NIGERIAN VETERINARY MEDICAL ASSOCIATION

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BOOK OF PROCEEDINGS

For the

36TH ANNUAL NATIONAL CONGRESS

Theme:

THE ROLE OF THE VETRINARIAN IN THE 21ST CENTURY CHALLENGES OF PRIVITISATION

Produced by:

KADUNA STATE CHAPTER

VENUE: AREWA HOUSE, KADUNA.

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Table 1. Composition of the Corn-bases ration

Ingredients	Percentages
Corn meat	20.00
Palm kernel cake	20.00
Wheat offal	20.00
Brewers' grain	37.80
Groundnut cake	1.85
Sodium chloride	0.25
Minovit supper	0.10
Total	100.00

Table 2. The reproductive parameters of WAD does fed on different nutritional planes.

Parameter	Group A - with high nutritional supplementation	Group B - with low nutritional supplementation	Group C - with no nutritional supplementation	Total
Number of does	20	20	20	60
Does that kid	17	16	10	43
As % of total does	85.0	80.0	50.0	71.67
Types of birth				
Singletons	12 (12)	12 (12)	7 (7)	31 (31)
Twins	12 (6)	8 (4)	6 (3)	26 (13)
Triplets	3 (1)	0 (0)	0 (0)	3 (1)
Total	27 (13)	20 (16)	13 (10)	60 (47)
Gestation Periods				
Short Gestation Periods	11 (64.71%)	8 (50.0%)	5 (50.0%)	24 (55.81%)
Normal Gestation Period	3 (17.65%)	3 (18.75%)	2 (20.0%)	8 (18.60%)
Long Gestation Period	3 (17.65%)	5 (31.25%)	3 (30.0%)	11 (25.58%)
Total	17 (100%)	16 (100%)	10 (100%)	43 (100%)
Birth weights (kg)				
Singletons	1.50 ± 0.06	1.30 ± 0.05	1.20 ± 0.10	1.35
Twins	1.30 ± 0.09	1.24 ± 0.09	1.08 ± 0.03	1.25
Triplets	0.53 ± 0.03	0	0	0.53 ± 0.03
Mean total birth weights	2.45	1.86	1.82	2.12
Standard Error Mean	0.23	0.22	0.25	

EFFECT OF TRYPANOSOMOSIS ON SOME BLOOD BIOCHEMICAL PARAMETERS IN RABBITS

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ABSTRACT

The effect of trypanosomosis on blood biochemical parameters were studied in 20 New Zealand white rabbits of 6 to 7 months of age.

Mean total protein and globulin levels in the infected rose significantly ($P<0.05$) while the mean albumin level decline in the same group simultaneously. Mean total plasma cholesterol, triglyceride concentration mean glutamate oxaloacetate transaminase (GOT) and the glutamate pyruvate transaminase (GPT) levels decreased significantly ($P<0.05$) in the infection group. The implications of these results are briefly discussed.

INTRODUCTION

Livestock production especially in Africa is hampered by many factors including diseases (FAO, 1976, Griffins, 1978). Trypanosomosis is one of the haemoparasitic diseases of economic importance to man and domestic animals (Anosa, 1983) it is regarded as the biggest factor limiting the number and productivity of animalos among the endemic livestock diseases in Africa (Hornby, 1952). Animal trypanosomosis is also important because of its socio-economic and socio-cultural effects, it has been a major hinderance to the efforts to settle the nomadic graziers (Ilemobade, 1981).

The pathogenic typanosomes of domestic ruminants and game animals are Trypanosoma vivax, T. congolense, and T. brucei (Soulsby, 1982). All the three species are found in the blood of infected animals but T. brucei is also found in the tissue.

Rabbits are monogastric herbivores of short reproductive cycle that could provide a bridge in terms of the animal protein requirement in Nigeria where less than half protein requirements is met as recommended by World Health Organization. However, generally very little attention has been paid to rabbit diseases especially the blood parasitic infections which continue to decimate their population. A good knowledge of these diseases is important in their control. This study was designed to study the effect of trypanosomosis on some blood biochemical parameters in rabbits.

MATERIALS AND METHODS:

Twenty male New Zealand white rabbits aged between six and seven months were bought from a rabbit farm at Molete, Ibadan.

Inoculum

The T. congolense used in this study was isolated from cattle in 1994 at Bassa, in Plateau State. The parasite was passaged in Swiss albino mice (Mus musculus) prior to sub-inoculation into experimental rabbits.

EXPERIMENTAL DESIGN

Experimental Animal

The rabbits were divided into 2 groups, A & B with those in group A as infected and those in group B as controls.

Group A consisted of 15 rabbits, each intraperitoneally infected with 10^6 T. congolense as described by Lumsden et al. (1973).

Group B consisted of 5 uninfected rabbits. Both groups were bled through the marginal ear vein weekly for 3 consecutive weeks for serology.

