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SMALL RUMINANTS AS A RESERVOIR OF CERTAIN BACTERIAL AND MYCOTIC PATHOGENS TO MAN

(With 3 Tables)

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(Received at 3/11/2001)

المجترات الصغيرة كمأوى لبعض البكتيريا والفطريات الممرضة للاسان

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تم جمع عدد ٢٥٠ عينة دم (١٥٠ من الأغنام، ٥٠ من الماعز و ٥٠ من الانسان) لمعر فسة نسبة الإصابة بمرض البر وسيلا. و كذلك تم أجراء الفحص البكتريولوجي على عـدد ١٥٠ مسحة شرجية من الأغنام و ٥٠ مسحة شرجية من الماعز و ٥٠ عينة براز من الإنسان. بالإضافة ألى أجراء الفحص الميكولوجي على عينات الصوف و الشعر والقشور الجادية التي تم جمعها من ٢٠٠ حيوان سليم ظاهريا، الأغنام (١٥٠)، الماعز (٥٠) و كذلك عدد ٥٠ مريض مصاب بأعراض جلدية. كانت النتائج الإيجابية لاختبارات البروسيد هي ٢,٦٧، ٣,٣٣ و ٢,٦٧ للأغذام ، صغر ، ٢ و ٢% للماعز و ٢ و صفر و ٢% للانسان أكل من . اختبار الروزينجال واختبار الأنتيجين الشريحي المحمض و اختبار الريفانول على الــترنيب. أسفر الفحص البكتريولوجي عن عزل كل من الأيشريشيا كولاي (٦١,٣٣%)، السهالمونيلا (٢,٦٧%)، الشيجيلا فلكسنبري (١,٣٣%) و الميكروب العنقودي الذهبي (٢%) من الأغنــلم و من الأيشريشيا كولاي (٥٨%) والسالمونيلا (٢%)، من الماعز و من الأيشريشيا كولاي (٦٠%)، السالمونيلا (٨%)، الشيجيلا فلكسنيري (٤%) و المبكروب العنقردي الذهبي (7%) من الإنسان. بينما أسف الفحص الميكولوجي عن عز كل من التريكوفيتو فيريكوزم (٣,٣٣)، التريكوفيتون مينتاجر وفابت (٢,٦٧%) و الكانديدا ألبيكانس (١,٣٣%) من الأغنام و التريكوفيتو فسيريكوزم (٤%)، الستريكوفيتون مينتاجر وفسايت (٦%) والكسانديدا ألبيكانس (٢%) من الماعز والتريكوفيتون فيريكوزم (٨%)، الــتريكوفيتون مينتاجروف ايت (٦%)، التريكوفيتون فيوليشيم (١٦%)، التريكوفيتون ربرم (١٠%)، ميكروســـبورم كسانز (١٨%)، الأبيدرموفيتون فلوكوزم (٤%) و الكانديدا ألبيكانس (١٠%) من الإنسان. هـذا و قد نوقشت الأهمية الصحية لكل من البكتيريا و الفطريات التي تم عزلها.

SUMMARY

A total of 250 blood samples (150 from sheep, 50 from goats and 50 from man) were subjected to serological examination for detection of positive reactors for Brucella. Also, bacteriological examination of rectal swabs collected from 150 sheep and 50 from goats as well as 50 stool specimens collected from human beings. In addition, mycological examination of wool, hair and skin scrapings collected from apparently healthy sheep (150) and goats (50) as well as human beings (50) suffering from clinical skin lesions. Serological examination by using of RBPT, BAPAT and Rivanol test revealed the following, positive reactors for brucellosis were 2.67, 3.33 and 2.67% for sheep, 0.0, 2.0 and 2.0% for goats and 2.0, 0.0 and 2.0% for man respectively. Bacteriological examination revealed the isolation of E. coli (61.33%). Salmonella spp. (2.67%), Shigella flexneri (1.33%), and Staphylococcus aureus (2,0%) from examined rectal swabs of sheep. Also, E. coli (58.0%) and S. typhimurium (2.0%) were isolated from examined goat samples. In addition, E. coli (60.0%), Salmonella spp. (8.0%), Shigella flexneri (4.0%), and Staphylococcus aureus (6.0%) were isolated from examined human samples. Moreover, mycological examination showed that the isolated pathogenic fungi were T. verrucosum (3.33%), T. mentagrophytes (2.67%), and Candida albicans (1.33%) from sheep samples. Moreover, T. verrucosum (4.0%), T. mentagrophytes (6.0%), and Candida albicans (2.0%) were isolated from goats. In addition, T. verrucosum (8.0%), T. mentagrophytes (6.0%), T. violaceium (16.0%), T. rubrum (10.0%), M. canis (18.0%), Epidermophyton floccosum (4.0%), and Candida albicans (10.0%) were isolated from human beings. The zoonotic importance of isolated pathogenic bacteria and fungi were discussed.

