

## EFFICACY EVALUATION OF NEW TEAT DIP FORMULATION (CYTEAL) FOR CONTROL OF BOVINE MASTITIS UNDER EXPERIMENTAL AND FIELD CONDITIONS

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### SUMMARY

The germicidal activity of commercial teat dip formulation (cyteal) (containing 0.1 g Hexamidine, 0.5 ml chlorhexidine digluconate and 0.30 g chlorocresol) was tested in vitro against *Staphylococcus aureus*, *Streptococcus* and *Corynebacterium bovis* field strains isolated from a dairy herd of 78 lactating cows. It was revealed that a concentration of 0.5% tested compound was effective against the isolated organisms.

A natural exposure study was conducted for 6 months in dairy herd to determine the efficacy of cyteal as post-milking teat antiseptic for the prevention of the new intramammary infections. There were 7.4% new intramammary infections on the dipped group and 21.6 on the control group, (65.7% reduction). The teat dip was effective in preventing new infections with *Staphylococci*, *Strept. agalactiae*, *Coryne. bovis* but did not appear to prevent new infection with coliforms.

The percentage of infected quarters on the dipped group was reduced from an initial (35.1%) to (22.2%) at the end of the study whereas the incidence in the control quarters increased from

(23.6%) to (35.8%) at the end of the study.

### INTRODUCTION

Control of bovine mastitis is essential to produce high quality milk efficiently and to maximize profits. Udder hygiene practices are necessary components of a complete programme for mastitis control (Pankey et al., 1984). The use of postmilking teat germicides is the most effective way to reduce the bacterial population on teat skin and to prevent new intramammary infections (IMI) in dairy cows (Farnsworth, 1984). Controlled studies are necessary to determine the availability of a teat sanitizer to reduce incidence of IMI (Hogan et al., 1990).

Chlorhexidine germicides are effective against wide range of bacteria including Gram negative organisms and are non irritating to teat skin, they are virtually non toxic, rapid acting (Grandner and Gary, 1983) and effective in the presence of organic matter such as blood, pus, uterine fluid on milk (Gerring et al., 1968). Post-milking teat

sanization with an effective teat dip formulation significantly reduced the rate of new IMI by contagious pathogens (Kesler, 1983), it has not been effective in controlling mastitis caused by environmental pathogens such as coliforms and streptococci other than *Strept. agalactiae* (Smith et al., 1985). Infectiveness of post-milking teat dips in prevention of mastitis by environmental pathogens is thought to be related to differences in the reservoir of mastitis pathogens and to the time when bacterial exposure takes place (Smith et al., 1985; Oliver et al., 1990; Goldberg et al., 1994). Reservoirs of environmental mastitis pathogens not only infect mammary glands but also the environment. Teat dipping reduces the number of environmental pathogens on teat skin immediately after milking. However, exposure of teats to environmental pathogens continues between milking when most of the teat dips have lost effectiveness (Drechsler et al., 1993).

The objective of the study reported here was to determine the efficacy of a postmilking teat germicide against *Staphylococcus aureus*, *Streptococcus agalactiae*, coliforms and *Coryne bovis* under in vitro and in vivo conditions for prevention of bovine mastitis during lactation

## MATERIAL AND METHODS

This work was conducted in two trials:

- A. Experimental conditions.
- B. Field conditions.

### A. Experimental conditions:

#### 1. Disinfectant used:

Cyteal solution, Medical Union Pharmaceuticals, Abu Sultan, Ismailia, Egypt. The solution

consisted of separate solution containing ingredients, 0.1 gm hexamidine, 0.5 chlorhexidine digluconate and 0.3 chlorocresol in 100 ml.

### 2. Strains used:

*Staphylococcus aureus*, *Streptococcus agalactiae*, coliforms, *Coryne bovis*. *Staphylococci*, coliforms and *Coryne. bovis* were isolated from infected milk samples of cows in the examined groups.

