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Impact of Indian Climate in Terms of Temperature Humidity Index on Reproductive and Semen Parameters of Barbari Bucks

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ABSTRACT

The present study was conducted to investigate the effect of temperature-humidity index on the reproductive and semen parameters of Barbari bucks. Twelve adult Barbari bucks aged between 1.5 to 4.0 years were selected and managed under a semi-intensive system. A total of 288 ejaculates were collected from the chosen bucks during the experiment. Detailed sexual characteristics and breeding behaviors were recorded during semen collection. Immediately after collection, the initial evaluation of the semen samples was carried out within 10 min, and then the sample was diluted with a Tris-based semen extender (TCEYG Buffer). The temperature humidity index (THI) was calculated to evaluate its effect on reproductive parameters. The result reveals that there was mild stress in the August and September months, whereas no stress was seen in other months of the study period. Correlation between climatic parameters with different semen parameters and the data showed that relative humidity was highly significant ($p \le 0.01$) and negatively correlated with pH. However, other climatic parameters were negatively correlated with seminal parameters.

Keywords: Barbari buck, Temperature humidity index, Heat stress, Semen.

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INTRODUCTION

The Barbari goat, a popular breed renowned for its high reproductive efficiency and adaptability to various climatic conditions, plays a pivotal role in meeting the ever-growing demand for meat and milk production worldwide. However, the reproductive performance of these bucks can be significantly influenced by environmental factors, particularly temperature

And humidity. **The climate** is the conditions of the <u>atmosphere</u> at a particular location over a long period. It is the long-term summation of the atmospheric elements and their variations that, over short periods, <u>constitute weather</u>. These elements are solar radiation, temperature, humidity, precipitation (type, frequency, and amount), atmospheric pressure and wind (speed and direction).

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The Temperature Humidity Index (THI) serves as a valuable indicator of thermal stress, reflecting the combined effect of temperature and humidity on animal comfort and productivity. In recent years, with the escalating concerns over climate change and its profound impact on livestock farming, researchers have intensified their focus on understanding how climatic variables, such as THI, influence the reproductive physiology of Barbari bucks. Elevated THI levels have been associated with decreased semen quality, altered hormonal profiles, and impaired testicular function in various goat breeds, including Barbari. Such adverse effects can lead to reduced fertility rates, delayed onset of puberty and ultimately, diminished reproductive performance, posing significant challenges to sustainable goat farming practices.

Despite the growing body of research elucidating the relationship between THI and reproductive parameters in livestock species, there remains a paucity of comprehensive studies specifically addressing Barbari bucks in the Indian climate. Given their economic importance and vulnerability to environmental stressors, it becomes imperative to investigate the precise impact of THI on key reproductive indicators, such as semen quality, libido, and sperm production, in Barbari bucks in Indian climatic conditions.

In goat breeding programs, male fertility is crucial since it directly impacts the herd's ability to reproduce and progress genetically and the seasonality of reproduction has a big impact on the quality of the semen produced. Temperature, humidity and sunshine are environmental elements that have an impact on the reproduction of animals. The temperature humidity index (THI) is the primary indication used to track an animal's state of thermal stress (Ribeiro et al., 2018). Elevated temperatures during the sweltering summer months are linked to poor animal reproduction. The study aimed to explore the effect of THI on the buck's fertility. Understanding how environmental factors, such as temperature, humidity and lighting, affect buck's health and productivity is essential.

MATERIALS AND METHODS

Location of study

The present study was carried out at the Department of Veterinary Gynaecology and Obstetrics, College of Veterinary Science and Animal Husbandry, NDVSU, Jabalpur (M.P), India. Prior consent was obtained for the use of experimental bucks from the ethical committee of the college body.

Ethical approval from the Institutional Animal Ethics Committee

The use of animals for this study was permitted by the aforementioned college's ethical committee (Ethical Approval No.: 14/IAEC/Vety/2023).

Experimental animals

For the present study, twelve sexually mature adult male Barbari bucks aged between 1.5 to 4.0 years were selected and managed under a semi-intensive system at a livestock farm in Amanala, NDVSU, Jabalpur, India. All the bucks were reared under uniform conditions of feeding, management and housing. All the bucks were previously trained to ejaculate into the artificial vagina while using a dummy doe.

Meteorological Variables

The record of environmental data was taken from the Meteorological Station, Jabalpur, M.P., India, situated at Latitude: 23.17°N; Longitude: 79.57°E and 410.87 MSL (meters above sea level). The data considered average dry bulb temperature (Tdb), relative humidity (RH) and sunshine hours (SH) during the entire study period. These values were recorded at 08.00 AM daily at the time of semen collection and averaged every month throughout the study period.

