

SUBCLINICAL MASTITIS IN FAYOUM GOVERNORATE

AMANY M.R. KAMEL ¹, M.M. BASSIONY, Y.A. FARAG ¹
AND A. FARID ²

¹ Animal Reproduction Research Institute, Agricultural Research Centre, Giza, Egypt.

² Faculty of Veterinary Medicine, Cairo University .

(Manuscript received 26 November 1995)

Abstract

Subclinical mastitis is a serious form of mastitis which affects the dairy herds, but without any clinical symptoms on the affected animal which usually appears healthy.

In the present study, California mastitis test (CMT) proved to be an efficient rapid field test for detection of subclinical mastitis. When applied on 720 milk samples collected from 180 dairy cows, 320 (44.4%) were positive (++) to the test.

The bacteriological examination of the 320 quarter milk samples revealed that staphylococci were the most predominant (62.8%), followed by streptococci, members of Gram-negative bacilli, *corynebacteria* and *anthracoides* in an incidence of (40.9%), (40.6%), (5.9%) and (1.7%), respectively. However, the season and age of the animals seem to influence the incidence of different secured microorganisms.

The antibiogram of the isolated bacteria revealed that gentamycin followed by chloramphenicol and / or oxytetracycline were the most effective antibiotics.

INTRODUCTION

Healthy mammary glands are essential for the secretion of milk, that is wholesome to drink and produced in sufficient quantity to be profitable to dairy men. Reduction in milk yield and production of abnormal milk is mostly due to mastitis, resulting in considerable economic losses to dairy industry throughout the world .

Since most mastitic cases occur in the subclinical form, the diseased animal continues, for a time, to be a dangerous source of infection.

The present work was conducted to detect cases with subclinical mastitis by rapid field test (CMT), and to enumerate different bacteria associated with it in bovine udders during the various seasons of the year. Moreover, *in vitro* antibiotic sensitivity test on the isolated bacteria was performed to ensure rapid and prompt recovery of affected udder.

MATERIALS AND METHODS

A total of 720 quarter milk samples were collected aseptically during the different seasons of the year from 180 cows with apparently normal udders kept at private and governmental dairy farms in Fayoum governorate.

All samples were subjected to CMT according to Schalm *et al.* (1971), and samples giving (++) and over were considered positive and were bacteriologically examined. The isolated organisms were identified according to Krieg and Holt (1984) and Sneath *et al.* (1986).

In vitro, antibiotic sensitivity test of the isolated bacteria was carried out according to Cruickshank *et al.* (1975).

RESULTS

California mastitis test on 720 milk samples revealed that, 320 (44.4 %) were positive (++ or more) and were considered as subclinical mastitic cases (Table 1).

Table 1. Incidence of subclinical mastitis.

Total No. of examined animals	Total No. of examined quarter milk samples	Apparently healthy quarter samples			
		Positive cases		Negative cases	
		No	%	No	%
180	720	320	44.4	400	55.6

The percentage is correlated to the total number of quarter milk samples 720

Culture examination revealed bacterial growth in the 320 quarter samples in an incidence of (100.0%) represented by (*Staphylococcus* spp., *Streptococcus* spp., Gram-negative bacilli, *Corynebacteria* and *Anthracoidea*) (Table 2), in the following incidence : (57.5%) *Staph. aureus*, (5.3 %) *Staph. epidermidis*, (18.4%) *Str. uberis*, (15.9%), *Str. agalactiae*, (4.4%), *Str. dysgalactiae*, (2.2%), *Str. faecalis*, (5.9%) *C.pyogenes*, (1.7%), *anthracoides*, (27.5%), *E.coli*, (7.2%), *Ps. Aureuginosa*, (3.4%), *P. vulgaris* and (2.5%) *K. pneumoniae* (Table 2) .

Table 2. Bacterial isolates from subclinical mastitic milk samples .

Organisms	No. of Isolates	Incidence %
(Gram positive cocci)		
1- <i>Staphylococcus</i> spp.	201	62.8
* <i>Staph. aureus</i>	184	57.5
* <i>Staph. epidermidis</i>	17	5.3
2- <i>Streptococcus</i> spp.	131	40.9
* <i>Str. uberis</i>	59	18.4
* <i>Str. agalactiae</i>	51	15.9
* <i>Str. dysgalactiae</i>	14	4.4
* <i>Str. faecalis</i>	7	2.2
3- <i>Corynebacteria</i>	19	5.9
* <i>C.pyogenes</i>	19	5.9
	5	1.7
4- <i>Anthracoidea</i>		
5- (Gram negative bacilli)	130	40.6
* <i>E.coli</i>	88	27.5
* <i>P.vulgaris</i>	11	3.4
* <i>K.pneumoniae</i>	8	2.5
* <i>Ps. aureuginosa</i>		7.2

* The percentage is correlated to the total number of examined samples.

