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Effect of mow procedure on physiological and biochemical properties of blood in sheep

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Abstract: *In this study18 female sheep aged between 20-25 months was used in this experiment in alqasim city .the mow procedure was performed in a closed area using traditional method with shearers***.**
The aim of study was to evaluate the stress response and the oxidant antioxidant balance against shearing process in sheep. Blood malonaldldhyde (MDA)and glutathione (GSH)concentration , were measured in 18 female sheep one day before a traditional mow procedure. The study show increase of respiration and heart rate . Circulating MDA concentrations were dramatically increased compared to initial values after shearing whereas GSH concentrations were significantly depressed .the variations of other biochemical parameters were not significant .these results demonstrate that MDA and GSH are the most powerful markers for evaluating the oxidant /antioxidant status and that mow was a stressful situation leading to an oxidative stress .
Key word: *sheep, mow, cholesterol*

I. Introduction

Shearing, no matter of the season is always accompanied by some degree of thermal stress. The latter drives a number of physiological and biochemical adjustments aimed at maintenance of the homeothermy that ultimately affects the level of production (1) . It has been considered insulation and adequate food supply to have survival significance for adaptation in cold environment. In producing domestic animals this extra need of energy maintenance have to be combined with the needs for production (2). .mow at ambient temperatures below the limit of thermoneutrality may reduce the availability of metabolizable energy for productive processes. Cold may directly influence the rate of milk secretion. (3) noted that cold exposure of ewes affected the distribution of blood flow within the mammary gland, but did not affect the total flow to the udder mow sheep contributes directly to the welfare of both animal and the the owner .shearing is necessary to enhance the physical welfare of the animal ,as domestic sheep do not shed their wool naturally (4),but can negatively affect the welfare of the animal if performed in an inappropriate way and time .mow can result in thermal stress when animals are exposed to wet conditions ,severe cold ,or intense suns-hine coupled with high temperatures. failure to shear ewes before confinement for lambing ,even in the winter may result in moisture and health problems in the barn. Shearing process causes stress on the animal because circulating corticoid concentrations increase(5)regardless of the method used ,and noise ,heat mind contact of the clippers induce this reaction .In addition ,the traditional method of up-ending sheep for shearing (resting on rump in upright position)contributes (6,7) Some shearers sheep by binding their legs ,a procedure which is stressful in itself {8},and may result in injuries ,but comparison of the over-all stressfulness of this method with up-ending has not been made .shearing is less stressful if done quickly (9) ,bearing in mind cuts resulting from hurried or careless shearing add to stress (10) infectious related mortality and morbidity .Moreover ,it is indicated that stress increases the oxidative stress which affects the survival and the metabolism efficiency (12,13,14) Overwhelming evidences indicate that oxidative stress can lead to cell and tissue injury .However ,the same free radicals that are generated during oxidative stress are produced during normal metabolism and thus are involved in both health and disease (15) .Under normal circumstances ,the generated reactive oxygen species (ROS)are detoxified by the antioxidant present in the body and the generated ROS and the present ant antioxidants are in equilibrium..However, owing to ROS overproduction or inadequate . The objective of this study was to determine the stress response of sheep during mow process as well as to evaluate the consequences on the oxidant-antioxidant status.

II. Materials and Methods

Chemicals

The chemicals used in the study were purchased sigma-Aldrich (Sigma-Aldrich C chemical co.st.louis,mo,usa)

Animals Nad Protocol Design

The study was conducted on 18 female ,20-25 months old , sheep in the alqassim city ..the mow procedure was performed in a closed area using traditional method with shearers . sheep were fasted and housed in a clean area before shearing .. the goat were shorn using mechanical hand pieces .the wool was removed beginning by removing the *goat belly wool .After setting the goat on its rump ,the belly was first shorn . the wool was removed starting from the wool-free area inside the sheep right hind leg to the* wool-free area just beside the sheep right fore leg in the same direction .all the wool from the goat belly from the breast bone to the udder on ewe was cut ,the wool on the ewes udder and vulva clipping was clipped carefully .After clipping the belly ,the wool from the insides of the hind legs was clipped On the goat right hind leg a couple of blows were taken from the hoof to the crotch ,on the inside of the left hind leg ,the blows started near the crotch and proceed out to the hoof ,once the entire fleece has been removed from the sheep ,the fleece was thrown ,clean side down ,on to a wool table by a shed hand .

