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Association of Lactation Numbers of Crossbred Cows with Their Serum Antisperm Antibody Titre

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ABSTRACT

Background: The objective of this study was to find out the association of lactation numbers of crossbred cow with their serum antisperm antibody titre, if any.

Methods: Total 163 numbers of normal breeding crossbred jersey crossbred cows belong to various lactational groups maintained in the Instructional Livestock Farm, College of Veterinary Science, Assam Agricultural University, Khanapara and various private cattle farms in Kamrup district of Assam, India were selected for the work. ELISA was carried out for detection of antisperm antibody (asp-abs) in the test sera. Data were analysed using Chi-square test.

Result: In the present study, out of 163 females 94 (57.67%) animal were found to be positive reactors. The percentage of positive reactors could be detected in 1st, 2nd, 3rd, 4th, 5th and above (5th, 6th, 7th and 9th) lactational group of cows were 16 (72.73%), 15 (50.00%), 23 (63.89%), 14 (43.75%), 26 (60.47%) respectively. The highest titre up to the extent of 1:320 was recorded in the sera of cows with 1st, 3rd and 4th lactations. A negative correlation ($r = -0.42^{NS}$) between the numbers of lactation and positive reactors for antisperm antibody (asp-abs) is found in crossbred cows.

Key words: Antisperm antibodies, Crossbred cows, Lactation numbers, Serum.

INTRODUCTION

The antigenicity of spermatozoa was first demonstrated as early as 1899. This finding opened up a new window for looking at the unexplained infertility from a different point of view. In the following years, many researches have been conducted worldwide in animal and human immune-infertility. Nevertheless, the results are conflicting. Farahani *et al*. (1981) indicated that antibodies against sperm apparently were not responsible for reduced fertility in animals. Spermatozoa can evade the body's immune system with the help of immunosuppressive molecules content in the seminal plasma. cervical mucus, uterine and follicular fluid that prevent the spermatozoa from recognition and antigen processing while passing through female genital tracts (Landers *et al.,* 1994). In contrast Stern *et al*. (1992) demonstrated that the antisperm antibodies may be present in seminal plasma, cervical mucus, uterine fluid, follicular fluid or blood sera and can inhibit the passage of spermatozoa through genital tract, prevent capacitation, reduce the ability to undergo acrosome reaction and interfere with zona binding and fertilization (Fijak and Meinhardt, 2006). The asp-abs may also induce infertility by sperm agglutination and decreasing sperm motility (Mathur *et al*. 1984). Risvanli *et al*. (2003) stated that the female genital tract can only produces asp -abs when the sperm come in contact with the blood of that particular cow. Therefore, the numbers of positive reactors to asp-abs in a population increases proportionately with the increasing parity and age of the cow due to the injury or inflammatory condition of the genital tract of the female during parturition or insemination (Cheema *et al*. 2016). Hence, the following study was carried out to find the possible relationship of serum asp-abs titre to the parity of cows.

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MATERIALS AND METHODS

The present study was conducted on 163 numbers of crossbred dairy cattle maintained in the Instructional Livestock Farm, College of Veterinary Science, Assam Agricultural University, Khanapara and various private cattle farms in Kamrup district of Assam, India from August, 2020 to July, 2021. Normal breeding Jersey crossbred cows of different lactations reared nearly under similar feeding and managemental practices were randomly selected for the study. The blood samples were collected aseptically and serum samples were stored at -20°C for detection of asp-abs.

Antigen from the bovine spermatozoa was prepared in the laboratory. 10 ml of freshly collected bull semen was

Table 1. Levels of asp-abs titre in serum in relation to numbers of lactation of cows.

Lactation number	Number of animal tested	Number of positive reactors	Number of negative reactors	Titre						Average titre
				1:10	1:20	1:40	1:80	1:160	1:320	
1th	22	16 (72.73)	6 (27.27)	4 (18.18)	6 (27.27)	1 (4.55)	2 (9.09)	2 (9.09)	1 (4.55)	1:62.50
2th	30	15 (50.00)	15 (50.00)	2 (6.67)	9 (30.00)	1 (3.33)	1 (3.33)	2 (6.67)	0	1:42.70
3th	36	23 (63.89)	13 (36.11)	6 (16.67)	9 (25.00)	3 (8.33)	2 (5.56)	2 (5.56)	1 (2.78)	1:50.40
4th	32	14 (43.75)	18 (56.25)	3 (9.38)	5 (15.63)	2 (6.25)	0	2 (6.25)	2 (6.25)	1:83.57
5th and above	43	26 (60.47)	17 (39.53)	5 (11.62)	10 (23.25)	7 (16.28)	4 (9.30)	0	0	1:32.69
Total	163	94 (57.66)	69 (42.33)							
Chi-square value		6.02NS					17.64NS			

Figures in parentheses indicate percentage.

