

STUDIES ON SOME BACTERIAL INFECTIONS OF CAMELS IN HALAIEB, SHALATEEN AND ABOU-RAMAD TRIANGLE

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SUMMARY

The bacteriological examination of 40 swabs from the nasal discharges of 20 local and 20 imported camels revealed that, 37 of them gave 47 bacterial isolates, out of them (25.53%) were Gram-positive and the others were Gram-negative. The most important identified bacterial spp. were *Staphylococcus aureus* (21.27%), *Staphylococcus epidermidis* (4.25%), *Pseudomonas aeruginosa* (4.25%) and *E. coli* (38.29%).

The examination of 40 faecal swabs from 20 local and 20 imported diarrhoeic camels revealed isolation of 50 bacterial isolates, all of them were Gram-negative. The most important isolates were, *Salmonella* spp. (4%), *E.coli* (42%), *Citrobacter* spp.(24%) and *Klebsiella* spp. (12%).

The prevalence of antibodies to brucella was studied serologically among 126 camels (95 local and 31 imported) using 3 different serological methods, namely, RBPT, STAT and ELISA. For local camels, the prevalence rate was 9.47%, 5.26% and 9.47%, respectively. In males the rate was higher than in females except by the STAT. Regarding imported camels, the prevalence rate was 6.67%, 9.67% and 25.80% using the same tests respectively. Also, in males the rate was higher than in females except by ELISA.

INTRODUCTION

Due to the few numbers of camels raised in Egypt, it is essential to import camels from other countries as Sudan, Somalia, Djibouti and Kenya, thus the occurrence of exotic diseases at any time could be expected. Also free movements of cam-

els throughout the borders lead to the transmission and spreading of diseases (Abd El-Aziz, 1996).

The area of the present study (Halaieb, Shalateen and Abo-Ramad triangle) represents the South Eastern border of Egypt and is considered the major point of entry of the imported camels. El-Shalateen market is considered the main market for trading of imported camels from Sudan. Camels enter Egypt via this region from three points of entry, one is legal called "Hederba" and the others are illegal.

Camels under desert condition are commonly suffering from either respiratory infection (Ghawi, 1978, Nawal et al., 1991 and Moustafa, 2004) or diarrhoea (Sayed et al., 1998).

The majority of camels imported to Egypt from Sudan through El-Shalateen Veterinary quarantine are coming from or passing through the Eastern regions of Sudan where brucellosis is recorded by (AbuñDamir et al., 1984 Agab et al., 1998 and Majid and Goraish, 2000).

The main objective of this work is therefore to investigate the prevalence of some bacterial infections among local and imported camels in this important border area of Egypt and the role of imported camels in the epidemiology of these diseases.

MATERIAL AND METHODS

Animals:-

Animals involved in the serosurvey for brucella antibodies were 126 dromedary camels (95 local and 31 imported) of different age (3-8 years) and different sex (77 females and 49 males) while the swabs for bacteriological examination were collected from 20 local and 20 imported camels suffering from respiratory manifestations in addition to anorexia, depression and decreased appetite and another 20 local and 20 imported camels showing diarrhea.

Samples for bacteriological examination:-

A total of 40 nasal and 40 faecal swabs were aseptically collected and rapidly immersed in screw capped tubes containing peptone water as a transport medium. The swabs were sent then to the laboratory with minimal delay for bacteriological examination.

Media for bacteriological examination :-

Peptone water medium (Hi Media), Tryptic soya broth [international diagnostic group (idg)], Blood agar; (Oxoid), MacConkey bile salt agar. (Biolife), Salmonella-Shigella agar; (Biolife); Mannitol salt agar (Oxoid), Triple sugar iron agar (Oxoid), Simmon's citrate agar (Oxoid), Christensen's urea agar (Difco), Brain heart infusion agar. (Difco), Dextrose phosphate, Nitrate reduction broth (Hi Media) and Sugar fermentation medium. (Oxoid).

