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**CLINICO – BACTERIOLOGICAL AND THERAPEUTIC
STUDEIES ON CORYNEBACTERIUM - PSEUDO
TUBERCULOSIS INFECTION IN CAMELS
IN ASSIUT GOVERNORATE –EGYPT**
(With 2 Tables and 5 Figures)

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دراسات إكلينيكية – بكتريولوجية وعلاجية عن عدوى كوريني السل الكاذب
في الجمال في محافظة أسيوط – مصر

طمي صديق على

في هذا البحث تم عمل الفحوصات الإكلينيكية على إجمالي (٣٥٠) جمل وأظهرت النتائج بأن عدد ٢٢ حالة (٦,٢٩ %) كانت مصابة بخراجات جلدية باردة على منطقة الرقبة والكتف والأرجل الخلفية. الغدد الليمفاوية أسفل قاعدة الرقبة للجمال المصابة كانت متضخمة – صديدية ومتليفة. الفحوصات المورفولوجية – البيوكيميائية – الميكروسكوب الاليكتروني للعترات المعزولة وللإختبار المرضى لهذه العترات المعزولة في الأرانب أشارت إلى أن الميكروبات المعزولة هو كوريني السل الكاذب. أظهرت نتائج الفحوصات البكتريولوجية بأن كوريني السل الكاذب كان منفرداً في عدد ١٦ حالة (٧٢,٧٣%) وكان مشتركاً مع الميكروب العنقودي في عدد ٤ حالات (١٨,١٨%) وكان أيضاً مع الميكروب السبحي في عدد ٢ حالة (٩,٠٩%) ومن نتائج الفحوصات البكتريولوجية كان ميكروب كوريني السل الكاذب هو الميكروب السائد في العترات المعزولة ولهذا يعتبر المسئول عن حالات الخراجات الجلدية الباردة والالتهاب الصيدي المتليف للغدد الليمفاوية في الجمال المصابة. التاريخ المرضي والملاحظات الإكلينيكية وانتشار العدوى الطبيعية لميكروب كوريني السل الكاذب في الجمال في شهري يوليو وأغسطس بمحافظة أسيوط يوضح بأن المناخ الحار يلعب دوراً مهم في نشاط الميكروب وظهور الحالات المرضية في الجمال ويحتمل أن يكون هناك علاقة بين الحشرات والميكروب في انتقال المرض بين الجمال. أظهرت نتائج إختبارات الحساسية للميكروبات المعزولة بأن المضاد الحيوي سيفالكسين هو أكثر تأثيراً وفاعلية على معظم ميكروبات كوريني السل الكاذب. إستخدام العلاج الجراحي مع المضاد الحيوي بجرعة ١٥ مجم/كجم من وزن الجسم في العضل يومياً لمدة ٢٠ يوم أعطى نتائج فعالة وإيجابية في حالة

الخراجات الجلدية الباردة ولكن في حالة الغدد الصديديّة المتليفة حدث ضمور في الحجم مع تيبس الصديد ولكن التضخم لم يزول تماما.

SUMMARY

Twenty-two (6.29%) cases of the total clinically examined camels (350) showed cold skin abscesses on the neck, shoulder and hind-legs. The superficial cervical lymphnodes of the infected camels were enlarged, suppurative and necrosed. Morphological, biochemical, electron microscopic examinations of the purified isolated strains and pathogenicity test in the rabbit pointed to that the isolated strains were *Corynebacterium Pseudotuberculosis*. Results of bacteriological examinations of the twenty-two pus samples yielded 16 cases (72.73 %) *Corynebacterium Pseudotuberculosis* alone, four (18.18%) *Corynebacterium Pseudotuberculosis* mixed with *Staphylococcus* spp. and two (9.09%) *Corynebacterium Pseudotuberculosis* associated with *Streptococcus* spp. *Corynebacterium Pseudotuberculosis* was the prominent bacterial isolated from the skin abscesses and suppurative lymphadenitis in camels. Case history, clinical observations and spreading of the natural infection with *Corynebacterium Pseudotuberculosis* between camels during July and August months in Assiut Governorate indicate the hot climate play an important role in the activation of the causative agent and facilitate the pathogenesis of the disease and also probably that there is a relation between the insects population and microorganism in the transmission of the disease in camels. Results of antibiotic sensitivity tests indicated that Cephalexine had strong inhibitory effect on most isolated strains of *Corynebacterium Pseudotuberculosis*. Surgical treatment with Cephalexine injection with a dose 15 mg / kg of body weight for twenty days gave good results in skin abscesses while, in suppurative necrosed lymphnodes, the infected lymphnodes were diminished in size and inspessiated but the enlargement not completely disappeared. From the present work, we concluded that camels are susceptible to take the infection with *Corynebacterium Pseudotuberculosis* and so wer recommended that the hygenic measures and control of insects must be taken in the consideration to prevention the spreading of the disease between camels.

