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Assessment of physiological and leucocyte responses to pulse chunni-incorporated complete diets in Nellore brown ram lambs

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Abstract

Thirty-two growing Nellore brown ram lambs (96.30 \pm 2.70 days of age and 13.27 \pm 0.03 kg body weight) were used to evaluate the effect of pulse chunni inclusion in sorghum straw based complete diets on growth performance, feed conversion efficiency and carcass characteristics. Lambs were fed finishing diets and assigned randomly to one of three dietary treatments for a period of 120 days under intensive system at Sheep unit, Livestock Research Station, Mamnoor, Warangal. Treatment diets were complete diet without any pulse chunni-control (T_1 ; N=8), 15% red gram chunni included complete diet-RGCC (T_2 ; N=8), 15% green gram chunni included complete diet-GGCC (T_3 ; N=8) and 15% Bengal gram chunni included complete diet-BGCC (T_4 ; N=8). Among physiological parameters studied, test diets did not alter rectal temperature, respiration rate and pulse rate of ram lambs in the morning as well as evening and these parameters were within the normal range, while period had highly significant (p<0.01) effect on overall values of physiological parameters in ram lambs. The lymphocyte (%) and monocyte (%) values throughout the experiment were statistically non-significant among four test groups. At the end of the experiment, significant (p<0.01) difference observed in neutrophil (%) and at the middle of the study, significant (p<0.01) difference observed in eosinophil (%) among four groups.

Keywords: Nellore ram lambs, pulse chunni based complete diets, physiological parameters, leucocytes

Introduction

Nutrition plays a pivotal role in determining the expression of an animal's genetic potential, particularly in terms of growth, health, and immune competence. Key nutrients such as protein, energy, vitamins, and minerals contribute significantly to maintaining and enhancing immune function. Existing literature suggests that, beyond basic nutrition, the composition and supplementation of complete diets have a substantial impact on the overall health and immunity of farm animals. Physiological responses in lambs such as respiration rate, pulse rate, and rectal temperature are influenced not only by diet but also by environmental factors and climatic conditions. Body temperature, in particular, serves as a direct indicator of an animal's capacity to regulate heat gain and heat loss under varying environmental stresses. Regular monitoring of these vital signs is essential for early detection of physiological or health abnormalities, enabling timely intervention and management.

In this context, the present study was undertaken to evaluate the influence of pulse chunnibased complete diets on the health status of Nellore Brown ram lambs, with a focus on key physiological parameters (rectal temperature, respiration rate, and pulse rate) and leukocyte indices as indicators of immune responsiveness.

Materials and Methods

Selection and Distribution of Experimental Ram Lambs

Thirty-two growing Nellore Brown ram lambs (96.30±2.70 days of age; 13.27±0.03 kg body weight) were selected from the Sheep Unit, Livestock Research Station, Mamnoor, for a 120-day growth trial.

The lambs were randomly allotted to four dietary treatment groups, with eight lambs per group, following a Completely Randomized Design (CRD). Care was taken to ensure uniformity in initial body weights among the groups.

Housing and Management

All lambs were maintained under hygienic and well-ventilated conditions in pens measuring 24' × 10', each provided with an adjacent open area of the same dimensions for daytime movement. The animals were not allowed to graze during the study period. Deworming was carried out at the beginning of the experiment and again after two months using Ivermectin (for external parasites) and Fenbendazole (for internal parasites). All lambs were vaccinated against Peste des Petits Ruminants (PPR) seven days after the first deworming. Proper sanitation and regular cleaning of the sheds were maintained. Each lamb was ear-tagged for accurate

identification and data recording.

Feeding and Watering

Four complete rations T_1 , T_2 , T_3 , and T_4 (Table 1) were randomly assigned to the respective groups for the 120-day feeding trial. The rations included:

- T₁: Control ration
- T2: RGCC-based complete ration
- T₃: GGCC-based complete ration
- T4: BGCC-based complete ration

Feeds were offered ad libitum twice daily at 9:00 AM and 3:00 PM. The feed offered and the refusals (recorded the following morning) were weighed using an electronic digital balance, allowing accurate measurement of individual daily feed intake. Clean, fresh drinking water was provided to all animals continuously in buckets throughout the experiment.

