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EFFECT OF FMD VACCINATION ON VARIOUS SEMEN CHARACTERISTICS OF SAHIWAL BULLS

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The breeding bulls reared for the purpose of artificial insemination needs timely prevention against various bacterial and viral diseases. Routine vaccination is thereby being practiced in all the bull studs and bull mother farms in India for this purpose but vaccination has become one of the major anaphylactic stress factors that affect the semen quality. Based on data available regarding FMD vaccination during the period of 2002 to 2004 in Artificial Breeding Complex, NDRI, Karnal; effect of FMD vaccination was studied on semen quality parameters of bulls. One month pre-vaccination and one month post-vaccination data were collected after data adjusted against season, period and age with respect to significance. The results showed that FMD vaccination had no significant ($P>0.05$) effect on ejaculate volume of semen (4.48 ± 0.24 ml vs. 4.70 ± 0.29 ml); total volume per day (7.07 ± 0.60 ml vs. 6.27 ± 0.68 ml); initial motility (55.41 ± 0.07 % vs. 48.07 ± 0.11 %) and total sperm output per ejaculate (3244.69 ± 238.21 vs. 2700.00 ± 294.77$\times10^6$) whereas there was a significant decrease in mass activity ($P<0.05$) (2.20 ± 0.12 vs. 1.73 ± 0.14) and sperm concentration per ml ($P<0.01$) (737.76 ± 41.21$\times10^6$/ml vs. 540.63 ± 50.99 $\times10^6$/ml) in Sahiwal bulls. The following study indicates that may be the secondary activities of accessory sex glands remain unaffected following vaccination but application of FMD vaccine has significant ($P<0.05$) adverse effect on most of the seminal attributes during post vaccination in Sahiwal bulls.
Keywords: FMD vaccination, semen characteristics, Sahiwal

INTRODUCTION

In India vaccination of breeding bulls especially those kept for the purpose of artificial insemination is a regular prophylactic measure. These bulls are vaccinated against various contagious diseases which are both bacterial and viral. Although vaccination of both Indian breeds and exotic bulls has become a prerequisite but the available reports on the effect of vaccination on various semen parameters are very contradictory. A number of researchers have earlier reported the increased incidence in sperm abnormalities following vaccination in various breeds (Venkataswami and Rao (1970); Rao and Venkataswami (1974) and Kammar and Gangadhar, (1998)). In contrast, Mangurkar *et al.* (2000) observed that vaccination did not affect the ejaculate volume, initial motility, pre freezing motility and post freezing motility.

During post vaccination period, occurrence of a febrile reaction causes rise in body temperature as well as that of the testis (Gahlot and Kohli 1981; Venkatareddy *et al.*, 1991 and Murugavel *et al.*, 1997). This rise in testicular temperature acts as a stress factor as there is a corresponding decline in the proportion of progressively motile and live spermatozoa and a subsequent increase in the incidence of morphologically abnormal spermatozoa, especially those with defective heads (Barth and Oko, 1989). The recovery is dependent upon the nature and duration of the thermal insult. The rise in body temperature has its deleterious effect on both the process of spermatogenesis (Venkatareddy *et al.*, 1991) and even on the fully formed sperm (Anderson, 2001). All stages of spermatogenesis are susceptible, with the extent of damage related to the extent and duration of the increased temperature (Waites and Setchell, 1990). This may result in decreased fertilization rates and an increased incidence of embryonic death (Burfening and Ulberg, 1968). The administration of vaccines to the animal can also lead to epididymal dysfunction and thereby cause increase in number of spermatozoal abnormalities in the initial stage resulting in reduced motility (Rao and Venkataswami, 1974 and Rao, 1976). Consequently the epididymal sperm reserves start declining, resorption of abnormal sperms increases (Rao *et al.*, 1980) which leads to a fall down in sperm concentration. Considering the type of vaccine, compared to bacterial vaccine, viral vaccine has more of deleterious effects. With the administration of viral vaccine both the metabolic activity and 'cold shock' resistance of sperm are reduced to a considerable level (Venkataswami *et al.*, 1972). Persual of literature showed that semen quality of exotic (Saxena and Tripathi, 1977; Gahlot and Kohli, 1981 and Gahlot *et al.*, 1990), crossbred bulls (Venkatareddy *et al.* 1991 and Mathur *et al.* 2003) and buffalo bulls (Tripathi and Saxena 1976) is adversely affected by vaccinations against FMD, HS and BQ. But still the information available in concern with the semen quality during post vaccination period in indigenous breeds like Sahiwal bulls is very less.