### Methods:

Two standard platinum loops from each isolate organism were suspended in 2 ml sterile normal saline solution from which one ml was inoculated into 9 ml of disinfectant solution and mixed thoroughly. A loopful was taken after 2.5, 5, 7, and 10 minutes and streaked on selected media tested organisms. The inoculated media were incubated at 37°C for 24 and 48 hours. Control plates were prepared using the same inoculum previously put into sterile saline solution. The growth of the microorganisms against the tested disinfectant was recorded in Table (2), Dennis and Gaunt, (1974).

### B. Field conditions:

A field study was conducted following procedures described by The National Mastitis Council (Hogan et al., 1991).

#### 1. Materials:

##### 1.1. Location:

This study was conducted at Seds Station for Animal Production belonging to Agriculture Research Institute in Beni Suf Governorate.

##### 1.2. Animals:

One hundred and twenty cows were used during the period of this study.

### 1.3. Housing and management:

Dairy animals were housed in six separate and partially sheltered yards on tied stalls, standing on earthy floor. Lactating cows were hand milked twice daily and received their needs of water 3-5 times/ day through common water troughs.

### 1.4. Disinfectant:

Cyteal solution, Medical Union Pharmaceuticals, Abu Sultan, Ismailia, Egypt.

### 2. Methods:

Clinical examination was applied to the udders of cows during sampling then 10 ml of individual quarter milk samples were collected aseptically into sterile screw capped bottle after rejecting the first two streams of milk at the start of the trial and one week later. Quarters were considered infected if the same pathogen was identified in two successive milk samples.

### 3. Experimental design:

Cows were divided into groups, each of thirty-nine lactating cows, the first one served as a control while the second was used as dipped group whereas the control group animals were kept under same prevailing managerial practices prevailing in the farm. In dipped group, the cows were kept in a clean and dry yard, tied to concrete manger and hand milked by special attendant. Strict sanitary measures including regular cleaning and disinfection of milking equipments and yard were applied. Teats of all cows were fully dipped twice daily in Cyteal containing 1% active ingredients soon after milking. Additional quarters milk samples were collected bimonthly of the beginning of the study for isolation and identification of microorganisms causing mastitis.

### RESULTS

Table (1): Percentage of infected quarters in control and treated group before the study .

Group	No. of cows	Infected quarters					Total
		Staph. aureus	Strept.agalactiae	Coliform	Coagulase +ve Staph.	Coryne. bovis	
Control	87	8.1	5.4	4.7	3.3	2.0	23.6
Dipped	37	12.8	9.4	5.4	4.7	2.0	35.1

Table (2): Germicidal sensitivity of isolated strains to teat sanitizer in vitro .

Germicidal conc.	Staph. aureus (minutes)				Strept. agalactiae (minutes)				Coliforms (minutes)				Coryne. bovis (minutes)			
	2.5	5.0	7.5	10	2.5	5.0	7.5	10.0	2.5	5.0	7.5	10.0	2.5	5.0	7.5	10.0
0.25	+ve	+ve	+ve	-ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve	-ve
0.5	+ve	+ve	-ve	-ve	+ve	+ve	-ve	-ve	+ve	+ve	-ve	-ve	-ve	-ve	-ve	-ve
1.0	+ve	-ve	-ve	-ve	+ve	-ve	-ve	-ve	+ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve

Table (3): Comparison of percentage of infection, persistence of infection, and new intramammary infection at start and at the end of the study .

Group of animals	Infected quarters	Start %	Infection in quarters			New intra-mammary infection (IMI)	Infection at the end of study
			Eliminated by Spontaneous recovery	antibiotic treatment	persisted until end of study		
Control	S. aureus	8.1	0.6	2.0	5.4	6.7	12.1
	St. agalactiae	5.4	---	1.35	4.0	4.7	8.7
	Coliforms	4.7	0.6	1.35	2.7	2.7	5.4
	Coagulase -ve	3.3	1.35	0.6	1.35	5.4	6.7
	Coryne bovis	2.0	--	1.35	0.6	2.0	2.7
	Total	23.6	2.7	6.7	12.6	21.6	35.8
Dipped	S. aureus	12.8	2.7	4.7	5.4	2.0	7.4
	St. agalactiae	9.4	1.35	4.0	4.0	1.35	5.4
	Coliforms	5.4	1.35	0.6	4.0	2.7	6.7
	Coagulase -ve	4.7	1.35	2.0	1.35	0.6	2.0
	Coryne bovis	2.0	--	2.0	--	0.6	0.6
	Total	35.1	6.7	13.5	14.8	7.4	22.2

