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Current situation of Peste des petits ruminants (PPR) virus disease in some Egyptian Governorates in the period between years 2014 to 2019

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ABSTRACT

Peste des petits ruminants (PPR) is a highly contagious disease of domestic and wild small ruminants. According to Office international des epizooties (OIE), it is now recognized as a disease of great economic importance, causing huge annual losses in farm animals. The present study recorded the current situation of PPR virus disease in some Egyptian Governorates in the period between years 2014 to 2019. A total of 432 swabs (nasal, ocular and mouth), 100 tissue samples (lung, spleen, intestine and lymph node) were tested by immunocapture ELISA for detection of PPRV antigen revealing that 133 (30.7%) of swabs and 38 (38%) of tissues were positive. Out of 1339 serum samples were tested by competitive ELISA (cELISA) for detection of PPRV antibodies, 853 (63.7%) were positive. The results showed that goat more sensitive for infection of PPR virus than sheep and the viral antigen detection increase in winter more than other seasons. Also the results showed that percentage of positivity increase in middle Egypt than other localities of Egypt.

INTRODUCTION

Small ruminant pests are one among the foremost economically important diseases.. it is an acute transboundary viral disease, highly contagious in sheep and goats with a high morbidity and mortality of up to 100% and 90%, respectively (Diallo et al. 2007 Khan et al. 2008).

The causative agent of the disease is the pesti des petits ruminants virus (PPRV), which belongs to the genus Morbillivirus of the Paramyxoviridae family (Ictvdb 2006). Infection is transmitted through contact between infected and susceptible animals, by the oral or respiratory route, as the virus is present in high concentrations in tears and oral secretions (Maxic and Yousef 2007). PPRV has

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been reported in various parts of Asia and Africa (Bao et al. 2008) and has had a disastrous impact on the agricultural development of a region (Couacy Hymann et al. 2007).

Peste des petits ruminants virus is now endemic in most of the Sahara and sub-Saharan Africa, Turkey, the middle East and India (Dhar et al. 2002 Banyard et al. 2010 Abubakar et al. 2011). PPR recently reported in areas previously thought to be free of the disease, such as Algeria, Morocco, China, the Democratic Republic of the Congo (DRC), Sierra Leone, and Tajikistan (Wang et al. 2009 Kwiatek et al. 2007 Banyard et al. 2014).

The highly contagious nature of PPRV and the spread and movement of small ruminants creates a serious transboundary problem that inhibits trade and increases economic losses in affected areas, where small ruminants are often more important than livestock in the production of small ruminants food (Banyard et al. 2010).

The PPRV virus has spread from an infested parts of Africa to neighboring countries and has devastated the livestock industry (FAO, 2009).

Following the eradication of rinderpest, measles and PPRV were also identified as potential global eradication targets for the virus for possible eradication. Epidemiological surveillance is essential for any disease eradication program (Mariner et al. 2017).

The final diagnosis of PPRV infection is based on the isolation of the virus and the detection of the PPR antigen and its specific antibodies. Various conventional diagnostic tests have been used to detect and identify PPR, including FAT, AC ELISA, agar gel precipitation test, and immune-peroxidase (Sing et al. 2004).

Infection with PPR is confirmed by the detection of antibodies directed against the highly antigenic Morbillivirus N protein (Kwiatek et al. 2007). Using monoclonal antibodies, competitive ELISAs have been used

for the serological diagnosis of PPR (Choi et al. 2003 Sing et al. 2004 Dechamma et al. 2006).

Serological positivity of PPR virus has been observed in the past in large ruminant populations (Abraham et al. 2005 Balamurugan et al. 2012 2014)

In Egypt

The first outbreak of PPR occurred in Egypt in January 1987 in goats in Kafr Hakim, Embaba, Giza Governorate, where 30% of the animals died and the PPR virus was isolated in Vero cells from lymph nodes and the spleen and virus isolated using the direct fluorescence antibody technique (FAT) (Ikram et al. 1988). An outbreak in 2006 in Aswan province again showed that infected goats are occasionally asymptomatic, while others develop severe clinical disease (El Hakim, 2006). Nahed et al. (2010) reported the reappearance of PPRV in some Egyptian governorates. The PPRV, which has been reported and circulated in Egypt for the last decade, belongs to line 4 (Mahmoud et al. 2017 Ahmed et al. 2021).