Table 1: Ingredient composition of experimental rations (kg/100 kg) used for growth study in Nellore ram lambs

		Cor	nplete diets	
Ingredients	T ₁ (Without any chunni: Control)	T ₂ (With Red gram chunni: RGCC)	T ₃ (With Green gram chunni: GGCC)	T ₄ (With Bengal gram chunni: BGCC)
Sorghum straw	50.0	50.0	50.0	50.0
Red gram chunni	-	7.5	-	-
Green gram chunni	-	-	7.5	-
Bengal gram chunni	-	-	-	7.5
Maize	13.0	14.0	15.0	13.0
Ground nut cake	21.5	20.5	19.5	21.5
Wheat bran	7.5	-	-	-
Molasses	5.0	5.0	5.0	5.0
Mineral mixture	2.0	2.0	2.0	2.0
Salt	1.0	1.0	1.0	1.0
Total*	100.0	100.0	100.0	100.0

^{*} Vitamin A supplement was added @ 10 g/100 kg complete diet.

Table 2: Chemical composition of experimental complete diets (On% DMB) fed to growing Nellore ram lambs

		Compl	ete diets*		
Nutrient	T ₁ (Without any chunni: Control)	T ₂ (With Red gram chunni: RGCC)	T ₃ (With Green gram chunni: GGCC)	T4 (With Bengal gram chunni: BGCC)	
		Proximate constituents	8		
Moisture	1.71	3.34	2.39	3.16	
Dry matter	98.29	96.66	97.61	96.84	
Organic matter	87.88	88.11	88.08	89.17	
Crude protein	13.79	13.68	13.88	13.78	
Ether extract	2.98	3.24	3.19	2.95	
Crude fibre	22.86	20.56	21.03	25.02	
Nitrogen free extract	48.94	48.63	49.99	47.04	
Total ash	12.12	11.89	11.92	10.83	
Calcium	1.57	1.61	1.13	1.15	
Phosphorus	0.83	0.66	0.70	0.80	
		Cell wall constituents	1		
Neutral detergent fibre	63.86	60.11	60.85	62.55	
Acid detergent fibre	33.82	34.12	33.15	35.95	
Cellulose	25.43	23.95	23.27	26.86	
Acid detergent lignin	6.25	7.33	5.85	4.83	
Silica	2.70	2.93	2.99	3.71	
Hemi cellulose	30.04	25.99	27.71	26.60	

^{*}Each value is an average of duplicate analysis

Physiological Parameters

The physiological responses rectal temperature (RT), respiration rate (RR), and pulse rate (PR) were recorded to assess the influence of pulse-chunni-based complete diets

(Harikrishna *et al.*, 2010) ^[5]. Measurements were taken in both the morning (7:30-8:30 AM) and evening (4:30-5:30 PM) at fortnightly intervals throughout the four-month experimental period.

Estimation Procedures Rectal Temperature (RT)

RT was measured using a clinical thermometer inserted into the rectum for one minute. Care was taken to place the thermometer obliquely in contact with the rectal wall. Temperature was recorded in °F.

Respiration Rate (RR)

RR was determined by counting the number of breaths per minute, felt by placing the back of the hand near the animal's nostrils. The values were expressed as breaths/min.

Pulse Rate (PR)

PR was measured by palpating the femoral artery on the inner side of the hind limb. Pulse rate was recorded as beats/min.

Blood Collection and Haematological Analysis

Whole blood samples (5 mL) were collected from the jugular vein of all experimental lambs at three intervals:

- At the beginning of the trial.
- At the mid-point, and.
- At the end of the experiment.

