HOW XYLAZINEADMINISTRATION AND ADHERENCE TO ANIMAL WELFARE IMPROVED CHEVON OF BUCKS EXPOSED TO ROAD STRESS

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Abstract
Thirty-two apparently healthy Sahel bucks were experimented upon in this study to assess the effect of xylazine an α-$_2$ adrenoceptor agonist *on meat quality characteristics in Sahel bucks exposed to long road transportation at high and low stocking rates. Bad handling practices would be adversely effect the psychology and this could be detrimental to meat quality. The animals at low stocking rate were stocked at 0.30m^{2}per animal; while the animal at high stocking density were stocked at 0.15 m^2 per animal. Each of experimental groups had a duplicate each stocked at a high or low stocking and were administered with xylazine at 0.01mg/kg, 0.015 mg/kg, 0.020 mg/kg intramuscularly prior and midway at staging point midway and none treated control groups. It means that all treatments had groups at low and high stocking rates respectively.The animals were then subjected to experimental journey of 996km from Sokoto (North-Western) to Abeokuta (South-Western) part to of Nigeria. The animals were rested for a day so that drug would be metabolized in the animal to a minimum tolerable level in meat. They were subsequently slaughtered and meat characteristics assessed were: live weight, carcass weight, shrinkage percentage, dressing percentage, empty body weight, ultimate pH of meat, water holding capacity and, meat colour. The doses of 0.02mg/kg,0.015mg/kg,0.01mg/kg at high and low stocking rates significantly increased carcass weight, dressing percentage empty body weight and colour (luminosity) when compared to the control.*
Conclusively, some of the meat characteristics were improved in most treatment groups of xylazineat low stocking rate (standard stocking)

Keywords: α-$_2$adrenoceptoragonist,meat,quality,transportation

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1. INTRODUCTION

Stress results in changes due to physiological disturbances caused by certain internal and external environmental factors termed as stressors (Gregory, 2004). Following physical trauma, during disease and psychological or emotional conflicts in food animals could pose as problem and could compromise meat qualityKannan *et al.*,(2002) also suggested that it is important to promote the welfare of food animals by attenuation of stress due to transportation. Stress of transportation adversely affect the health of the animal and could cause compromise in meat quality.
Ali and Al-Qarawi, (2002) and Ali *et al.* (2006)suggesteddrugs that are likely to influence the neurotransmitters and calm the animals. Some drugs suggested for amelioration of road transportation stress of domestic animals are alpha-adrenoceptor agonists, benzodiazepines, opioids and some supplements. Theα-$_2$adrenoceptor agonists were xylazine, medetomidine and detomidine. Just recently propofol an alkyl phenol was proven to be a drug of choice in the amelioration of short term transportation stress in West African bucks Biobaku *et al.* (2016c). While nutritional supplements such as vitamin C, tryptophan, and magnesium were also recommended .Adenkola *et al.* (2009) andBiobaku *et al.* (2016b) used Vitamin C to ameliorate transportation stress in pullets, pigs and goats in Northern zone of Nigeria. It has been established that consumption of meat from stressed animals is detrimental to health due to the presence of some stress induced endogenously produced biochemical constituents and hormones in the meat (Irwin *et al.*2012).The stress in animals could cause change in surge of stress hormone cortisol and might cause an exudates and dark firm

syndrome in meat which could lead to condemnation and economic loss. The objective of this study is to evaluate the effects of xylazine treatment on meat characteristics in bucks exposed to road transportation.

2. MATERIALS AND METHODS

Experimental Animals and Management

Apparently healthy Sahel bucks, 18 to 24 months of age, and weighing between 10-14 Kg were used for this study. The animals were acclimatized for two weeks and were prophylactically treated against helminthes using albendazole (Albenzole, Agbara Industries, Lagos, Nigeria) and a combination of penicillin and streptomycin (penstreptomycin® made by Kepro, Holland). The stocking rate was more than 2 m^2per animal. The bucks were fed with groundnut hay, cowpea husk and corn residue while clean fresh water was provided *ad libitum*.