THI Calculation

For evaluating the effect of the temperature humidity index (THI) on reproductive parameters, average dry bulb temperature (Tdb), relative humidity (RH) and sunshine hours (SH) of every month were collected from the Meteorological station. The THI was calculated as per the formula given by Ranjan et al. (2020). The data so obtained was analyzed accordingly for the effect of the THI index on reproductive behavior and semen parameters (Table:01). The THI was estimated using the following formula:

 $THI = Tdb - [0.55 - (0.55 \times RH/100)] \times (Tdb - 58)$

**THI- Temperature Humidity Index; DBT/Tdb(°F)- Dry Bulb Temperature in Degree Fahrenheit; RH- Relative Humidity (Ranjan et al., 2020). The THI was classified into four groups as follows

Table 1: Classification of the calculated values for the THI:

S. No.	THI Value	Stress Level
1.	<72	No heat stress
2.	72 to 79	Mild heat stress
3.	80 to 89	Moderate heat stress
4.	>90	Severe heat stress

Andrological Behaviour

Sexually mature Barbari bucks and does were kept separately and managed under a semi-intensive system. Mounting behavior and libido of 12 Barbari bucks were examined before semen ejaculation and during semen collection based on the following parameters:

Sexual aggressiveness (SA)

The behavior of the bucks during the approach of the doe was assessed visually and classified as follows, as per the parameters described by Barik et al. (2009) (Table:02)

- a) Aggressive: Extremely eager to mount and approach the doe with full vigor.
- b) Active: Approached the doe with less vigor and aggression.
- c) Dull: Proceeded with a dull expression and took a longer time to mount.
- d) Shy: Exhibited mild sexual interest and was reluctant to mount.

Table 2: Scorecard for Sexual Aggressiveness of Barbari bucks

S. No.	Sexual Aggressiveness	Score
1.	Aggressive	4
2.	Active	3
3.	Dull	2
4.	Shy	1

Reaction time (RT)

Reaction time is the amount of time spent by a buck between being exposed to a doe or other female animal to mounting. The reaction time was arrived as such as per the parameter described by Barik et al.(2009). (Table:03)

Table 3: Categorization of Reaction Time for Barbari bucks

S. No.	Reaction Time (sec.)	Score
1.	<20	4
2.	21-40	3
3.	41-60	2
4.	61-80	1
5.	>80	0

Semen collection and evaluation

The bucks were trained to ejaculate in the artificial vagina (AV) before 15 days start of the experiment. A collection of semen was done with AV (length = 20 cm and diameter = 4.5 cm) into the calibrated glass collection cup throughout the study period. A total of 288 ejaculates were collected routinely from 12 bucks for 6 6-month period. Immediately after collection, the semen samples were placed in a water bath (37°C) and the initial evaluation was carried out within 10 min, which included volume, color, consistency, pH and mass activity of the ejaculates. Semen was extended in Tris-Egg yolk-citrate-Fructose (TCF) diluent at the rate of 1:10, having 10, 12 and 15% (v/v) egg yolk and 6% glycerol (v/v) to make final dilution.

Sperm concentration:

Sperm concentration was determined manually by using a Hemocytometer. Diluting fluid was prepared manually for the dilution of the semen sample. The number of sperm enumerated in eighty small squares was multiplied by 10⁴ to obtain the number of sperm per cubic millimeter.

Sperm motility

In diluted semen (10 μ l), progressive motility was assessed by placing a drop of the semen sample on a pre-warmed (37°C) glass slide and a coverslip was put over it. Sperm motility of the ejaculate was assessed under high magnification (40x) of the Phase Contrast Microscope and expressed in percentage from 0-100%.

Sperm viability

One drop of Eosin (2%) (Sodium Citrate Dihydrate-2.9g, Eosin stain-5.00g, Double distilled water-100ml) and three to four drops of Nigrosine stain (10%) (Sodium Citrate Dihydrate-2.9g, Nigrosine stain-10.00g, Double distilled water-100ml) were poured and mixed in a glass slide, then a small drop (10 μ L) of semen sample was mixed with it.

The smear was prepared on a clean, grease-free slide (at 37°C) with the mixture on different slides. Under high magnification (40x) of the Phase Contrast Microscope, a total of 200 sperms were counted, keeping a note of dead and live sperms and the percentage of each was worked out. The stained spermatozoa (eosinophilic) obtained a pinkish color and were categorized as dead and the unstained ones, against a dark background of Nigrosin, were counted as live.

