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Comparative Evaluation of Stress and Haematological Parameters of Meat Goats Grazing Southern-Pine Silvopasture and Open Pasture During Summer

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ABSTRACT

Milder climatic conditions in silvopasture may impact the health of grazing animals differently than open pasture. However, little is known about the potential differences in stress and other health indicators among grazing animals due to the pasture system. We hypothesized that meat goats grazing in silvopasture would be less stressed and exhibit better hematological parameters than those grazing in open pasture during summer. The objective of the study was to evaluate stress and hematological parameters of Kiko does raised in silvopasture and open pasture. Twenty Kiko does (age: 2-5 years, weight: 45.9 ± SE 1.31 kg) were split into silvopasture and open pasture groups and rotationally stocked in each system from June to September 2025 for 70 days. Hair samples were collected on the first day, every four weeks, and on the last day of the study, and analyzed for cortisol to determine animal stress. Blood samples were collected on the first day, every two weeks, and on the last day of the study and analyzed for 15 hematological parameters. The silvopasture group had a 21.21% lower cortisol level ($p < 0.05$) than the same parameters of the open-pasture group on Day 70 of the study, despite similar results in prior observations. Does reared in silvopasture had lower neutrophil (15.94%, $p < 0.01$), monocyte (20%, $p < 0.05$), and eosinophil (31.25%, $p < 0.001$) vs. the open-pasture group. Results suggest that meat goats grazing in silvopasture during summer can be less stressed than in open pasture.

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1. Introduction

Goats (*Capra hircus* L.) are predominantly raised in open pastures, pastures without trees, in the southeastern United States (Karki, 2020). Grazing animals perform well when the surrounding temperature falls within their comfort zone, which is the range of air temperatures at which they can maintain a constant body temperature (Brown–Brandl, 2018). When temperature or the temperature-humidity index exceeds the upper or lower critical thresholds, animals experience stress (Karki and Goodman, 2009). For example, rearing animals in open pastures during summer may lead to heat stress (Karki and Goodman, 2009; Pent and Fike, 2018; Pent et al., 2018, 2019). Fonseca et al. (2016) reported that goats can tolerate a thermoneutrality zone of 20–30 °C. Over the past decade, the average summer air temperature in the southeastern US has ranged between 27–29 °C, with the daytime average being 33–34 °C. Maximum temperature reaches 38 °C, particularly between 12:00–16:00 h (NOAA, 2025), exposing open pasture-reared animals to an uncomfortable environment that affects their health (Thomsen et al., 2023). USDA (n.d.) estimated 10% economic loss in the livestock industry each year in the southeastern US due to heat stress-induced animal mortality and reduced productivity. Therefore, the provision of shade to protect animals from heat stress is crucial (Collier et al., 1982; Danso et al., 2024).

Allowing animals to graze in a silvopasture system during summer has the potential to reduce heat stress, as it creates milder climatic conditions compared with open pasture (Karki & Goodman, 2012; Karki & Goodman, 2014; Poudel et al., 2024). Silvopasture is an agroforestry system in which selected trees and forages are grown in a single management unit and grazing animals are integrated to utilize the understory forage (Greene et al., 2023; Karki & Goodman, 2012, 2014; Poudel et al., 2022; Thomsen et al., 2023). A previous study has reported lower total solar radiation (14-58%, $p < 0.05$) and lower air temperature (1.03%, $p < 0.0001$) in loblolly pine (*Pinus taeda* L.) silvopasture compared to open pasture during summer in Chipley, Florida, United States (Karki and Goodman, 2012). The milder climatic condition in the silvopasture system enhanced the grazing duration and even distribution of cattle compared to open pasture (Karki &

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Goodman, 2009), indicating less stress and better animal comfort in the former system. However, such literature for goats grazing silvopasture and open pasture is rare.

Animals under stress release cortisol, a stress hormone produced in the adrenal cortex (Pragst & Balikova, 2006; Russell et al., 2012; Romero et al., 2014). Under stressful conditions, cortisol is first released into the bloodstream, where it remains for a short time, a few minutes to hours (Riek et al., 2019). Riek et al. (2019) reported that the blood cortisol levels increased after 10 minutes of adrenocorticotropic hormone (ACTH) administration, peaked at 70 minutes, and returned to baseline concentration levels after 170 minutes in cattle. Because of its brief presence in the bloodstream, blood cortisol is not a suitable indicator for long-term stress evaluation. Circulating cortisol in blood diffuses into growing hair through passive diffusion from blood capillaries to hair follicles (Cook, 2012; Heimbürge et al., 2020) located 0.3–0.4 cm below the skin surface (Heimbürge et al., 2018; Lepselter & Elman, 2004).

