



Shelter designing and season affects scrotal thermal profile vis-a-vis semen quality in Frieswal breeding bulls

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ABSTRACT

Twenty-four Frieswal bulls (Holstein Friesian × Sahiwal) were randomly distributed according to their housing: animals housed individually in sheds with partition (n = 12; PS) and without partition (n = 12; PW) wall. The present study revealed that the alteration in design of bull housing affected THI leading to changes in scrotal thermal gradient in crossbred Frieswal bulls during different seasons. Scrotal thermal gradient was higher in PW bulls than PS bulls during afternoon period in both the seasons. The semen quality was better in the bulls with higher thermal gradient in well-ventilated sheds during hot-humid season. The season and design of bull housing affected sperm morphology during different seasons. Higher total antioxidant capacity was estimated in the bulls of PW than in bulls of PS during hot-humid season, but no difference was noticed during winter. It was concluded that the season and design of housing can affect scrotal thermal gradient and semen quality in crossbred breeding bulls.

Keywords: Breeding bull, Housing, Infrared thermography, Season, Semen quality

The quality of male germplasm plays an important role in number of inseminations per successful conception. Several studies have reported that the semen quality of bull was affected by season, especially in tropical climates (Koivisto *et al.* 2009, Snoj *et al.* 2013), however, other studies have not been able to detect a seasonal effect (Brito *et al.* 2002, Prastowo *et al.* 2019). Heat stress can occur in bulls of any breed kept under widely different environmental conditions; seasonal effects on sperm quality can be observed in many countries (Morrell 2020).

Spermatogenesis is temperature-dependent and optimal spermatogenesis occurs when testicular temperatures are maintained 4–6°C lower than the core body temperature (Mieusset and Bujan 1995, Kastelic *et al.* 2018). This fact is supported by studies including Johnston *et al.* (1963), who reported a decrease in semen quality caused by thermal stress in bulls, and by Kastelic (2014), who verified that the increased difference between the body temperature and testicular temperature favoured spermatogenesis. Recording of scrotal surface and ocular area temperature by infrared thermography (IRT) may better reflect the effects of ambient temperature on the reproductive capacity of bulls. However, there is still a lack of precise information investigating the influence of combined effect of season and shed design on scrotal thermoregulation and semen characteristics of Frieswal bulls. Therefore, the objective of the present study was to use scrotal IRT to determine

effects of environmental conditions and thermal stress on semen quality of Frieswal bulls.

MATERIALS AND METHODS

The study was conducted during Hot-humid and winter season at ICAR-Central Institute for Research on Cattle, Meerut, India. For this study, 24 Frieswal (Holstein Friesian × Sahiwal) crossbred bulls were randomly distributed according to their housing: animals housed in sheds with partition (n = 12; PS) and without partition (n = 12; PW) wall. The experimental bulls were kept in individual pens having covered and open area with equal floor space/bull in both types of sheds. The concrete wall (1.8 m height) partitioned the individual bull pens in PS, however, galvanized iron pipes were used for partition in PW bull pens. All the bulls were managed and fed under standard farm conditions.

Ambient temperature and relative humidity were recorded using data loggers every one hour inside the pens. The THI (temperature humidity index) was calculated according to the following formula described by McDowell (1972):

$$THI = 0.72 (Cdb + Cwb) + 40.6$$

Semen samples were collected by artificial vagina technique twice a week from each bull during entire experimental period. The fresh ejaculates were subjected to evaluation for volume (ml), initial progressive sperm motility (%), sperm concentration ($\times 10^6$ /ml), sperm morphology and acrosome integrity. The frozen semen samples were examined for post-thaw sperm motility

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(PTM) after 24 hr period of freezing. To evaluate the sperm morphology, Eosin-Nigrosin stain was used to differentiate viable (live) and non-viable (dead) spermatozoa (Campbell *et al.* 1953).

Scrotal surface and ocular temperature of each bull was recorded with infrared thermal imaging camera before semen collection in the morning and during afternoon on the same day at fortnightly interval during the study period. The blood samples from experimental bulls were collected once during each season for estimation of total antioxidant capacity (TAC). Antioxidants in samples were analysed by Antioxidant Assay Kit (Cayman Chemical).

The experimental data were analysed using t-test to determine significant differences in all the parameters recorded between groups using the SPSS/PC computer programme (Version 20.0, SPSS, Chicago, IL, USA). The differences with values of $p < 0.05$ were considered to be statistically significant.

