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Epidemiology of Camel Calf Diarrhoea in Sudan: Seroprevalence of Camel Rotavirus Infection

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Abstract: The epidemiology of camel calf diarrhoea in four different areas in Sudan; River Nile (North), Gedarif (East), Sennar and Blue Nile (Central to South) and Kordofan (West) was studied. Data about the epidemiology of camel calf diarrhoea, its treatment regimen adopted by the owners was collected and analyzed. A total of 383 camel herds were investigated about the incidence of camel calf diarrhoea during wet and dry seasons over three years (2000-2002) in the focused areas. The overall morbidity rate of camel calf diarrhoea in the four areas of study was 83% while mortality and case fatality rates were 39.9% and 43.3%, respectively. The morbidity, mortality and case fatality rates of camel calf diarrhoea were found to be almost the same in the four areas focused during wet and dry seasons with slight increase during wet season. With regards to different treatment regimens adopted to diarrhoeic camel calves by the owners, 38.2% of cases were left without treatment, 58.51% received antibiotics while other drugs (symptomatic treatments, traditional medicines) constituted a very minor percentages. A serological survey was conducted using group A rotavirus antibody detection ELISA on 530 camel sera. The overall percentage of positive samples was 48.1%. Seropositivity was detected in all areas of study with slightly higher percentage in Sennar and Blue Nile States. The overall percentage of high antibody titer (4+) was 31.4% and 3+ was 22%. Most of the seropositive samples were collected from camels of 18-36 month of age and adult camels with slightly higher percentage in males than females (56.5% males and 43.5% females). A correlation was found between the seropositivity and the clinical status of diarrhoea. The highest percentage of seropositivity was found in clinically healthy camel calves (69.7%). The results showed the high prevalence of camel calf diarrhoea and the major role of rotavirus in the disease in Sudan.

Key words: antibodies, camel calf, diarrhoea, seroepidemiology, group A rotavirus

Introduction

Camels play an important role in the economy of Sudan. The estimates of camel population in Sudan make it one of the richest countries where approximately twenty percent of world camel population is kept (Schwartz, 1992). Calf mortalities are considered one of the major constraints to higher productivity in camels of which calf diarrhoea is regarded the major cause. Thirty percent morbidity and 100% mortality due to the disease was previously reported (Schwartz and Dioli, 1992).

In Sudan, Abbas *et al.* (1992) reported that camel calf diarrhoea affects about 33% of the neonates causing 23% mortality in northeast Sudan. According to Abbas *et al.* (1993), 43.4% of questionnaried groups of pastoralists in eastern Sudan complained from camel calf mortality while 7.4% of them considered it their disease priority.

Only few reports on viral causes of camel calf diarrhoea were published. Mahin *et al.* (1983) reported that using counter immunoelectrophoresis 27 out of 55 sera collected from 1-3 years old camels in Morocco were found positive for rotavirus antibodies. Elsayd *et al.* (1992) reported that high titers of antibodies against rotavirus were found in camel milk. Mohamed *et al.* (1998) was the first to report the detection of group A rotavirus in diarrhoeic camel calf faeces in eastern Sudan.

Generally there is a lack of detailed study on the role of rotavirus in camel calf diarrhoea. In this communication, the epidemiology and the detection of rotavirus antibodies in the different areas focused in Sudan are targeted.

Materials and Methods

Area of Study: The area of study selected represents River Nile State in the north, Gedarif State in the east, Sennar and Blue Nile States in the middle to South and North and West Kordofan States in the west of Sudan. Each of these areas were visited twice per year during dry season (February-May) and wet season (June-October) for three successive years (2000-2002) to collect data and serum samples from diarrhoeic, recovered and clinically healthy camel calves and adults.

Data Collection: A total of 383 camel herds were investigated in 186 field visits during dry season and 197 visits during wet season. The camel owners in the areas of study were interviewed. Data on the incidence of camel calf diarrhoea, morbidity, mortality and case fatality rates of the disease as well as seasonality, age and sex of the affected calves and the treatments applied by camel owners were collected over three years period. The number of camel herds visited for data collection in the areas of the study was 25 in River Nile state, 45 in Gedarif State, 53 in Sennar and Blue Nile states and 260 in North and west Kordofan states. The data was statistically analyzed.