Key words: Corynebacterium- Pseudotuberculosis in Camels

INTRODUCTION

The Camel in Africa and Arabia is considered as a wealth reserve and forms an important part of the social structures of many nomadic people in Egypt. According to the last official Egyptian reports, there are 267,000 camels in Egypt, which are one humped and belong to the species camel dromedaries, most of them are located in Upper - Egypt (Abdel-El Salam, 1993). Skin diseases of camels due to bacterial infection were considered as one of the economic losses, in addition the lower prices of sales camels with skin diseases (peculiar appearance) in the commercial markets should not be neglected. Reports on infectious diseases of camels in Egypt are still apparently brief.

Corynebacterium Pseudotuberculosis is well known as the specific cause of caseous lymphadenitis in sheep and goats (Stoops *et al.*, 1984 and Schreuder *et al.* 1994). This microorganism was also encountered as an etiological agent of cutaneous ulcerative lymphangitis with or without regional lymph nodes involvement in cattle and buffaloes (Abou-Zaid and Hammam, 1994; Ali and Zaitoun, 1999) also horses (Gillespie and Timoney 1981). On the other hand, as early as (1934) Carpano could isolate *Corynebacterium* like organism from cases of ulcerative dermatitis in cattle, buffaloes and camels. Ismail *et al.* (1985) recorded an outbreak of *Corynebacterium Pseudotuberculosis* in 21 dromedaries, Cattle and buffaloes in El- Sharkia Governorate. In (1986) Nashed and Mahmoud were isolated an organism resembling *Corynebacterium Pseudotuberculosis* from lymph nodes of camels in Assiut Governorate. The aim of the present work was directed to study: (1) The clinical picture of camels had grossly cold skin abscesses and suppurative lymphadenitis, (2) Isolation and identification of the etiological agents responsible for such lesion, (3) Therapeutic trials with choice drugs according to the results of antibiotic sensitivity tests.

MATERIAL and METHODS

1 - Animals:

During July and August months (1999), a total number of (350) camels of different villages in Assiut- Govenonate were clinically examined and the cases showed cold skin abscesses and suppurative lymphadenitis subjected to detailed clinical examinations describing the clinical findings, Owner's, complaint and history of infected cases

including patient data and prior treatment were also taken. Camels were at different ages from 5 to 10 years and from both sexes.

2 – Samples and culturing methods:

Closed suppurative skin lesions of the infected camels were surgically incised and pus material was swabbed under complete aseptic conditions. The collected swabs were directly inoculated into tube containing brain heart infusion broth then incubated at 37°C for 24 – 48 hours. The incubated tubes were plated on sheep blood agar media. The growth colonies were picked up and subjected to morphological and biochemical identification as methods described by Cruickshank, *et al.* (1975) and Carter (1986). The purified isolated strains were examined with electron microscope in Assiut-University unit. On the other hand, the collected pussy materials from each incised lesion was also directly cultured on sabaroud dextrose agar plates supplemented with chloramphenicol for mycological examinations.

3 – Pathogenicity test:

Purified isolates having morphological; cultural and biochemical characters were subjected to pathogenicity test.