As hair grows, approximately 1.0–1.5 cm per month in goats, it takes around 6–12 days to reach the growing hair follicle above the skin surface, after which the hair can be collected and used for cortisol extraction (Endo et al., 2017). Del Rosario González-De-La-Vara et al. (2011) reported elevated hair cortisol in cattle on day 14 following ACTH application on day 1, demonstrating that hair cortisol can be used to assess stress over a retrospective period of a couple of weeks. Stalder and Kirschbaum (2012) reported that hair samples collected as close to the skin surface as possible can be used to determine cortisol concentration, an indicator of stress level in animals over the previous two weeks to a couple of months. Gupta et al. (2023) reported that approximately 1 cm of hair from the skin surface represents the amount of cortisol produced during the previous month in animals. Once accumulated in hair, cortisol may remain stable throughout its life (Stalder & Kirschbaum, 2012). However, Wester et al. (2016) reported that environmental conditions, such as sunlight exposure, decrease the hair cortisol concentration. Cortisol concentration in hair samples decreased by 54% ($p < 0.001$) when samples were exposed to 40 h of natural sunlight (Wester et al., 2016). Once collected, hair samples can be stored for at least 5 years without cortisol degradation if properly stored and protected from sunlight (Berger et al., 2024). Moreover, hair sampling is painless and minimally affected by handling, further enhancing its suitability and accuracy for stress assessment (Heimbürge et al., 2020; Nejad et al., 2013; Nejad et al., 2022; Pragst & Balikova, 2006). Poudel et al. (2022) reported lower hair cortisol in ewes grazed in black walnut (Juglans nigra L.) silvopasture by 51.9% ($p < 0.0001$) in 2020 and by 109.6% ($p < 0.0001$) in 2021 compared to grazing in open pasture during summer in Blacksburg, Virginia, USA. However, no information is available regarding the potential of the southern pine (*Pinus* spp.) silvopasture in reducing hair cortisol in goats.

Few studies have reported the impact of stress on various hematological parameters of different animal species. Dhabhar et al. (1994) found a higher neutrophil count (9%) in stressed rats subjected to an hour-long restraint compared to the control group ($p < 0.05$). Similarly, Lamsal et al. (2024) found 15% higher RBC in does kept outdoor versus indoor during winter ($p < 0.05$). However, prolonged stress compromises the hematological cell producing system, leading to a reduction in the release of hematological cells compared to those animals reared within their comfort zone. Lamsal et al. (2025) found lower platelet volume in ewes by 21% ($p < 0.01$) kept outdoor vs. indoors during winter in the southeast US. Lopes et al. (2022) found lower lymphocyte ($p < 0.05$) count in heat-stressed Nellore heifers reared in open pasture versus. silvopasture during summer in Brazil.

Although mild stress activates the immune system, prolonged stress in animals cause excessive cortisol release, which acts as an immunosuppressant, resulting in reduced production of immune cells such as neutrophils, basophils, and eosinophils (Diaz-Jimenez et al., 2021; Obeagu, 2025; Van De Wouw et al., 2021). These cells are responsible for an immediate response to sudden injury and infections and function as the first line of defense against invading pathogens. Suppression of their production due to stress increases the susceptibility of animals to pathogens, jeopardizing their health (Obeagu, 2025). However, studies are limited regarding the role of silvopasture systems in maintaining healthy hematological parameters, including immune cells, and reducing stress in meat goats during summer. We hypothesized that goats reared in silvopasture would be less stressed and exhibit better hematological parameters than those reared in open pasture. The objective of the study was to evaluate hair cortisol and hematological parameters of Kiko does reared in silvopasture and open pasture.

2. Methods

2.1. Study Site

The study was conducted over 10 weeks using three silvopasture plots (1-acre each; 32°44'39"N - 32°44'42.8"N, Longitude 85°73'09"W - 85°74'27"W) located at the Atkins Agroforestry Research and demonstration sites, and five open pasture plots (0.5-acre each; Latitude 32°44’02”N - 32°44’07.6”N, Longitude 85°74’03”W - 85°74’09”W) located in the facility of Caprine Research and Education Unit at Tuskegee University. Silvopasture plots consisted of longleaf pine (*Pinus palustris* Mill.) and loblolly pine (*Pinus taeda* L.) (210 trees/ha, 12 m tall, 0.8 m DBH, where DBH refers to tree diameter measured at breast height, 1.37 m above the ground surface). Both study sites were dominated by bahiagrass (*Paspalum notatum* Fluegge) and MaxQ tall fescue (*Festuca arundinacea* Schreb. Holub). Each plot consisted of water lines and a water trough, one mineral feeder, and two mobile shelters (port-a-hut metal shelters).