Experimental Method

Trypanosomes were detected by the dark ground buffy coat method (Murray et al; 1977). Total protein was determined by using Biuret method (Coles, 1974), albumin was determined by bromocresol green-binding method while globulin was determined as described by Toro and Ackerman (1975).

Chemical calorimetric methods as demonstrated by Edwards et al (1972) were used in the analysis of serum cholesterol and triglycerides. Glutamate oxaloacetate transaminase (GOT) and glutamate pyruvic transaminase (GPT) concentrations were determined by calorimetric methods as described by Toro and Ackerman (1975).

Statistical Analysis

Results are presented as mean ± standard error of the mean. P values of ≤0.05 were considered significant.

RESULTS

The results of changes in the biochemical parameters of uninfected control and infected rabbits are presented in Table I and Figs. 1 and 2. The mean total protein level gradually increased in the infected rabbits (Fig.1). The increase was significant ($P<0.05$) while the mean globulin levels increased significantly as compared to the levels of the control rabbits. A decrease in mean values of albumin globulin ratio (AGR) was also observed in the infected rabbits.

The plasma total cholesterol concentration declined significantly ($P<0.05$) following infection (Fig. 2). A significant decrease was also observed in the mean plasma triglyceride concentration in the infected rabbits when compared with the uninfected controls. The mean GOT values rose gradually in the infected rabbits when compared to values in uninfected controls. This increase is significant when compared with that of the control rabbits $P<0.05$. The mean GPT values increase significantly throughout the period of infection when compared to that of controls.

TABLE 1: BLOOD BIOCHEMICAL VALUES OF T. CONGOLENSE INFECTED RABBITS.

PARAMETERS	MEAN VALUES IN CONTROLS	MEAN VALUES 8 DAYS PI	MEAN VALUES 15 DAYS PI
Total Protein (g/dl)	5.30±0.50	6.23±0.13	6.26±0.20
Globulin (")	2.80 ± 0.12	3.76 ± 0.07	3.86 ± 0.18
Albulin (")	2.51 ± 0.13	2.40 ± 0.18	2.40 ± 0.12
AGR	0.91 ± 0.07	0.66 ± 0.06	0.62 ± 0.06
Cholesterol (mg/dl)	d86.14 ± 14.72	76.50 ± 15.5	58.00 ± 2.00
Triglyceride (mg/dl)	66.71 ± 7.50	65.33 ± 3.30	61.33 ± 15.72
GOT (u/L)	46.57 ± 4.81	49.33 ± 10.84	55.66 ± 12.68
GPT (u/L)	83.85 ± 19.0	100.30 ± 24.03	127.00 ± 26.08

DISCUSSIONS

In this study, total protein and globulin levels increased. This corroborates the reports of Anosa and Isoun (1976) in T. vivax infected goats and sheep, and Facer (1976) in T. brucei infected rabbits. Increase in the globulin levels may be due to changes in IgM and IgG fractions at various stages of the disease in the rabbits as described by Luckins, 1972 and Kalu et al., 1989. It has been suggested that the decrease in the plasma albumin levels may be due to the uptake of albumin-bound fatty acids and lipoproteins (Vickerman and Tetley, 1979) and haemodilution (Katunguka-Rwakishaya et al, 1992a). The result of these changes was decrease in the albumin/globulin fractions of proteins. The infected rabbits showed significant decrease in plasma cholesterol and triglycerides concentrations, with resultant decrease in serum total lipids. This, again confirms the findings of previous workers (Katunguka-Rwakishaya et al, 1992b and Roberts, 1975, 1977). It has been proposed that cholesterol is essential for trypanosome growth (Black and Vanderweed, 1989) and the findings of Traoure-Leroux et al., 1987 and Katunguka-Kwakishaya et al., 1992c suggested that higher blood cholesterol concentration promotes the parasitic growth and multiplication. Trypanosomes need the nergy derivable from the fatty acid metabolism (Tizard et al., 1978). Trypanosomes therefore obtain more fatty acids in addition to those circulating freely in plasma by uptake of albumin bound fatty acids (Vickerman and Tetley, 1979) and Lipoproteins, especially low and high density lipoproteins. The increase in both GOT and GPT as observed in this study agrees with the findings of Gray (1963) in T. vivax infection in sheep, but not with the report of Welide et al, 1974 in T. congolense infection of cattle which showed decreased values. The elevation in GOT and GPT values ordinarily suggested specific organ damage in trypanosomosis as follows, to the liver and heart tissue for the GOT and the liver alone for the GPT. This, according to Cornelius et al. (1959), is because the two enzymes are exclusive to the tissue by these organs, and their level in circulation reflects the extent of damage or necrosis of the tissues of these organs and hence the severity.

The present study has demonstrated the series of biochemical changes observed in trypanosomosis due to T. congolense in rabbits and also related same to the severity of the disease.

