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MICROBIOLOGICAL EVALUATION OF SHEEP'S AND GOAT'S MILK IN ASSIUT GOVERNORATE (With 9 Tables)

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التقييم الميكروبيولوجي للبن الأغنام والماعز في محافظة أسيوط

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تم جمع ١٠٠ عينة من لبن الأغنام والماعز من أماكن وقرى مختلفة بمحافظة أسيوط وذلك لفحصها ميكروبيولوجيا. وقد تم تقدير العدد الكلى للميكروبات ، الكوليفورم ، الفيكالكوليفورم ، الايشيريشيا كولاي (*E. coli*) ، الانتيروكوكاي ، المكور العنقودي الذهبى والخمائر والفطريات بالإضافة إلى عزل المكورات العنقودية الأخرى ووجد الميكروبات اللاهوائية. وقد أوضحت النتائج أن متوسط العدد الكلى للميكروبات كان 1.4×10^7 ، 1.2×10^6 لكل مل بالنسبة للبن الأغنام والماعز على الترتيب ، بينما تم عزل الكوليفورم ، الفيكالكوليفورم و *E. coli* من ١٠٠ ، ٥٨ ، ١٢% من لبن الأغنام على التوالي ومن ٩٨ ، ٨٢ ، ١٠% من لبن الماعز على الترتيب ، وقد توأجت هذه البكتريا بأعداد متفاوتة. كذلك تم تقدير العدد الكلى للانتيروكوكاي (*Enterococci*) في العينات المفحوصة وكان متوسط عددها 1×10^6 /مل في ٤٦% من لبن الأغنام ، 9×10^4 /مل في ٨٠% من لبن الماعز. كذلك تم عزل المكور العنقودي الذهبى من ٤٢ ، ٨٤% بمتوسط عددي 2×10^4 ، 1×10^3 /مل من هذه الألبان على التوالي . وقد توأجت ميكروب الـ *Staph. epidermidis* ، الـ *Micrococci* في ٥٢ ، ١٦% من عينات لبن الأغنام، بينما تم عزلها من ٣٤ ، ٤٠% من عينات لبن الماعز بالترتيب. كذلك توأجت الميكروبات اللاهوائية في ٦٢ ، ٣٢% من العينات المفحوصة على التوالي. ويتقدير العدد الكلى للخمائر والفطريات وجد أن متوسطها كان 2×10^4 ، 1×10^3 /مل من لبن الأغنام والماعز على التوالي. وقد نوقت الخطورة الصحية لهذه الميكروبات على الصحة العامة وكذلك الشروط والخطوات التي يجب إتباعها لمنع تلوث هذه الألبان بهذه الميكروبات.

SUMMARY

One hundred random samples of sheep's and goat's milk were collected from different localities and villages in Assiut province. The samples were examined microbiologically for Aerobic plate, Coliforms, Fecal coliforms, *E. coli*, Enterococci, *Staph. aureus* and total yeast and mold counts as well as isolation of other *Staphylococci* and detection of anaerobes. The obtained results reveal that the average counts of total bacteria were 14×10^9 and 12×10^8 /ml of sheep's and goat's milk samples, respectively. Coliforms, Fecal coliforms and *E. coli* were existed in 100, 58 and 12% of sheep's milk samples and in 98, 82 and 10% of goat's milk samples, respectively. These organisms were detected in variable numbers. Enterococci were enumerated in 46 and 80% of the examined sheep's and goat's milk with average counts of 1×10^5 and 9×10^4 /ml, respectively. *Staph. aureus* could be detected in 42 and 84% of the examined sheep's and goat's milk samples with average counts of 2×10 and 1×10^3 /ml of the samples, respectively. *Staph. epidermidis* were isolated from 52 and 34% of sheep's and goat's milk samples, respectively. While, micrococci were existed in 16 and 40% of the samples of both types, respectively. Anaerobic bacteria were detected using stormy fermentation test in 62 and 32% of the examined sheep's and goat's milk samples, respectively. Total yeasts and molds, were detected in all of the examined sheep's and goat's milk samples with average counts of 2×10^4 and 1×10^3 /ml of the samples, respectively. The public health hazard and preventive measures were discussed.