Antigens used for diagnosis of brucellosis:

Rose Bengal and standard serum agglutination antigens were supplied by Vet. Serum and Vaccine Research institute Abassia, Cairo, Egypt. while *Brucella abortus* lipo poly saccharide (LPS) antigen was supplied by the Molecular Biology Unit, Faculty of Vet. Medicine, Cairo University, Egypt.

Bacteriological methods for examination of nasal and faecal swabs:- Isolation and identification of microorganisms was carried out according to Cruickshank et al., (1975) and Carter, (1984).

Serological methods for detection of antibodies to brucella in camel sera :

Three tests were used, namely, Rose Bengal Plate Test (RBPT) (Morgan et al., 1969), Serum tube Agglutination Test (STAT) (Alton and Jones, 1967) and the modified indirect ELISA using protein-A conjugate according to Chand et al., (1988).

RESULTS

1. Bacteriological study on the nasal and faecal swabs from clinically affected local and imported camels

As shown in Tables (1), out of 40 nasal swabs ex-

amined, 37 were bacteriologically positive giving 47 bacterial isolates, out of them 25.53% were Gram-positive and the other 74.46% were Gram-negative. All of the 40 faecal swabs were bacteriologically positive and revealed 50 bacterial isolates, all of them were Gram-negative.

Nasal swabs from local camels (Table2) revealed 20 bacterial isolates, out of them 20% were *Staphylococcus aureus*, 10% *Staphylococcus epidermidis*, 35% were *E.coli*, 15% *Citrobacter* spp., 10% *Klebeseilla* spp.and 10% were *Proteus* spp., while imported camels revealed 27 isolates, out of them 22.22% were *Staphylococcus aureus*, 7.40% *Pseudomonas aeruginosa*, 40.7% *E.coli*, 22.22% *Citrobacter* spp., 3.70% *Klebeseilla* spp.and 3.70% were *Proteus* spp.

Faecal swabs from local camels (Table3) revealed 24 bacterial isolates, out of them 37.5% were *E.coli*, 20.83% *Citrobacter* spp., 4.16% *Salmonella* spp. and 12.5% for each of *Enterobacter* spp., *Klebseilla* spp. and *Proteus* spp., while imported camels revealed 26 isolates, out of them 46.15% were *E.coli*, 26.92% *Citrobacter* spp., 3.8% *Salmonella* spp., 11.53% *Klebseilla* spp., 7.69% *Enterobacter* spp. and 3.84% were *Proteus* spp.

Table (1): Prevalence of the total bacterial isolates from nasal and faecal swabs of clinically affected camels.

| | Total samples | Positive samples | Total number of isolates | Gram-positive isolates | | Gram-negative isolates | |
|--------------|---------------|------------------|--------------------------|------------------------|-------|------------------------|-------|
| | | | | No. | % | No. | % |
| Nasal swabs | 40 | 37 | 47 | 12 | 25.53 | 35 | 74.46 |
| Faecal swabs | 40 | 40 | 50 | 0 | 0 | 50 | 100 |

No. = Number of isolates.

%. = Percent of isolates to the total isolates of each group

Table (2): Bacterial isolates from nasal swabs of camels.

| Isolated species | | Local camels | | Imported camels | | Total | |
|------------------|------------------------------------|--------------|-----|-----------------|-------|-------|-------|
| | | No. | % | No. | % | No. | % |
| 1 | <i>Staphylococcus aureus.</i> | 4 | 20 | 6 | 22.22 | 10 | 21.27 |
| 2 | <i>Staphylococcus epidermidis.</i> | 2 | 10 | 0 | 0 | 2 | 4.25 |
| 3 | <i>Pseudomonas aeruginosa.</i> | 0 | 0 | 2 | 7.40 | 2 | 4.25 |
| 4 | <i>E. coli.</i> | 7 | 35 | 11 | 40.74 | 18 | 38.29 |
| 5 | <i>Citrobacter spp.</i> | 3 | 15 | 6 | 22.22 | 9 | 19.14 |
| 6 | <i>Klebseilla spp,</i> | 2 | 10 | 1 | 3.70 | 3 | 6.38 |
| 7 | <i>Proteus spp.</i> | 2 | 10 | 1 | 3.70 | 3 | 6.38 |
| Total isolates. | | 20 | 100 | 27 | 100 | 47 | 100 |

No. = Number of isolates. % = Percent of isolates to the total isolates of each group.

