Bacteriological and pharmacological studies on clinical mastitis in cows at Ismailia province

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Abstract

This study was performed to identify the bacterial agents causing clinical mastitis in cows and studying their antibiotic resistance patterns as well as determination of some biochemical alterations in blood and milk and application of certain field treatment regimens. A total number of 113 cows (103 mastitic and 10 apparently healthy) belonging to dairy farms at Ismailia Governorate. The results of bacteriological examination revealed that *E.coli* was isolated in high percentage (45.71%) followed by ,*Staphylococcus aureus, Streptococcus aglactiae, Streptococcus uberis, Enterobacter aerogenes, Corynebacterium pyogenes (Arcenobacterium), Klebsiella pneumoniae and Proteus mirabilis* in percentage (16.57% 12.57%, 9.14%, 5.14%, 2.28%, 3.42% and 5.14). The result of serum biochemical analysis showed that Ca, Zn and Fe levels of serum in mastitic cows were significantly lower than healthy normal cows, but Cu value were higher ($P \leq 0.01$) than normal cows. No significant difference were observed in serum Mg between healthy and mastitic cows. While Calcium level of milk of mastitic cows were significantly ($P \leq 0.01$) lower than that of healthy normal one, in contrast Fe and Zn was significantly higher than normal cows. Mg level was not significantly different.

All isolated bacteria were sensitive to Ceftiofur, Ciprofloxacin and Enrofloxacin. While most of them were resistant to Penicillin and Amoxicillin as well as intermediate to Streptomycin and Neomycin. Field treatment based on antibiotic sensitivity showed that administration of Ceftiofur hydrochloride 125 mg intra-mammary was better than the negative control, and the 5-days Ceftiofur treatment regimens have higher bacterial cure rates than the standard 2-days Ceftiofur treatment regimen.

Periodical bacteriological examinations of milk are recommended and caring of the used instrument to avoid spreading of the infection among cows, good hygienic milking practices for preventing new infections and using appropriate antibiotic treatment for clinical cases to reduce the prevalence of infection .

Introduction

Mastitis continues to be the most frequent and costly disease of dairy cattle. Financial losses occur for both sub-clinical and clinical stages of the disease. Losses caused by clinical mastitis include discarded milk, transient reductions in milk yield and premature culling

(Fetrow, 2000). Clinical cases of mastitis are often severe; systemic signs are present and the condition is obviously painful. Clinically affected glands frequently suffer partial or complete damage and do not resume normal function. Additional losses associated with clinical mastitis are the costs of treatment and culling of cows due to permanent udder damage .In very severe cases, gangrene may develop in the mammary gland and the animal may die. Thus, mastitis has a major impact on both economy and animal welfare. Bacteriological culture of milk is the gold standard method for, determining the cause of clinical mastitis in dairy cows (Ding well et al., 2003).

Although, a wide range of microorganisms may cause mastitis, most cases are reported to be due to *staphylococci* (Adam et al., 2007), *Streptococcus agalactiae, Escherichia coli, Corynebacterium bovis and Klebsiella spp.* (Tenhagen et al., 2006)

Mastitis cause an increase in passage of plasma protein due to increase vascular permeability as a consequence of chemotaxis of leukocytes to inflamed tissue resulting in an increased Na, Cl, P and decreased K also, reduced synthesis of fat, casein and lactose.This done because of cell degeneration in the mammary tissue (Hamit and Erdal , 2005).

Determination of Ca and Mg in mastitic milk may give important clues about the status of mastitis. Administration of Zn and Cu during mastitis treatment might be helpful for controlling and preventing mastitis by increasing the phagocytic activity of polymorphonuclear and mononuclear cells in the udder . It was also suggested that Zn given in feed contributes to the formation of creatinin which prevents the entry of bacteria via the teat canal (Burton and Erskine , 2003).

Increasing antimicrobial resistance has become a serious concern worldwide and antimicrobial use in animal agriculture is currently under scrutiny. Mastitis is the most common reason for antibiotic use in dairy herds and thus, antimicrobial resistance of mastitis pathogens has received more attention (Rajala-Schultz et al., 2004).

The principles of successful treatment of clinical mastitis should include early detection of mastitis, isolation and identification of the pathogen and the antibiotic sensitivity test of the isolated microorganisms.

