National Agriculture and Water Research Center, Ministry of Agriculture and Water, Saudi Arabia.

# THE SUSCEPTIBILITY OF CAMELS TO NATURAL INFECTION WITH FOOT AND MOUTH DISEASE VIRUS

(With 5 Tables)

# M. A. FARAG; A. AL-SUKAYRAN\*; K.S. MAZLOUM and A.M.AL-BOKMY

\*Department of Foot and Mouth Disease, Veterinary Serum and Vaccine Research Institute, Abbassia, Cairo - Egypt. (Received at 29/9/1998)

قابلية إصابة الجمال بالعدوى الطبيعية بفيروس مرض الحمى القلاعية

د.مختار عامر فرج ، عبداللة محمد الصقيران ، كمال صابر مظلوم أحمد منصور البقمي

أوضحت العديد من الدراسات السابقة على قابلية الجمال (أحادية السنام) للاصابة الطبيعية بغيروس مرض الحمى القلاعية عن وجود تناقض في نتائجها بين الباحثين، في هذة الدراسة وعلى أبقار وجمال واغنام وماعز غير محصنة بلقاح الحمى القلاعية ترعى في ثلاث مزارع مصابة بأوبئة تنتمى للنوع السيرولوجي (A) و (O) لفيروس مرض الحمى القلاعية وتقع في محافظات موبوءة بهذا المرض في المنطقة الوسطى والشرقية بالمملكة العربية السعودية، دلت نتائج الدراسات السيرولوجية على عدم وجود أجسام مناعية ترسيبية ضد الأنتجن المصاحب للعدوى بفيروس مرض الحمى القلاعية (VIA) في أمصال الجمال، بينما وجدت في ٥٥% من أمصال الأبقار (٢٩من ٥٠)، و ١٤% من أمصال الأغنام (٤٤ من ٢٠٧)، علاوة على ذلك، اكدت الدراسة من ٢٠٠٧) و ١٣ % من أمصال الماعز (٣٦ من ٢٧٣)، علاوة على ذلك، اكدت الدراسة أن جميع الأمصال التي تحتوى على أجسام ترسيبية (Precipitating Antibodies) أنتجن عادليت العيرة الى مرض الحمى القلاعية، بينما لم يتم الى تعادل لنفس الفيروسات عند فحص ٩٠ عينة من أمصال الجمال، وعند فحص عينات السائل المريئي البلعومي لـ ١٩ رأس من الأبقيار و العدن المناس الأبقيار و المناس من الأبقيار و المنافز تحتوى في أمصالها على أجسام ترسيبية والمنام وعند فحص عينات السائل المريئي البلعومي لـ ١٩ رأس من الأبقيار و ٢٠ من الماعز تحتوى في أمصالها على أجسام ترسيبية و ٢٠ من الماعز تحتوى في أمصالها على أجسام ترسيبية و ٢٠ من الماعز تحتوى في أمصالها على أجسام ترسيبية و ٢٠ من الماعز تحتوى في أمصالها على أجسام ترسيبية و ٢٠ من الماعز تحتوى في أمصالها على أجسام ترسيبية و ٢٠ من الماعز تحتوى في أمصالها على أجسام ترسيبية و ٢٠ من الماعز تحتوى في أمصالها على أجسام ترسيبية و ٢٠ من الماعز تحتوى في أمصالها على أجسام ترسيبة و ٢٠ من الماعز تحتوى في أمصالها على أجسام ترسيبية و ٢٠ من الماعز تحتوى في أمصالها على أجسام ترسيبية و ٢٠ من الماء و ١٠ من الماء و ٢٠ من الماء و ١٠ من الماء و ١٠ من الماء و ١٠٠ من الماء و ١٠ من الماء و ١٠ من الماء و ١٠ من الماء و ١٠ من الماء و ١٠

(VIA) للأنتجن المصاحب للعدوى بفيروس مرض الحمى القلاعية (VIA) تسم عزل ٧ فيروسات محمولة (Carrier Strains) للنوع السيرولوجى (A) و ٣٢ للنوع السيرولوجى (O) ، بينما لم يتم عزل اى فيروس من عينات الجمال ولا النتائج المتحصل عليها على أن الجمال لم تكون اجسام مناعية ، بينما دورها كحامل لفيروس مرض الحمى القلاعية لم يتم تقنينة بعد ،