Sample Collection: A total of 530 serum samples from diarrhoeic, as well as some recovered and clinically healthy calves and adult camels were collected. Blood was withdrawn from the jugular vein and serum was separated either by light centrifugation or by leaving the vaccutainer tubes overnight at room temperature. Serum samples were stored at –20°C until tested. The number of samples and their origin are presented in Table 1.

Sero-epidemiology of Rotavirus in Camels: A competitive ELISA kits for group A rotavirus antibody detection (Bio X diagnostics-Belgium) were used according to the manufacturer instructions.

Results

Incidence of Camel Calf Diarrhoea in Sudan: The incidence of camel calf diarrhoea in the four areas of study, seasonality, morbidity, mortality, case fatality rates and the treatments adopted by the owners were recorded and analyzed. Cases of calf diarrhoea were reported in 318 out of 383 camel herds giving an incidence of 83%. The overall morbidity, mortality and case fatality rates due to camel calf diarrhoea in the four areas of study were 83%, 39.9% and 43.3%, respectively (Fig.1). Epidemiological data were collected by interviewing camel herders in these areas, on the occurrence of camel calf diarrhoea during the three years of the study (2000-2002). It was noticed that there was no significant difference in morbidity, mortality and case fatality rates among the four different areas of study (P>0.05).

Table 1: Location, age, sex and source of serum samples collected for detection of rotavirus antibodies

Age/ month	Sex	States: River Nile State D	R	H	Gedarif State D	R	H	Sennar and Blue Nile State D	R	H	Kordofan State D	R	H	Subtotal D	R	H	Total
0-3 month	M	5	1	4	5	2	2	9	1	-	47	14	4	66	18	10	94
	F	5	2	1	8	2	2	11	-	-	42	17	-	66	21	3	90
>3-6	M	2	-	5	-	5	1	-	-	-	11	8	-	13	13	6	32
month	F	-	2	6	-	2	3	-	-	-	6	5	1	6	9	10	25
>6-9	M	-	1	2	-	1	-	-	-	-	5	1	-	5	3	2	10
month	F	-	-	2	-	2	-	-	-	-	1	4	3	1	6	5	12
>9-18	M	-	1	2	1	-	11	-	-	-	4	0	3	5	1	32	38
month	F	-	1	2	-	-	8	-	-	15	3	2	-	3	3	25	31
>18-36	M	-	-	3	-	-	2	-	-	-	-	-	120	-	-	125	125
month	F	-	-	9	-	-	1	-	-	28	1	-	-	1	-	38	39
Adult	M	-	-	2	-	-	2	-	-	-	-	-	1	-	-	5	5
	F	-	-	11	-	-	8	-	-	-	1	-	9	1	-	28	29
Subtotal	M	7	3	18	6	8	18	9	1	16	67	23	128	89	35	180	304
	F	5	5	31	8	6	22	11	-	43	54	28	13	78	39	109	226
Total		69			68			80			313			530			530