The objective of this work was to identify the causative bacterial agents of clinical mastitis in cows and to study their antibiotic resistance patterns as well as determination of some biochemical alterations in blood and milk associated with mastitic cows and application of certain field treatment regimens.

Materials and methods

Animals :

This study was carried out on 113 cows (103 mastitic and 10 apparently healthy cows) in some dairy farms at Ismailia governorate.

Animals showed the signs of mastitis one week to two month after parturition.

Apparently healthy cows subjected in this study were proved to be free from subclinical mastitis by California mastitis test.

Clinical examination :

All the examined cows were subjected to clinical examination for the presence or absence of any systemic reaction. Physical inspection and manual palpation of the affected quarters were carried out.

Samples:

A- Milk samples: About ten milliliters of milk were collected into separate tubes from each affected quarter and quarters of apparently healthy cows after the udder was thoroughly washed with clean warm water, then dried with clean paper towel. The teats orifice was disinfected by ethyl alcohol 70%, after that the first few squirts of milk was discarded then two milk samples from each infected quarter were collected for bacteriological and chemical examination. All samples were transported to the laboratory in ice box.

B- Blood samples: (10 ml) of blood were collected from the jugular vein under aseptic conditions from 10 apparently healthy and from 25 mastitic cows. They were kept for separation of serum at room temperature for two hours. Then they were centrifuged at 3000 rpm for 15 min. Both serum and milk samples were stored at -20°C until analysis. Ca, Zn, Fe, Cu, and Mg concentrations in blood serum and milk (except Cu in milk) were determined by atomic absorption spectrophotometer according to the method described by (AOAC) Association of official analytical Chemists (1984). Lactose in milk was determined according methods described by (Davis and Mac Donald, 1953).

Bacteriological examination:

The milk samples were activated by incubation for 12h at 37 °C , then centrifuged at 3000 r.p.m. for 30minutes . All the samples were examined bacteriologically by streaking loopful of centrifuged milk sediment from each sample on surface of nutrient agar, MacConkey agar and blood agar containing 7.5% defiberinated sheep blood . All plates were observed after incubation 24-48 hr, at 37 °C and any growth were recorded. Biochemical tests of isolated strains were determined by using criteria of (Quinn et al., 1994 and 2002)

Antibacterial sensitivity test:

Sensitivity of the isolates was determined using Mueller–Hinton agar against different chemotherapeutic agents including (amoxycillin, penicillin, ampicillin, chloramphenicol, ceftiofur, streptomycin, enrofloxacin, ciprofloxacin, gentamycin , neomycin, erythromycin, tetracycline and doxycycline). Isolates categorized as susceptible, intermediate or resistant according to methods and criteria described by National Committee for Clinical Laboratory Standards (NCCLS, 2002).

Field treatment regimens :

Based on the result of antibiotic sensitivity test, some of the infected quarters with most prevalent bacterial pathogens were allocated in different treatment regimens as follow:

1- Two days standard therapy group : infected udder were treated once daily with 125 mg of Ceftiofur hydrochloride (Pfizer Animal Health, USA) intra-mammary infusion per quarter for ٢ consecutive days.

2- Five days extended therapy group: infected udder were treated once daily with 125 mg of Ceftiofur hydrochloride (Pfizer Animal Health, USA) intra-mammary infusion per quarter for 5 consecutive days.

3- Non treated infected control group infected udder do not receive any medication and served as a control.

Mammary quarter foremilk samples were obtained 14 days after the last treatment for bacteriological evaluation. A bacteriological cure was defined as a treated infected mammary quarter that was bacteriologically negative for the presence of previously identified bacteria at 14 days after the last treatment (Wilson et al. 1986).

Results

Infected cows showed typical signs of udder inflammation in the form of hotness, swelling and tenderness. In addition to systemic reaction in the form of pyrexia (40.3±0.6 °C), anorexia, dullness and ruminal stasis were recorded in some cows. However some cows showed only local inflammatory reaction in the mammary gland. Moreover, the milk was serous amber secretions containing small barely like flacks. Blood was found in some milk samples.

Rate of clinical mastitis in the examined dairy herds based on quarter levels are shown in table (1). Types and incidence of bacterial isolates from mastitic cows milk are present in table (2).