#### SUMMARY

The results of previous studies on the susceptibility of camels (Camelus dromedarius) to natural infection with Foot and Mouth Disease (FMD) reported by different authors are contradictory. Comparative serological studies carried out on non - vaccinated animals (cattle, sheep, goats and camels) raised in three different farms located in endemic FMD provinces in the Central and Eastern regions of Saudi Arabia and exposed to type (A) and (O) FMD outbreaks, revealed that none of the tested camel sera positive for VIA antibodies, while 58%(29/50) cattle, 14%(44/307) sheep and 13%(36/273) goats reacted positive. Moreover, all sera with precipitating antibodies against VIA antigen confirmed the presence of antibodies against A Sau 41/91 and O1 Manisa \68 strains of FMD virus. None of the examined camels' sera showed any neutralizing antibodies against the two viruses. In the animals of the second outbreak, out of 20 sheep and 39 goats' sera that reacted negative against VIA antigen, 8 (40%) and 17(44%) respectively neutralized O1 Manisa\ 68 strain of FMD virus. Testing oesopharyngeal fluids (O.P.) collected from 19 cattle, 34 sheep, 29 goats that reacted positive against VIA antigen showed the isolation of seven type A, and 32 type O FMD carrier viruses. On the contrary, 30 probang samples collected from camels tested negative. The results obtained from the present study indicated that camels did not show any antibody development. However, their role as carriers of FMD viruses can not be ruled out

Key words: Foot and Mouth Disease-Virus-Carrier-Serology-Camels-Saudi Arabia.

# INTRODUCTION

There have been contradictory reports concerning the susceptibility of camels to both natural and experimental infections with

Foot and Mouth Disease (FMD) virus. KOWALEVSKY, 1912 in Kazakhstan and MOUSSA et al., 1987 in Egypt had shown that camels are susceptible to FMD. Additional experimental studies in Egypt following intranasal inoculation of serotype (O) FMD virus strains, showed only slight or inapparent clinical signs (Nassr et al., 1980., Omar, 1982 and Moussa, 1988). On the contrary, in various FMD epizootics countries (Ethiopia, Oman and Egypt), when camels were kept for several weeks in close contact with severe cases of FMD in cattle, sheep and goats, the camels did not develop any clinical signs of FMD (Richard, 1976., Hedger et al., 1980 and Omar, 1982). The aim of the present study was to, if possible, resolve the question of the susceptibility of camels to natural infection with FMD virus and its possible role in the epizootiology of the disease.

# **MATERIALS and METHODS**

Three field outbreaks of FMD (in which camels were in contact with the affected animals) at backyard in the Eastern region, at large breeding farm in the Central region and at a small breeding farm near Riyadh were investigated by the authors. The relevant epidemiological information of each outbreak recorded in Table 1. On the animals of the three farms, no clinical signs observed in all camels and no vaccination against FMD was applied. Serotype A FMD virus antigenically related to A Sau 41/91, and two O serotypes virus related to O1 Manisa (FMD-WRL, R.P. KITCHING personal communication) were isolated from the three outbreaks respectively.

# Serum samples:

Twenty-five camels and 35 cattle sera were collected from the animals of the first outbreak. Six-hundred camels, 247 sheep and 228 goats of the animals of the second outbreak. Twenty camels, 15 cattle, 60 sheep and 45 goats' sera collected from the animals of the third outbreak. The animals of the first and the third outbreaks were sampled 2 months after the end of the outbreaks, while, the animals of the second outbreak were sampled 5 months after the end of the outbreak. Cattle, sheep, goats and camels' sera screened for the presence of VIA antibodies.

# Immunodiffusion test:

The immunodfiffusion test was carried out in 100 mm Petri dishes using FMD virus infection associated (VIA) antigen and reference positive

serum as previously described (Hafez et al., 1994).

#### Virus neutralization

Outbreak1: All cattle that had VIA antibodies as well as all camels' sera tested for the presence of neutralizing antibodies against A Sau 41\91 FMD virus.