MATERIALS AND METHODS

The present study was carried out with 6 Sahiwal breeding bulls maintained at Artificial Breeding Complex, NDRI, Karnal, Haryana, India, under standard managemental practices. The observations were recorded for the time span of two years, from 2002 to 2004 and based on this data effect of FMD vaccination was studied on semen quality parameters of bulls. The basic criterion of bulls' selection was that those which produced freezable quality semen and gave at least 4 ejaculates each during pre-vaccination and post-vaccination period. Semen was collected in the morning by AV technique and on each collection; two ejaculates were taken with a 20 to 30 min gap in between two successive ejaculates. Each ejaculate was preceded by a period of sexual preparation consisting of at least two false mounts separated by about one minute restraint. The semen quality evaluation was done one month before and one month after vaccination. A total of 49 ejaculates were taken before vaccination, which served as control. FMD (concentrated tetravalent) vaccine (Intervet Ltd., India) was administered @ 2.5ml by SC injection route, which contained FMD virus types O, A, C and Asia 1 strains. A total of 32 ejaculates were collected after vaccination to study the effect of vaccination stress, if any. These ejaculates were subjected to semen evaluation (Ejaculate Volume, Mass Activity, Initial Motility) by standard methods Tomar *et al.*, 1966. Sperm concentration was evaluated by using improved Neubauer's chamber method. The data regarding all the semen quality parameters was adjusted against the season, period and age with respect to significance and then the adjusted data were subjected to least square analysis (Snedecor and Cochran, 1994) to study the effect of non genetic factors on the semen quality parameters.

RESULTS AND DISCUSSION

It is desirable that the bulls kept for AI purpose should be maintained in a disease free area. The bulls are routinely vaccinated against bacterial and viral diseases e.g., foot-and-mouth disease (FMD), haemorrhagic septicaemia (HS) and black quarter (BQ) in the countries where these diseases are prevalent. This is being practiced in all the bull studs and bull mother farms in India. However, the available reports on the effect of such vaccination on semen quality are conflicting. Vaccination is one of the major anaphylactic stress factors that affect the semen quality (Gahlot and Kohli, 1981; Venkatareddy *et al.*, 1991 and Murugavel *et al.*, 1997). Viral vaccination produces more deleterious effect than that with bacterial vaccines (Venkataswami *et al.*, 1972). Foot and Mouth Disease (FMD) vaccination adversely affects the semen quality (Saxena *et al.*, 1976) of exotic and crossbred bulls (Saxena and Tripathi, 1976; Saxena and Tripathi, 1977; Gahlot *et al.*, 1990 and Venkatareddy *et al.*, 1991). Most of the earlier authors (Venkataswamy and Rao, 1970; Rao, 1974 and Tripathi and Saxena, 1976) studied the effect of formalised gel FMD vaccine.

The least squares means values for various quantitative and qualitative attributes of semen of 6 sahiwal bulls during pre and post-vaccination periods and analysis of variance for the effect of vaccination are depicted in Table- 1 and 2, respectively and graphically represented in Figs-1 and 2, respectively. In general a declining trend in all the semen quality parameters was observed after vaccination.

The mean ejaculate volume (ml) of sahiwal bulls was 4.48 ± 0.24 ml before vaccination and post vaccination the volume was 4.70 ± 0.29 ml, but there was no significant (P>0.05) variation in semen volume during pre- or post- vaccination period in sahiwal bulls, thus signifying that vaccination did not affect the volume of semen. These findings are similar to the reports of Rao (1974); Tripathi and Saxena (1976); Saxena and Tripathi (1977; Kammar and Gangadhar (1998); Mangurkar *et al.* (2000) and Singh *et al.* (2003), on semen volume following vaccination. The major portion in the semen is the seminal plasma which is contributed by the accessory sex glands (Roberts, 1986). The secondary activities of accessory sex glands remain unaffected following vaccination (Radhakrishnan *et al.*, 1975). So, this can be considered as a possible cause for no change in the ejaculate volume. However, Venkatareddy *et al.* (1991) in Ongole, Jersey and Ongole x Jersey breeds reported an increase volume of semen. There was no significant (P>0.05) variation in total volume per day during pre- or post- vaccination period in sahiwal bulls, thus suggesting that vaccination that did not affect the total volume per day. Whereas Mass Activity (MA) was significantly (P>0.05) decreased 2.20 ± 0.12 vs. 1.73 ± 0.14 sahiwal bulls after vaccination. Pre-vaccination progressive motility (%) was 55.41 ± 0.07 %, and after vaccination the value was 48.07 ± 0.11 % respectively. IM was decreased after vaccination, but it was not significant (P>0.05). Similarly, Verma (1996) had reported decrease in motility following administration of the drugs. There was inverse correlation between progressive motility and the tail abnormalities (r=-0.64). However, there was a positive correlation between live sperm count and progressive motility (r=0.433). The decrease in sperm motility may be due to the