D: Diarrheic R: Recovered H: Healthy M: Male F: Female

Table 2: Detection of group A rotavirus antibodies in camel sera using ELISA

Age/ month	Sex	River Nile State +	2+	3+	4+	-	Gedarif State +	2+	3+	4+	-	Sennar/Blue Nile +	2+	3+	4+	-	Kordofan States +	2+	3+	4+	-	Total +	2+	3+	4+	-	T+	%+
0-3	M	-	-	1*	-	9	3	1	-	-	5	-	-	-	-	10	7	3	4	-	51	10	4	5	-	76	19	20
	F	1	-	1	1	5	3	-	-	-	9	3	-	-	-	8	3	6	2	1	48	10	6	3	2	70	21	23.1
>3-6	M	1	-	-	1	5	-	1	-	-	5	-	-	-	-	-	-	1	-	-	18	1	2	-	1	28	4	12.5
	F	-	2	-	-	6	-	-	1	1	3	-	-	-	-	-	-	1	1	-	9	-	3	2	1	18	6	25
>6-9	M	-	-	-	1	2	-	-	-	-	1	-	-	-	-	-	1	1	1	1	3	1	1	1	2	6	5	45.5
	F	-	-	-	-	2	-	-	-	-	2	-	-	-	-	-	1	1	-	-	4	1	1	-	-	8	2	2
>9-18	M	1	-	-	1	1	2	2	-	6	2	2	3	6	3	2	-	2	1	-	4	5	7	7	10	9	29	76.3
	F	-	-	1	-	2	-	1	1	4	2	2	1	3	7	2	-	-	1	-	4	2	2	6	11	10	21	67.7
>18-36	M	-	1	1	-	1	-	1	-	-	1	-	-	-	-	-	20	17	18	26	39	20	19	19	27	40	85	68
	F	-	2	2	3	2	-	-	1	-	-	11	4	3	5	5	1	-	-	-	-	12	6	6	8	7	32	82.1
Adult	M	1	-	-	-	-	1	-	-	1	-	-	-	-	-	-	-	-	-	-	-	2	-	-	1	-	3	100
	F	-	-	4	8	-	-	-	1	6	1	-	-	-	-	-	1	3	2	4	2	1	3	7	18	3	29	90.6
Sub-	M	3	1	2	3	18	6	5	-	7	14	2	3	6	3	12	28	24	24	27	115	39	33	32	40	15	14	47.5
total	F	1	4	8	12	17	3	1	4	11	17	16	5	6	12	15	6	11	6	5	67	26	21	24	40	94	111	48.9
Total		4	5	10	15	35	9	6	4	18	31	18	8	12	15	27	34	35	30	32	182	65	54	56	80	275	255	48.1

+, 2+, 3+, 4+, -: Degree of positivity according to optical density reading of test sample. F: Female M: Male T+: Total positive

Table 3: Resutls of ELISA for detection of group A rotavirus antibodies in sera collected from camel calves with different clinical status

Clinical status	No. of samples: +	2+	3+	4+	-	Total positive	Percentage of positive
Diarrhoeic	16	12	11	5	125	44	26.3
Recorvered	3	2	2	2	64	9	12.3
Healthy	47	39	43	73	88	202	69.7

+, 2+, 3+, 4+: Degree of positivty. -: Negative

Seasonality of Camel Calf Diarrhoea in Sudan: The seasonality of camel calf diarrhoea with regard to morbidity, mortality and case fatality rates during dry compared to wet seasons in three of the four areas of study is shown in Fig. 2.

During wet season, the morbidity, mortality and case fatality rates were 85.4%, 41.6% and 45.3%, respectively. On the other hand morbidity, mortality and case fatality rates during dry season were 80.4%, 37.9% and 41.2%, respectively. The statistical analysis of the morbidity, mortality and case fatality rates during wet and dry seasons showed that there was no significant difference between the different areas of the study (P>0.05).

Treatment of Camel Calf Diarrhoea: The treatment adopted to diarrhoeic camel calves by the owners during the study period in the four areas of study are presented in Fig. 3.

The percentage of calves left without treatment was 38.20% while antibiotics (tetracycline, penicillin and sulpha derivatives) were used

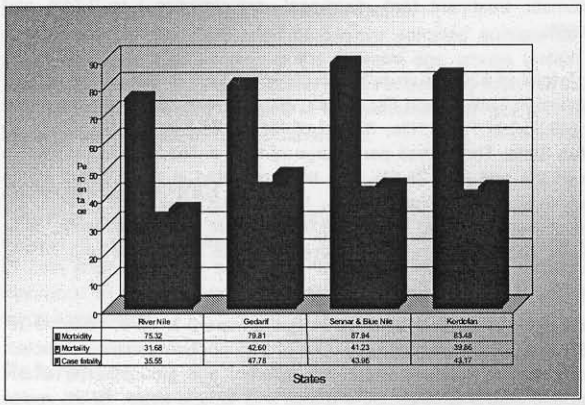

Fig. 1: Epidemiological parameters of camel calf diarrhoea infour different states