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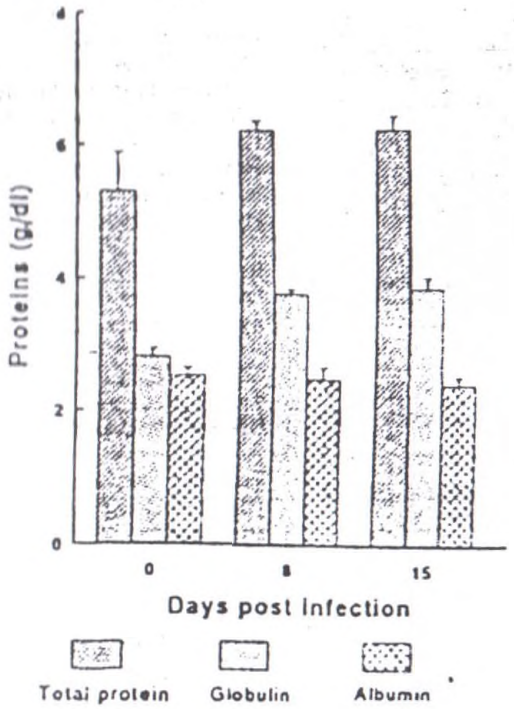

Fig.1: Mean changes in total protein, albumin and globulin levels in T. congolense infected rabbits.

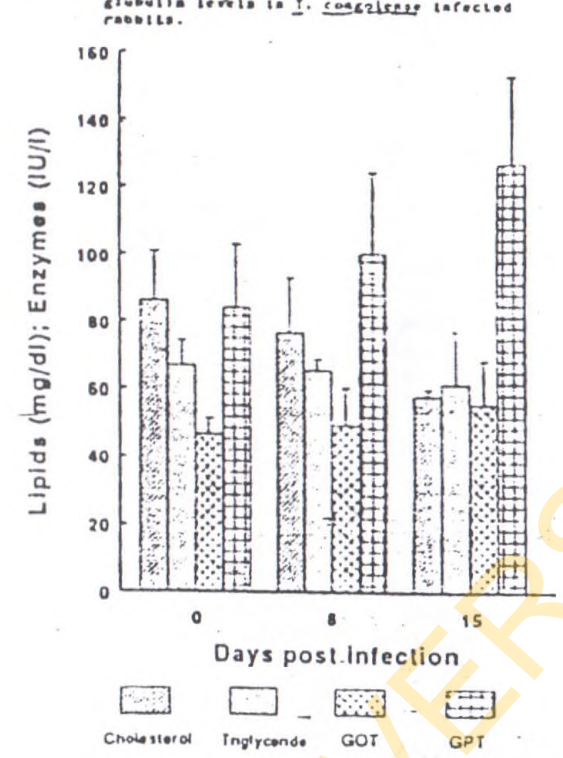

Fig 2: Mean Changes in plasma cholesterol triglyceride, and transaminases in T. congolense infected rabbits.

THE EFFECT OF FEED SUPPLEMENTATION ON THE WEIGHT CHANGES, LIVER ENZYMES AND SOME MINERALS IN ADULT WEST AFRICAN DWARF DOES.

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ABSTRACT

A total of twenty clinical healthy aged West Africa Dwarf (WAD) does were used for this study. The does were divided into two groups. Their feeding was supplemented with corn-based concentrated rations.

There was significant increase in the body weight of all the animals just before parturition as compared with their weight at service ($P<0.05$).

There was also a significant increase in the level of the liver enzyme found in the serum after parturition than before parturition ($P<0.05$). There was no significant difference in the level of some mineral (Na, K, Ca) in the serum before and after parturition.

It is advisable to give feed supplementation to does to improve mothering ablity, prevent abortion and sustain multiple births.

INTRODUCTION.

High birthweight is desirable in goats as it is correlated with live weight increase and so affect the time taken to reach slaughter weight (Datta et al., 1963). Diwali, 1943). High birthweight also has an influence on mortality rate; survivors are heavier at birth than those that died.

Litter size and birthweight seem to be influenced by the weight of the doe. Since birthweight influences survival rate, the attention paid to the nutrition of does prior to mating and during pregnancy, should result in an increase in the peerventage of kids reared (Adu et al., 1979).

When a higher plane of nutrition was given in late pregnancy, it resulted in higher doe or wew live weight as well as higher kid or lamb birth weight (Wilkinson and Chestnutt, 1988, Osuagwuh, 1984). Approximately, 70% of fetal growth in sheep and goats takes place during the last six weeks of gestation (Wallace, 1948). The level of energy feeding in pregnancy has for long been clearly established as a major factor affecting birthweight and viability of lambs especially those born as multiples (Robinson, et al. 1979, Osuagwuh, 1991).

It has been established that does or ewe weight change varied greatly between those fed on a high energy ration and those fed on a low energy ration. Those fed with low energy ration lost weight and those on high energy ration gained weight during pregnancy in any of the groups. Weight changes relate to litter size with ewe or does with multiple kids/lambs losing more weight or gaining less than those with single keds/lambs (Khalaf et al., 1979).