Table 1. Least-square means ± S.E. for FMD vaccination effect on various semen characteristics of Sahiwal bulls

Parameters	Pre-vaccination		Post-vaccination	
	N	LSM ± SE	N	LSM ± SE
Ejaculate Volume (ml)	49	4.48 ± 0.24	32	4.70 ± 0.29
Volume (ml)/ day	31	7.07 ± 0.60	24	6.27 ± 0.68
Mass Activity (0-5 scale)	49	2.20 ± 0.12	32	1.73 ± 0.14
Initial Motility (%)	49	55.41 ± 0.07	32	48.07 ± 0.11
Sperm concentration (million/ml)	49	737.76 ± 41.21	32	540.63 ± 50.99
Sperm concentration per ejaculate(million)	49	3244.69 ± 238.21	32	2700.00 ± 294.77

Table 2. Least-square ANOVA for FMD vaccination effect on various semen characteristics of Sahiwal bulls (M.S.S. Values)

S. No.	Parameters	Source of Variation	d.f.	M.S.S.
1	Volume (ml)	Vaccination	1	1.00
		Error	79	2.71
2	Volume (ml)/ Day	Vaccination	1	8.73
		Error	53	10.98
3	Mass Activity (0-4)	Vaccination	1	4.27*
		Error	79	0.66
4	Initial Motility (%)	Vaccination	1	342.77
		Error	79	111.62
5	Sperm concentration (million/ml)	Vaccination	1	752258.2**
		Error	79	83225.83
6	Sperm concentration per ejaculate(million)	Vaccination	1	5743359.84
		Error	79	2780369.88

** P<0.01; *, P<0.05

anaphylactic stress effect of vaccination, which was depicted by the significant rise in body temperature as well as temperature of testes causes derangement in epididymal functions and spermatogenesis by vaccination induced testicular degeneration (Venkatareddy *et al.*, 1991). Sperm cell develops the capacity for motility during their passage through the epididymis (Moulikrishan and Rao, 1986). Epididymal dysfunction following vaccination could be the possible cause for decline in motility. Effect of temperature on the fully formed epididymal spermatozoa could give rise to secondary abnormalities (Venkataswami and Rao, 1970). Although heating seems to affect Sertoli and Leydig cell function, germ cells are the most sensitive to heat (Waites and Setchell, 1990). All stages of spermatogenesis are susceptible, with the extent of damage related to the extent and duration of the increased temperature (Waites and Setchell, 1990). Spermatocytes in meiotic prophase are killed by heat, whereas spermatozoa that are more mature usually have metabolic and structural abnormalities (Setchell *et al.*, 1971). Increase in temperature of testis usually decreases the proportion of progressively motile and live spermatozoa, and increases the incidence of morphologically abnormal spermatozoa. Gwazdauskas *et al.* (2006) reported differences in the embryo quality for embryos obtained after IVF with semen samples from bulls that had an intense response to scrotal insulation, may be due to occurrence of abnormal spermatozoa. Decline in motility may also be due to the increase in sperm tail and mid-piece abnormalities. Rao (1976) had reported low sperm motility associated with high incidence of sperm tail defects as a result of epididymal dysfunction and poor handling in the laboratory. In a personal observation by Anderson (2001), one bull recovered from FMD, the concentration of spermatozoa was normal, but the motility was poor (20 percent); nine weeks later the motility was better (80 percent), since then the bull maintained good sperm production. But on the contrary, Venkataswamy and Rao (1970); Rao (1974); Sexena *et al.* (1976); Tripathi and Saxena,