The result of serum biochemical analysis showed that, Calcium, Zn and Fe levels of serum in mastitic cows were significantly lower than healthy normal cows at ($P \leq 0.05$), ($P \leq 0.01$) and ($P \leq 0.01$) respectively, but Cu value were significantly higher ($P \leq 0.01$) than normal cows (table 3). With regard to Mg in serum, no significant difference were observed between healthy and mastitic cows.

Calcium level of milk in mastitic cows were significantly ($P \leq 0.01$) lower than healthy normal cows milk. In contrast Fe was significantly higher ($P \leq 0.01$) than normal cows. Also Zn level was significantly ($P \leq 0.05$) higher than normal cows milk. On the other hand Mg level not significantly differ between normal and mastitic milk (table 4).

Table (5) showed the results of antibiotic sensitivity test of the isolated bacteria against different chemotherapeutic agents. All isolated bacteria were sensitive to ceftiofur, ciprofloxacin and enrofloxacin.

While most of them were resistant to penicillin and amoxicillin, but intermediately sensitive to streptomycin and neomycin.
Field treatment showed that ceftiofur treatment regimens were better than the negative control, and 5-days ceftiofur treatment regimen has higher bacterial cure rates than the standard 2-days ceftiofur treatment regimen for all bacterial pathogen except streptococci infection where, no difference were recorded between 2days and 5 days Ceftiofur treatment regimens (table 6).
Table (1): Rate of clinical mastitis in dairy herds based on quarter levels

Affected quarter	Number N =103	%
Cow with one affected quarter	55	53.39
Cow with two affected quarter	29	28.15
Cow with three affected quarter	14	13.59
Cow with four affected quarter	5	4.8
Total	103	

The percentage were calculated according to the total positive samples (103).

Table(2): Types and incidence of bacterial isolates from mastitic cows milk

Types of isolates	Positive isolates/ cows numbers		Positive isolates/ affected quarter numbers	
	No.	%	No.	%
Escherichia coli	80/103	77.66	80/175	45.71
Staphylococcus aureus	29/103	٢٨,١٥	29/175	16.57
Streptococcus aglactiae	22/103	٢١,٣٥	22/175	12.57
Streptococcus uberis	16/103	١٥,٥٣	16/175	9.14
Enterobacter aerogenes	9/103	8.73	9/175	5.14
Arcenobacterium pyogenes	4/103	٣,٨٨	4/175	2.28
Klebsiella pneumoniae	6/103	٥,٨٢	6/175	3.42
Proteus mirabilis	9/103	8.73	9/175	5.14

Table (3):Mean levels of the elements in blood serum of cows with normal and clinical mastitis

Element	normal healthy (n=١0)	Mastitis n=25	Significance
Ca (mg/dl)	13.51±0.42	11.28±0.42	*
Cu (mg/l)	0.42±0.02	0.97±0.04	**
Zn (mg/l)	1.05±0.07	0.77±0.04	**
Mg (mg/dl)	2.79±0.09	2.78±0.16	NS
Fe (mg/l)	3.14±0.27	2.29±0.22	**

N.S = non significant
* significant at p ≤0.05
** significant at p ≤0.01

Table (4):Mean levels of some elements and of lactose in milk of normal and mastitic cows

Element	normal healthy (n=١0)	Mastitis n=25	Significance
Ca (mg/dl)	101.30±2.42	72.77±2.65	**
Zn (mg/l)	3.51±0.17	4.21±0.24	*
Mg (mg/dl)	13.88±0.69	12.70±0.66	NS
Fe (mg/l)	0.42±0.07	0.68±0.02	**
Lactose %	4.75±0.53	2.55 ±0.11	**

