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EFFECTS OF ASCORBIC ACID SUPPLEMENTATION ON GROWTH PERFORMANCE, CARCASS CHARACTERISTICS AND MEAT QUALITY OF KACHHI SHEEP UNDER HOT CLIMATIC CONDITIONS

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Abstract

Sheep are often neglected and limited research has been done to raise them intensively and provide protection from the effects of extreme heat, like the average temperature (38 °C to 42 °C) recorded in Sindh province during summer months. This study evaluated the effects of ascorbic acid supplementation on growth performance, carcass characteristics, and meat quality of Kachhi lambs under hot climatic conditions. Fifteen male Kachhi lambs with same age and weight were randomly allocated into three groups (n=5): a control (KCC) kept intensively at 35°C to 45°C, positive control (KCPC) at 30°C to 35°C, and (KCAA) supplemented group receiving 1g/day of ascorbic acid at 35-45°C. Over 90 days, body weight, conformation (chest girth, body length, height at withers), carcass traits, and meat quality were assessed. The lambs were weighed using a weighing machine and conformation measurements were recorded using a measuring tape in centimeters. At the end of the experimental trial, the lambs were slaughtered to estimate carcass characteristics and meat quality. The results indicate that supplemented lambs showed significantly improved growth performance, carcass characteristics and meat quality attributes compared to both control groups. These results suggest that ascorbic acid supplementation enhances the productivity and meat quality of sheep under thermal stress.

Keywords: Carcass yield, Dietary interventions, Environmental management, Proximate nutrient evaluation, Sheep productivity, Thermal stress response

INTRODUCTION

Pakistan being an agricultural country has a large rural based communities and agriculture-based industry, in which livestock plays an important role in the economy and livelihood for rural farmers (1). Sheep and goats are vital for farmers, providing meat, milk, skin, wool, and fiber. In Pakistan, sheep provide income to rural farmers and have excellent export potential, contributing to foreign exchange earnings. The adaptability of its breeding systems makes sheep a significant livestock animal with a major role in supporting marginal rural communities (2). In Sindh Province, more than a dozen sheep breeds are raised, with the Kachhi sheep being one of them. The breed is originated from the Tharparkar District and neighboring desert areas, with thin-tailed, medium-sized, white body coat, and often has brown or blackish head markings, small ears, and a prominent Roman nose (3).

Climate change is a critical concern, particularly for rural populations in developing countries that rely heavily on natural resources for their livelihoods. One immediate consequence of climate change is heat stress, which directly affects livestock by reducing growth, body weight, and meat quality. Heat stress decreases protein synthesis and muscle mass, leading to negative impacts at the carcass yields (4). Sheep are commonly raised outdoors (extensive) or in partly controlled (semi-intensive) systems and their health, production and fertility are deleteriously affected by heat stress. The annual estimated profitable loss caused by environmental stress ranges from 0.1 to 4% of total earnings. The estimated range of temperature tolerance for sheep is between 12°C and 32°C, any fluctuation in temperature from that range will depreciate animal health and consequently, production (5). Sheep exposed to extreme temperatures experience reduced



growth performance, body weight, average daily gain, and total body solids, resulting in lower productivity. Reduced feed intake under heat stress leads to lower body and carcass weights, diminished longissimus muscle development, and altered fat deposition. As a result, notable changes are produced in carcass characteristics and meat quality (6-11).

Carcass characteristics are the structures of the animal carcass that are estimated after slaughtering to evaluate the quality and value for meat production. These characteristics usually include carcass weight, dressing percentage and organs weight; they provide significant information regarding yield, quality, and overall market value of the meat. However, Meat quality refers to the attributes of meat determines its value to consumers. These characteristics can be allocated into nutritional, sensory and safety characteristics, which effect consumer fulfillment and approval. In small ruminants, heat stress has been linked to decreased meat quality and production characteristics. The animals exposed to increased environmental temperatures have higher pH, reduced meat tenderness and water holding capacity. The stress before slaughter depletes glycogen levels, impacting meat quality by altering water holding capacity and electrolyte balance, resulting in higher pH and shorter shelf life (12-15). Heat stress impacts meat composition by increasing moisture loss, altering protein content, and changing fat composition. Elevated temperatures cause dehydration, leading to greater moisture loss in both live animals and meat. Prolonged heat stress disrupts protein metabolism and affects fat content due to changes in eating behavior and nutrient utilization (16). Moreover, heat stress can influence mineral absorption and retention, potentially altering the ash content of the meat due to decreased feed intake (17, 18).

To mitigate heat stress, environmental modifications such as shade and evaporative cooling are effective. Vitamins and feed additives, including ascorbic acid (Vitamin C), support immune function and protect against oxidative damage at cellular levels (19). Although ruminants can synthesize ascorbic acid from D-glucose or D-galactose, several reports suggest a decline in its tissue concentrations during various stress and disease conditions. However, beneficial effects have been observed from its supplementation on the maintenance of health and fertility in animals (20, 21). Despite the known effects of heat stress on livestock, the impact of ascorbic acid supplementation on sheep in hot climates remains understudied. Considering the above facts this study evaluates the growth performance, carcass characteristics and meat quality of Kachhi sheep supplemented with ascorbic acid under different management systems.

