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PERFORMANCE OF IMPORTED SHEEP TO KUWAIT UNDER SIMULATED PROCEDURES TO TRANSPORTATION PRIOR TO SLAUGHTERING

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ABSTRACT

More than three hundred imported Australian sheep were used to determine the ability to categorize and differentiate stressed from a lesser stressed animals that transported all the way from Australian ports to Kuwait. Accuracy of differentiating the two groups went up to 90%. Such accuracy encouraged the team members to proceed with the second phase dealing with the initiation of the Stress Score System. This will help the decision maker to give the right grouping decisions for weather animals should go to slaughtering, meat factory, live sales or for housing in the farm. Simulated procedures were used to create stress on experimental animals in two ways. Animals were stressed due to altitude change and kept in multiple level cages that are movable with the movement of the animal and categorized into three sub groups (H = Third floor) , (M = Second floor) (L = Lower or ground floor). The other group of animals was kept under simulated measure of tough handling processes ((Th= Tough handling) or normal handling processes (Nh = Normal handling). Animals exposed to simulated actions for location and handling showed a decline in cortisol levels by time. This was shown clearly by plotting the linear trend graph for the response of serum cortisol level in animals exposed to the several stress actions. Animals in L floor fluctuate lesser than the other groups. All animals exposed to tough handling had higher serum cortisol level than the Nh group. Animals from H and M floors gained lesser weights than those from L floor throughout the experiment time. Meanwhile, animals in Nh group gained lesser weight than those in Th group. Animals under higher stress in all groups showed darker meat, less tenderness and poorer taste preference. Ratios of liver, kidney and heart weights to the half carcass weight were measured. Liver and kidney were of greater ratios under higher stress while heart ratios were the opposite. Cortisol level for animals that head covered at slaughter time was lower than those without a head cover while color, taste and tenderness did not show significant difference due t the short time of the stress effect.

KEYWORDS: Lamb, stress, transportation, handling, simulation, physiological parameters.

INTRODUCTION

Handling of livestock is an important issue that is strongly believed to affect the performance of the livestock in general in the ante-mortem stage **(Scanga et al 1998). Temple Grandin in 1995** observed, during twenty years of work on livestock, that many people use sheer force rather

than using behavioral principles in restraining the animals. He found that animals could become bruised or injured under the conventional handling like using the regular squeezed chutes. In 1981 a survey on seven major feedlots by **Brown *et al.* (1981)** indicated that in five of the feedlots, 1.6% - 7.8% of animals were bruised. Bruises is not the only concern since this will heal by marketing time but pain and trauma that affect sheep performance and reduce weight gain **(Grandin, 1990).** Crowded animals specially those transported from ship to farm could suffer stress where **Grandin (1980)** found that some animals exposed to this type of stress could collapse and even die if operator fails to release them immediately. The finance manager of a major farm in Kuwait reported that they loose not less than 12% of the shipment of sheep. These losses are either during shipping from the original home land, during downloading in the port, during housing in the farm or during the transportation to Central Slaughter House (CSH).

This type of information drives the breeders and those who have interest in this industry to pay more attention to this loss and spend more efforts to decrease it. Unfortunately, neither in Kuwait nor in the neighboring countries any efforts were spent in this field from the angle of the degree of stress that the animals suffered through handling procedures at any stage. Not only in this aspect but also the performance of sheep in regard of carcass weight which will be very crucial to the wholesaler who will sell later by weight. It might affect the percentage of fat which could affect the percentage of red meat in the carcass. Cortisol level will be checked as a reference for stress intensity in blood.

This project aims at studying some of the stress affecting sheep and how it is important to handle sheep with care and professionalism. It is a step towards a process of initiating the Stress Score System due to transportation and handling after boarding in Kuwait. The study is restricted to the activities after arrival to port of Kuwait since it is almost impossible to gain precise information about several parameters before boarding especially in the handling procedures. Several phases will be conducted after this study including the initiating of the SSS (Phase I) and then the study of the physiological effects of the animal stress due to handling and transportation procedures (Phase II) and finally the third phase that will deal with the evaluation of the management measures to reduce the losses due to stress of transportation.

MATERIALS AND METHODS

Identifying the Stressing Procedures and Stressed Animals

In this task a careful attention was paid to each single handling procedure for the sheep. Actions of handling were varied due to the stress intensity on these animals. Staff members monitored the handling

procedures and were able to identify the stressed animals during different procedures of handling. Border line sheep were ignored while those showing the stress symptoms were tagged with red and those were not were tagged with different color e.g. yellow. This was done in animals' handling from the shipping vessel to the farm and during housing in the farm and from the farm to the slaughter house. During the following month the staff started to apply this procedure in the farm. Stressed animals were grouped rather than individual. Each group of sheep was tagged with color that reflects the way of handling in each handling procedure. This was based on: 1- Actions of handlers during any time (calling), 2- Roughness of flooring, 3- Steepness of connecting channels, 4- Crowd of sheep at any stage, 5- Position in the trailer, 6- Duration from the port to the farm and from the farm to the slaughter house, 7- Position in the vessel, 8- Number of sick or dead sheep in the pen.

Simulated procedures to the unloading activity of sheep from the vessel to the port were done on 200 sheep. Stressed animals were tagged with red at that time while the non stressed ones will be tagged with yellow tags.

An extensive monitoring practice was done on several hundreds of sheep prior to tagging any of the sheep groups. Sheep were tested for the cortisol level in their blood to assure the accurate grouping. This work took four to five months until staff became confident with grouping the sheep into stressed (R) or non-stressed group (Y). After this, staff started tagging and continued the rest of the tasks.