Energy requirements for pregnancy take into account the fact that most of the growht of the fetus takes place in the last two months before birth. It is therefore suggested that the goat requires an extra 6 megajoules (mj) of metabolizable energy (ME) per day during the last two months or pregnancy. There is evidence that increasing energy intake during the last two months of pregnancy by 4 - 7mj of metabolizable energy per day improves subsequent performance (Wilkinson and Stark 1987).

Undernutrition, in terms of energy or protein level in late pregnancy, can result in a substantial depression of birth weight and this is associated with increased perinatal losses. Nutrient requirements for the fetus follow a similar trend to fetal growth being very low in early pregnancy and increases markedly in the last trimester (Everitt, 1968). Osuagwuh (1984) found out that basal ration in the form of quality forage may not successfully maintain pregnancy.

In the early state of pregnancy, the amount of nutrient deposited are small and it is only in the last third stage of pregnancy that it becomes necessary to make special provision in the diet for the growth of the fetus. The net energy needed for the growth of the uterus and its contents is small in relation to the maintenance requirements of the mother herself during the early stage of pregnancy but the net requirements for protein, calcium, phosphorus and other mineral elements increase quite appreciably in the later stages of pregnancy (McDonald et al, 1981)

MATERIALS AND METHODS

A total of twenty healthy West African Dwarf does aged between 4 and 5 years were used for this study. The animals were kept at the Reproductive Physiology Unit of the Department of Veterinary Surgery and Reproduction, University of Ibadan. The animals were certified to be healthy and of good reproductive features with body weight ranging between 12 -18kg.

The animals were dewormed with albendazole[R] (Pfizer, Ikeja, Nigeria). They were also vaccinated against peste de petit ruminants (PPR) using tissue culture rinderpest vaccine (TCRV) (NVRI, Vom Jos, Nigeria).

Asuntol[(R)] Solution (Bayer Leverlusen Germany) was used to bathe the animals against ectoparasites and routine vetèrinary attention was provided.

FEED SUPPLEMENTATION

All the animals, in addition to being fed with dry cassava peelings (Manihot esculanta) and Elephant grass (Pennisetum Purpureum), received a conr-based concentrate for two weeks before mating. This was done to increase the level of nutritional supplementation of feed given to female animals 3-4 weeks before mating (flushing). The concentration contain 20% corn meal, 40% Brewer's grain, 15% palm kernel cake, 20% wheat offals, 3% groundnut cake, 10% salt and 1% minovit (minerals). The daily intake of the corn-based concentrate is 314gm throughout the time of the experiment. Clean fresh water was provided ad libitum.

BODY WEIGHT AND BLOOD COLLECTION

The body weight of the animals were determined weekly using a suspension balance. Blood samples were collected via the jugular veins of the animals before and after parturition for the analysis of some minerals and enzymes levels in the serum.

DATA ANALYSIS

The results obtained from the animals were compared with standards obtained from literatures and subjected to standard deviation and students 't' test.

RESULT

The weight changes in the West African Dwarf does before and aftrer feed supplementation are presented in Table 1.

The result showed that there is a significant increase in the body weight of all the animals after been fed with a corn-based concentrate for two weeks ($P<0.05$). At the end of the study (after parturition) mean body weight of all the animal is 40.29± 3.32 as compared with 16.14 ± 1.95 which was the mean body weight of the animals before feed supplementation i.e at service. The animals had gained an average body weight of 3.97 ± 2.14.

Table 1 shows the results of the analysis of the serum for some liver enzymes namely glutamic pyruvic Ttansaminase (GPT), alkaline phosphatase (ALP) and glutamic oxaloacetic transaminase (GOT). The result showed that the values of all the enzymes were lower just before parturition. The values increased significantly ($P<0.05$) after parturition. It is showed that GOT
34.67± 3.712 < 150.30 ± 22.40, ALP 106.67 ± 4.67 < 261.30 ± 53.50 and GPT
25.30 ± 10.50 < 91.00 ± 32.00.

The results of the serum minerals before and after parturition is presented in Table 1. The results show that there is no significant increase in the levels of the serum minerals just before and after parturition ($P>0.05$).

Table 1. Weight of does, serum metal lons and serum metal lons before and after parturition.

Serum liver enzymes	Before Parturition (Mean ± SEM)	After Parturition (Mean ± SEM)
Weight	16.14 ± 1.95	20.40 ± 3.2
GOT	34.67 ± 3.71	150.30 ± 22.40
ALP	106.67 ± 4.67	* 261.30 ± 52.50
GPT	25.30 ± 10.50	91.00 ± 32.00
Na^+	141.00 ± 1.53	135.67 ± 1.20
Ca^{++}	4.33 ± 0.58	5.13 ± 1.26
K^+	8.87 ± 0.09	8.37 ± 0.50

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DISCUSSION

The changes in live weight at service and parturition indicate that concentrate supplementation in this study has a positive effect on the weight of the West African dwarf (WAD) goats. These is also an evidence of good reproductive performance in the goats since there was no abortion or still birth in the course of the study. The agrees with previous studies by Akusu (1987) and Osuagwuh (1991). They found out that without that feed supplementation, pregnant does may either abort or give birth to weak kids.