MATERIALS AND METHODS

SITE OF STUDY

The experiment was carried out at Department of Livestock Management, Faculty of Animal Husbandry and Veterinary Sciences Sindh Agriculture University Tandojam. Located at 25° 25' 37.85" N latitude and 68° 32' 10.28" E longitude, in a semi-arid area, and 29 meters above sea level. The average annual ambient temperature ranges from 9.4 to 42.9 °C. The study was conducted from April to June 2023.

EXPERIMENTAL ANIMALS

Fifteen male Kachhi lambs of about 3-4 months of age were purchased during April to June 2023 from local animal market near Hyderabad and surrounding districts of Sindh province.

ANIMAL MANAGEMENT, DIET AND SUPPLEMENTATION

The lambs were housed in clean and well-ventilated animal shed and had *ad libitum* access to good quality drinking water. All the lambs were provided with a basal diet 70% roughage and 30 % concentrates which contains approximately 60-70 % moisture, 15-20 % Crude protein, 25-35% Crude fiber and 3-6 % Ether Extract at animal shed. However, the group 3 (KCAA) lambs were supplemented with Ascorbic Acid (Vitamin C) @ 1g/day/lamb in addition to basal diet.

EXPERIMENTAL DESIGN

For the experimental trial, animals were kept for ninety (90) days with a fifteen-day acclimatization period for adaptation before starting the experiment. The lambs were randomly divided into 03 groups (5



each) based on their body weight. Group 1 referred to as (KCC) Kachhi control group and kept intensively at Livestock Experiment Station at an average temperature (35°C to 45°C), the group 2 was named (KCPC) Kachhi positive control group was maintained between the range of 30°C to 35°C by placing air cooler at animal shed. However, the group 3 (KCAA) Kachhi supplemented with Ascorbic Acid (Vitamin C) @ 1g/day/lamb and kept intensively at Livestock Experiment Station at an average temperature (35°C to 45°C). The environmental conditions, such as temperature and humidity were monitored with the help of Automatic Temperature Clock Humidity (HTC-1)

PARAMETERS STUDIED

GROWTH PERFORMANCE

The lambs were weighed at the beginning of the experiment using an electrical weighing machine (Yameto, China). Thereafter body weight was taken on a fortnightly basis till the end of the experiment. The body conformation measurements, such as chest girth, body length, and height at the withers, were recorded using a measuring tape in centimeters. Chest girth was measured as the circumference of the chest immediately behind the elbow. Body length was measured from the shoulder point to the tip of the pin bone. However, body height of Kachhi lambs was measured from the withers to the end of the hoof (22).

CARCASS CHARACTERISTICS

At the end of the experimental trial, the lambs were slaughtered by Muslim or Halal method. The slaughtering process involves using a well-sharpened knife and a swift incision were made across the animal's throat, cutting the trachea, esophagus, carotid arteries, and jugular veins. The purpose is to ensure the swift flow of blood out of the body and a quick loss of consciousness for the animal. After decaying skin and removing the viscera and offal, the carcass characteristics such as carcass weight, organ weight (Head, Neck, Shoulder & Thorax, Loin & flank, Legs, Liver & Lungs) were taken with the help of weighing balance as suggested by (23). The dressing percentage was computed using the formula given below.

$$\text{Dressing percentage, (\%)} = \frac{\text{Carcass Weight}}{\text{Live weight during slaughtering}} \times 100$$

MEAT QUALITY

pH Value

The pH value of meat samples was evaluated using a method described by Ockerman 1985 (24). Total 10 grams of frozen minced meat sample were taken in a glass beaker and homogenized in 90 ml of distilled water; a pH meter probe along with temperature electrode was inserted into the homogenized meat sample. The results shown on the screen of the pH meter base were noted and presumed to be the pH (concentration of hydrogen ion) in each meat sample.

Water Holding Capacity (WHC)

The water holding capacity of meat samples were determined with the help of scientific protocol established by Wardlaw and McCaskill 1973 (25). Total 8 grams of meat sample was mixed with 12 ml of NaCl (0.6 % molar solution) in a centrifuge tube. The material was then placed in a centrifuge machine and spin at 10,000 rpm for 15 minutes. After centrifugation, the uppermost liquid was then transferred into a measuring cylinder for calculation. Water holding capacity was measured by varying the amount of NaCl (0.6% molar solution) consumed with the amount of transparent liquid. The following formula was used to calculate the water holding capacity of meat samples.

$$\text{WHC (\%)} = \frac{\text{Actual Weight} - \text{Supermatant Volume}}{\text{Actual Weight}} \times 100$$

Moisture Content

Moisture content in meat samples was determined as described by (AOAC, 2000). The weight of an empty aluminum dish was taken and considered as (W^1). Then, 5 grams of meat sample was put into the same dish and weighted as (W^2). The aluminum dish was placed in a 100°C hot air oven for 4 hours to dry the



sample. The dried sample was then cooled in a desiccator for 1 hour. Finally, the weight of the aluminum dish containing the dried sample (W^3) was determined. The moisture content of meat samples was calculated using the formula given below.

