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THEME:

**THE ROLE OF
HORTICULTURE IN FOOD
SECURITY AND SUSTAINABLE HEALTH
IN ERA OF CLIMATE CHANGE AND
COVID-19 PANDEMIC**

 14TH - 18TH NOVEMBER, 2021

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THEME: The Role of Horticulture in Food Security and Sustainable Health in Era of Climate Change and Covid-19 Pandemic



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EFFECT OF PLANT GROWTH HORMONE (BIOGRO) ON BEHAVIOR AND SERUM BIOCHEMISTRY OF WISTAR RAT

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ABSTRACT

With the purpose of studying data on spontaneous customary changes in wistar rats and effect of biogro (bio fertilizer) on organ enzymes (serum biochemistry), we administered food laced with dried okra treated with the biogro. This study was carried out at three different places which are the animal experimental house, water hyacinth, National Horticultural Research Institute (NIHORT) Ibadan, Oyo state, Nigeria, the Clinical pathology laboratory, University of Ibadan, Nigeria and the histopathology laboratory at the Department of Veterinary Pathology, Faculty of Veterinary Medicine, University of Ibadan, Nigeria. A total of 32 wistar rats were used for the experiment, with the rats divided into eight groups. Four rats occupy a unit/cell of the cage, making it eight treatments replicated four times. The experiment was laid out in complete randomized design (CRD) and data were analyzed with one-way analysis of variance (ANOVA). The biogro treatments (treated on okra plants) were administered daily as it was included in the feed given for five days (acute toxicity test). On the termination of the experiment, all the rats were observed not to have any behavioral changes as they all appear and were normally behaved, and sacrificed, blood samples were taken for analysis. At the end of the experiment it was observed that the treatment had no significant effect on both the blood parameter AST = Aspartate amino transferase, TB = Total bilirubin, EO = Eosine, ALP = Alkaline Phosphate, ALT = Alanine amino transferase, TP = Total protein, ALB= Albumin, GLB = Globulin.

Key words: Biogro, serum biochemistry, wistar rats, plant hormone

INTRODUCTION

Serum biochemical assessments are relevant tools in evaluating the physiological and pathological status of mammals and birds, as they provide information for the proper diagnosis of diseases, making a prognosis, evaluating the efficacy of instituted therapy, and toxicity of drugs and chemical substances (Stockham and Scott, 2008). Serum biochemical assessment helps to predict pathological processes in the vital internal organs of the body such as the liver, muscle, heart, pancreas and kidney (Stockham and Scott 2008). It also helps to establish the presence or absence of disease of an organ, and determine the nature and extent of a disease process (static, progressive or regressive) by serial performance of laboratory tests of the internal organs.

Several physiological factors have been known to affect the serum biochemistry parameters in apparently healthy rats. These factors are breed, sex, age, reproductive status; time of feeding, diurnal variations, nutritional state and management; and

geographical/climatic factors such as temperature, humidity, altitude and day length (Weiss and Wardrop, 2010). Alterations in serum biochemistry values can be attributed to poor sample handling, stress during blood sample collection, blood sampling techniques, presence of an inhibitor such as anticoagulant and stress of capture (Stockham and Scott, 2008).

In available literature, there are reasonable amount of haematological and serum biochemistry values of rodents such as the birch mouse (Wolk 1985), juvenile laboratory rat (Ihedioha et al., 2004), African soft-furred rat (Kagira et al., 2005), wild grasscutter (Opara et al., 2006) and adult wild African giant rat (*C. gambianus*) (Oyewale et al., 1998a, 1998b; Olayemi and Adeshina, 2002; Cooper and Erlwanger, 2007), but only little information is available on the haematology and serum biochemistry values of the juvenile wild African giant rat (Nssien et al., 2002; Onwuka et al., 2003). Due to the variations in the haematological and serum biochemistry values,

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there is need for each clinic to establish its own reference range of values for relevant animal population. Also, due to the popularity of this rodent, efforts have been made to domesticate and keep them as pet, and attempts have also been made to use them as laboratory animals (Olayemi and Adeshina 2002).

Justification

Agrochemicals or even plant hormones such as fertilizing elements are essential to boost agricultural/horticultural production in Nigeria. The usual practice is to recommend "appropriate" doze due to observed increase in yield. However, there is the need to continue to monitor the residual effects of these agrochemicals or plant hormones so as to ensure they have no residual effect or at least, that the residuals are within safe limits recommended by regulatory authorities such as WHO and FAO. Hence, our recommendations will no longer be based on quantity (yield) only but also on food quality. The objectives of the study are: 1) To observe treated animal for behavioral changes and 2) To investigate the serum biochemistry of treated animal for possible effect.

