

#### **TECHNICAL SUPPORT**

#### Laboratory technical support/ training received from Reference Laboratories

Myanmar received technical support and training from the Reference Laboratories which include:

- Laboratory training
- Proficiency testing
- Reference materials and
- SOPs supported by AAHL (Real time PCR)

# Technical support expected from OIE/ FAO partners and Reference Laboratories

- Proficiency testing Real-time PCR
- Provision of reference materials
- Procurement of diagnostic kits, reagents and lab consumables and
- SOPs (ELISA)

#### Expected training on laboratory diagnosis of ASF from OIE/ FAO/ Partners and Reference Laboratories

Myanmar expects support on the training of their staff through laboratory hands-on training and on-line training course. The priority topics identified for the training are on the use of PCR and sequencer.

#### Other comments to improve ASF diagnosis

- Need technical support;
- Need automatic nucleic acid extraction machine (MagMax automatic extraction machine)

# Papua New Guinea

# **BACKGROUND DETAILS**

Department: National Agriculture Quarantine and Inspection Authority (NAQIA)

Name of the National Laboratory: National Animal Health and Food testing laboratory (NAHTL).

Contact Person: Dr Orlando Mercado

Position: Animal Health Program Manager

Email: OMercado@naqia.gov.pg

# Laboratories involved in diagnosis of ASF at the national and provincial level:

- ASF laboratory testing done at the National Animal Health and Food Testing Laboratory only

National Laboratory is accredited to Biosafety level 3 (BSL-3): No

# **DIAGNOSTIC TESTS**

Diagnostic tests for agent identification/ detection of virus

- Real-time PCR
- Antigen ELISA Ingenesa/Ingezim commercial test ki
- Field test Lateral Flow Device (Rapid Test Kits) for ASF antigen detection.

Antigen ELISA is the most common test.

**Approximate number of ASFV Real-time PCR performed per day:** Capacity set-up is currently in progress with national collaborative partners.

Extraction of DNA from field samples (for other diseases): Extract manually.

# Diagnostic tests for (ASF serological tests) detection of immune response

- Antibody ELISA Ingenesa/ Ingezim commercial test kits.
- Lateral flow device (antibody)

Laboratory tests that are accredited to ISO/IEC 17025: None.

#### **BIOSAFETY AND QUALITY CONTROL**

#### Biocontainment classification of the laboratory to perform routine diagnosis of ASF

Molecular tests/ handling of suspected	PC1 laboratory set-up using Class II
samples	Biosafety Cabinets.
Virus isolation	NA
Serology	PC1 laboratory set-up using Class II
	Biosafety Cabinets.

# Quality control (QC) implemented in Laboratories

QC is implemented in the laboratory by:

- All porcine specimens are checked against corresponding lab submission forms which includes a Veterinary Specimen Advice Card and a Specimen Inventory List of the specimens being submitted.
- All porcine specimens are physically inspected individually to observe condition of specimens for suitability for testing
- Inconsistencies or non-compliance issues to do with porcine specimens and lab submission forms are communicated back to the consignor.
- All submissions are registered in the central laboratory accession system. Each consignment is given a unique lab accession number and all individual specimens are also given unique specimen ID numbers.
- All EDTA blood specimens and sera are stored at minus -20 degrees celcius.
- Commercial kit instructions are followed to set-up, run and record test results.
- ASF dedicated MS Excel Forms are used to document all ELISA testing.
- Internal Positive and Negative quality controls provided with the kits are used.
- All ELISA test results are vetted before reporting.
- Same samples are submitted to accredited regional reference lab (i.e. CSIRO ACDP) for ELISA, IFAT and PCR Testing
- Field and lab data from NAHFTL and CSIRO ACDP lab test reports are consolidated for reporting to the OCVO by dedicated personnel. Comparison studies were used to assess performance of ASF LFD/RTK versus ASF ELISA versus ASF PCR

#### LOGISTICS AND CHALLENGES

#### Dealing with surge of samples due to increased ASF outbreaks

- Intervention was required to ensure that there were dedicated trained lab personnel to receive, process, test and issue test reports.
- The traditional reporting system of NAHFTL had to change to accommodate for improved turnaround time and also consolidated reporting.
- Field data had to be incorporated to allow for traceability of specimens to sampling locations.

