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SERO-PREVALENCE OF PESTE DES PETITS RUMINANTS (PPR) VIRUS IN SHEEP AND GOATS IN PUNJAB PROVINCE OF PAKISTAN

H. A. KHAN, M. SIDDIQUE, M. J. ARSHAD[1], Q. M. KHAN[2] AND S. U. REHMAN

Department of Veterinary Microbiology, University of Agriculture, Faisalabad, [1]National Veterinary Laboratory, Islamabad, [2]National Institute of Biology and Genetic Engineering, Faisalabad, Pakistan

ABSTRACT

Peste des petits ruminants (PPR) is an acute febrile viral disease of sheep and goats characterized by mucopurulent nasal and ocular discharge, necrotizing and erosive stomatitis, enteritis and pneumonia. The disease is endemic in Pakistan and causes huge economic losses due to high rates of mortality and morbidity in infected sheep and goats. In the present study, 660 serum samples of sheep and goat were collected from 24 districts of Punjab Province of Pakistan. Competitive Enzyme Linked Immunosorbant Assay (cELISA) was used to detect the presence of antibodies in the serum against PPRV. Findings suggested that the sero-positive cases were significantly higher in sheep than in goats (51.29 versus 39.02%; P=0.002). The overall sero-prevalence of PPRV in small ruminants was 43.33%. Highest prevalence (35.71-100%) was observed in Southern districts, while no antibodies were found in serum from some of Northern and Eastern districts of the Punjab Province.

Key words: Peste Des Petits Ruminants, seroprevalence, small ruminants.

INTRODUCTION

Peste Des Petits Ruminants (PPR) is an acute and highly contagious viral disease of small ruminants. This disease is characterized by high fever, oculo-nasal discharge, pneumonia, necrosis and ulceration of mucous membranes and inflammation of gastrointestinal tract, leading to severe diarrhoea. PPR occurs in an epizootic form, it may have dramatic consequences with morbidity of 80-90% and mortality between 50 and 80% (Lefevre and Diallo, 1990). The virus that causes PPR belongs to the morbilli virus group of the paramyxoviridae family. It is closely related to rinder pest virus which makes the PPR an important disease of small ruminants and has created tremendous problems due to its apparent similarity to rinder pest (Lefever and Diallo, 1990).

The transmission of virus requires close contact between susceptible and infected animals in the febrile stage (Braide, 1981). The discharges from eyes, nose, mouth and the loose faeces contain large amounts of the virus. Fine infected droplets are released into the air from these secretions and excretions, particularly when affected animals cough and sneeze (Taylor, 1984; Bundza *et al*., 1988). Animals in close contact inhale the droplets and are likely to become infected.

The disease is endemic in the Arabian Peninsula (Taylor *et al*., 1990), the Middle East and in the Indian subcontinent (Shaila *et al*., 1996). The existence of Peste des petits ruminants has been recognized in Pakistan since 1991 (Amjad *et al*., 1996) as an epidemic in Punjab Province (Athar *et al*., 1995). Seroprevalence of PPR in unvaccinated sheep and goats in Southern, Central and Northern parts of Punjab Province of Pakistan has been described in the present paper.

MATERIALS AND METHODS

Study area

The study was conducted in the areas of Punjab Province of Pakistan designated as pockets of PPR by participatory disease surveillance teams (PDS). A total of 24 districts of Southern, Northern, Western, Eastern and Central districts of Punjab, Province of Pakistan were selected (Table 1). From randomly selected villages of each district, a total of 660 blood samples were collected (232 from sheep and 428 from goats) over a period of 10 months from December 2005 to September 2006. The area was divided into five different regions known to be PPR affected.

Blood was collected by jugular-vein puncture and left to clot overnight at 4°C. Serum was decanted into sterile tubes and kept on ice for transportation to the laboratory. In the laboratory, serum was centrifuged, transferred to screw capped serum tubes and stored at -20°C.

Competitive Enzyme Linked Immunosorbant Assay

The PPR competitive ELISA kit (collectively produced by Biological Diagnostic Supplies Ltd, Flow Laboratories and Institute for Animal Health Pirbright,

Surrey, England) was provided by the National Veterinary Laboratory, Islamabad, Pakistan. The kit is based on a standard competitive enzyme linked immunosorbant assay (cELISA) to determine the presence of anti-PPR antibodies in serum, as described by Singh *et al*. (2004). The test is based on the competition between the anti-H protein of PPR virus monoclonal antibodies and the serum samples for binding the PPR antigen (Libeau *et al*., 1992). The presence of antibodies to PPR virus in the serum samples blocks reactivity of the monoclonal antibodies which causes reduction in the expected colour following the addition of enzyme labeled anti-mouse conjugate and chromogen solution. The negative and positive cut-off values were used from the controls of the test procedure. The ELISA micro-plates were read using an immunosunkan reader with an inference filter of 492 nm. The reader was connected to a computer loaded with ELISA Data interchange (EDI) software, which was used to automate the reading and calculation of percentage of inhibition (PI) values. The optical density (OD) values were converted to percentage inhibition by using the following formula:

$$PI = 100 - \frac{\text{OD control/test serum}}{\text{(OD monoclonal control)}} \times 100$$

The samples with PI >50% were considered as positive.

