PREVALENCE OF PESTE DES PETITS RUMINANTS (PPR) INFECTION IN SINDH PROVINCE OF PAKISTAN- A ONE YEAR STUDY

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خلاصير

موجو دہ ریسر چ پاکستان کے صوبہ سندھ میں بکریوں اور بھیڑوں میں پائے والی بیاری کاٹا (PPR) کی موجو دگی، اسباب اور کنٹر ول کرنے کے کار آمد طریقے معلوم کرنے کے لئے کی گئی ہے۔ جس کے دوران ای بیاری کی ۵۴؍2وباء (Outbreaks) کا جائزہ لیا گیاجو صوبہ سندھ کے ۹۲ میں سے ۱۲ اضلاع میں پائی گئی سب سے زیادہ وباء ۲۷۔ ۱۵ فیصد مٹیاری ضلع میں اور سب سے کم عرصے کی وباء عمر کوٹ ضلع میں ریکارڈ کی گئی جو کہ ۵ دن تھی اس کے علاوہ لیے عرصہ کی بیاری لاڑکانہ میں پائی گئی سب سے زیادہ وباء ۷۔ ۱۵ فیصد مٹیاری ضلع درجہ حرات میں اضافہ ۲ ۱۰ - ۲ سے ۲۰ سب ۲ ریکارڈ کی گئی ہو کہ ۵ دن تھی اس کے علاوہ لیے عرصہ کی بیاری لاڑکانہ میں پائی گئی جو کہ ۲۷ دن تھی۔ بیاری کی نشانیوں میں جسم کے درجہ حرات میں اضافہ ۲ ۲ ۱۰ - ۲ سے ۲۰ سند کار دیکار گئی۔ آئکھوں کی سوزش، آئکھوں اور ناک سے گاڑی مر طوبت، کھانی اور دست پائے گئے، منہ میں مسوڑوں پر لال سوجن

Abstract

The current study was carried out in twenty-six (26) districts of Sindh province of Pakistan, including six districts of Karachi for determination of different epidemiological parameters and nature of the Peste des petits ruminant virus (PPRV) in sheep and goats. A total of 847 outbreaks of Peste des petits ruminants (PPR) were investigated in twenty-six (26) districts of Sindh province during the year 2016. The highest 27.51% of PPR disease outbreaks were recorded in Matiari district. The shortest (05 days) length of PPR disease outbreak was observed in the Umerkot district, while the longest (62 days) outbreak duration was observed in district Larkana. Clinical investigation of the affected animals discovered high body temperature ranging between 102.2°F-107.6°F, conjunctivitis, mucopurulent nasal and ocular discharges beside with a cough and diarrhea. The necrotic mouth abrasions were found in all the sick animals. Red inflamed areas were found on the lower gums, the inner side of upper and lower lips with the dorsal side of the tongue. Statisticalanalysis of the ELISA results between different age groups with ANOVA reveals no significance. The results from the current study indicated that the PPR infection is endemic in the province. It was concluded from the study that there is the urgent need of a comprehensive strategy to improve the disease reporting through active surveillance activities, vaccination, disease awareness and quarantine guidelines are crucial to reduce economic losses and the socio-economic impact of livestock farmers especially subsistence farmers.

Keywords: Small ruminants' diseases, PPR, ELISA.

Introduction

Pakistan comprises three idiosyncratic geographic regions that are the northern Mountains with western ranges and high plateaus Indus plain and sandy desert (the Cholistan, and Thal). The altitude, climate and soil productivity control biodiversity and dictate animal/agriculture production systems. Livestock is asignificant component of agriculture, which is predominantly active in arid and hyper-arid zones with limited resources. Agriculture is one of the main sources of income in rural areas of thecountry, contributed 19.5 % to the gross domestic production and the largest (42.3%) employer of Pakistan's total labor force according to the 2016-17(Economic Survey of Pakistan). More than half the growth of the industry depends on livestock, which therefore makes it too important subject to neglect. According to the Pakistan Economic Survey, the livestock sector accounted for 58.33% of 3.43% total agricultural growth recorded during the 2016-17 financial year. Per year gross milk production from sheep and goat has estimated 39000 and 891000 tones and mutton 701000 tones during the year 2016-17(Economic Survey of Pakistan). Sheep & goat are important ruminants offering economic support to an incredible number of landless cultivators in dry to thehyper-arid ecological situation of Pakistan. Goat dairy products are recommended for its healthy value for human beings, especially children. Three kinds of the production system are discovered nationally according to the ecological situation, i.e. nomadic, transhumant and stationary or family businessZahur et al., (2014) Kunbhar et al., (2016), and Ahmed et al., (2016).