This study aimed to:

Throw some light on the current situation of PPR in Egypt

MATERIAL AND METHOD

1-Samples: From 2014 to 2019, different samples (lung, spleen, intestine and lymph node, nasal, oral and lacrimal swabs, and blood without anticoagulant) were collected from different species (sheep and goats) showing PPRV clinical signs (clinical cases of PPR showing signs including anorexia, mucopurulent nasal discharge, erosive and necrotic stomatitis and diarrhea) from 24 different Governorates of Egypt . Collected samples were submitted to the animal health Research Institute (AHRI) for virological examination. The samples were properly prepared according to (OIE Manual 2019). All data of samples were shown in **table (1)**

Table (1): Number of collected samples from different Governorates in Egypt

Governorates	No.of serum	No. of swabs	No.of tissues
Cairo	-	14	4
Giza	14	29	13
Qalyobia	24	17	5
Menofeya	50	20	30
Gharbia	-	3	-
KafrElsheigh	13	25	4
Behera	10	26	9
Demiatta	2	1	1
Dakahlia	53	124	11
Sharkia	10	14	-
Alexandria	22	16	2
Port Said		3	2
Ismailia	7	4	-
Sues	9	-	-
Fayoom	8	-	-
Beni-Suef	25	44	-
Qena	5	3	-
Menia	20	9	4
Aswan	6	11	-
Assiot	-	6	-
Red Sea	14		
Matrooh	978	54	10
South Sainai	3	3	4
New Vally	66	6	1
Total	1339	432	100

Detection of PPR antigen by Immunocapture ELISA:

Immunocapture ELISA kit for detection of PPRV antigen in the tissue samples, buffy coat and swabs that produced by ID Vet Innovative Diagnostics.

This sandwich ELISA is designed to detect PPRV in different matrix. It uses an anti-

nucleoprotein (N) capture antibody and an anti -N monoclonal HRP antibody for revelation. It can be used on live animal or post-mortem diagnosis. Viral antigen being located mainly in the epithelial cells of the digestive, respiratory, and urinary tracts (Abubakar et al. 2011 Couacy- Hymann et al. 2009 Muhammad et al. 2012). The kit used as its enclosed in-

structions.

3- Competitive ELISA for detection of PPR Viral antibody

c-ELISA kit (ID Vet Innovative Diagnostics) was used to detect the antibody against the nucleoprotein of peste des petits ruminants virus in the serum samples collected from clinically affected and apparently healthy animals. This kit was used for detection of antibodies of PPR virus. The micro plates were percolated with PPR nucleoprotein (NP).

All procedure was carried according to the instruction of the manual included with the kit. Tested sera presenting a competition percentage $\leq 50\%$ are considered to be positive, $> 50\%$ and $\leq 60\%$ are considered to be doubtful and $> 60\%$ are considered to be negative (Choi

et al. 2005).