NS : Non-significant (p>0.05).

brought to the laboratory and centrifuged at 3000 rpm for 10 minutes. The supernatant was discarded and the spermatozoa pellet was washed thrice with Phosphate buffer Solution (PBS, pH-7.4). Approximately 1 gm of washed spermatozoa was dissolved in 5 ml of PBS. Lysis of sperms was made by repeated freezing and thawing and centrifuged at 5000 rpm fir 15 minutes. The supernatant was collected and distributed in screw capped vials and stored at -20°C and used as antigen for the ELISA. The antigen was titrated and a titre of 1: 100 was found to be appropriate for ELISA test. ELISA test was performed by following the procedure as describe by Sarma *et al*., 2009. Data were analysed statistically following chi-square test using Microsoft office excel.

RESULTS AND DISCUSSION

Under the present study an attempt was made to find out the association between the production of asp-abs titre and lactations of the cows. The numbers of positive reactors in each of the lactational group along with their levels of titre have been presented in Table 1. Out of 163 animals 94 (57.67%) animal were found to be positive reactors. The total numbers and percentage of positive reactors could be detected in 1st, 2nd, 3rd, 4th, 5th and above (5th, 6th, 7th and 9th) lactational group of cows were 16 (72.73%),15 (50.00%), 23 (63.89%), 14 (43.75%), 26 (60.47%) respectively. Amongst the 22 cows of 1st lactation, as high as 72.73 per cent found positive reactors up to 1:160 and 1:320 levels of titre with an average of 1:62.5. The highest average asp-abs titre (1:83.57) could be recorded in 32 cows belongs to 4th lactational group where 6.25 per cent cows exhibited 1:160 and another 6.25 per cent cows exhibited 1:320 levels of titre. A similar trend of positivity in cows of 2nd (50.00%) and 3rd lactational (63.89%) group was observed with the average of 1:42.70 and 1:50.40 levels of titre, respectively. In contrast to the reports of Fayemi, (2005), the present study could be recorded a lower levels of average asp-abs titre of 1:31.00, 1:42.20, 1:23.30 and 1:22.50 in cows belongs to 5th, 6th, 7th and 9th lactational group. Moreover, in the cows of groups 7th and 9th lactations 50.00 per cent and 57.67 per cent, respectively were positive reactors in which the titre did not exceed 1:40. Results depicted in the Table 2 suggest that there is a negative correlation (r = -0.42) between the numbers of lactation and positive reactors for antisperm antibody (asp-abs) in Crossbred jersey cows which is non significant (p>0.05). The finding of present investigation was in accordance with the reports of Cheema *et al*. (2016) where significant increase in asp-abs in serum and cervical mucus was demonstrated with increase in number of inseminations, but not with the age and parity of cows. Moreover, Farahani *et al*., (1981) also failed to correlate the incidence and/or titre of the asp-abs with fertility status of the animal and stated that the asp-abs as natural antibodies which did not require exposure to sperm antigens. Ahuja *et al*. (2016) have also opined the asp-abs are as naturally developing antibodies and a constant component of blood in female cattle but in contrast to others they found the asp-abs titre to increase with the increasing age or number of lactations of the cows. The detection of lower asp-abs titre in cows with more numbers of lactations in this study might either be due to lack of exposures of spermatozoal antigen to the blood of the cow through the intact and normal mucosa of the genital tract leading to failure of asp-abs production or decreased production of asp- abs titre possibly due to their older age and senility besides other factors such as nutrition, environment and lactational stress leading to poor immune response (Risvanli *et al*., 2003).

Table 2: Correlation coefficient between number of lactation in cows and positive reactors.

	Number of lactation	Positive reactors
Number of lactation	1	-0.42
Positive reactors	-0.42	1

CONCLUSION

The present study reveals no significant association of number of lactations and the asp-abs titre in jersey crossbred dairy cows.

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Conflict of interest

All authors declare that they have no conflict of interest.

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