The journey began at the livestock farm unit of the Faculty of Agriculture, Usmanu Danfodiyo University, Sokoto, in the Semi-arid zone of North-western Nigeria and ended at Abeokuta, in the Rain Forest zone of South-western Nigeria. The distance travelled during the experimental journey was 996 km at an average speed of 40 km/h.

Experimental Design

Thirty-two animals were randomly separated into eight groups of four animals each. Each group was administered with 0.01mg/kg,0.015mg/kg and 0.02mg/kg at high stocking and same doses were used at low stocking rates respectively. Two groups other groups were designated as high stocking control and low stocking control in this groups they were not treated with any agent .The animals that were stocked at low stocking rate were stocked in conformity with the Animal Disease Control Act of 1988 of Nigeria and is in agreement with standard international regulations for transportation of goats. Animals that were stocked at high stocking density were considered as non adherence to standard international regulation in animal transport. This is with intent of creating a condition similar to the stocking rate most transporters and livestock sellers use in transporting animals routinely in comparison to the standard in the other parts of the world.

Administration of Xylazine and Dose Extrapolation

Xylazine (XYL-M2® Berendonk Drug Company, Belgium) an injection solution was used in this study and all were administered using intramuscular route. The dose of 0.01 mg/kg xylazine was adopted based on a previous study (Sanhouri *et al.*1992). Other doses used were 0.015 mg/kg and 0.02 mg/kg as previously adopted by Biobaku *et al*.,(2016a).Thus, graded doses administered were within the safety therapeutic margin. Xyalzine was administered at two intervals at the beginning of the experimental journey at Sokoto and mid-way into the journey at Jebba.

Loading, stocking, Induction of Transportation Stress and Ethics

Prior to transportation, health certificate and a movement permit number were obtained from the Veterinary unit of the Ministry of Forestry and Animal Health, Sokoto State, Nigeria. This was done in conformity with guidelines stipulating standard animal transportation welfare.

The bucks were handled with care during loading and off loading and throughout the journey. The animals stocked at high stocking were estimated at 0.15 m^2 per animal. The area occupied by animals at low stocking density was 0.30 m^2 per animal. Which was in conformity with international guidelines on transportation of animals as previously described by Biobaku *et al.* (2016a).The animals stocked at high density were not given food and water. The explanation for the difference between the high and low from this point of view is that the animals stocked at high density is the crude way by which some marketers that are not conformist embark upon and is not encouraged. The animals transported at low density were given access to water and feed. This is to adhere to animal

transportation welfare regulations. In this study animals there were different treatment groups in attempt to address psychological and physical stress and control groups. This is done in an attempt to make comparison of good and bad practices in animal transportation practices in tandem with the use of pharmacologic agent xylazine in mitigation of transportation stress. The feed included wheat bran and cowpea husk which were given 12-hours into the journey and at a staging point at the quarantine post at Jebba midway into the journey where samples were collected for analysis and this was considered the staging point.