Statistical analysis

The data were analyzed statistically by applying z-test to the proportion at 95% confidence interval.

RESULTS AND DISCUSSION

The overall PPR antibody seroprevalence recorded in sheep was 51.29% and in goats it was 39.02% which is significantly higher with P=0.002 at 95% confidence interval (Table 1). The highest seroprevalence was 100% in sheep and goat in districts of Cholistan, Bhawalpur and Bhawalnagar, followed by 90% (Chakwak), 79.17% (Bhakkar), 62.75% (Toba Tek Singh), 52.63% (Multan), 49.37% (Faisalabad), 40% (Jhang) 48.84% (Sahiwal), 42.86% (Mandi Bhaudin), 41.94% (Hafizabad), 35.71% (Kanewal), 37.50% (Dera Gazi Khan), 24.14% (Sargodha), 17.86% (Mianwali) and 13.7% (Attock). PPR antibodies were not detected in sheep population in Northern and Northwestern (Gujrat, Sargodha, Khushab, Attock) and Eastern parts of Punjab Province (Gujranwala, Gujrat, Lahore). In goat population, PPR antibodies were not detected in Northern and Eastern (Rawalpindi, Khushab, Gujrat, Gujranwala, Lahore, Okara, Pak Pattan), Central (Jhang) and Western parts (Dera Ghazi Khan) of Punjab Province.

This study provided valuable data on the serologic status of PPR in sheep and goats in Punjab province of Pakistan. Infection with PPRV was demonstrated in 18 districts of Punjab which are known to be the pockets of PPR and 6 districts in surroundings of the above mentioned districts. Variation in prevalence is probably related to the intensity of movements of nomads of small ruminants.

The presence of PPRV infection in Punjab has been reported already by Tahir *et al*. (1998) and Hussain *et al*. (1998). The overall prevalence of PPRV was 43.33% of the ruminant population. In this study, sheep population showed a relatively higher level of serum antibodies against PPR than goats. This may be attributed to a higher recovery rate and greater longevity of sheep verses goat which is in contrast to the serological profile reported by Taylor *et al*. (1990). According to anecdotal reports from the field veterinarians, the animal owners widely used the rinder pest virus vaccine to protect small ruminants against PPRV infection in some parts of Pakistan before rinder pest virus vaccination was stopped in year 1999-2000. This might be one reason for the higher percentage of PPRV-positive animals found in this study.

The overall prevalence of antibodies was higher in Southern and Western than Northern and Eastern parts of Punjab. This might be due to the nomadic grazing in Southern and Western parts of the Punjab. Climatic conditions and seasonal forage availability dictate grazing patterns in the area of southern and northern Punjab. Livestock migrate between alpine pastures and the Pothwar Plateau in the foothills of the Himalayas. The livestock spend April in subtropical and temperate forest grazing areas below 2,000 meters. They utilize the alpine areas from June to October, when low temperatures retard plant growth, and then herders descend towards the plains or low valleys. During winter, livestock graze in Pothwar scrub ranges, abandoned cultivated lands, or browse in valleys along water channels, roads and grazing grounds between agricultural fields (Dost, 1998). So, the nutritional status of the animals improves during the rainy season due to increase availability of fodder that may lead to the increased resistance. Wosu (1994) recorded similar observation in humid zone of Southern Nigeria.

These results also show that with help of competitive ELISA, the measurement of prevalence of antibodies to PPR is efficacious for the laboratory diagnosis. The rinderpest tissue culture vaccine was being used for the control of PPR in Pakistan since