Sperm abnormality

A manually made Rose Bengal stain (Rose Bengal stain {HIMEDIA}-3g, Formalin-1ml, Double distilled water-100ml) that offered a high degree of plasma membrane sharpness for visualizing the sperm cell structure was used to determine the proportion of aberrant sperm, which assisted in identifying morphological defects. For this, a thin smear of the semen sample was prepared on a clean, grease-free slide and air dried it. The slide was placed in a jar containing Rose Bengal stain for 5-7 minutes. Washed the slide in running tap water, dried it and examined it under high magnification (100×) of a Phase Contrast Microscope. A total of 200 sperm were counted from various microscopic fields on the slide and each abnormality was noted. The proportion of each was calculated and expressed as a percentage.

Statistical analysis

The data were analyzed using SPSS 24 software, San Jose California USA, www.sigmaplot.com. Data from different experiments were presented as Mean \pm SE. The pair-wise comparison of means was carried out using Fisher's multiple comparison test as per the standard statistical method described by Snedecor and Cochran (1994). The difference at P \leq 0.05 was considered to be satisfactory significant.

RESULTS AND DISCUSSION

Temperature Humidity Index (THI)

The THI was classified into four groups: 1. <72- No heat stress; 2. 72 to 79- Mild heat stress; 3. 80 to 89- Moderate heat stress and 4. >90- Severe heat stress (Table:01).

THI was calculated for the evaluation of the effect of temperature humidity index on reproductive parameters and the result revealed (Table:04) that there was mild stress in August and September, whereas no stress was seen in other months, i.e., October, November, December and January. On the other hand, the highest THI was seen in the first week of September (78.55) and the lowest was seen in the third week of December (47.30).

Table 4: Effect of average THI during different months of the study

		Environme	_	Heat		
S. No.	Month	Temperature (Tdb°C)	RH (%)	SH (Hrs.)	THI (%)	stress level
1.	August	26.10	86.89	02.69	75.74	Mild
2.	September	26.46	86.21	04.87	77.58	Mild
3.	October	22.92	70.35	08.34	70.85	Nil
4.	November	18.96	75.46	05.17	64.97	Nil
5.	December	15.53	65.39	05.53	57.77	Nil
6.	January	13.42	85.89	04.79	56.44	Nil

Tdb°C -Dry Bulb Temperature in Celsius; RH-Relative Humidity; SH-Sunshine hours:

THI-Temperature Humidity Index.

Ranjan et al. (2020) studied the effect of THI on sexual behavior and semen quality in Barbari bucks in Indian climatic conditions (CIRG, Makhdoom) and based on the THI values, they recorded that there was no heat stress during November, December and January; however, mild heat stress during October and moderate heat stress during September. These results are comparable with the present findings, as in the current study, mild heat stress is reported during the August and September months. However, Rahman et al. (2016) also studied the effect of heat stress on bucks' adaptability and semen characteristics in Black Bengal bucks and found that the average temperature was 20.50°C and the average relative humidity was 70.5% in the morning. Hence, the THI value was 19.95 in the morning during their study period, which indicates all the experimental bucks were in the absence of heat stress. This result is also similar to the findings of the present study.

Reproductive parameters of breeding bucks

The mean libido score of Barbari buck was found from 03.35 ± 0.10 (September) to 03.79 ± 0.07 (October) during these six months (Table:05). The analysis of data revealed significantly higher (p<0.05) libido scores during October (03.79±0.07), August, November, followed by January. However, the significantly lowest libido score was revealed during December and September.

Table 5: Mean libido score of Barbari buck at monthly intervals

S. No.	Month	Libido score
1.	August	03.73°±0.07
2.	September	$03.35^{b}\pm0.10$
3.	October	$03.79^{a}\pm0.07$
4.	November	$03.73^{a}\pm0.06$
5.	December	$03.67^{a}\pm0.07$
6.	January	$03.60^{a}\pm0.07$

Mean value bearing different superscripts (a, b) differ significantly columnwise (p<0.05)

Semen parameters

Ejaculate volume was highest in November, however, the mass motility, progressive motility and live-dead sperm count (Mean \pm SE) were significantly higher (P < 0.05) in fresh semen during August in comparison to other months. Significantly (P < 0.05) poorest quality semen was obtained during December and January in the present study. The mean ejaculate volume and pH of Barbari buck semen were found from 0.62 \pm 0.02 (December) to 0.75 \pm 0.03 (November) and 06.61 \pm 0.01 (January) to 06.69 \pm 0.01 (December), during the study period (Table:06).

Table 6: Mean±SE of seminal attributes of Barbari bucks at the pre-freeze stage.