Where: W^1 = Empty dish weight, W^2 = Empty dish weight + Sample and W^3 = Empty dish weight + Dried sample

Protein Content

The protein content of meat samples was determined by using Kjeldahl procedure described by AOAC 2000 (26). The Kjeldahl method is most commonly used approach for determining the nitrogen concentration of organic compounds, which is subsequently used to estimate protein content, as proteins contain approximately 16 % nitrogen. Total 2 grams of meat sample was placed in a Kjeldahl digestion flask contains 30 ml of concentrated sulfuric acid (H_2SO_4). The acid digests the sample, breaking down the organic content and releasing nitrogen in the form of ammonium sulfate. The mixture of potassium sulphate (K_2SO_4 7g) and copper sulphate ($CuSO_4$ 0.35 g) was added to facilitate the digestion process. The mixture was slowly heated until it became transparent. After digestion, the solution was allowed to cool by placing it in a distillation device. A 40 % solution of sodium hydroxide ($NaOH$) was added to the flask to convert the ammonium sulfate to ammonia. Then, in the presence of sodium hydroxide solution, 5 ml of the diluted sample was subjected to four minutes extraction was performed with a Kjeldahl distillation apparatus. The produced vapors were collected in 5ml of a solution containing 2% of boric acid with bromocresol green utilized as the indicator substance. Following this, the sample underwent titration using 0.1 N of HCl to measure the level of ammonia. The concentration of nitrogen was calculated using a specific formula based on the titration results.

$$\text{Nitrogen (\%)} = \frac{1.4 \times (V^1 - V^2)}{\text{Meat sample} \times \text{Diluted sample}} \times \text{Normality of HCL}$$

Where: V^1 = Volume of HCL used during the titration of the sample, V^2 = Volume of HCL used in titrating the blank solution. The protein content was calculated by changing the acquired nitrogen concentration by the conversion factor (CF) of 6.25.

$$\text{Protein \%} = \text{Nitrogen \%} \times \text{conversion factor (CF)}$$

Ether Extract Content

The Ethers extract (Fat content) of meat sample was determined using Soxhelt extraction unit, described by AOAC 2000 (26). The glass fiber thimble was dried in an oven and weighted with the help of an analytical balance. Following that, 2 grams of meat sample was taken and placed in the pre-weighed dried thimble. The Soxhlet apparatus was assembled, with the extraction flask on the bottom, the condenser on top and the thimble with the sample was fitted inside the Soxhlet extraction chamber. 150 ml of petroleum ether was added to a distillation flask, which was then connected to a condenser. The solvent was boiled smoothly using an electric heater. Following extraction of six hours, the distillation flask was detached, left to cool and then weighed. The ether extract content in the meat was determined using this formula:

$$\text{Ether extract (\%)} = \frac{W^2 - W^1}{W^3} \times 100$$

Where: W^1 = Weight of empty distillation flask, W^2 = Weight of the distillation flask + extracted fat and W^3 = Weight of the meat sample

Ash content

The ash content of meat samples was evaluated using the gravimetric method described by AOAC 2000 (26). The weight of an empty crucible dish was taken with a weighing balance. To ignite the sample, 5 grams of meat were placed in a pre-weighed crucible dish and heated in a muffle furnace at 550°C for 4

hours. After heating, the ashed meat was then allowed to cool in a desiccator for 1 hour and the crucible dish was weighed again. The ash content in the meat samples was determined using this formula:

$$\text{Ash(\%)} = \frac{(W^3 - W^1)}{(W^2 - W^1)} \times 100$$

Where: W^1 = Empty crucible dish weight, W^2 = Weight of the crucible dish with the meat sample before ashing and W^3 = Weight of the crucible dish with the ashed meat sample.

STATISTICAL ANALYSIS

Data were analyzed using one-way analysis of variance (JMP version 7). Before conducting the analysis, assumptions (e.g. normality, homogeneity of variances and independence of observations) were checked for all variables. The data were prepared through cleaning and addressing missing values. Differences among treatments were identified using Tukey's post-hoc test with significance thresholds at $p < 0.05$.