Table 1: Sero-prevalence of PPR in different areas of Punjab, Pakistan

Sr.No	District/ Areas	Sheep T	Sheep +ve	Goat T	Goat +ve	Total	Total +ve	%
Southern districts								
1	Cholistan	20	20	10	10	30	30	100.00
2	Bahawalpur	14	14	10	10	24	24	100.00
3	Bhawalnagar	2	2	6	6	8	8	100.00
4	Multan	7	3	31	17	38	20	52.63
5	Khanewal	11	4	3	1	14	5	35.71
	Total	54	43	60	44	114	87	76.32
	Positive %		79.63		73.33			76.32
Northern districts								
6	Attock	23	0	50	10	73	10	13.70
7	Chakwal	-	-	20	18	20	18	90.00
8	Rawalpindi	-	-	13	0	13	0	0.00
9	Sargodha	14	0	15	7	29	7	24.14
10	Khushab	14	0	4	0	18	0	0.00
11	Mianwali	-	-	28	5	28	5	17.86
12	Mandi Bhaudin	9	3	5	3	14	6	42.86
13	Gujrat	6	0	6	0	12	0	0.00
	Total	66	3	141	43	207	46	22.22
	Positive %		4.55		30.50			22.22
Western districts								
14	D.G.Khan	8	6	8	0	16	6	37.50
15	Bhakkar	25	22	23	16	48	38	79.17
	Total	33	28	31	16	64	44	68.75
	Positive %		84.85		51.61			68.75
Eastern districts								
16	Gujranwala	4	0	6	0	10	0	0.00
17	Lahore	6	0	15	0	21	0	0.00
18	Hafizabad	4	4	27	9	31	13	41.94
19	Pakpattan	-	-	15	0	15	0	0.00
20	Okara	-	-	15	0	15	0	0.00
21	Sahiwal	16	10	27	11	43	21	48.84
	Total	30	14	105	20	135	34	25.19
	Positive %		46.67		19.05			25.19
Central districts								
22	Jhang	5	4	5	0	10	4	40.00
23	Faisalabad	17	10	62	29	79	39	49.37
24	T.T.Singh	27	17	24	15	51	32	62.75
	Total	49	31	91	44	140	75	53.57
	Positive %		63.27		48.35			53.57
	G. Total	232	119	428	167	660	286	43.33
	Positive %		51.29		39.02			43.33

95% C.I. for sheep 44.26-57.42, C.I. for goats 35.19-44.66, C.I. for overall population 39.92-47.61

many decades. As Pakistan was provisionally declared free from rinderpest, the vaccine production was stopped due to hindrance in the sero-monitoring of the rinderpest in global rinderpest eradication programme. There is very small vaccination to the small ruminants against PPR with the Nigerian strain provided by the FAO for ring vaccination. A large number of populations remain unvaccinated due to limited number of doses. In this study, the population of small ruminants under consideration seem to be lacking this facility due to far-flung areas and the ignorance of the farmers. About 70-80% of small ruminant population is, therefore, at risk of infection because very less doses of imported vaccine are available. Vaccination strategies for the control of PPR would need to account for the dynamics of sheep and goats population.

REFERENCES

Amjad, H., Q. U. Islam, M. Forsyth, T. Barrett and P. B. Rossiter, 1996. Peste des petits ruminants in goat in Pakistan. Vet. Rec., 139: 118-119.

Athar, H., Q. U. Islam, F. Azim, A. Shakoor, A. Maqbool and N. I. Chaudhary, 1995. An outbreak of peste des petits ruminants-like disease among goats in Punjab (Pakistan). Pakistan Vet. J., 15: 140-143

Braide, V. B., 1981. Peste des petits ruminantss. World Anim. Review, 39: 25-28.

Bundza, A., A. Afshar, T. W. Dukes, D. J. Myers, G. D. Susi and A. W. E. Becker, 1988. Experimental PPR (goat plague) in goats and sheep. Canadian J. Vet. Res., 52: 46-52.

Dost, M., 1998. Grassland and Pasture Crops: Country Pasture/Forage Resourse Profile. Available from: www.fao.org/ag/ AGP/AGPC /doc/ Counprof/ Pakistan.htm. Accessed 2006 June12.

Hussain, M., M. Afzal, R. Muneer, M. Ashfaq and E. U. Haq, 1998. An outbreak of peste des petits ruminants in goats in Rawalpindi. Pakistan Vet. J., 18: 224-226

Lefever, P. C. and A. Diallo, 1990. Peste des petits ruminants virus. Rev. Sci. Tech. Off. Int. Epiz., 9(4): 951-965.

Libeau, G., A. Diallo, D. Calvez and P. C. Lefever, 1992. A competitive ELISA using anti-N monoclonal antibodies for specific detection of RP antibodies in cattle and small ruminants. Vet. Microbiol., 31: 147-160.

Shaila, M. S., S. David, M. A. Foryth, A. Diallo, L. Goatley, R. P. Kitching and T. Barret, 1996. Geographical distribution and epidemiology of PPR viruses. Virus Res., 43: 149-153.

Singh, R. P., B. P. Sreenivasa, P. Dhar, L. C. Shah and S. K. Bandyopadhyay, 2004. Development of monoclonal antibody based competitive-ELISA for detection and titration of antibodies to peste des petits ruminants (PPR) virus. Vet. Microbiol. 98: 3–15.

Tahir, M. T., R. Ahmad, I. Hussain and M. Hussain, 1998. Counter-immunoelectrophoresis-A rapid technique for the diagnosis of peste des petits ruminants. Pakistan Vet. J., 18: 55-56

Taylor, W. P., 1984. The distribution and epidemiology of PPR. Prev. Vet. Med., 2: 157-166.

Taylor, W. P., S. Al-Busaidy and T. Barrett, 1990. The epidemiology of peste des petits ruminants in the Sultanate of Oman. Vet. Microbiol., 22(4): 341-352.

Wosu, L. O., 1994. Current status of peste des petits ruminants (PPR) disease in small ruminants. A review article. Stud. Res. Vet. Med., 2**:** 83-90.