

Effect of post AI intrauterine treatment with *E. coli* lipopolysachharide and autologous serum on characteristics of cervico-vaginal mucous in subclinical endometritic cows

P. SINGLA¹, JAGIR SINGH², G. S. DHALIWAL³ AND AJEET KUMAR⁴

Department of Animal Reproduction, Gynaecology & Obstetrics
Punjab Agricultural University, Ludhiana-141004 (Punjab)

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ABSTRACT

Sub-clinical bacterial endometritis is a common cause of repeat-breeding in cows. The present study was conducted on 36 cows with confirmed sub-clinical endometritis. Based on the treatment protocol, these cows were divided randomly into three groups of 12 animals each. Group 1 (control), Group 2 and 3 cows were infused intrauterine with 20 ml of phosphate buffer saline (PBS), *E. coli* lipopolysaccharide (LPS) 100µg and *E. coli* LPS 100µg + 5 ml of autologous serum, respectively, 8-12 hr after insemination. Cervico-vaginal mucus (CVM) was collected before artificial insemination (AI) and 12 hr after treatment. It was observed that colour and consistency of CVM became normal in significant number of animals. A significant decrease in pH of CVM was also observed in treated animals.

Key Words: Lipopolysachharide, autologous serum, sub-clinical endometritis, cervico-vaginal mucus, cows

Bacterial endometritis is one of the common causes of altering the physical properties of the CVM (Dohman *et al.*, 1996). Normal CVM is important for sperm transport in genital tract (Rowson *et al.*, 1972). Intrauterine antibiotics/antiseptics have been used for the treatment of endometritis with variable success (Shukla and Pandit, 1989). The disadvantages of antibiotics/antiseptics are compulsory milk disposal, development of microbial resistance, higher cost and inhibition of phagocytic activity of polymorphonuclear cells (Vandeplasseche, 1984). Taking these problems in to consideration, alternative therapies have been used for the treatment of uterine infection. One is them is the use of immunomodulators. Oyster glycogen has been used endometritis, but, there was loss of one complete estrous cycle (Anderson *et al.*, 1984). Therefore, LPS was tried as post AI therapy in the sub-clinical endometritis cows in the present study to save one estrous cycle.

Thirty six normal cycling crossbred cows maintained at dairy farm, PAU, Ludhiana; Habowal Dairy Complex and animals brought to PAU Clinics were included in the study.

Sub-clinical endometritis was confirmed on the basis of bacterial culture and isolation from uterine swabs. The animals were divided randomly into three groups of 12 animals each. Group 1 (control), group 2 and group 3 cows were infused intrauterine with 20 ml phosphate buffer saline (PBS), 100 µg of LPS* and 100µg LPS+5ml autologous serum, respectively, 8-10 hrs after insemination. Cervico-vaginal mucus was collected using AI glass pipette and syringe aseptically before and 12 hrs after treatment. Color, consistency and pH of CVM were assessed. Colour was assessed with the naked eye. Consistency was judged and graded as thin (++) and thick (+++). pH was noted with the help of pH paper.

Bacteria isolated from uterine swabs are presented in Table-1. Microorganisms isolated from uterine swabs of cows with subclinical endometritis were mainly *E. coli*, *Staph. aureus* and *Proteus* spp. whereas the incidence of *Streptococci* spp., *Klebsiella* spp., *Pseudomonas* spp., *Corynebacterium* spp. and *Bacillus* spp. were relatively less. Almost similar bacterial isolates in bovines with endometritis had been reported by Shukla (1988), Singla *et al.* (1991) and Singh (1996). On the contrary, other workers reported that *Staph. aureus*, *Actinomyces pyogenes*, *Streptococcus* spp. and *Bacillus* spp. predominated in repeat breeding bovines (Sharda *et al.*, 1991, Goswami *et al.*, 1992 and Saini, 1993). More incidences of *E. coli* and *Staph. aureus* in the subclinical endometritis could

¹Veterinary Officer

²Gynaecologist

³Professor & Head

⁴Assistant Professor,

¹Corresponding author

Table 1. Bacteria isolated from uterine swabs in cows with sub clinical endometritis