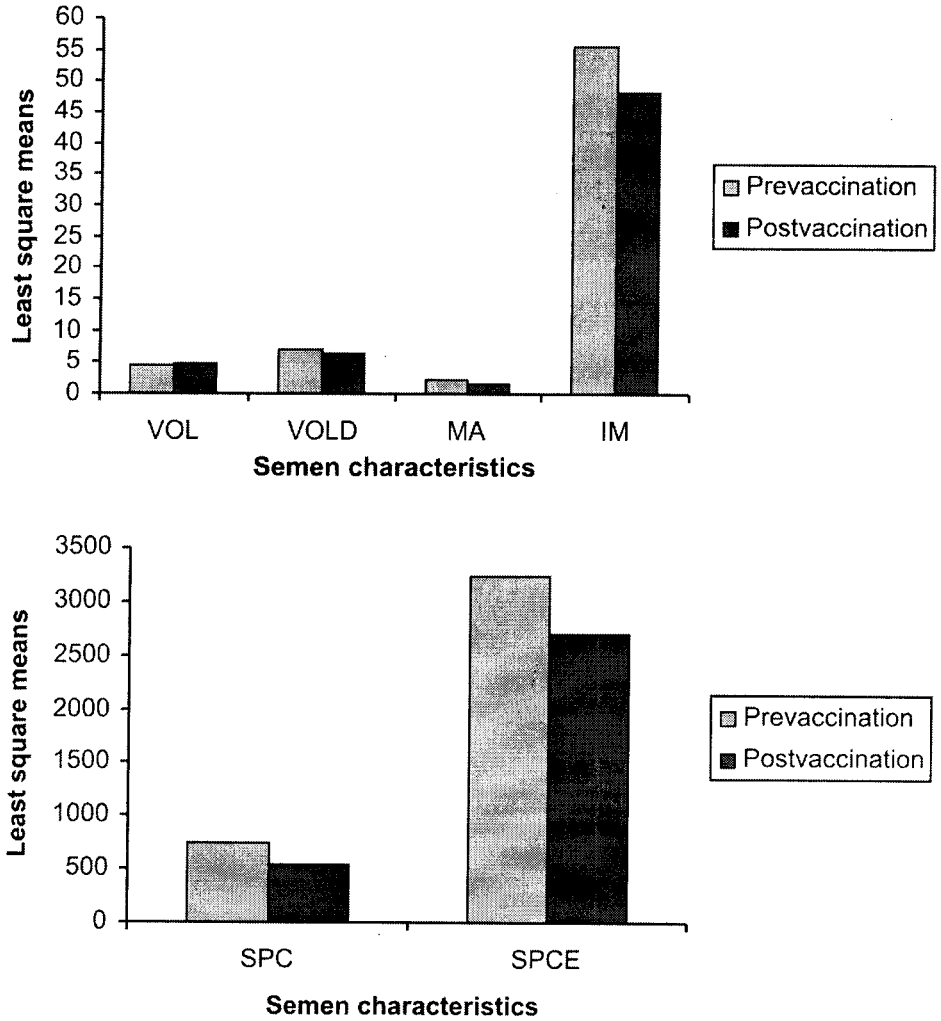

Fig. 2. Effect of FMD vaccination on semen characteristics of Sahiwal bulls

(1976); Venkatareddy *et al.* (1991); Kammar and Gnagadhar (1998) and Singh *et al.* (2003) have reported significant reduction in the motility of bull semen after vaccination. Saxena and Tripathi (1977); Mangurkar *et al.* (2000) and Venkatareddy *et al.* (1991) have reported increase in sperm abnormalities and live sperm count and percent of cold shock resistant sperms during post-vaccination period.

The mean sperm concentration before vaccination was 737.76 ± 41.21 $\times 10^6$/ml in Sahiwal bulls. After vaccination the values were 540.63 ± 50.99 $\times 10^6$/ml. There was significant decrease (P>0.01) in sperm concentration of both the breed following vaccination. Our findings are akin to the earlier reports of Venkataswamy and Rao (1970); Venkatareddy *et al.* (1991) and Singh *et al.* (2003). On the contrary, Kammar and Gnagadhar (1998) reported no adverse effect of vaccination on sperm concentration during post vaccination period. Similar trend like sperm concentration was observed in the case of total sperm output was decreased 3244.69 ± 238.21 vs. 2700.00 ± 294.77 in sahiwal bulls after vaccination, but it was not significant (P>0.05). The decreased sperm concentration may be due to the adverse effects of therapeutic agents on germinal cells resulting into increase in dead spermatozoa, which are absorbed by leucocytes through phagocytosis (Mann and Mann, 1981). The adverse effects of vaccination may be like the adverse effects produced by therapeutic agents or like degenerative changes in germinal epithelium. The increased resorption of abnormal spermatozoa leads to reduction in epididymal sperm reserves (Rao *et al.*, 1980), thus decreasing concentration. So the decreased sperm concentration following vaccination may be probably due to an increase in resorption of spermatozoa in the epididymis.