RESULTS AND DISCUSSION

The ambient temperature ranged from 23 to 40.5°C during Hot-humid season and from 3.5 to 24.5°C during winter in PS bull sheds. However, it ranged from 23 to 38.5°C during Hot-humid season and from 4.0 to 21.5°C during winter in PW bull sheds. The results revealed that the experimental bulls of all the sheds were under moderate thermal stress during Hot-humid season; however, they were in comfortable zone during winter season (Fig. 1). Mean THI values were significantly higher ($p < 0.05$) in PS sheds than in PW sheds during both the seasons, which might be due to open area towards south direction and entry of maximum sunrays into the shed (Table 1). In agreement to the present study, Samer (2011) recorded improvement in THI in well-ventilated animal sheds. Although minimum lower value of THI and mean least THI was observed in PW sheds in the morning, during afternoon period mean THI was significantly ($p < 0.05$) higher in PS than in PW sheds during Hot-humid season (Table 1).

Scrotal thermal gradient (TG) was significantly higher ($p < 0.05$) in PW bulls than the PS bulls during afternoon period, however, no difference was observed between these groups during morning period in both the seasons (Table 1). The results of the present study revealed that

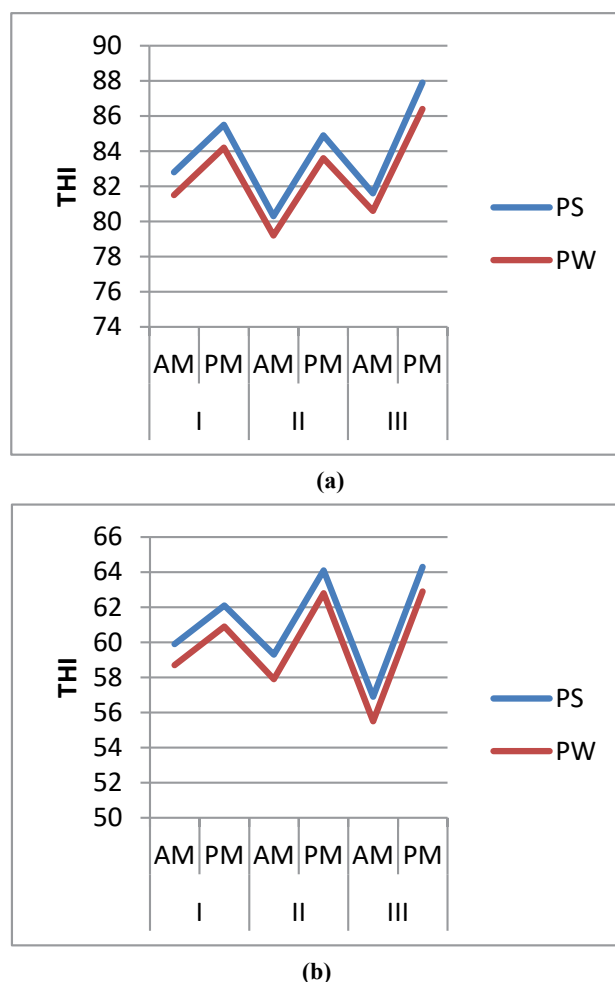


Fig. 1. Pattern of THI at different intervals in PS and PW bull sheds during (a) Hot-humid and (b) winter season (AM, Morning period; PM, Afternoon period).

TG was better during winter than in Hot-humid season irrespective of design of shed. During Hot-humid season, higher ($p < 0.05$) TG in PW than in PS indicated efficient scrotal thermoregulation which might be due to better ventilation and evaporative cooling in the sheds. PW bulls had significantly ($p < 0.05$) lower ocular temperature during afternoon period in winter season. In agreement to the present findings, Ahirwar *et al.* (2017) reported that the season and management had significant ($p < 0.05$) effect on

Table 1. Mean values ($\pm$ SE) for THI, scrotal thermal gradient and ocular temperature

Parameter	Period of day	Hot-humid season		Winter season	
		PS	PW	PS	PW
THI	AM	81.55 $\pm$ 0.31 ^a	80.42 $\pm$ 0.33 ^b	58.69 $\pm$ 0.43 ^a	57.39 $\pm$ 0.43 ^b
	PM	86.07 $\pm$ 0.40 ^a	84.72 $\pm$ 0.39 ^b	63.46 $\pm$ 0.31 ^a	62.14 $\pm$ 0.29 ^b
THI range	AM	73.94 - 86.25	72.86 - 85.24	53.42 - 64.65	52.19 - 62.92
	PM	76.6 - 93.16	75.45 - 91.00	59.82 - 68.61	58.60 - 67.17
TG (°C)	AM	2.29 $\pm$ 0.20	2.58 $\pm$ 0.18	4.9 $\pm$ 0.24	5.19 $\pm$ 0.17
	PM	0.91 $\pm$ 0.14 ^b	1.34 $\pm$ 0.19 ^a	1.6 $\pm$ 0.18 ^b	2.12 $\pm$ 0.19 ^a
Ocular temperature (°C)	AM	36.89 $\pm$ 0.42	36.05 $\pm$ 0.26	35.72 $\pm$ 0.57	35.2 $\pm$ 0.47
	PM	39.35 $\pm$ 0.34	38.99 $\pm$ 0.40	39.33 $\pm$ 0.34 ^a	36.19 $\pm$ 0.40 ^b