Assessment of stress level by Measuring the Cortisol Level in Blood as an Index

All tagged sheep were tested for cortisol level in their blood. Blood sample was drawn from a vein (vein puncture), usually the jugular vein on the sides of the neck of the sheep. The puncture site was cleaned with antiseptic, with pressure by hand to restrict blood flow through the vein. This caused vein below the tourniquet to distend (fill with blood). A needle was inserted into the vein, and the blood was collected in an air-tight vial or a syringe. During the procedure, the tourniquet was removed to restore circulation. Once the blood had been collected, the needle was removed, and the puncture site was exerted with pressure to stop any bleeding.

Blood samples were analyzed for level of cortisol, using COBAS INTEGRA 400 device.

Blood samples were collected and kept in ice box in the field and transferred later to animal physiology laboratory at KISR. Serum was separated in the laboratory and stored in the freezer for cortisol level determination using Enzyme Immuno Assay EIA (**Cook *et al.*, 1997**).

Measurement or Carcass Weight, Percentage of Fat, Meat Color, Hardness and Taste.

All tagged sheep (colored) that are under the simulated situation of the study were loaded in the trailers aiming to the slaughter house. Sheep were slaughtered and carcass weights were measured along with recording the meat color and hardness. Meat hardness was checked using TAXT2 Texture analyzer whereas the color of meat were checked using color checker CIE lab system using the rib eye muscle between the 12th and 13th vertebrate or using other available methods. Half of the carcasses (the left half) were frozen to check for the percentage of fat in the carcass. A sample of the hand muscle of half of all the tagged carcasses were tested for taste preferences.

Statistical Analysis

Collected data from all the above tasks were analyzed using Least Square Means analysis of variance using General Linear Model of the statistical analysis system (SAS 1998). All tested parameters were treated as dependent variables. Age of the sheep, weight of sheep at origin, sex of sheep, body condition score and the score of stress intensity were used as independent variables whereas the cortisol level was used as dependent variable. Significance of the difference between least square means was determined using Student–Neuman–Keuls method. Interaction of the tested independent variables was taken into consideration if any.

RESULTS AND DISCUSSION

Categorizing the Stressed Animals

A close look was made to all animals that were unloaded from the carrying vessels. Stressed animals were identified according to the location on the ship, flooring, steepness of the connecting channel, crowd and calling; team members started to call groups of animals and categorize them into high and low stressed animals. Groups of animals were located in certain enclosures in the quarantine area in the port which made easier to the team to take blood samples from the categorized ones. Blood samples were collected from 295 animals, where half of them called as high stressed ones and the other half as the low stressed ones. Results from this analysis showed that ninety percent of the categorization procedure was correctly grouped as showing higher cortisol level in serum Table 1. In spite of the difference in intensity of stress between the groups, all the highly stressed animals showed higher cortisol levels in serum compared with the lower stressed ones (Graph 1).

Accuracy and efficiency in categorizing animals under stress will enable the decision makers in the industry to sort the imported animals depending on their stress condition and how suitable they are for either

slaughter, keep for recovery or send to meat factory. These decisions will help the industry to avoid low quality of end products with lesser shelf life and low taste grade and color for meat of the imported sheep. This means a lot to the industry that always seeks better color and taste for meet to satisfy its own customers.

Table 1. Mean Cortisol Levels mcg/dl for Sheep unloaded from a Vessel Traveled from Australia to Kuwait Port and Categorized as lower and highly stressed animals according to the position on board and to the handling procedures followed.

Visit number	Stress	Number of animals	Mean of cortisol level
1	Low	25	4.1
1	High	25	7.7
2	Low	30	4.1
2	High	30	8.24
3	Low	25	4.75
3	High	25	10.70
4	Low	30	5.90
4	High	25	9.12
5	Low	15	4.85
5	High	15	4.55
6	Low	25	2.80
6	High	25	3.90

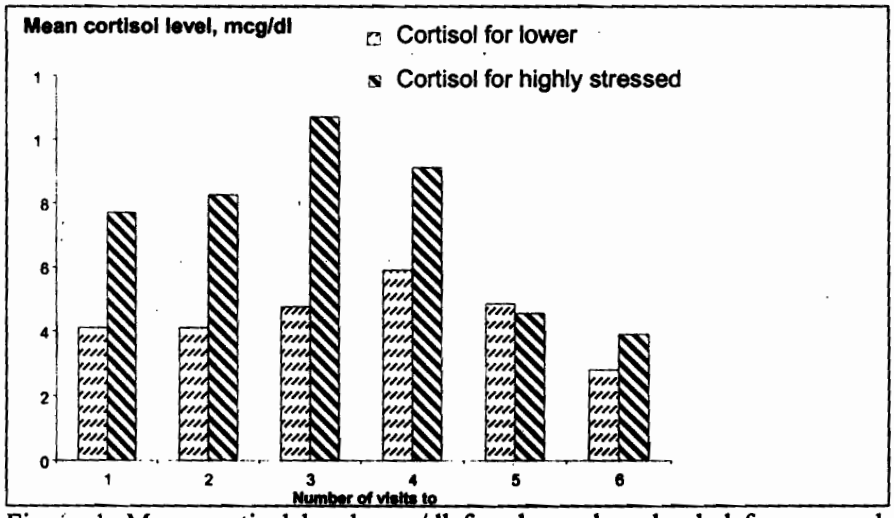

Figure 1. Mean cortisol level mcg/dl for sheep downloaded from vessel traveled from Australia to Kuwait port and categorized as lower and highly stressed animals according to the position on board and to the handling procedures followed.