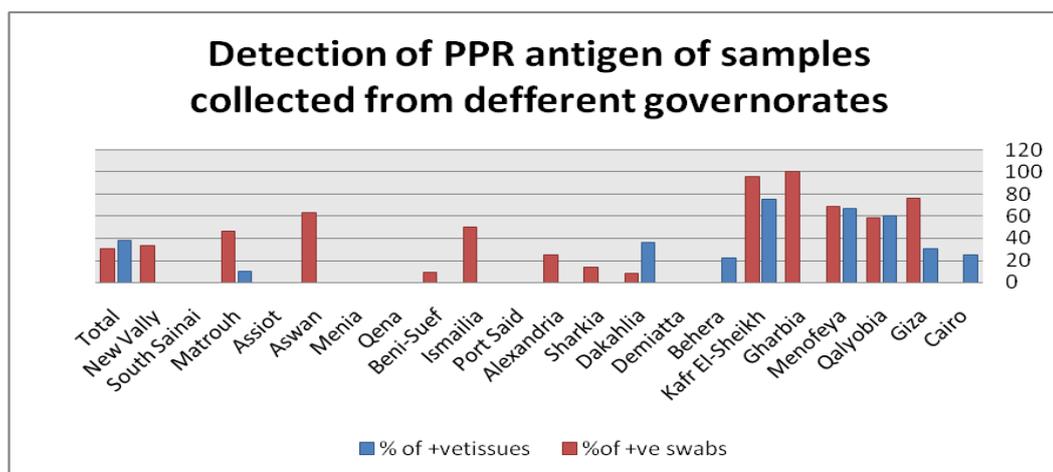
RESULTS

1-Detection of PPRV antigen by immunocapture (IC-ELISA)

Detection of PPRV antigen in 432 swabs and 100 tissue samples by IC- ELISA revealing that 133 (30.7%) of swabs and 38 (38%) of tissues were positive as shown in (table 2 and graph 1)

Table (2): Detection of PPR antigen in samples collected from different Governorates

Governorates	Swabs			Tissues		
	Total no of swabs	Positive	% of positive	Total no of tissues	positive	% of positive
Cairo	14	-	0	4	1	25
Giza	29	22	75.86207	13	4	30.76923
Qalyobia	17	10	58.82353	5	3	60
Menofeya	26	18	69.23077	30	20	66.66667
Gharbia	3	3	100	0	0	0
Kafr El-Sheikh	25	24	96	4	3	75
Behera	26	-	0	9	2	22.22222
Demiatta	1	-	0	1	0	0
Dakahlia	118	10	8.474576	11	4	36.36364
Sharkia	14	2	14.28571	0	0	0
Alexandria	16	4	25	2	0	0
Port Said	3	0	0	2	0	0
Ismailia	4	2	50	0	0	0
Beni-Suef	44	4	9.090909	0	0	0
Qena	3	0	0	0	0	0
Menia	9	0	0	4	0	0
Aswan	11	7	63.63636	0	0	0
Assiot	6	-0	0	0	0	0
Matrouh	54	25	46.2963	10	1	10
South Sainai	3	-	0	4	0	0
New Vally	6	2	33.33333	1	0	0
Total	432	133(30.7%)	30.78704	100	38(38%)	38



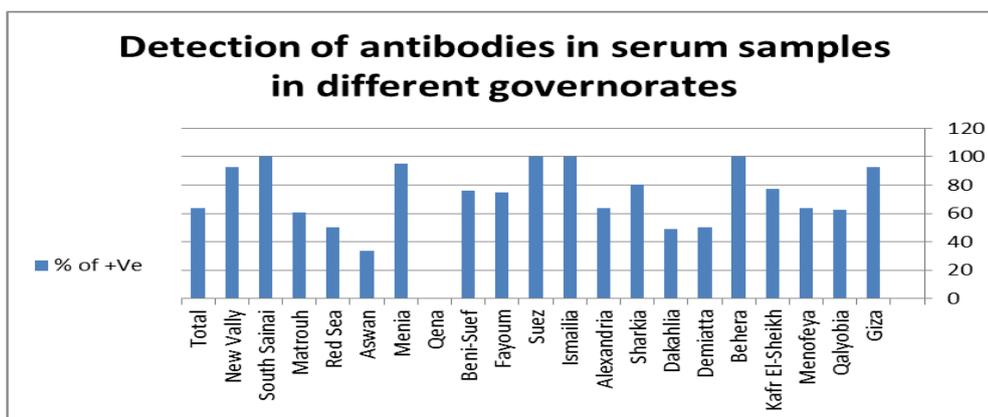
Graph 1: Detection of PPR antigen in samples collected from different Governorates.

Detection of PPRV antibodies by ID Screen Competition ELISA

Out of 1339 serum samples 853 samples were positive by percentage 63.7% as shown in (table 3 and graph 2).