2.2. Study Animals

Twenty Kiko does aged 2–5 years were used in the study. Based on the initial live weight, FAMACHA score, body condition score (BCS), and EPG (number of gastrointestinal nematode egg per gram of animal feces), animals were divided into two groups: silvopasture and open pasture. Animals were rotated among plots within each site based on 50% forage depletion, assessed by visual observation and forage height measurement with a grazing stick. Animals with a FAMACHA score of 3 or above and an EPG greater than 1000 were dewormed with cydectin (0.4 mg/kg body weight) (Kaplan, 2014) one day after each data collection. Animals in both groups had free access to clean drinking water, mineral mix (Purina goat minerals, Arden Hills, MN, USA), and shelters throughout the study. Daily animal care and management were performed in accordance with the animal care and use protocol approved by the Tuskegee University Animal Care and Use Committee (TUACUC), protocol number R08_2024-06.

2.3. Data Collection

2.3.1. Weather Parameters

Temperature and humidity at the silvopasture site were measured on-site using a Govee Smart Thermo-Hygrometer. For the open pasture site, secondary weather data collected for Tuskegee University by the nearby Visual Crossing weather station (https://www.visualcrossing.com/weather-

history/Tuskegee/metric/2025-06-23/2025-09-4), located 4.83 km from the study site, were used. An LI-191 light quantum sensor (Li-COR Biosciences, NE, USA) was used to measure photosynthetically active radiation (PAR) at both sites. Ten wooden pegs were installed along the diagonal line of each silvopasture plot, and five wooden pegs at each open pasture plot (Fig. 1). On the measurement day, the LI-191 sensor was placed at the base of each peg, perpendicular to the diagonal, at each measurement point in every plot. PAR data were collected between 12:00 and 14:00 h on clear and sunny days, one or two days after moving animals out of each plot. At each collection point, data were logged for one minute, with the LI-1500 set to automatically record readings every six seconds. The LI-191 sensor integrated PAR over its 1 m duration, providing a representative measurement.

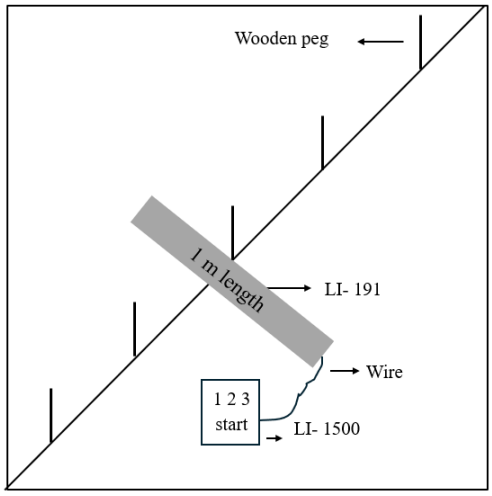

Figure 1. *Schematic diagram showing measurement points (represented by wooden pegs) along the diagonal of a study plot. LI-191 light sensor was placed perpendicular to the diagonal at each measurement point to record photosynthetically active radiation, June - September 2025, Tuskegee University, Tuskegee, Alabama, USA.*

2.3.2. Forage Sampling

Ten random forage samples per plot were collected from both research sites using a 0.25 m² quadrat one day before introducing animals into the plots at each rotation. While setting the quadrat for sample collection, forages rooted inside the quadrats were included in, and those rooted outside were excluded from the quadrat before clipping. Then forages collected inside the quadrat were clipped to 10 cm above the ground surface using scissors. Collected samples were kept in individual pre-labeled paper bags, brought to the Agroforestry and Grazing Land Ecology Lab (AG Lab) located at the Caprine Research and Education Unit (CREU) facility, and oven-dried at 60 °C for 72 hours. Dried samples were weighed to determine the productivity, then ground to pass through 2 mm mesh. Ground samples were analyzed for crude protein (CP), acid detergent fiber (ADF), and neutral detergent fiber (NDF) using the Fourier transform infrared (FTIR) Spectroscopy. Total digestible nutrient (TDN) was estimated based on NDF values: TDN (%) = (105.2-0.667*NDF%)*0.88.

2.3.3. Hair Sample Collection and Analysis

Hair samples were taken from a 10-cm^2 area of the flank region of all animals on Days 1 (the day study was initiated), 28, and 70 (last day) of the study. Hair samples were collected from the same spot on each sampling period using the shave-reshave method. Hair from the designated area was clipped as close to the skin as possible using a battery-operated clipper. Collected hair samples were individually placed in clean, airtight aluminum foil, and stored in a dark place at room temperature for 1-3 months until processing for cortisol extraction. The collected hair samples were finely chopped to 2-4 mm in length using surgical scissors. Then 35 mg of chopped hair was washed with a 1.5 mL aliquot of 100% isopropanol to remove any contamination or surface-bound cortisol released by animal sweating. The samples were dried at room temperature for 72 hours. To extract cortisol from the hair samples, 1.5 mL of 100% methanol was then added to the same tube and vortexed for about 24 hours. The vortexed mixture of 750 µL of supernatant was transferred to a fresh tube and then dried. After drying, the ELISA buffer was added to the same tube with the cortisol extract. Cortisol concentration was quantified with a commercial salivary cortisol ELISA kit (Cayman Chemical, MI, USA) according to the manufacturer's instructions. Eight standards with different dilutions were prepared, and each standard was replicated twice to ensure the validation of the ELISA assay. The samples (no dilution) were pipetted into the wells followed by the addition of the cortisol-AChE tracer and the cortisol ELISA monoclonal antibody. The ELISA plate was incubated overnight (18 h) at 4 °C. The wells were washed, Ellman's reagent was added to the wells, and the wells were kept on an orbital shaker for 90 minutes at room temperature. When a yellow color developed, the intensity of color was measured at 415 nm wavelength in a microplate reader (Varioskan Lux Multimode Microplate Reader) for determining the cortisol concentration, with three replications for each sample.