#### Experience in shipping samples to overseas OIE Reference Laboratories for ASF

- Yes, but it took very long time.

#### Main challenges in the diagnosis of ASF and other pig diseases

- Lack of human resources
- Lack of fund
- Availability of diagnostic kits, reagents and consumables
- Availability of reference materials (antisera against different strains)
- Lack of technical capacity

- Poor laboratory infrastructure/ limited space.
- Poor reporting and submission of samples

# **TECHNICAL SUPPORT**

#### Laboratory technical support/ training received from Reference Laboratories

PNG received technical support and training from the Reference Laboratories which include:

- Laboratory training through fellowships and other training methods including presentations, meetings, and lab exposure training in regional reference lab.
- Proficiency testing through Proficiency Testing Program offered by regional reference lab.
- Supply of reference materials and all relevant lab reagents.
- SOP drafting and finalization.
- Scientific and technical support and advice
- Laboratory equipment and consumables
- Facilitating relevant import permits for various diagnostic samples and intercepted pork products
- Linkages to experts and collaborative partners

#### Technical support expected from OIE/ FAO partners and Reference Laboratories

- Proficiency testing ASF ELISA and ASF PCR and tests for other diseases
- Provision of reference materials as a means to improve confidence in reporting lab test results and also meet ISO17025 requirements
- Advice and procurement of diagnostic kits, reagents and lab consumables. There should be recommendations on the brand or type of equipment and diagnostic kits and consumables needed based on independent performance assessment or validation studies.
- Assistance with developing relevant and appropriate SOPs
- Work experience training for confidence and competency development
- External assessments as a means to assess effectiveness of activity.

#### Expected training on laboratory diagnosis of ASF from OIE/ FAO/ Partners and Reference Laboratories

- PNG expects support on the training of their staff through laboratory hands-on training and mentorship through Fellowships. on-line training course.
- LFD/RTK, ELISA and PCR trouble shooting including reagents and equipment troubleshooting.
- Diagnostic training on PNG's priority target exotic and endemic animal diseases for both terrestrial and aquatic animals including disease antigen and antibody profile and mechanisms of these diseases. This would help lab personnel to determine the relevant and appropriate lab tests to be done as well as the necessary specimens required.
- Specialization of different lab disciplines and veterinary diagnostician/pathologist to allow for sound animal disease investigation.

#### Other comments to improve ASF diagnosis

- There is need of a dedicated PCR suite at NAHFTL.
- Need to build and equip a new and proper PC2 lab facility to accommodate the storage, testing and disposal of porcine specimen waste and lab generated waste in accordance with minimum international standards.
- This is to ensure we improve the containment of animal or zoonotic diseases to the environment or the public.

# **Philippines**

# **BACKGROUND DETAILS**

Department: Department of Agriculture – Bureau of Animal Industry (DA-BAI)

Name of the National Laboratory: Animal Disease Diagnosis and Reference Laboratory (ADDRL), Veterinary Laboratory Division (VLD), Bureau of Animal Industry (BAI).

Contact Person: Dr Edna A. Felipe/ Dr Rachel R. Azul.

Position: Office- In-Charge, VLD (Veterinarian IV) / Offficer-In-Charge, ADDRL (Veterinarian III)

Email: <a href="mailto:edua.felipe@bai.gov.ph">edua.felipe@bai.gov.ph</a> / <a href="mailto:rachel.azul@bai.gov.ph">rachel.azul@bai.gov.ph</a>

# Laboratories involved in diagnosis of ASF at the national and provincial level:

- National Reference Lab for confirmation is the ADDRL-VLD-BAI
- Regional Labs which provide provisional diagnosis on initial outbreak in the region and send samples for confirmation at National Lab. But once the labs are proficient in real time PCR diagnosis, they are allowed to test and release results through the National ASF Task Force : These 6 laboratories are the Regional Animal Disease Diagnostic Laboratories (RADDLs) located in Region 2 (RADDL2); Region 3 (RADDL3) ;Region 5 (RADDL5); Region 8 (RADDL8); Region 10 (RADDL10) and Region 12 (RADDL12).
- Regional Labs which provide rapid PCR tests (isothermal assays) for screening and/or send samples for confirmation to National or RADDLs nearest to them : There are 9 labs, namely RADDL CAR, RADDL1, RADDL 4A, RADDL 4B, RADDL 6, RADDL 7, RADDL 9, RADDL 11, RADDL 13