Glutamic pyruvic transaminase (GPT), alkaline phosphatase (ALP) and glutamic oxaloacetic transaminase (GOT) are liver enzymes found in the serum. They are usually nonfunctional plasma enzymes because they perform no physiologic function in the blood. Their substrates frequently are absent from plasma. Their presence in plasma at levels elevated above normal values suggests an increased rate of tissue destruction. Low levels of nonfunctional enzymes found ordinarily in plasma arise apparently from the routine normal destruction of erythrocytes, leukocytes and other cells.

From the results there was a significant increase in the level of the liver enzymes just before parturition and after parturition. Despite the increase, the level of the GOT and ALP still fall within the normal range reported by Boyd (1962) and Wroblewski and LaDue, (1955). Hoe and O'shea, (1965) and Kaneko (1989) has also reported a slightly elevated level of alkaline phosphatase in pregnant, sheep, goat and cow. It is evident from the result that there is no abnormal increase in the level of all these serum enzymes. The goes on to confirm that there is no case of liver disease in all the animals. Since both GOT and ALP levels in the serum of animals can be used as a determinant of liver malfunctioning, increase in both enzymes has been associated with liver diseases. Significant elevations of serum GOT have also been observed in muscular dystrophies of nearly all animal species (Cornelius et al., 1959).

The increase in the level of GPT in the study might be due to secondary hepatic necrosis, and parturient haemorhage (Thorpe et al., 1968 and Benjamin, 1979). How (1961) also reported increase in GPT in cases of excessive bleeding and serious uterine infection post-partum. Therefore where there is doubt of parturition in animal serum GPT can reveal the status of such an animal.

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EFFECT OF ADDITION OF SODIUM BICARBONATE TO 'DUSA' A CONCENTRATE SUPPLEMENT, ON FEED INTAKE AND LIVEWEIGHT RESPONSES OF SHEEP FED TWO ROUGHAGE TYPES.

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ABSTRACT

Twenty four Yankasa rams with initial average weight of 16.55kg and aged about 12 months were grouped into six groups of four rams per group and each group balanced for weight, thereafter randomly assigned to one of the six experimental feeds. These were: Untreated maize husk (UMH) (20g/kg BW) which served as control for UMH roughage; UMH (20g/kg BW plus sodium bicarbonate (SB/Dusa (DS) mixture (20g/kg BW + 2g/kg BW); UMH plus DS (20g/kg BW + 20g/kg BW; untreated rice straw (URS) (20g/kg BW) which also served as control for URS roughage; URS plus DS/SB mix (20g/kg BW + 2-g/kg + 20g/kg BW mix); URS plus DS (20g/kg BW + 20g/kg BW). Animals were fed DS as concentrate while SB was used as additive.

Feeds offered and rejected were weighed daily while the animals were weighed weekly. The experiment started by mid November, 1995 and lasted 63 days. Data generated were analysed using missing data technique as some rams died during the experiment. The data were subjected to Analysis of variance and the treatment means tested using Duncan Multiple Range test.

Results showed that intake of UMH varied from 66.96 g d^{-1} to 85.38 g d^{-1} while that of URS varied from 78.38 d^{-1} to 94.22 g d^{-1}. There was no significant difference between intake of SB/DS mix in T-2 and T-5 and also between intake of DS alone in T-3 and T-6 respectively (P>0.01). The total intake for T-1 to T-3 were 28.09g/kg $^{wo.75}$; 40.47g/kg $^{wo.75}$ and 71.9g/kg $^{wo.75}$ while that of T-4 to T-b were 26.35g/kg $^{wo.75}$; 39.69g/kg $^{wo.75}$ and 81.98g/kg $^{wo.75}$ respectively. T-1 and T-4 lost weight while the rest treatments that had supplement gained weight.

It may be concluded from these results that addition of $NaHCO_3$ to animal feed may substantially enhance the efficiency of rumen digestion.

Crop residues are a major basal roughage for livestock but their utilization is limited by low digestibility and nutrient content which inhibit intake and productivity. Appropriate supplementation is a means that has been used to enhance intake and utilization of crop residues and the consequent productivity of livestock (Greenhalgh, 1980).

