Research Application Summary

Harnessing higher education institutions' capabilities in controlling trans-boundary animal diseases at the livestock-wildlife interface: the case for African swine fever in Uganda

Masembe, C.,*¹ Ademun, A.R.O.,² Stahl, K.,⁴ Atimnedi, P. & Muwanika, V. B.³ ¹College of Natural Sciences, School of Biological Sciences, Department of Zoology, Entomology and Fisheries Sciences, Makerere University, P. O. Box 7062, Kampala, Uganda, ²Ministry of Agriculture Animal Industry and Fisheries, National Animal Disease Diagnostics and Epidemiology Centre, P. O. Box 513, Entebbe, Uganda ³ College of Agriculture and Environmental Sciences, School of Forestry, Geographical and Environmental and Sciences, Department of Environmental Management, Molecular Genetics/Biology Laboratory, Makerere University, P.O. Box 7062, Kampala, Uganda ***Corresponding Author:** cmasembe@cns.mak.ac.ug

Abstract

Uganda is the third largest pig producing country in Africa. However the pig industry in Uganda is currently threatened by African swine fever virus (ASFV) disease, which is a fatal, contagious viral haemorrhagic disease of domestic pigs, causing up to 100% mortality, and the disease has neither vaccine nor treatment. Harnessing the technological, research potential and community outreach strategies, Makerere University in partnership with government agencies, is using a community participatory epidemiology and molecular characterization approach to develop community-derived control strategies and genetic characteristics of the ASF virus. Through this effort the effectiveness of available molecular ASF diagnostic approaches and home ranges of domestic and wild pigs are being investigated. Seventy seven blood samples were collected from pigs during reported ASF outbreaks in selected pig farms in Uganda. The samples were analysed using a variety of molecular diagnostic approaches available at the molecular genetics laboratory at Makerere University. Preliminary results indicate that the samples tested positive for ASFV. In addition tracking collars (GPS/GSM) were used to monitor the movement patterns of free-ranging domestic pigs and wild pigs. Domestic pigs were mostly active during the nights, early mornings and in the evening. The bush pigs were active from the evening and throughout the night wandering between game reserves and farmlands. There was an overlap between free-ranging domestic pigs and bush pig activity times and this might be a contributing factor to the ASFV disease epidemics. The study involves training two MSc and four undergraduate students. It is anticipated that the trained students will add to the pool of home-grown scientists that can contribute to disease diagnostics prevention and control.

Keywords: African swine fever, bush pig, diagnostics, domestic pigs, tracking collars, Uganda

Masembe, C et al.

Résumé

L'Ouganda est le troisième plus grand pays producteur du porc en Afrique. Pourtant, l'industrie du porc en Ouganda est actuellement menacé par le virus de la maladie de la peste porcine africaine (PPA), qui est une maladie mortelle, contagieuse virale hémorragique des porcs domestiques, causant jusqu'à 100% de mortalité, et la maladie n'a ni vaccin, ni traitement. Utilisant les stratégies de sensibilisation technologiques, le potentiel de recherche ; et de l'engagement communautaire, l'Université de Makerere en partenariat avec les organismes gouvernementaux, utilise une épidémiologie participative communautaire et l'approche de la caractérisation moléculaire pour développer des stratégies de contrôle inspirées par la communauté et les caractéristiques génétiques du virus de la peste porcine africaine. Grâce à cet effort l'efficacité des approches de diagnostic moléculaire de la PPA disponibles et l'élevage en liberté des porcs domestiques et des porcs sauvages ont été étudiés. Soixante-dix-sept échantillons de sang ont été prélevés sur des porcs pendant les épidémies de peste porcine africaine signalées dans élevages de porcs sélectionnés en Ouganda. Les échantillons ont été analysés en utilisant une variété d'approches de diagnostic moléculaire disponible au laboratoire de génétique moléculaire à l'Université de Makerere. Les résultats préliminaires indiquent que les échantillons ont testé positifs pour le virus de la PPA. En outre, les colliers de repérage (GPS / GSM) ont été utilisés pour surveiller les habitudes de déplacement des porcs domestiques en liberté et des porcs sauvages. Les porcs domestiques étaient surtout actifs pendant les nuits, tôt le matin et le soir. Les porcs sauvages étaient actifs dans la soirée et toute la nuit à errer entre les réserves de chasse et des terres agricoles. Il y avait un chevauchement entre les périodes d'activité de porcs en liberté et des porcs sauvages, ce qui serait un facteur contribuant aux épidémies de la PPA. Cette étude contribue à la formation de deux étudiants Master et quatre étudiants de premier cycle. Il est prévu que les étudiants formés vont s'ajouter au nombre de scientifiques formés localement qui peuvent contribuer à la diagnostique, la prévention et le contrôle des maladies.