RESULTS AND DISCUSSION

GROWTH PERFORMANCE

The results of the study clearly showed that both environmental conditions and dietary supplementation significantly influenced growth performance in Kachhi lambs. The significant differences were observed among all groups. The Lambs belonging to KCAA group were maintained under intensive conditions with temperatures of 35–45°C and supplemented with ascorbic acid (1 g/day/lamb) showed significantly ($p < 0.01$) higher values for final body weight and improved body conformation traits such as chest girth, body length and height ($p < 0.05$). In comparison, growth performance was lessened in lambs of the KCPC and KCC groups. The growth performance of KCPC group, kept at a moderate temperature of 30 to 35°C without ascorbic acid were slower than those in KCAA group due to the absence of antioxidant support. Moreover, growth performance was significantly attenuated in the KCC group due to heat stress and without any supplementation. This emphasizes the importance of environmental management in combination with nutritional supplementation for enhancing lamb growth performance (Table I). The better growth performance recorded in the KCAA lambs can be ascribed to the role of potent antioxidant that is essential for ameliorating oxidative stress during heat load. Free radicals are released in larger quantities inside the animal's body, overpowering its naturally protective guards. It happens most commonly when heat stress or high temperatures. The supplementation of ascorbic acid helps to neutralize these free radicals by donating an electron and converting them into less reactive molecules, thereby preventing cellular damage which leads less oxidative stress and preventing tissue damage.

In addition, ascorbic acid recycles other antioxidants such as vitamin E by reducing their oxidized forms back into active states which to improve an overall antioxidant defense system. It also serves a cofactor to enzymes in the production of collagen which is necessary for structural integrity and repair from oxidative damage. Furthermore, ascorbic acid may impact cellular signaling pathways through alteration of activities from antioxidant enzymes and inhibition such pro inflammatory processes via nuclear factor-kappa B (NF-κB) manipulation. Together, these help to shield cells against oxidation and ensure the integrity of cellular function (27). This view agrees with previous work done by (28), who stated that in some specific environmental & physiological conditions, the level of ascorbic acid synthesized by the animal may not be enough to meet its need. The antioxidant supplementation thus has great beneficial effect against stress and averts tissue damage. Present findings are consistent with (29) the higher temperatures that lambs are exposed to promoted higher rates of heat stress which in turn affect physiological and metabolic processes such as decreased feed intake, energy directed to thermoregulation instead of feeding and depressed nutrient absorption as well. These variables collectively hinder growth and development, resulting in reduced body measurements (chest girth, body length and height). Several studies corroborate the positive effects of ascorbic acid supplementation on growth performance under heat stress. For instance (30-34) reported that lambs supplemented with ascorbic acid exhibited improved growth performance such

as body weight, average daily gain and conformation and alleviated the adverse effects of heat stress in lambs during the summer.

Table I. Body weight and body conformation (chest girth, body length & body height) of Kachhi lambs

Parameter (Kg)	KCC	KCPC	KCAA	SE	p-Value
Initial body weight (x)	11.60 ± 1.55	12.00 ± 2.15	11.40 ± 1.78	0.825	0.873
Final body weight (y)	15.60 ± 1.91 ^b	17.70 ± 1.68 ^{ab}	19.60 ± 1.67 ^a	0.787	0.012
Total weight gain (y-x)	4.00 ± 1.17 ^b	5.70 ± 0.57 ^{ab}	8.20 ± 0.12 ^a	0.344	0.01
Initial chest girth (x)	53.60 ± 3.28	54.00 ± 2.91	53.60 ± 3.36	1.428	0.974
Final chest girth (y)	58.80 ± 2.38 ^b	60.80 ± 1.92 ^{ab}	63.60 ± 1.81 ^a	0.920	0.001
Total girth gain (y-x)	5.20 ± 2.16 ^b	6.80 ± 1.30 ^b	10.0 ± 1.87 ^a	0.812	0.004
Initial body length (x)	43.40 ± 1.516	44.40 ± 3.36	43.60 ± 2.40	1.137	0.808
Final body length (y)	48.60 ± 1.67 ^b	51.60 ± 2.88 ^{ab}	53.60 ± 3.28 ^a	1.208	0.038
Total length gain (y-x)	5.20 ± 0.83 ^c	7.20 ± 0.83 ^b	10.0 ± 1.41 ^a	0.476	0.001
Initial body height (x)	46.20 ± 2.38	46.80 ± 3.56	46.00 ± 3.08	1.363	0.911
Final body height (y)	50.60 ± 1.14 ^b	53.20 ± 2.48 ^{ab}	54.60 ± 1.81 ^a	0.848	0.018
Total height gain (y-x)	4.40 ± 2.07 ^b	6.40 ± 1.67 ^{ab}	8.60 ± 1.67 ^a	0.812	0.011

*KCC – Kachhi Control; KCPC – Kachhi Positive Control; KCAA – Kachhi Ascorbic Acid; SE –Standard Error, a,b Values within a row with different superscripts are significantly different at $p < 0.05$

CARCASS CHARACTERISTICS

After slaughter on the 90th day of the experimental trial, statistically significant differences ($P < 0.05$) were observed in carcass characteristics between the groups. The results are presented in (Table II). The findings indicate that KCAA lambs, which were supplemented with ascorbic acid and exposed to high environmental temperatures (35 to 45°C), had significantly higher pre-slaughter, carcass and organs weight compared to KCPC lambs without supplementation and kept at 30 to 35°C. Emphasizing that under heat stress conditions, ascorbic acid is an effective growth promoter and carcass quality enhancer which is important in ensuring health and productivity of the lambs in hot climatic conditions. In contrast, it was found out that KCC lambs when subjected to high temperatures without providing ascorbic acid supplements showed the lowest carcass weight, underscoring the negative impact of heat stress on growth and meat production when antioxidants are not provided. There was, however, no statistical difference across the groups for dressing percentage ($P > 0.05$), the trend towards a higher dressing percentage in KCAA lambs suggests potential economic advantages, as higher dressing percentages are typically associated with increased meat yield. These results highlight the importance of ascorbic acid supplementation in improving both the health outcomes and economic value of lambs exposed to high environmental temperatures. This is due to the beneficial effects of ascorbic acid such as antioxidant activity can be attributed to the neutralization of free radicals that arise from heat stress.