One ml of 24 hours broth culture was injected subcutaneously in rabbit and another rabbit was inoculated also subcutaneously by the same dose of sterile broth and used as control. The two rabbits put under observation. Culture on broth was made from pus material from abscess of rabbit and plated on sheep blood agar.

4 – Antibiotic sensitivity test:

The purified isolated strains were tested for the presence of different members of Antibiotic discs as mentioned in Table (2) and inhibition zone were measured and interpreted according to method of Baure *et al.* (1966). The choice antibiotic was used for treatment of some infected camels.

RESULTS

1 – Clinical findings:

Twenty-two (6.29%) cases of the clinically examined camels (350) showed cold skin abscesses on the neck, shoulder and hind-legs and skin necrosis, enlargement and suppurative of the superficial cervical lymphnodes of camels. The skin abscesses were rounded in shape similar nipples (Fig. 1) approximately 15 cm in diameter as large orange in size and also were painless, firm, and movable. The skin abscesses when incised were filled with thick whitish odourless creamy

pus tinged with blood. The superficial cervical lymphnodes of infected camels were enlarged; necrosed, painless, firm and elevated semi-circular in shape (Fig. 2). The affected lymphnodes when incised were contained odorless, thick whitish caseated pus material tinged with blood. The body temperature of the infected camels were ranged from 37.2 – 38 C°, while the pulse and the respiratory rates were within the normal range.

2 – Bacteriological examinations:

Sixteen (72.73%) of the bacteriological samples yielded *Corynebacterium Pseudotuberculosis* alone; four (18.18%) of samples yielded *Corynebacterium Pseudotuberculosis* with *Staphylococcus* spp. and two (9.09 %) of samples *Corynebacterium Pseudotuberculosis* with *Streptococcus* spp. As shown in Table (1). The collected microscopical slides from twenty-two pus samples showed small Gram's positive rods mixed with other cocci. Colonies of the isolated strains of *Corynebacterium Pseudotuberculosis* were a narrow zone of haemolysis. There was no-growth on the McConkey agar plates. The colonies consisted of gram-positive, coryneform microorganisms (Fig. 3). Biochemical analysis of the pure isolates of *Corynebacterium Pseudotuberculosis* should that catalase, urease, glucose and maltose tests were positive, while, all the other biochemical tests including nitrate reduction were negative. By scanning electron-microscope the pure isolates of the *Corynebacterium Pseudotuberculosis* were short rods arranged in parallel rows (Palisades) and dephtheroid arranged in (V) and (L) shaped (Fig. 4).

3 – Mycological examinations:

Results of the mycological examinations yielded non-pathogenic mycotic agents.

4 – Pathogenecity test:

The inoculated rabbit showed subcutaneous abscess within one week and *Corynebacterium Pseudotuberculosis* re-isolated from pus material of the abscessiated rabbit (Fig. 5).

5 – Antibiotic sensitivity test:

Results of the antibiotic sensitivity test (Table 2) cleared that Cephalxine had strong inhibitory effect on most isolated strains of the *Corynebacterium Pseudotuberculosis*. Surgical treatment with Cephalxine 15 mg/kg, B.W.daily for twenty days, intra muscular, gave a good result in skin abscesses while, in the necrosed suppurative

lymphnodes were inspissated and diminished in size but the enlargement not completely disappeared.