2.3.4. Blood Sample Collection and Analysis

Blood samples were collected on the first day of the study prior to allocating animals to their respective sites, then fortnightly during the study period from the jugular vein using Vacutainers into 2 mL tubes containing EDTA (ethylenediaminetetraacetic acid). Immediately after collection, the tube was gently shaken a couple of times to mix the blood with EDTA and prevent clotting, then stored in a cool box until sample collection from all research animals was completed. The samples were then brought to the Lab and stored in a refrigerator until analysis, which was conducted within a couple of hours of collection. The tube was gently shaken 10–15 times using a test tube shaker, then analyzed in ProCyte Dx hematology analyzer (IDEXX Laboratories, Inc., Westbrook, ME, USA) for 15 hematological parameters.

2.4. Data Analysis

All data sets were analyzed using SAS 9.4, and hypotheses were tested at 95% confidence level. Air temperature and relative humidity data were first divided into two categories, dawn to dusk and dusk to dawn, before being analyzed. Air temperature, relative humidity, forage quality, and hematological data were analyzed using the generalized linear model (GLM) with MANOVA (multivariate analysis of variance) option to account for the correlation among the dependent variables within each data set. Photosynthetically active solar radiation and hair cortisol data were analyzed using the mixed model.

The general GLM model used to analyze various data sets is presented below:

$$Y_{(1\text{-}n)ij} = \mu + \alpha_i + (\alpha\beta)_{ij} + e_{ij}$$

MANOVA h = Animal group and the interaction of animal group and observation date. Where,

Y(1-n) ij = dependent variable from the ith group and jth observation data, μ = grand mean, αi = group effect, (αβ)ij =interaction of ith group and jth observation date, eij = error associated with the ith group and jth observation date. Y(1-n)ij for each data set is described below.

1. Weather parameters: $Y_{(1\text{-}2)ij}$ = temperature and humidity at the i^{th} research site and j^{th} observation date.
2. Forage quality: $Y_{(1\text{-}3)ij}$ = forage quality from the i^{th} research site and j^{th} observation date.
3. Hematological parameters: $Y_{(1\text{-}15)ij}$ = hematological variables of animals from the i^{th} group and j^{th} observation date.

The model used for the Mixed Procedure is presented below:

$Yij = \mu + \alpha i + (\alpha\beta)ij + eij$

Yij = dependent variable from the ith group and jth observation data, μ = grand mean, αi = group effect, (αβ)ij =interaction of ith group and jth observation date, eij = error associated with the ith group and jth observation date. Yij for each data set is described below.

1. Photosynthetically active solar radiation: Y_{ij} = photosynthetically active solar radiation from the i^{th} research site and jth observation date.
2. Hair cortisol: Y_{ij} = hair cortisol of animals from the i^{th} research group and j^{th} observation date.

3. Results

3.1. Climatic Parameters

Lower air temperature was observed in the silvopasture during dawn to dusk; however, humidity was higher during that period than in open pasture plots (Table 1). Lower PAR (74%, $p < 0.0001$) was observed in silvopasture compared to open pasture (Fig. 2).

Table 1. *Temperature and humidity in open pasture (Open) and silvopasture (Silvo), June-September 2025, Tuskegee University, Tuskegee, Alabama, USA.*

Observation days	Dawn - dusk		Dusk - dawn	
	Open	Silvo	Open	Silvo
	LSMean ± SE			
	Temperature (°C)			
01-14	28 ± 0.2	28 ± 0.2	24 ± 0.3	23 ± 0.3
14-28	28 ± 0.2^{a**}	27 ± 0.2^{b}	25 ± 0.3^{b}	27 ± 0.3^{a**}
28-42	29 ± 0.2	29 ± 0.2	25 ± 0.3	26 ± 0.3
42-56	26 ± 0.2^{a*}	25 ± 0.2	24 ± 0.3	24 ± 0.3
56-70	27 ± 0.2^{a****}	25 ± 0.2^{b}	22 ± 0.3	23 ± 0.3
	Humidity (%)			
01-14	74 ± 1.0^{b}	78 ± 1.0^{a*}	91 ± 1.4	94 ± 1.4
14-28	75 ± 0.9^{b}	82 ± 0.9^{a****}	92 ± 1.3^{a***}	85 ± 1.3^{b}
28-42	77 ± 0.9	79 ± 0.9	92 ± 1.3	90 ± 1.3
42-56	82 ± 1.0^{b}	89 ± 1.0^{a****}	95 ± 1.2	92 ± 1.2
56-70	68 ± 0.9^{b}	79 ± 0.9^{a****}	91 ± 1.1	87 ± 1.1