During high temperatures, the feed intake of animals usually declines as a thermo regulatory mechanism which results in lower subcutaneously and intramuscularly deposition (35). The findings of this study are consistent with those reported by (30, 36,37,38) who found that antioxidants like ascorbic acid can improve carcass characteristics and dressing percentages in lambs and prevent potential economic losses in the livestock industry due to reduced carcass yield. The findings of this study are further supported by Abd-El-Monem *et al.*, (2008) who observed that ascorbic acid supplementation enhanced the final growth margin, hot carcass weight, and organ weights (such as head, liver, and lungs) in growing lambs (39). However, the results of this study contrast with those of Rana *et al.*, (2014), who reported no significant effect ($P > 0.05$) of heat stress on the carcass weight of sheep (6). The discrepancy between these findings may be attributed to differences in sheep breeds and study designs. Their study involved varying durations of thermal stress exposure and indigenous Bangladeshi sheep breed which could explain the differences in outcomes. The ability of different sheep breeds to cope with the duration and severity of heat stress likely plays a role in these variations. The carcass characteristics of Kachhi lambs can be improved with ascorbic acid supplementation leading to provide economic benefits to the farmers. It helps to produce lambs with higher meat yields by improving pre-slaughter weight, carcass weight, and organ weight and increased yield

directly converts into higher market value because carcass weight is a contributing factor in pricing. Furthermore, lambs with superior carcass characteristics are expected to command better prices in the market due to their high quality and increased meat yield. This improved market value can improve the cost-effectiveness of farming operations, as farmers are capable to sell their products at best prices. Moreover, supplementation with ascorbic acid can reduce the need for additional management practices by alleviating the negative effects of heat stress and growth-related issues. This will also reduce the overall management cost.

Table II. Carcass characteristics of Kachhi lambs

Carcass Characteristics (Kg)	KCC	KCPC	KCAA	SE	p-Value
Pre- slaughter weight	15.60 ± 1.91 ^b	17.70±1.68 ^{ab}	19.60± 1.67 ^a	0.787	0.012
Carcass weight	6.96± 0.86 ^b	8.00 ± 1.36 ^{ab}	9.26 ± 1.52 ^a	0.573	0.045
Dressing percentage (%)	44.64 ± 1.58 ^a	44.96 ± 3.73 ^a	47.0 ± 3.93 ^a	1.460	0.484
Weight of head	0.94 ± 0.12 ^b	1.07 ± 0.14 ^{ab}	1.25 ± 0.12 ^a	0.058	0.010
Weight of neck, shoulder & thorax	2.68 ± 0.55 ^b	3.26 ± 0.58 ^{ab}	3.64 ± 0.42 ^a	0.236	0.041
Weight of loin & flank	2.79 ± 0.44 ^b	3.18 ± 0.41 ^{ab}	3.57 ± 0.27 ^a	0.172	0.024
Weight of legs	0.72 ± 0.21 ^b	0.96 ± 0.31 ^{ab}	1.24 ± 0.30 ^a	0.126	0.039
Weight of liver	0.39 ± 0.05 ^b	0.44 ± 0.050 ^{ab}	0.50 ± 0.06 ^a	0.024	0.017
Weight of lungs	0.19 ± 0.04 ^b	0.23 ± 0.05 ^{ab}	0.28 ± 0.04 ^a	0.022	0.042

KCC – Kachhi Control; KCPC – Kachhi Positive Control; KCAA – Kachhi Ascorbic Acid; SE – Standard Error, a,b Values within a row with different superscripts are significantly different at $p < 0.05$

MEAT QUALITY

Stresses like heat stress and pre-slaughtering stress reduce meat quality traits by depleting glycolysis, needed for meat maturation under anaerobic circumstances. Changes in meat quality are mainly associated with the fluctuations in glycogen level as during stress condition its amount decreases due to contraction glucose loss. This has deleterious effects on meat quality causing shorter shelf life (12-15). Supplementation with ascorbic acid could be beneficial as it has anti-stress properties that could be employed to improve meat quality and welfare of animal (40, 41). This study assessed the meat quality of Kachhi lambs focusing on pH value, water holding capacity (WHC), moisture, protein, ether extract and ash content under hot environmental conditions supplemented with ascorbic acid.

