ISSN Print: 2664-844X
ISSN Online: 2664-8458
NAAS Rating (2026): 4.97
IJAFS 2026; 8(8): 366-369
www.agriculturaljournals.com
Received: 02-06-2026
Accepted: 04-07-2026

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Seasonal variation in serum enzyme activities of Nagavali sheep under tropical conditions

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DOI: https://www.doi.org/10.33545/2664844X.2026.v8.i8e.1788

Abstract
This study evaluated seasonal variation in serum aspartate aminotransferase (AST), alanine aminotransferase (ALT), and alkaline phosphatase (ALP) activities in Nagavali sheep maintained under tropical conditions. One hundred apparently healthy sheep (50 ewes and 50 rams), aged at least one year, were sampled during summer, monsoon, and winter. The temperature-humidity index (THI) was highest in summer (80.02 ± 0.43), followed by monsoon (70.51 ± 0.41) and winter (67.89 ± 0.27). Serum AST activity was significantly higher in summer than in monsoon and winter in both sexes and the total herd. ALT and ALP activities were also generally higher in summer and lower in winter. These findings indicate that seasonal thermal load is associated with measurable changes in serum enzyme activities in Nagavali sheep. The reported values may provide useful baseline information for clinical interpretation and management of this recently recognized breed under tropical production systems.

Keywords: Alkaline phosphatase; alanine aminotransferase; aspartate aminotransferase; heat stress; Nagavali sheep; temperature-humidity index

Introduction
Sheep contribute substantially to food production, fibre, rural employment, and household income. As homeothermic animals, they maintain thermal balance through physiological and behavioural adjustments. Blood biochemical traits are useful indicators of metabolic status, but they may vary with breed, sex, age, nutrition, health, reproductive status, season, diet, and management [1, 2, 3, 4]. Among climatic variables, ambient temperature and humidity strongly influence livestock productivity and welfare. Increasing thermal load can alter feed intake, water balance, respiration, metabolism, production, and reproduction [5, 6].
The Nagavali sheep, also known as Vizianagaram or North Coastal sheep, is a recently recognized breed from the north coastal region of Andhra Pradesh. Breed-specific reference information on seasonal serum enzyme activity remains limited. The present study therefore aimed to assess seasonal changes in serum AST, ALT, and ALP activities in rams, ewes, and the total herd, and to relate these changes to the prevailing THI.

Materials and methods
Study location and animals
The investigation was conducted at the Livestock Research Station, College of Veterinary Science, Garividi, Sri Venkateswara Veterinary University, Vizianagaram district, Andhra Pradesh, India (18.28946° N, 83.55393° E). One hundred apparently healthy Nagavali sheep aged one year or older and maintained under comparable management were selected. The study population comprised 50 ewes and 50 rams. Sampling was conducted in May, August, and December, representing summer, monsoon, and winter, respectively.

Ethical approval
Animal sampling was performed after approval by IAEC College of veterinary science, Tirupati. under approval number [281/go/ReBi/S/2000/CPCSEA/CVSc/TPTY/010/] and all procedures followed applicable institutional animal-welfare requirements.

Temperature-humidity index

Dry- and wet-bulb temperatures, minimum and maximum temperatures, and relative humidity were recorded daily at 13:00 h using a digital thermo-hygrometer. THI was calculated as THI = 0.72(Tdb + Twb) + 40.6, where Tdb is dry-bulb temperature and Twb is wet-bulb temperature (°C) [7].

Blood collection and serum preparation

Approximately 5 mL of blood was collected aseptically from the jugular vein of each sheep between 07:30 and 08:30 h in each season. For serum separation, 2.5 mL was transferred to a clot-activator vacutainer. Samples were labelled, transported to the laboratory on ice, allowed to clot for 2 h, and centrifuged at 5000 rpm for 5 min. Serum was transferred to labelled microcentrifuge tubes and stored at -20 °C until analysis.

Serum enzyme assays

Serum AST, ALT, and ALP activities were measured using commercial Erba diagnostic kits and a Thermo Scientific Multiskan spectrophotometer according to the manufacturer's instructions. AST and ALT were determined using kinetic methods, and enzyme activities were expressed as IU/L [21].

