Seroprevalence of PCV2 in north eastern hill states of India

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ABSTRACT

This study describe the seroprevalence of porcine circovirus type 2 (PCV2) in north eastern hill (NEH) states of India. Information on PCV2 from NEH states is lacking. Serum samples (306) were collected from the pigs and different epidemiological parameters like age, sex, and litter size of drift or sounder, system of rearing and different clinical symptoms from November 2017 to May 2018. Samples were screened for PCV2 infection by commercially available ELISA kit (INGEZIM PCV IgG and INGEZIM PCV IgM, Ingenasa, Madrid, Spain). Out of 306 serum samples tested, 151 samples were positive for PCV2 infection. An overall PCV2 prevalence of 49.35% was recorded in NEH states of India. Highest prevalence was found in Sikkim (94%) and lowest prevalence in Nagaland (4%). This study revealed that PCV2 is prevalent in all the north eastern states of India which necessitates the need for implementation of prevention and control measures.

Key words: ELISA, NEH, PCV2, Pig, Seroprevalence

The pig population in India is 8.8 million, which is 1.09% of the world’s pig population of 967.38 million (FAOSTAT 2017). Pig population contributed by the north eastern hill (NEH) states are Manipur (2.69%), Mizoram (2.38%), Meghalaya (5.27%), Nagaland (4.89%), Tripura (3.52%) and Sikkim (0.29%) (19th Livestock Census 2012). The smallholder pig farming system is always facing challenges and thus offering ample opportunities for its improvement for better livelihood (Haldar et al. 2017).

Porcine circovirus disease (PCVD) is one of the most significant diseases that affect industrial pig production worldwide (Allan and Ellis 2000). Porcine circovirus type 1 (PCV1) is non-pathogenic whereas porcine circovirus type 2 (PCV2) is pathogenic in nature. PCV1 and PCV2 belong to the Circoviridae family and Circovirus genus. The members of Circoviridae family are small (17 nm), non-enveloped, icosahedron shaped viruses with a single-stranded covalently closed circular negative stranded deoxyribonucleic acid (DNA) molecule. Pigs are susceptible to PCV2 infection which leads to morbidity and mortality resulting in considerable economic losses to pig industry. A PCV2 prevalence study in NEH states is lacking. In India, only scanty studies are being conducted to track the epidemiological pattern of PCV2. Hence, an epidemiological investigation is necessary. The aim of this study was to investigate the seroprevalence of PCV2 in all NEH states and to understand the epidemiology of PCV2 infection which will ultimately help in devising control strategy against the disease. The NEH states are sharing boarders with Tibet, China, Bhutan, Myanmar, Bangladesh and Nepal. Thus, prevalence studies of major viral diseases of swine in these states are important.

MATERIALS AND METHODS

Population screened: The study was conducted in Mizoram, Manipur, Meghalaya, Nagaland, Tripura, Arunachal Pradesh and Sikkim which are having subtropical climate and the study period was from November 2017 to May 2018. Blood samples without an anticoagulant were randomly collected from a representative subset of the pig population and data were categorized under sex, age, size of drift or sounder, management system followed. Blood samples were transported to laboratory and serum was separated by centrifuging at 2,500 rpm for 5 min and kept at -20°C until use.

Laboratory testing of the samples: All the serum samples were screened for PCV2 infection by using commercially available ELISA kit (INGEZIM PCV IgG and INGEZIM PCV IgM, Ingenasa, Madrid, Spain). Out of 306 serum samples tested, 151 samples were positive for PCV2 infection. An overall PCV2 prevalence of 49.35% was recorded in NEH states of India. Highest prevalence was found in Sikkim (94%) and lowest prevalence in Nagaland (4%). This study revealed that PCV2 is prevalent in all the north eastern states of India which necessitates the need for implementation of prevention and control measures.
available ELISA kit (INGEZIM PCV IgG and INGEZIM PCV IgM, Ingenasa, Madrid, Spain) as per manufacturer’s instruction. Results were recorded as IgM positive or IgG positive or both IgM and IgG positive.

Seroprevalence study: Seroprevalence under different category were calculated using the formula:

\[
\text{Prevalence (P)} = \frac{\text{No. of animals found positive for either IgM or IgG or both IgM and IgG}}{\text{Total number of animals tested in that category}} \times 100
\]

Statistical analysis: Statistical analysis was done using Statistical Package for the Social Sciences (SPSS version 20, 2011). Different groups were analysed using Chi-square test \((\chi^2\) test). Recorded Chi square statistic and significance noted at 5% level.