Assessment of Carcass and Goat Meat (Chevon) Characteristics

Prior to slaughtering, the animals were rested for twenty-four hours. This is in conformity with international standard. This was as stipulated in the Animal Disease Control Act of 1988 of the Federal Republic of Nigeria. The animals were rested for a day to ensure that they metabolized the xylazine to the most probable minimum tolerable level before slaughter.Eighteen animals were slaughtered in all, three per group using "halal" method which involved severing the jugular vein, carotid arteries, trachea and oesophagus as adopted by Biobaku*et al*. (2016b). After slaughtering, the neck region where the incision for slaughter was made was tightened firmly with a rope. The animals were then inflated using a pumping machine (Yamaha brand, China). The essence of this is to ease the process of shaving of the slaughtered animal. The animals were shaved using shaving blade to remove the hair from the skin. The skin is also consumed as delicacy in South-west, Nigeria. The shaving operation was carried out using a shaving blade at an angle less than $90^{o}$to the skin, in a direction of the hair strands. A piece of herbal soap called Dudu Osun® (Tropical Naturals, Lagos, Nigeria), containing *Aloe vera* plant latex soothers and honey was used with water for washing and lubrication during shaving.The difference of weights prior to journey and after journey multiplied by hundred is considered the shrinkage percentage. The carcass weight in South-western Nigeria was obtained after removing the head and legs and with the intact shaved skin and after removing the testis and the scromatic sac. The removal of internal offal comprising basically the plug (the laryngeal cartilage, trachea, lungs and heart) or organs in the thorax and other organs in the abdominal cavity . After the removal of the organs the carcass bearing the skin in this study is considered the dressed carcass. The carcass weight without the internal organs was considered as the empty body weight. The dressing percentage was obtained by taking the carcass weight and dividing it by the live weight multiplied by one hundred as carried out by Tamir and Awuk,(2015).Cooking loss test was also conducted using a chop from the *semimembranosous* muscle measuring 5 cm in length and width and 1 cm thickness and weight of 4 gram as previously carried by Kannan *et al.*,(2002);Jibir *et al.*, (2012); and as adopted by Biobaku *et al*.,(2016b). The percentage loss in weight after boiling was deduced by subtracting the final weight after boiling from the initial weight multiplied by a hundred, using the method of Biobaku *et al*., (2016b) Meat pH was determined using a chop from the *semimembranosous* muscleas previously described by (Pethick *et al.*, 2005).Water holding capacity of the *semimembranosous* muscle was assessed using the filter paper method of Wierbicki and Deatherage, (1958). Meat colour assessment of *rectus abdominis* muscle was carried out using the method of Colomer-Rocher *et al*. (1987) and as adopted by Jibir *et al.*, (2012), which possess values to express magnitude of luminosity and extend of congestion of meat. The higher the luminosity the lower its congestion and redness and the lower the luminosity the higher its congestion and redness.

Statistical Analysis

The values of various parameters were analyzed by the analysis of variance using Graphpad Prism version 5.03 and $P<0.05$ was considered as level of significance.

3. RESULTS AND DISCUSSION

Figure 1 shows the effect of xylazine treatment on carcass weight in Sahel bucks exposed to long term transportation stress. The Groups of xylazine at low stocking density were significantly (P<0.05) higher than the control and other treatment groups.

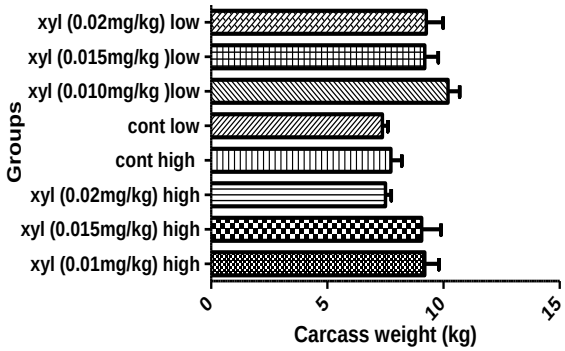

Fig.1. Effect of xylazine treatment on carcass weight of Sahel bucks

xyl indicates treatment of xylazine at a dose in mg/kg injected, while low or high indicates stocking rate or density

Stocking was considered as a condition of animals during transportation and was factored in during the pharmacologic amelioration of stress of transportation using xylazine. Stocking also could influence the welfare of animals at transportation which could affect meat characteristics. Although the influence of the drug on the transported animals could be influenced by some factors such as:stocking rate, ambient temperature, wind speed, individual inherent hereditary traits and breed and social status of animal in the group, psychology of the animal's temperament among others which could affect the direct pharmacologic actions of agent used in mitigation of stress. High stocking predisposed to more stress which could be physical or psychological or both, this coupled with long transportation stress would further cause adverse effect meat quality. The carcass weights in the animals at low stocking were higher than the weights of carcasses of animals in higher stocking. This is due to anxiety due to movement of the vehicle and contact with one another. There could be higher expenditure of energy due to muscle contraction during competition for space in the vehicle. Xylazine improve carcass weight due to its myo-relaxing and sedative effect which is attributable to be due to the sensitivity of α-$_2$ receptors in the central and peripheral nervous system. This suggestion is in agreement with Sanhouri *et al*., (1992); Khan *et al*. (1999) and Mogoa *et al.* (2003). Since the animals were calm and not adrenalized, this might have minimized the adverse impact of the general adaptive syndrome (GAS) that would affected homeostasis and the weight of the animal and the carcass weight as previously suggested by Biobaku *et al.*,(2016a)

Figure 2 shows the effect of xylazine on shrinkage percentage of chevon of Sahel bucks exposed to long term transportation stress. The animals at low density that were treated with xylazine had significantly (P<0.05) higher shrinkage percentage than other groups.