The probable causes for febrile reaction during vaccination (Hanly *et al.*, 1998) can be 1. adjuvant used (Contamination, Quality) and 2. antigen used (not purified bacterial antigen, viral antigen not clarified). The degenerative changes in testicular structure can be attributed not only to the post-vaccination febrile reaction but to additional possible factor related to the immunogenic nature of the vaccine as well, since inflammatory reaction was observed in the intertubular space (Sahatpure and Patil, 2003). When testicular temperature increases, metabolism increases at a greater rate than blood flow and hence the testes become hypoxic. Therefore, the testes are very susceptible to temperature increases due to endogenous or exogenous factors (e.g. fever, high ambient temperature). As testicular temperature increases, the proportion of defective spermatozoa increases; recovery is dependent upon the nature and duration of the thermal insult. The testis usually operates on the brink of hypoxia (Setchell, 1978). Increased temperature increases metabolism, with a concurrent need for increased oxygen to sustain aerobic metabolism. However, studies in rams (Setchell, 1978) have shown that blood flow changes little in response to increases in testicular temperature and consequently the testes become hypoxic. Increasing blood oxygen saturation is not practical since the blood is nearly completely saturated under normal conditions. Although increasing blood flow would increase the delivery of oxygen, it would also bring considerable additional heat into the testes. The increased body temperature may affect not only spermatogenesis but also fully formed sperm (Anderson, 2001).

From the above discussions, it is clear that the application of FMD vaccine has an adverse effect. Mass activity and sperm concentration have been affected adversely in sahiwal bulls. Febrile reaction due to vaccination results in to the testicular degeneration although of very mild nature. So, the spermiograms affected following vaccination suggests that in bovines, the semen collection and preservation should be suspended till normal fertility of sperm is restored to avoid the failure of conception from AI using such semen.

REFERENCES

Anderson, J. 2001. The semen of animals and its use for artificial insemination. Greenworld publishers, Lucknow, India, First Ind. Reprint.

Barth, A.D. and R.J. Oko. 1989. Abnormal morphology of bovine spermatozoa. Iowa state university press, Ames, pp.17 and 37.

Burfening, P.J. and L.C. Ulberg. 1968: Embryonic survival subsequent to culture of rabbit spermatozoa at 38°C and 40°C. J. Reprod. Fertil., 15: 87-92.

Gahlot, P.S. and T.S. Kohli. 1981. Effect of Rinderpest vaccination (freeze dried) on the semen quality of jersey bulls maintained in the arid zone. Indian Vet. J. 58: 1001-1002.

Gahlot, P.S., B.L. Bishnoi and T.S. Kohli. 1990. Semen studies during vaccination stress in jersey bulls. Indian J. Anim. Reprodn. 11(2): 109-110.

Gwazdauskas, F.C., A.H. Walters, R.G. Saacke and R.E. Pearson. 2006. Assessment of early embryonic development following bovine IVF with abnormal spermatozoa. Symposium on frontiers in reproduction: concepts and application in genomic era & 16th annual meeting of the Indian society for the study of reproduction and fertility (ISSRF). Feb 23-25, pp.47.

Hanly, W.C., B.T. Bennett and J.E. Artwohl. 1997. Overview of adjuvants. AWIC-Resource-Series, 3: 1-8.

Kammar, N.F. and K.S. Gangadhar. 1998. Effect of foot-and-mouth vaccination on reaction time and some seminal characteristics in Surti bulls. Indian J. Anim. Reprodn. 19(2): 149-50.

Mangurkar, B.R., Y.P. Phadnis and M.R. Bhosrekar. 2000. Effect of Foot and mouth disease vaccination on semen characteristics of exotic and cross-bred bulls. Ind. J. Anim. Reprod. 21(2): 135-137.

Mann, T. and C.L. Mann. 1981. Male Reproductive Funtion and Semen. Springer-Verlag Berlin. Heidelberg, New York, USA.

Mathur, A.K., S. Tyagi, D.K. Mandal and S.P. Singh. 2003. Effect of multiple vaccinations on the semen quality of Frieswal bulls. Indian J. Anim. Sci. 73(8): 864-866.

Moulikrishan, K. and A. Ramamohan Rao. 1986. Epididymal dysfunction in buffalo bulls. Ind. Vet. J. 63: 1013-1016.

