# تشوهات تناسلية في كباش مارينو مستوردة نتيجة اصابتها بميكروب الكوريني أوفز ٢ ـ عزل وتصنيف الميكروبات السببة

#### اللخص

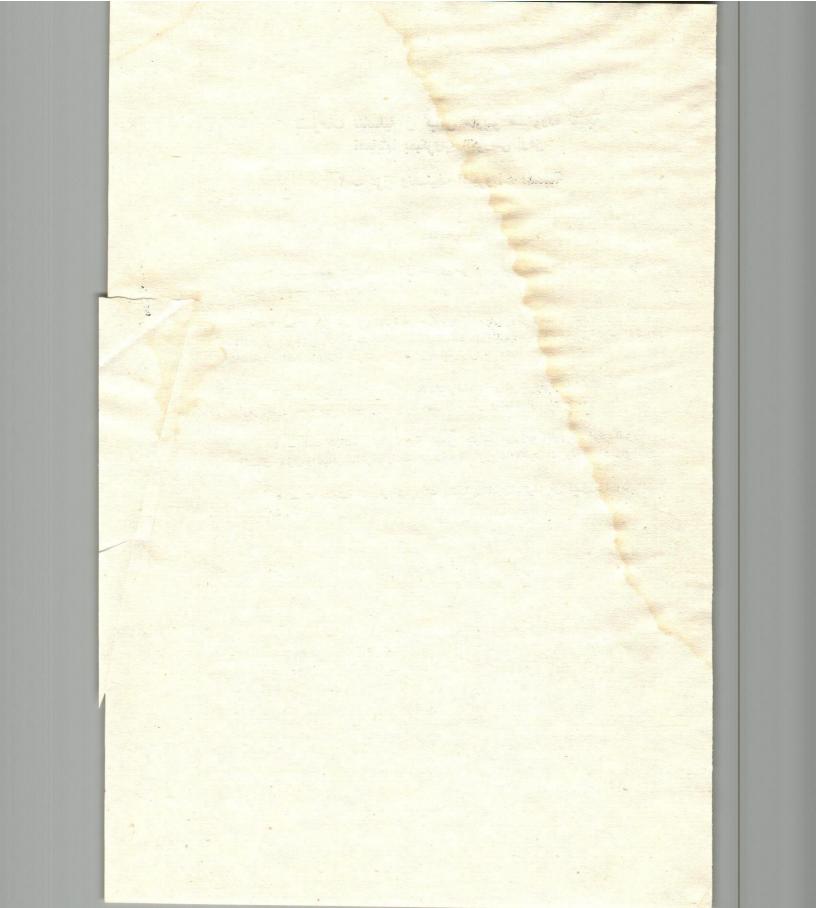
۱ - تم جمع ۱۳۳ عینة سائل منوی من کباش ذات خصی سلیمة ظاهریا واخری متورمة و فحصت بکتریولوجیا •

٢ - تم عزل خمسون عترة من ميكروب الكورينى أوفز وخمسة عترات من ميكروب الكورينى بيوجينز وثمانية عترات من الميكروب السبحى المذبب للدم وأربعة عترات من الميكروب العنقودى الذبب وثلاثة عترات من الميكروب القولونى .

٣ - تم ذبح ١٣ كبش مصاب بالتهاب الخصية والبربخ وعزل منها ميكروب الكوريشي أوفز بصورة نقية من الآفات المرضية بالجهاز التناسلي والغدد الليمناوية .

٤ - تمت دراسة بكتريولوجية لجميع عترات ميكروب الكوريني أوفز المعزولة من السائل المنوى والكباش المدبوحة وكذلك تصنيف للميكروبات الأخرى المعزولة من السائل المنوى م

٥ - تم مناقشة انتشار المبكروبات بالسائل المنوى بالرغم من فشل عزلها من الخصي
 المسابة .



## REPRODUCTIVE DISORDERS CAUSED BY CORYNEBACTERIUM OVIS INFECTION IN IMPORTED MERINO RAMS

#### II. Isolation and Identification of the Causative organisms

(with one table and one figure)

By

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#### SUMMARY

Out of 133 semen samples collected from normally and clinically abnormal genitalia, 50 strains of *C. ovis* was recovered in association with *C. Pyogenes* in 5 cases haemolytic streptococci in 8, Staph. aureus in 4 and E. coli in 3. C. ovis was also isolated in pure culture from pathological lesions in the reproductive organs and the regional lymph nodes of 13 slaughtered rams.