DISCUSSION

In the present work, cold skin abscesses and enlargement, suppurative and necrosis of the superficial cervical lymphnodes (Fig. 1 & 2) were the predominant clinical findings of the infected camels. Similar clinical signs were previously reported in camels by Ismail *et al.*, (1985) and (1990) in Egypt, Domenech *et al.* (1977) in Ethiopia and Schwartez *et al.* (1982) in Kenya. Results of morphological, biochemical, scanning electron microscope of the purified isolated strains (Fig. 3, 4 & 5) respectively were agreed completely with *Corynebacterium Pseudotuberculosis* as described by Soliman *et al.* (1970) in different animals. Results of bacteriological examinations (Table 1) of the all collected pus samples pointed that *Corynebacterium Pseudotuberculosis* were isolated alone from 16 (72.73%) cases out of 22 infected camels; 4 (18.18%) were mixed with *Staphylococcus* spp. and associated with *Streptococcus* spp. In 2 cases (9.09%). The obtained results of bacteriological examinations showed that *Corynebacterium Pseudotuberculosis* was the prominent pathogenic bacterial isolates which responsible for the observed skin abscesses and suppurative lymphadenitis of the infected camels. The obtained results in the present work were nearly similar to the result obtained by experimental studies which made by Torkey *et al.* (1982) and Khater *et al.* (1983) in cattle and Domenech *et al.* (1977) in Ethiopian camels. The latter authors found that the animals inoculated with *Corynebacterium Pseudotuberculosis* showed suppurative skin lesions followed by necrosis and lymphadenitis. These observations may explain the picture of the clinical finding of the infected camels and the isolated microorganism is consider the main cause of the skin abscesses and suppurative lymphadenitis of the infected camels. This opinion was attributed to that the microorganism is a pyogenic bacteria penetrates into the skin-tissues then spread inside the animal body throw cutaneous passways like wise blood and lymphatic vessels (metastasis) producing a filtrable toxin. Extensive caseous necrosis in the lymphnodes of camels were described microscopically by Nashed and Mahmoud (1986) the authors mentioned that the microscopical lesions consists of caseous necroses with a lymphoid and ephthelioid cell reaction.

Case history, clinical observations and spreading of the natural infection with *Corynebacterium Pseudotuberculosis* between camels during July and August months in Assiut Governorate indicate that hot climate play an important role in the activation of the causative agent and facilitate the pathogenesis of the disease and also probably that there is a relation between the insects population and microorganism in the transmission of the disease in camels. Results of the antibiotic sensitivity tests (Table 2) pointed that Cephalexine had strong inhibitory effect on the most of isolated strains of *Corynebacterium Pseudotuberculosis*. Surgical treatment with the choice drugs when applied on three cases with a dose 15 mg / kg B.W daily for 20 days intramuscular gave good results in skin abscesses while, in the necrosed suppurative lymphnodes, the lesions were inspessiated and diminished in size but the enlargement not completely disappeared. From the present work, we concluded that camels are susceptible to take the infection with *Corynebacterium Pseudotuberculosis* and so we recommended that hygenic measure and control of insects vectors must be taken in consideration to prevention the transmission of the disease between camels.

REFERENCES

- Abdel-Salam, M.M. (1993):* Trace elements variation in blood serum of adult she-camel under patho-physiological condition. M.V.Sc., clinical and laboratory diagnosis Fac. of Vet.Med., Assiut univ.
- Abou-Zaid, A.A. and Hammam, H.M. (1994):* Studies on some skin affection in cattel 2-ulcerative lamphangitis. 6th sci., Cong 20-22 Nov., Fac. Vet. Med. Assiut- Egypt. 523-535.
- Ali, H.S. and Zaitoun, A.M. (1999):* Studies on cutaneous suppurative lamphangitis in Buffaloes at Assiut Governorate-Egypt. Assiut. Vet. Med. J. Vol, 41, No: 81 pp.208-222.
- Baure, A.W.; Kirbe, W.M.M.; Sherris, J.C. and Turck, M. (1966):* Sensitivity testing by standard single disc method. Amer.J. of Clin. Path. 45: 493-496
- Carpano, M. (1934):* Ulcerative dermatitis of ruminant and its relation diphtheria of man. Bull. Minist. Agric. Tech. Sci. Ser. Cairo, No: 118.
- Carter, G.R. (1986):* Essential of veterinary Bacteriology and Mycology. 3rd edit. Lea Febiger, Philadelphia.