# National Laboratory accredited to Biosafety level 3 (BSL-3): No

# **DIAGNOSTIC TESTS**

# Diagnostic tests for agent identification/ detection of virus

- Real-time PCR
- Field test
  - Isothermal Assays : i) POCKIT Insulated Isothermal PCR (ii PCR) by Genereach, Taiwan (ii) Convective PCR (cPCR) by Shweitzer Biotech Co (SBC), Taiwan
  - Lateral Flow Device: INgezim ASFV CROM Ag
- Antigen ELISA INgezim PPA DAS (Eurofins-Ingenasa)

Real-time PCR test is the most popular and common tests followed by Field tests and Antigen ELISA.

The National Lab does not perform sequencing. They sent samples for sequencing to the OIE Reference Lab (The Pirbright Institute) during initial ASF outbreak in 2019.

**Approximate number of ASFV Real-time PCR performed per day:** 3-5 tests per day (1 test= One - 96 well PCR plate (with 43 pooled samples and 5 controls in duplicate). This is due to the increased environmental sample submission for repopulation purposes.

**Extraction of DNA from field samples (for other diseases):** Extract manually and using automatic nucleic acid extraction machine.

# Diagnostic tests for (ASF serological tests) detection of immune response

- Antibody ELISA INgezim PPA COMPAC (Eurofins-Ingenasa)
- Lateral flow device (antibody)

Laboratory tests that are accredited to ISO/IEC 17025: None.

# **BIOSAFETY AND QUALITY CONTROL**

#### Biocontainment classification of the laboratory to perform routine diagnosis of ASF

Molecular	BSL2
Virus isolation/FAT/Ag ELISA	BSL2
Serology	BSL2

# Quality control (QC) implemented in Laboratories

QC is implemented in the laboratory by:

- using real time PCR protocols based from OIE Terrestial Manual (we have adapted the ASF taqman PCR assay of CSIRO ACDP by King, et.al ,2003 and 1-step Taqman Real-time PCR assay of The Pirbright Institute, UPL/Fernandez-Pinero et al, 2013)
- using commercial Antigen and Antibody ELISA recommended by ASF reference Lab (INgezim-Eurofins/Ingenasa)
- Molecular and Serological tests are conducted in BSL2 lab
- inclusion of Extraction and PCR controls for every PCR run and these are monitored daily in a PCR progressive monitoring sheet
- annual calibration and PM of equipment used in conduct of PCR and serology
- annual participation in CSIRO ACDP Proficiency Test for ASF and other swine diseases vii) laboratory is ISO: 9001 accredited

# LOGISTICS AND CHALLENGES

# Dealing with surge of samples due to increased ASF outbreaks

Following measures were undertaken to deal with the surge of samples due to increasing ASF outbreaks.

- Building capacity of the different regional laboratories in performing and setting up the PCR laboratories through trainings;
- Government proclamation of a State of Emergency which led to budget allocation for ASF testing needs of the national and regional laboratories;
- Technical assistance-diagnostics kits and consummables from FAO and US DTRA;
- Accreditation of Private and government laboratories to perform ASFV testing.

#### Experience in shipping samples to overseas OIE Reference Laboratories for ASF

- Yes, but it took very long

# Main challenges in the diagnosis of ASF and other pig diseases

- Lack of human resources
- Lack of fund
- Availability of diagnostic kits and consumables
- Availability of reference materials
- Lack of technical capacity
- Poor laboratory infrastructure/ limited space
- Poor reporting and submission of samples
- IATA Accreditation on shipping dangerous goods
- Cost for setting up certain diagnosis of animal diseases is very expensive and frequency of using is almost zero, thus we are unable to develop diagnosis test for certain animal diseases.

# **TECHNICAL SUPPORT**

# Laboratory technical support/ training received from Reference Laboratories

Philippines received technical support and training from the OIE Reference Laboratories which include:

- FAO and OIE sponsored laboratory and online trainings;
- 2012 to present Proficiency testing from CSIRO AAHL/ACDP;
- 2012 to 2020 SOPS from CSIRO AAHL/ACDP thru trainings;
- 2012 to 2019 SOPS from The Pirbright Institute;
- 2019 to present Reference materials from CSIRO AAHL/ACDP c/o FAO, 2019-2020.