Blood was collected in the morning before feeding, under aseptic conditions, using hypodermic needles and sterile tubes containing EDTA as anticoagulant. Samples were immediately stored in ice packs at approximately 5 °C and transported to the laboratory for analysis. All laboratory procedures were performed using clean glassware and

analytical-grade reagents. For Differential Leucocyte Count (DLC), samples were analyzed using an automated haemoanalyser (ABX MICROS-60).

Statistical Analysis

Statistical analysis of the data was carried out according to the procedures suggested by Snedecor and Cochran (1994) [17]. Least-square Analysis of variance was used to test the significance of various treatments and the difference between treatment means was tested for significance by Duncan's Multiple Range and F-Test (Duncan, 1955) [4].

Results and Discussion Physiological Parameters

The effect of feeding sorghum straw-based complete diets containing different pulse chunnies on physiological parameters rectal temperature (RT), pulse rate (PR) and Respiration Rate (RR) of growing Nellore ram lambs was assessed. The fortnightly mean (±SE) values recorded during morning and evening are presented in Tables 3-8.

Rectal Temperature

The fortnightly rectal temperature values (°F) of lambs in all dietary groups (T_1 - T_4) recorded in the morning and evening showed normal physiological ranges. Across the experimental period, the morning RT ranged between approximately 101.11 to 103.03 °F, while the evening RT ranged between 101.31 to 103.38 °F among treatments.

Table 3: Fortnightly morning rectal temperature of Nellore ram lambs fed experimental complete diets

		Initial Rectal	Fortnightly Morning Rectal Temperature [#] (Mean ± SE; °F)									
Complete diets	N	Temperature (°F)	1	2	3	4**	5	6	7**	8 (Final Rectal Temperature)		
T ₁ (Without any chunni: Control)	8	102.20±0.05	102.42±0.07	102.65±0.10	102.62±0.16	102.22a±0.18	102.70±0.16	102.66±0.10	101.11 ^a ±0.40	102.95±0.09		
T ₂ (With Red gram chunni: RGCC)	8	102.30±0.03	102.65±0.13	102.5±0.09	102.86±0.18	102.88°±0.11	102.76±0.11	102.67±0.09	102.83 ^{ab} ±0.19	102.93±0.17		
T ₃ (With Green gram chunni: GGCC)	8	102.23±0.03	102.57±0.10	102.76±0.12	102.56±0.06	102.22 ^{ab} ±0.20	102.63±0.09	102.61±0.11	103.03 ^b ±0.09	102.56±0.11		
T ₄ (With Bengal gram chunni: BGCC)	8	102.22±0.04	102.67±0.13	102.72±0.14	102.88±0.17	102.81 ^b ±0.17	102.78±0.09	102.61±0.02	102.78 ^{ab} ±0.10	102.87±0.08		
SEM		0.021	0.057	0.059	0.078	0.099	0.058	0.044	0.074	0.064		
P-Value		0.416	0.421	0.545	0.352	0.011	0.816	0.942	0.007	0.105		

[#]Each value is an average of eight observations; N=Number of animals in each treatment; P=Probability Value; SEM: Standard Error Mean abMeans with different superscripts column wise differ significantly **(p<0.01)

Table 4: Fortnightly evening rectal temperature of Nellore ram lambs fed experimental complete diets

		Initial Rectal												
Complete diets	N	Temperature (°F)	1	2	3	4	5	6	7	8 ^{NS} (Final Rectal Temperature)				
T ₁ (Without any chunni: Control)	8	102.83±0.09	102.95±0.14	103.35±0.22	103.18±0.17	103.13±0.07	101.37±0.41	103.31±0.05	103.17±0.06	103.10±0.06				
T ₂ (With Red gram chunni: RGCC)	8	102.86±0.09	103.00±0.07	103.11±0.13	103.17±0.11	103.18±0.07	101.48±0.36	103.02±0.08	103.23±0.06	103.20±0.08				
T ₃ (With Green gram chunni: GGCC)	8	102.83±0.16	103.01±0.08	103.20±0.09	103.16±0.09	103.10±0.10	101.31±0.28	103.10±0.11	103.38±0.07	103.26±0.05				
T ₄ (With Bengal gram chunni: BGCC)	8	102.78±0.05	103.02±0.11	103.32±0.08	103.18±0.05	103.13±0.09	103.13±0.08	103.21±0.03	103.11±0.10	103.32±0.05				
SEM		0.051	0.053	0.072	0.056	0.044	0.050	0.043	0.041	0.035				
P-Value		0.967	0.965	0.545	0.998	0.922	0.318	0.086	0.102	0.131				