Statistical analysis

Seasonal variation in serum AST, ALT and ALP activities was analysed separately for rams, ewes and the total herd using one-way ANOVA. Significant differences among seasonal means were identified using Duncan's multiple range post-hoc test. Statistical significance was accepted at $P \leq 0.05$.

Results

Seasonal temperature-humidity index

The mean THI was highest in summer (80.02 ± 0.43), followed by monsoon (70.51 ± 0.41) and winter (67.89 ± 0.27) (Figure 1). Based on the thresholds cited in the source manuscript, summer conditions represented moderate heat stress, whereas monsoon and winter conditions were comparatively comfortable.

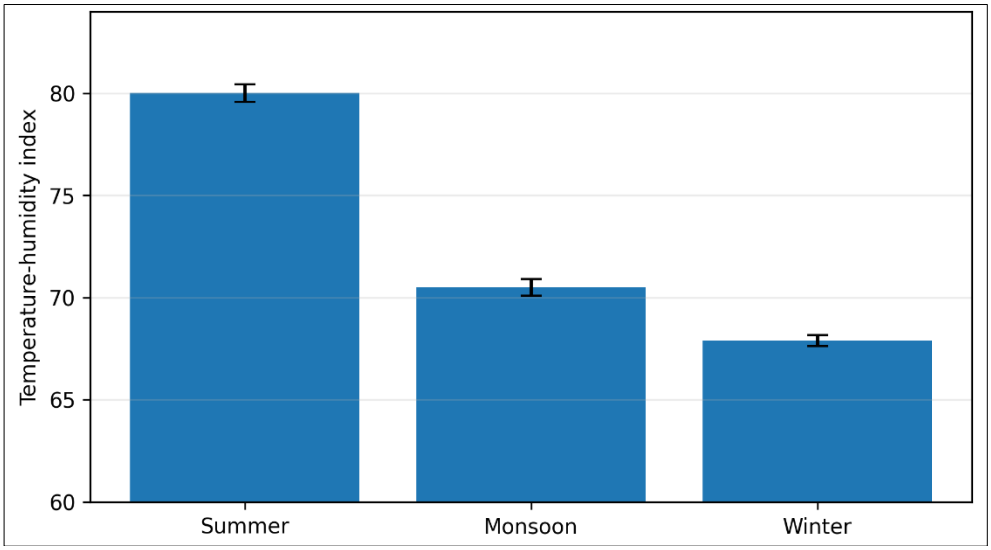

Fig 1: Seasonal variation in temperature-humidity index (mean ± SE) at the study location

Serum enzyme activities

Season significantly affected serum AST activity in rams, ewes, and the total herd (Table 1). AST was highest in summer, intermediate in monsoon, and lowest in winter. ALT activity was lower in winter than in summer in rams and the total herd; in ewes, winter values were lower than both summer and monsoon values. ALP activity was also higher in summer and monsoon than in winter.

Table 1: Seasonal serum AST, ALT, and ALP activities in Nagavali sheep (mean ± SE)

Enzyme and season	Rams	Ewes	Total herd
Aspartate aminotransferase, IU/L			
Summer	195.61 ± 3.95^{a}	202.00 ± 4.92^{a}	198.80 ± 3.16^{a}
Monsoon	171.66 ± 4.01^{b}	169.61 ± 3.63^{b}	170.64 ± 2.69^{b}
Winter	94.44 ± 2.62^{c}	92.07 ± 2.79^{c}	93.26 ± 1.91^{c}
Overall	153.90 ± 4.09	154.56 ± 4.39	154.23 ± 2.99
Alanine aminotransferase, IU/L			
Summer	56.38 ± 1.23^{a}	52.91 ± 1.46^{a}	54.65 ± 0.96^{a}
Monsoon	52.49 ± 1.37^{ab}	53.05 ± 1.32^{a}	52.77 ± 0.95^{a}
Winter	48.32 ± 1.14^{b}	45.88 ± 1.34^{b}	47.10 ± 0.89^{b}
Overall	52.40 ± 0.77	50.62 ± 0.83	51.51 ± 0.57
Alkaline phosphatase, IU/L			
Summer	86.28 ± 1.23^{a}	98.21 ± 1.46^{a}	92.25 ± 0.95^{a}
Monsoon	78.52 ± 1.37^{a}	85.15 ± 1.32^{a}	81.84 ± 0.95^{a}
Winter	58.35 ± 1.14^{b}	65.58 ± 1.34^{b}	61.97 ± 0.88^{b}
Overall	74.38 ± 0.77	82.98 ± 0.83	78.68 ± 0.57