- Cruickshank, R.; Duguid, J.P. Marmion, B.P. and Swain, R.H.A. (1975):* Medical Microbiology. 12th Edit. Churchill living stone, Edinburgh london and New York..
- Domench, J., Guidot, T. G. and Richard, D. (1977):* Les maladies pyogenes du dromadaire en Ethiopie. Symptomatologie-etiologie. Rev. Elev. Med. Vet. Pays Trop. 30(3): 251-258.
- Gilespie, J.H. and Timoney, J.F. (1981):* Hagan and Bruner's infectious diseases of domestic animals. 7th Edit, Cornel University press. Thaca, New York.
- Ismail, M.M.; Ezzat, J.; El- Jakee, Z.E., El-Sayed and Abd-Elrahmen, M. (1990):* Microorganisms associated with closed abscesses of camels in Egypt. Vet. Med. J. Giza, 38, 53-62.
- Ismail, M.M; Enany, F.R., El-Seedy and Shouman, M.T. (1985):* Oedematous skin disease of camel in EL-Sarkia Governorate. Proc. 1st int. Conf. Appl. Sci- Zagazig.
- Khater, A.R.; Deeb, S.; Bayoumi, A.H. and Salem, H. (1983):* Studies on experimental infection with Corynebacterium Pseudotuberculosis (ovis). II-Pathological changes in cattel. Assiut Vet. Med. J. Vol., 11, No: 21: 85-89.
- Nashed, S.M. and Mahmoud, A.Z. (1986):* Microbiological and Histopathological studies for Rare cases of Coryneba-cterium infection in camels. Assiut. Vet. Med. J. Vol, 18, No: 36, PP. 83-86.
- Schreuder, B.E.C.; Ter- Laak, E.A. and Dercksen, D.P. (1994):* Eradication of caseous lymphadenitis in sheep with help of a newly developed Elisa technique. Vet. Rec. Vol, 135, No, 8: 74-176.
- Schwartz, S. Scjartz, H.J. and Wilson, A.J. (1982):* Fotografische dokumentation wichtiger Kamelkrankhetiten in Kenia. Der parakat. Tierarzt 11, 985-989.
- Soliman, K.N.; Zaki, M.M; Sayour, E.M. and El-Hiniaidy (1970):* The role of the Genus Corynebacterium in animal diseases with special references to species of significances in Egypt.
- Stoops, S.G.; Renshow, H.W. andThilsted, J.P. (1984):* Ovine caseous lymphadenitis: disease prevalence, lesion distribution and thoracic manifestations in a population of mature culled sheep from western united states.
- Torkey, H.A.; El-Shenawi, M. and Okaila, M. (1982):* An investigation into on absecure skin diseases in a caw. World cong. On disease of cattle: 927-933.

Table (1) Results of bacteriological examinations

Number of samples	C.ovis pure	%	c.ovis +	%	c.ovis +	%
			Staph.spp		Strept.spp	
22 pus samples from closed abscesses and suppurative lymphnodes	16	72.73	4	18.18	2	9.09

Table (2) : Antibiotic sensitivity tests on some isolated strains (n = 10)

<i>Antibiotic disc</i>	<i>Inhibition zone</i>									
	1	2	3	4	5	6	7	8	9	19
Tetracycline	3+	2+	3+	2+	1+	1+	1+	2+	1+	2+
Ampicillin	1+	2+	—	1+	2+	—	1+	—	1+	—
Penicillin	2+	—	2+	1+	1+	1+	1+	—	—	—
Cloxacillin	3+	2+	2+	2+	3+	1+	1+	2+	2+	1+
Gentamycin	3+	3+	2+	2+	3+	3+	3+	2+	2+	3+
Cephalexine*	3+	3+	3+	2+	3+	3+	3+	3+	3+	3+
Erythromycin	3+	3+	3+	3+	2+	3+	2+	2+	3+	3+
Kanamycin	3+	3+	2+	3+	2+	3+	3+	2+	3+	1+
Neomycin	3+	2+	3+	3+	2+	1+	3+	3+	1+	2+
Sterptomycin	2+	1+	1+	—	1+	2+	—	—	—	1+