# Technical support expected from OIE/ FAO partners and Reference Laboratories on diagnosis of ASF and other pig diseases

- Proficiency testing ASF, CSF, PRRS, Swine Influenza
- Provision of reference material Positive and negative control antigen for PCR of ASF Differential diagnosis (CSF, PRRS, SI, PCV2) and reference positive/negative control antisera for serological confirmatory tests and cell lines for virus isolation of CSF, PRRS and ASF
- Diagnostic kits, reagents and lab consumables ASF IB strips, ASFV IPT plates, Protein A conjugate for Immunoblotting test and Immunoperoxidase test
- SOPs Virus isolation/HAD, IBT, IPT, IFA; Preparation of IQC materials for PCR , preparation of PT panels, BIO banking or sample banking; Validation/Verification of diagnostic tests
- Other (please specify)

# Expected training on laboratory diagnosis of ASF from OIE/ FAO/ Partners and Reference Laboratories

- Philippines expect support on the training of their laboratory staff through wither laboratory hands-on training or via on-line training course.

 The topics identified to be covered in the future training programme are: Virus isolation/HAD; Cell culture; Serological tests such as IBT, IPT and IFAT; QA and QC; In depth analysis and interpretation of PCR results (with real time samples from different labs)- i.e. setting threshold, baseline, use of Shewart Chart, setting CT value cut-offs, etc Validation /Verification of diagnostic tests efficiency of rapid PCRs/isothermal PCR assays.

### Other comments to improve ASF diagnosis

- Early detection through disease recognition in the field;
- collection of appropriate samples and use of appropriate and validated diagnostic test/kits to improve diagnosis;
- Training the laymen, field personnel and field vets on the disease recognition and correct interpretation of lab results will also help improve diagnosis.

# Singapore

#### **BACKGROUND DETAILS**

Department: National Parks Board

Name of the National Laboratory: Centre for Animal and Veterinary Sciences (CAVS).

Contact Person: Dr OCharlene Fernandez

**Position: Director** 

Email: Charlene FERNANDEZ@nparks.gov.sg

#### Laboratories involved in diagnosis of ASF at the national and provincial level:

- Centre for Animal and Veterinary Sciences (CAVS).

National Laboratory is accredited to Biosafety level 3 (BSL-3): No

#### **DIAGNOSTIC TESTS**

Diagnostic tests for agent identification/ detection of virus

- Real-time PCR
- Conventional PCR
- Sequencing Sanger, Illumina, Nanopore.

Real-time PCR the most common test used followed by Conventional PCR and Sequencing.

**Approximate number of ASFV Real-time PCR performed per day:** around 100 per year for ASF Real-time PCR.

**Extraction of DNA from field samples (for other diseases):** Extract using automatic nucleic acid extraction machine.

#### Diagnostic tests for (ASF serological tests) detection of immune response

- Antibody ELISA – Commercial test kit - ID Vet.

Laboratory tests that are accredited to ISO/IEC 17025: ASF Antibody ELISA

#### **BIOSAFETY AND QUALITY CONTROL**

#### Biocontainment classification of the laboratory to perform routine diagnosis of ASF

Molecular tests/ handling of suspected samples	BSL2
Virus isolation	NA
Serology	BSL2

# Quality control (QC) implemented in Laboratories

QC is implemented in the laboratory by:

- Participate in Proficiency Testing program with ACDP and APHA.

- Accreditation to ISO 17025:2017

### LOGISTICS AND CHALLENGES

#### Preparation for the possible increase of sample testing due to incursion of ASF

- Emergency Plan in place.

# **Experience in shipping samples to overseas OIE Reference Laboratories for ASF** - No.

# Main challenges in the diagnosis of ASF and other pig diseases

- Availability of reference materials (antisera against different strains)

# **TECHNICAL SUPPORT**

#### Laboratory technical support/ training received from Reference Laboratories

Singapore received technical support and training from the Reference Laboratories which include:

- Proficiency Testing (PT) ACDP (PCR), APHA (ELISA)- done annually (date varies each year)
- Test methods The Pirbright Institute- email communications in 2018 & 2019
- Laboratory training through fellowships and other training methods including presentations, meetings, and lab exposure training in regional reference lab.