Key words: Ruminants, Bacterial and Mycotic pathogens. MAN.

INTRODUCTION

Small ruminants may be infected with various agents of zoonotic importance or being a symptomless carrier. Their excretion may contain many pathogenic microorganisms which results in human infection, especially farmers and herdsmen who are usually in contact with them as well as consumers.

The epidemiological literatures showed that many infections could affect both small ruminants and man among which *Brucella melitensis*. Salmonella, *E. coli*, and dermatophytes as *T. verrucosum*, and *T. mentagrophytes* (Mitra *et al.*, 1998, Bettelheim *et al.*, 2000; Reviriego *et al.*, 2000, and Urio *et al.*, 2001).

Sheep and goats can transmit certain pathogenic and potentially pathogenic agents to human beings through contamination of human food with secretions and excretions of such animals, handling of sheep and goats carcasses during slaughtering and skinning or as a result of ingestion of milk or meat of infected animals, and/or contact with diseased animals (Acha and Szyfres, 1991).

This work is under-taken to give a further light on the role of infected and apparently healthy sheep and goats as a reservoir of certain pathogenic microorganisms (bacteria and fungi) to man and conclude the preventive measures available to overcome such problems in order to promote human health.

MATERIAL and METHODS

1. Sampling:

1.1.Collection of blood samples from sheep, goats and human beings:

A total of 200 blood samples were collected from sheep (150) and goats (50) as well as (50) from human, with a history of contact with sheep and goats. About 10 ml of whole blood was obtained in sterile test tubes. These samples were kept in a slant position about two hours far away from sunlight, then transferred to the laboratory.

1.2. Rectal swabs and stool samples for bacteriological examination:

A) Sheep and goats:

Two hundred rectal swabs (150 from sheep, 50 from goats) were collected.

B) Human beings:

Fifty stool specimens were collected from man, in contact with sheep and goats.

1.3. Hair, wool and skin scraping for mycological examination:

A) Sheep and goats:

A total of 200 samples of wool and hair were collected from apparently healthy sheep (150) and goats (50). Each sample was placed separately in a sterile foil and taken to the laboratory.

B) Human beings:

Fifty human patients clinically diagnosed as having tinea with a history of contact with farm animals especially sheep and goats, were mycologically investigated. The infected area was thoroughly cleaned with 70% ethyl alcohol. The lesions were scraped with sterile scalpel blade. The resultant scales together with some of the blucked hairs were placed separately in a sterile foil and taken to the laboratory.

2. Serological examination of blood samples for brucellosis:

2.1. Collection of sera:

The blood samples were kept over night at 4 $^{\rm O}$ C to allow separation of serum. When the clot retracted, serum was removed by Pasteur pipette, then centrifuged at 3000 rpm. for 10 minutes. The clear serum was transferred into test tubes for each samples and labeled unless immediately used, they were stored at - 20 $^{\rm O}$ C in the deep freezer.

2.2. Serological tests:

A)Buffered acidified plate antigen test (BAPAT):

The test was carried out according to Alton et al. (1988).

B) Rose Bengal plate test (RBPT):

The test was carried out according to Morgan et al. (1978).

C) Rivanol test (Riv.T):

The test was carried out according to the method described by Alton *et al.* (1988).

3. Bacteriological examination of rectal swabs and stool samples:

3.1. Isolation of Gram negative bacteria:

Escherichia coli, Salmonella and Shigella: isolation, purification of the culture and identification of the isolates (Morphologically, detection of motility and biochemical characteristics) were carried out according to scheme described by Koneman *et al.* (1988) and Quinn *et al.* (1994). Serological typing of Salmonella organisms was carried out according to Kauffmann-White scheme (Kauffmann, 1974).

3-2. Isolation of Gram positive bacteria:

Isolation and identification of *Staphylococcus aureus* was carried out according to the methods recommended by Koneman *et al.* (1988) and Quinn *et al.* (1994).

- 4. Mycological examination of wool, hair and skin scrapings samples:
 - 4.1. Isolation and identification of dermtophytes:

These was carried out according to Frey *et al.* (1979), Monica Cheesbrough (1993) and Crissey *et al.* (1995).