RESULTS AND DISCUSSION

The present study revealed that seroprevalence of PCV2 in NEH states was 49.35%. Previous studies reported the prevalence of PCV2 in some of the NEH states. Kikon et al. (2017) had reported 51.57% seroprevalence of PCV2 in Nagaland from 223 samples tested whereas Mukherjee et al. (2018) had reported 96.2% seroprevalence of PCV2 in Meghalaya from 1,899 samples tested. The high prevalence rate of PCV2 may be due to the stable nature of virus that can be shed through oro-nasal secretions, faeces, urine, colostrum, milk and semen (Patterson et al. 2015).

Disease outcome is related to immunological status, stressors, co-infection and virus load (Duy et al. 2013). Further, they had reported that the main diseases associated with PCV2 are systemic involvement showed a higher prevalence (88.24%) whereas samples from animals with no clinical symptoms showed prevalence of 34.33% (Table 4). PCV2 is considered to be the key agent in the development of a number of diseases like enteric form, respiratory form, systemic form, mange, all of which have been reported to be common in free-range pigs and their offspring. But, Correge (2001) reported that higher prevalence of PCV2 in Arunachal Pradesh had reported that high density of pig population in some districts and unorganized type of pig farming leads to seroprevalence of viral infection.

Size of drift or sounder: PCV2 prevalence was more in group where 5–10 animals were kept (60.47%) and lowest in group a single animal was kept (38.64%) (Table 2). Similar observations were noticed by Fraile et al. (2009) who opined that size and location of the farm has significant effects on the risk of PCV2 infection in the piglets and the bigger the farm, the easier the transmission of PCV2.

Sex wise: The seroprevalence of PCV2 was more in female animals compared to male animals (Table 3). Soria and Segales (2012) opined that, in a farm, the breeding animals are the ones that maintain the infection and transmit to their offspring. But, Correge (2001) reported that higher susceptibility of castrated males to PCV2 infection.

Symptoms wise: Samples from animals having multi-systemic involvement showed a higher prevalence (88.24%) whereas samples from animals with no clinical symptoms showed prevalence of 34.33% (Table 4). PCV2 is considered to be the key agent in the development of a number of diseases like enteric form, respiratory form, systemic form, post weaning multi-systemic wasting syndrome (PMWS).

Table 1. State wise PCV2 seroprevalence

| State                | Samples | IgM | IgG | IgM+IgG | Total | %   |
|----------------------|---------|-----|-----|---------|-------|-----|
| Tripura              | 14      | 0   | 4   | 1       | 5     | 35.71 |
| Mizoram              | 87      | 1   | 39  | 7       | 47    | 54.02 |
| Manipur              | 103     | 0   | 45  | 5       | 45    | 43.69 |
| Nagaland             | 25      | 0   | 0   | 1       | 4     | 100  |
| Meghalaya            | 21      | 0   | 2   | 2       | 4     | 19.05 |
| Arunachal Pradesh    | 6       | 0   | 2   | 0       | 2     | 33.33 |
| Sikkim               | 50      | 0   | 47  | 0       | 47    | 94   |
| Total                | 306     | 1   | 139 | 11      | 151   | 49.35 |

Table 2. Drift/Sounder size PCV2 seroprevalence

| Size of Drift or Sounder | Samples | IgM | IgG | IgM+IgG | Total | %   |
|--------------------------|---------|-----|-----|---------|-------|-----|
| Single                   | 54      | 1   | 15  | 6       | 22    | 40.74 |
| 2 to 5                   | 101     | 0   | 35  | 5       | 40    | 39.6 |
| 5 to 10                  | 43      | 0   | 26  | 0       | 26    | 60.47 |
| Above 10                 | 108     | 0   | 63  | 0       | 63    | 58.33 |
| Total                    | 306     | 1   | 139 | 11      | 151   | 49.35 |