The Simulated Study and the Cortisol Levels

Table 2 is showing the least square means of cortisol level in the serum of sheep exposed to stress under simulated procedure for a carrying vessel. Cages with three levels of altitude (H=high, M=middle and L=low) were made and animals were kept in these cages taking into consideration the same space allowance for each in a single cage. Cages were shaky upon the movement of the animals giving the same feeling of a moving vessel. Animals were showing a fluctuating cortisol levels which was typical for this cortical hormone response (Figure 2). Cortisol levels in all experimental sheep showed a gradual decrease in the response which might indicate the toleration of the sheep to the stress effect for this period of time (Figure 3). Animals onboard traveling that period of time all over from Austarlia to Kuwait not only were exposed to this experimental stress factor but also had other stresses like climate and odor which were absent here. This might make animals taking even longer time to tolerate the combination of these stressogenic factors. Such combined factors are worth to be studied for the sake of knowing the period needed for the toleration and the time needed for the recovery.

All Animals exposed to the three types of stresses tended to respond gradually with reducing response intensity by the end of the fifteenth experimentation days. In spite of similarity of the general response for the three groups of animals, sheep that supposed to be under the higher stress effect (H) tended to tolerate faster than those at the lower and medium ones (M, L) (Figure 4). That was shown clearly on the sharper linear trend line of the serum cortisol response for animals of the higher level group (H) while the other two groups (M, L) showed almost similarity in the response during the fifteen days period (Figure 4).

Different experimental groups were exposed to either high (Hh) or low (Lh) stress effect due to handling procedures including calling, housing and downloading for the same time period of fifteen days. Tables 3, 4, 5 and 6 showed that animals exposed to rough handling measures (H) had higher serum cortisol levels during all the experimental days at all the tested times around the day. Animals of lower stress actions (L) tended to have lesser fluctuation levels than those exposed to the higher stress levels (Figure 5). Similar to the observed toleration response at the end of the fifteen days period, there was a toleration response within a single day to the stress effect for both Hh and Lh groups (Figure 5). Animals from both groups like animals exposed to stress due to altitude showed toleration over the fifteen days period for the stress even if it was not complete one for the (Hh) group (Figure 5).

Table 2. Least square means of cortisol levels mcg/dl in the serum of sheep exposed to stress under simulated procedure for a carrying vessel.

	Day1	Day5	Day8	Day10	Day 12	Day15
Control	6.2	5.5	5.8	5.5	4.4	4.2
Medium	5.5	5.9	6.1	5.4	4.3	4.2
Higher	7.2	6.9	6.3	4.4	4.2	4.1

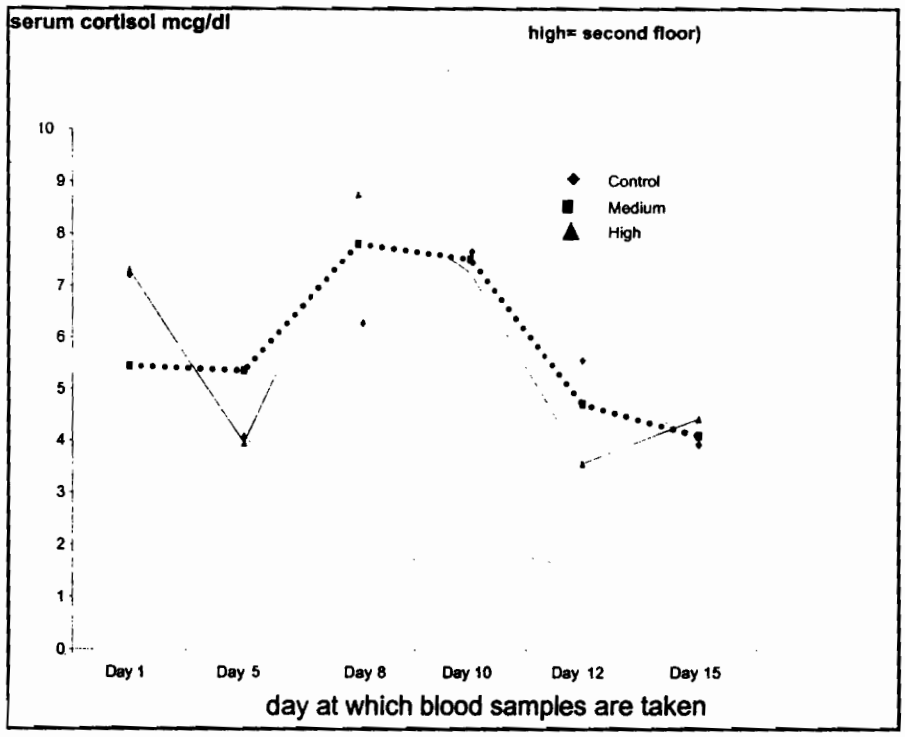

Figure 2. Least square means of serum cortisol level mcg/dl in imported sheep exposed in simulated way to different levels of stress due to altitude (low = ground floor, medium first floor and high = second floor).