^{ab}LSMeans in the same row within the same period with different superscripts differ. (*p < 0.05, **p < 0.01, ***p < 0.001, ****p < 0.0001).

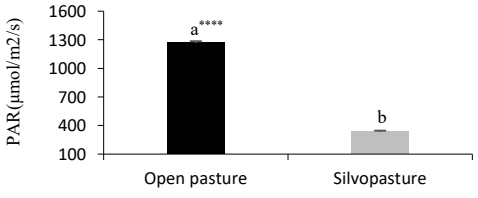

Figure 2. *Photosynthetically active radiation (PAR) (LSMean ± SE) in open pasture and silvopasture, June- September 2025, Caprine Research and Education Unit, Tuskegee University, Tuskegee, Alabama, USA (***p < 0.0001).*

3.2. Forage Quality

Higher CP was found in forages collected from silvopasture (10%) compared to open pasture (9%) (Table 2). However, NDF, ADF, and TDN were similar in the forage samples collected from both systems.

Table 2. *Quality of forages from open pasture and silvopasture, June-September 2025, Caprine Research and Education Unit, Tuskegee University, Tuskegee, Alabama, USA.*

Forage quality	Open pasture	Silvopasture
	LSMEAN ± SE	
CP†	9 ± 0.1^{b}	10 ± 0.1^{a****}
NDF	66 ± 0.5	66 ± 0.5
ADF	42 ± 0.3	41 ± 0.3
TDN	53 ± 0.2	53 ± 0.3

†CP- Crude protein; NDF - Neutral detergent fiber; ADF - Acid detergent fiber; TDN - Total digestible nutrients. ^{ab}LSMeans in the same row with different superscripts differ (****p < 0.0001).

3.3. Hair Cortisol

Both groups had similar cortisol concentrations at the start of the study (Fig. 3). Cortisol levels increased in both groups at later observation dates, and group difference occurred on Day 70, with silvopasture group showing lower concentrations (21.21%, $p < 0.05$).

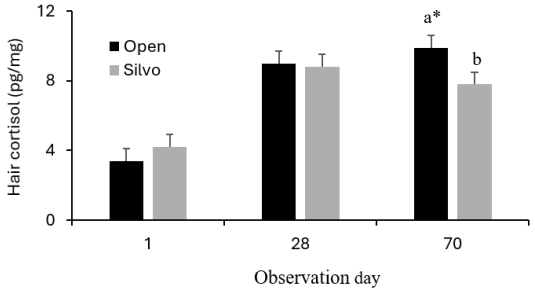

Figure 3. *Hair cortisol (LSMean ± SE) of Kiko does grazing in open pasture (Open) and silvopasture (Silvo), June-September 2025, Caprine Research and Education Unit, Tuskegee University, Tuskegee, Alabama, USA (*p < 0.05).*

3.4. Hematological Parameters

Table 3. *Hematological parameters of Kiko does grazing in open pasture (Open) and silvopasture (Silvo) (Day 1), June- September 2025, Caprine Research and Education Unit, Tuskegee University, Tuskegee, Alabama, USA.*

Hematological parameters	Open	Silvo	Normal range
	Day 1		
	LSMean ± SE		
Red blood cell (M/μL)	11 ± 0.9	12 ± 0.9	9.4 – 15.1
Hematocrit (%)	25 ± 1.9	23 ± 1.9	22.0 – 39.0
Hemoglobin (g/dL)	7 ± 0.5	7 ± 0.5	10.0 – 14.9
Mean corpuscular volume (fL)	22 ± 1.6	20 ± 1.6	14.0 – 22.3
Mean corpuscular hemoglobin (g/dl)	6 ± 0.2	6 ± 0.2	5.0 – 7.0
Mean corpuscular hemoglobin concentration (g/dl)	30 ± 1.8	32 ± 1.8	32.0 – 34.0
Reticulocytes (K/μL)	2 ± 1.2	2 ± 1.2	0.0 – 15.0
White blood cell (K/μL)	12 ± 0.9	12 ± 0.9	6.03 – 19.58
Neutrophil (K/μL)	6 ± 0.7	6 ± 0.7	1.72 – 10.61
Lymphocyte (K/μL)	3 ± 1.1	4 ± 1.1	2.68 – 11.54
Monocyte (K/μL)	0.6 ± 0.1	0.6 ± 0.1	0.06 – 0.89
Eosinophil (K/μL)	1 ± 0.2	1 ± 0.2	0.03 – 1.29
Basophil (K/μL)	0.1 ± 0.0	0.1 ± 0.0	0.00 – 0.24
Platelets (K/μL)	398 ± 29.2	478 ± 29.2	246 – 912
Mean platelet volume	8 ± 0.1	8 ± 0.1	NA