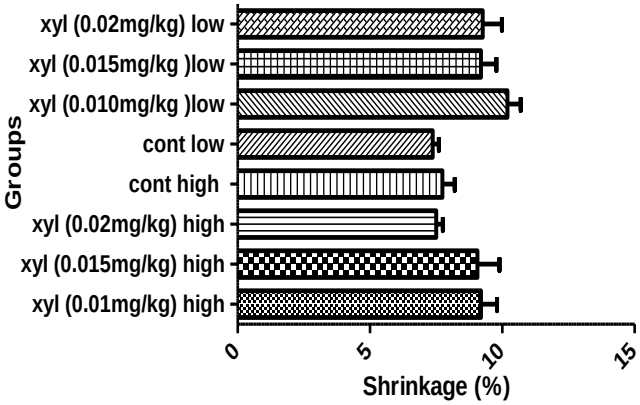

Fig. 2. Effect of xylazine treatment on shrinkage percentage of Sahel bucks

xyl indicates treatment of xylazine at a dose in mg/kg injected, while low or high indicates stocking rate or density

Shrinkage percentage in the animals treated with xylazine at low stocking rate was similar to this one at the high stocking rate. It is evident that environmental conditions might be the primary cause. In this study, induction of stress was carried out in the harmattan in the semi-arid along the trans-Savannah to the Tropical rain forest of South-western, Nigeria. Due to higher dry air circulation between the animals in spite the low stocking in and the use

of ameliorative xylazine resulted in higher shrinkage percentage. In contrast, animals at high stocking density must have snuggled due to the cuddling of the animals. This might have improved the warmth of the animals and might have minimized the circulation of dry air that would have caused shrinkage in the animals. In the same vein, animals that were administered with xylazine at low density might have decreased the rate of metabolic activity which could affected the appetite and decrease feed intake and rumen motility in the goats. This could be due to the central action of α_2-adrenoceptors agonist which has effect on the vagal action. This assumption is in line with the previous observation of Van Miert *et al.*, (1994). This might interfere with the satiety center to cause a decrease in the amount of feed intake at rest 12-hours post loading , this could improve physiologic adaptation.This adaptation of decrease in feed intake might have influence on the health of animals, this would further be assisted by adherence to Animal Disease Act of 1988 which is in tandem with international standard. This could also influence the weights and result in deficit in weight and shrinkage of the animals in the treatment groups. The decrease in appetite and demand for food at long transportation might improve homeostatic adaptation in long transportation stress in animals, since increased demand could cause physical stress due to hunger and could predispose to hypoglycaemia.

Figure 3 shows the effect of xylazine treatment on dressing percentage of Sahel bucks exposed to long term transportation stress. The group administered with 0.02mg/kg at low density stocking rate had significantly ($P<0.05$) higher dressing percentage when compared to other groups. The dose (0.015mg/kg) of xylazine at high density stocking had significantly ($P<0.05$) higher dressing percentage when compared to other groups at high stocking density.

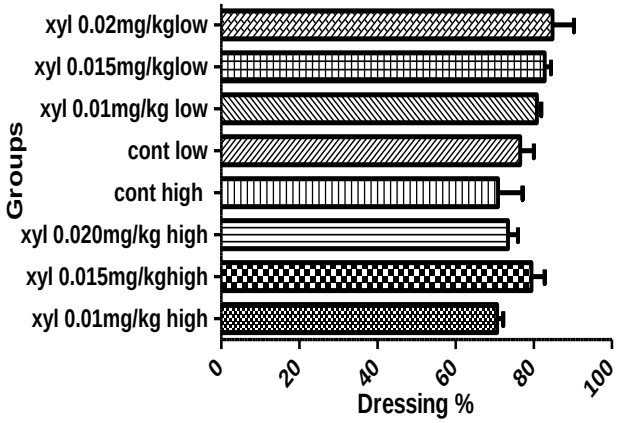

Fig. 3. Effect of xylazine treatment on dressing percentage of Sahel bucks

xyl indicates treatment of xylazine at a dose in mg/kg injected, while low or high indicates stocking rate or density