^{*}Each value is an average of eight observations; N=Number of animals in each treatment; P=Probability Value

Statistical analysis revealed no significant (p>0.05) differences in RT among the four treatment groups. The inclusion of different pulse chunnies did not influence the thermal response of lambs either in the morning or evening,

except for slight variations during the 4th and 7th fortnights in morning readings. These results align with earlier findings in calves and sheep (Rokde, 2007; Harikrishna *et al.*, 2010; Rangamma, 2020; Likhitha, 2022; Pandu *et al.*, 2021) [16, 5, 15,

NSNon-significant, SEM: Standard Error Mean

^{8, 11]}, and also corroborate reports that flooring or environmental differences did not significantly alter RT (Kulkarni *et al.*, 2000; Thiruvenkadan *et al.*, 2009; Divate, 2014; Deshmukh, 2017; Mohit *et al.*, 2019; Ramachandran *et al.*, 2020) ^[7, 20, 3, 2, 9, 14]. The RT values remained within the normal physiological range for sheep (Harikrishna *et al.*, 2010) ^[5].

Pulse Rate

The fortnightly pulse rate values (beats/min) recorded in the morning and evening across treatments T_1 - T_4 ranged from 44.13 to 55.87 in the morning and from 61.12 to 65.37 in the evening. Statistical analysis indicated no significant (p>0.05) differences among dietary treatments. Lambs fed diets with or

without pulse chunnies showed similar pulse rates throughout the study. Pulse rate consistently remained higher during the evening compared to morning observations.

These findings agree with previous studies in ram lambs and sheep (Kulkarni *et al.*, 2000; Thiruvenkadan *et al.*, 2009; Divate, 2014; Deshmukh, 2017; Mohit *et al.*, 2019c; Ramachandran *et al.*, 2020; Pandu *et al.*, 2021; Likhitha, 2022) ^[7, 20, 3, 7, 2, 8, 11, 9, 14]. However, contrary to the present results, Rokde (2007) ^[16], Harikrishna *et al.* (2010a) ^[5] and Rahman *et al.* (2013) ^[12] reported significant differences in PR under yeast-based diets or different management systems. Pulse rate values observed in this investigation fall within the normal physiological limits for sheep (Rangamma, 2020) ^[15].

Table 5: Fortnightly morning pulse rate of Nellore ram lambs fed experimental complete diets

		Initial Pulse Rate (Beats/min)		Fortnightly Morning Pulse Rate# (Mean ± SE; Beats/min)									
Complete diets	N		1	2	3	4	5	6	7	8 ^{NS} (Final Pulse Rate)			
T ₁ (Without any chunni: Control)	8	46.62±0.32	44.75±0.31	45.38±0.26	46.87±0.35	54.75±0.31	55.75±0.16	55.37±0.18	55.87±0.29	55.50±0.26			
T ₂ (With Red gram chunni: RGCC)	8	46.50±0.26	44.13±0.29	45.13±0.22	46.62±0.26	54.13±0.22	55.12±0.22	55.25±0.16	55.75±0.16	55.12±0.29			
T ₃ (With Green gram chunni: GGCC)	8	46.50±0.18	44.38±0.18	45.37±0.46	46.37±0.26	54.25±0.36	55.13±0.29	55.13±0.22	55.87±0.29	55.75±0.31			
T ₄ (With Bengal gram chunni: BGCC)	8	46.75±0.25	44.25±0.25	45.25±0.25	46.75±0.25	54.87±0.35	55.63±0.26	55.38±0.26	55.63±0.32	55.38±0.26			
SEM		0.126	0.133	0.150	0.139	0.162	0.126	0.103	0.133	0.142			
P-Value		0.889	0.391	0.932	0.643	0.283	0.162	0.813	0.904	0.488			