Mot Clés : la peste porcine africaine, les porcs sauvages, la diagnostique, les porcs domestiques, des colliers de repérage, l'Ouganda

The epidemiology of African swine fever

Agriculture is the major economic activity in Uganda. According to the 2008 economic indicators, the contribution by agriculture to the national GDP was 21.5% (UBOS, 2009). Livestock production in Uganda contributes 5% and 14.6% to total GDP and agricultural GDP, respectively (UBOS, 2004). Livestock is an integral part of the agricultural system in many parts of the country. The increasing human population and middle class workers have given a strong impetus to pig production in central, eastern and northern Uganda. Pigs are kept in both municipal and remote villages, where they assume great significance for food security in the families. However, pig production faces a number of constraints, one of which is infectious diseases like African Swine Fever (ASF).

140

141

In Uganda, 85% of the population lives in rural areas and depends on agriculture, with the livestock sub-sector steadily increasing in response to the demand. The country has the 2nd largest and most rapidly growing pig production in Africa, with a pig population that stood at 3.5 million in 2012 (UBOS, 2012). More than 75% of these pigs are found in smallholder farms in the rural areas in systems with low biosecurity levels. Pig keeping is of great importance as 44% of world meat protein consumption is derived from pork and pork products (FAO, 2001). For the smallholder pig keeper, pigs are considered 'walking banks'. This is due to the fact that pigs grow to maturity in a short time (8-12 months); produce several piglets (6-12 piglets); and can be easily sold with relative ease at any stage. Indeed preliminary field studies in Gulu district (northern Uganda) show that pork is one of the cheapest forms of animal protein to raise, since pigs can be raised successfully on food waste and other inexpensive fodder (ASFUganda, unpublished).

Despite all the above good attributes from pig keeping, the pork industry is faced with a serious, and most feared lethal threat of (ASF) disease, which has neither vaccines nor treatment; and it is an OIE notifiable Transboundary Animal Disease (TAD). In order to control this disease, there is an urgent need to investigate the ASF disease transmission epidemiology (Kukielka *et al.*, 2016), and community biosecurity participatory integrated approach so as to design ASF control strategies for improved pork production, food security and livelihoods. This can be done by validating the available diagnostic platforms, and investigating the role of wildlife in the epidemiology of the disease.

African swine fever is endemic in Uganda with outbreaks occurring at regular intervals (>20 outbreaks confirmed during a 10 months period of October 2010-June 2011). ASF is caused by a double stranded DNA virus (ASFV) classified within the Asfarviridae family, genus Asfivirus. ASFV has approximately 200,000 base pairs, and exists in 22 genotypes, which have been previously classified by sequencing of a relatively conserved P72 region of the genome. In countries where proper control measures are in place, effective control is by diagnosis and slaughter to eliminate infected animals, followed by compensation. Given Uganda's low economic level, this approach is not possible at the moment. Subsequently, the affected farmers lose their pigs to the disease, putting their livelihoods into food and economic insecurity.

African Swine Fever is indigenous to the African continent where it is thought to occur in one of three possible cycles; (i) an ancient sylvatic cycle, (ii) a pig/tick cycle, and (iii) a more recently described domestic pig cycle without the involvement of other hosts or vectors (Bastos *et al.*, 2003). ASF epidemiology is complex with different epidemiological patterns of infection occurring in Africa and Europe. ASF occurs through transmission cycles involving domestic pigs, wild boars, wild African suids, and soft ticks (Sánchez-Vizcaíno *et al.*, 2009). In regions where Ornithodorus soft-bodied ticks are present, the detection of ASFV in these reservoirs of infection contributes to a better understanding of the epidemiology of the disease. This is of major importance in establishing effective control and eradication programmes (Basto *et al.*, 2006).

Masembe, C et al.

The ancient sylvatic cycle is well described from eastern and southern Africa (Costard *et al.*, 2009). It involves soft ticks and wild suids, especially warthogs, who are natural hosts for the virus. Studies have shown that about 80% of free-living warthogs are infected with ASF virus in areas where ticks are present and can act as vectors (Sánchez-Vizcaíno *et al.*, 2012). Historically the sylvatic cycle dominated, and ASF outbreaks occurred more or less frequently as a result of spill over to domestic pigs at the wildlife-livestock interface.

The domestic pig cycle is one of the three cycles where the ASF is maintained within domestic pigs through direct contact, which is predominant, and indirectly between domestic pigs which is the most important route of transmission and the infection are mainly by ingestion and inhalation (Penrith *et al.*, 2009). The opinion about how long a domestic pig can stay infected varies amongst scientists. A pig with acute ASF shed high amounts of virus and if it survives it can continue to shed virus for about 30 days even if the infection may persist longer. According to Penrith (2009) the virus can persist for a maximum of three to four months while Costard *et al.* (2009) believe that it can persist for at least six months.