Key words: Sheep's and Goat's milk

INTRODUCTION

In recent years, sheep's and goat's milks have become of growing concern and important role in human diet all over the world. Such milk can fulfill the increasing demands on foods accompanied the unlimited growing number of population. However, there is no production of sheep's and goat's milk for sale in many countries including Egypt. Most of such milks produced and consumed locally in villages by large number of people.

Unlike cow's or buffalo's milk which have stringent hygiene and other regulations that control their production, distribution and

processing. Sheep's and goat's milks are still in need to more concern and regulations to control their production under hygienic measures. Consumption of raw sheep's and goat's milk by relatively large number of people may represent a public health hazard. It has been reported that sheep's and goat's milk could be a reservoir of staphylococci and streptococci. Also, such milk could transmit tuberculosis, brucellosis and salmonellosis (Campell and Marchall, 1975; Anon, 1983; El-Leboudy and Gamel, 1994 and Cosentino and Palmas, 1997). Moreover, it has been recorded that goat's milk has shown to transmit Louping-ill virus (Reid *et al.*, 1984).

Although, the extensive research works that have dealt with hygienic production and microbiological evaluation of cow's and buffalo's milk, there are relatively few investigations concerned with hygienic production and quality of sheep's and goat's milk (Chubb *et al.*, 1985; Anyam and Adekeye, 1995 and Cosentino and Palmas, 1997). This paper gives data on the microbiological quality of raw sheep's and goat's milks available at producers level in some villages in Assiut province.

MATERIAL and METHODS

Collection of samples:

One hundred samples of raw sheep's and goat's milk were collected randomly (50 samples each) from different localities and villages in Assiut province. Each sample (250 ml) was collected in a sterile glass stoppered bottle. The samples were surrounded by ice in an insulated box and transported to the laboratory with a minimum of delay. In the laboratory, the samples were mixed thoroughly and prepared for microbiological examination according to A.P.H.A. (1985).

Microbiological examination:

- 1- Aerobic plate count was determined according to A.P.H.A. (1985).
- 2- Coliforms and Fecal coliforms (MPN/ml) was performed according to A.O.A.C. (1975).
- 3- Test for *Escherichia coli* was done according to A.O.A.C. (1975).
- 4- Enterococci count was estimated by using K.F. medium as described by Deibel and Hartman (1976).
- 5- Enumeration and isolation of staphylococci and micrococci.

- a) Enumeration of coagulase positive *Staph. aureus* was performed using Baird-Parker's medium (Baird-Parker, 1962), using surface plating technique (Thatcher and Clark, 1975).
- b) Isolation of other staphylococci and micrococci was carried out according to Finegold and Martin (1982).
- 6- Detection of anaerobes was done using stormy fermentation test as described by Cruickshank *et al.* (1969).
- 7- Total yeast and mold count was carried out according to Harrigan and Margaret (1976).

RESULTS

The obtained results are recorded in Tables 1 - 9.

DISCUSSION

The recorded results in Table 1 reveal that all of the examined samples of sheep's and goat's milk contained viable bacterial counts of 4.2×10^7 and 5.3×10^3 /ml as a minimum and 8×10^{10} and 7×10^9 /ml as a maximum with average counts of 14×10^9 and 12×10^8 /ml of the examined samples, respectively. Lower numbers of total bacteria in sheep's milk were recorded by Pla *et al.* (1992). Also, lower findings of viable counts in goat's milk were reported by Abo El-Naga *et al.* (1985). While, Collinge (1985) stated that the recommended total counts of cooled goat's milk must be less than 50,000/ml. The relatively high counts of total bacteria in the examined samples reflects the neglected hygiene adopted during milking and handling of such milks.

The data presented in Tables 2 and 3 prove that all of the examined samples of sheep's milk were contaminated by coliforms in numbers ranged from 10^2 - $> 10^3$ /ml. Most of the examined samples (74%) had coliforms in numbers exceeding 10^3 /ml. Fecal coliforms were existed in 58% of the examined samples in numbers varied from < 10 - 10^3 /ml. Most of positive samples (37.93%) had counts of 10 - 10^2 /ml. The rest of positive samples were equally distributed among < 10 and 10^2 - 10^3 /ml (31.03% each). Also, the results point out that 12% of the examined samples of sheep's milk contained *E. coli* in numbers of less than 10/ml.