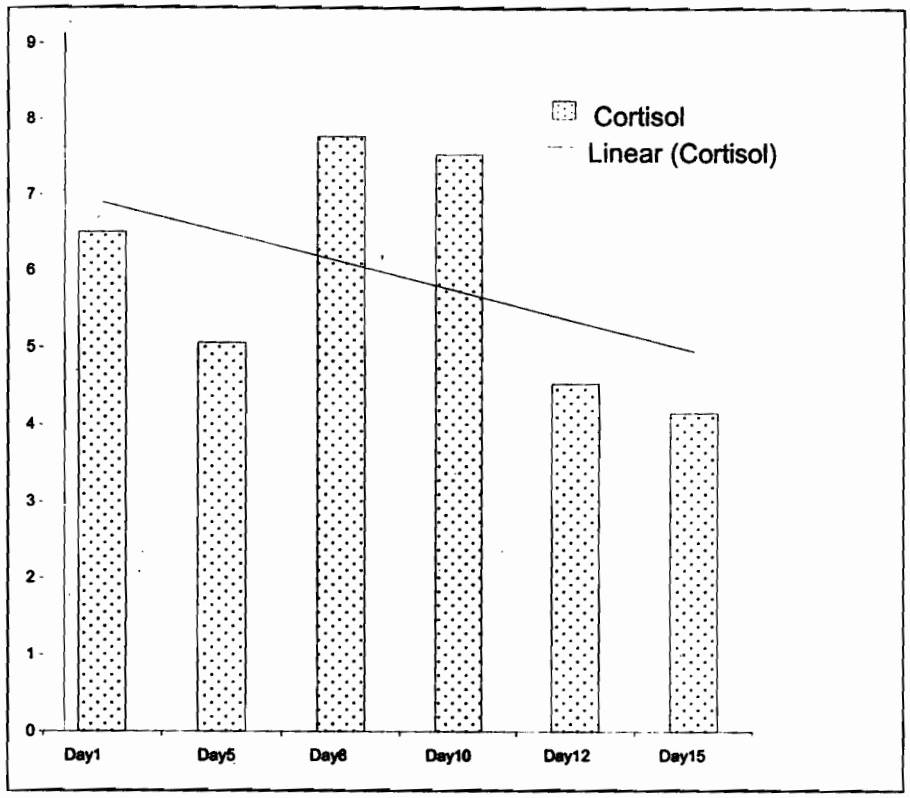

Figure 3: Overall serum cortisol level in imported sheep in three level (ground, first floor and second floor) cages for 15 days.

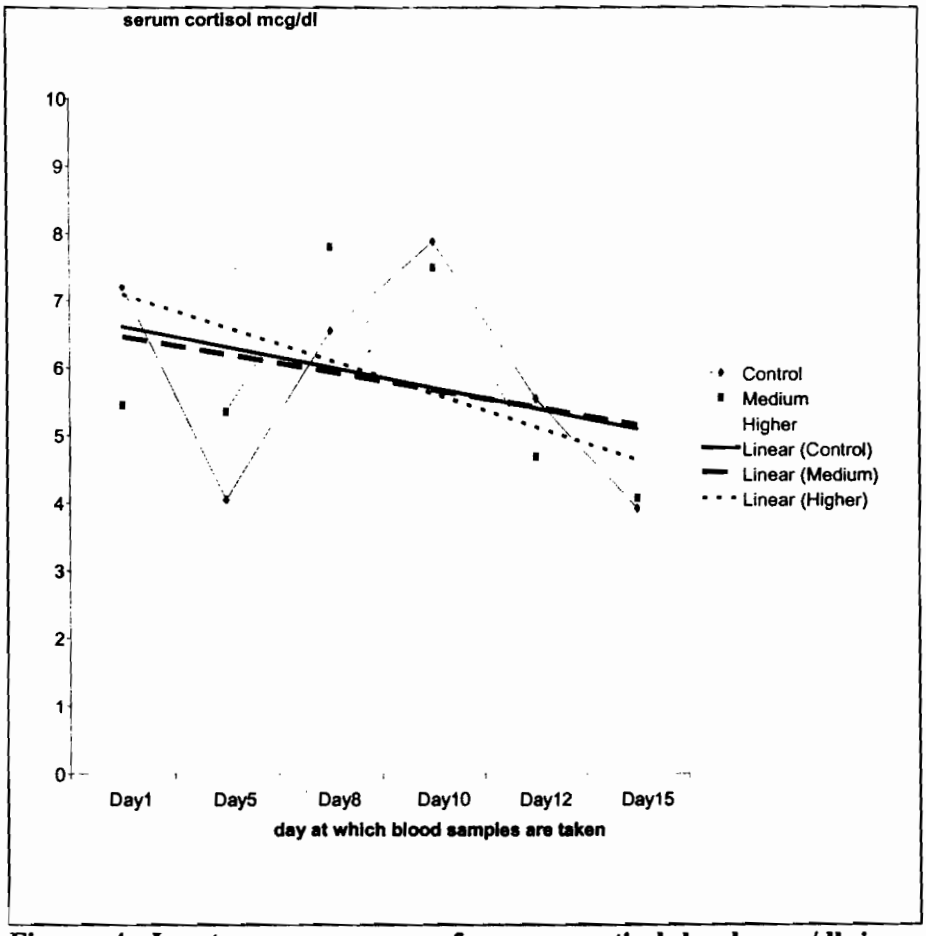

Figure 4. Least square means of serum cortisol level mcg/dl in imported sheep exposed in simulated way to different levels of stress due to altitude (low = ground floor, medium first floor and high = second floor)

Table 3. Least square means of cortisol levels mcg/dl of imported sheep exposed either to high or low stress level due to general handling processes at different times of the first day in the enclosures.

	Day1 8am	Day1 9am	Day1 10am	Day1 11am	Day1 12pm
Low stress	4.2	3.8	4.4	4.9	3.1
High stress	14.3	7.13	5	6.8	5.3

Table 4. Least square means of cortisol levels mcg/dl of imported sheep exposed either to high or low stress level due to general handling processes at different times of the fifth day in the enclosures.

	Day5 8am	Day5 9am	Day5 10am	Day5 11am	Day5 12pm
Low stress	2.6	3.4	3.3	3.11	3.12
High stress	5.2	6.4	7.7	6.9	8.2

Table 5. Least square means of cortisol levels mcg/dl of imported sheep exposed either to high or low stress level due to general handling processes at different times of the eighth and tenth day in the enclosures.

	Day8 5pm	Day8 10pm	Day10 8am	Day10 9am	Day10 10am	Day10 11am	Day10 12pm
Low stress	4.5	5	6.8	10.6	5.34	3.3	3.3
High stress	11.6	9.8	11.9	14.5	10.7	5	3.5

Table 6. Least square means of cortisol levels mcg/dl of imported sheep exposed either to high or low stress level due to general handling processes at different times of the twelfth and fifteenth day in the enclosures.