Heart and respiratory rest were recorded during the shearing procedure .Blood samples were taken from each animal ,an One day before and just after the shearing ,by puncture of the jugular vein into heparin zed tubes for measuring malonaldehyde (MDA) ,reduced glutathione (GSH) ,glucose ,total cholesterol and as well as the plasma total antioxidant activity the circulating MDA and GSH concentrations were measured from whole blood (2ml) whereas the other biochemical parameters were determined on plasma obtained after blood centrifugation (1000g,10 min at room temperature)and stored at -30 c.

Biochemical Analyses

Blood malonaldehyde concentration (MDA)

The circulating MDA concentration ,an index of lipid per-oxidation was measured by the double heating method of DRAPER and HADLEY{22} .the method is based on the spectrophotmetric measurement of the purple color gene d-rated by the reaction of thiobarbituric acid (TBA) with MDA Briefly ,2,5 ml of a trichloroacetic acid solution (10% w/v) was added to the whole blood (0.5 ml) and the mixture was placed in a boiling water bath for 15 min .after cooling to room temperature and centrifugation (1000gfor 10 min at 4 c)the supernatant fraction (2 ml)was transferred to a test tube containing 1 ml of the TBA solution (0.67%,w/v)min cooed to room temperature and finally the absorbance was measured at 532 nm using a Shimadzu UV160 l spec based on the absorbance coefficient of the TBA-MDA complex (=1.56.10 cm)

Blood reduced glutathione (GSH) concentration

The blood GSH concentration was measured as described by BEUTLER et al {5} Briefly ,after haemolysis in distilled water(0.2 ml of blood samples in 1.8ml of distilled water) the precipitating solution (3 ml) (1.67%w/v meta –phosphoric acid .0.2% EDTA m30% Nacl)was added and the mixture was allowed to stand for approximately 5 min and then filtered (what man no 42) thereafter a 0.3 m disodium hydrogen phosphate solution (8ml)and 5,5- dithiobis ,2-nitrobenzoic acid (DTNB)(1 ml) were added to the filtrate (2 ml)and the absorbance was measured at 412 nm in the shimadzu UV 160 1 spectrophotometer ,the blank reactant was precipitating-with the phosphate solution (8 ml) the 3/5 diluted precipitating solution (2 ml)and DNTB (1 ml)and a GSH Standard solution (0.4 g/l)was used as a reference

Plasma glucose and total cholesterol concentrations

Plasma glucose concentrations were estimated by the glucose oxidase /peroxides method using the commercially available kit (n ct f400 ch , chema Diagnostic, Italy) .

Statistical Analysis

ALL data were presented as mean ± Standard Error (SE)for parametric variables .the comparisons of parameters were performed with Student s t-test Data were analyzed using the SPSS for Windows computing program (version 10,0) and p≤0,05 was considered statistically significant {65}.

III. Results

AS reported in Table I. the heart rate and reparatory rates slightly increased the shearing procedure compared to the initial values .

animal	Before mow	After mow
Heart rate beat/min	73 ±4a	95 ±5b
Respiratory rate	22 ± 2a	34 ±8b

Table au l: heart and respiratory rates measured in the female sheep (n=18) during The mow procedure .Results are expressed as means ±standard errors

Among the biochemical parameters investigated sheep (table 11),the plasma glucose and cholesterol concentrations did not exhibit significant variations the

Biochemical parameter	Before mow	After mow
MDA	52.2 ± 21a	102± 24 b
GSH g/l	177.5 ± 10.4a	111 ± 5.5b

Glucose mg/dl	77.32± 12 a	72.9 ±9.3 a
Cholesterol mg/dl	88.97± 7.2 a	89.65±6.8 a

Table Au 11: Effects of the traditional procedure on some biochemical parameters (blood MDA and GSH concentrations , glucose and cholesterol concentrations in female sheep (n=18) .Results are expressed as means ±standard errors . the result show no significant differences (p< 0,05) after mow the sheep in value of glucose and cholesterol .

