

## ASSAYING OF PURIFIED COLOSTRAL IMMUNOGLOBULINS AGAINST *TOXOCARA VITULORUM* AS DETERMINED BY INDIRECT HAEMAGGLUTINATION TEST

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Antibody titre against *Toxocara vitulorum* was determined by indirect haemagglutination (IHA) test in the purified colostrum immunoglobulins of 10 cross-bred cows. All cows had a varying amount (1:4-1:32) of indirect haemagglutinating antibodies in their colostrum. At 0 hour (after parturition), it was the highest (GMT = 13.92) which gradually declined (GMT = 1.74) at 36 hours and became nil at 48, 60 and 72 hours.

### INTRODUCTION

Various parasitic infections have been reported among the probable causes of death in calves. Ascariasis has been observed to be the most common helminth infection in neonatal calves (Silva, 1959). One of the most important host defence considerations in young domestic animals is the passive transfer of immunoglobulins from dam to neonate via colostrum. The present study reports the titration of antibodies against *Toxocara vitulorum* in colostrum immunoglobulins as determined by indirect haemagglutination (IHA) test.

### MATERIALS AND METHODS

**Preparation of antigen and sensitisation of erythrocytes:** Sonicated antigen of *T. vitulorum* was prepared as described by Kent (1960). Indirect haemagglutination test was performed to assess the antibody titres against *T. vitulorum* following the method of Akhtar *et al.* (1991). Packed human group 'O' erythrocytes (0.1 ml) were suspended in 3.2 ml of the sonicated antigen. The suspension was left in a water bath at 37°C for

three hours with frequent shaking. The suspension was centrifuged at 1500 rpm for three minutes. The packed antigen sensitised erythrocytes were washed twice with phosphate buffered saline (PBS). A 0.2% concentration of erythrocytes was prepared in PBS.

**Colostrum samples:** Seventy colostrum samples, seven from each of the 10 cross-bred cows at 12 hours interval upto 72 hours were processed for purification of colostrum immunoglobulins according to Akhtar (1990).

Two-fold serial dilutions of all the purified colostrum immunoglobulins were made using 4-channel pipette in the microtitration plates. Fifty  $\mu$ l of the sensitised erythrocytes (0.2%) were added to each well of the plate. The plates were incubated at 37°C for 20 minutes. The results of haemagglutination were observed and titres were recorded in comparison with the control. The maximum dilution of each sample causing haemagglutination was taken as the endpoint. The IHA antibody titre was expressed as reciprocal of the dilution of the sample in the wells.

## RESULTS AND DISCUSSION

Transfer of colostral immunoglobulins from dam to the newborn calf is the most important means of providing protection to the young against diseases. But the level of antibodies in the colostrum from dam is not always commensurate with passive protection of the calf due to the intervention of numerous factors. An attempt is made to ascertain the level of natural antibodies against *T. vitulorum* in the colostrum of crossbred cows using IHA.

The antibody titres at 24 hours after parturition ranged from 1:8 to 1:16. Among 10 samples, the titres were 1:16 in 1 sample, 1:8 in 5 samples while 4 samples showed no antibody titre. The GMT was 3.73. At 36 hours, the antibody titre ranged from 1:4 to 1:8. Among 10 samples, 2 had titre 1:8, 1 had 1:4 while 7 samples showed no antibody titre against *T. vitulorum*. The GMT was 1.74. The immunoglobulins purified from colostrum at 48, 60 and 72 hours interval after parturition showed no antibody titre against *T. vitulorum* (Table 1).

Table 1. Antibody titres against *Toxocara vitulorum*

Interval (hours)	IHA antibody titre						Geomean titre*
	0	2	4	8	16	32	64
0				3	6	1	13.92
12			2	3	4	1	9.18
24	4			5	1		3.73
36	7		1	2			1.74
48	10						0
60	10						0
72	0						0

\*Ten colostrum samples were taken, one from each cow at each hour.

Antibody titres against *T. vitulorum* in purified colostral immunoglobulins just after parturition (0 hour) ranged from 1:8 to 1:32. Among 10 samples processed, the titres were: 1:32 in one sample, 1:16 in 6 samples and 1:8 in 3 samples. The geomean titre (GMT) was calculated as 13.92 whereas the antibody titres at 12 hours interval after parturition ranged from 1:4 to 1:32. Among 10 samples, one had antibody titre 1:32, 4 had 1:4, 3 had 1:8 while 2 samples had 1:4. The GMT was found to be 9.18.

The results indicate that all cows had antibodies in their colostrum against *T. vitulorum* upto 36 hours. The antibodies in the immunoglobulins from colostrum just after parturition show maximum titre and as the time increases, the titre decreases (Fig. 1).

The availability of passive immunity to the calf depends on the immunologic competence of the dam and the dam's ability to produce antibodies against potential pathogens and to concentrate these antibodies in the colostrum. Such a high level of

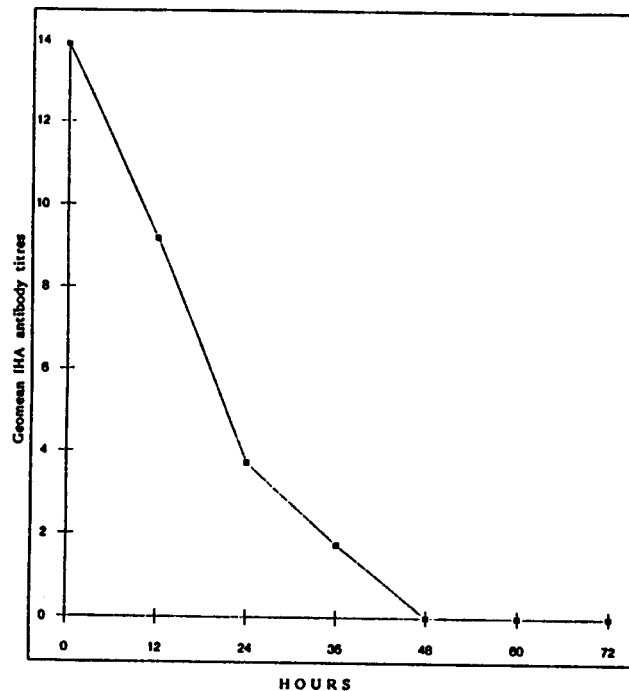


Fig. 1. Geomean IHA antibody titres against *Toxocara vitulorum* in purified colostrum immunoglobulins.

antibodies against *T. vitulorum* in the purified colostrum immunoglobulins suggests that calves can be passively immunised against Ascariasis by feeding colostrum to the calves in early age particularly before 36 hours. In circumstances where colostrum is not available, purified colostrum immunoglobulins can successfully be used to confer the passive immunity as the shelf-life of purified colostrum immunoglobulins is much greater than that of colostrum.

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