N.S = non significant
* significant at p ≤0.05
** significant at p ≤0.01

Table (5): Aantibiotic sensitivity test of isolated bacteria from mastitic cows

Antibacterial agents	*E.coli*				*Staph aureus*				*Streptococcus agalactia*				*Streptococcus uberis*				*Enetrobacter aerogenes*				*Arcenobacterium pyogenes*				*Klebseilla pneumoniae*			
	No. of strains(80)				*No. of strains*(29)				*No. of strains*(22)				*No. of strains*(16)				*No. of strains* (9)				*No. of strains* (4)				*No. of strains* (6)			
	R		S		R		S		R		S		R		S		R		S		R		S		R		S	
	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%	No	%
Amoxicillin	80	100	0	0	20	68.96	9	31.1	22	100	0	0	16	100	0	0	9	100	0	0	2	50	2	50	6	100	0	0
Penicillin G.	80	100	0	0	8	27.5	21	72.4	22	100	0	0	16	100	0	0	9	100	0	0	3	75	1	25	6	100	0	0
Ampicillin	75	93.7	5	6.3	15	51.72	14	48.27	22	100	0	0	16	100	0	0	9	100	0	0	1	25	3	75	6	100	0	0
Chloramephenicol	60	75	20	25	15	51.72	14	48.27	11	50	11	50	8	50	8	50	9	100	0	0	0	0	4	100	6	100	0	0
Ceftiofur	0	0	80	100	0	0	29	100	0	0	22	100	0	0	16	100	0	0	9	0	1	25	3	75	0	0	6	100
Streptomycin	65	81.3	15	18.7	18	62.1	11	37.9	7	31.81	15	68.18	7	93.75	9	56.25	4	44.44	5	55.55	1	25	3	75	3	50	3	50
Enrofloxacin	5	6.3	75	93.7	2	6.89	27	93.1	2	9.1	20	90.9	5	22.72	11	68.75	5	55.55	4	44.44	0	0	4	100	2	33.33	4	66.66
Ciprofloxacin	6	7.5	74	9.25	1	3.44	28	93.1	0	22.75	17	77.27	4	25	12	75	3	33.33	6	66.66	0	0	4	100	1	16.66	5	83.34
Gentamycin	7	8.75	73	91.25	1	3.44	28	93.1	0	45.95	12	54.54	8	50	8	50	3	33.33	6	66.66	1	25	3	75	6	100	0	0
Neomycin	10	12.5	70	87.5	1	3.44	28	93.1	0	54.54	10	45.45	7	43.75	9	56.25	2	22.22	7	77.77	1	25	4	75	3	50	3	50
Erythromycin	30	37.5	50	62.5	5	17.24	24	82.75	0	45.45	12	54.54	3	18.75	13	81.25	8	88.88	1	11.11	2	50	2	50	3	50	3	50
Tetracycline	80	100	0	0	8	27.58	21	72.4	0	50	11	50	7	43.75	9	56.25	7	77.77	2	22.22	1	25	3	75	2	33.33	4	66.66
Doxycycline	55	68.75	25	31.25	29	100	0	0	0	5.91	9	40.9	6	37.5	10	62.5	9	100	0	0	0	0	4	100	4	66.66	2	33.33

Amoxycillin (Am - 10 µg) , Pencillin G (P - 10),Ampicilli (Amp 10 µg) , Chloramephenicol (10 µg), Ceftiofur(30 µg), Streptomycin (S - 10 µg) , Enrofloxacin (Enr - 10 µg) Ciprofloxacin (Cip - 5 µg), Gentamycin (Gn - 10 µg), Neomycin (N - 30 µg), Erythromycin (E - 15 µg), Tetracyclin (TE - 30 µg) and Doxycyclin (Do - 100 µg).

Table (6): Comparison of different treatment regimens with Ceftiofur of clinically mastitic cows and their quarters cure rates.

Isolated bacteria	No. of affected quarters	No. of cured quarters after 14 days of Ceftiofur treatment					
		2 days treatment regimen		5 days treatment regimen		Control non treated	
		No.	%	No.	%	No.	%
Escherichia coli	40	8/15	53.33	13/15	86.66	0/10	0.0
Staphylococcus aureus	25	3/10	٣٠	8/10	80	0/5	0.0
Streptococcus Aglactiae	19	5/7	71.42	5/7	71.42	0/5	0.0
Streptococcus Uberis	15	4/5	80	4/5	80	0/5	0.0
total	99	20/37	54.05	30/37	81.08	0/25	0.0

Discussion

Mastitis is one of he most important diseases affecting dairy animals and despite the knowledge regarding its prevention and control, economic losses still occur (Anwar et al., 2003). The results showed that mastitis was found more often in one quarter (53.39 %.) and these agreed with Rajala-Schultz et al. (2004) who stated that the majority of the cows had infection only in one quarter and occurs in other quarters by variable percentages.Mean while Lam et al . (1997) and Rotz et al. (2003) reported that the transmission of intramammary infection occurs not only among cows but also among quarters within a cow.More over Kikkers et al. (2004) reported that infected quarters could be due to random distribution .