Higher doses of xylazine of 0.02mg/kg at low density and 0.015mg/kg at high stocking density influenced the dressing percentage. This could mean that dressing percentage possess a dose-dependent relationship with xylazine. The characteristics of meat improved upon by a dose-dependent factor. This could be associated to the α-2 receptors in the central and peripheral nervous system. Xylazine must have acted onα-2D and other α-2 receptors at the central nervous system, most probably the hypothalamus more specifically the satiety center. This could have interfered with the rate of ingestion of feed and might result to an after effect on weight. The effect on the weight would invariably influence the dressing percentage of the meat. Similarly, this would also affect the rate of ingestion of feed during resting and this would improve the empty body weight of the animal.

Figure 4 shows the effect of xylazine treatment on empty body weight of Sahel bucks exposed to long term transportation stress. The groups administered with 0.01mg/kg at low stocking density and 0.01mg/kg at high stocking density had significantly higher ($P<0.05$) empty body weight when compared to other treatment groups.

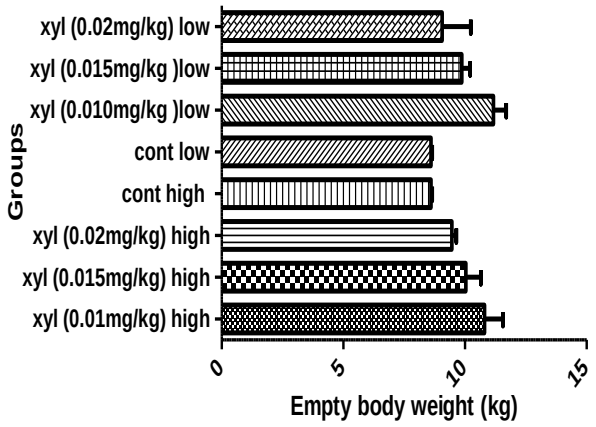

Fig. 4. Effect of xylazine treatment on empty body weight of Sahel bucks

xyl indicates treatment of xylazine at a dose in mg/kg injected, while low or high indicates stocking rate or density

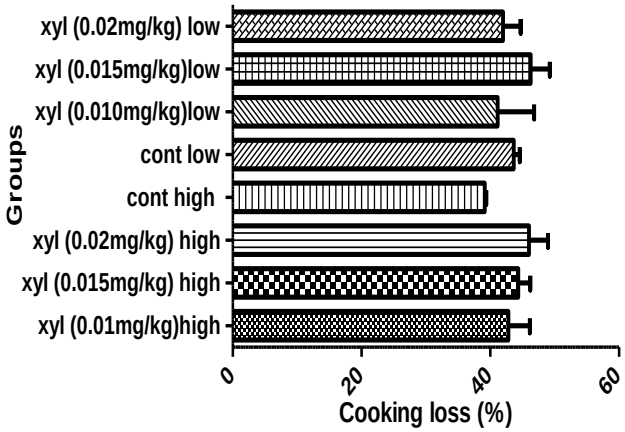

Fig. 5. Effect of xylazine treatment on cooking loss of chevon of Sahel bucks

xyl indicates treatment of xylazine at a dose in mg/kg injected, while low or high indicates stocking rate or density

The effect of ameliorative xyalzine resulted low gastrointestinal motility and centrally affected the satiety center resulting to less *ingesta* taken and the relative increase in empty body weight. Therefore, similarity exists in the phenomena relating the dressing percentage and empty body weight in this study. However in this study the major contrast of empty bodyweight and dressing percentage is that empty body weight is not dose-dependent.