The isolation rate was highest during summer months "157 (32.30%)", followed by spring "154 (31.69%)", winter " 105 (21.60%)" and autumn "70 (14.40%)". Moreover, *Staph. aureus* was the most frequently recovered from the subclinical milk samples during different seasons followed by *E.coli*, *Str. agalactiae* and *Ps. aureuginosa* (Table 3).

Table 3. Number and incidence of isolated micro-organisms from subclinical mastitic milk samples during the different seasons of the year.

Isolated micro - organisms	Spring	Summer	Autumn	Winter	Total
<i>Staph aureus</i>	59 (32.06%)	56 (30.35%)	23 (12.5%)	46 (25.0%)	184 (37.86%)
<i>Staph epidermidis</i>	5 (29.41%)	6 (35.29%)	6 (35.29%)		17 (3.50%)
<i>Str. agalactiae</i>	17 (33.33%)	17 (33.33%)	9 (17.64%)	8 (12.86%)	51 (10.49%)
<i>Str. dysgalactiae</i>	7 (50.0%)	7 (50.0%)			14 (2.88%)
<i>Str. uberis</i>	30 (50.89%)			29 (49.15%)	59 (12.14%)
<i>Str. fecalis</i>	3 (42.86%)	4 (57.14%)			7 (1.44%)
<i>C. pyogenes</i>	5 (26.32%)	9 (47.37%)	5 (26.32%)		19 (3.91%)
<i>Anthracoidea</i>	1 (20.2%)	2 (40.0%)		1 (20.0%)	5 (1.03%)
<i>E. coli</i>	13 (14.77%)	45 (51.14%)	20 (22.72%)	10 (11.36%)	88 (18.11%)
<i>Ps. aureginosa</i>	8 (34.78%)	4 (17.39%)	5 (21.74%)	6 (26.09%)	23 (4.73%)
<i>P. vulgaris</i>		7 (63.64%)		4 (36.36%)	11 (2.26%)
<i>K. pneumoniae</i>	6 (75.00%)		2 (25.00%)		8 (1.65%)
Total	154 (31.69%)	157 (32.30%)	70 (14.40%)	105 (21.60%)	488

* The percentage is correlated to the total number of the isolates .

The highest incidence of subclinical mastitis together with the highest rate of isolation of bacteria, 229 (47.2%) out of 486 were met with in the milk samples of cows during the 3rd lactation period. Meanwhile, the lowest recovery rate 102 (20.99 %) was obtained in the first lactation period (Table 4).

The antibiogram of the isolated bacteria was shown in Table 5, where gentamycin, chloramphenicol and oxytetracycline were the most effective .

DISCUSSION

To date, mastitis constitutes a great economic loss to milking herd. Its serious effects are derived from the subclinical form, which is devoid of obvious symptoms and secretes apparently normal contaminated milk for a long time, during which infection can spread in the herd .

In Table 1, it is interesting to note that out of 720 quarter milk samples collected from 180 cows, 320 milk samples showed positive CMT (++ or more) in an

Table 4. Relationship between the age of the apparently normal animals and the isolated micro-organisms from their milk samples.

Isolated micro - organisms	Total	Age of cows with a pparently healthy udders		
		1st lactation period	2 nd lactation period	3 rd lactation period
<i>Staph. aureus</i>	184	32 (17.39%)	62 (33.69%)	90 (48.91%)
<i>Staph. epidermidis</i>	17	3 (17.65%)	5 (29.41%)	9 (52.94%)
<i>Str. uberis</i>	59	12 (20.34%)	18 (30.51%)	29 (49.16%)
<i>Str. agalactiae</i>	51	10 (19.61%)	14 (24.45%)	27 (52.94%)
<i>Str. dysgalactiae</i>	14	3 (21.43%)	5 (35.71%)	6 (42.86%)
<i>Str. feacalis</i>	7	2 (28.57%)	2 (28.57%)	3 (42.86%)
<i>C. pyogenes</i>	19	6 (31.58%)	6 (31.58%)	7 (36.84%)
<i>Anthracooides</i>	5	2 (20.00%)	2 (40.00%)	1 (20.00%)
<i>E. coli</i>	88	20 (22.73%)	26 (29.45%)	42 (47.73%)
<i>Ps. aureginosa</i>	23	7 (30.43%)	9 (39.13%)	7 (30.43%)
<i>P. vulgaris</i>	11	3 (27.27%)	3 (27.27%)	5 (45.45%)
<i>K. pneumoniae</i>	8	2 (25.00%)	3 (37.50%)	3 (37.50%)
Total	486	32 (17.39%)	155 (31.92%)	229 (47.12%)

* The percentage is correlated to the total number of isolates.