pH VALUE

The important aspect of meat quality is meat pH because it affects the capacity of muscle proteins to bind with water and subsequent juiciness and tenderness of meat. Through the help of pH value, the acidification of meat is measured, and it provides useful information regarding the prospective quality of meat (42). The results are presented in (Fig. 1 Panel A). Significant differences in pH value were observed among the groups ($p < 0.02$). The results indicates that KCPC and KCAA lambs had a pH of (5.64 ± 0.11 ; 5.6 ± 0.15). In contrast, KCC lambs, exhibited the highest pH value (5.9 ± 0.18). The elevated pH in KCC lambs indicating that heat stress can deplete muscle glycogen, leading to higher pH values, which negatively affect meat quality by reducing juiciness and tenderness (43, 44). The current findings are in line with (16) they observed that long-term heat stress produced by seasonally high temperature dramatically elevates the pH of muscles in sheep and goats. Likewise, Kadim *et al.*, (2007) found that the meat pH of Somali and Merino sheep was higher (5.77) during hot season than (5.60) in cold season (45). The current findings contradict with the findings of Rana *et al.*, (2014), who found that thermal stress have non-significant influence ($p > 0.05$) on meat pH. The sex, age and breed of sheep could be the factors contributing to difference. Their study was conducted on indigenous Bangladeshi sheep breed involved varying durations of thermal stress exposure which could explain the differences in outcomes.

WATER HOLDING CAPACITY (WHC)

Water holding capacity is a key quality attribute, influences the meat's ability to retain moisture during processing, cooking, and storage. Significant differences ($p < 0.01$) in WHC were found among the groups. KCAA lambs, supplemented with ascorbic acid, had the highest WHC ($50.6 \pm 1.14\%$), followed by KCPC lambs ($49.8 \pm 0.83\%$). However, KCC lambs had the lowest WHC ($48 \pm 1.58\%$). The results are shown



in (Fig. 1 Panel B). The lower WHC noticed in KCC lambs may be because the denaturation of proteins by heat stress interfered with the water binding ability of meat. Protein denaturation is caused by exposure to high temperatures, as they are associated with the WHC of the meat and any kind of damage to the protein leads to reduce the ability of meat to bind water (46). These findings are in agreement with several other studies reporting protection of proteins against heat damage through supplementation with ascorbic acid, hence improving WHC. Ascorbic acid as an antioxidant that minimizes oxidative stress, which can result in protein damage. It stabilizes collagen, the protein responsible for maintaining firmness in meat by preventing its breakdown under heat stress. Also, ascorbic acid will help maintain the integrity and functionality of the muscle proteins responsible for water retention. By reducing protein degradation and protecting these proteins, ascorbic acid helps improve the meat's ability to hold water and making meat more tender and juicier (41, 47). The current findings contradict with Zimmerman *et al.*, (2013), who found that the stress did not affect the WHC of meat (48). This disparity could be attributed to the difference in the level of stress tolerance in Hemsin and of sheep breeds.

MOISTURE CONTENT

Moisture content is an important determinant of meat quality. Significant differences were observed among the groups ($p < 0.02$). Fig. 1 Panel C showed highest moisture content ($73.6 \pm 1.14\%$) in KCPC lambs, maintained at 30 to 35°C, followed by KCAA lambs ($72.2 \pm 2.04\%$) supplemented with ascorbic acid. Supplementation improves meat moisture content by reducing oxidative stress, improving collagen synthesis, stabilizing pH, and maintaining the integrity of muscle cell membrane. These effects jointly lead to enhanced water retention and reduced drip loss, resulting in higher moisture content and improved meat quality (12). While KCC lambs, had the lowest ($70 \pm 1.87\%$) moisture content. The reduced moisture content in KCC lambs may be attributed to the negative effects of heat stress on WHC and resulting in drier meat with shorter shelf life. This is in line with Farouk & Swan (1998) and Offer *et al.*, (1989) which showed that moisture content is directly affected by environmental stressors that affect muscle water retention (49, 50). High temperature causes muscle fibre shrinkage resulting to moisture loss and reduced meat quality. Additionally, Lawrie & Ledward (2006,) highlighted that lower moisture content is often associated with reduced tenderness and palatability, further underscoring the importance of maintaining optimal environmental conditions and dietary supplementation to preserve moisture content in meat (51).

PROTEIN CONTENT

Protein content varied significantly among the groups ($p < 0.01$). KCAA lambs, supplemented with ascorbic acid, showed the highest protein content ($20.8 \pm 1.30\%$), followed by KCPC lambs ($19.2 \pm 1.92\%$). However, KCC lambs had the lowest protein content ($16.8 \pm 2.28\%$) See Fig. 1 Panel D. The higher protein content in KCAA lambs may be due to the protective effect of ascorbic acid against heat stress, preserving muscle protein integrity. In contrast, the lower protein content in KCC lambs can be attributed to heat-induced protein denaturation. The results are in line with Koohmaraie *et al.*, (2003), they reported that the significant protein accretion occurs probably due to hyperplasia (increase in cell number), hypertrophy (increase in cell size) and a decrease in protein degradation while the protein synthesis levels remain the same (52). The results are further supported by Rana *et al.*, (2014) who found that the group that was not exposed to heat showed significantly ($P < 0.05$) higher crude protein content compared to the eight hours heat exposure group (6).