Overall, the silvopasture group had lower neutrophil (15.94%, $p < 0.01$), monocyte (20%, $p < 0.05$), and eosinophil (31.25%, $p < 0.001$) vs. the open-pasture group. However, other parameters were similar for both groups. Both groups had similar hematological parameters on the first day (Table 3). The interaction between the system and observation day occurred for reticulocytes, eosinophils, and monocytes during the study (Tables 4, 5, 6).

Table 4. *Hematological parameters of Kiko does grazing in open pasture (Open) and silvopasture (Silvo) systems (Days 14 & 28), June- September 2025, Caprine Research and Education Unit, Tuskegee University, Tuskegee, Alabama, USA.*

Hematological parameters	Open	Silvo	Open	Silvo
	Day 14		Day 28	
	LSMean ± SE			
Red blood cell (M/µL)	10 ± 0.9	11 ± 0.9	10 ± 0.9	11 ± 0.9
Hematocrit (%)	22 ± 1.9	23 ± 2.0	22 ± 1.9	27 ± 2.0
Hemoglobin (g/dL)	7 ± 0.5	7 ± 0.5	7 ± 0.5	7 ± 0.5
Mean corpuscular volume (fL)	23 ± 1.6	21 ± 1.6	24 ± 1.6	22 ± 1.6
Mean corpuscular hemoglobin (g/dl)	7 ± 0.2	7 ± 0.2	7 ± 0.2	7 ± 0.2
Mean corpuscular hemoglobin concentration (g/dl)	34 ± 1.8	34 ± 1.8	31 ± 1.8	33 ± 1.8
Reticulocytes (K/µL)	6 ± 1.2[a*]	1 ± 1.2[b]	5 ± 1.2	2 ± 1.2
White blood cell (K/µL)	15 ± 0.9	13 ± 0.9	15 ± 0.9	12 ± 0.9
Neutrophil (K/µL)	8 ± 0.7[a*]	6 ± 0.7[b]	8 ± 0.7[a*]	6 ± 0.7[b]
Lymphocyte (K/µL)	7 ± 1.1	5 ± 1.1	4 ± 1.1	5 ± 1.1
Monocyte (K/µL)	0.9 ± 0.1	0.9 ± 0.1	0.9 ± 0.1	0.7 ± 0.1
Eosinophil (K/µL)	1.9 ± 0.2	1.2 ± 0.2	1 ± 0.2	1 ± 0.2
Basophil (K/µL)	0.1 ± 0.0	0.2 ± 0.0	0 ± 0.0	0 ± 0.0
Platelets (K/µL)	457 ± 29.2	446 ± 29.2	430 ± 29.9	493 ± 29.9
Mean platelet volume	8 ± 0.1	8 ± 0.1	8 ± 0.1	8 ± 0.1

[ab]LSMeans in the same row for the same day with different superscripts differ (*$p < 0.05$). The normal range is presented in Table 3.

Table 5. *Hematological parameters of Kiko does grazing open pasture (Open) and silvopasture (Silvo) (Day 42 & 56), June- September 2025, Caprine Research and Education Unit, Tuskegee University, Tuskegee, Alabama, USA.*

Hematological parameters	Open	Silvo	Open	Silvo
	Day 42		Day 56	
	LSMean ± SE			
Red blood cell (M/µL)	11 ± 0.9	12 ± 0.9	12 ± 0.9	12 ± 0.9
Hematocrit (%)	26 ± 1.9	28 ± 1.9	27 ± 1.9	26 ± 1.9
Hemoglobin (g/dL)	8 ± 0.5	9 ± 0.5	8 ± 0.5	8 ± 0.5
Mean corpuscular volume (fL)	24 ± 1.6	22 ± 1.6	22 ± 1.6	22 ± 1.6
Mean corpuscular hemoglobin (g/dl)	7 ± 0.2	7 ± 0.2	7 ± 0.2	7 ± 0.2
Mean corpuscular hemoglobin concentration (g/dl)	29 ± 1.8	32 ± 1.8	31 ± 1.8	32 ± 1.8
Reticulocytes (K/µL)	2 ± 1.2	1 ± 1.2	1 ± 1.2	1 ± 1.2
White blood cell (K/µL)	12 ± 0.9	14 ± 0.9	12 ± 0.9	12 ± 0.9
Neutrophil (K/µL)	7 ± 0.7	6 ± 0.7	6 ± 0.7	6 ± 0.7
Lymphocyte (K/µL)	4 ± 1.1	4 ± 1.1	4 ± 1.1	5 ± 1.1
Monocyte (K/µL)	0.9 ± 0.1	0.8 ± 0.1	0.8 ± 0.1	0.6 ± 0.1
Eosinophil (K/µL)	1.9 ± 0.2[a*]	1.2 ± 0.2[b]	1.7 ± 0.2	1.1 ± 0.2
Basophil (K/µL)	0.0 ± 0.0	0.0 ± 0.0	0.1 ± 0.0	0.1 ± 0.0
Platelets (K/µL)	460 ± 29.2	513 ± 29.2	482 ± 29.2	484 ± 29.2
Mean platelet volume	8 ± 0.1	8.0 ± 0.1	7.9 ± 0.1	8 ± 0.1