Figure 5 shows the effect of xylazine treatment on cooking loss of chevon of Sahel bucks exposed to long term transportation stress. The groups administered with xylazine 0.01mg/kg at low stocking density had significantly ($P<0.05$) higher cooking loss while the group administered with 0.02mg/kg at high stocking density was significantly higher ($P<0.05$) amongst the groups of higher stocking rate groups . The control group however, had significantly lower ($P<0.05$) cooking loss percentage than other treatment groups.

Cooking loss is the decrease in percentage weight of the meat sequel to boiling or roasting. This could be attributable to changes in juiciness, amount of connective tissue, and water holding capacity of the meat after cooking.

This parameter was improved by higher doses of 0.015mg/kg of xylazine at low density and the dose of 0.02mg/kg at high density. We speculate that the myo-relaxing effect of xylazine might cause the myofibrils to be relaxed and hence releasing fluids and electrolytes to the outer more concentrated hot water at boiling. This would subsequently result in loss in weight of meat after cooking and increasing in cooking loss percentage.

Xylazine decreased metabolic activities in the myocytes due to relaxation and sedation. Xylazine myo-relaxing effect caused the meat to have lower tendency to hold fluid within the myocytes and contractile unit.

This decreased the water holding capacity of the chevon. The animals are expected to be adrenalized due to fear of fight and fright due to the transportation stress. Xylazine prevented the agitation in the animals and exudation due to soreness of the meat brought about by injury at transportation. This supports the previous work of Minka and Ayo, (2010).But in this study myorelaxing and sedative effect could be the likely explanation for this outcome. While in the previous study the cytoprotective and anti-oxidative effect of ascorbic acid could be the explanation to the observation. The similarity of the two studies could be due to the decrease in excitability attributable to ascorbic

acid as stated by Ayo *et al.*, (2006) and the decrease in the excitability due to xylazine as previously observed by Biobaku *et al.*, (2016a) which might be the reason for calmness and the effect on electrolytes in the tissues and the chevon cooking loss characteristics.

Figure 6 shows the effect of xylazine treatment on water holding capacity of chevon of Sahel bucks exposed to long term transportation stress.The treatment groups of xylazine at both stocking densities had significantly ($P<0.05$) lower water holding capacity when compared to the control group at both stocking rates.

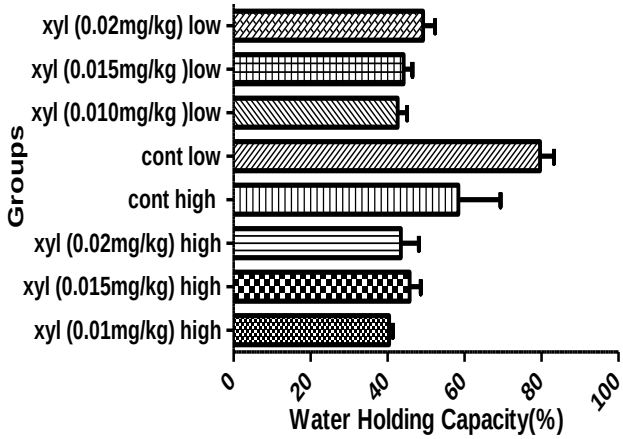

Fig. 6. Effect of xylazine treatment on water holding capacity chevon of Sahel

xyl indicates treatment of xylazine at a dose in mg/kg injected, while low or high indicates stocking rate or density

Although xylazine resulted in decrease in water holding capacity we could corroborate that it prevented the dark dry firm syndrome and meat exudation due to transportation stress.

Figure 7 shows the effect of xylazine treatment on chevon colour of Sahel bucks exposed to long term transportation stress. The 0.02mg/kg at low stocking density had significantly ($P<0.05$) higher colour (luminosity) when compared to other treatment groups and to the control group. While the xylazine treated groups had significantly higher ($P<0.05$) luminosity values when compared to control at higher and low stocking