A bacteriological study of all the recovered C. ovis strains as well as the identification of other isolates was performed.

Dissimination of the isolates in the semen, despite of failure of isolation from testicles, was discussed.

#### INTRODUCTION

Sheep as a whole, is not a main breeding animal in this country, but flocks are known among the young breeders, where they graze and drink along the banks of irrigating and draining canals. Although genital affections in rams had been investigated by many authors in many other parts of the world, yet little attention had been paid to this problem in Egypt.

Regarding brucella infection, Brucella melitensis was isolated from testicular and epididymal lesions by ANDREEVSKII (1940). SIMMONS and HALL (1953) procured Br. ovis from semen and/or epididymal lesions and MAYER and CAMERON (1956) demonstrated Br. abortus as a cause of epididymitis in rams.

On the other hand, other minor micro-organisms were traced, where Pasteurella pseudotuberculosis was found by JAMIESON and SOLTYS.

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(1947), from cases with suppurative epididymoorchitis in rams. A Grammegative bipolar pleomorphic organism was reported by DODD and HART-LEY (1955), from semen and suppurative epididymitis in rams, for which ROBERTS (1956-b) suggested the name *Histophilus ovis*. Later, LOLIGER (1958) isolated *E. coli* from semen and genital lesions of affected rams. However Streptococci and Bacteroids were isolated from the genital lesions in rams by BOGDAN (1960) and EKDAHL, MONEY and MARTIN (1968).

The Corynebacterium group of organisms were increminated in genital tract affection, thus *C. ovis* was recovered from epididymal lesions by MCGO-WAN and SHULTZ (1956), the incidence of which was found to be 12.5% among rams by BOGDAN (1960). KHALIMBEKOV, FARZALLEV, and PANKOV (1961); GALLOWAY (1966); EKDAHL *et al.* (1968) and HUGHES and GLAXTON (1968) reported the isolation of the same organism from epididymal lesions and liver abscesses of affected rams. *C. pyogenes* was found by EKDAHL *et al.* (1968) and HUGHES and GLAXTON (1968) to be a cause of epididymal lesions in rams in conjunction with *C. ovis*.

The aim of this contribution is to investigate the bacterial causes of reproductive affections among imported Merino rams at Abis, Koom Oshiem and Yousofia sheep breeding stations of the Meat and Milk Organisation.

#### MATERIALS AND METHODS

The first ejaculate from 133 rams were collected asceptically, as far as possible, in a sterile container, by using separate sterile artificial vagina. These rams were with either apparently normal or diseased genitalia as manifested by oedema of the scrotal sacs, orchitis or epididymitis.

Genital lesions as well as inguinal and prefemoral lymph nodes of 13 slaughtered rams were also incorporated in this investigation. Material was transferred to the laboratory on ice with minimum delay.

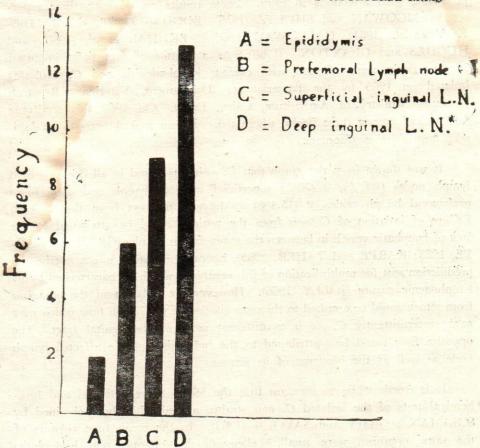
Semen, lesions and lymph nodes were plated on nutrient agar, 10% horse blood agar, Mac-Conkey agar, Albimi brucella agar (RENOUX, 1954), and Sabouraud's agar containing 40 mg Streptomycin sulphate, 30 IU Penicillin per ml of medium. Inoculated plates were incubated under the same condition at 22°C and 37°C for 7 days. Albimi agar plates were incubated in 10% of CO<sub>2</sub> atmosphere at 37°C for a week.