[ab]LSMeans in the same row for the same day with different superscripts differ (*$p < 0.05$). The normal range is presented in Table 3.

Table 6. *Hematological parameters of Kiko does grazing open pasture (Open) and silvopasture (Silvo) (Day 70), June- September 2025, Caprine Research and Education Unit, Tuskegee University, Tuskegee, Alabama, USA.*

Hematological parameters	Open	Silvo
	Day 70	
	LSMean ± SE	
Red blood cell (M/µL)	13 ± 0.9	12 ± 0.9
Hematocrit (%)	26 ± 1.9	24 ± 1.9
Hemoglobin (g/dL)	8 ± 0.5	8 ± 0.5
Mean corpuscular volume (fL)	21 ± 1.6	20 ± 1.6
Mean corpuscular hemoglobin (g/dl)	7 ± 0.2	7 ± 0.2
Mean corpuscular hemoglobin concentration (g/dl)	34 ± 1.8	36 ± 1.8
Reticulocytes (K/µL)	1 ± 1.2	1 ± 1.2
White blood cell (K/µL)	14 ± 0.9	12 ± 0.9
Neutrophil (K/µL)	5 ± 0.7	5 ± 0.7
Lymphocyte (K/µL)	5 ± 1.1	5 ± 1.1
Monocyte (K/µL)	1.3 ± 0.1[a*]	0.7 ± 0.1[b]
Eosinophil (K/µL)	1.9 ± 0.2	1.3 ± 0.2
Basophil (K/µL)	0.1 ± 0.0	0.1 ± 0.0
Platelets (K/µL)	488 ± 29.2	463 ± 29.2
Mean platelet volume	8 ± 0.1	8 ± 0.16

[ab]LSMeans in the same row with different superscripts differ (*$p < 0.05$). The normal range is presented in Table 3.

4. Discussion

4.1. Hair Cortisol

The hypothesis that Kiko does in silvopasture would be less stressed was accepted since the study found lower hair cortisol concentration in the silvopasture group compared to the open-pasture group on Day 70 of the study. The lower cortisol concentration in goats grazing silvopasture may be due to milder microclimatic conditions compared with open pasture. The current study found 74% ($p < 0.0001$) lower photosynthetically active solar radiation and 2.44% ($p < 0.0001$) lower diurnal air temperature in silvopasture compared to open pasture, which might have created a less stressful environment for grazing animals, resulting in reduced cortisol production. Ripamonti et al. (2025) also found 20% ($p < 0.05$) lower hair cortisol in beef cattle grazing oak (*Quercus cerris* L.) forest, where the black globe humidity index (calculated using total solar radiation intensity, air temperature, and relative humidity) was limited to 75, compared to those grazing an open pasture where the index rose above 80 in June in Tuscany, Italy, during summer.

Additional studies have reported the effect of difference in microclimatic parameters on hair cortisol level in different ruminant species. Ataallahi et al. (2019) found 29.09% ($p < 0.05$) lower hair cortisol concentration in Korean native Hanwoo cattle during autumn, when the ambient temperature was 10 °C, compared to that in summer when the ambient temperature was 26 °C in Kangwon National University research farm in the Republic of Korea. Nejad et al. (2018) also observed lower hair cortisol (75%, $p < 0.0001$) in Holstein dairy cows raised under temperatures of 21-22 °C versus those raised under temperatures of 26.6-38 °C in a Daejeon feedlot facility in the Republic of Korea. Similarly, Lamsal et al. (2024) found 125% and 140% ($p < 0.01$) lower hair cortisol in does reared in indoor vs. outdoor during winter on Day 34 and Day 69, respectively. The lower hair cortisol level in indoor raised animals in this study could be due to less cold stress during winter. In another study, Lamsal et al. (2024) found hair cortisol level 280% ($p < 0.001$) lower in indoor-raised (21.6 °C) vs. outdoor-raised (19.1 °C) ewes on Day 34 of the study during winter in Tuskegee, Alabama, USA. Similarly, Agradi et al. (2023) found hair cortisol levels 98% lower ($p < 0.001$) in indoor-raised Frisa female goats kept at the thermoneutral zone (15-20 °C) compared to those raised

outdoors in pasture (24-27 °C) during summer for a month in the Rezzano valley of Italy.