Within each enzyme and column, means with different superscript letters differ significantly at $P \leq 0.05$. AST, aspartate aminotransferase; ALT, alanine aminotransferase; ALP, alkaline phosphatase.

Discussion

The higher summer THI observed in this study indicates greater thermal load during that season. Similar seasonal increases in THI have been reported in small ruminants [9]. Sheep and goats are generally considered relatively tolerant of harsh climates, but prolonged exposure to high THI can still affect production, physiology, and welfare [10, 6].

Using the classification cited in the source manuscript, THI values of 72 or below indicate no heat stress, 73-77 mild stress, 78-89 moderate stress, and values above 90 severe stress [7]. Accordingly, the summer THI of approximately 80 represented moderate heat stress, whereas monsoon and winter conditions were below or near the comfort threshold [8].

Aspartate aminotransferase

Serum AST activity was markedly higher during summer in rams, ewes, and the total herd. This pattern agrees with reports of elevated AST during hot seasons in indigenous sheep and other small ruminants [5, 3]. Increased AST may reflect altered hepatic and muscular metabolism, greater membrane permeability, or tissue strain under thermal stress. Temperature-related acceleration of enzymatic reactions has also been proposed [12].

Alanine aminotransferase

ALT activity was generally higher in summer and monsoon and lower in winter. Comparable seasonal increases have been documented in sheep and goats [5, 15]. Because ALT participates in conversion of alanine to pyruvate, increased activity during summer may be associated with enhanced gluconeogenic demand or altered liver function under heat stress [14, 13].

Alkaline phosphatase

ALP activity was higher during summer and monsoon than during winter. ALP originates mainly from liver, bone, and other tissues, and seasonal variation may reflect changes in hepatic function, bone turnover, growth, and energy metabolism. Similar thermal effects on enzymatic activity have been described in goats and sheep [17, 18, 19, 20].

The present results should be interpreted with caution because the study involved a single flock and three discrete seasonal sampling periods. Potential effects of age, physiological state, nutrition, and repeated sampling were not modelled separately. Nevertheless, the findings provide preliminary breed-specific information that may assist clinical interpretation and seasonal management of Nagavali sheep.

Conclusion

Summer conditions were associated with the highest THI and increased serum AST, ALT, and ALP activities in Nagavali sheep. Winter values were consistently lower, particularly for AST and ALP. Seasonal thermal load should therefore be considered when interpreting serum enzyme profiles in this breed. The values reported here may serve as preliminary baseline data, although confirmation in larger, multi-flock studies using repeated-measures or mixed-effects models is warranted.

Acknowledgement

The authors acknowledge the staff of the Livestock Research Station, the in-charge of the Livestock Farm Complex, and the Associate Dean, College of Veterinary Science, Sri Venkateswara Veterinary University, Garividi, for permission, facilities, and support.

Funding

No external funding was received for this study. The research was carried out as part of the Master's degree programme of the first author using the facilities available at Sri Venkateswara Veterinary University.

Conflict of interest

The authors declare that they have no conflict of interest.

Author contributions

G.N.S.: conceptualization, investigation, supervision, and manuscript preparation; S.S.: sample processing and data collection; K.A.K. and K.P.: methodology, interpretation, and critical review; T.S.R.: technical support and critical review. All authors read and approved the final manuscript.

Data availability

The data generated during this study are available from the corresponding author on reasonable request.

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