4.2. Isolation and identification of yeasts:

These carried out according to *Lodder and* Kreger Van-Rij (1970), Frey *et al.* (1979) and Monica Cheesbrough (1993).

RESULTS and DISCUSSION

Data showed in Table (1) illustrated that, Brucella positive reactors in examined sheep was 2.67%. This result is nearly similar to that recorded by Bassiony and Ibrahim (1997), but lower than that reported by Mainar-Jaime and Vazquez-Blond (1999), while higher than the result obtained by Ding (1993) and Reviriego et al (2000). The higher incidence of Brucellosis reported by some authors may be attributed to the fact that most of their samples were obtained from infected flocks having a history of abortion. However, in the present study the samples were collected from flocks with and without history of brucellosis. The incidence of ovine Brucellosis by Rose Bengal plate test (RBPT), Buffered Acidified Plate Antigen test (PAPA) and Rivanol test was 2.67, 3.33 and 2.67% respectively (Table 1).

Table (1) revealed that, Brucella antibodies were detected at a percentage of 2.0 in examined goats. This result is agree with that obtained by Bassiony and Ibrahim (1997), but lower than that reported by Mainar-Jaime and Vzquez-Boland (1999), while higher than the result recorded by Reviriego *et al.* (2000). In addition, the incidence of caprine Brucellosis by RBPT, BAPA and Riv. test was 0.0, 2.0 and 2.0% respectively (Table, 1).

Brucellosis in goats and sheep are manifested by abortion, which occurs most frequently in the third or fourth month of pregnancy, arthritis and orchitis. Mastitis in goats with small nodules in the mammary glands (Acha and Szyfres, 1991).

It was evident from Table (2) that Brucella positive reactors among human beings was 2.0%. This result is lower than that recorded by Ding (1993), and Wallach *et al.* (1997), and higher than that reported by Abo Shehada *et al.* (1996). In addition, the percentage of Brucella positive reactors in human being examined by RBPT, BAPAT and Rivanol test was 2.0, 0.0, and 2.0% respectively, (Table, 1). Brucellosis is an acute or chronic contagious disease of animals and man, characterized by septicemia, intermittent or irregular fever, sweating of peculiar odour occurs at night, anorexia, general malaise, followed by localization of infection in lymph nodes and genital organs (Acha and Szyfres, 1991).

Data presented in Table (2) illustrated that *E. coli* could be isolated from sheep and goats at a percentage of 61.33 and 58.0 respectively. Concerning with the obtained result of sheep is agreement with that recorded by Randall *et al.* (1997), and higher than that reported by Bettelheim *et al.* (2000). *E. coli* infection in sheep and goats causing white diarrhoea and septicaemic illness in lambs, with neurologic symptoms, ascitis, and hydropericarditis and gastrointestinal disorder (Acha and Szyfres, 1991).

It was evident from Table (2) that *E. coli* could be isolated from human beings at a percentage of 60.0. This result is higher than that reported by Keskimaki et al (2000), and Olorunshola *et al.* (2000). The main clinical signs of *E. coli* infection in man including gastroenteritis. diarrhoea, endometritis, septicemia, urogenital affection and pneumonia (Joklik *et al.*, 1980).

Salmonella could be isolated from sheep at a percentage of 2.67 (Table 2). The isolated Salmonella species from sheep were *S. typhimurium* (2.0%) and *S. enteritidis* (0.67%). In addition, Table (2) illustrated that Salmonella was isolated from goats at a percentage of 2.0 and identified as *S. typhimurium* (2.0%). These results are agreement with Acha and Szyfres, 1991, who stated that *S. typhimurium* was the most common serotypes found in gastroenteritis cases in sheep and goats.

Salmonella was isolated from human beings at a percentage of 8.0 (Table, 2). This result is higher than that reported by Gonera (2000), Seas *et al.* (2000), and Urio *et al.* (2001). The isolated *Salmonella species* from human beings were *S.typhimurium* (4.0%), *S.enteritidis* (2.0%) and *S.typhi* (2.0%) (Table, 2). Signs of Salmonellosis in man include fever, headache, abdominal pain, nausea, vomiting, diarrhoea, gastroenteritis and septicemia (Joklik *et al.*, 1980; and Acha and Szyfres, 1991).

Shigella flexneri was isolated from sheep at a percentage of 1.33 (Table, 2). In addition, the results presented in Table (2) revealed that *Shigella flexneri* was isolated from human beings at a percentage of 4.0, which is nearly similar to that recorded by Seas *et al.* (2000). and lower than that reported by Urio *et al.* (2001). Shigellosis in man is manifested by bacillary dysentery, tenesmus, fever, abdominal pain, gastroenteritis

and diarrhoea which characterized by watery faeces tinged with blood, pus and mucous. (Joklik *et al.*, 1980 and Acha and Szyfres, 1991).