Peste des petits ruminants (PPR) is a word derived from theFrenchlanguage means the pest of small ruminants clinicalsimilarities with RinderpestWohlsein and Saliki;(2006). There is critical crevice in the study of disease transmission of PPR episodes because of poor observation, theabsence of accessible information because of ashortage of flare-ups report from the field and absence of the familiarity with PPR ailment among veterinarian and para-vets particularly working in private sector. The expansive based and controlled investigations of PPR in various geographic areas and period were not easy to decide the general circumstance and effect of PPR in Sindh province of PakistanNizamani et al., (2015). Serological reconnaissance gave a down to earth apparatus to quantify thelevel of infection course in the defenseless populace without immunizationMunir et al., (2009).

Due to deprived disease awareness and reporting in the province, there are serious gaps to design and implement disease control and eradication strategy. The current study was planned to fill the gap in this regard. The study was carried out in twenty-six (26) out of twenty-nine (29) districts of Sindh province of Pakistan, including six districts fromKarachi (east, west, south, central, Malir and Korangi) for the estimation of baseline data in the determination of different epidemiological parameters and nature of the Peste des petits ruminant virus (PPRV) circulating in the province. Three districts Tando Mohammed Khan, Kandkote and Kambar Shahdadkot were excluded from the study because of no outbreak report from these districts during the year 2016.

MaterialsandMethods

Outbreak investigation: A total 847 outbreaks of Peste des petits ruminants (PPR) was investigated in twentysix (26) districts of Sindh province during the year 2016.

Clinical observation: Clinical examination of the diseased animals was conducted in each outbreak. Rectal temperature, mouth examination for erosive /necrotic mouth abrasions, ocular and discharge, evidence of diarrhea were recorded and outbreak control measures were suggested to the livestock owners.

Epidemiological observations: Epidemiological information from all outbreaks like the index case, the length of outbreak, possible source, morbidity rate, mortality rate, asign of disease, recovery period and post-infection complication, etc., were recorded on prescribed proforma.

Collection, processing,and preservation of laboratory samples: Ocular, nasal and epithelial swab samples were taken from PPR suspected sheep and goat from each outbreak were pooled in one container (one suspected animal from each outbreak). These pooled swabs from 847 outbreaks were tested through IcELISA.Pair swabs were collected through inserting a sterile BD swab under the conjunctiva of each eye, into the oral cavity and deep into each nostril. The newsterile swab was used for each sampling. The cotton area of the swab was separated out from swab stick thru using sterile forceps and scissors.The swab was placed into a sterile Eppendorf tube having 1.5 ml of sterile phosphate buffer saline (PBS; 0.01 M, pH 7.4). After squeezing the swab was removed from 1.5 ml Eppendorf tube. Thesupernatant was collected after centrifugation at 10,000 rpm for 3-5 minutes at 39.2°F and stored at -94 °F for future analysis.

Confirmation of the PPR infection: Immuno-capture Enzyme-Linked Immuno-Sorbent Assay (Ic-ELISA) test was performed at Central Veterinary Diagnostic Laboratory Tando Jam. (CVDL) andNational Veterinary Laboratory, Islamabad as already applied by others Munir et al., (2009), Diop et al;(2005) and Abubakar et al., (2008).Thekit was used, which was conjointlyformed Biological Diagnostic Supplies (BDSL) and Flow Laboratories and CIRAD, EMVT, France.

The standardized reagents, assay protocol and manual were found with the kit. The cutoff point was determined by observing 04 antigen blank wells (B) having extreme optical density (OD) values were rejected, that is 02 wells with lowest OD values and 02 wells with highest OD values. The remaining four wells withintermediary OD values were taken into account. Cut off was calculated as two times the mean OD of these intermediate wells. Samples with high OD than the cut off were considered as positive, while samples with low OD as the cut off were considered as negative. A sample positive in the duplicate wells was regarded as positive and otherwise retested before considering the resultsAbubakar et al., (2008).