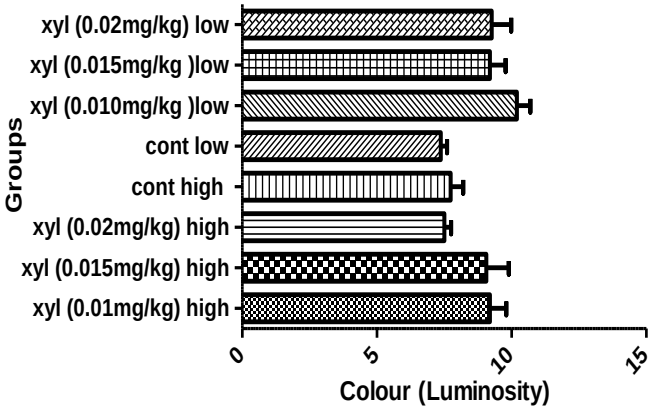

Fig. 7. Effect of xylazine treatment on chevon colour of Sahel buck

xyl indicates treatment of xylazine at a dose in mg/kg injected, while low or high indicates stocking rate or density

The xylazine improved meat appearance by having higher luminosity values; higher luminosity values mean pinker better appearance of the meat as previously suggested by Jibir *etal.*(2012);Biobaku *et al.*(2016b) Darker meat appearance meat tends to be due to congested capillaries and necrosis in muscle tissue. The xylazine might have caused analgesic and myo-relaxing effect prevented muscle damage at the cellular level. Preventing centrally acting effect elicited at pre and post junction myo-neural level had suggested a cyto-protective effect in the muscles by preventing the detrimental effect of catecholamine out flow from the peripheral nervous system. Xylazine due to itsα-$_2$ agonistic effect on the pre and postsynaptic effect in the central neuron could cause sedative effect which could protect the neurons from stress due to anxiety. The sedation would decrease metabolic rate and influence circulation of blood and possibly at the capillaries level this would affect perfusion to the muscles due to anxiety prompted by the sympathoadrenal system.This could affect the oxymyoglobin dynamics in the muscles, thus the muscles would not be congested due to sedation and improvement in myocytes oxymyoglobin concentration. This might influence the colour of the muscles and the meat would be lighter. The colour chart had

higher luminosity value and a better and more wholesome chevon.

Figure 8 shows the effect of xylazine treatment on ultimate pH of chevon of Sahel bucks exposed to long term transportation stress. The treatment group with xylazine had significantly ($P<0.05$) higher pH than the control group at low density.

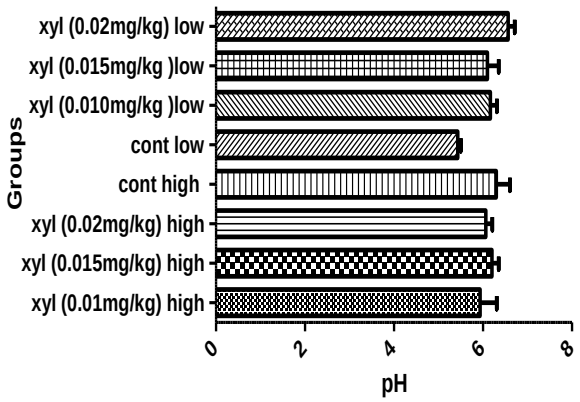

Fig. 8. Effect of xylazine treatment on ultimate pH of chevon of Sahel bucks.

xyl indicates treatment of xylazine at a dose in mg/kg injected, while low or high indicates stocking rate or density

Xylazine treatment must have affected the metabolism and this must have prompted the homeostasis to shift from the normal. The effect on the pancreas and dynamics of glucose metabolism might also be responsible for these changes in ultimate pH. In the same vein the xylazine might have decrease in respiratory rate and medulla which might haveaffected the physiological mechanisms that occur during contraction and relaxation of muscles in the animals exposed to stress. The non treated groups however, had their muscles rigorously involved in contraction and relaxation which needs high muscle energy expenditure. This requires higher glucose demand, glycogenolysis and increased level of lactic acid deposition in the muscles which would further decrease the pH of the meat.

4. CONCLUSION

Xylazine in this study suggests that if used to ameliorate transportation stress of transportation at therapeutic doses in goats improved some chevon characteristics. The meat characteristics improved are carcass weight, dressing percentage, and empty body weight and meat colour luminosity respectively. Other characteristics were influenced but the due to the detrimental effect of long transportation stressed still prevailed on the other characteristics chevon namely; shrinkage, water holding capacity, pH, and cooking loss percentage.

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