The addition of bicarbonates such as sodium bicarbonate (SB) into the feed of ruminants has been known to enhance productivity (Solvay, 1983). Experiments have been conducted especially in developed countries in the use of SB in ruminant diet (Orsbourn et al., 1970, Emmanuel et al., 1970; Mould et al., 1983; Kellaway et al., 1973; and Kellaway et al., 1977). It is recognised that rumen fermentation is impaired and animal performance lowered when the nitrogen content of the diet is less than 1.2% (Conrad and Hibbs, 1968). The feeding of energy and protein supplements is known to enhance the utilisation of poor quality feeds like crop residues such as rice straw and maize husk by maximizing roughage degradation and optimizing rumen microbial protein synthesis (Anderson, 1978; O'Donovan, 1983). Because of high cost, scarcity and other logistic problems, the use of concentrates such as cotton seed cake and soya bean meal cannot be justified especially in Nigeria, where these commodities are virtually out of the reach of peasant farmers. It has therefore become imperative to look inward for some other substitutes that are relatively cheaper, easily available and within reach of the livestock producers. In Nigeria, cereal milling waste locally called 'Dusa' is widely used throughout the year, especially during the dry season when used judiciously with available roughage types such as untreated maize husk (UMH) or untreated rice straw (URS). This could further be enhanced with the inclusion of sodium bicarbonate (SB) as an additive. In the Northern Guinea Savannah vegetational zone of Nigeria where this experiment was conducted, the inclusion of SB in ruminant diet is yet to be exploited. In this zone, UMH and URS form a large percentage of ruminant basal diet.

The objective of this study was to investigate the effect of SB on the intake and weight responses of yearling rams fed a basal diet of UMH or URS and Digitaria smutsii using DS as a concentrate supplement.

MATERIALS AND METHODS

Background of research station

The study was conducted at the National Animal Production Research Institute of the Ahmadu Bello University, Shika, Zaria, Nigeria. Shika lies between latitudes 11 and 12^0N, and between longitudes 7 and 8^0E and has an altitude of 64m above sea level. Shika is situated within the Northern Guinea Savannah zone and has an

(*Ocimum basilicum* L.) and osage orange (*Maclura pomifera*, Rob.), as well as their pure constituents for antimutagenic potential against UV-and EtBr-induced mutations. Antigenotoxic potentials are estimated by applying prokaryotic and eukaryotic tests. Results obtained by our *E.coli* assay system, designed and validated to detect bioantimutagens and their mechanisms of action, are confirmed with Ames test (*Salmonella typhimurium*) and eukaryotic test (*Saccharomyces cerevisiae*). The most significant result indicates that monoterpenoids from cultivated sage inhibit UV-induced mutagenesis by modulating DNA repair pathways. Antioxidants, an integral part of plant extracts, evidenced in sage fractions with high content of diterpenoids, in extract of osage orange and its pure constituent pomiferin, significantly inhibited EtBr-induced mutagenesis, probably by inhibiting metabolic activation of promutagen. Preliminary experiments indicate that ethereal oil of sage suppressed in vivo mytomicine C-induced chromosome aberration in mice. Further study will show whether the plant antimutagens are useful as anticarcinogens.

O7-7 Pseudouridine, an antimutagenic component in beer toward N-methyl-N'-nitro-N-nitrosoguanidine and N-methyl-N-nitrosourea

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It is important to identify food and drinks which could protect against cancer. We have been investigating antimutagenic properties of beer. Previously, we reported that beer is antimutagenic against several food-derived mutagens including heterocyclic amines. We describe here the isolation and identification of pseudouridine from beer as an antimutagenic substance against N-methyl-N'-nitro-N-nitrosoguanidine (MNNG). All of the 17 samples of beer tested showed inhibition of the MNNG mutagenicity in Sa lmonella. Extensive fractionations through chromatographies of the active components from a freeze-dried beer sample gave several antimutagenic fractions. One of them has been revealed to contain pseudouridine, as characterized by UV, NMR, and co-chromatography in HPLC. Authentic pseudouridine inhibited the mutagenicity of MNNG in a dose-dependent manner. Another methylating agent N-methyl-N-nitrosourea was also inhibited by the presence of pseudouridine. The amount of pseudouridine in the beer sam p l e, estimated at about 0.4 mg/100 ml beer, can account for only a few percent of the total antimutagenicity of beer. Thus, the major active components in beer remain to be identified. A search for a similar antimutagenicity among pseudouridine analogs s howed that spongouridine, but not uridine, is antimutagenic against MNNG. Pseudouridine is the first example among nucleosides to be shown to possess an a nti mutagenic property.

O7-8 Inhibition of aflatoxin B1-induced clastogenicity and hepatocarcinogenicity by kolaviron (*Garcinia biflavanones*) in rats

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The effect of kolaviron, an antioxidant [1] and hepatoprotective [2] *Garcinia* biflavanones from *Garcinia* kola seeds cultivated widely in West Africa, on aflatoxin B_1 (AFB_1)-induced carcinogenicity and induction of micronuclei in rat peripheral blood was investigated. The frequency of micro nucleated reticulocytes peaked at 12 h after a single i.p treatment of rat with AFB_1 at a dose of 2 mg/kg. Administration of kolaviron at a dose of 200mg/kg for 3 days consecutively inhibited micronucleus induction by AFB_1. Similarly, GSH, cysteine and ascorbate at doses of 800mg/kg, 400mg/kg and 200mg/kg administered at different times before treatment with AFB_1 mitigated micronucleus induction by AFB_1. AFB_1 induced the activity of γ-glutamyl transferase and alanine aminotransferase (markers of hepatic damage) significantly. Histological examinations of the liver slices correlated with the changes in the activities of these enzymes. Kolaviron attenuated the AFB_1-induced elevation in the activities of these enzymes ($P<0.001$). Kolaviron also inhibited the AFB_1-induced formation of malondialdehyde and lipid soluble fluorophores at a dose of 100mg/kg body weight ($P<0.01$). The results suggest that kolaviron like other antioxidants (GSH, cysteine and ascorbate) protects against the clastogenicity and carcinogenicity of AFB_1 by either inhibition of reactive oxygen species from AFB_1 and /or elimination of active oxygen and other reactive metabolites.