ETHER EXTRACT CONTENT

Ether extract content, representing fat content, also differed significantly among the groups ($p < 0.02$). KCAA lambs had the highest fat content ($2.82 \pm 0.37\%$), followed by KCPC lambs ($2.52 \pm 0.37\%$). However, KCC lambs, had the lowest fat content ($2.14 \pm 0.23\%$) as shown in Fig. 1 Panel E. The higher fat content in KCAA lambs could be linked to the antioxidative properties of ascorbic acid, which may enhance lipid deposition. On the other hand, the lower fat content in KCC lambs might result from metabolic alterations

Panel A: pH Value

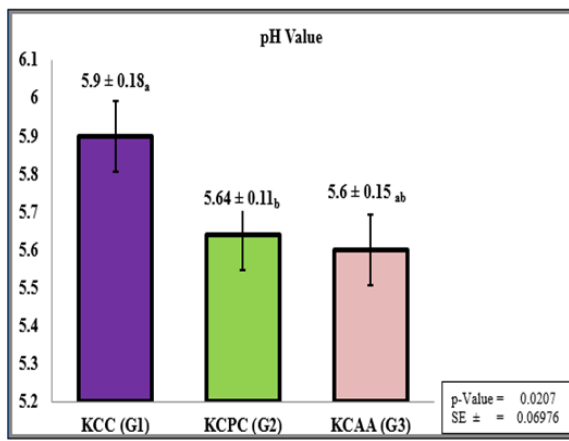


Figure-1 Panel: A Graphical presentation of pH values of meat. The bars of the graphs represent the groups KCC – Kachhi Control; KCPC – Kachhi Positive Control; KCAA – Kachhi Ascorbic Acid

Panel B: Water Holding Capacity

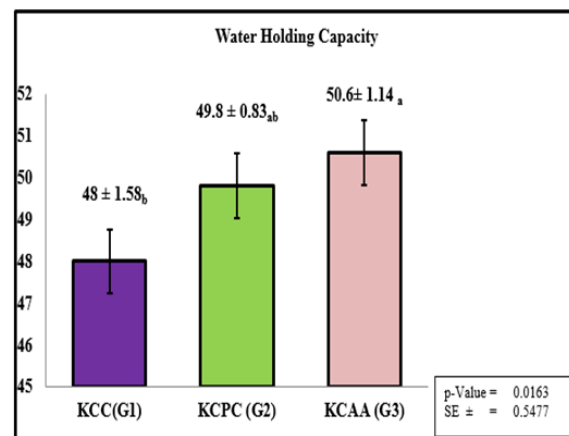


Figure-1 Panel: B Graphical presentation of Water Holding Capacity of meat. The bars of the graphs represent the groups KCC – Kachhi Control; KCPC – Kachhi Positive Control; KCAA – Kachhi Ascorbic Acid

Panel C: Moisture content

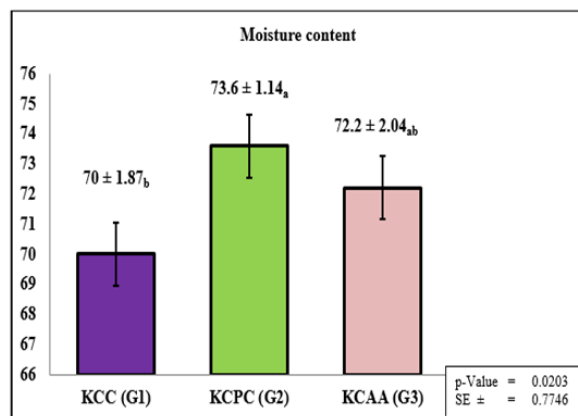


Figure-1 Panel: C Graphical presentation of moisture content of meat. The bars of the graphs represent the groups KCC – Kachhi Control; KCPC – Kachhi Positive Control; KCAA – Kachhi Ascorbic Acid

Panel D: Protein content

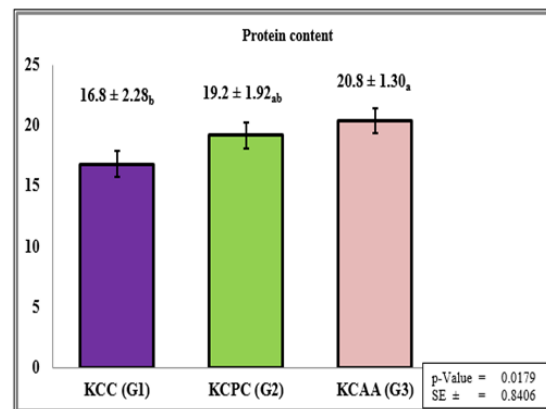


Figure-1 Panel: D Graphical presentation of Protein content of meat. The bars of the graphs represent the groups KCC – Kachhi Control; KCPC – Kachhi Positive Control; KCAA – Kachhi Ascorbic Acid