Results and Discussion

Epidemiological parameters

Disease length & sign of disease: Table-01 describes averagelengths and percentages of signs observed and calculated from eight hundred forty-seven (847) PPR disease outbreaks in twenty-six (26) districts of Sindh Province. The shortest length of PPR disease outbreak was observed in the Umerkot district, which was five (05) days, and the longest outbreak duration was observed in district Larkana, which was (62) days. It was concluded that the fever, nasal and ocular discharge was found in 100 % cases, while acough respiratory distress and diarrhea were less common in animals affected with PPR infection.

Clinical investigation of the affected animals discovered high body temperature ranging between 102.2°F-107.6°F, conjunctivitis, mucopurulent nasal and ocular discharges beside a cough and diarrhea. Red inflamed areas were found on the lower gums, the inner side of upper and lower lips and, the dorsal side of the tongue. The affected animals also showed signs of severe dehydration and hindquarters were soiled with diarrhea material.

Symptoms observed during outbreaks in Sindh province were also reported by many researchers i.e. Nizamani et al; (2015), Hamdy and Dardiri; (1976), Taylor et al., (1990), Roeder et al., (1994), Sande et al., (2011) and Almeshay et al., (2017). It was revealed from the study that due to PPR infection, 100 % pregnant animals were aborted in Ghotki, Jacobabad and Jamshoro districts, the same pattern was reported by Abubakar et al., (2008), while zero abortion rate was observed in Badin, Mirpur Khas, Sukkur, and Umerkot districts. The sequence of clinical signs development of PPR infection in sheep and goat was observed after a variable incubation period of 04-06 days, a sudden onset of fever, depression, loss of appetite, and nasal discharge, which becomes thicker and yellow in later stage, often forms a crust that blocks the nostrils resulting respiratory distress as reported earlier by Taylor et al., (1990), Roeder et al., (1994), Abubakar et al., (2008), Sande et al., (2011), and Almeshay et al., (2017). The eyes were also affected with glued eyelids together with discharge. There are augmented ulcerative mouth lesions found on the lower gums, dental pad, hard palate, cheeks, and tongue also reported by other researchers like Roeder and Obi. (1999). Sande et al., (2011), Zahur et al., (2014), Almeshay et al., (2017), and Shahab Uddin et al., (2017). Severe diarrhea with dehydration and weight loss was reported in some animals. Usually pneumonia usual in later stages of infection and abortionwere also reported from study area, which is similar to other part of country reported by Abubakar et al., (2008) and Zahur et al; (2014). The severe infection was observed in young stock and goats are affected more than sheep similar reported earlier from the country and other part of world Roeder and Obi (1999), Diop et al., (2005), Sande et al., (2011), Zahur et al; (2014), Kunbhar et al., (2016), Almeshay et al., (2017), and Shahab Uddin et al., (2017). No sign was reported in companion animals of sheep and goat during outbreaks in the study area. The male ELISA positive percentage from goat and sheep was 97.6% and 96.9% respectively, while in the female ELISA positive percentage from goat and sheep is 98.9% and 96.7% respectively. ANOVA analysis among various age groups revealed that no significant difference. These similar conclusions reflect that PPR is a regional problem in South Asia and required a collaborative regional effort for control.

Rate of abortion: It was revealed from the study that due to PPR infection, 100 % pregnant animals were aborted in Ghotki, Jacobabad, and Jamshoro districts, while 0% abortion rate was observed in Badin, Mirpur Khas, Sukkur and Umerkot districts as presented in Table-2.However investigations are needed to study other causes of abortion in sheep and goats during PPR outbreaks; FMD, brucellosis, chlamydia and physical trauma need to be considered.