### Technical support expected from OIE/ FAO partners and Reference Laboratories

- Proficiency testing ASFV PCR and ELISA
- Provision of reference materials Positive control materials for ASF PCR and ELISA
- SOP (Test method) Virus isolation

#### Expected training on laboratory diagnosis of ASF from OIE/ FAO/ Partners and Reference Laboratories

- Singapore expects support on the training of their staff through laboratory hands-on training and on-line training course.
- The priority topics identified for the training are molecular diagnostics including PCR and sequencing Virus isolation

#### Other comments to improve ASF diagnosis

- No comments

# Timor Leste

# **BACKGROUND DETAILS**

Department: Veterinary Diagnostic Laboratory

Name of the National Laboratory: Veterinary Diagnostic Laboratory

Contact Person: Dr Felisiano da Conceicao

Position: Chief of the Department

Email: maularavets@yahoo.com

#### Laboratories involved in diagnosis of ASF at the national and provincial level:

- ASF laboratory testing done at the Veterinary Diagnostic Laboratory only which performs field tests. For the confirmation, samples are referred to Australian Centre for Disease Preparedness (ACDP).

National Laboratory is accredited to Biosafety level 3 (BSL-3): No

#### **DIAGNOSTIC TESTS**

Diagnostic tests for agent identification/ detection of virus

- Only Field test using LAMP is used for ASF diagnosis. Samples referred to ACDP for confirmation.

Approximate number of ASFV Real-time PCR performed per day: Not applicable.

Extraction of DNA from field samples (for other diseases): Extract manually (LAMP).

#### Diagnostic tests for (ASF serological tests) detection of immune response

- They do not perform any testing for detection of immune response.

Laboratory tests that are accredited to ISO/IEC 17025: None.

# **BIOSAFETY AND QUALITY CONTROL**

#### Biocontainment classification of the laboratory to perform routine diagnosis of ASF

Molecular tests/ handling of suspected	NA
samples	
Virus isolation	NA
Serology	NA

# Quality control (QC) implemented in Laboratories

QC is implemented in the laboratory by:

- Positive and negative control

-

#### LOGISTICS AND CHALLENGES

Dealing with surge of samples due to increased ASF outbreaks

- Information not provided.

# Experience in shipping samples to overseas OIE Reference Laboratories for ASF

- Yes.

# Main challenges in the diagnosis of ASF and other pig diseases

- Lack of human resources
- Availability of diagnostic kits, reagents and consumables
- Availability of reference materials (antisera against different strains)

# **TECHNICAL SUPPORT**

# Laboratory technical support/ training received from Reference Laboratories

Timor Leste received technical support and training from the Reference Laboratories but detail of the training was not provided.

# Technical support expected from OIE/ FAO partners and Reference Laboratories

- Proficiency testing – LAMP and PCR test

# Expected training on laboratory diagnosis of ASF from OIE/ FAO/ Partners and Reference Laboratories

- Timor Leste expects support on the training of their staff through laboratory hands-on training.
- The priority topics identified is molecular diagnosis of ASF.

# Other comments to improve ASF diagnosis

- No comments

# Thailand

# **BACKGROUND DETAILS**

Department: Department of Livestock Development (DLD).

Name of the National Laboratory: National Institute of Animal Health. National Laboratory is accredited to Biosafety level 3 (BSL-3).

Contact Person: Dr Prakit Boonpornprasert.

Position: Veterinarian (Veterinary Virologist)

Email: prakitb@dld.go.th

#### Laboratories involved in diagnosis of ASF at the national and provincial level:

- National Institute of Animal Health National reference lab. for ASF diagnosis and confirmation (qPCR, VI, Sequencing, ELISA, IPT)
- Veterinary research and development center (Upper Northern/Lower Northern/Upper Northeastern/Lower Northeastern/Western/Eastern/Upper Southern/Lower Southern region) -Regional labs for ASF diagnosis (qPCR) and refer samples to national Lab for further diagnosis

#### National Laboratory is accredited to Biosafety level 3 (BSL-3): Yes

#### **DIAGNOSTIC TESTS**

#### Diagnostic tests for agent identification/ detection of virus

- Real-time PCR
- Conventional PCR
- Virus isolation/ HAD test (Haemadsorption)
- Sequencing Sanger sequencing and next generation sequencing (Illumina)
- Field test

**Approximate number of ASFV Real-time PCR performed per day:** Average 50 tests per day but the capacity can increase to 100 tests/ day.