Total No. of examined camels = 40 (20 local + 20 imported).

Table (3): Bacterial isolates from faecal swabs of diarrheic camels.

| Isolated species | | Local camels | | Imported camels | | Total | |
|------------------|--------------------------|--------------|-------|-----------------|-------|-------|-----|
| | | No. | % | No. | % | No. | % |
| 1 | <i>Enterobacter Spp.</i> | 3 | 12.5 | 2 | 7.69 | 5 | 10 |
| 2 | <i>Salmonella Spp.</i> | 1 | 4.16 | 1 | 3.84 | 2 | 4 |
| 3 | <i>E. coli.</i> | 9 | 37.5 | 12 | 46.15 | 21 | 42 |
| 4 | <i>Citrobacter Spp.</i> | 5 | 20.83 | 7 | 26.92 | 12 | 24 |
| 5 | <i>Klebseilla Spp.</i> | 3 | 12.5 | 3 | 11.53 | 6 | 12 |
| 6 | <i>Proteus Spp.</i> | 3 | 12.5 | 1 | 3.84 | 4 | 8 |
| Total isolates. | | 24 | 100 | 26 | 100 | 50 | 100 |

No. = Number of isolates. % = Percent of isolates to the total isolates of each group.

Total No. of examined camels = 40 (20 local + 20 imported).

Table (4): Prevalence of brucella antibodies among local and imported camels using different serological tests.

| Camels | Number of examined camels | | | RBPT positive Camels | | | | | | STAT Positives camels | | | | | | ELISA positive Camels | | | | | |
|----------|---------------------------|-----|-----|----------------------|------|-----|-------|-----|------|-----------------------|------|-----|------|-----|------|-----------------------|-------|-----|-------|-----|-------|
| | total | M | F | total | | M | | F | | total | | M | | F | | total | | M | | F | |
| | No. | No. | No. | No. | % | No. | % | No. | % | No. | % | No. | % | No. | % | No. | % | No. | % | No. | % |
| Local | 95 | 33 | 62 | 9 | 9.47 | 3 | 9.09 | 6 | 9.67 | 5 | 5.26 | 1 | 3.03 | 4 | 6.45 | 9 | 9.47 | 4 | 12.12 | 5 | 8.06 |
| Imported | 31 | 16 | 15 | 3 | 9.67 | 2 | 12.5 | 1 | 6.66 | 3 | 9.67 | 2 | 12.5 | 1 | 6.66 | 8 | 25.80 | 4 | 25 | 4 | 26.66 |
| Total | 126 | 49 | 77 | 12 | 9.52 | 5 | 10.20 | 7 | 9.09 | 8 | 6.34 | 3 | 6.12 | 5 | 6.49 | 17 | 13.49 | 8 | 16.32 | 9 | 11.68 |

M = Male. F = Female.

RBPT. = Rose Bengal Plat Test.

STAT. = Serum Tube Agglutination Test..

ELISA. = Enzyme Linked Immuno Sorbent Assay.

N.B : In STAT, the end titer 1:40 or above is considered positive according to (Fayed *et al*, 1982).