Moulikrishan, K. and A. Ramamohan Rao. 1988. Epididymal dysfunction in buffalo bulls. Ind. Vet. J. 63: 1013-1016.

Murugavel, K., C. Veerapandian and A. Subramanian. 1997. Effect of black quarter vaccination on semen quality in Murrah bulls. Indian J. Anim. Sci. 67: 597-598.

Radhakrishnan, R., V. Venkataswami and S.R. Pattabiraman. 1975. Further report on the effect of protective vaccination on semen quality of breeding bulls. Indian Vet J. 52: 620-625.

Rao Narsimha, A.V. 1974. Vaccination stress in crossbred bulls. Indian Vet. J. 51: 74-75.

Rao, A.R. 1976. Sperm akinesia in a hollikar bull. Indian Vet. J. 53: 414-418.

Rao, A.R., A. Bane and B.K. Gustafasson. 1980. Changes in the morphology of spermatozoa during their passage through the genital tract in dairy bulls with normal and impaired spermatogenesis. Theriogenology, 14(1): 10-12.

Rao, V.J. and V. Venkataswami. 1974. Histo-pathology of testicular tissue of crossbred bulls in Foot and Mouth disease vaccination. II. Indian Vet. J. 48: 1101.

Roberts, S.J. 1986. Veterinary Obstetrics and Genital Diseases. Wood stock, Vermount.

Sahatpure, S.K. and R.K. Patil. 2003. Effects of Haemorrhagic septicaemia vaccine with two different adjuvants on the histological structures of testis and epididymis in crossbred bulls. National Seminar on Frozen Semen Technology, Dhoni, Kerala, India, pp.227-230.

Saxena, V.B. and S.S. Tripathi. 1977. Effect of foot and mouth disease vaccination on semen quantity, quality and preservability in Jersey bulls. Indian Vet. J. 54: 959-964.

Saxena, V.B., S.S. Tripathi, M.C. Verma and R. Singh. 1976. Effect of vaccination on semen quality parameters in crossbred bulls. Curr. Sci. 45: 154.

Setchell, B.P. 1978. The scrotum and thermoregulation. In: B.P. Setchell (ed.) The mammalian testis. Ithaca: Cornell University Press, pp.90-108.

Setchell, B.P., J.K. Voglmayr and N.T. Hinks. 1971. The effect of local heating on the flow and composition of rete testis fluid in the conscious ram. J. Reprod. Fertil. 24: 81-89.

Singh, R., H.K. Verma and S. Kumar. 2003. Effect of the foot and mouth disease vaccination on the semen quality of buffalo bulls. Indian J. Anim. Sci. 73: 1319-1323.

Snedecor, G.W. and W.G. Cochran. 1994. Statistical Methods (6th Ed) Oxford and IBH Publ. Co., New Delhi.

Tomar, N.S., B.S. Mishra and C.B. Johari. 1966. Seasonal variations in reaction time and semen production, and prediction of some semen attributes on initial motility of spermatozoa in Hariana and Murrah bulls. Indian J. Dairy Sci. 19: 87-93.

Tripathi, S.S. and V.B. Saxena. 1976. A note on the foot-and-mouth vaccination stress on quantity, quality and preservability of semen of Murrah buffalo bulls. Indian J. Anim. Sci. 46: 44-47.

Venkatareddy, J., A. Venkatamunichetty, S.V. Ramachandran and Sreeraman 1991: Effect of foot and mouth disease vaccination on semen quality. Indian J. Anim. Prod. 12: 13-14.

Venkataswami, V. and V.J. Rao. 1970. Preliminary report on the effect of foot and mouth disease vaccination on semen quality of crossbred bulls-1. Indian Vet. J. 47: 23-29.

Venkataswami, V., J. Pattabiraman Daniel and Sunderavadanam. K. 1972. Effect of vaccination on spermatozoa resistance and on their metabolic activity. Indian Vet. J. 49: 1012-1016.

Verma, H.K. 1996. Studies on the effect of therapeutic agents on the seminal attributes of buffalo bulls. Ph.D. Thesis, PAU, Ludhiana, India.

Verma, H.K. 1996. Studies on the effect of therapeutic agents on the seminal attributes of buffalo bulls. Ph.D. Thesis, PAU, Ludhiana, India.

Waites, G.M.H. and B.P. Setchell. 1990. Physiology of the Mammalian Testis. In: G.E. Lamming (ed.) Marshall's Physiology of Reproduction, 4th Ed., Vol. 2, Reproduction in the Male. Edinburgh: Churchill Livingstone, pp.1-105.