^{*}Each value is an average of eight observations; N=Number of animals in each treatment; P=Probability Value

Table 6: Fortnightly evening pulse rate of Nellore ram lambs fed experimental complete diets

		Initial Pulse Rate- (Beats/min)		F	ortnightly I	Evening Puls	se Rate# (Me	ean ± SE; Bo	eats/min)	
Complete diets	N		1	2	3	4	5	6	7	8 ^{NS} (Final Pulse Rate)
T ₁ (Without any chunni: Control)	8	61.75±0.25	61.25±0.25	62.75±0.16	62.50±0.18	62.87±0.35	65.25±0.25	64.37±0.32	64.62±0.37	64.25±0.52
T ₂ (With Red gram chunni: RGCC)	8	61.63±0.18	61.38±0.18	62.00±0.26	62.12±0.12	62.50±0.26	65.00±0.26	64.25±0.25	64.50±0.18	63.87±0.51
T ₃ (With Green gram chunni: GGCC)	8	61.75±0.16	61.87±0.12	62.63±0.26	62.38±0.18	62.63±0.26	65.13±0.23	64.50±1.18	64.75±0.25	64.50±0.32
T ₄ (With Bengal gram chunni: BGCC)	8	61.38±0.18	61.75±0.31	62.75±0.31	62.25±0.16	62.75±0.25	65.37±0.18	64.12±0.29	64.87±0.29	65.00±0.26
SEM		0.098	0.119	0.135	0.083	0.138	0.114	0.130	0.138	0.215
P-Value		0.504	0.192	0.145	0.437	0.811	0.701	0.783	0.811	0.315

^{*}Each value is an average of eight observations; N=Number of animals in each treatment; P=Probability Value

Table 7: Fortnightly morning respiration rate of Nellore ram lambs fed experimental complete diets

		Initial		Fortnightly Morning Respiration Rate# (Mean ± SE; No./min)									
Complete diets	N	Respiration Rate (No./min)	1	2	3	4	5	6	7	8 ^{NS} (Final Respiration Rate)			
T ₁ (Without any chunni: Control)	8	16.38±0.18	16.25±0.16	16.38±0.18	16.12±0.16	16.25±0.16	17.25±0.16	17.62±0.18	16.85±1.29	16.75±0.16			
T ₂ (With Red gram chunni: RGCC)	8	16.37±0.18	16.25±0.16	16.12±0.12	16.25±0.18	16.12±0.12	17.12±0.12	17.62±0.18	16.50±1.12	16.38±0.18			
T ₃ (With Green gram chunni: GGCC)	8	16.25±0.16	16.13±0.12	16.25±0.16	16.37±0.18	16.38±0.18	17.38±0.18	17.37±0.18	16.62±0.86	16.63±0.18			
T ₄ (With Bengal gram chunni: BGCC)	8	16.50±0.19	16.25±0.16	16.37±0.18	16.13±0.22	16.5±0.19	17.37±0.18	17.75±0.16	16.75±0.16	16.75±0.16			
SEM		0.087	0.074	0.081	0.098	0.083	0.081	0.088	0.083	0.087			
P-Value		0.809	0.920	0.669	0.788	0.437	0.669	0.516	0.437	0.391			

^{*}Each value is an average of eight observations; N=Number of animals in each treatment; P=Probability Value

^{NS}Non-Significant, SEM: Standard Error Mean

NSNon-significant, SEM: Standard Error Mean

NSNon-significant, SEM: Standard Error Mean

Respiration Rate

Morning respiration rates (No/min) ranged between 16.12 and 17.75, while evening values ranged between 17.12 and 18.25 across all treatments during the experimental period.