**Extraction of DNA from field samples (for other diseases):** Extract using automatic nucleic acid extraction machine and manual extraction.

#### Diagnostic tests for (ASF serological tests) detection of immune response

- Antibody ELISA ID Screen<sup>®</sup> African Swine Fever Indirect (ID-VET) and INgezim PPA COMPAC (EUROFINS)
- Immunoperoxidase test (IPT)

Laboratory tests that are accredited to ISO/IEC 17025: Detection of African swine fever using Real-time PCR (processing).

#### **BIOSAFETY AND QUALITY CONTROL**

#### Biocontainment classification of the laboratory to perform routine diagnosis of ASF

Molecular	BSL2
Virus isolation	BSL3
Serology	BSL2

#### Quality control (QC) implemented in Laboratories

QC is implemented in the laboratory by:

- Internal controls (positive/negative extraction control and positive/negative reaction control) for Real-time PCR;
- Interlaboratory comparison (National Lab and Regional Labs);
- Proficiency test (National Lab and OIE reference laboratory

# LOGISTICS AND CHALLENGES

# Preparation for possible increase of sample testing in case of ASF incursion

- Building capacity of the regional laboratories in performing and setting up the PCR laboratories through trainings;
- Strengthen the laboratory capacities via mobile labs and portable qPCR.
- We have laboratory network partnership (private labs/ university labs/ authorised labs) to deal with the surge of samples.
- If the ASF is introduced into the country, we will set up mobile labs (portable qPCR) at the point of outbreak.

# Experience in shipping samples to overseas OIE Reference Laboratories for ASF

- Yes, but it took very long

# Main challenges in the diagnosis of ASF and other pig diseases

- Lack of fund
- Availability of diagnostic kits and consumables

# **TECHNICAL SUPPORT**

# Laboratory technical support/ training received from Reference Laboratories

Thailand received technical support and training from the OIE Reference Laboratories which include:

- Laboratory training; Proficiency testing; Reference materials; Diagnostic reagents and lab consumables; and SOPs.

# Technical support expected from OIE/ FAO partners and Reference Laboratories

- Proficiency testing (ASF) PCR and Serology
- Procurement of diagnostic kits, reagents and lab consumables from International Atomic Energy Agency (IAEA)
- SOPs from OIE Reference Laboratories.

# Expected training on laboratory diagnosis of ASF from OIE/ FAO/ Partners and Reference Laboratories

- Thailand expects support on the training of their staff through laboratory hands-on training on laboratory diagnosis and interpretation.

#### Other comments to improve ASF diagnosis

- No comment

# Vietnam

### **BACKGROUND DETAILS**

### Department: Department of Animal Health

Name of the National Laboratory: National Centre for Veterinary Diagnosis

Contact Person: Dr Nguyen Dang Tho

Position: Head of Virology Office- In-Charge, VLD (Veterinarian IV) / Offficer-In-Charge, ADDRL (Veterinarian III)

Email: thovet99@yahoo.com

#### Laboratories involved in diagnosis of ASF at the national and provincial level

- National Centre for Veterinary Diagnosis
- Regional Animal Health Office No.1 Laboratory
- Regional Animal Health Office No.2 Laboratory
- Regional Animal Health Office No.3 Laboratory
- Regional Animal Health Office No.4 Laboratory
- Regional Animal Health Office No.5 Laboratory
- Regional Animal Health Office No.6 Laboratory
- Regional Animal Health Office No.7 Laboratory

#### National Laboratory is accredited to Biosafety level 3 (BSL-3): No

#### **DIAGNOSTIC TESTS**

#### Diagnostic tests for agent identification/ detection of virus

- Real-time PCR
- Conventional PCR
- Virus isolation/ HAD test (Haemadsorption)
- Sequencing Sanger, NGS
- Antigen ELISA INgezim PPA DAS 2.0
- Florescent antibody test (FAT)

Real-time PCR test is the most popular and common tests used by Vietnam followed by Conventional PCR, Virus isolation, sequencing and Antigen ELISA.

Approximate number of ASFV Real-time PCR performed per day: 500 rxn.

# **Extraction of DNA from field samples (for other diseases):** Extract using automatic nucleic acid extraction machine.