Outbreaks 2 and 3: All animals that had VIA antibodies screened for strain O1 Manisa/68 FMD neutralizing antibodies. Moreover, of the reacted negative animals 50 camels, 20 sheep and 39 goats from the second outbreak, and 20 camels from the third outbreak also tested. The detection of neutralizing antibodies against strain A Sau 41/91 and O1 Manisa /68 FMD virus in the selected serum samples carried out as described elsewhere (GOLDING et al., 1976).

Probang samples:

Of the animals that reacted positive against VIA antigen of the three outbreaks, probang samples were collected from 19 cattle, 34 sheep and 29 goats. In addition, 10 camel samples from each outbreak also collected. Sample collection was carried out using probang cups the samples were treated with equal amount of trichlorotrifluoro-ethan as previously described by Sutmoller & Cottral (1967).

# Enzyme- linked immuno -sorbent assay (ELIAS):

Rabbit and guinea pig immune sera against the 7 serotypes of FMD virus, standard inactivated antigens for each serotype, horse radish peroxidase conjugated rabbit anti-guinea pig serum and the orthophenylenediamine substrate tablets were kindly provided by the FMD World Reference Laboratory (WRL) as an ELISA FMD - typing kit. The indirect sandwich ELISA described by Roeder & Leblanc Smith (1987) with slight modification (Anon, 1989). Was applied for serotyping of the isolates carrier strains of FMD virus.

#### Virus isolation in cell cultures

Primary calf kidney cell cultures (PCK) were used to isolate the carrier strains of FMD virus from probang samples as previously described by Hafez et al., (1993).

#### **RESULTS**

Testing of sheep, goats and camel sera for the presence of precipitating antibodies against VIA antigen.

None of the tested camel sera was positive for VIA antibodies, while 57%(20/35) cattle, 6% of both the sheep (16/247) and goats (14/228) and 60%(9/15) cattle, 47% (28/60) sheep and 49%(22/45) goats sera of the three outbreaks respectively reacted positive (Table 2).

Testing of some sheep, goats and camel sera for the detection of neutralizing antibodies against A Sau 41/91 and O1 Manisa \ 68 of FMD virus.

Examination of all sera with precipitating antibodies against VIA antigen by virus neutralization test confirmed the presence of antibodies against A Sau 41/91 and O1Manisa/68 FMD virus. In addition, testing of 25, 50 and 20 camels from the three outbreaks as well as 20 sheep and 39 goats that non of them had VIA antibodies. None of the camel sera had neutralizing antibodies, while antibodies against serotype O1Manisa/68 of FMD virus were detected in 8 (40%) and 17 (44%) of the sheep and goat sera respectively (Tables 3 and 4).

Isolation and typing of FMD virus from probang samples:

Seven type A, and 32 type (O) FMD carrier viruses were isolated from the probang samples collected from VIA positive cattle, sheep and goats. On the other hand, all the probang samples collected from camels tested negative (Table 5).

# **DISCUSSION**

The presence of VIA antibodies in 58%(29/50) cattle, 14%(44/307) sheep and 13%(36/273) goats of the animals of the three farms after two to five months from the end of the outbreaks. The detection of neutralizing antibodies against serotype A Sau 41/91 and O1 Manisa\ 68 FMD virus in all animals that had VIA antibodies, and 40% (8/20) sheep and 44%(17/39) goat sera reacted negatively for VIA antigen of the animals of the second outbreak. Althe isolation of type A and O FMD carrier virus from some individual cattle, sheep and goats, confirmed the prevalence of the two viruses within the three farms. The

relatively high numbers of isolated type O and A FMD carrier viruses from cattle 74%(14/19), sheep 44%(15/34) and goats 34%(10/29) positive for the presence of precipitating antibodies against VIA antigen proved that the persisted infection in the three farms were types A and O FMD viruses (Berger et al., 1990., Neitzert et al., 1991).

Based on: A) The absence of clinical signs of FMD in camels despite the presence of clinically affected cattle, sheep and goats, and the circulation of the viruses in the three farms.