Panel E: Either Extract (Fat) content

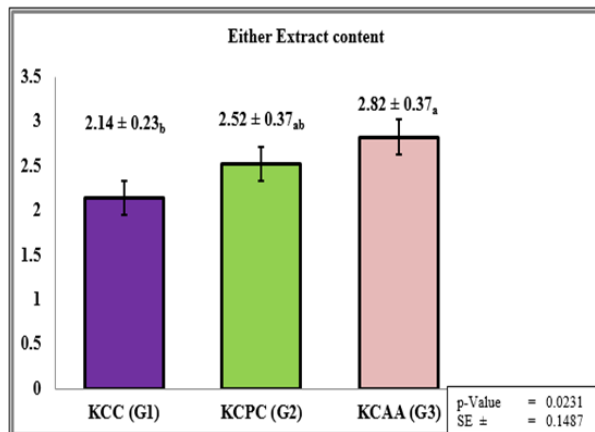


Figure-1 Panel: E Graphical presentation of Either Extract content of meat. The bars of the graphs represent the groups KCC – Kachhi Control; KCPC – Kachhi Positive Control; KCAA – Kachhi Ascorbic Acid

Panel F: Ash content

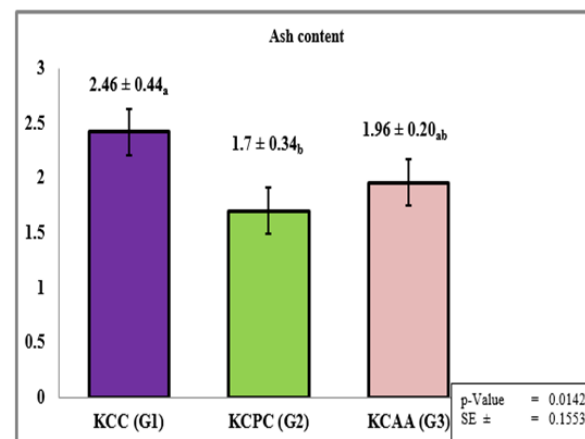


Figure-1 Panel: F Graphical presentation of Ash content of meat. The bars of the graphs represent the groups KCC – Kachhi Control; KCPC – Kachhi Positive Control; KCAA – Kachhi Ascorbic Acid

Fig. 1. Meat quality parameters of Kachhi lambs

due to heat stress, affecting fat accumulation. The findings are congruent with Gallardo *et al.*, (2015), who found that supplementing lambs with antioxidants improved the fat content of lamb meat (53). Furthermore, Pogge & Hansen, (2013a) findings imply that ascorbic acid can modify the lipid content of meat (54). The supplementation causes a shift in ascorbic acid synthesis, causing more glucose available for lipid deposition, variation in glucose turnover rate, a variable that characterizes a more dynamic view of carbohydrate metabolism.

ASH CONTENT



Ash content, which reflects the mineral composition of meat, varied significantly among the groups ($p < 0.01$). KCC lambs, showed the highest ash content ($2.46 \pm 0.44\%$), while KCAA lambs ($1.96 \pm 0.20\%$) and KCPC lambs ($1.7 \pm 0.34\%$) had the lowest as shown in Fig. 1 Panel F. The elevated ash content in KCC lambs may result from heat stress-induced mineral retention, consistent with the findings of Gallardo *et al.*, (2015) who reported that environmental stressors could alter the mineral composition of meat (53).

CONCLUSION

This study highlights the essential role of antioxidant supplementation, particularly ascorbic acid, in improving growth performance and mitigating the effects of heat stress in livestock, with significant implications for managing animals in hot climates. The observed enhancements in body weight, conformation traits, carcass quality, and meat characteristics in Kachhi lambs supplemented with ascorbic acid (KCAA group) demonstrate its effectiveness in supporting animal health and productivity during elevated temperatures. These findings suggest that ascorbic acid, when included in a balanced diet with green fodder and concentrate rations, can significantly boost growth, meat quality, and overall resilience to heat stress. Practically, integrating ascorbic acid into feeding strategies offers a valuable approach for livestock producers in hot regions, helping to alleviate heat stress and improve animal health, growth rates, and meat quality. This strategy is particularly useful in hot climates where heat stress is a recurring challenge, contributing to more sustainable and efficient livestock management. Additionally, the research opens opportunities for further studies to refine antioxidant supplementation levels and combinations across different environmental conditions and livestock species. Future research could also examine the long-term impacts of such supplementation on animal health, reproductive performance, and farm profitability. Exploring the integration of other nutritional and technological solutions, such as heat-tolerant breeds or cooling systems, could further enhance livestock management practices. In conclusion, supplementing livestock diets with ascorbic acid, especially in hot climates, is a scientifically supported method for improving animal welfare and productivity. This study advances our understanding of nutritional interventions to combat heat stress and provides practical, actionable solutions that can enhance the sustainability and profitability of livestock farming in challenging environments.

Conflict of Interest:

Authors have no conflict of interest.

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