Table 5. In - vitro antibiogram of isolated bacteria from milk of subclinical mastitic cows.

Antimicrobial Agents	Staphylococcus spp. (201)						Streptococcus spp. (131)						Gram-Negative enteric bacilli. (107)						C. Pyogenes. (19)					
	R			S			R			S			R			S			R			S		
	No	%	I	No	%	I	No	%	I	No	%	I	No	%	I	No	%	I	No	%	I	No	%	I
Ampicillin	131	65.2	6	3.0	64	31.8	79	60.3	11	8.4	41	31.3	32	30.0	12	10.8	63	59.2	10	52.6	31	5.8	6	31.6
Chloramphenicol	7	3.5	4	2.0	190	94.5	2	1.5	4	3.1	125	95.4	9	8.5	12	10.8	89	80.8	1	5.3	2	10.5	16	84.2
Erythromycin	20	2.0	34	16.9	147	73.1	68	51.9	4	3.1	59	45.0	54	50.8	29	28.9	24	22.3	7	36.8	5	26.3	7	36.8
Gentamycin		0.0		0.0	201	100.0		0.0	3	2.3	128	97.7	3	3.1	8	7.7	96	89.7		0.0	1	5.3	18	84.3
Neomycin	22	10.9	9	4.5	170	84.6	64	48.9	21	16.0	46	35.1	58	54.6	19	17.7	30	27.7	13	68.4	4	21.1	2	10.5
Oxytetracycline	17	8.5	37	18.4	147	73.1	14	10.7	11	8.4	106	80.9	26	23.9	25	23.4	56	52.3	2	10.5	4	21.1	13	68.4
Penicillin G	151	75.1	7	3.5	43	21.4	66	50.4	19	14.5	46	35.1	94	87.7	7	6.9	6	5.4	6	31.6	3	15.8	10	52.6
Streptomycin	89	44.3	19	9.5	93	46.3	54	41.2	36	27.5	41	31.3	32	30.0	19	17.7	56	52.3	8	42.1	8	42.1	3	15.8
Sulphamethoxazole/Trimethoprim		0.0		0.0	201	100.0	93	71.0	16	12.2	22	16.8	54	50.8	20	18.5	33	30.8	13	68.4	3	15.8	3	15.8

R = Resistant

I = Intermediate

S = Susceptible

incidence of (44.4%), and this result nearly coincides with the results obtained by Fustes *et al.* (1985) and Lau *et al.* (1986).

On the other hand, bacteria were isolated from 320 (100.0 %) samples, and similar results were obtained by Misra *et al.* (1973), Maihotra *et al.* (1976), Verma *et al.* (1978), Rao *et al.* (1981), Anderson *et al.* (1982) and Kalorey *et al.* (1983).

Of different isolated bacteria, *Staphylococcus* spp. was the most prevalent with an incidence of (62.8%), followed by *Streptococcus* spp., members of Gram-negative bacilli, *Corynebacterium* spp. and *anthracoides* with an incidence of (40.9%), (40.6%), (5.9) and (1.7%), respectively (Table 2). The aforementioned data are similar to those reported previously by Kalorey *et al.* (1983), Misra *et al.* (1973), Abd-El-Karim and El-Ashmawy (1979) and Abd-El-All (1989).

The effect of the season of the year was distinctly clear on both the incidence of subclinical mastitis and the recovery of mastitic pathogens. Table 3 shows that, the highest incidence was during summer (32.30 %), followed by spring (31.69%), winter (21.60 %) and autumn (14.40 %) which are in agreement with Abd-El-All (1989). However, the effect of age on the increased incidence of subclinical mastitis may be attributed to the prolonged exposure of older cows to infection, as well as the lowering of the resistance with advancing age.

Finally, the in vitro antibiogram of bacteria revealed that, gentamycin followed by chloramphenicol and/or oxytetracycline were the most effective, and could be used with confidence in treatment of subclinical mastitis.