- B) Undetectable specific precipitating and neutralizing antibodies against VIA and type A and O1 Manisa \ 68 FMD viruses in all the tested camel sera.
- C) Unsuccessful isolation of FMD carrier virus from the camel probang samples. Accordingly, camels can not be naturaly infected with FMD virus. The virus was not be able to establish itself in the tissue of the upper respiratory tract and replicate to enter the blood stream, so camels failed to produce antibodies against the circulated FMD virus. The present serological results are in accordance with the results of a serological survey carried out in Egypt in which all the 364 tested camels' sera were negative for VIA test (Farag, 1983). In addition, camels intranasally inoculated with the Egyptian type O1 FMD virus strain did not show any thermal, clinical or serological response to this experimental infection in which an infection route similar to the possible natural route was used (FARAG, 1983). Similarily, no FMD virus was isolated from 454 camels probang samples collected from different Egyptian provinces where FMD outbreaks had been recorded (Tantawi et al., 1984). Recent study In Egypt following intranasal inoculation of camels with serotype (O) FMD virus, the virus was re-isolated from the O.P. and faeces for 6 days following virus inoculation (Moussa, 1988). Studies carried out in Egypt, Sultanate of Oman, Ethiopia and Kenya proved that camels can not be infected with FMD virus by contact (Omar, 1982., Hedger et al., 1980., Richard, 1976 and Paling et al., 1979).

The results of our investigations together with the divergent results reported by various authors from different countries, suggest that camels (*Camelus dromedarius*) are not susceptible to natural infection with FMD, but could be considered as mechanical carrier or resorvoir in the epizootiology of foot and mouth disease, as camels show neither any thermal nor any clinical signs of the disease (Farag, 1983., Buchnev et al.,1987., Doli & Stimmelmayer, 1992., Fassi-Fehri,1987., Mcgrains &

Higgins, 1986., Shommein & Osman, 1987 and Wilson, 1984) and excrete the virus over a certain period (6 days) following inoculation, with virus titers to low as compaired to that of the inoculated dose (Nasser et al., 1980, Omer, 1982 and Moussa, 1988). At the meantime, camels do not seem to produce antibodies against the virus.

#### REFERENCES

- Anon, (1989): Foot and Mouth Disease virus antigen detection: ELISA manual. Foot and Mouth Disease World Reference Laboratory working protocol. 12 pp.
- Buchnev, K.N., Tulpbaev S.Zh. & Sansyzbaev ,A.R. (1987): "Infectious disease in camels in the USSR. Rev. sci. tech. off. int. Epiz. 6,487-495.
- Berger, H.G., Straub, O.C., Ahl, R. & Marquardts, O. (1990): Identification of Foot and Mouth Disease virus replication in vaccinated cattle by antibodies to non-structural virus proteins. Vaccine 8, 213 -216
- Doli, M. & Stimmelmayer, R. (1992): "Important camel disease". In: Schwarz, H.J. & Doli, M. (Editors): The one-Humped camel in Eastern Africa: A Pictorial Guide to Disease, Health Care and Managment". Josef Margraf Publishers, Weikerscheim, Germany, pp. 155-224.
- Farag, M.A.(1983): Significance of (VIA) virus infection associated antigen as a tool for epizootiological survey to Foot and Mouth disease in Egypt. Master thesis, Cairo University, 71 pp.
- Fassi Fehri, M.M. (1987): "Disease of camels" Rev. sci.tech. Off. int. Epiz. 6,337-354.
- Fernandez, A.A., De Mello, P.A., Gomer, L. and Rosenberg, F. J. (1975): The use of virus infection associated antigen (VIA) in the detection of cattle exposed to FMDV. Bol. Centr. Panam. Fieber Aftosa. 17 / 18,19-22
- Golding, S.M., Hedger, R.S. and Talbot, P. (1976): Radial immunodiffusion and serum neutralization techniques for the assay of antibodies to swine vesicular disease. Res. Vet. Sci. 20, pp.142-147.