In the present study, most of clinical mastitis in cattle were originated from environmental bacteria .*Escherichia coli* was the most predominant isolates (45.71%), this high incidence of isolates was nearly in agreement with Eman et al. (2006) who isolated *E.coli* at percentage 51.1% as a causative agent of mastitis. while, Nahed et al. (2003) isolated it at a percentage of 55.29% .Bramley , (1992) stated that *E.coli* is one of the most frequently isolated pathogens from both clinical and chronic infections. Other bacterial isolates were *Staphylococcus aureus, Streptococcus aglactiae , Streptococcus uberis, Enterobacter aerogenes ,Arcenobacterium pyogenes ,Klebsiella pneumoniae and Proteus mirabilis* in a percentage of (16.57%, 12.57%, 9.14%, 5.14%, 2.28%, 3.42% and 5.14) respectively. Similar bacterial pathogens were isolated from mastitic milk by Douglas et al., (2000); Phuektles et al.,(2001); Gregory and Hoedemaker (2002) and Anwar et al., (2003). Also, Eman, (2006) isolated similar organisms from the milk of dairy cows.

One of the most common types of chronic mastitis are caused by *Staphylococcus aureus* . This organisms persist in mammary glands , teat canals and teat lesions of infected cows and are considered

contagious (Jones et al., 1998). Our result revealed the isolation of *Staphylococcus aureus* by (16.57%) . This result relatively agreed with Anwar et al, (2003) and Eman et al, (2006) who isolated this organisms in percentage 20% & 20.7% respectively .The present result are in disagreement with the result of Sargeant et al.(2001); Abdel Hafeez (2002) and Shitandi and Kihumbu (2004), who isolated this *Staphylococcus aureus* bacteria in percentage 90.1%, 45.6% and 50% respectively.

Streptococcus spp. is not an active tissue invader but it multiplies in the milk within the udder elaborating an irritant causing an inflammatory reaction (Schalm et al,. 1971). In the present study *Streptococcus aglactiae* & *Streptococcus uberis* were isolated in percentage (12.57% & 9.14%) and this relatively agreed with(Shitandi and Kihumbu,2004) .

Zinc is essential for the activity of more than 100 enzymes in the body, and zinc-containing enzymes are involved in major pathways of nucleic acid, protein, carbohydrate and lipid metabolism. It is an essential component of the antioxidant enzyme Cu-Zn superoxide dismutase. Zinc is required for the formation of keratin that lines the teat canal and helps in killing organisms that enter through the teat canal (Bitman et al., 1991).

Concerning the biochemical constituent of serum in both normal and mastitic cows, our results revealed that the mean zinc concentration in blood from cows with clinical mastitis were significantly ($P \leq 0.01$) lower than that of healthy cows this result agreed with Ranjan et al. (2005). More over Ram Naresh et al. (2001) reported significantly lower blood zinc concentration in bovine mastitis.

The present result also showed that the mean value of copper in clinical cases of mastitis was significantly higher than in those in the blood of healthy cows($p \leq 0.01$) this result goes parallel with that reported by (Hamit and Erdal , 2005). Copper acts as an antioxidant as well as prooxidant. It is an integral part of several proteins such as ceruloplasmin and the enzyme superoxide dismutase. These proteins act as an antioxidants by scavenging oxygen free radicals and could serve as an endogenous modulator of the inflammatory response. Singh and Bansal, (2001) have also shown that cows supplemented with copper at 20 ppm in feed/day in addition to basal diet had more uninfected quarters compared to nonsupplemented cows. On the other hand Conner et al. (1986) mentioned that higher concentration of copper was found in blood from cows with mastitis. The ceruloplasmin contains more than 95% of the circulating copper in animals. Its level in blood has been

reported to increase many folds during inflammation. Therefore, increase in blood copper level in the clinical cases of mastitis might be due to the elevated level of ceruloplasmin in inflammatory udder conditions.

Calcium, and iron levels in serum of mastitic cows were significantly ($P \leq 0.05$ and $P \leq 0.01$) lower than healthy normal cows, but with regard to Mg in serum, no significant difference were observed between healthly and mastitic cows. These results were agreed with that reported by Hamit and Erdal , (2005) . In the same context Erskine et al. (1993) observed that plasma Zn and Fe concentrations were decreased in *Escherichia coli* induced mastitis in experimental animals. This observation coincided with our result and supported by the bacteriological isolation in this study where, *E.coli* infection was the predominant bacterial cause of mastitis in examined quarters.