Length in Days / Dist	Signs of Disease							
District	Days Av.	Ocular discharge %age	Nasal discharge %age	Fever %age	Necrotic Stomatitis %age	Cough %age	Respiratory distress %age	Diarrhea %age
Badin	12	100	100	100	96	89	81	83
Dadu	16	100	100	99	59	37	35	30
Ghotki	18	100	100	100	88	47	41	41
Hyderabad	16	100	92	80	55	97	23	37
Jacobabad	17	100	100	100	77	89	88	80
Jamshoro	19	100	100	100	84	88	85	78
Karachi City District (Six districts))	16	100	100	92	75	67	49	67
Khairpur	16	100	100	90	80	38	21	43
Larkana	62	100	100	97	94	48	39	72
Matiari	12	100	100	100	78	60	20	31
Mirpur Khas	15	100	100	100	100	38	16	62
Naushahro Feroze	14	100	100	100	57	83	20	46
Shaheed Benazir Abad	16	100	100	96	53	44	31	32
Sanghar	17	100	100	100	59	84	41	44
Shikarpur	20	100	100	100	87	63	66	40
Sujawal	24	100	100	95	91	66	48	78
Sukkur	21	100	100	100	91	55	52	61
Tando Allahyar	23	100	100	100	98	36	16	61
Tharparkar	25	100	100	92	91	49	36	77
Thatta	16	100	100	100	73	49	25	43
Umerkot	05	100	100	100	100	73	82	82

Table 1. Length of the outbreak and sign of PPR disease.

Table 2. Rate of abortion in the affected sheep & goat.

Sr.	District	Number of pregnant animals	Number of abortion	Rate of abortion (%)
1	Badin	14	0	0
2	Dadu	107	60	56
3	Ghotki	07	07	100
4	Hyderabad	22	11	50
5	Jacobabad	20	20	100
6	Jamshoro	08	08	100
7	Karachi six districts	16	04	25
8	Khairpur	02	01	50
9	Larkana	20	12	60
10	Mirpur Khas	06	0	0
11	Matiari	26	07	27
12	Naushahro Feroze	14	04	29
13	Sanghar	11	05	45
14	Shikarpur	10	05	50
15	Shaheed Benazirabad	14	04	29
16	Sujawal	15	08	53
17	Sukkur	06	0	0
18	Tando Allahyar	14	04	29
19	Tharparkar	53	22	42
20	Thatta	25	13	52
21	Umerkot	04	0	0

Disease outbreak detail: The highest number of PPR disease outbreaks were recorded in the Matiari district, which is 27.51%. This district is located on the national highway between the Shaheed Benazir Abad district (2.72% outbreaks) & the Hyderabad district (2.36% outbreaks). The second highest outbreaks were found is 25.27% in the Karachi City District, which is highly human populated and used to be one large district but now dived into six districts. The frequency of the PPR outbreaks in different districts are presented in Fig-1.



Age wise morbidity & mortality rate in goat:Data revealed from the current study that morbidity rate in goat ages less than 4 months was highest, which is 100 % in the Khairpur and Larkana districts followed by 89 % Shikarpur, 78.9% Sukkur and, 75% in the Matiari district. The lowest morbidity rate was observed 23 % in Naushahro Feroze and 40 % in the Thatta districts. Similarly, in the age between 04 to 12 months, the highest morbidity rate was also observed 90 % in the Larkana district followed by 71.4 % Thatta, 58.3 Sukkur and 56.9 in the Tando Allahyar, the lowest morbidity rate (13%) was found in Jamshoro, 15.3% in Hyderabad and 16 % in the Jacobabad district. Relatively less infection was observed in above one-yearold goats, the highest morbidity 59.5 % in the Tando Allahyar district followed by 52.6% Sukkur and 47.2 in the Khairpur district, while lowest (16.4 %) morbidity ratewas observed in the Sanghar, followed by 19.4 % in Mirpur Khas and 20.5 in the Shaheed Benazir Abad district. The 100 % mortality rate was observed in below 4 months old animal at Larkana district, while 0% mortality was observed in Mirpur Khas, Sanghar Tando Allahyar, Thatta, and Umerkot districts as shown in Table 03.

Disease Morbidity & Mortality rate age-wise Sheep: Table 04 shows the age wise morbidity and mortality in sheep in the study area. The morbidity rate in less than 04 months oldsheep was highest (90 %) in the Ghotki district followed by 65% in Tando Allahyar, 64.8 % in Thatta, 63.6% in Larkana and 63.8% in the Karachi city district. The 0% morbidity was observed in between 4 to 12 months old sheep at Jacobabad and Shaheed Benazir Abad district. Similarly, the highest mortality was observed in below 4 months old sheep, which is 100% at the Badin district followed by 85 % in Ghotki, 80 % in Larkana, and 70.9% in the Thatta district. The lowest (0%) mortality rate was observed in Badin, Dadu, Jacobabad, Sujawal, and the Tando Allahyar district. About ninety eight percent of the attended outbreaks were confirmed positive through ICELISA, which clearly shows the severity of PPR infection in Sindh province as reported earlier Nizamani et al; (2015), and Zahur et al; (2014). The mortality in some animals may be credited to secondary infection due to poor immunity after