\[\chi^2_{23}, 5.2868; \chi^2_{14}, 4.4677; \chi^2_{23}, 7.3248. \text{Significant at 5\% level (P<0.05).}\]
and porcine dermatitis and nephropathy syndrome (PDNS), porcine respiratory disease complex (PRDC), proliferative and necrotizing pneumonia (PNP) with clinical signs of weight loss/emaciation, tachypnoea, dyspnoea, icterus and pallor in pigs of weaning and post-weaning age (Segales 2012). Lymphadenomegaly may also be present (Afolabi et al. 2017). Association of PCV2 with reproductive disorders like mummified foetus, abortion, repeat breeding, still birth and necrotizing myocarditis of the foetus had been reported (Salogni et al. 2016). Baro et al. (2015) narrated about the intestinal disorders in growing and finishing pigs associated with PCV2 referred as PCV2-enteric disease (PCV2-ED). Anoopraj et al. (2014) described about PCV2 etiology in pneumonia and wasting disease of pigs. Cantile and Youssef (2016) described that PCV2 produces nonsuppurative or granulomatous encephalitis with gliosis. PCV2 is also responsible for skin lesions of red to purple colour in affected pigs and can cause PDNS (Segales 2012). Coming to the asymptomatic animals, the prevalence was 34.33%. Segales (2009) reported PCV2 positive animals in an apparently normal farm with no symptoms of the disease and concluded that the dynamics of the PCV2 infection can be very similar on farms with or without disease. Similarly, Shen et al. (2010) could detect PCV2 viremia in healthy sows and their pre-suckle piglets.

**Age wise:** The highest seroprevalence (79.66%) was recorded in 6–12 months age group and lowest was in 3–5 months age group (43.59%) (Table 5). The seroprevalence was more in growers and adult animals. This was in agreement with the earlier reports (Csank et al. 2011).

**Management system:** Extensive system of management showed 75% and intensive system of management showed 45.78% of PCV2 seroprevalence (Table 6). There is a strong link between the occurrence of diseases and pig production systems and farm scales. Smallholder pig production systems are usually linked to poor hygiene and low biosecurity with few barriers to potential contacts between the pigs, humans and wildlife. This facilitates disease transmission from wildlife to pig, pig to pig and pig to human. The reasons for more prevalence in extensive system may be due to lack of observation leading to delayed diagnosis of serious diseases, lack of investment in pig health by their owners, uncontrolled movement of pigs, difficulty in applying preventive or control measures, contact with wild pigs, pigs from other herds and carcasses of pigs (Thomas et al. 2013).

This comprehensive study on PCV2 infection revealed an overall seroprevalence of 49.35% in NEH states of India. The present study warrants in depth studies in this region involving larger pig population as well as devising control measures to prevent the occurrence of this economically important viral disease of pigs.

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**REFERENCES**

Allan G M and Ellis J A. 2000. Porcine circoviruses: A review. *Journal of Veterinary Diagnostic Investigation* 12: 3–14.

Anoopraj R, John J K, Sethi M, Somvanshi R and Saikumar G. 2014. Isolation and identification of porcine circovirus 2 from cases of respiratory disease and postweaning multisystemic wasting syndrome in pigs. *Advances in Animal and Veterinary Science* 2(6): 365–68.
Baro J, Segalés J and Martínez J. 2015. Porcine circovirus type 2 (PCV2) enteric disease: An independent condition or part of the systemic disease? *Veterinary Microbiology* **176**: 83–87.

Cantile C and Youssef S. 2016. Nervous system. *Jubb, Kennedy and Palmer’s Pathology of Domestic Animals.* 6th edn. Elsevier. Pp 250–406.

Corre J. 2001. La Maladie de l’Amaigrissement du Porcelet (MAP). *Journées de la Recherche Porcine* **33**: 283–90.

Csank T, Pisl J, Pollakova J, Holoda E and Harvan M. 2011. Prevalence of porcine circovirus 2 infection in pig population in Slovakia. *Acta Virologica* **55**(3): 267–71.

Duy D T, Huong L T T and Bryant J E. 2015. Porcine circovirus in Vietnam: Ruminations on a puzzling swine virus. *Vietnam Journal of Preventive Medicine* **25**: 162.

FAOSTAT. 2017. http://faostat3.fao.org/home/E

Fraile L, Calsamiglia M, Mateu E, Espinal A, Cuxart A, Seminati C, Martin M, Domingo M and Segales J. 2009. Prevalence of infection with porcine circovirus-2 (PCV-2) and porcine reproductive and respiratory syndrome virus (PRRSV) in an integrated swine production system experiencing postweaning multisystemic wasting syndrome. *Canadian Journal of Veterinary Research* **73**(4): 308–12.

