Research article

Risk Assessment of African Swine Fever Virus in Pork in Phnom Penh, Cambodia

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Abstract African swine fever (ASF) is an acute infectious and deadly viral disease that affects domestic and wild pigs of all breeds and ages. ASF virus (ASFV) can spread vastly to non-infected pig population, but it cannot be transmitted from pigs to humans. In Cambodia, first ASF outbreak was transmitted from bordering countries and reported in five provinces. While estimated 70% of ASF widespread all over the country, local farmers experienced a greater economic loss. Among the four components of risk analysis, the study determines the risk assessment, and hazard identification plays a crucial step in risk assessment. The objectives of this study aimed to detect the presence of ASFV in pork on local markets through qualitative risk assessment approach and propose possible measurable recommendations to prevent ASF outbreak. The study was conducted during the period of ASF outbreak in August 2019, and the qualitative detection of ASFV was conducted on pork tissue samples selected from wet markets and supermarkets in Phnom Penh. Sample extractions were isolated from 30 pork tissue samples and detected virus by iiPCR. The qualitative result on detection of ASF virus is confirmed by PCR technique. The ASFV is found in pork tissue samples in wet markets and supermarkets. Of the 30 samples, 21 (70%) were found positive with ASFV, 6 in 9 (20%) tissue samples from supermarkets and 15 in 21 tissue samples from wet markets (50%) confirmed the presence of ASFV. With this result, it indicates that likelihood of the ASF virus transmission would be very likely to occur and the spread of ASF virus in pork tissue samples in wet markets and supermarkets is significantly prevalent, and the virus is likely to spread quickly. Scientifically, there is no vaccine to prevent ASF, and as recommended by FAO, the influenced policy-based implementation is required in place to minimize further production losses. The implementations must be strengthened through strict farm biosecurity guideline and slaughter of infected pigs, strict import regulation (border and movement control of live pigs) and heavy penalty to illegal import of live pigs. Based on this result, it may contribute to bring consumers' and relevant

stakeholders' awareness to reduce high risk through early detection of ASFV at the slaughterhouses and markets by risk assessment approach. Further studies on risk management and risk communication to complete the risk analysis of ASFV in pork are highly recommended.

Keywords African swine fever virus, risk assessment, pork, markets, Cambodia

INTRODUCTION

African swine fever (ASF) is an acute infectious and deadly viral disease that affects domestic and wild pigs of all breeds and ages, and caused by a DNA virus belonging to *Asfarviridae* family (Guberti et al., 2018). ASF virus (ASFV) can spread vastly to non-infected pig population, but it cannot be transmitted from pigs to humans (FAO, 2008). Although humans are not susceptible to ASF, its outbreak causes noticeable socio-economic consequences to infected countries (Bellini, Rutili and Guberti, 2016). In Cambodia, first ASF outbreak transmitted from bordering countries was reported in Ratanakiri province by the Ministry of Agriculture, Forestry and Fisheries, MAFF (FAO, 2019). ASFV has continued its transmission and spread quickly to Tboung Khmum, Svay Rieng, Takeo and Kandal provinces. While estimated 70% of ASF widespread all over the country, local farmers in these five areas experienced a greater economic loss. Majority of small- and medium-scale pig farms stop to operate temporarily, and a small number of commercial farms are being operated (Siem Reap PDAFF, 2019).

In such cases, risk analysis, the process composed of hazard identification, risk assessment, risk management and risk communication, is required. FAO/WHO (2006) defined risk assessment as the term that is generally used to describe the entire process of making a public health decision regarding a specific drug or agent and it is the scientific evaluation of known or potential adverse effects resulting from human exposure to food borne hazards. According to Venn et al. (2014) as cited in the Codex Alimentarius Commission (CAC, 1999), risk assessment is defined as integrated elements of the structure of risk analysis and to be based on the following steps as shown in Fig. 1 and Fig. 2.

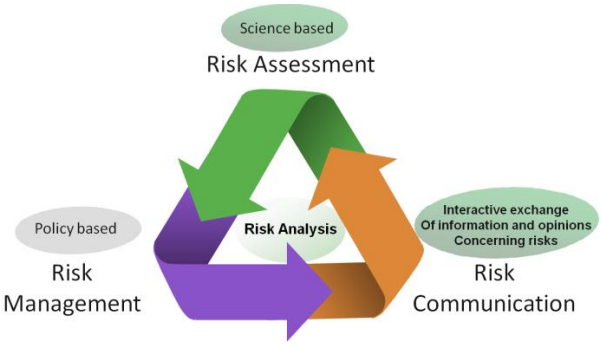

Fig. 1 Structure of risk analysis (Venn et al., 2014 cited in CAC, 1999)