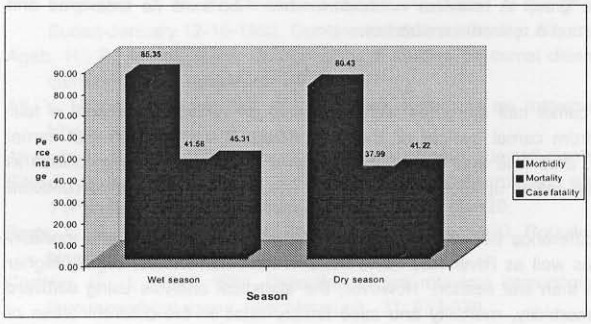

Fig. 2: Theepidemiological indices of camel calf diarrhoea in dry and wet season

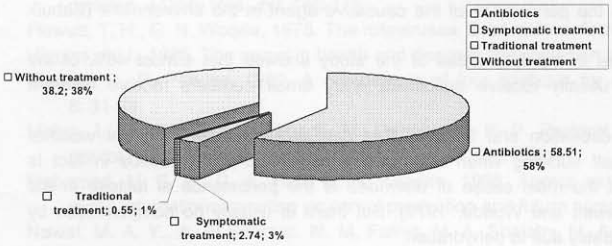

Fig. 3: Different regimens adopted by the owners for treating diarrhoeic camel calves

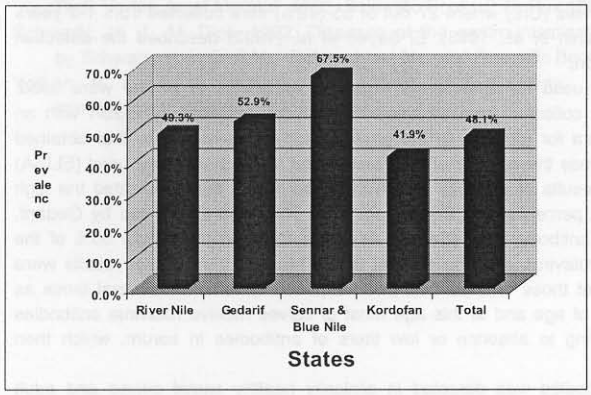

Fig. 4a: Sero-prevalence of group A rotavirus antibodies in different states

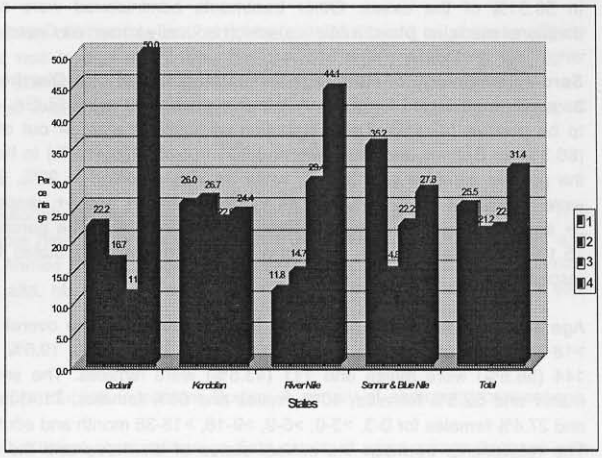

Fig. 4b: the distribution of degree of positivity of group Arctavirus antibodies in camel sera in the different states

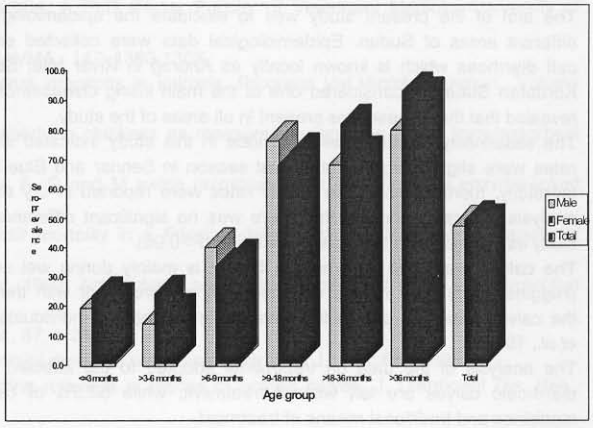

Fig. 5: The distribution of sero-prevalence of group A rotavirus in camel in different age and sex groups