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O7-9 Chemoprotective effects of resveratrol against oxidative cell death and DNA damage

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Oxidative stress induced by reactive oxygen species (ROS) has been implicated in a wide variety of clinical abnormalities and disorders. Recent studies have revealed that ROS can cause cell death via apoptosis. Resveratrol (3,4',5-trihydroxy-stilbene), an antioxidant found in grapes, has been considered to be responsible in part for the protective properties of red wine against coronary heart disease. In this study, we have investigated the effects of resveratrol on oxidative DNA damage and cell death induced by hydrogen peroxide or beta-amyloid peptide. Thus, the compound inhibited the strand scission in ϕ174 RFI supercoiled DNA induced by hydrogen peroxide in the presence of transition metal ion. In another experiment, PC12 cells treated with hydrogen peroxide underwent apoptotic death as determined by morphological features, internucleosomal DNA fragmentation and positive *in situ* terminal end-labeling (TUNEL staining). Resveratrol pretreatment attenuated hydrogen peroxide-induced oxidative cell death. Likewise, beta-amyloid peptide-induced apoptosis and intracellular accumulation of reactive oxygen species were inhibited by resveratrol. Resveratrol mitigated the NF-κB activation transiently induced by hydrogen peroxide or beta-amyloid in PC12 cells. Resveratrol also inhibited TCDD-induced expression of cytochrome P450 1A1 and 1B1 in human breast epithelial (MCF10A) cells which are known to catalyze the hydroxylation of 17-beta estradiol at C-2 and C-4 positions, respectively. Since the resulting catechol estrogens can undergo redox cycling to produce reactive oxygen species, the blockade of their formation by resveratrol through inhibition of the corresponding P450 isoforms may provide another mechanism underlying chemoprotective effects of this phytochemical against oxidative cell death.

O7-10 The relationship between nitric oxide (NO) concentrations and regulation of cyclo-oxygenase (COX-2) expression by soy isoflavones

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Plasma NO concentrations and expression of cyclooxygenase in spontaneously hypertensive rats (SHRs) fed formulated mixture of soy isoflavone glycosides (genistein 4.0%, daidzein 15.3%, glycitein 11.9%; 10g/kg diet) in a diet for 30 days were investigated. During the 30-day study period, tail systolic blood pressures in control SHR group increased significantly from 162.4 ± 2.3 to 177.9 ± 5.4 mmHg ($p < 0.05$), whereas isoflavone supplemented group had a marked antihypertensive effect (160.1 ± 1.8 to 160.2 ± 4.9 mmHg). The plasma concentration of NO was significantly elevated in the isoflavone group compared to that of the control group (48.4 ± 8.9 vs 29.8 ± 2.0 micro mol, $p < 0.05$). Feeding isoflavones resulted in increased concentration of NO in plasma, which was accompanied by a significant decrease in tail systolic blood pressure compared to the control group. Therefore, vasodilating effect of isoflavones through NO seems to be responsible for antihypertensive action of isoflavones in SHRs. Recently it has been shown that cyclooxygenase-2 is involved in vasodilating effect of estrogen. Western blot analysis of COX-2 expression in heart of isoflavone fed animals showed that COX-2 is also involved in vasodilating effect of isoflavones. The regulation of COX-2 by NO will be discussed in cell culture system.

O7-11 Antimutagens from Philippine medicinal plants

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An antimutagen was isolated from the leaves of Carmona retusa (Vahl)Masam. Results of the micronucleus test that, at a dosage of 24.4 mg / kgmouse, the pure isolate reduced by 68.4% the number of micronucleated polychromatic erythrocyte (MN-PCE) induced by the mutagen tetracycline. Its structure was elucidated to be 4-hydroxy-7,8,11,12,15,7',8',11',12',15'-decahydro-κ, Ψ-carotene.
Another antimutagen was isolated from the flowers of Cucurbita maxima Duchesne. At a dosage of 100 mg / kg mouse, the isolate decreased the mutagenicity of tetracycline by 64.7% using the micronucleus test. Spectral analysis showed that the antimutagen is 24-α-ethyl-5α-cholesta-7,*trans*-22-dien-3β-ol or spinasterol. At a concentration of 15μg / mL, spinasterol also decreased the incidence of skin tumors by 55.6% and decreased the number of tumors by 65.0% when applied immediately after croton oil. β-Sitosterol was isolated as an antimutagen from the leaves of Mentha cordifolia Opiz, while its glucoside was isolated as an antimutagen from Cassia alata Linn. At a dosage of 0.5 mg / kg mouse, β-sitosterol inhibited the mutagenicity of tetracycline by 65.3%. At the same dosage, it did not exhibit chromosome-breaking activity on normal mice. β-Sitosteryl-β-D-glucoside also exhibited a 79.4% antimutagenic activity at a dosage of 75 mg / kg mouse.