Table (3): Detection of PPR antibodies in serum samples from different Governorates

Governorates	Total No of serum	No of positive samples	positive %
Giza	14	13	92.85714
Qalyobia	24	15	62.5
Menofeya	50	32	64
Kafr El-Sheikh	13	10	76.92308
Behera	10	10	100
Demiatta	2	1	50
Dakahlia	53	26	49.0566
Sharkia	10	8	80
Alexandria	22	14	63.63636
Ismailia	7	7	100
Suez	9	9	100
Fayoum	8	6	75
Beni-Suef	25	19	76
Qena	5	-	0
Menia	20	19	95
Aswan	6	2	33.33333
Red Sea	14	7	50
Matrouh	978	591	60.42945
South Sainai	3	3	100
New Vally	66	61	92.42424
Total	1339	853	63.70426



Graph 2: Detection of PPR antibodies in serum samples from different Governorates.

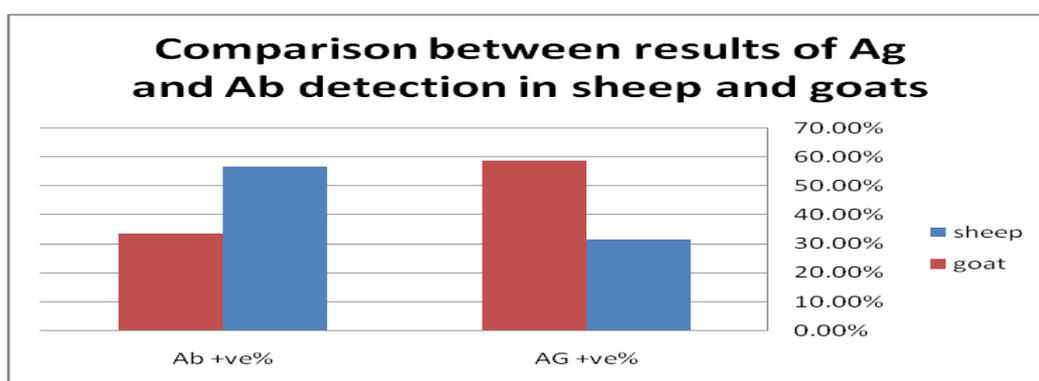
Results of antigen and antibody detection according to species

The percentage of antigen and antibodies in goat was higher than in sheep. The positive percent of Ag detection in goat was 33.5% and

31.4 % in sheep and the positive percent of Ab detection in goat was 58.5%% and 56.6 % in sheep as shown in (table 4 and graph 3).

Table (4) : Comparison between results of antigen and antibody detection in sheep and goat.

	sheep		Goat	
	Ag detection	Ab detection	Ag detection	Ab detection
No of samples	350	970	182	369
No of positive samples	110	637	61	216
%of positivity	31.4%	56.6%	33.5%	58.5%



Graph 3: Comparison between results of antigen and antibody detection in sheep and goats.

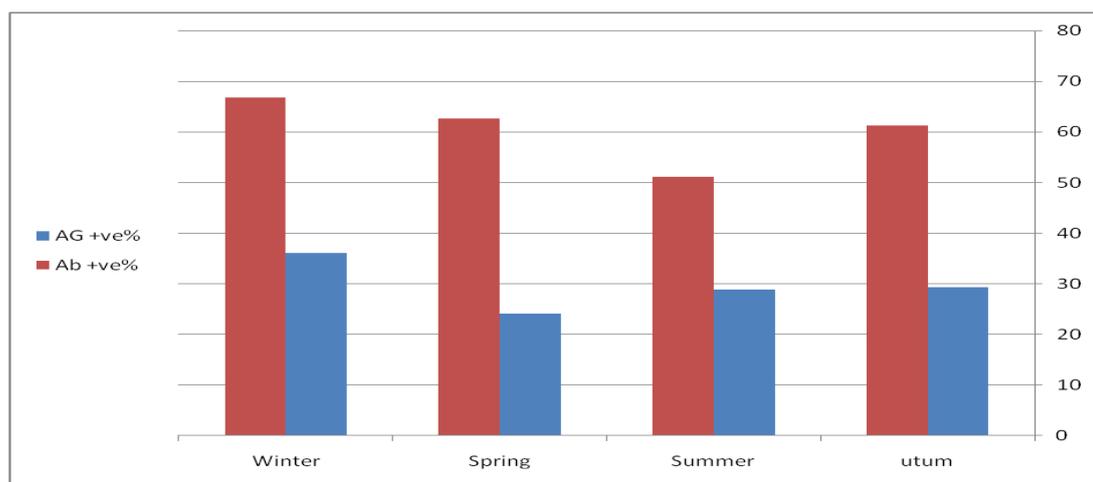
4- Percentage of positivity according to season