	Day12 10pm	Day12 03am	Day15 8am	Day15 9am	Day15 10am	Day15 11am	Day1 12pm
Low stress	2.5	7.2	4.1	4.2	2.44	1.13	3.12
High stress	5.7	3.4	6.4	5.1	4.2	6.1	8.2

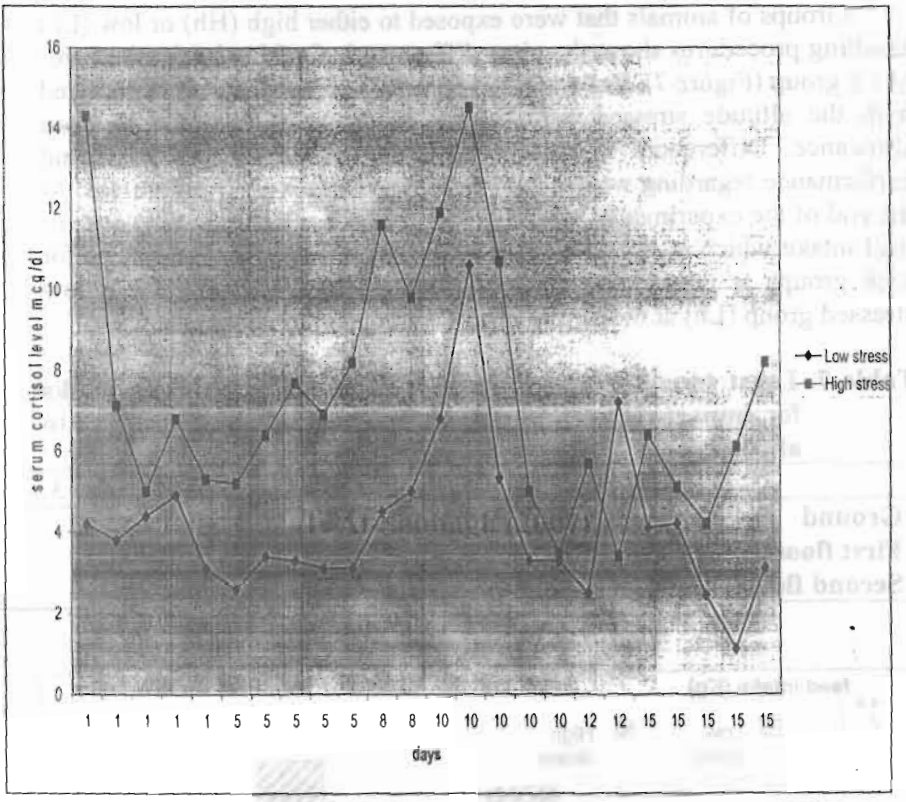

Figure 5. Least square means of serum cortisol level of imported sheep kept under simulated procedure of either low of high stress due to calling and altitude.

Performance and Feed intake

Feed intake was calculated for the three groups of animals that are (H), (M) and (L) at three different time periods and showed similarity. Although amount of feed intake were almost the same for the three different groups (Table7), weight gain was different (Figure 6). Animals from (L) group gained weight at the first time period whereas the (M) group maintained their weight and on the other side the (H) group started loosing weight. In the second test period, results showed that the (L) group maintained its weight whereas both (M) and (H) groups were loosing weights. The third tested period showed weight loss of all the three groups. Toleration of stress was observed at the end of the fifteen days period but yet all the three groups are still loosing weights which might be a nutritional cost for the toleration that might be made up from the reserved energy of the animal upon the fixed feed amount offered to each group.

Groups of animals that were exposed to either high (Hh) or low (Lh) handling procedures showed a clear difference in feed intake for the favor of Lh group (Figure 7). Experimental animals were smaller here compared with the altitude stressed animals yet they were offered similar feed allowance. Differences in feed intake were clear here (Table 6) and performance regarding weight gain was compatible with it in figure 8. At the end of the experimental period, toleration took place on the expense of feed intake which is expressed here in the decrease of the weight loss for both groups at the second test period until it was shown for the low stressed group (Lh) at the third period with an increase of weight gain.

Table 7. Least square means of feed intake at three different weeks for imported sheep exposed to three levels of stress due to altitude.

	Feed intake 1	Feed intake 2	Feed intake 3
Ground	4.9+-.05	4.95+-.05	4.95+-.03
First floor	4.8+-.05	5+-.05	5+-.03
Second floor	4.8+-.05	4.85+-.05	4.8+-.03

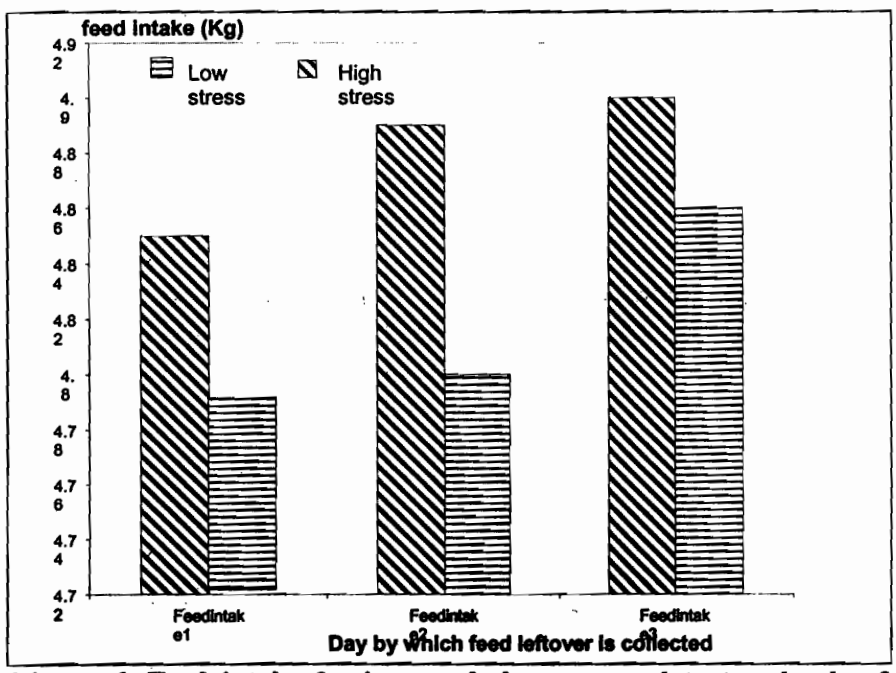

Figure 6. Feed intake for imported sheep exposed to two levels of stress in simulated procedures for fifteen days.