By contrast blood MAD concentrations were significantly dramatically enhanced after mow compared to the initial values (p< 0,05) whereas the blood GSH concentrations were significantly depressed .

IV. Discussion

Stress has been defined as the cumulative response of animal resulting from interaction with its environment receptors(15)Biological consequences of stress have adaptive purpose , intending to keep homeostasis balance (16,17) . However , they may lead to the development of pre-pathological state and eventually, to a pathological state. the pre-pathological state has been proposed to be clinical defined by change in biological functions without obvious negative consequences for lifespan(18) the stress-induce impairment of biological functions can compromise animal well-being ,health and life(19) some hormone involved for modifications of biological functions(20)this way ,it has been reported that stressful conditions stimulate the hypophysal-adrenal axis.

Nowadays the interest of ROS in biology and medicine has been increased because of their strong relationship with aging and disease processes.

However ,excessive generation of free radicals can occur due to endogenous biological or exogenous environmental factors ,such as exposure to radiation ,pollution or chemical substances(21)For example pesticides have been reported to cause alteration in antioxidants or free radical scavenging systems(22) .when ROS production and various antioxidant systems are imbalanced (23)cellular injury and tissue damage occur leading to alterations of macromolecules (membrane lipids, proteins and DNA) changes in, intracellular calcium and intracellular ph ,and finally to cell death(24) .the lipid attack mediated by free radicals ,named as lipid peroxideation (LP) ,is complicated radical chain reaction leading to the formation of various products including lipid hydro peroxides ,conjugated dynes and thiobarbituric acid-reactive substances (TBARS)such as malondialdehyde(MDA) which can be measured and used as markers for LP (25,26,27) Since membrane phospholipids are the major targets of oxidative damage .lipid per oxidation is often the first parameter analyzed for proving the involvement of free radical damage and the plasma MDA concentrations have been reported to directly correlate with the severity of stress (28). Although there is no report available on oxidative stress and shearing procedure ,it has been shown that some other management procedures cause oxidative stress .29found an increase of plasma TBARS and a decrease of plasma lipid soluble antioxidants in moderately heat stressed mild-lactating cows during summer .AVCI et al. avci et al (30)suggested that transport might play an important role in oxidative stress by reducing GSH and increasing MDA concentration in sheep .in the present study ,shearing procedure has induced strong elevations of blood MDA concentrations occurrence of an oxidative stress ,that result could be explained by the occurrence of free radicals due to the stress factors during the shearing process.

in ruminants ,plasma glucose come approximately at 44% from organic acid absorption from the rumen (predominantly propionate)and subsequent conversion to glucose in the liver at 33% from post- ruminal glucose absorption ,and at 23% from other carbon sources such as amino acids and subsequent conversion to glucose in the liver .catecholamine's and glucocrticoide increase the glycogenolysis and the gluconeogenesis [31,32,33] and secondarily the glycemia . the plasma glucose concentration been reported to be increased In stressed animals(8,28,37,49) however in this study ,shearing procedure unchanged plas-ma glucose concentration . (34) reported that maximum glucose concentration is achieved two hours after .cholesterol is synthesized mainly in the liver but its concentrations also depend on the diet supply (35).the result is agreement with AVCI e1 al.(36). Which reported that the plasma cholesterol concentration didn't change in transported sheep . (37).

0the other investigated parameters glucose and cholesterol concentrations in response to shearing could be an additional factor responsible for oxidative stress A more complete identification of the physiological changes during shearing could be beneficial for further researches in terms of correct management practices within sheep industry .

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