REFERENCES

1. Abd-El-All, B.E. 1989. Studies on bacteria causing mastitis in cattle in Assiut. Thesis, Ph. D. Sc., Assiut Univ.
2. Abd-El-Karim, A.M. and El-Ashmawy. A.A. 1979. Diagnosis of subclinical mastitis in Iraqi dairy cattle. Assuit Vet. J., 6 : 282 - 286 .
3. Anderson, K.L., A.R. Smith, B.K. Gustaffon, S.L. Spahr and H.L. Whitmore. 1982. Diagnosis and treatment of acute mastitis in large dairy herd. J. Am. Vet. Med. Assoc., 181 : 690 - 693 .
4. Cruickshank, R., J.P. Duguid, B.P. Marmion and R.H.A. Swain. 1975. Medical Microbiology. 12 th Ed., Vol. 11 Churchill Living Stone, London, U.K.
5. Fustes, E. C., Avila and L. Ortega. 1985. Bovine mastitis effect on milk production and the livestock economy in Cuba. Revista de Salud Ani, 7 (1) : 91 - 100. Vet. Bull., 56 Abst. No 744. pp. 88 .
6. Kalorey, D.R., J.H. Purhit and P.M. Dholokia. 1993. Studies on the incidence of subclinical mastitis, its etiology and in-vitro sensitivity of isolates. Ind.J. Ani.Sci., (9) : 961 - 963 .
7. Krieg, N.R. and J.G. Holt. 1984. Bergey's Manual of Systematic bacteriology. Vol. 1, Williams and Wilkins, London, U.K .
8. Lau, H.D., N.P. Singh and S.J. Hess. 1986. Comparison of indirect test for diagnosis of subclinical mastitis in buffaloes. Boletim de Pesquisa, Embrapa, Centro de Pesquisa Agropecuaria do Trópico Unido, No 77, 13 pp. Vet . Bull., 57 , Abst. No. 2532. pp. 354.
9. Malhotra, B.R., D.R. Ghodekar and V.K.N. Nembudripad. 1976. Studies on Leucocyte count of buffalo milk. Ind. J. Dairy Sci., 29 : 59 - 61 .
10. Misra, P.K., S.N. Panda and S.K. Misra. 1973. Incidence and etiology of subclinical bovine mastitis in Orissa. Ind. J. Anim. Hlth, 12 : 175 - 180 .
11. Rao, K.P.S. , E.G. Jagannath and V.K.N. Namburipad. 1981. Use of a new mastitis detector card (a modification of bromothymol blue card) for early detection of mastitis in milking herds. Ind . Vet. J. , 58 : 326-329.

12. Schalm, O.W., J.E. Carrol and N.C. Jain. 1971. Bovine mastitis. 1st Ed., Leaand Febiger, Philadelphia, U.S.A.
13. Sneath, P.H.A. , N.Mair, M.F. Sharpe and J.G. Holt. 1986. Bergey,s Manual of Systematic Bacteriology, Vol. 2 , Williams and Wilkins, London U.K.
14. Verma, T.N. L.N. Mandal and B.K. Sinhn. 1978. Studies on subclinical mastitis with special reference to bacterial correlation and its public health importance. Ind. J. Public. Hlth., 22 : 249-253. Vet . Bull., 50, Abst. No. 3916. pp. 543 .

التهاب الضرع تحت الاكلينيكي في محافظة الفيوم

١٢. Schalm, G.W. and W.C. Ross. 1981. *Textbook of Veterinary Clinical Medicine*. 1st Ed. Lea and Febiger, Philadelphia, U.S.A.
١٣. Snieszko, P.H.A., N.M. Malik, M.E. Ghany and J.G. Holt. 1985. *Practical Manual of Systematic Bacteriology*. Vol. 3. Williams and Wilkins, London, U.K.
١٤. Verma, T.N., L.N. Mandal and B.K. Sinha. 1978. *Studies on subclinical mastitis with special reference to bacterial correlation and its public health importance*. Bull. Vet. Res. 25: 20-25.
١٥. معهد بحوث التناسليات - مركز البحوث الزراعية - الجيزة - مصر.
١٦. كلية الطب البيطري - جامعة القاهرة - الجيزة .

يعتبر التهاب الضرع تحت الاكلينيكي واحدا من أخطر أنواع التهابات الضرع التي تؤثر في القطعان الحلابة بدون أية أعراض إكلينيكية على الحيوان الذي يظهر عادة بشكل صحي سليم.

شمل هذا البحث اختبار كاليفورنيا للتشخيص السريع لاحتمال وجود التهاب الضرع تحت الإكلينيكي، وقد أظهرت النتائج ان ٣٢٠ عينة لبن بنسبة (٤٤,٤%) كانت إيجابية للاختبار (++) من إجمالي ٧٢٠ عينة لبن جمعت من ١٨٠ بقرة حلب.

ولقد تم فحص ٣٢٠ عينة لبن بكتريولوجيا ، ووجد أن الميكروب العنقودي هو السائد بنسبة (٦٢,٨%) ويليه الميكروب السبحي والميكروبات الجرام والكريتي باكتريا والانثراكويدز بنسبة (٤٠,٩%) (٤٠,٦%) ن (٥,٩%) ن (١,٧%) على التوالي .

ولقد وجد أن الموسم وكذلك عمر الحيوان لهما تأثير على مختلف الميكروبات المعزولة من هذه الحالات ، كما أجريت اختبارات حساسية للبكتريا المعزولة من العينات المصابة بالتهاب الضرع تحت الإكلينيكي وكان الجنتاميسين ويلي الكلورامفينيكول و الأوكسي تتراسيكلين هي الأكثر فاعلية بصفة عامة.