PPR infection. However, the results of the IcELISA directed to the fact that morbidity and mortality are most likely due to PPR than the other issues during an outbreak. During study morbidity rate in less than 04 months old goat was highest, which was also reported by Abubakar et al., (2011), that is 100 % in Khairpur and Larkana districts followed by 89 % in Shikarpur, 79% in Sukkur and, 75% in Matiari districts.

Morbidity rate			Mortality rate					
<4 months	4-12 months	>12 months	<4 months	4-12 months	>12 months			
Badin District								
69	51.1	36	17.2	10.6	2			
	•	Dadu I	District	•				
42.4	48.8	45.1	86.1	46.8	44.6			
		Ghotki	District					
50	37.5	33.3	80	33.3	22.2			
	•	Hyderaba	d District	•				
69.7	15.3	25.5	43.5	53.8	14.3			
		Jacobaba	d District					
60.	16	29	45	58	18			
		Jamshoro	District					
67	13	22.5	40.5	43.8	24			
	I	Karachi City Dist	rict (six districts)					
32.6	50	37.2	80	70.6	50			
		Khairpur	District					
100	49.2	47.2	75	21.9	12			
	.,	Larkana	District					
100	90	44.3	100	60.5	10			
Mirpur Khas District								
75	48.6	19.7	50	5.6	0			
		Matiari	District					
64	38.2	30.4	46.9	42.9	8.6			
		Naushahro Fe	eroze District					
23.4	30	27.7	63.6	25	15.4			
	l	Sanghar	District					
47.6	42.4	16.4	70	7.1	0			
		Shikarpu	r District					
89	35	21	45	58	18			
		Shaheed Benazi	r Abad District					
57.1	51.6	20.5	31.3	12.5	33.3			
	•	Sujawal	District					
50	54.1	35.1	26.3	40	38.5			
	1	Sukkur	District	1				
78.9	58.3	52.6	80	28.6	50			
(2.2	5(0)	Tando Allah	iyar District	15.0	0			
03.2	50.9	39.5 Therperks	00./	15.2	0			
52.1	34.2	32.4	61 2	43.5	29.2			
	51.2	Thatta l	District	13.5	27.2			
40	71.4	38.5	100	10	0			
	, 1, 1	Umerkot	District	-0				
28.6	23.5	31.3	50	25	0			

Table 3.Age-wise morbidity & mortality rate in Goat (%)

The 100 % mortality rate was observed in below 4 months old animal at the Larkana district as reported by earlier by Zahur et al; (2014), while 0% mortality was observed in Mirpur Khas, Sanghar Tando Allahyar, Thatta, and the Umerkot districts. The morbidity rate in less than 04 months old sheep was highest (90 %) in Ghotki district, subsequently 65% in Tando Allahyar, 64.8% in Thatta, 63.6% in Larkana, and 63.8% in six districts of the Karachi. The reason of susceptibility among the young goat is long-term malnutrition due to the imbalance nutritional supplement for their growth as reported by Shahab Uddin et al., (2017). The amplified susceptibility of young sheep and goats could be due to poor immunity, malnutrition, and poor management system. Zero morbidity was observed in the sheep between 04 to 12 months old at Jacobabad and the Shaheed Benazir Abad district. Similarly, the highest (100%) mortality rate was observed in below 04 months old sheep at the Badin district subsequently 85 % in Ghotki, 80 % in Larkana, and 70.9% in the Thatta districts. While zero mortality was observed in Badin, Dadu, Jacobabad, Sujawal, and in the Tando Allahyar districts. Around 80% mortality was noticed during 10 - 12 days in acute outbreaks during study. Similar patterns for morbidity and mortality have also been reported by many researcher like Abubakar et al., (2008), Zahur et al; (2014), Nizamani et al: (2015) and Mailto et al., (2017). Roeder and Obi (1999) reported that in endemic areas, the mortality rates decline as low as 20% with continuous outbreaks. It was reported by Roeder and Obi (1999) and Chowdhury and Mutalib (2003) that Various factors like breed, age, production system, health condition, the exposure history and virulence of PPRV strain also play the role in varying morbidity and mortality in between 0 - 90%.