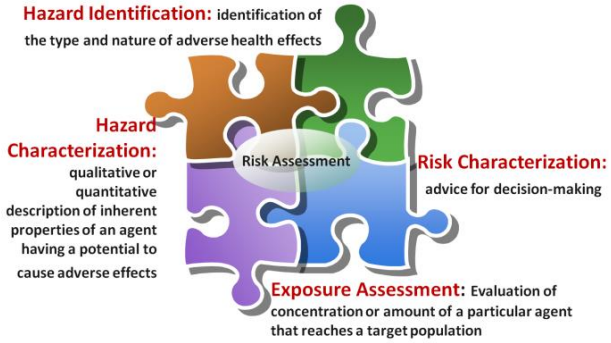

Fig. 2 Risk assessment (Venn et al., 2014 cited in CAC, 1999)

ASFV can be transmitted and spread from infected pigs directly to uninfected populations. The cause of transmission is associated with virus-containing matter (blood, faeces, urine or saliva from infected pigs) that contact directly with brought-to-farm infected pork, farm inputs and in-farm used materials (bedding, pig cage, feed, equipment, clothing and footwear, means of transportation (vehicles), studied by Thomson (1985). In the research of Mellor et al. (1987), it has shown that ASFV can be airborne within a short distance, approximately less than 2 kilometers.

ASFV has a remarkable ability to survive for long periods in a protein environment, and therefore meat from pigs slaughtered in the infective stages of ASF or die naturally of the disease provides a good source of virus. Pork slaughtered from infected pigs with ASF provides a favorable protein environment for the virus to survive in the tissues for a long period of time. The virus is strongly resistant to high temperatures. The studies of McKercher et al. (1978) and Plowright et al. (1994) confirmed that fresh, frozen, processed, or preserved (salted and dried) pork may contain the infective virus. As ASFV is transmitted in a closer distance, it may be possible to prevent the occurrences by applying strict biosecurity rules and observation. While taking precautionary measures in use, it is necessary to limit accesses for people/ farm workers/ visitors and vehicles to enter the areas or farms where pigs are kept. This is also ensuring that the farm managers/ owners and veterinarians are disinfected before entering the critical areas/ farms with protective clothing and footwear, and free from inadvertent feeding of leftovers and pork (Penrith and Vosloo, 2009).

OBJECTIVE

This study aims to detect the presence of ASF virus in pork in Cambodian markets through risk assessment approach, and propose possible measurable recommendations to prevent ASF outbreak.

METHODOLOGY

The study was conducted during the period of ASF outbreak, and the qualitative detection of ASF virus was conducted on pork tissue samples using PCR technique certified by OIE approval number 20130108 for registration of diagnostic kits (GeneReach Biotechnology Corp., 2019). The samples were selected from 7 wet markets and 3 supermarkets in Phnom Penh in August 2019. Primary information was gathered by interviewing the key informants of the city slaughterhouses and sellers in both markets. The distribution of live pig movement and slaughtered pork was from smallholder famers, middlemen and traders who source for pigs from the small-scale farms and neighboring countries.

Sample extractions were isolated from 30 pork tissue samples and detected virus. The analysis samples were evaluated based on the insulated isothermal PCR technique that automates sample lysis, nucleic acid extraction and amplification and detection of the target sequences. Pork tissue samples were pretreated by weighting 40 mg of each pork tissue sample and placing them into a clean 1.5 ml microcentrifuge tube with 0.5 ml of Sample Storage Solution atc-lysis. The pork tissues were homogenized in the grinder and spined the tube for 1 minute in mini-centrifuge before transferring 200 µl of supernatant to an Extraction Cartridge. Then, Transfer Cartridge was prepared by placing one Premix and one Transfer Cartridge for each sample; labeling the sample ID and Premix ID on the side of Transfer Cartridge; removing Transfer Cartridge Cap; turning the notched side of the cartridge away, and snapping the Premix vial into well #3 of the Transfer Cartridge. The Extraction Cartridge was prepared by placing one Extraction Cartridge (B) for each sample; removing the Extraction Cartridge from the aluminum pack, labeling the sample ID on the side of Extraction Cartridge; slowly peeling off the aluminum film; turning notched side, and loading 200 µl of homogenized sample to the sample well of the Extraction Cartridge. After the preparations of Transfer Cartridge and Extraction Cartridge were completed, the Extraction lot number of the Extraction Cartridge was entered into the PCR; then the loaded Extraction Cartridge was place into the selected slot before the Reagent lot number of the Premix Reagent was entered. The Transfer Cartridge to the selected was loaded into the slot and the analysis were ready to run in the PCR.