Month	Volume (ml)	рН	Mass Motility (%)	Concentration (million/ml)	Progressive motility (%)	Viability (%)	Abnormality (%)
August	0.64°±0.03	06.64 ^{ab} ±0.01	04.15°±0.09	2891.04 ^{ac} ±49.8	74.27 ^a ±2.15	82.8a ±1.57	5.65±.061
September	$0.66^{ab}\pm0.03$	$06.64^{ab} \pm 0.01$	03.67 ^b ±0.09	3209.17 ^b ±35.98	68.23 ^{ab} ±2.12	79.02 ^{abc} ±1.7	5.60±.052
October	$0.66^{ab}\pm0.03$	06.65 ^{ab} ±0.01	04.13°±0.11	3233.33 ^b ±33.03	68.33 ^{ab} ±2.79	81.67 ^{ab} ±1.29	5.72±.032
November	0.75 ^b ±0.03	06.68 ^b ±0.01	$04.08^a \pm 0.10$	3022.50°±58.57	68.75 ^{ab} ±1.83	77.58 ^{bc} ±1.42	5.68±.036
December	0.62°±0.02	06.69 ^b ±0.01	03.75 ^b ±0.09	2831.46 ^a ±70.02	67.08 ^b ±2.13	76.71°±1.49	5.71±.045
January	0.66 ^{ab} ±0.03	06.61°±0.01	03.65b±0.09	2218.13 ^d ±65.41	54.27ª ±1.51	65.54 ^d ±1.33	5.73±.044

Mean value bearing different superscripts (a,b,c,d) differ significantly column-wise (p<0.05)

The result of the current observations for the volume (ml) of Barbari buck is comparable with the finding of Baghel (2013) and Anand (2016) who reported semen volume 0.68±0.03 and 0.74±0.12 ml, respectivly in fresh semen of Barbari bucks. The findings of the present study for mass motility are quite comparable with the findings of Baghel (2013), who reported 3.82±0.06 and Anand (2016), who found 4.00±0.07 mean mass motility in fresh semen of Barbari bucks. Whereas, Hahn et al. (2019) concluded that after semen collection, pH was within the reference range, *i.e.*, 6.4–7.0 in 65% of the sample. However, about 35% showed a slightly increased pH value (7.2).

The monthly research findings for progressive sperm motility are supported by the fact that in November, fresh and frozen-thawed semen had significantly higher sperm motility when compared to other months. Conversely, in January and February, Barbari bucks produced significantly lower-quality semen (Ranjan et al., 2020). Qureshi et al. (2013) found that the sperm concentration was the highest during November in bucks of dairy goats and the

data are quite similar to the monthly findings of the present study.

The data of mean sperm viability of this study is quite comparable with the non-eosinophilic (live) spermatozoa count of Barbari breeds reported by Singh (2003), who recorded 72.40±1.67%, Tiwari (2000) found 77.55±2.21% live viable sperm in Barbari buck semen during September-November. The data of sperm abnormality percentage is also comparable with the findings of Baghel et al. (2016) in the Barbari breed of bucks, who reported the mean value of percent abnormal spermatozoa in 6 ejaculates ranging from 3.50±0.34 to 4.83±0.48 with a mean value of 4.07±0.19 during winter months.

Correlation among various seminal parameters with environmental variables

The correlation between climatic parameters with macroscopic and microscopic semen parameters was investigated and depicted in Table No. 07.

Table 7: Correlation among various seminal parameters with environmental variables in Barbari bucks

Parameters	Libido Score	Volume	pН	Mass motility	Concentration	Progressive motility	Sperm Viability	Sperm Abnormality
Temp. (°C)	0.393	0.111	-0.155	0.426	0.745	0.760	0.786	-0.785
RH (%)	0.224	-0.015	-0.845^{*}	-0.177	-0.283	-0.133	-0.206	-0.503
SH (Hours)	0.117	0.134	0.235	0.099	0.361	-0.153	0.051	0.619
THI	0.404	0.141	-0.224	0.386	0.722	0.706	0.727	-0.788

^{*}Correlation is significant at the 0.05 level (2-tailed)

The data from the table shows that the temperature was negatively correlated with pH and sperm abnormality. Relative humidity was negatively correlated with volume, mass motility, concentration, individual abnormality, sperm viability and abnormality. However, relative humidity was highly significant(p<0.05) and negatively correlated with pH. The sunshine hour was negatively correlated with individual abnormality, whereas THI was negatively correlated with pH and sperm abnormality.

CONCLUSION

The comfortable Temperature Humidity Index during the October and November months had a positive effect on the buck's libido, with higher semen volume and sperm concentration. On the other hand, mass motility, individual motility and sperm viability were highest during August and sperm abnormalities were substantially lower in September.It is suggested that semen preservation should be carried out in November-December as these months are favorable for semen production and bucks are also in an auspicious environment without any heat stress in tropical regions.

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CONFLICT OF INTEREST

The authors don't have any conflict of interest in the conduct of this experiment.

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