- Hafez, S.M., Farag, M. A., Mazloum, K.S.& Al-Bokmy, A.M.(1993):
  Application of double sandwich enzyme linked immunosorbent assay for the diagnosis of Foot and Mouth disease in Saudi Arabia.
  Dtsch, Tierarztl. wschr. 100. 103 106
- Hedger, R.S., T.R. Barnett & D.F. Gray (1980): Some virus diseases of domestic animals in the Sultanate of Oman. Trop. Anim. Hlth. Prod. 12, 107 114
- Kowalevsky, M.J.M. (1912): Le Chameau et ses maladies d'apres les observations d'auteurs russes .J. Med . Vet. Zootechn, Lyon, France 15, 462 466.
- Mcgrane, J.J & Higgins, A.J. (1986): "Infectious disease of the camels: Viruses, bacteria and fungi". In Higgins, A.J. (Editor) "The camel in Health and Diseas". Bailliere Tindall. London. 92-110
- Moussa, A. A. M. (1988): The role of camels in the epizootiology of FMD (Foot and Mouth Disease) in Egypt. In: FAO, The camels: Development research. Proc. of Kuwait seminar, Kuwait, 1986/10/20-23, pp.162-173
- Moussa, A., A., A.Daoud, A. Omer, N. Metwally, M. El-Nimr & J.W. Mc Vicar (1987): Isolation of Foot and Mouth Disease virus from camels with ulcerative disease syndromes. J. Egypt. Vet. Med. Ass. 47 (1%2), 219 229
- Nassr, M., A.A Moussa, M. Abdeir Metwally and R. El-S. Saleh (1980):
  Secretion and persistence of Foot and Mouth Disease virus in faeces of experimentally infected camels and rams. J. Egypt. Vet. Med. Ass. 40 (4), 3-13
- Neitzert, E., Beck, E., De Mello, P.A. Gomes, I. & Bergmann, I.E. (1991):

  Expression of the Aphthovirus RNA polymerase gene in

  Eschrechia coli and its use together with other bioengineered nonstructural antigens in detection of late persistent infection.

  Virology, 184, 799 804
- Omar, A. (1982): Detection of Foot and Mouth Disease virus among carriers in farm animals. Ph.D.Thesis (Microbiology) Vet. Med. Cairo university, 145 PP
- Paling, R.W., Jessottt, D.M. & Heath, B.R. (1979): The occurrence of infectious disease in mixed farming of domesticated wild herbivores and domestic herbivores, including camels in Kenya.I. Viral disease: a serologic survey with special reference to FMD. Journal of Wild life Disease, 2:351-358

- Richard, D. (1976): "The disease of the dromedary in Ethiopia" Ethiopean Vet. Bull. 37,46-47.
- Roeder, P. L. & Le Blanc Smith, P.M. (1987): Detection and typing of Foot and Mouth Disease virus by enzyme linked immunosorbent assay: a sensitive, rapid and reliable technique for primary diagnosis. Res. Vet. Sci. 43,225-232
- Shommenin, A.M. & Osman, A.M. (1987): "Diseases of camels in the Sudan". Rev. sci.tech.Off. int.Epiz. 6,481-486
- Sutmoller, P. & Cottral, G.E. (1967): Improved techniques for the detection of FMDV in carrier cattle. Arch. ges. Virusforsch. 21, 170 177
- Tantawi, H.H., Moussa, A. A. M., Omer, A. & Arafa, M.A. (1984):

  Detection of Foot and Mouth Disease virus carrier among farm animals in Egypt. Proc. First Scientific Congress, Faculty of Veterinary Medicine, Assiut University, 65 79
- Wilson, R. T (1984): "The camel". longman, London, pp. 118-133.

Table 1: Number of animals showing clinical signs of Foot and Mouth Disease during the occurrence of the three out

Animal	Ва	ckyar	Larg	ge breedi	ng farm	Smal	ll breedi	ng farm
Specises	A	%	P	Α	%	P	A	%
Camels	0	0	700	0	0	25	0	0
Sheep	-		3000	1000	33	220	140	64
Goats			900	375	42	190	100	53
Fattening bulls Cows	55 15	92 75	-		-	20	20	100

P: Population.

A: Affected animals

<sup>%:</sup> Percentage of affected animals.