MATERIALS AND METHODS

This study was carried out at the animal experimental house, water hyacinth National Horticultural Research Institute Ibadan Oyo state, Clinical pathology laboratory, University of Ibadan and Histopathology laboratory of the Department of Veterinary Pathology, Faculty of Veterinary Medicine, and University of Ibadan, Nigeria.

A total of 32 wistar rats were used. The rats were divided into eight groups (treatments) with four rats occupying a unit/cell of the cage, making it eight treatments replicated four times and the rats were acclimatized for one week before commencement of treatments. Feed and water were given to the animals during acclimatization ad-libitum. The biopro treatments were administered daily as it was included in the feed given for five days (acute toxicity test). On the termination of the experiment, all the rats were sacrificed and organs of interest were carefully removed through cervical dislocation for diagnosis. These include the liver and kidney for

histopathological examination. The experiment was laid out in completely randomized design (CRD) and data were analyzed with one-way analysis of variance (ANOVA), group means were compared for significance at 95% confidence level (P 0.05) by Duncan's Multiple Range Test (DMRT) from SAS 2003.

Blood Sample Collection

Blood samples for the serum biochemical determinations were collected from the orbital sinus of the 32 wistar rats following the procedure of Stone (1954). These samples were collected between the hours of 8 and 11 in the morning on the same day. Blood samples (1 ml) for haematology were collected into sample bottles containing ethylene diamine tetra acetic acid (1 mg/ml of blood). For serum biochemical analysis, 3 ml of blood was put into plain glass tubes and sera harvested within 1 h after centrifuging the clotted blood with a clinical table centrifuge at 3000 rpm for 15 min. The haematological and serum biochemical analyses were carried out immediately upon blood collection.

Biochemical Parameters

Serum samples were analyzed for the activities of aspartate aminotransferase (AST), alanine aminotransferase (ALT) and for the concentrations of total protein, albumin, cholesterol, total and direct bilirubin, alkaline phosphate and urea by the Roche Diagnostic Hitachi 902 Analyzer.

Haematological and serum biochemistry procedures Haematological and serum biochemistry determinations were carried out following standard procedures. PCV was determined by the micro-haematocrit method (Thrall and Weiser, 2002), Hbc was determined by the cyanomethaemoglobin method (Higgins et al. 2008). RBC count and total leukocyte count (TLC) were carried out by the haemocytometer method (Thrall and Weiser, 2002), differential leukocyte count was done by making a blood smear on a clean glass slide and staining it following the Leishman technique. The different cells of the leukocytic series were enumerated by the battlement counting method (Thrall and Weiser, 2002). The mean corpuscular values – mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular



haemoglobin concentration (MCHC) – was calculated using the standard formulae (Coles, 1986). Serum biochemistry determinations were carried out using commercial test kits, Quimica Clinica Aplicada (QCA) test kits (QCA, Spain), Randox test kits (Randox, UK) for total proteins and albumin, and a digital colorimeter. Serum ALT and AST activities were determined by the Reitman–Frankel method (Reitman and Frankel, 1957). Serum ALP activity was determined by the phenolphthalein monophosphate method (Babson et al., 1966), while total serum proteins were determined by the direct Biuret method

(Lubran, 1978). Serum albumin was determined by the Bromocresol green method (Doumas et al., 1971). Serum globulin was calculated as the difference between serum total proteins and serum albumin (Colville, 2002), while serum total bilirubin was determined by the Jendrassik–Grof method (Doumas et al., 1973). BUN was determined by the Berthelot–Searcy method (Fawcett and Scott, 1960), while serum creatinine was determined by the modified Jaffe method (Blass et al., 1974). Serum cholesterol was determined by the enzymatic colorimetric method (Allain et al., 1974).