* Cephalexine = velosef (Squibb Egypt)

3+ (I Z > 23 mm)

2+ (I Z > 15-18 mm)

1+ (I Z < 15 mm)

Fig. (1)
Skin abscess at the end of the neck
and shoulder were,

- Rounded
- Large orange in size
- Similar nipple

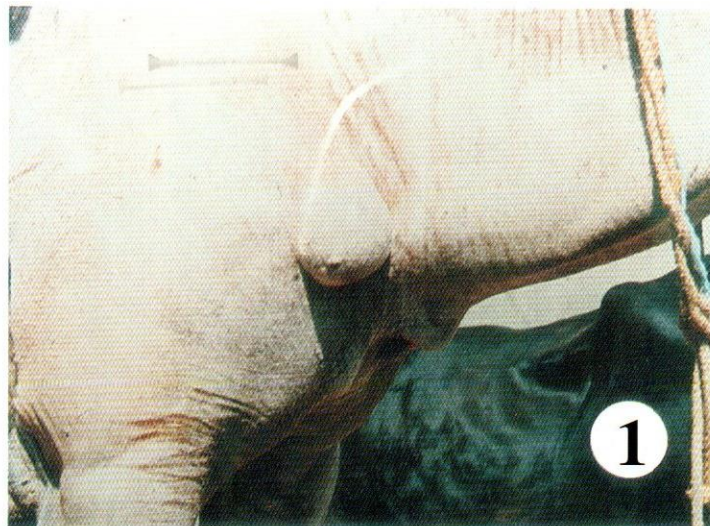


Fig. (2)
Superficial cervical lymphnodes were,

- Enlarge d
- Necrosed
- Elevated semi-circular in shape

Fig. (3)
Gram-positive Coryneform
microorganism

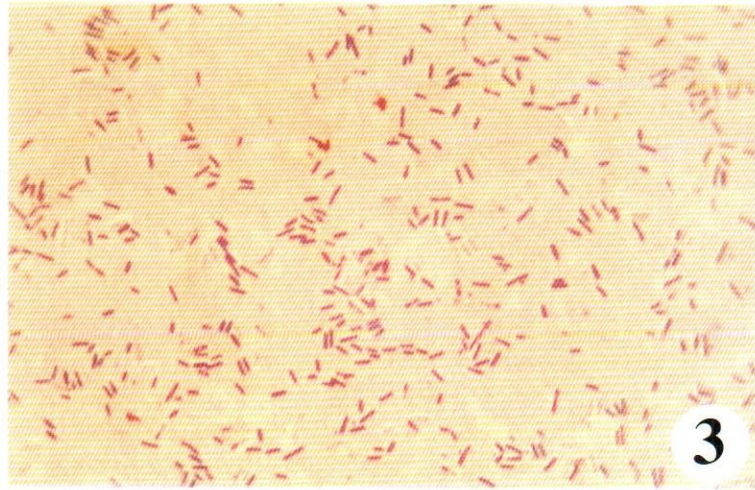


Fig. (4)
Scanning electron-microscope show, M.o.s.
• Short rods
• Arranged in parallel rows (palisades)
• Dephththeroid in shape

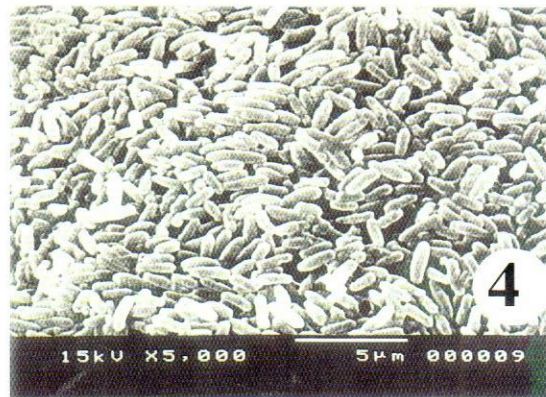


Fig. (5)
Subcutaneous abscess in rabbit with
Pale yellow pus