Table (4): Efficacy of new teat dip for prevention of new IMI in Experimental

Organism	Group of Animals	New intramammary (IMI) infection	
		% of quarters	% of infection
Staphylococcus aureus	Control Dipped	6.7 2.02	69.8
Streptococcus agalactiae	Control Dipped	4.72 1.35	71.36
Coliforms	Control Dipped	2.7 2.7	--
Coagulase negative Staphylococci	Control Dipped	5.4 1.35	75.0
Corynebacterium bovis	Control Dipped	2.02 0.67	33.4

## DISCUSSION

The germicidal activity of commercial teat dip formulation containing 0.1g Hexamidine, 0.5ml chlorhexidine and 0.3g chlorocresol was tested in vitro against field strains isolated from dairy herds. Besides, natural exposure study for 6 months to evaluate its efficacy.

Infection status of lactating cows was confirmed by bacteriological examination of individual quarter milk samples and cows were classified into 2 groups (control and dipped). The percentage of infection was 23.6 and 35.1 for two groups, respectively at the start of study with isolation of *Staphylococcus aureus* at a percentage of 8.1 and 12.8, *Str. agalactiae* 5.1 and 9.4 Coliforms 4.7 and 5.4, coagulase negative *Staphylococci* 3.3 and 4.7, *Coryne, bovis* (2.0 and 2.0) for the two examined groups, respectively. These results agree with those reported by Oliver et al. (1990); Drechster et al. (1993) and Oliver et al. (1993).

Germicidal activity of teat sanitizers against representative strains isolated from examined lactating cows in control and dipped group is shown in (Table 2). It was clear that Cyteal at a concentration of 0.5% kills all tested strains (*Staphylococcus aureus*, *Str. agalactiae*, Coliforms and *Coryne. bovis*) within 7.5 minutes. Similar results were obtained by Pankey et al. (1983), Reem Desoky et al. (1993) who mentioned that the concentration of choice of disinfectant under test was realized after 7.5 minutes exposure time.

According to the results obtained, cyteal was

tested in vivo at concentration of 1% as a post-milking teat antiseptic. The use of such lethal concentration has many advantages including cost, reduced irritation and minimal residue in milk.

Table (4) shows that the percentage of infection at the end of study was reduced from 35.1 to 22.2 in dipped group, comparing to increased the percentage from 23.6 to 35.8 in control group. The results agree with those reported by Pankey et al. (1978) and Sears et al. (1991).

Regarding the persistence of infections contracted during lactation, the percentage of spontaneous recovery was higher in dipped than control group 6.7 and 2.7 respectively. These results are in agreement with Wesen and Schultz (1970) and Goldberg et al. (1993) who mentioned that post-milking teat antiseptic reduced the incidence of existing infection and enhanced spontaneous recovery in dipped group.

Table (3 and 4) list the new infections during lactation and the percentage of infections caused by each microorganism. There were 7.4 and 21.6% new intrammary infection in control and dipped group respectively (65.7%) reduction in dipped group. Similar results were recorded by Neave et al. (1969); Oliver et al. (1990) and Goldberg et al. (1993).

The percentage of reduction was also calculated for individual pathogens, the post-milking teat dip resulted in a 69.8 reduction in the number of new infections with *S.aureus*, 71.3% reduction for *Str. agalactiae*, 75% for coagulase negative

Staphylococci and 33.4% for *Coryne. bovis*. The number of infections with Coliforms remained the same or slightly increased. Practically similar results were recorded by Stewart and Philpot (1982) who found that 77.1% reduction for *Staph. aureus*, 18.0% for *Strept. agalactiae*, 71.2 for coagulase negative Staphylococci and 14.5 for *Coryne, bovis*, while the increase in new infection with coliforms amounted to 50.5%.

Conclusively, the examined antiseptic showed high efficacy as post-milking teat antiseptic as it reduced the number of existing infection and new IMI and enhanced spontaneous recovery in infected quarters.

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