Concerning goat's milk as presented in Tables 2 and 4, coliforms existed in 98% of the examined samples in numbers varied from 10^1 - 10^3 /ml. The highest frequency distribution (46.94%) lay within the range of 10^2 - 10^3 , while 34.69% of the positive samples had counts more than 10^3 /ml. Fecal coliforms contaminated 82% of the examined samples in variable numbers. Most of the positive samples (43.90%) had fecal coliforms in numbers varied from 10^1 - 10^2 , while 19.51% of positive samples had fecal coliforms over 10^3 /ml. *E. coli* existed in 10% of the examined samples in numbers ranged from $< 10^1$ - 10^2 /ml. Most of the positive samples (80%) had *E. coli* below 10/ml.

Lower incidence of coliforms in goat's milk was obtained by Sethi *et al.* (1974) and Roberts (1985). Jensen and Hughes (1980) obtained lower incidence of *E. coli* in goat's milk, and quite different findings concerning coliforms, that 49% of the examined samples had coliforms of 10/ml and most of the positive samples had *E. coli* below 100/ml. The relatively higher incidence of coliforms in the examined sheep's and goat's milk samples besided the existence of fecal coliforms and *E. coli* should be considered a real indicator of fecal pollution with possible existence of associated pathogens. Furthermore, presence of coliforms and fecal coliforms beyond certain level could be of public health hazard, as they may cause dreadful diarrhea disease (Robert *et al.*, 1977). Also, the public health hazard of *E. coli* has been emphasized by several investigators, as they have been implicated in human cases of gastroenteritis & epidemic diarrhea in infants, as well as, in cases of food poisoning (Marier *et al.*, 1973 and Mossel, 1975).

The results in Table 5 point out that enterococci could be detected in 46% of the examined samples of sheep's milk in counts ranged from 1×10^2 to 1×10^6 with an average of 1×10^3 /ml. While, goat's milk reveal a high incidence of enterococci (80%), and the counts ranged from 5×10^2 to 7×10^5 with an average counts of 9×10^4 /ml. Enterococci could be isolated from goat's milk examined by Tzanetakis and Tzanetaki (1989). The public health hazard of enterococci can not be denied as they have been implicated in several cases of food poisoning outbreaks (Seidel and Muschter, 1967 and I.C.M.S.F., 1978).

It is evident from Table 6 that *Staph. aureus* could be detected in 42% of the examined samples of sheep's milk in numbers varied from < 100 to 2×10^2 with average counts of 2×10^1 /ml. While, goat's milk show a higher incidence of *Staph. aureus* (84%). The counts of the

organism ranged from <100 to 3×10^4 with average numbers of 1×10^3 /ml. The results presented in Table 7 prove that *Staph. epidermidis* and micrococci could be isolated from 52 and 16% of the examined sheep's milk samples, respectively. Concerning goat's milk (Table 7), *Staph. epidermidis* and micrococci were detected in 34 and 40% of the examined samples, respectively. *Staph. aureus*, *Staph. epidermidis* and micrococci could be isolated from ewe's milk examined by Saad and Zaki (1993); Anyam and Adekeye (1995); Burriel (1997) and Cosentino and Palmas (1997). Also, these organisms could be isolated from goat's milk examined by Sethi *et al.* (1974); Mallikeswaran and Padmanaban (1989) and Deinhofer and Pernthaner (1993). Food poisoning outbreaks due to *Staph. aureus* have been well documented and reported by several investigators (Volvich *et al.*, 1964; Enhuber, 1971 and Galbraith *et al.*, 1982).