O7-12 6-Formylindolo[3,2-*b*]carbazole reduces the DNA-adduct levels and the genotoxic effects of benzo[*a*]pyrene *in vitro* and *in vivo*

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Light exposure of tryptophan solutions, including cell culture media, gives rise to tryptophan photoproducts, some of which have very high Ah-receptor affinity and are potent inducers of cytochrome P450 1A1 (CYP1A1). We have earlier shown that two of the most active photoproducts, 6-formyl- and 6,12-diformylindolo[3,2-b]carbazole were inhibitors of the cytochrome P450-dependent mutagenicity of benzo[a]pyrene (BaP) in Ames *Salmonella* assay and of the CYP1A1-dependent mutagenicity caused by benzo[a]p yrene-trans(-)7,8-dihydrodiol (BPD) in Chinese hamster cells expressing rat CYP1A1 (XEM2-cells) (Rannug et al. Env Mol Mutagen, 20, 1992, 289). In the present study further mutagenicity tests were carried out in XEM2 cells treated with BPD in the presenc e and absence of the photoproduct 6-formylindolo[3,2-b]carbazole (FICZ). The DNA adduct levels were analyzed by means of ^{32}P-postlabelling. Parallel to the reduction in mutagenicity DNA-adduct levels were reduced in the presence of FICZ. A 10 w e eks treatment of transgenic mice (XPA-/-, p53+/-) with BaP±FICZ was carried out. The DNA-adduct levels in liver, lung and spleen were determined. As a measure of chromosome damage, the frequency of micronuclei (MN) in polychromatic erythrocytes was also determined. With BaP, the DNA-adduct levels increased in all organs with time and the combined BaP/FICZ treatment resulted in lower adduct levels. In the BaP treated groups, MN frequencies were significantly elevated over the control groups. The BaP/FICZ treated groups showed lower MN frequencies, which were not significantly different from the control level. Consequently, the antimutagenic activity seen in vitro was confirmed in vivo.

O7-13 Chemoprevention against dietary mutagens by induction of phase II enzymes and by ingestion of Brassica vegetables in humans

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To elucidate the impact of consumption of Brassica vegetables on detoxifying enzymes in humans two series of intervention studies were carried out. In the first series induction of glutathione *S*-transferases (GST) was investigated with 5 different Brassica varieties. Participants (n=10) received 300g cooked vegetables for 5 days. Average GST activity in plasma was induced 1.8 fold with red cabbage and 1.7 fold with Brussels sprouts. White cabbage and broccoli were ineffective. ELISA tests showed that the GSTα isoenzyme was not affected by the diets whereas the GSTπ isoenzyme was consistently enhanced .The impact of food processing was elucidated and it was found that cooked Brassica vegetables were more effective than uncooked material. No correlations between GST induction and gender or GST genotypes could be seen. In the second experimental series meat (beef, chicken) derived urinary mutagenicity was determined in reversion assays with a heterocyclic amine sensitive bacterial strain (*S. typhimurium* 1024). A clear-cut decrease in meat induced urinary mutagenicity after consumption of red cabbage and Brussels sprouts was observed. These phenomena are probably due to induction of glucuronidation, a major pathway in the detoxification of heterocyclic amines. Overall, our findings suggest that Brassicas protect humans against mutagens since impaired GSTπ is associated with certain cancers in humans and recent animal studies show that enhanced glucuronidation is paralleled with protection against cancer inducing heterocyclic amines.

O7-14 Prevention of AOM-induced colon cancer by lemongrass

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The possible anticancer activity of lemongrass has been shown; i.e. significantly inhibition of chemical-induced mutagenesis in Salmonella typhimurium, antimetastatic effect on fibrosarcoma transplantable mice. We have reported the increase in activities of xenobiotic phase II metabolizing enzymes such as, glutathione-S-transferase, quinone reductase and UDP-glucuronosyl transferase in liver and small intestine of rats which were fed with lemongrass extract supplemented to diet.This investigation, crude ethanolic extract of lemongrass was sequentially partitioned according to polarity into 4 fractions, the first hexane-soluble, the second ethyl acetate-soluble, the third butanol-soluble and the fourth ethanolic-soluble fractions. The hexane-soluble fraction appeared to be the most interesting because it had the strongest antimutagenicity and was the most potent DT-diaphorase inducer. The hexane-soluble fraction was studied for its inhibitory effects on azoxymethane (AOM)-induced ACF formation in Wistar rats. Rats on the hexane-soluble fraction diet before AOM treatment were found significantly to have fewer colonic aberrant crypt foci than AOM treated rats. Administration of the fraction supplemented to the diet significantly