4.2. Hematological Parameters

The hypothesis that hematological parameters would be better in the silvopasture group was accepted, as the study found that hematological parameters were maintained within the normal range in the silvopasture group but above the normal range in the open pasture group. The eosinophil and monocyte cell counts being within the healthy range in the silvopasture group but above the normal range in open pasture group could be due to less stressful conditions in silvopasture versus an open–pasture system, as the study found 74% ($p < 0.0001$) lower photosynthetically active solar radiation and 2.44% ($p < 0.0001$) lower diurnal air temperature in the former than in the latter site. Alam et al. (2013) reported lower neutrophil (8.21%, $p < 0.05$), eosinophil (62.16%, $p < 0.05$), and monocyte (96%, $p < 0.01$) counts in Black Bengal goats reared at lower temperatures (21.67 °C) compared with those exposed to higher temperatures (28.17 °C) for eight hours daily over 18 days. Park et al. (2021) found lower eosinophil counts (67%, $p < 0.001$) within a healthy range in dairy cattle exposed to a temperature-humidity index of 65 versus 80 during an indoor experiment in the Republic of Korea.

A lower reticulocyte count observed in the silvopasture group on Day 14 of the current study could be due to a less stressful environment compared to open pasture, since stress also triggers the production of reticulocytes (Waltz et al., 2014). During heat stress, red blood cells are allocated toward the skin to dissipate heat from the body; in turn, bone marrow produces and releases reticulocytes into the bloodstream to protect other body tissues from hypoxia (Collin et al., 2001). Waltz et al. (2014) found lower (67%, $p < 0.05$) reticulocytes in pigs reared at 24 °C versus 32 °C for 7 days. The current study did not find any difference in lymphocyte counts between the two groups. However, Lopes et al. (2022) reported contradictory findings, with higher (150%, $p < 0.05$) lymphocyte count in Nellore heifers reared in silvopasture versus open pasture during summer in Brazil. The lower proliferation of lymphocytes in the open pasture group in that study could be due to severe stress-induced immunosuppression (Obeagu, 2025), as the maximum temperature reported at the study site was 33 °C. However, temperatures for the silvopasture and open-pasture sites were not reported separately.

Another possible reason behind improved hematological parameters in the silvopasture group in the current study could be the availability of higher-quality forage, as forage samples from silvopasture had higher crude protein (CP) content (10% CP) than those in open pasture (9% CP), although forage CP content from both sites seems to fulfil the maintenance requirements of mature does (NRC, 2007). Eosinophil and monocyte counts above the normal range observed in does grazing open pasture in the current study could be due to the activation of immune cells under more severe environmental conditions combined with lower forage CP, as moderate protein restriction increases the secretion of first-line defense cells (Wojtowicz et al., 2024). Contrary to the current study, Astuti et al. (2021) reported similar eosinophil and monocyte counts in Friesian cows fed elephant grass (*Pennisetum purpureum* Schumach) containing 7% CP compared with another group fed guinea grass (*Panicum maximum* Jacq.) containing 5% CP.

Similar red blood cell (RBC), hemoglobin, and hematocrit levels in both groups in the current study could be due to the animal's body prioritizing RBC production and oxygen transport as critical functions. Tölü et al. (2025) reported similar RBC, hemoglobin, and hematocrit levels in Karalabey Merino lambs and ewes fed oats (*Avena sativa* L.) containing 22% CP compared to those fed triticale (*Triticosecale* Wittmack) containing 25% CP. Contradictorily, Astuti et al. (2021) reported higher hematocrit level but within a healthy range in Friesian cows (22%, $p < 0.05$) fed elephant grass containing 7% CP versus another group fed guinea grass containing 5% CP. In the same study, authors also found higher hemoglobin concentration (33%, $p < 0.05$) in the healthy range in cows fed elephant grass versus guinea grass.

5. Conclusions

Kiko does grazing silvopasture had lower hair cortisol concentration on Day 70, compared to their counterparts grazing open pasture (21%, $p < 0.05$). Overall, the silvopasture group had lower neutrophils (15.94%, $p < 0.01$), monocytes (20%, $p < 0.05$), and eosinophils (31.25%, $p < 0.001$) compared to the open pasture group. Monocytes and eosinophils were within the normal range in the silvopasture group; however, they were above the normal range in the open pasture group. Results indicated that silvopasture creates a milder microclimate and provides a more comfortable environment, thereby reducing stress and improving hematological parameters in grazing meat goats during summer. The findings of this study may apply to meat goats and similar species raised under similar climatic and grazing conditions comparable to those in the current study.

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