Garam G B, Bora D P, Borah B and Das S K. 2016. Seroprevalence of Rotavirus infection in pig population of Arunachal Pradesh. *Vetenary World* **9**(11): 1300–04.

Hadam K L H, Puro K, Bhattacharjee U, Sen A, Samir S, Ghatak G, Sanjukta R, Shakuntala and Rajkhowa D J. 2016. Expression of markers of innate immune response in indigenous pig of northeast India in comparison of crossbred and Hampshire. *Indian Journal of Hill Farming* **29**: 140–44.

Haldar A, Das D, Saha B, Pal P, Das S, Das A, Rajkhowa D, Hazarika A and Datta M. 2017. Smallholder pig farming for rural livelihoods and food security in north east India. *Journal of Animal Research* **3**: 471–81.

Kikon L J, Rajkhowa T K, Arya R S, Singh Y D and Ravindran R. 2017. Seroprevalence of porcine circovirus type-2 (PCV2) and molecular diagnosis of PCV2 associated reproductive failure in pig population of Nagaland. *Indian Journal of Veterinary Pathology* **41**: 79–83.

Mukherjee P, Karam A, Singh U, Chakraborty A K, Huidrom S, Sen A and Sharma I. 2018. Seroprevalence of selected viral pathogens in pigs reared in organized farms of Meghalaya from 2014 to 16. *Veterinary World* **11**: 42–47.

Nath B G, Pathak P K, Ngachan S V, Tripathi A K and Mohanty A K. 2013. Characterization of smallholder pig production system: productive and reproductive performances of local and crossbred pigs in Sikkim Himalayan region. *Tropical Animal Health and Production* **45**: 1513–18.

Patterson A R, Madson D M, Halbur P G and Opriessnig T. 2011. Shedding and infection dynamics of porcine circovirus type 2 (PCV2) after natural exposure. *Veterinary Microbiology* **149**: 225–29.

Pegu S R, Sarma D K, Rajkhowa S and Choudhary M. 2017. Seroprevalence and pathology of important viral pathogens causing reproductive problems in domestic pigs of NE India. *Journal of Entomology and Zoology Studies* **5**: 1816–18.

Rajkhowa T K, Mohanrao G J, Gogoi A and Hauhnar L. 2016. Indian porcine reproductive and respiratory syndrome virus bears discontinuous deletion of 30 amino acids in nonstructural protein 2. *Virus Disease* **27**(3): 287–93.

Salogni, C, Lazzaro M, Giacomini E, Giovannini S, Zanoni M, Giuliani M, Ruggeri J, Pozzi P, Pasquali P, Boniotti M B and Alborali G L. 2016. Infectious agents identified in aborted swine fetuses in a high density breeding area: a three year study. *Journal of Veterinary Diagnostic Investigation* **28**: 550–54.

Segales J. 2009. Epidemiology and transmission of PCV2 and of porcine circovirus. https://www.pig333.com/articles/epidemiology-and-transmission-of-pcv2-and-of-porcine-circovirus_386/

Segales J. 2012. Porcine circovirus type 2 (PCV2) infections: clinical signs, pathology and laboratory diagnosis. *Virus Research* **164**: 10–19.

Shen H, Wang C, Madson D M and Opriessnig T. 2010. High prevalence of porcine circovirus viremia in newborn piglets in five clinically normal swine breeding herds in North America. *Preventive Veterinary Medicine* **97**(3–4): 228–36.

Soria S L and Segales J. 2012. Updated on the epidemiology of PCV2 and implications. https://www.pig333.com/articles/updating-on-the-epidemiology-of-pcv2-and-implications_6619/

Thomas L F, Glanville W A, Cook E A and Fevre E M. 2013. The spatial ecology of free-ranging domestic pigs (*Sus scrofa*) in western Kenya. *BMVC Veterinary Research* **9**: 46.

SPSS. 2011. IBM Statistics. Version 20.

19th Livestock Census. 2012. *All India Report*. Ministry Of Agriculture, Department of Animal Husbandry, Dairying and Fisheries, Krishi Bhawan, New Delhi. pp. 75–80.