Means with different superscripts between columns under specific season differ significantly ($p < 0.05$). THI, Temperature humidity index; TG, scrotal thermal gradient; AM, Morning period; PM, Afternoon period.

Table 2. Effect of season and housing on semen quality in crossbred Frieswal bulls

Parameter	Hot-humid season		Winter season	
	PS	PW	PS	PW
Semen volume (ml)	4.22±0.10 ^b	5.41±0.12 ^a	5.17±0.13	5.17±0.14
Sperm concentration (million/ml)	763.33±29.83 ^b	850.58±30.62 ^a	1064.31±41.80 ^a	932.14±35.99 ^b
Sperm concentration per ejaculate (million)	3341.02± 172.12 ^b	4776.14 ±211.09 ^a	5464.47 ±247.67	4936.99 ±247.40
IPM (%)	45.57±1.46 ^b	58.14±1.15 ^a	53.12±1.39	54.67±1.21
PTM (%)	37.05±1.58	39.87±1.06	33.04±1.35	34.13±1.44

Means with different superscripts between columns under specific season differ significantly ($p < 0.05$). IPM, Initial progressive sperm motility; PTM, Post thaw motility.

scrotal surface and ocular temperature in buffalo breeding bulls. It has been found that due to rise of microclimatic temperature, the temperature of body surface including scrotum rose concomitantly in Frieswal breeding bulls (Sirohi *et al.* 2017). PW bulls had significantly ($p < 0.05$) lower ocular temperature during afternoon period in winter season; however, no significant effect was observed on ocular temperature in the experimental bulls during Hot-humid season.

Mean semen volume, sperm concentration/ml of semen, sperm concentration per ejaculate and initial sperm progressive motility were significantly ($p < 0.05$) higher in PW bulls than the PS bulls during Hot-humid season (Table 2). The bulls kept under PS housing had significantly ($p < 0.05$) higher sperm concentration/ml of semen than PW bulls during winter season. In the present study, more ventilated housing favoured quality semen production during Hot-humid season. In agreement to the present study, Landaeta-Hernandez *et al.* (2020) reported that semen characteristics and quality varied throughout the year with there being important genotype $\times$ seasonal interactions. Factors such as environment, housing and breed have been shown to influence sperm quality in some studies (Suriyasomboon *et al.* 2005, Snoj *et al.* 2013). No difference ($p > 0.05$) was observed for PTM values of frozen samples processed from the bulls of both the groups and during both the seasons.

Total sperm abnormalities including head, mid-piece and tail were within the normal range without any difference between two groups during both the seasons. During winter season, although mid-piece sperm abnormalities were significantly ($p < 0.05$) higher in PW (10.0±0.99) than in PS (7.35±0.78) bulls, total sperm abnormalities in bulls of both the groups were within the normal prescribed limits for further processing of semen for freezing. Alves *et al.* (2021) reported that scrotal heat stress affected male fertility by increasing sperm morphological abnormalities. In summer, reactive oxygen species production significantly increases in sperm, leading to lipid peroxidation and major sperm defects (Nichi *et al.* 2006). Per cent acrosome integrity was significantly ($p < 0.05$) higher in PS (82.21±0.94) bulls than in PW (77.71±1.58) bulls during winter season.

Significantly ($p < 0.05$) higher TAC was estimated in the bulls of PW (7.88±0.28 mM) than in the bulls of PS (1.26±0.24 mM) during Hot-humid season. The bulls of PS (5.87±0.24 mM) and PW (5.42±0.13 mM) had no

difference for TAC during winter season. The testicular heat stress activates an increase in cell metabolism associated with higher activity of the epithelial cells from seminiferous tubule (Setchell 1998) leading to hypoxia that results in oxidative stress (Paul *et al.* 2009). The results of the present study revealed that more ventilated sheds made comfortable shelter in PW which favoured better antioxidant response.

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