Determination of Ca and Mg in mastitic milk may give important clues about the status of mastitis. Ca and Mg contents of milk originate from the blood. These elements are found in insoluble form (75%) or colloidal Ca phosphates and as colloidal complexes with casein in milk (Hamit and Erdal , 2005) . A single milking has been shown to remove nearly 50% of the keratin of the teat canal and daily replacement of keratin is essential for udder health. In cattle, zinc supplementation was found to be beneficial in enhancing fertility, and it reduced incidence of new intramammary infections (Spain, 1993).

Indeed, increased passage of some elements to milk that was reported during acute mastitis resulted from inflammation induced increase in vascular permeability(Hamit and Erdal ,2005).

In this study the calcium level of milk in mastitic cows were significantly ($p \leq 0.01$) lower than healthy normal one. In contrast iron was significantly ($P \leq 0.01$) higher than normal cows. On the other hand magnesium level was not significantly differ between normal and mastitic milk .This result goes parallel with that reported by Hamit and Erdal , (2005).

Varying observations were reported regarding Zn levels in the milk of clinical and subclinical mastitis cows . Janota et al.(1978) found a slight decrease while Banga et al. (1989) showed that Zn values did not significantly change in the milk but increased in the inflammatory tissue. In experimentally induced mastitis, Zn levels in milk were found to be increased (Lohuis et al.,1988). Our result indicated that Zn level in mastitic milk was significantly ($P \leq 0.05$) higher than normal cows milk. this result agreed with that reported by(Hamit and Erdal ,2005).

Regarding , the lactose content in milk the result indicated that there was a significant decrease in lactose content in mastitic milk than normal health one . This result agreed with Auldist et al. (1995) and Bansal et al .(2005).

Important considerations for antibiotic treatment of clinical bacterial mastitis are using an antibiotic with an appropriate spectrum of activity, selecting an antibiotic that attains and maintains an effective

therapeutic concentration at the site of infection (milk, mammary tissue, or blood); treating for the appropriate duration; and avoiding adverse local or systemic effects and violative residues (Peter and Dawn, 2003).

Concerning , the results of antibiotic sensitivity test of the isolated bacteria against different chemotherapeutic agents. All isolated bacteria were sensitive to Ceftiofur, ciprofloxacin and enrofloxacin. While most of them were resistant to penicillin and amoxicillin as well as intermediate to streptomycin and neomycin. This result agreed with Gianneechini et al. (2002); Erskine et al.(2002) and Makovec & Ruegg, (2003)

The most common route of administration of antimicrobials in mastitis is the intramammary (IMM) route .The advantages of this route are high concentrations of antibiotics achieved in the milk compartment of the mammary gland (Gruet et al., 2001), and low consumption of the antimicrobial substances as the drug is administered straight to the infection site (Satu ,2002).

All two Ceftiofur treatment regimens were better than the negative control, and the 5-d Ceftiofur extended therapy treatment regimens have higher bacterial cure rates than the standard 2-d Ceftiofur treatment regimen. This result agreed with that reported by Oliver et al. (2004) , Wenz et al. (2005) and Sabry and Salama, (2008). Also the present result indicated that standard 2-d Ceftiofur treatment not different from 5-d extended regimen in streptococcus spp infection .This observation was also reported by Oliver et al. (2004) who reported that none of the treatment regimens (2-daya and 5-days) of ceftiofur therapy were different from one another for *Strept. uberis* and *Strept. dysgalactiae.* The result of the 2-days standard Ceftiofur therapy treatment regimen was better than the negative control for *Strep. dysgalactiae* and *Strep. Uberis*. For all other pathogen/pathogen groups, the 5-days Ceftiofur extended therapy treatment regimen was better than the negative control. It should be , however noted that, sensitivity test in the laboratory does not always indicated the relative effect of the drug when infused into an infected quarter, and this depends on many factors that take place inside the mammary glands such as PH, tissue protein binding and milk components as calcium (Soheir and Magda, 1997).

From this study, it can be concluded that *E.coli* , *Staphylococcus aureus, Streptococcus aglactiae and Streptococcus uberis* are the main isolated bacteria, associated with mastitis besides *Enterobacter aerogenes* , *Corynebacterium pyogenes, Klebsiella pneumoniae and Proteus mirabilis* with variable percentage.