Statistical analysis indicated non-significant (*p*>0.05) differences in respiration rate among the treatment groups. Feeding pulse chunnies did not exert any influence on RR. As with other physiological parameters, RR was consistently higher during the evening, likely due to elevated ambient temperatures. These findings concur with earlier reports in lambs and sheep (Kulkarni *et al.*, 2000; Thiruvenkadan *et al.*, 2009; Divate, 2014; Deshmukh, 2017; Mohit *et al.*, 2019; Tharuntej *et al.*, 2020; Pandu *et al.*, 2021; Rangamma, 2020; Likhitha, 2022) [7, 20, 3, 2, 8, 11, 9]. However, Harikrishna *et al.* 2010a [7, 5, 15] and Rokde 2007 [16] observed significant increases in RR under yeast-supplemented diets. The RR

values recorded in this study fall within the normal physiological standards for sheep.

Effect of Period (Morning vs Evening) on Physiological Parameters

The overall effect of time of recording was significant (p<0.05) for rectal temperature, respiration rate and pulse rate. All three parameters were consistently higher during the evening than morning. This trend is well supported by earlier reports (Harikrishna *et al.*, 2010a; Rangamma, 2020; Likhitha, 2022) [5, 15, 8], indicating that higher afternoon ambient temperatures elevate the physiological responses of sheep.

The elevated evening readings are attributed to greater heat load during the day, indicating normal thermoregulatory adjustments in the animals.

Table 8: Fortnightly evening respiration rate of Nellore ram lambs fed experimental complete diets

Complete diets	N	Initial Respiration Rate (No/min)		For	tnightly Ev	ening Respi	ration Rate#	(Mean ± SE;	No./min)	
			1	2	3	4	5	6	7	8 ^{NS} (Final Respiration Rate)
T ₁ (Without any chunni: Control)	8	17.25±0.25	17.12±0.12	17.62±0.18	17.63±0.18	17.50±0.19	17.75±0.16	18.25±0.16	17.75±0.16	17.88±0.12
T ₂ (With Red gram chunni: RGCC)	8	17.38±0.26	17.50±0.18	17.50±0.19	17.88±0.29	17.50±0.18	17.62±0.18	18.12±0.12	17.50±0.18	17.37±0.18
T ₃ (With Green gram chunni: GGCC)			17.25±0.16	17.25±0.16	17.25±0.16	17.63±0.18	17.87±0.22	18.13±0.22	17.63±0.37	17.50±0.18
T ₄ (With Bengal gram chunni: BGCC)	8	17.50±0.18	17.37±0.18	17.12±0.12	17.38±0.18	17.37±0.18	17.50±0.18	18.12±0.13	17.62±0.18	17.62±0.18
SEM		0.109	0.083	0.089	0.109	0.089	0.095	0.079	0.117	0.088
P-Value		0.837	0.437	0.153	0.189	0.824	0.555	0.933	0.911	0.225

^{*}Each value is an average of eight observations; N=Number of animals in each treatment; P=Probability Value

Leucocyte Indices

The mean values of lymphocytes, neutrophils, eosinophils, monocytes and basophils recorded at the beginning, middle

and end of the study are presented in Table 9. Overall analysis revealed no significant (p>0.05) differences among treatments for most leucocyte indices, with a few exceptions

Table 9: Leucocyte indices of Nellore ram lambs fed experimental complete diets (Mean \pm SE)