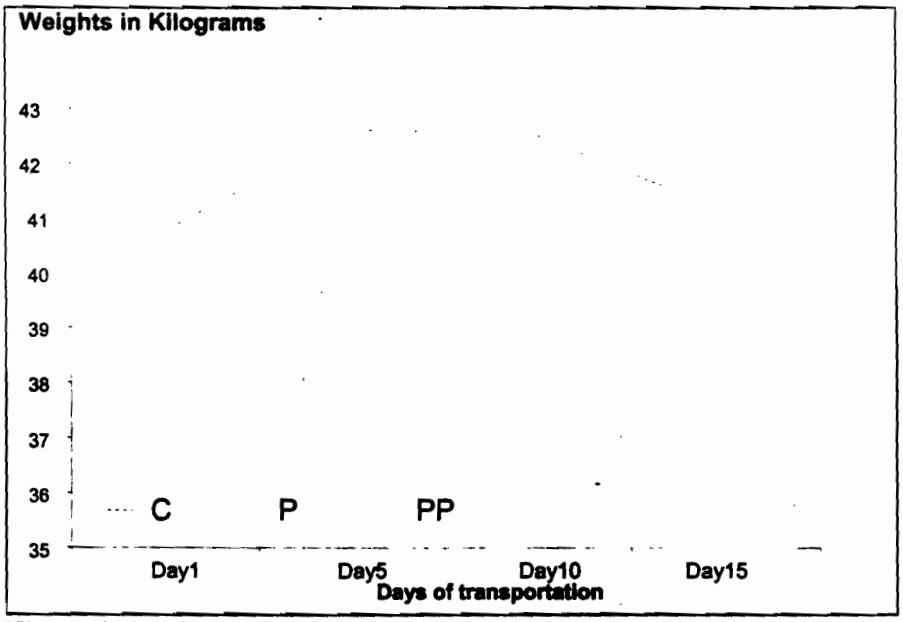

Figure 7. Performance of sheep weights under simulated effects if transportation for 15 days.

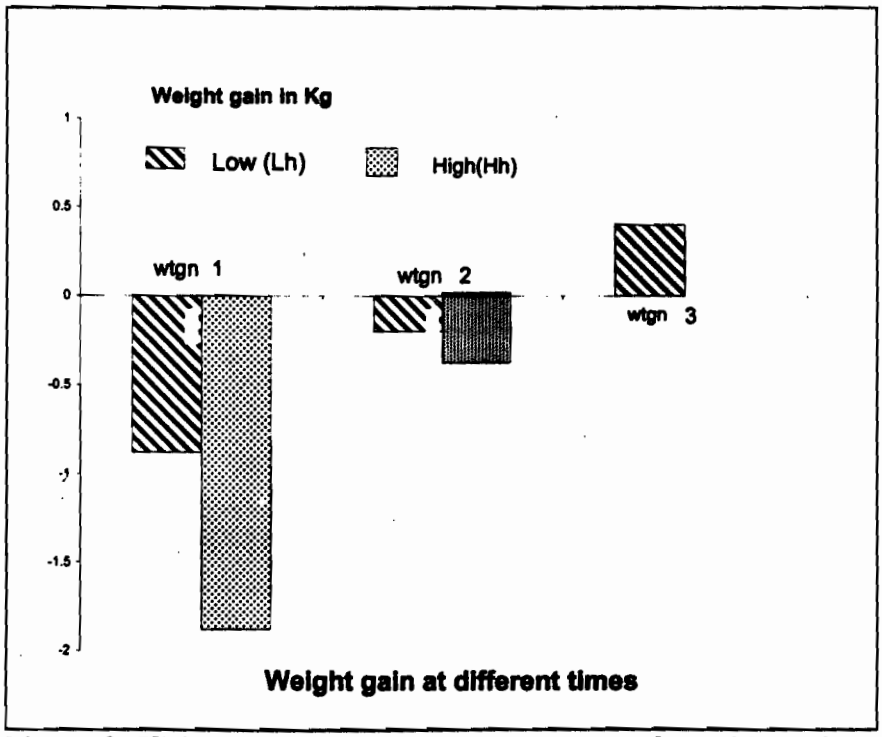

Figure 8. Weight gain of imported sheep exposed to either high handling procedures (Hh) or low (Ll).

Performance of Sheep Regarding Carcass and Quality

Stressed animals, due to altitude, (H, M and L) were monitored after slaughter and tested for color, toughness of the meat and taste. Animals in the low level (L) were lighter than both (M) and (H) groups, not only that but tender and tastier (Figure 9). Although groups (M) and (H) have almost the same meat color, group (M) has tender meat and tastier (Figure 9).

Color of meat for group of animals that were under higher stress due to handling (Hh) was darker with less tenderness and poorer taste than the (Lh) (Figure 10).

Internal organs were evaluated by checking the ratio of its weight to the carcass weight. Liver ratio for animals of all groups was almost similar, with the superiority of the (H) group. Kidney ratio followed the same pattern where the highest ratio is for (H) and the lowest for (L) (Figure 11). This could be due to the physiologic response of the body due to stress that increase the need for the function of the liver and kidney which needs more investigation to clarify that effect, a matter proposed in the third phase of the project.