### Diagnostic tests for (ASF serological tests) detection of immune response

- Antibody ELISA - INgezim PPA CROM 2. ID Screen® African Swine Fever Indirect.

- Indirect fluorescent antibody test (IFAT)

### Laboratory tests that are accredited to ISO/IEC 17025: Real-time PCR, PCR, ELISA

#### **BIOSAFETY AND QUALITY CONTROL**

#### Biocontainment classification of the laboratory to perform routine diagnosis of ASF

Molecular	BSL2
Virus isolation/FAT/Ag ELISA	BSL2
Serology	BSL2

#### Quality control (QC) implemented in Laboratories

- Internal controls are used;
- Test value of positive control are recorded.

#### LOGISTICS AND CHALLENGES

#### Dealing with surge of samples due to increased ASF outbreaks

Following measures were undertaken to deal with the surge of samples due to increasing ASF outbreaks.

- Work day and night on shift system.

#### Experience in shipping samples to overseas OIE Reference Laboratories for ASF

- Yes

#### Main challenges in the diagnosis of ASF and other pig diseases

- Availability of reference materials (antisera against different strains)
- Lack of technical capacity
- Poor laboratory infrastructure/ limited space

#### **TECHNICAL SUPPORT**

#### Laboratory technical support/ training received from Reference Laboratories

Vietnam received technical support and training from the OIE Reference Laboratories which include:

- laboratory training, Proficiency Testing (PT), reference materials, SOPs supported by IAEA Animal Health laboratory.
- There is ongoing Laboratory Twinning Project between ACDP and RAHO6 for swine diseases including ASF diagnostic capacity building.

# Technical support expected from OIE/ FAO partners and Reference Laboratories on diagnosis of ASF and other pig diseases

- Proficiency testing (PT) Real-timePCR
- Provision of reference material
- Diagnostic kits, reagents and lab consumables
- SOPs

# Expected training on laboratory diagnosis of ASF from OIE/ FAO/ Partners and Reference Laboratories

- Philippines expect support on the training of their laboratory staff through laboratory hands-on training or via on-line training course.
- The topics identified to be covered in the future training programme are virus isolation; Cell culture; and Sequencing.

#### Other comments to improve ASF diagnosis

- Development of ASF vaccines in good progress in Vietnam.
- Concerned about non availability of DIVA tests.

# Annex 2: Questionnaire to review the status of African swine fever (ASF) laboratory diagnostic capacities in South-East Asia

The Standing Group of Experts on ASF for Asia Pacific (SGE-ASF Asia Pacific) identified several priority topics related to ASF. Laboratory diagnostic capacity development was one of the identified priorities. Much work has been done across the South-East Asia region to build capacity over the past years, however there is still a need to further enhance the laboratory and the field diagnostic capacity, harmonize laboratory diagnostic techniques, facilitate the sharing of information and coordinate laboratory activities amongst national laboratories, Reference Laboratories and laboratory experts to strengthen ASF control.

The third ASF coordination meeting will provide platform to share experiences and challenges in ASF diagnosis, assess laboratory capacity and discuss measures to enhance laboratory diagnostic capacity and coordinate laboratory activities to strengthen ASF control in the region. Therefore we have prepared a questionnaire to review the current laboratory diagnostic capacity and identify challenges faced in ASF diagnosis by the member countries.

Through this questionnaire, the OIE SRRSEA and FAORAP expect:

- to understand the existing laboratory activities and diagnostic tests used by the countries for ASF diagnosis;
- to understand the challenges faced by countries in ASF diagnosis;
- to understand the areas of support required by the member countries to enhance ASF diagnosis from OIE/FAO and other partners;
- to seek recommendations to enhance diagnostic capacity of the members and to strengthen ASF control in the region.

This questionnaire was developed based on the recommendations of the first meeting of the Regional Laboratory Expert on ASF and other pig diseases in Asia and the Pacific on 24 June 2021 and further improved during the preparatory meeting of the Third ASF Coordination meeting among the Experts from ASF Regional Reference Laboratories (Australian Centre for Disease Preparedness and China Animal Health and Epidemiology Centre) and staffs of FAORAP and OIE (SRRSEA& RRAP) on 10 August 2021.