The results in Table 8 point out that anaerobic bacteria could be detected in 62 and 32% of the examined samples of sheep's and goat's milk, respectively. While, the data presented in Table 9 show that all of the examined samples of sheep's and goat's milk were contaminated by yeasts and molds. Their counts varied from 4×10 and 1×10 /ml as minimum and 2×10^5 and 2×10^4 /ml as maximum with average counts of 2×10^4 and 1×10^3 /ml of the both types of the examined samples, respectively.

Contamination of all of the examined samples of sheep's and goat's milks by yeasts and molds besides existence of anaerobic bacteria reflects the bad hygiene adopted during milking and handling of such milks.

It is worthwhile to state that, the results of such study prove that sheep's and goat's milks are still in need to more concern and regulations to control their production under hygienic measures, as well as, educational programs for owners on the hygienic measures which must be taken during milking and handling of such milks

Table 1: Aerobic plate count in the examined samples of sheep's and goat's milk.

Examined Samples	Positive samples		Counts / ml		
	No./50	%	Min.	Max.	Average
Sheep's milk	50	100	4.2×10^7	8×10^{10}	14×10^9
Goat's milk	50	100	5.3×10^3	7×10^9	12×10^8

Table 2: Incidence of coliforms, fecal coliforms and *E. coli* in the examined samples of sheep's and goat's milk.

Examined Samples	Positive samples					
	Coliforms		Fecal coliforms		<i>E. coli</i>	
	No./50	%	No./50	%	No./50	%
Sheep's milk	50	100	29	58	6	12
Goat's milk	49	98	41	82	5	10

Table 3. Frequency distribution of positive samples of sheep's milk based on their coliforms, fecal coliforms and *E. coli* counts (MPN/ml).

Counts / ml	Positive samples					
	Coliforms		Fecal coliforms		<i>E. coli</i>	
	No./50	%	No./29	%	No./6	%
< 10	0	0	9	31.03	6	100
10 - 10 ²	0	0	11	37.93	0	0
10 ² - 10 ³	13	26	9	31.03	0	0
> 10 ³	37	74	0	0	0	0
Total	50	100	29	100	6	100

Table 4. Frequency distribution of positive samples of goats's milk based on their coliforms, fecal coliforms and *E. coli* counts (MPN/ml).

Counts / ml	Positive samples					
	Coliforms		Fecal coliforms		<i>E. coli</i>	
	No./49	%	No./41	%	No./5	%
< 10	0	0	4	9.76	4	80
10 - 10 ²	9	18.37	18	43.90	1	20
10 ² - 10 ³	23	46.94	11	26.83	0	0
> 10 ³	17	34.69	8	19.51	0	0
Total	49	100	41	100	5	100

Table 5. Incidence and counts of enterococci in the examined samples of sheep's and goat's milk.

Examined samples	No. of examined		Positive samples		Counts / ml	
	samples	No.	%	Min.	Max.	Average
Sheep's milk	50	23	46	1×10^2	1×10^6	1×10^3
Goat's milk	50	40	80	5×10^2	7×10^5	9×10^4

Table 6. Incidence and counts of *Staph. aureus* in the examined samples of sheep's and goat's milk.

Examined samples	No. of examined samples	Positive samples		Counts / ml		
		No.	%	Min.	Max.	Average
Sheep's milk	50	21	42	< 100	2×10^2	2×10
Goat's milk	50	42	84	< 100	3×10^4	1×10^3

Table 7. Incidence of *Staph. epidermidis* and micrococci in the examined samples of sheep's and goat's milk.

Examined samples	No. of Examined Samples	Positive samples			
		Staph. Epidermidis		Micrococci	
		No.	%	No.	%
Sheep's milk	50	26	52	8	16
Goat's milk	50	17	34	20	40

Table 8. Incidence of anaerobes in the examined samples of sheep's and goat's milk.

Examined samples	No. of examined Samples	Positive samples	
		No.	%
Sheep's milk	50	31	62
Goat's milk	50	16	32

Table 9. Total yeast and mold counts in the examined samples of sheep's and goat's milk.

Examined samples	Positive samples		Counts / ml		
	No./50	%	Min.	Max.	Average
Sheep's milk	50	100	4×10^4	2×10^5	2×10^4
Goat's milk	50	100	1×10^4	2×10^4	1×10^3

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