Table (5): Prevalence of brucella antibodies among local and imported camels using serum tube agglutination test.

| Camel origins | Number of examined camels | | Serum tube agglutination test (STAT) reactors. | | | | | | | | | | | | | | | | | | |
|---------------|---------------------------|-------|--|-------|---|------|---|------|---|------|---|------|---|-------|---|-------|---|---|---|---|---|
| | Total | M No. | F No. | Total | | 1/10 | | 1/20 | | 1/40 | | 1/80 | | 1/160 | | | | | | | |
| | | | | M | F | M | F | M | F | M | F | M | F | M | F | Total | | | | | |
| Local | 95 | 33 | 62 | 3 | 6 | 9 | 1 | 2 | 3 | 1 | 0 | 1 | 1 | 1 | 2 | 0 | 3 | 3 | 0 | 0 | 0 |
| Imported | 31 | 16 | 15 | 2 | 1 | 3 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 2 | 0 | 2 | 0 | 1 | 1 |
| Total | 126 | 49 | 77 | 5 | 7 | 12 | 1 | 2 | 3 | 1 | 0 | 1 | 1 | 1 | 2 | 2 | 3 | 5 | 0 | 1 | 1 |

M = Male. F = Female.

No. = Number of reactors.

N.B : In STAT, the end titer 1:40 or above is considered positive according to (Fayed *et al*, 1982).

2. Serological study on the prevalence of brucella antibodies among local and imported camels:

As shown in Tables (4 and 5), Rose Bengal Plate Test was positive in 9.47%, 9.67% and 9.52% of the local, imported and total examined camels respectively, while it was 10.20% and 9.09% in the male and female camels respectively. Serum tube agglutination test revealed a positive rate of 5.26%, 9.67% and 6.34% in local, imported and total examined camels respectively, while it was 6.12% and 6.49% in the male and female camels respectively. Indirect ELISA revealed a positive rate of 9.47%, 25.80% and 13.49% for local, imported and total examined camels respectively, while gave 16.32% and 11.68% for male and female camels respectively.

DISCUSSION

In general, camels do not suffer from respiratory diseases, however, when it occurs it is usually initiated by predisposing factors. A number of bacterial species have been found in camels with respiratory disease, however, little is known whether these agents are truly responsible for the diseases (Wernery and Kaaden, 1995).

The data in (Table 1), revealed that bacteriological investigation of 40 nasal swabs from (20 local and 20 imported) camels with nasal discharges revealed that, 37 (92.5%) of them were bacterio-

logically positive. From Table (2), it is clear that the most predominant and important isolates among local camels were *Staphylococcus aureus*, *Staphylococcus epidermidis* and *E.coli*, while among imported camels, *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *E.coli* were the most important isolated species. As shown in Table (2), *Citrobacter* spp., *Klebsiella* spp. and *Proteus* spp. Were also isolated from the local and imported camels. These results were found to be nearly similar with that previously reported by El-Magawary et al., (1986); Al-Ani et al., (1998) and Alhendi, (1999).

It is interesting to mention that, most bacterial isolates recovered in the present study from the nasal discharges of camels were previously isolated from apparently healthy camels as reported by Chauhan et al., (1987) and Nawal et al., (1991) as well as from pneumonic camels as previously reported by Thabet, (1993) and Seddek, (2002). This indicates that, there is a positive correlation between the type of bacterial isolate from the nasal cavity and the pneumonic lungs of camel (Al-Ani et al., 1998).

In (Table 3), the bacteriological investigation of 40 faecal swabs from (20 local and 20 imported) diarrheic camels revealed that, all of them were bacteriologically positive giving 50 Gram-negative bacterial isolates. Faecal samples examination from local and imported camels revealed isolation of *E. Coli*, *Citrobacter* and *Salmonella*

spp. from diarrheic camels: in addition to *Enterobacter*, *Klebsiella* and *proteus* spp.: however with varying percentages, Table (4).

The obtained results are nearly in agreement with those previously obtained by (El-Magawary, 1980, and Sayed et al., 1998) and can be attributed to the exposure of camels to many stress factors as long transport, lack of food and water and climatic changes which lower the animal resistance and facilitate the invasion and or flourishing up of these normal inhabitant bacteria (Sayed et al., 1998). *Escherichia coli* constitutes a large part of the normal commensal aerobic intestinal flora but, it is also the cause of various diseases of great economic magnitude, especially in young animals as enteropathogenic *E. coli* strains which produce enterotoxins causing enteritis and dehydration (Wernery and Kaaden, 1995). Isolation of *salmonella* spp. is in agreement with some authors in Egypt as Kamel and Lotfi, (1963) who examined intestinal lymphnodes and faecal samples and isolated *Salmonella* spp. from 3.1% of the dromedaries examined and Selim, (1990) who found that, 3% of healthy dromedaries showing no signs of diarrhea were *Salmonella* carriers, compared to 17% of dromedaries with enteritis. They concluded that, dromedary is an important reservoir for *Salmonella* and could therefore represent a health hazard for man.