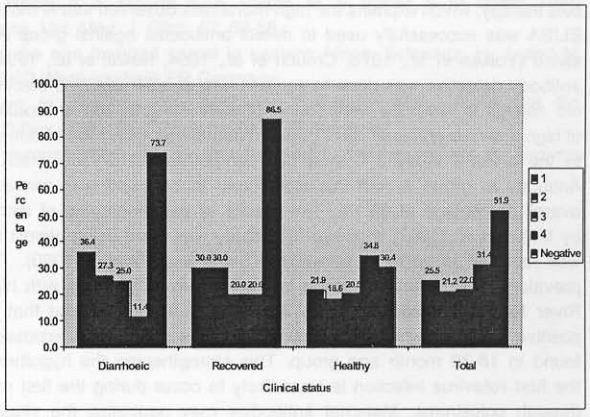

Fig. 6: Sero-prevalence and degree of positivity of group A rotavirus in camel calves with different clinical status

in 58.51% of the cases. Other treatments administered were symptomatic treatment (anti diarrhoeal, anthelmintics) in 2.74% and traditional medicine (*Acacia Nilotica*) which is locally known as Garad in 0.55%.

Sero-epidemiology of Rotavirus Associated Camel Calf Diarrhoea:: A total of 530 serum samples collected from camel calves as well as adults were tested for group A rotavirus antibodies using ELISA. As shown in Table 2 and Figures 4a, 4b, a total of 255 sera were found to be positive (48.1%). The distribution of positives was 34 out of 69 (49.3%) in River Nile, 37 out of 68 (54.4%) in Gedarif, 53 of 80 (66.3%) in Sennar and Blue Nile and 131 out of 313 (41.9%) in Kordofan State. The overall percentage of high antibody titers (4+) within the positive samples was 31.4%, while 3+ was detected in 22%, 2+ in 21.2% and + in 25.5%. The titers detected in the different states were 11.8%, 14.7%, 29.4% and 44.1% for +, 2+, 3+ and 4+, respectively in River Nile State. 24.3%, 16.2%, 10.8% and 48.6% for +, 2+, 3+ and 4+, respectively were detected in Gedarif State. The percentage of titers for 1+ to 4+ in Sennar and Blue Nile States were, 34%, 15.1%, 22.6% and 28.3%, respectively while in Kordofan States the titers detected for + to 4+ were, 26%, 26.7%, 22.9% and 24.4%, respectively.

Age and Sex Distribution of Positive Sampled Animals: The overall percentages of positive samples in age groups 0-3, >3-6, >6-9, >9-18, >18-36 month of age and adults were 15.7%, 3.9%. 2.7%, 19.6%, 45.9% and 12.5%, respectively. Out of 255 positive samples detected, 144 (56.5%) were males and 111 (43.5%) were females. The sex distribution of positives within the different age groups were, 47.5% males and 52.5% females, 40% males and 60% females, 71.4% males and 28.6% females, 58% males and 42% females, 72.6% males and 27.4% females for 0-3, >3-6, >6-9, >9-18, >18-36 month and adults, respectively (Fig. 5).

The relationship between the clinical status of diarrhoea and the seropositivity to group A rotavirus is shown in Table 3 and Figure 6. Out of 167 diarrhoeic camel calves tested, 26.3% showed detectable group A rotavirus antibodies while 12.3% of 73 recovered and 69.7% of 290 clinically healthy camel calves and adults were positive for group A rotavirus antibodies.

Discussion

The aim of the present study was to elucidate the epidemiology of camel calf diarrhoea with emphasis on rotavirus infection in four different areas of Sudan. Epidemiological data were collected either from camel owners or the pastoralists. It was noticed that camel calf diarrhoea which is known locally as *Khorag* in River Nile, Sennar, Blue Nile and Gedarif States while it is called *Reet* or *Sabba* in Kordofan States is considered one of the main killing diseases of camel calves up to 6 month of age. The analysis of the data collected revealed that the disease was present in all areas of the study.