Bacterial isolate	No. of isolates	Per cent
<i>E. coli</i>	14	31.11
<i>Staph. aureus</i>	12	26.67
<i>Proteus spp.</i>	8	17.77
<i>Streptococci spp.</i>	4	8.88
<i>Klebsiella spp.</i>	3	6.67
<i>Pseudomonas spp.</i>	2	4.44
<i>Corynebacterium spp.</i>	1	2.22
<i>Bacillus spp.</i>	1	2.22
Total no. of isolates	45	100.00

indicate that, these bacteria are more commonly present in the animals excreta like dung etc. Unhygienic conditions at the time of per-rectal examination or at the time of A.I. may lead to the entry of these organisms into uterus causing subclinical endometritis

Colour, consistency and pH of CVM were recorded before and after treatment and the data is presented in Table-2. Before treatment, CVM was turbid in about 83, 75 and 67% animals in groups 1, 2 and 3, respectively. Upon LPS treatment in group 2, turbidity of CVM decreased significantly ($P < 0.05$) from 75 to 25% animals, whereas, treatment in group 3 showed maximum efficacy in clearance of CVM turbidity. However, PBS treatment in control group did not result any change in the colour of CVM. The cervical mucus of healthy animals is usually colourless and transparent (Roberts, 1986 and Arthur *et al.*, 1989). Cloudy, milky and turbid cervical discharge is generally observed due to uterine and cervical infections (Pandey *et al.*, 1983, Shankar *et al.*, 1984, and Saini, 1993).

Consistency of CVM before treatment was thick (+++) in about 42, 33 and 25% animals in group 1, 2 and 3, respectively. After treatment in group 2 and 3, the consistency of CVM changed to normal (thin, ++) in 83% and 92% animals, respectively. However, the changes in the consistency of CVM were non significant in control group after PBS infusion. In the earlier studies, the incidence of thin mucus in repeat breeder cows was either similar (Deo and Roy, 1971) or was lower (62-66%) than in the normal bovines (87-92%; Bishnoi 1975, and Saini 1993). Enkhia and Kohli (1982), however, reported that the CVM in majority of repeat breeding cows (65%) was watery. The difference in the cause and type of inflammation (Runnells *et al.*, 1965) might probably contribute to the variation in the

Table-2. Colour, consistency and pH of cervico-vaginal mucus before and after treatment with PBS, LPS and LPS + autologous serum.

Group	Pre treatment						Post treatment					
	Colour		Consistency		pH		Colour		Consistency		pH	
	Clean	Turbid	Thin (++)	Thick (+++)			Clean	Turbid	Thin (++)	Thick (+++)		
1 (n=12)	2 (16.66%)	10 (83.38%)	7 (58.3%)	5 (41.66%)	7.71 ± 0.05 ^a	2 (16.66%)	10 (83.38%)	8 (66.66%)	4 (33.33%)	7.68 ± 0.06		
2 (n=12)	3 (25.00%)	9 (75.00%)	8 (66.66%)	4 (33.33%)	7.71 ± 0.04 ^a	9 (75.00%)	3 (25.00%)	10 (83.33%)	2 (16.66%)	7.23 ± 0.05 ^b		
3 (n=12)	4 (33.33%)	8 (66.66%)	9 (75.00%)	3 (25.00%)	7.68 ± 0.05 ^a	11 (91.67%)	1 (8.33%)	11 (91.61%)	1 (8.33%)	7.20 ± 0.04 ^c		

Values with common Superscript within or between groups vary non-significantly.

consistency of CVM. The CVM consistency primarily depends upon the degree of cross linkage in the epithelial glycoprotein macromolecules of estrual cervical mucus, which in turn brought forth periodic changes in the penetrability of spermatozoa in cervical canal (Hafez, 1993).

pH of CVM before treatment were 7.71 ± 0.05 , 7.71 ± 0.04 and 7.68 ± 0.05 in group 1, 2 and 3, respectively. pH decreased marginally to 7.68 ± 0.06 in control group. However, significant decrease in pH of CVM was observed after treatment in groups 2 (7.23 ± 0.05) and 3 (7.20 ± 0.04). The pH of estrual CVM in repeat breeder cows with subclinical endometritis ranged from 7.40 to 8.00 (Akhtar and Singh, 1979; Wani *et al.*, 1982; Rane *et al.*, 1992 and Saini, 1993). The pH of normal CVM is around 7.00-7.20. It was concluded that treatment with LPS or LPS + autologous serum improved colour, consistency and pH of CVM.

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