The highest percentage of antigen and antibodies were found during winter season by

percentage 36.1% of Ag detection and 66.8% of Ab detection as shown in (table 5 and graph 4)

Table (5) Percentage of positivity in different seasons

Winter		Spring		Summer		Autumn	
Ab detection	Ag detection						
+ve							
105(66.8%)	39(36.1%)	64(62.7%)	2(24.1%)	89(51.1%)	47(28.8%)	630(61.3%)	48(29.4%)



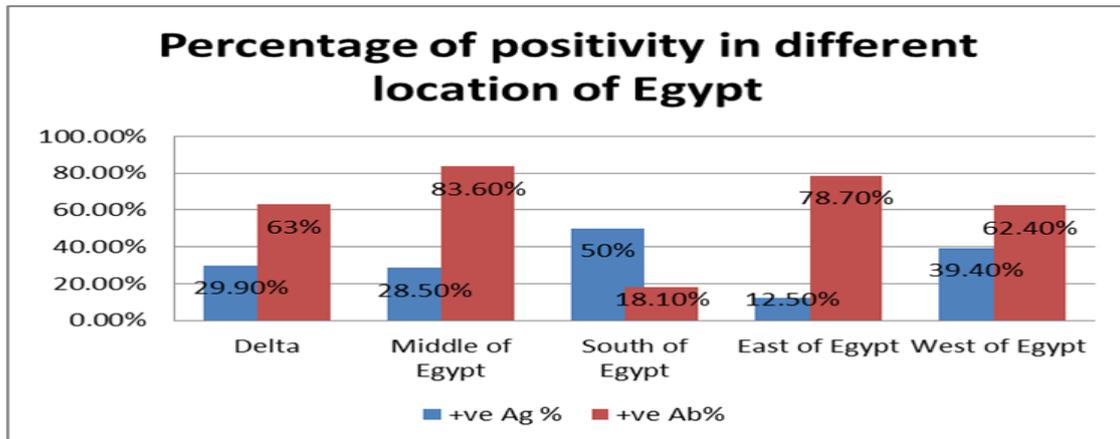
Graph 4: Percentage of positivity in different seasons.

5-Detection of PPR Ag and Ab in different localities in Egypt: The results showed high positivity of Ag detection in south of Egypt by

50% and high positivity of Ab detection in middle of Egypt by 83.6% as shown in (table 6 and graph 5)

Table 6: Percentage of positivity in different location of Egypt.

Governorates	Ag detection			Ab detection		
	No of samples	No of positive samples	%of positivity	No of sample	No of positive samples	%of positivity
Delta	304	91	29.9%	184	116	63%
Middle of Egypt	105	30	28.5%	61	51	83.6%
South of Egypt	14	7	50%	11	2	18.1%
East of Egypt	16	2	12.5%	33	26	78.7%
West of Egypt	71	28	39.4%	1044	652	62.4%



Graph 5 : Percentage of positivity in different location of Egypt.

DISCUSSION

Pesti des petites ruminant (PPR) is a disease of sheep and goats with high economical important. The large numbers of small ruminants raised in enzootic areas make PPR a serious disease that threatens the livelihoods of poor farmers (Diallo et al. 2007). In Egypt, small ruminants are one of the main sources of meat production, infection by the PPR virus causes high morbidity and mortality and leads to serious economic losses (ElAllawy et al. 1993).