The in vitro sensitivity test is very important to determined the drug of choice for treatment the affected quarter to avoid the spread of the bacteria among other quarter or other cows . Ceftiofur therapy was effective in eliminating clinical infection in lactating dairy cows caused by several different mastitis pathogens, and that extended Ceftiofur therapy enhanced treatment efficacy.

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الملخص العربي

دراسات بكتيريولوجية وفارماكولوجية على مرض التهاب الضرع الاكلينيكى فى الأبقار بمحافظة الإسماعيلية

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الهدف من هذه الدراسة هو تحديد الأسباب البكتيرية لالتهاب الضرع الظاهري فى الأبقار ودراسة مدى استجابتها للمضادات الحيوية المختلفة مع قياس مستوى بَعْض المكونات الكيماوية الحيوية في الدمْ واللبنِ وتطبيقِ بَعْض نظمِ العلاج الحقلى باستخدام المضاد الحيوى المناسب. وقد تم تجميع عينات لبن ودم من ١١٣ بقرة (١٠٣ تعانى من التهاب ضرع ظاهرى و١٠ سليمة ظاهريا) من مزارع لإنتاج الألبان في محافظة الإسماعيلية .

أظهرت النتائج تواجد ميكروب اشيرشيا كولاى بنسبة(45.71%) وباقي الميكروبات بنسب متفاوتة .الميكروب العنقودي الذهبي، والميكروب السبحي اجلكتيا والميكروب السبحى ابرص والكورينى بكتيريا والكلبسيللا نيمونيا والبورتيس ماريبلس (١٦,٥٧% ١٢,٥٧ %، ٩,١٤%، ٥,١٤%، ٢,٢٨%، ٣,٤٢% و٥,١٤) على التوالى. كما أوضحت نتيجةَ تحليلِ المصلِ الكيمياوي الحيوي بأنّ مستوى الكالسيوم و الزنك و الحديد في الابقارِ التى تعانى من التهاب ظاهرى للضرع اقل منْ الأبقارِ الطبيعيةِ وان مستوىَ عنصر النحاس أعلى معنويا منْ الأبقارِ الطبيعيةِ.بينما لم يسجل إختلاف فى مستوى الماغنسيوم فى المصل بين الأبقار المريضة والسليمة. وعند دراسات بعض مكونات الحليب وجد ان مستوى كالسيوم حليبِ الأبقار المصابة اقل بشكل ملحوظ منْ حليبِ الأبقارِ الطبيعيةِ ، وعلى النقيض من ذلك فان مستوى الحديد والزنك كَانَ أعلى جداً منْ الأبقارِ الطبيعيةِ من الناحية الأخرى لم يتأثر مستوى الماغنسيوم. وأظهرت نتائج اختبار الحساسية للميكروبات المعزولة بأنها جميعا كَانتْ حسّاسة إلى سفتيفيور والسيبروفلوكساسين والانروفلوكساسين,. وان معظم البكتيريا المعزولة كَانت مقاومة إلى البنسيلينِ والاموكسيسلين بالإضافة إلى أنها متوسّطة الاستجابة تجاه كل من النيوميسين والاستربتوميسين . وعند إجراء بعض العلاجات الحقلية مستندة على نتيجةِ اختبار الحسّاسيةِ للمضادات الحيوية، أسفرت النتائج عن أن استخدام عقار السيفتيوفيور هيدروكلورايد ١٢٥ ملجم موضعيا داخل الربع المصاب كَان أفضل من المجموعة المصابة والغير معالجة، وان استخدام عقار السيفتيوفيور هيدروكلورايد ١٢٥ ملجم لمدة خمسة أيام متتالية أفضل من استخدامه لمدة يومين وذلك بناء على معدل الشفاء البكتيري للربع المصاب. لذا يوصى بالفحص المعملى البكتيرى الدوري و يجب الاهْتمام بالآلات المستعملة لتَجَنُّب انتشار العدوى بين الأبقارِ، كذلك أتباع الشروط الصحّيةِ اثناء عملية الحلبِ لمَنْع الإصابات و العدوى مع استخدام العلاج بمضادات حيوية فعالة لحالاتِ التهاب الضرع الظاهرى لتَخفيض معدل انتشار العدوى.