Identification of isolates was performed after BREED, MURRAY, and SMITH (1957). Special attention was paid to the morphological and cultural characteristics, biochemical activities, and biological behaviour in guinea-pigs for the *C. ovis* strains isolated in this investigation.

Isolates from all collected semen samples are illustrated in the following table :

SEMEN	No of samples examined	Isolates   Isolates							
		C. ovis	C. pyogenes	Haem. strptrcocci	Staph. aureus	Staph. epidermidis	E. coli	Sarcina sp.	Gaffkia sp.
Clinically normal genitalia	83		2	3	1	15	in The	13	4
Clinically abnormal genitalia	50	50	3	5	3	2	3	2	2

### FREQUENCY OF ISOLATION OF C. CVIS IN SLAUGHTFRED RAMS



Site of Isolation

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Frequency of C. ovis isolates from lesions in organs and lymph nodes of slaughtered animals are shown in the following graph.

All semen samples and lesion of slaughtered rams were negative to members of the genus Brucella, pathogenic yeasts and fungi.

#### DISCUSSION

It is clear from the table that C. ovis was not isolated from any semen sample collected from clinically normal genitalia, while it was found in nearly all samples collected from clinically abnormal genitalia and mixed with other organisms in 20 samples. Out of 146 semen and lesions in the organs and lymph nodes, 63 strains (43.2%) of C. ovis were obtained. The high incidence of C. ovis was rather stricking and disclosed its role in producing such affections in rams. These results agreed with those reported by MGOWAN and SHULTZ (1956), BOGDAN (1960), KHALIMBE-KOV et al. (1961), GALLOWAY (1966), EKDHAL et al. (1968), and HUGHES and GLAXTON (1968). As a matter of fact these imported Merino flocks were affected with caseous lymphadenitis and C. ovis was isolated on 1965 by the Bacteriology Department, Veterinary Research Institute, Ministry of Agriculture, Cairo, Egypt. On 1968 this syndrome was noticed among these flocks, where the organism took 3 years to establish such reproductive disorders.

It was shown from the graph that C. ovis was found in all deep inguinal lymph nodes (100%), 9 (70%) superficial inguinal lymph nodes, 6 (46%) prefemoral lymph nodes, 2 (15.4%) epididymi and non from the testicles. Failure of isolation of C. ovis from the testicles could be attributed to the lack of lymphatic vessels in between the semineferous tubules (GRAU and KAR-PE, 1963; KARPE and TAHER, 1965), hence; the testicle might not be the prediliction seat for multiplication of this organism, which is characterised by its lymphogenic nature (JOLLY, 1966). However the isolation of the organism from semen could be ascribed to the regurgitation of the lymph flow which may carry intermittently C. ovis to different parts of the genital tract. The opposite flow could be attributed to the induration of the affected lymph node as well as the blockage of its sinuses.

It is worth while to mention that the fermentative activities and biochemical tests of the isolated *C. ovis* strains, agreed with those obtained by SOLIMAN, AGAMY and SAYOUR (1963). In the mean time colonies of the same organism were small, ellepsoid, dry opaque, non haemolytic,

creamy in colour and when touched by the platinum loop, they moved as a whole with difficulty from the surface of the medium. Subsequently on subcultures, colonies became rosette-like in shape, crumbly and surrounded by a narrow zone of beta haemolysis. These findings differed somewhat from what had been reported by the same authors. Concerning the biological behaviour of these isolates in guinea pigs through their injection via the intra-dermal and subcutaneous routes, they gave a constant pattern i.e. death within 24-72 hours with post mortem picture of general toxaemia. This pattern of pathogenicity in g. pigs coincides with 19 strains out of the 25C. ovis strains investigated by SOLIMAN et al. (1963).

The occasional recovery of other organisms as C. pyogenes, Staph. aureus, Streptococci and E. coli from some cases could be considered as secondary invadors or mixed infection with C. ovis. This was in agreement with the findings of EKDHAL et al. (1968), HUGHES and GLAXTON (1968), BOGDAN (1960) and LOLIGER (1958).

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