The present study recorded the current situation of PPRV in various governorates between 2014 and 2019. Sheep and goats suffered fever, anorexia, diarrhea, expectorant nasal discharge, ocular discharge, dyspnea and ulcerative stomatitis, these concordants were recorded (Ahmed et al. 2005 Kul et al. 2008). The PPR must be confirmed by laboratory methods. Rapid diagnosis of the disease is made using an enzyme-linked immunosorbent assay (ELISA) immunocapture (OIE 2019). Commercial Immunocapture ELISA (Ic ELISA) can detect and quantities cell-free amounts of PPRV antigen (Mohammed et al. 2008 OIE 2019). Ic ELISA is a very sensitive and rapid test for the detection of PPR viral antigen in tissue samples, swabs and can detect a very low PPR viral antigen titer; (Housawil et al. 2004 Choi et al. 2005). In this study, detection of PPR viral antigen by IC ELISA revealed that out of 432 swabs and 100 tissue samples, 133 swab samples and 38 tissue samples were positive for PPR antigen with a per-

centage of 30.7 and 38 respectively (Table 2 and Graph 1). These results were consistent with those of (Abd ElRahim et al. 2010) who detected the PPR viral antigen in occulo-nasal smears by ELISA and found that 30/40 (75%) of the smears were positive. Other studies by (Abubakar et al. 2011) demonstrated PPR viral antigen in lungs and lymph nodes using a commercial Ic-ELISA.

Detection of the PPR antigen in different provinces showed that the highest percentage of positivity was found in Gharbia, then Kafr Elsheikh, then Giza and the lowest positive percentage was found in Sharkia, then Beni-Suef and Dakahlia. Competitive ELISA has been used as a standard technique for the detection of antibodies against PPR (Khan et al. 2008 Mohammed et al. 2008 Munir et al. (2009).

Detection of viral antibodies against PPR can confirm the diagnosis of PPR (Abid Mehmood et al. 2009) In this study, the serological diagnosis of antibodies against PPRV by ID Screen ELISA showed that out of 1339 serum samples, 853 samples were positive for PPRV antibodies with a positive percentage of 63.7% (Table 3, Figure 2) The results also showed that a high percentage of positivity was found 100% in Behera, Ismailia, Suez, South Seni, then Menia, Giza and New Vally. To differentiate between sheep and goats, 350 sheep and 182 goat samples 110 and 61 samples were positive for the detection of the PPR antigen with a percentage of 31.4 and 33.5%

and out of 970 serum samples from sheep and 369 serum samples from goat 637 and 216 samples were positive for the detection of PPR antibodies with a percentage of 56.6 and 58.5%, respectively (**Table 4 and Figure 3**). The results showed that goats are more susceptible than sheep agree with (**Al-Dubaib 2009 Mervat et al. 2015**).

The study also revealed that the percentage of positive detection of Ag in winter, spring, summer and autumn was respectively 36.1, 24.1, 28.8 and 29.4 % (**Table 5 graph 4**) and these results revealed that PPR virus infection increased in the winter. This is due to the increased activity of the virus in the low temperature and rainy season (**OIE 2019**). The results of antibody detection in different season in table 4 graph 3 showed that high number positive samples of Ab detection were recorded in winter, spring autumn and summer with a percentage 66.8, 62.7, 29.4 and 28.8% respectively and these results means that high positivity of antibody detection found in winter and this agree with results of antigen.

From table 5 graph 4 we found that the high percentage of positivity of Ag detection was found in south of Egypt followed by west of Egypt then Delta, Middle of Egypt end by East of Egypt with percentage of positivity 50, 39.4, 29.9, 28.5 and 12.5% respectively.

The excessive percentage in west governorates due to the fact (PPR) is a contagious transboundary sickness and this governorates are border provinces so the animals can migrate from neighboring nations to them and high percentage in Delta governorates due to wetness and migration of animals to markets between them . While the high percentage of positivity of Ab detection was found in Middle of Egypt followed by East of Egypt then Delta, West of Egypt end by south of Egypt with percentage of positivity 83.6, 78.7, 63, 62.4 and 18.1% respectively.

CONCLUSION

The present study concluded that this situation needs a lot of studies and regulations to control PPRV infection in Egypt.

Restriction of movement of sheep and goat population during appearance of PPRV infection should be applied.

Control by vaccination process is very important.

PPRV infection in Egypt needs continuous screening by reliable diagnostic systems.

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