The seasonality of camel calf diarrhoea in this study indicated slight difference between the areas focused. The morbidity and mortality rates were slightly higher during wet season in Sennar and Blue Nile as well as River Nile State while in Kordofan States, slightly higher morbidity, mortality and case fatality rates were reported in dry season than wet season. However, the statistical analysis using software analysis programs showed that there was no significant difference in morbidity, mortality and case fatality rates in the different areas of study as well as in the two seasons studied ($P>0.05$).

The calving seasons of camels in Sudan is mainly during wet season (July-October) with some calving during November to December (Higgins 1985). The results obtained was in agreement with the previous reports that the incidence of calf diarrhoea increases during the calving seasons due to the increase in susceptible individuals with the persistence of the causative agent in the environment (Babuik *et al.*, 1985).

The analysis of the data on treatments adopted to the affected calves in the four areas of the study showed that almost 40% of the diarrhoeic calves are left without treatment, while 58.5% of cases usually receive antibiotics. Very small numbers receive different medicines and traditional means of treatment.

It was noticed that most of the camel owners ignore the microbial causation and believes that calf suckling during the hot weather naturally causes calf diarrhoea and accordingly they used to stop calf suckling when diarrhoea is observed. This practice in fact is valuable in case of rotavirus induced diarrhoea, as it was proved that the main cause of diarrhoea is the persistence of lactose in the lumen causing osmotic drain attracting body fluids into the lumen (Flewett and Woode, 1978). But there is usually no compensation by fluid therapy, which explains the high mortalities observed that is most probably due to dehydration.

ELISA was successfully used to detect antibodies against group A rotavirus in human and various species including bovine, chicken and swine (Yolken *et al.*, 1978, Crouch *et al.*, 1984, Nawal *et al.*, 1994, Bartz *et al.*, 1980 and Corthier and Franz, 1981). In camels, rotavirus antibody detection was done in serum using counter immuno electrophoresis (CIE) where 27 out of 55 (49%) sera collected from 1-3 years old camels in Morocco were found positive for rotavirus antibodies (Mahin *et al.*, 1983). El Sayed *et al.* (1992) described the detection of high rotavirus antibody titer (1:256) in camel milk using virus neutralization.

In the present study a commercial competitive ELISA kit, which was used for detection of rotavirus antibodies in bovine were used. Antibody to group A rotavirus was found to be existing in camel sera collected from all areas of study as detected by ELISA with an overall percentage of 48.1%. The results of seromonitoring of camel sera for rotavirus in the present study were similar to that obtained by Mahin *et al.* (1983). However this study was done using almost ten times the number of sera tested and the technique we used (ELISA) was reported to be more sensitive (Khattar and Pandey, 1990). The results of antibody ELISA obtained in this study indicated the high prevalence of rotavirus infection in the four areas of study with highest percentage in Sennar and Blue Nile States, followed by Gedarif, River Nile and Kordofan States, respectively. It was noticed that high antibody titers (3+ and 4+) were found in more than 50% of the positive samples, which may indicate that the animal had exposed to rotavirus infection several times. Most of the positive results were found in 18-36 month age group. This strengthening the hypothesis that those animals had been exposed to rotavirus several times as the first rotavirus infection is most likely to occur during the first month of age and at this age most of calves receive rotavirus antibodies through colostrums. Maternal antibodies may neutralize the virus leading to absence or low titers of antibodies in serum, which then increases due to subsequent exposures.

We noticed that the highest percentage of positivity for rotavirus antibodies was detected in clinically healthy camel calves and adult camels, then in diarrhoeic and finally recovered camel calves. This may be attributed to the fact that usually the antibody titers during the infection are low and increase to reach high level in recovered and clinically healthy animals in which previous infections were more

likely to be occurred specially in endemic areas.

Concerning the sex distribution of positive sampled animals for detection of rotavirus antibody, males were found to possess slightly higher positives than females in the different age groups tested. This was in agreement with the previous report describing the higher percentage of male bovine and camel calves affected by rotavirus induced diarrhoea in Sudan (El Nour, 1994, Ali 2003).

This is the first study to determine the prevalence of group A rotavirus antibodies in camels in Sudan, a study to investigate group C rotavirus antibodies is highly recommended.

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