Heart ratio was inversely related to the altitude sheep exposed to in the cage (Figure 11). Second group of handling stress followed the same pattern where animals exposed to higher stress actions (Hh) had higher liver and kidney ratios than low ones (Lh) whereas heart ratios are lower for the (Hh) than (Lh) (Figure 12).

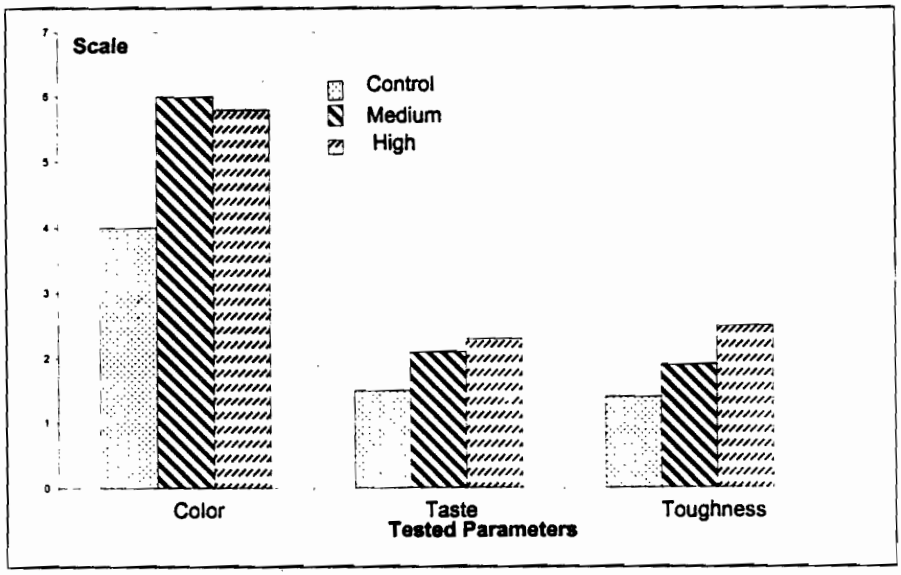

Figure 9. Color, taste and toughness of meat of imported sheep exposed to three levels of stress due to altitude.

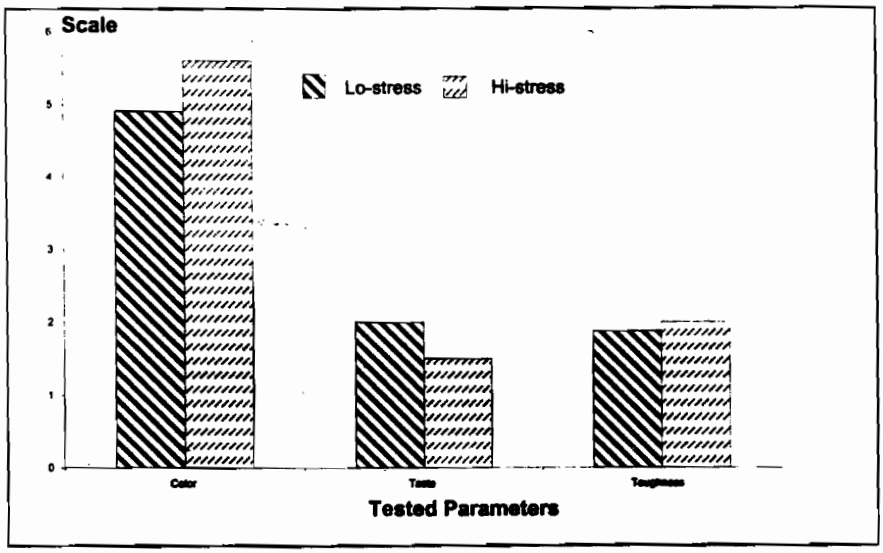

Figure 10. Color, taste and toughness of meat of imported sheep exposed to either high or lower stress due to general handling processes.

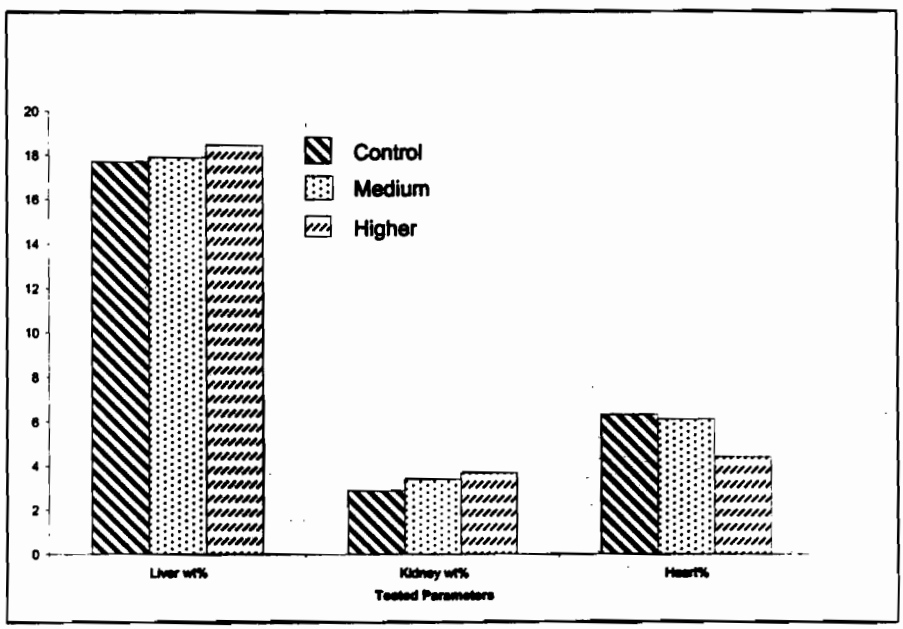

Figure 11: Ratio of liver, kidney and heart weight to half carcass for imported sheep exposed to three levels of stress due to altitude for fifteen days.

