Pakistan Veterinary Journal

ISSN: 0253-8318 (PRINT), 2074-7764 (ONLINE)
Accessible at: www.pvj.com.pk

SHORT COMMUNICATION

Detection of Porcine Circovirus Type 2 (PCV2) in Mosquitoes from Pig Farms by PCR

Xiaohong Yang, Lidan Hou, Jing Ye, Qigai He and Shengbo Cao*

State Key Laboratory of Agricultural Microbiology and Laboratory of Animal Virology, College of Veterinary Medicine Huazhong Agricultural University, Wuhan, Hubei, 430070, P. R. China
*****Corresponding author: sbcao@mail.hzau.edu.cn

ARTICLE HISTORY

Received: April 28, 2011
Revised: June 15, 2011
Accepted: July 03, 2011

Key words:
Mosquitoes
PCV2
Polymerase chain reaction (PCR)
Vector

ABSTRACT

To investigate, whether mosquitoes could be potential vectors that maintain or transmit PCV2, 59 mosquito samples were collected from suspicious PCV2-infected pig farms in Hubei and Anhui province, China. Total DNA from these mosquitoes was extracted and then tested for presence of PCV2 nucleic acid by PCR. Four (6.78%) samples showed the positive result. Subsequently, the positive PCR product was cloned into pEASY-T1 vector and sequenced. Sequence analysis displayed that homology between the PCR products and PCV2 strains were more than 94%. These results demonstrate that PCV2 nucleic acid exists in these mosquitoes, which suggests that mosquitoes could serve as mechanical transmission vectors of PCV2.

To Cite This Article: Yang X, L Hou, J Ye, Q He and S Cao, 2012. Detection of porcine circovirus type 2 (PCV2) in mosquitoes from pig farms by PCR. Pak Vet J, 32(1): 134-135.

INTRODUCTION

Porcine circovirus (PCV) is a non-enveloped, circular single-stranded DNA virus that belongs to the genus *circovirus* in the family of *circoviridae*. This virus is usually divided into two groups: porcine circoviruses type 1 (PCV1) and porcine circoviruses type 2 (PCV2) (Allan *et al*., 1998). PCV2 are associated with many diseases, such as postweaning multisystemic wasting syndrome and porcine dermatitis nephropathy syndrome, which are also related to porcine respiratory disease complex and many other diseases (Chae, 2005). In recent years, the presence of PCV2 nucleotide in domestic pigs, mice, rats (Lorincz *et al*., 2010) and wild boars (Schulze *et al*., 2003) has been reported. However, the presence of PCV2 in cattle, horse, sheep and humans is still controversial (Allan *et al*., 2000; Ellis *et al*., 2001).

Mosquitoes, as one of the most well known vectors of pathogens, have great impact on public health. They can carry pathogen and transmit diseases including protozoan diseases i.e., malaria, filarial diseases, e.g., dog heartworm and viral diseases such as dengue, Japanese encephalitis and yellow fever (Hemingway *et al*., 2006). So far, there hasn't been any information whether mosquitoes can maintain or transmit PCV2. Thus, the purpose of our research is to investigate the presence of PCV2 in mosquitoes, and its roles in transmission of PCV2.

MATERIALS AND METHODS

Mosquito (n=59) samples were collected from suspicious PCV2-infected pigs in 9 farms in Hubei and Anhui province in China, and all of them were identified as mosquitoes-Culex. Total DNA of these mosquitoes were extracted and then detected for PCV2 nucleic acid by PCR. The nucleotide sequences for the sense primer (5'-CACGGATATTGTAGTCCTGGT-3') and anti-sense primer (5'-GACAGTATATCCGAAGGTGCGG-3') were specific for conserved regions of the ORF2 gene. PCR was performed under the following conditions: denaturation for 5 minutes (min) at 95°C; 30 cycles of 1 min at 95°C, 30 s at 56°C, and 45 s at 72°C; the final step for 10 min at 72°C. The amplified PCR products were verified in 0.8% agarose gel.

RESULTS AND DISCUSSION

Among all the samples being tested, four (number 5, 13, 14, 15) out of 59 showed the positive bands (Fig. 1), while no signal was detected in negative control (water as sample). Subsequently, the positive PCR product was cloned into pEASY-T1 vector and sequenced. The result of sequence analysis showed that the matching rate was more than 94% between the PCR products and PCV2 strains, indicating that PCV2 exist in these mosquitoes.