The data presented in Table (2) revealed that *Staph. aureus* could be isolated from sheep at a percentage of 2.0. Also, *Staph. aureus* could be isolated from human beings at a percentage of 6.0 (Table 2), which is lower than that reported by Rao *et al.* (1987). Signs of Staphylococcal infection in human beings are suppurative diseases, pyogenic lesions on the skin, septicemia, food poisoning signs which include nausea, vomiting, abdominal pain and diarrhoea, and pneumonia (1980 and Acha and Szyfres, 1991).

Table (3) showed that *T. verrucosum* could be isolated from apparently healthy sheep at a percentage of 3.33, which is lower than that reported by Seddek *et al.* (1994), and higher than that recorded by Ali-Shtayeh *et al.* (1989) and Haggag (1998). In addition, *T. verrucosum* was isolated from apparently healthy goats at a percentage of 4.0 (Table 3), which is agreement with that recorded by Ali-Shtayeh *et al.* (1988) and Haggag (1998), and lower than that reported by El-Sayed (1980). *T. verrucosum* infection in sheep manifested by skin lesions in the form of grayish crusty and alopecic areas in the head and face. In goats the lesions were typically circular and frequently occurred on the ear pinna with active center (Acha and Szyfres, 1991).

T. verrucosum was recovered from human patients at a percentage of 8.0 (Table 3). This result is similar to that obtained by Greatorex (1988), and lower than the result recorded by Abou-Eisha and El-Attar (1994) and Khosravi *et al.* (1994), while higher than that reported by Ali-Shatyeh and Arda (1986), and Spiewak and Szostak (2000).

T. mentagrophytes could be isolated from apparently healthy sheep at a percentage of 2.67 (Table 3), which is nearly similar to that reported by Haggag (1998), and lower than that recorded by Mitra *et al.* (1998).while higher than the result obtained by Ali-Shtayeh *et al.* (1989) and Seddek *et al.* (1994). In addition, *T. mentagrophytes* was isolated from apparently healthy goats at a percentage of 6.0 (Table, 3), which is similar to that recorded by Ali-Shtayeh *et al.* (1988) and higher than that obtained by Haggag (1998) and Mitra *et al.* (1998).

As shown in Table (3) *T. mentagrophytes* could be isolated from human patients at a percentage of 6.0. This result is agree with that recorded by Al-Sogair *et al.* (1991), and lower than that obtained by Greatorex (1988), Nowicki et al (1994), and Costa (1999), while higher than the result reported by Ali-Shtayeh and Arda (1986), Khosravi *et al.* (1994) and Ingordo *et al.* (2000). *T. mentagrophytes* causes an inflammatory infection on the body of human beings and as uncommon infection in tinea cruris and confined to the thigh. Also, *T. mentagrophytes* was widely detected by many authors in tinea pedis cases (Cervetti *et al.*, 1992). It produce more acute intensely inflamed and irritating lesions that caused by any of the anthropophilic species.

T. violaceium was isolated from human beings at a percentage of 16.0 (Table, 3), which agree with that reported by Haggag (1998), and lower than that obtained by Ali-Shtayeh and Arda (1986), and Ali-Shtayeh *et al.* (1998), while higher than the result recorded by Lupa *et al.* (1999). *T. violaceium* is the most important anthropophylic dermatophyte causing tinea capitis, tinea corporis as well as encountered in many cases of tinea cruris (Ali-Shtayeh *et al.*, 1998).

Table (3) revealed that *T. rubrum* could be isolated from human beings at a percentage of 10.0, which is nearly similar to that obtained by Haggag (1998), and lower than that recorded by Greatorex (1988), Costa (1999), and Lupa *et al.* (1999), while higher than the result reported by *Ingordo et al.* (2000). *T. rubrum* is frequently encountered in many cases of tinea cruris causing an annular lesions in the crural folds and on the inner aspect of one or both thighs. Also, it is frequently encountered in tinea pedis where the infection occur as scaly erythematous lesions between the toes, sole and the sides and top of the foot (Kwon-Chung and Bennett, 1992).

Epidermophyton floccosum could be isolated from human beings at a percentage of 4.0 (Table 3), which is agreement with that recorded by Cervetti *et al.* (1992), and *Haggag (1998)*, and higher than the result reported by *Ingordo et al. (2000). Epidermophyton floccosum is* an anthropophylic dermatophytes, encountered in many cases of tinea pedis and tinea corporis (Al-Sogair *et al.*, 1991 and Cervetti *et al.* 1992).