The results of serosurvey for brucellosis which demonstrated in (Table 4) are found to be agree

with those obtained by Fayed et al., (1982) and Nada, (1990) as they detected 6.6% using STAT, Zagloul and Kamel, (1985) 8.1% and Hamdy, (2000) 9.5% and 6.8% using RBPT and STAT respectively. In contrast, lower percentages (4.0%, 4.7%, 1.7% and 7.6%) were reported by El-Nahas, (1964), Thabet et al., (1993), (AbouñEisha, (2000) and AL-Gaabry and Mourad, (2004) respectively, while higher results (24.2) was reported by Ayoub et al., (1978) and AbouñZaid, (1998) who recorded 10.4% positive cases using RBPT and 12.3% using STAT with 5% phenolized saline as well as Ghazy et al., (2001) who recorded 24.3% by RBPT and 18.6% by STAT.

The data in (Table 4) revealed that, male camels had slightly higher prevalence rate than females. These results agreed with those of Okoh (1979) who found that 3 out of 232 camels were positive reactors to brucella, all of them were males; AbuñDamir et al., (1984) who examined 740 camels from 3 regions of Sudan using RBPT, STAT and CFT and found an incidence of 5.6% in males and 4.5% in females and Majid and Goraish., (2000) who found that, female's incidence is slightly higher than males except those camels examined in Gadarif state in eastern of Sudan. On the other hand, the obtained results are in contrast with those reported in Egypt by El-Nahas, (1964) (2% in males and 4% in females), Ayoub et al., (1978) (14% in males and 25% in females), Ahmed et al., (1999) (9.2% in males and 13.7%

in females).

The higher prevalence rate in males than females in this study may be attributed to the continuous movement of male camels either for grazing or during the trading activities and come in contact with other camels, sheep and goats from different herds and different localities which may increase the possibility of transmission of the disease, but the movement of female camels is usually restricted within a limited area with less continuous contact with other animals. Also Al-Khalaf, S. and El-Khaladi, A. (1989) reported that, male camels are considered the main source of transmission of brucellosis, they travel from farm to farm specially during mating time. On the other hand AbuñDamir et al., (1984) stated that, it is well known that female cattle are more susceptible to brucella infection than males, the difference in susceptibility of the sexes between camels and cattle may reflect biochemical or behavioural differences between the species or indicate that, the organism which infect camels is less fastidious than that which infects cattle, moreover Adamu et al., (1997) found that, the difference in infection rates between males and females was not significant suggesting that sex may not be a determining factor in camel brucellosis.

Concerning the source of camels tested either local or imported, the study revealed that, imported camels had a seroprevalence to brucellosis higher than that of local camels Table (4). This result

agrees with that of Radwan et al.,(1983) who found that, the incidence in local camels in Saudi Arabia was 2.8%, whereas in imported Sudani camels it was 4.2%. Our results disagree with those obtained by Atwa, (1997), who investigated 1258 camels sera imported from Kenya and Djibouti at Seuz quarantine and 116 sheñcamels from Egypt using RBPT and found an incidence of 4.05% and 7.75% for imported and local camels respectively. This may be explained that, the majority of camels imported to Egypt from Sudan through ElñShalateen Veterinary quarantine are coming from or passing through the Eastern regions of Sudan (Kassala) which have high incidence rates for brucellosis as recorded by Majidi and Goraish, (2000), also similar result reported by AbuñDamir et al.,(1984), who found that the highest positive numbers of serum samples for brucellosis (7.5%) was from the Eastern region of Sudan. The incidence rates of brucellosis in Sudan were much higher than in Egypt as recorded in some studies by Agab et al.,(1998), who recorded an incidence rate of 30% and Majid and Goraish, (2000) who recorded a rate of 13.9% to 43.9% using RBPT.