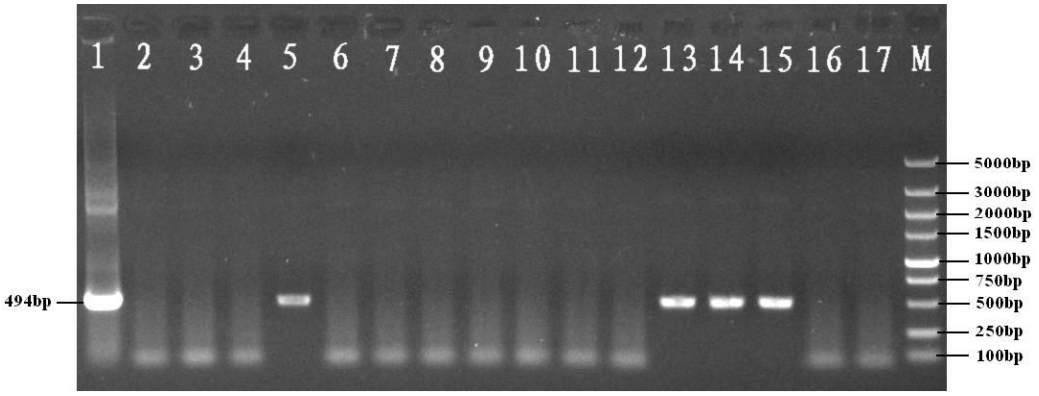

Fig.1: Detection of PCR products for PCV2. Total DNA of these mosquitoes were extracted and then detected for PCV2 nucleic acid by PCR. The amplified PCR products were verified in 0.8% agarose gel. M□DL5, 000 DNA marker; 2~16: PCR products; 1: positive control; 17: negative control.

Beside domestic pigs, whether non-pig animals, such as mice, rats, ringdoves, sparrows, mosquitoes and flies, can act as natural reservoirs for PCV2 remains unclear. In natural, the biological transmission of PCV2 arises only among pigs, but not via other carriers such as insects or other animals living in pig herd (Yang *et al*., 2007). Under laboratory conditions, PCV2 could replicate in mice after inoculation intraperitoneally, intranasally or *per os* and the virus could transmit directly between mice (Cságola *et al*., 2008). These findings displayed that mice might be a kind of natural reservoir and could transmit PCV2. Our results showed that PCV2 can be present in mosquitoes-Culex, which deduced that mosquitoes could serve as mechanical transmission vectors of PCV2. It has been reported that mosquitoes-Aedes vexans can serve as mechanical vectors of Porcine Reproductive and Respiratory Syndrome Virus (PRRSV), but are not likely to serve as biological vectors of PRRSV (Otake *et al*., 2003) for PRRSV can not survive in this mosquitoes. However, they could survive in mosquitoes-Culex tritaeniorhychus (Shi *et al*., 2009). Mosquitoes still can act as a potential vector that transmits PRRSV.

The findings of our research demonstrated that PCV2 could present in mosquitoes, and also indicated that mosquitoes could serve as the potential vector during the transmission of PCV2. However, whether mosquitoes play an important role in the transmission of PCV2 is still need further research.

Acknowledgement

This work was supported by grant # NYCYTX-009.

REFERENCES

Allan G, B Meehan, D Todd, S Kennedy, F McNeilly, J Ellis, EG Clark, J Harding, E Espuna, A Botner and C Charreyre, 1998. Novel porcine circoviruses from pigs with wasting disease syndromes. Vet Rec, 142: 467-468.

Allan, GM, F McNeilly, I McNair, MD Curran, I Walker, J Ellis, C Konoby, S Kennedy and B Meehan, 2000. Absence of evidence for porcine circovirus type 2 in cattle and humans, and lack of seroconversion or lesions in experimentally infected sheep. Arch Virol, 145: 853-857.

Cságola A, D Cadar and T Tuboly, 2008. Replication and transmission of porcine circovirus type 2 in mice. Acta Vet Hung, 56: 421-427.

Chae C, 2005. A review of porcine circovirus 2-associated syndromes and diseases. Vet J, 169: 326-336.

Ellis JA, C Konoby, KH West, GM Allan, S Krakowka, F McNeilly, B Meehan and I Walker, 2001. Lack of antibodies to porcine circovirus type 2 virus in beef and dairy cattle and horses in western Canada. Can Vet J, 42: 461-464.

Hemingway J, B J Beaty, M Rowland, TW Scott and BL Sharp, 2006. The innovative vector control consortium: improved control of mosquito-borne diseases. Trends Parasitol, 22: 308-312.

Lorincz M, A Csagola, I Biksi, L Szeredi, A Dan, T Tuboly, 2010. Detection of porcine circovirus in rodents. Acta Vet Hung, 58: 265-268.

Otake S, SA Dee, RD Moon, KD Rossow, C Trincado and C Pijoan, 2003. Evaluation of mosquitoes, Aedes vexans, as biological vectors of porcine reproductive and respiratory syndrome virus. Can J Vet Res, 67: 265-270.

Schulze C, G Neumann, I Grutze, A Engelhardt, C Mirle, F Ehlert and A Hlinak, 2003. Porcine circovirus type 2-infection in an European wild boar (*Sus scrofa*) in the state of Brandenburg, Germany. Deut Tierarztl Woch, 110: 426-428.

Shi KZ, M Wen, B J Zhou, KG Wang and DS Wang, 2009. Isolation and identification of the porcine reproductive and respiratory syndrome virus (PRRSV) from *Culex tritaeniorhynchus* (Diptera: Culicidae) in pig farms. Acta Entomol Sin, 52: 509-513.

Yang CY, TC Chang, HC Su, PL Wang, LJ Ye, JY Lin, YJ Lin and MT Chiou, 2007. Biological transmission of porcine circovirus type 2 in pig herds. Taiwan Vet J, 33: 66-71.