Ment

# What kind of information from Members would be useful for RLs and leading laboratory in plaining/providing training or technical support



Background

Objective

Survey Methodology

Plenary session

Findings

- Diagnostic tests
- Biosafety and quality control
- Challenges
- Technical support
  - o Existing
  - o Next
- Way Forward

Annexes

# **BACKGROUND DETAILS**

Country:

Department:

Name of the Laboratory:

**Contact Person:** 

Position:

Email:

- 1. List the name of laboratories involved in diagnosis of ASF at the national and provincial level? For example National Reference Lab for confirmation and other laboratories in the provinces to provide provisional diagnosis and refer samples to national laboratory?
- 2. Have your National Laboratory accredited to Biosafety level 3 (BSL-3)?

Yes | No

# **DIAGNOSTIC TESTS**

- 3. Does your country/territory have the capacity to conduct the following laboratory tests for (ASF Virological or molecular) agent identification? *Identify and rank as per usage/ frequency of use?* 
  - a. Conventional PCR
  - b. Real-time PCR
  - c. Antigen ELISA (Indicate in-house or commercial/ brand)

- 4. If you laboratory is using Real-time PCR, approximate number of real-time PCR tests performed per day in your laboratory?
  - a. Conventional PCR
  - b. Real-time PCR
  - c. Antigen ELISA (Indicate in-house or commercial/ brand)
- 5. Do you extract DNA from field samples manually or using automatic nucleic acid extraction machine?
  - Extract manually
  - Extract using automatic nucleic acid extraction machine
- 6. Does your country/territory have the capacity to conduct the following laboratory tests for (ASF serological tests) detection of immune response? *Identify and rank as per usage/ frequency of use?* 
  - a. Antibody ELISA (Indicate inhouse or commercial/ brand)
  - b. Immunoblotting test (IBT)
  - c. Immunoperoxidase test (IPT)
  - d. Indirect fluorescent antibody test (IFAT)
  - e. Lateral flow device (antibody)
- 7. Lists the tests (for ASF diagnosis) in your laboratory that are accredited to ISO/IEC/17025?

#### **BIOSAFETY AND QUALITY CONTROL**

8. What is the biocontainment classification of the laboratory where you perform routine diagnosis for ASF?

Molecular	
Virus isolation/FAT/Ag	
ELISA	
Serology	

- 9. What are the biosecurity measures implemented in case you perform field tests for ASF diagnosis?
- 10. How is quality control being implemented in your Laboratories?

# LOGISTICS AND CHALLENGES

- 11. For ASF infected country, how did you deal with surge of samples due to increased ASF outbreaks?
- 12. For ASF free country, how do you prepare for possible increase of sample testing in case of an ASF incursion in your country?
- 13. Does your country (National Reference Laboratory) have successful experience in shipping samples to overseas OIE Reference Laboratories for ASF (ACDP or CAHEC in Asia and/ or other OIE ASF Reference Laboratories?
  - Yes | Yes, but it took very long time | No

# 14. What are the main challenges in your country in the diagnosis of ASF and other pig diseases?

- a. Lack of human resources
- b. Lack of fund
- c. Availability of diagnostic kits and consumables
- d. Availability of reference materials
- e. Lack of technical capacity
- f. Poor laboratory infrastructure/ limited space
- g. Poor reporting and submission of samples
- h. Others (specify)

# **TECHNICAL SUPPORT**

# **15.** Has your country/territory received laboratory technical support/ training from Reference Laboratories/ Leading Laboratories?

Yes | No

If Yes, describe the type of support; for example, laboratory training, Proficiency Testing (PT), reference materials, SOPs etc. and indicate which lab was supporting and the date of the activity?

# 16. What technical support do you expect from OIE/ FAO partners and Reference Laboratories?

- a. Proficiency testing
- b. Provision of reference materials
- c. Procurement of diagnostic kits, reagents and lab consumables
- d. SOPs
- e. Other (please specify)

# 17. Provide details on each of answer provided above? (e.g. proficiency test for which test(s))

# **18.** What type of training on laboratory diagnosis of ASF do you expect from OIE/ FAO partners and Reference Laboratories?

- a. Laboratory hands-on training
- b. On-line training course
- c. Others (please specify)

# **19.** If you could avail training on laboratory diagnosis, what topics you wished to be covered in the training?

# 20. Any other comments to improve ASF diagnosis in your country?

Thank You