Results in Table (4) indicate that, RBPT is more sensitive than STAT test for serodiagnosis of brucellosis in camels, this comes in agreement with Adamu et al., (1997) and Gameel et al.,(1993), who concluded that, the STAT failed to eradicate brucellosis because many infected camels may give negative reactions thus the combined use of

RBPT with STAT is more reliable in diagnosis of brucellosis in camels. Also with Yagoub et al., (1990) who suggested that, RBPT may be a more satisfactory rapid technique for diagnosis and control of brucellosis. This may attributed to the fact that RBPT is a highly sensitive test; Nicolett, (1982) which can detect low titer as in cases of chronic brucellosis, that can not be considered positive by the quantitative tests. Moreover, samples positive to RBPT and negative to STAT mean that, these animals were suffering from chronic brucellosis with low antibody titer not exceed 1/40 (Tables 4 and 5).

Indirect ELISA, Table (4) detected a prevalence rate higher than that detected by RBPT and STAT. This is in agreement with Hamdy, (2000) and Azwia et al., (2001) and suggest that , ELISA is more sensitive than RBPT and STAT due to it's ability to detect antibodies of all iso types (IgM, IgG1 and IgG2) as mentioned by Nielsen et al., (1988).

The relatively high incidence of seropositive camels to brucellosis in the area of study may be due to the close contact of these camels with other farm animal species (sheep and goats) which graze in the same areas and drink from the same water sources. Abo ElñHassan et al., (1991), Barsoum et al., (1995) and AbouñZaid, (1998) detected high percentages of positive reactors camels in contact with other farm animals than those kept in closed farm. Also Radwan et al., (1995),

concluded that, contact between camels and small ruminants was incriminated in the transmission of brucellosis to camels. The majority of locally examined camels were Rashidi breed which had a higher sero prevalence of brucellosis than other camels sharing the same ecoñsystem as recorded by Abbas and Agab, (2002). On the other hand, cross reactivity with other bacterial species and the lack of cultural identification of *Brucella* microorganisms can not be neglected

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دراسات عن بعض الإصابات البكتيرية فى الجمال فى مثلث حلايب - شلاتين - أبورماد

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أدى فحص ٤٠ مسحة أنفية من ٣٠ جمل (سلالة محلية) و ٢٠ جمل مستورد إلى عزل ٤٧ عترة بكتيرية من ٣٧ من هذه الجمال. ومن هذه السلالات البكتيرية ٥٣.٣٥٪ وجدت موجبة الجرام والباقى سالبة الجرام . ومن هذه السلالات المعزولة كان ٣٧, ٢١ ستافيلوكولس أوريس ، ٢٥, ٤٪ ستافيلوكولس أبيدرمس ٣٥, ٤٪ سودومونس أيروجينوزا ، ٣٩, ٣٨٪ شرشياكولوى .

وعند فحص المسحات البرازية (٤٠ عينة) من جمال تشهر عليها الإسهال ثم عزل ٥٠ معزول بكتيرو وجدت جميعها سالبة الجرام . وكان أهم المعزولات كما يلى :

سلالات سالمونيلا (٤٪) وشرشياكولوى (٤٣٪) ستروياكتير (٣٤٪) وكلبسيلا (١٣٪) وبالفحص السيرولوجى لميكروب البروسيلا بين ١٣٥ جمل بواسطة إختبارات مسلية مختلفة وجدت إيجابية فى ٩٠, ٤٧٪ ، ٥٠, ٣٦٪ ، ٩٠, ٤٧٪ الإختبارات الروزبنجال . إختبار التلبد الأنبونى والأليزيا على الترتيب وقد وجد أن الحالات الإيجابية كانت أكثر فى الجمال الذكور عن الإناث .