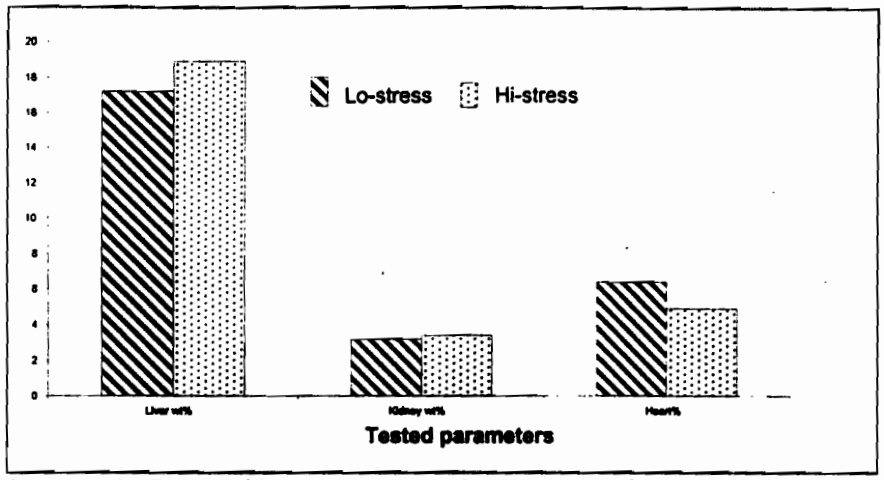

Figure 12: Ratio of liver, kidney and heart to half carcass of imported sheep exposed to either high or low level of handling process once of Head Covered Versus Non-covered during Slaughter.

Animals from the same group were treated either by covering their heads with a black bag (C) where they can breath but can not see through it, or non covered (NC). Animals that were head covered at slaughtering showed lower cortisol level at slaughter time than those who were not covered (Figure13).

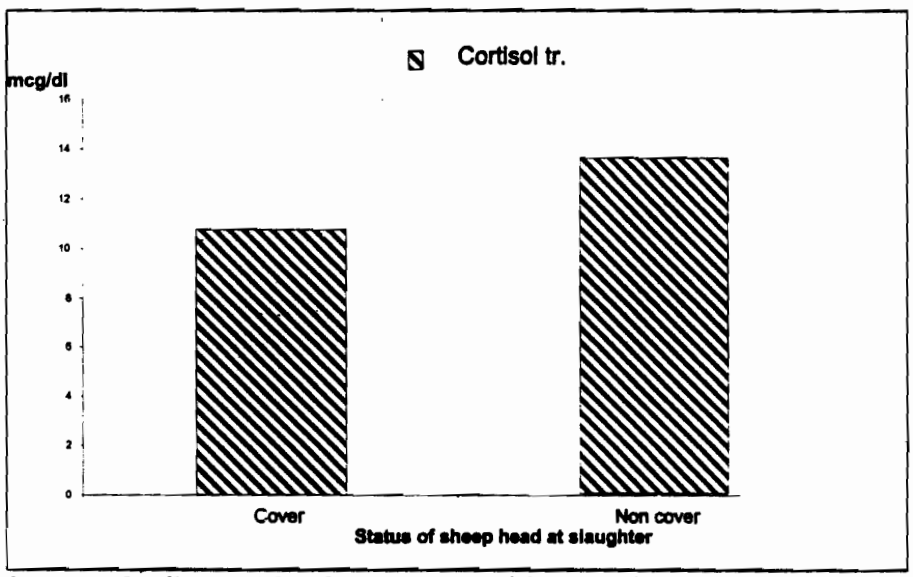

Figure 13: Cortisol levels in serum of imported sheep either head covered or non- covered at slaughtering time.

Color Taste and Tenderness were almost the same between the two groups with the superiority of the head covered animals. The absence of difference in color taste and tenderness could be due to the short time effect of the increase of cortisol level where it was not enough to play its role in decreasing the glycogen that was needed to produce the lactic acid that increase the acidity of the end products (Figure 14).

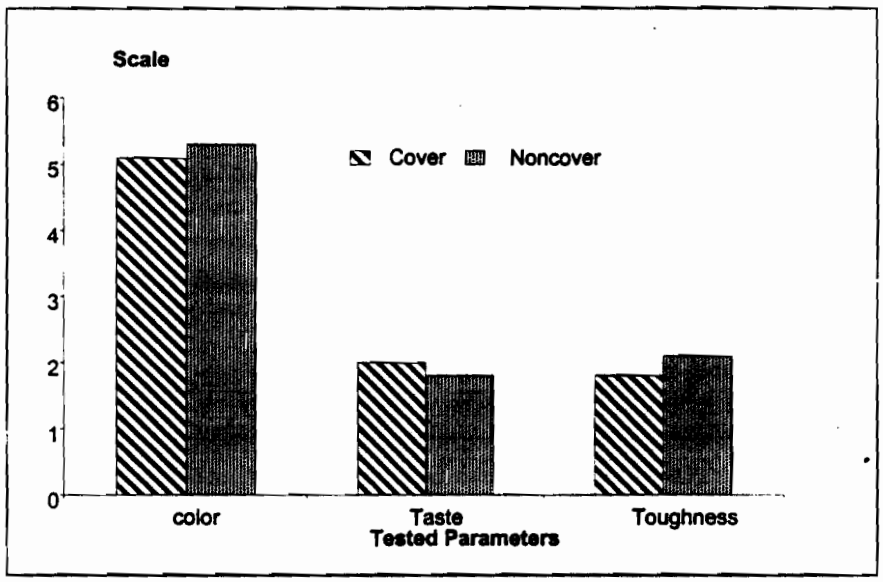

Figure 14. Color, taste and toughness of meat of imported sheep head covered versus non-covered at slaughter time.

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