			Leucocyte Indices# (Mean ± SE)													
Complete	N	Lyn	nphocytes	(%)	Ne	eutrophils ((%)	Eo	sinophils (%)	Monocytes (%)					
diets		Initial	Middle	At the End	Initial	Middle	At the End**	Initial	Middle**	At the End	Initial	Middle	At the End			
T ₁ (Control)	8	63.81±2.15	60.21±2.50	63.02±2.61	40.38±0.69	42.20±0.93	$42.56^{b}\pm0.34$	0.02 ± 0.01	0.16 ^a ±0.07	0.28±0.19	0.08 ± 0.05	1.40±0.08	0.68 ± 0.15			
T ₂ (RGCC)	8	65.47±2.84	62.30±2.93	63.08±2.02	39.66±0.99	41.98±0.91	44.08 ^b ±0.52	0.07 ± 0.06	0.15 ^a ±0.07	0.10 ± 0.08	0.16 ± 0.14	1.10±0.13	0.56 ± 0.21			
T ₃ (GGCC)	8	64.18±2.65	62.43±2.96	62.78±1.96	40.05±1.75	42.45±0.92	43.83 ^b ±0.40	0.08 ± 0.06	0.13a±0.07	0.18 ± 0.08	0.09 ± 0.06	1.14±0.07	0.88±0.17			
T ₄ (BGCC)	8	60.63±2.82	60.96±1.99	64.80±0.89	40.31±1.75	39.36±0.73	40.23°±0.96	0.53±0.38	$0.50^{b}\pm0.11$	0.40±0.17	2.10±1.26	1.31±0.06	0.77±0.17			
SEM		1.293	1.259	0.949	0.553	0.475	0.399	0.100	0.050	0.071	0.340	0.050	0.088			
P-Value		0.606	0.917	0.882	0.970	0.068	0.000	0.238	0.01	0.500	0.085	0.099	0.624			

^{*}Each value is an average of eight observations; N=Number of animals in each treatment; P=Probability Value; SEM: Standard Error Mean ab Means with different superscripts column wise differ significantly **(p<0.01)

Lymphocytes

Lymphocyte percentages ranged between 60.63-65.47% across treatments and time periods. All values were within the normal reference range. Findings agree with Swensen and Ithane (1984) and Mohit *et al.* (2019c) ^[9] but were higher than those reported by Nagalakshmi & Dhanalakshmi (2015) ^[10], Rahman *et al.* (2018) ^[13], Beigh *et al.* (2018) ^[1] and Pandu *et al.* (2021) ^[11].

Neutrophils

Neutrophil percentages ranged between 40.23-44.08%. Although overall treatment effects were non-significant, a significantly (p<0.01) lower neutrophil count was observed in T_4 at the end of the experiment compared to T_1 - T_3 . These values agree with Swensen and Ithane (1984), Mohit *et al.*

(2019) [9] and Pandu *et al.* (2021) [11], but were higher than Rahman *et al.* (2018) [13] and Beigh *et al.* (2018) [1].

Eosinophils

Eosinophil percentages ranged from 0.10-0.40%, with a significantly (p<0.01) higher value in T₄ during the middle of the experiment. These low values indicate absence of parasitic infestation. Values were similar to Swensen and Ithane (1984) and Mohit *et al.* (2019) ^[9], but lower than Rahman *et al.* (2018) ^[13], Beigh *et al.* (2018) ^[11] and Pandu *et al.* (2021) ^[11].

Monocytes

Monocyte values (0.56-0.88%) showed no significant differences among treatments at the end of the experiment. These values align with Swensen and Ithane (1984) but were

^{NS}Non-significant, SEM: Standard Error Mean

lower compared to Rahman *et al.* (2018) [13], Mohit *et al.* (2019) [9] and Pandu *et al.* (2021) [11].

Basophils

Basophils were absent in all samples across treatments and time periods. According to Jain (1993), presence of basophils indicates disease; thus, the zero-basophil count confirms the animals remained physiologically healthy throughout the experiment.

Summary

- Dietary inclusion of different pulse chunnies did not significantly affect rectal temperature, pulse rate, respiration rate or major leucocyte indices.
- All physiological and haematological parameters remained within normal physiological ranges.
- Evening measurements of RT, PR and RR were consistently higher due to ambient environmental conditions.
- Minor variations in neutrophils and eosinophils were observed but did not indicate compromised health.
- Zero basophil counts throughout the study confirm overall good health and absence of disease.

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Conflict of Interest

Not available

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