Morbidity Rate			Mortality Rate				
<4 months	4-12 months	>12 months	<4 months	4-12 months	>12 months		
	•	Badin	District				
40	71.4	38.5	100	10	0		
Dadu District							
60	67.9	35	66.7	27.3	0		
		Ghotki	District				
90	61	32.5	85	24	10		
		Jacobaba	d District				
15	7	0	1	0	0		
		Karachi City Dist	rict (six districts	5)			
63.8	54.8	14.4	70.4	26.1	22.2		
		Larkana	District				
63.6 57.1 13.9		80	30	26.3			
		Shaheed Benaz	ir Abad District				
37.5	0	0	0	0	0		
	•	Sujawal	District				
50	31.3	37.5	57.1	0	0		
	·	Thatta	District				
64.8	56.3	14.8	70.9	26.7	22.2		
Tando Allahyar District							
65	65 33.3 34.8		46.2	0	0		
		Tharpark	ar District				
60.7	18.1	16.7	41.2	46.2	6.3		

Table 4. Age Wise Morbidity and Mortality in Sheep

Laboratory analysis: Table 05 shows the result of IcELISA. Statisticalanalysis of the ELISA results between different age groups with ANOVA reveals no significance as presented in Table 06. Most of the results were achieved on the basis of the number of sample from various district. Findings are in agreements with earlier studiesAbubakar et al., (2008)., Zahur et al; (2014), Kunbhar et al., (2016).

Specie	Total Sample	Male			Female		
	Collected	Total	Positive	Negative	Total	Positive	Negative
Goat	785	246	240	06	539	533	06
Sheep	62	32	31	01	30	29	01
Total	847	278	271	07	569	562	07

Table 5: Results of IcELISA for the detection of PPRV antigen.

Table	6: Age-wise results of IcELISA for the detection of PPRV Antigen.	
ANOVA: Single	Factor SUMMARY	

0						
Groups	Count	Sum	Average		Variance	
Total Eliza positive	2	833	416.5	254184.5		
<4months	2	176	88	10082		
4-12 months	2	503	251.5	94612.5		
>12months	2	154	77	9248		
ANOVA						
Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	153850.5	3.0	51283.5	0.6	0.7	6.6
Within Groups	368127.0	4.0	92031.8			

Conclusion

The present circumstance of PPR infection in Sindh province is endemic and need comprehensive efforts to identify all the outbreak. Animal movement, the lack of veterinary facilities, the lack of quarantine and poor managemental practices are the main sources of PPR outbreaks in a susceptible population. There is agreement that this disease can occur in any season but an increase in outbreaks in the study area was recorded during the year 2016, in the late spring period and winter seasons (November and March), which is in the correlation to the movement of the animasl, lambing/kidding, and scarcity of natural feed resources. The outcomes demonstrated that PPR outbreaks were seen throughout the year because of extensive movements of the ruminants within and out of the province. Most of the outbreaks occurred usually every year during the draught period and the winter season in the province. There is a marked increase during the local or religious festivals like Eid-ul Azha, because of substantial movement for the purpose of sacrifice and close contact. The majority of farmers are convinced that the seasonal pattern is continuously changing and there is no longer a fixed time for aparticular weather condition. Rainfall has also decreased, which results in a scarcity of nutritional resources for animals as well as for humans.

It was concluded from the study that there is the urgent need of a comprehensive strategy to improve the disease reporting through active surveillance activities, vaccination, disease awareness and quarantine guidelines are crucial to reduce economic losses and the socio-economic impact of livestock farmers especially subsistence farmers. The prevention and control of TADs like PPR is a serious challenge due to the shortage of resource that is trained manpower, diagnostic facilities, early reporting, thermos-stable vaccine, lack of awareness, the loose management system, unrestricted movement and another economic constraint.

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Authors' contributions

Author-1 is the cross ponding author, and leader in the research.

Author-2 helped in field work and sample collection.

Author-3 helped in diagnostic lab.

Author -4 help in drafting manuscript.

Author-5 played supervisory role in designing study.

The authors of the manuscript declare that they have no competing interests.

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