Table (3) showed that *M. canis* was isolated from human beings at a percentage of 18.0%, which is agreement with that obtained by Khosravi *et al.* (1994) and Haggag (1998), and lower than that reported by Nowicki *et al.* (1994), while higher than the result recorded by Costa (1999), and Spiewak and Szostak (2000). The existence of *M. canis* infection among animals as well as the dissemination of the infected scales and hairs in the environmental surroundings are the most attributable factors responsible for the predominance of *M. canis* and its wide spread distribution among human beings. (Ogbonna *et al.*, 1986).

C. albicans could be isolated from apparently healthy sheep at a percentage of 1.33 (Table 3). This result is agree with that recorded by *Haggag* (1998), and lower than that reported by Seddek *et al.* (1994). In

addition, Table (3) revealed that *C. albicans* was isolated from apparently healthy goats at a percentage of 2.0, which is nearly similar to that reported by Haggag (1998).

C. albicans was isolated from human patients at a percentage of 10.0 (Table 3), which is lower than that obtained by Greatorex (1988), and Cervetti et al (1992), while higher than the result reported by Pliego-Castaneda *et al.* (2000). C. *albicans* was almost exclusively found in conjunction with chronic paraonychia and onychomycosis (Acha and Szyfres, 1991).

From the aforementioned results, it can be concluded that pathogenic bacteria and fungi isolated from clinically infected and apparently healthy sheep, goats and human beings were similar in their cultural characteristics, microscopical examination and biochemical reactions. So, clinically infected and apparently healthy sheep and goats play an important role as a reservoir of some pathogenic bacteria and fungi, which could be transmitted from such animals to human contact or consumers. Accordingly, the following hygienic measures should be adapted including hand washing after handling of infected animals and immersion in mild antiseptic after washing, pasteurization or boiling of milk for human consumption, hygienic disposal of animal excreta, recognization and rapid treatment of the diseased sheep and goats, vaccination of sheep and goats against infectious diseases, isolation and slaughtering of seropositive reactors for brucellosis.

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ueiee	non of brucer	ia inteci	100.										
	No. of			Serological tests									
	examined	RBPT		BAPAT		Rivanol test							
Origin	samples	+ve	%	+ve	%	1/25	1/50	1/100	1/200	1/4			
Sheep	150	4	2.67	5	3.33	_	t	1	1	1			
Goats	50	0	0.00	1	2.00	-	-	-	1				
Human beings	50	1	2.00	0.00	0.00	-		1		-			

 Table 1: Results of three serological tests on sera of sheep, goats and human beings for detection of Brucella infection.

RBPT = Rose Bengal plate test.

BAPAT = Buffered acidified plate antigen test.

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	SHEEP (No. = 150)		GO (No.	ATS = 50)	HUMAN BEINGS (No. = 50)	
Isolates	+ve	%	+ve	%	+ve	%
E. coli	92	61.33	29	58.0	30	60.0
Salmonella spp.	4	2.67	I	2.0	4	8.0
S. typhimurium	3	2.00	1	2.0	2	4.0
S. enteritidis	1	0.67	-	0.0	1	2.0
S. typhi		0.00		0.0		2.0
Shigella flexneri	2	1.33	-	0.0	2	4.0
Staph. Aureus	3	2.00	-	0.0	3	6.0
Total	101	67.33	30	60.0	39	78

 Table 2: Pathogenic bacteria isolated from rectal swabs of sheep, goats and stool samples of human beings.

 Table 3: Pathogenic fungi isolated from wool and hair samples of apparently healthy sheep and goats, and hair and scale samples of human patients.

	SHEEP (No. of examined samples = 150)		GOA (No. of er sample	ATS camined s = 50)	HUMAN BEINGS (No. of examined samples = 50)		
Isolates	+ve	%	+ve	%	+ve	%	
T. verrucosum	5	3.33	2	4.0	4	8.0	
T. mentagrophytes	4	2.67	3	6.0	3	6.0	
T. violaceium	_	0.00	_	0.0	8	16.0	
T. ruhrum	-	0.00		0.0	5	10.0	
M. canis	Į _	0.00	-	0.0	9	18.0	
Epidermophyton) _	0.00	_	0.0	2	4.0	
floccosum	2	1.33	1	2.0	5	10.0	
Candida alhicans							
Total	11	7.33	6	12.0	36	72.0	