Table 1. Effects of treatments on serum biochemical parameters of wistar rats

Treatment	ALT (U/L)	ALB(G/DL)	GLB (G/DL)	TP (x10 ⁶ /mm ³)	AST (U/L)	EOS (x10 ⁶ /mm ³)	ALP (x10 ⁶ /mm ³)
T1	7.63a	1.93b	4.33a	0.63a	34.33ab	2.00a	25.67c
T2	7.27a	2.63ab	4.33a	0.57a	37.00ab	1.67a	25.33c
T3	7.37a	2.83ab	19.50a	0.53a	38.00ab	2.00a	32.33ab
T4	7.43a	2.60ab	4.30a	0.60a	40.00ab	1.33a	33.0a
T5	6.57c	2.57ab	4.37a	0.63a	30.07ab	1.67a	33.33a
T6	7.20a	2.77ab	4.47a	0.57a	25.00b	2.33a	26.33bc
T7	7.70a	2.97a	4.40a	0.43a	35.00ab	2.00a	27.33abc
T8	8.37a	3.43a	5.43a	0.40a	44.00a	2.00a	32.00ab

Means within the same column followed by the same letter(s) are not significantly different at P < 0.05 using DMRT

AST = Aspartate amino transferase, TB = Total bilirubin, EO = Eosine, ALP = Alkaline Phosphate, ALT = Alanine amino transferase, TP = Total protein, ALB= Albumin, GLB = Globulin

RESULTS AND DISCUSSION

There was significant difference in Aspartate amino transferase (AST) of animals treated with biogro and biofart, while there was no significance difference from observation made among the other treatments (total protein, Eosine, Globulene and Alkaline Phosphate) and the control. However, the rats treated with T5 recorded highest means value while the lowest mean value was obtained from T2. There were no significant differences in ALT, total protein, globulin and eosin in all the treatments compared with the control but T8 was highest in mean value and lowest in the control with respect to ALT. In Total protein, control had highest mean value while T8 recorded lowest mean value, and in Eosin the highest mean value was observed in animals treated with T6 and lowest in T4 as seen in Table 1.

The results showed that there was significant difference in ALP between treatments, highest in mean value was observed control while T2 recorded lowest mean value. There was significant difference in the control and treatments for albumin where T8 had the highest mean value and treatment with T1 had the lowest mean value from Table 1.

All rats treated with biogro showed a marked increase in body weight gain and the rats showed no signs of poisoning and this is in agreement with the work of Mansour et al. (2007) that there were no signs of Toxicity noted in rats treated with plant growth hormones.

The trend in lymphocytes revealed highest mean value in animals treated with T2 and lowest in T7 as shown in Table 1, the normality found in serum levels of protein for all the rats dosed at all dosages of



treatment shows that the treatment is not toxic to the organs or health of the animals as reported in the findings of Das and Mukherjee (2000) that toxicants may cause stress-mediated mobilization of protein to cope with the detrimental condition so imposed. The protein mobilized is one of the strategies employed to meet the energy required to sustain increased physical activity, biotransformation and excretion of the toxicants.

Amino transferases (ALT & AST) are produced in the liver and are good markers of damage to liver cells but not necessarily the severity of the damage as reported by (Olav et al., 2007; Rej, 1989). The non-significant levels of EOS, TP, GLB and ALT in treated groups could be as a result of suppression of production by the liver which is in agreement with Obaineh and Mathew (2009) that they are normally present at low levels in the blood so if the liver cells are damaged, it would be expected that some of the enzymes leak into the blood and increase the levels.

Decrease levels of albumin may be due primarily to reduction in synthesis by the liver and secondarily to reduced protein intake which further confirms no hepatic damage and agree with the works of Luskova et al. (2002) and Jyotsna et al. (2003) as observed in WBC in the study.

In contrast to serum enzymes, albumin apart from being a useful indicator of the integrity of glomerular membrane is also an important indicator in determining the severity of disease as also stated by Lawrence and Amadeo (1989) and (Mukut et al. (2001). In the histopathological evaluation of the liver and kidney of rats treated with biofarm and biogro showed no injury as there was no visible lesion observed neither was any severe damage in terms of necrosis observed in any of the organs of the treated animals and this is in agreement with the work of McCormack (2011). In the various treatments, the only observable change is an increased vascular congestion ranging from mild to moderate. The treatments do not have a structural change on the tubules and other anatomic structures in the kidney. The toxicity effects of treatments are not acute as the clinical symptoms did not manifest in test animals.

CONCLUSION AND RECOMMENDATION

The report above has shown that the treatments had no significant effect on the behavior, serum biochemistry and on the health of the test animals hence, it is safe for use at the recommended dosage. It is hereby recommended based on this finding that the test product is safe for use at the recommended standard dosage.

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