Table 2: Results of VIA immunodiffusion test for serum samples collected from camels, sheep and goats

	Be	Backyard	-	Large br	Large breeding farm	1	Small	Small breeding farm	m	Total number	number of positive
Animal specises		,	~							animals	ls
	No* tested	positive	%	No*.tested	positive	%	No* tested	positive	%	Total positive	%
Camels	25	0_	0	009	0	0	20	0	0	0	0
Cattle	35	20	57%	**	1	1	15	6	%09	29	28%
Sheep	1	-	1	247	91	%9	09	28	47%	44	14%
Goats	-	1	1	228	14	%9	45	22	46%	36	13%

No\*. Number of samples tested.

%: Percentage of positive.

Table 3: Neutralizing antibody titers against A Sau 41/91 FMD virus in camels and cattle sera collected from the animals of the first outbreak.

Spe	Animal Specices	Samples programments of the second se	Samples positive for Immunodiffusion test	Sample 1 Immunod	Sample negative for mmunodiffusion test	_ #
	No*.tested Positive	Positive	%	No*.tested Positive	Positive	%
Camels				25	0(1)	%0
Cattle	20	20(2)	100%	1	1	1

No\*.: Number of samples tested.

(1): No neutralization of the test virus at serum dilution of 1:10

(2): Sero titers ranging from 1:80 to 1:320 (Average 1: 168)

Table 4: Neutralizing antibody titers against O1 Manisa /68 FMD virus in camels, cattle, sheep and goats sera collected from the animal second and third outbreaks.

	-		Constitution of the last									
		Large brea	Large breeding farm (Second outbreak)	(Second	outbreak)			Small b	Small breeding farm (Third outbreak)	m (Third.	(Appart)	
Animal specises	•	to VIA toof			TA VIA	*			9		outol can	
	1	-		- 1	-ve via lest			+ve VIA test	st		-v VIA test	
	S N	positiv	%	No	Positiv	%	NO NO	Positiv	%	No	Positive	%
-Transco								0				
Camels	1	1	1	20	000	%0	1			30	0111	100
Cattle			-							707	0(1)	%5
Catho	-	***	1	1	1	!	6	006	100%	1		
Sheep	16	16(3)	1000%	20	0 (4)	4007	00		2007		-	-
		70.07	0/001	707	8 (4)	40%	87	28 (5)	%001	1	1	1
Goats	14	14 (6)	100%	39	17(7)	44%	22	22 (8)	10007			
	The state of the s	Section of the local Contraction	- Carlo Carlo Constitution of the Constitution	Personal Printers of the Personal Printers of			44	(0) 77	200	1	1	1

No\*: Number of animals tested.

(1): No neutralization of the test virus at serum dilution of 1:10

(2): Sero titers ranging from 1:40 to 1:320 (Average 1:173)

(Average 1:216) (3): Sero titers ranging from 1:80 to 1:320

(Average 1:124) (Average 1:40) (5): Sero titers ranging from 1:40 to 1:320 (4): Sero titers ranging from 1:20 to 1:80

(Average 1:100) (Average 1:118) (7): Sero titers ranging from 1:40 to 1:320 (6): Sero titers ranging from 1:20 to 1:320

(Average 1:91)

(8): Sero titers ranging from 1:20 to 1:320

Table 5: Isolation and typing of carrier strains of FMD from probang samples collected from camels, cattle, sheep and goats.

	Ba	Backyard		Large br	Large breeding farm	u	Small	Small breeding farm		Total muchae	f. monition
Animal specises				)				ini Simonia		rota mumber of	animor of positive
	Not tooted	Danising	10	NT de L		L				animia	2
	TAO . ICOICO	LUSTILVE	0/	No. rested	positive	%	No*.tested	positive	%	Total positive	%
Camels	10	0	%0	10	0	00%	10		/00	2000	2
Cottle		448					0.1		020	0	%5
Calife	10	**/	20%	!	1	1	6	7***	780%	7.1	740/
Sheen				1.4	1000	1000			10/0	14	0/4/
		**		14	***/	20%	20	*	40%	15	7077
Goats	1	1	1	14	4**	200%	15	2887	1004		0/44
The state of the s	Section of the last of the las					0/17	CI	0	41%		

No\*: Number of animals tested.

\*\*: Serotype (A) of FMD virus was detected.

Serotype (O) of FMD virus was detected \*\*\*