Difficulties in detection of ASF Virus

There are several tests that can be used to diagnose ASFV but almost each of them has shortcomings. For example the lateral flow devices pose an uncertainty on the intensity of the "positive line", while the conventional PCR (Mullis and Faloona, 1987) diagnosis requires robust resources. To circumvent these challenges, total genomic DNA was extracted directly from blood samples using the DNeasy Blood and Tissue Purification Kit (QIAGEN) following the manufacturer's guidelines. A tetracore Realtime PCR (http://www.tetracore.com/) assay was used to screen the blood DNA samples for presence of ASF viral DNA. Another platform (Blood Direct) using the blood directly to conduct an ASF specific PCR was carried out. There is however need to collect several field material and test them on a panel of tests, and the best performing test should then be rolled out to partner institutions involved in animal disease diagnosis.

The preliminary results on pig movements and interactions

GPS collared pigs revealed that the free ranging domestic pigs actually moved between homesteads and gathered around waste disposal centers, and occasionally raided crops in the gardens. This roaming provides domestic pigs with an opportunity to share feeding grounds with the other free ranging domestic pigs in a particular area, and hence spreading pathogens, including the deadly ASFV. Bush pig movement patterns showed that the wild pigs actually moved across the wildlife-livestock interface, from the swamps during the night into homesteads, which kept domestic pigs and then back to the swamp at daybreak. This provides another potential overlap between domestic and wild pigs, creating an avenue for pathogen exchange.

How can Higher Education Institutions contribute to disease control?

The ASF diagnostic approaches and pig movement investigations have been done under the leadership of Makerere University (HEI), whose mandate is teaching,

142

Fifth RUFORUM Biennial Regional Conference, 17-21 October 2016, Cape Town , South Africa 143 research and outreach. The results obtained above have potential uptake by the Uganda Ministry of Agriculture Animal Industry and Fisheries for improving surveillance, disease prevention and control. Indeed the Higher Education Institutions are at the forefront of research that aims to improve livelihoods. Therefore the role of HEI's in community development should not be neglected.

Acknowledgements

Authors acknowledge the valuable work of the field veterinary staff in Greater Masaka and Greater Gulu regions, the Uganda Wildlife Authority staff and the larger ASF Uganda team that made this work possible. The study was partly financed by USDA-APHIS, Sida, RUFORUM, and the Wellcome Trust [105684/Z/14/Z]. This paper is a contribution to the 2016 Fifth African Higher Education Week and RUFORUM Biennial Conference.

References

- Bastos, A. D. S., Penrith, M. L., Cruciere, C., Edrich, J. L., Hutchings, G., Roger, F., Couacy-Hymann, E. and Thomson, G. R. 2003. Genotyping field strains of African swine fever virus by partial p72 gene characterisation. *Arch. Virol* 148 (4): 693–706. (doi:10.1007/s00705-002-0946-8).
- Kukielka, E., Jori, F., Martínez-López, B., Chenais, E., Masembe, C., Chavernac, D. and Ståhl, K. 2016. Wild and domestic pig interactions at the wildlife-livestock interface of Murchison Falls National Park, Uganda, and the potential association with African swine fever outbreaks. *Frontiers in Veterinary Science* 3: 31. DOI=10.3389/fvets.2016.00031
- Catley, A., Alders, R. G. and Wood, J. L. 2012. Participatory epidemiology: approaches, methods, experiences. *The Veterinary Journal* 191 (2):151-160.
- Mullis, K. B. and Faloona, F. 1987. Specific synthesis of DNA in vitro via a polymerase catalyzed chain reaction. *Methodology in Enzymes* 155: 335-350.
- Mendoza, G., 1995. A primer on marketing channels and margins. In: Scott, J. (Ed.), Prices, Products, and People: Analyzing Agricultural Markets in Developing Countries.Lynne Rienner Publishers.
- Uganda Bureau of Statistics (UBOS), 2004. Uganda Bureau of Statistics. Ministry of Finance, Planning and Economic Development.
- Uganda Bureau of Statistics (UBOS). 2009. Statistical Abstract. Kampala, Uganda: UBOS. <u>Onlineathttp://www.ubos.org/onlinefiles/uploads/ubos/pdf%20docu</u> <u>ments/2009Statistical %20 Abstract.pdf (last access 26/03/2014)</u>
- Sánchez-Vizcaínoj, M., Martinez-López, B., Martinez-Avilés, M., Martins, C., Boinas, B., Vial L., Michaud, V., Jorif. Etter, E., Albina, E. and Roger, F. 2009. Scientific Review on African swine fever. CFP/EFSA/AHAW/2007/02, pp: 1–141.
- Sánchez-Vizcaíno, J.M., Mur, L. and Martínez-López, B. 2012. African swine fever: An epidemiological update. *Transboundary and Emerging Diseases* 59 (1): 1-9.
- Penrith, M-L., Guberti, V., Depner, K. and Lubroth, J. 2009. Preparation of African swine fever contingency plans. Rome: FAO Animal Production and Health Manual No.8.