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RESPONSE OF ADMINISTRATION OF PANICUM MAXIMUM ETHANOL LEAF EXTRACT ON TISSUE WEIGHT GAIN, BIOCHEMICAL AND HAEMATOLOGICAL INDICES IN RABBITS

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ABSTRACT
This study evaluated the response of administration of graded doses of *Panicum maximum* ethanol leaf extract on tissue weight gain, biochemical and haematological indices in rabbits. There were Twelve (12) rabbits divided into four (4) groups of three (3) per group. Group 1, served as the control which received distilled water. Groups 2, 3, and 4 were the treatment groups which received the graded doses of *P. maximum* at 300, 600 and 900 mg/kg body weight respectively and were treated for 14 days *per os*. At the end of dosing period, blood samples were collected for haematology and serum biochemistry assay. Organs (heart, lungs, liver, kidney) were harvested for relation organ weight. Data obtained was analysed using SPSS expressed as mean ± Standard error of mean. The result showed a consistent increase in organ weight as the doses of *P. maximum* extract increased. The dressed weight, thigh weight, breast, shoulder, back cut, drum stick and fore arm weights significantly increased compared with the untreated as the dose of the extract increased. PCV and RBC decreased while the TWBC increased but within normal reference range. There was mild hyperproteinaemia, mild hyperbilirubinaemia and moderate hypercholesteremia with elevated triglyceride from 47.47±1.59 to 79.23± 0.54. AST, ALT levels increased but were within normal reference range. ALP decrease was within normal reference range indicating safety of the organ. Bilirubin, urea and creatinine did not deviate from reference range, indicating kidney safety. Whole triglycerides remained at normal reference range. This shows that *P. maximum* is a safe grass for animals.

KEYWORD: Haematology, *Panacum ma ximum*, Rabbit, Serum Biochemistry, Weight gain.

INTRODUCTION
Panacum. maximum Jacq. commonly known as Guinea grass is one of the most important fodder and pasture species (Bogdan, 1977). As a cultivated grass, it is much valued for its good persistence (Ng *et al.*, 1977). It is one of the most successful weeds (Batianoff and Andrew, 1998) and also a good colonizer (Boonman, 1993; Martinez *et al*., 1997). *P. maximum* is a monocot belonging to the family Poaceae (grass family) *P. maximum* is widespread throughout Southern Nigeria, on roadsides, waste places and in the wetter parts of the North (Stanfield, 1970). It is a very variable species in East Africa and numerous natural types exist, some of which have been described as botanical varieties. *P. maximum* is a perennial, tufted grass with a short, creeping rhizome. The stems of this robust grass can reach a height of up to 2 m. As the stems bend and nodes touch the ground, roots and new plants are formed. The leaf sheaths are found at the bases of the stems and are covered in fine hairs. It remains green until late into winter. The leaf blades are up to 35 mm wide and taper to a long fine point. The inflorescence is a large multi-branched, open panicle with loose, flexure branches. Figure 1 shows the picture of *P. maximum.*

Fig. 1: Picture of *panacum maximum.*

P. maximum is one of the nutritious forage and pasture grass in the tropics (Kanife, 2011). The anti-diabetic and antibacterial activities of ethanol leaf extract have been reported. (Doss 2011, Ajoku 2015). In West Africa, extract form leaf has been used in curing various diseases such as malaria, infections, rheumatism pain, inflammation and diabetes, traditionally (Doss 2011, Ajoku 2015). The anti-diabetic, antiplasmodial and analgesic activities of the extract have also been reported (Antia, 2010). The ethanolic leaf extract possess anti-inflammatory and antipyretic properties (Okokon, 2011). The antibacterial activity of *P. maximum* leaf on selected bacterial strains compared well with the standard drug like ciprofloxacin. *P. maximum* popularly referred to as *P. maximum* is native to African countries such as Kenya, Ethiopia, Cameroon, Cote D'Ivoire, Ghana, Guinea, Liberia, Nigeria, Sierra Leone, Angola, Malawi, Mozambique, Tanzania, Togo, Uganda, Zambia and Zimbabwe. It is a robust perennial with an extensive root system reaching a depth of 4.5m. The plant has been used extensively in livestock nutrition and has a huge potential as a feedstock for bio fuel processing. The grass yield is highest in dry matter and is better than most tropical grasses. Phytochemical screening of *P. maximum* has been carried out by researchers using Gas Chromatography-Mass Spectrometry Igwe *et al* (2016; 2016a) This analytical method is one of the valuable methods used by many researchers to identify plant compounds (Igwe *et al* 2020; 2020a). This work is therefore aimed at evaluating the weight gain, haematology and serum enzymes as biochemical markers. Rabbit was used as the animal model.

MATERIALS AND METHODS

Collection Material

Fresh leaves of *P. maximum* were sourced from Michael Okpara University of Agriculture, Umudike in Abia State, Nigeria. The leaves were identified using Google plant identifier and was confirmed in the Department of Forestry and Environmental Management of the same University.

Plant Materials and Extraction

Identified leaves were ground using mechanical grinding into coarse powder, soaked in ethanol and was left for 48 hours. Cold maceration was used which involved soaking the plant sample in ethanol for 48 hours with constant agitation every two hours. Filtered with Whatman filter paper and was concentrated using hot-air oven at 30^0C then refrigerated at 4^oC for the experiment.

Experimental design

Twelve (12) male rabbits were divided into four (4) groups of three (3) per group kept in different cages. They were treated for 14 days. Group 1 (Control) received distilled water. Group 2 (Low dose) received 300mg/kg body weight of plant extract. Group 3 (Medium dose) received 600mg/kg body weight of plant extract. Group 4 (High dose) received 900mg/kg body weight if plant extract. On the last day (Day 15), the body weight was determined, and blood sample was collected from the heart before sacrificing each of them for haematology and serum biochemistry. Organs (Heart, lungs, spleen, liver and kidney) were all taken from each animal for the relative organ weight.

Data analysis

Data obtained were analyzed using Statistical Package for Social Sciences (SPSS) version 22. Values were expressed as mean ± standard error of mean (SEM). Results were subjected to one-way analysis of variance (ANOVA) to compare the different doses with the control group. Duncan Post-hoc statistics was used to separate the significant means while the statistical confidence was set at 95% ($p < 0.05$).

RESULTS

Influence of *P. maximum* crude extract on the carcass weight of of male rabbit.

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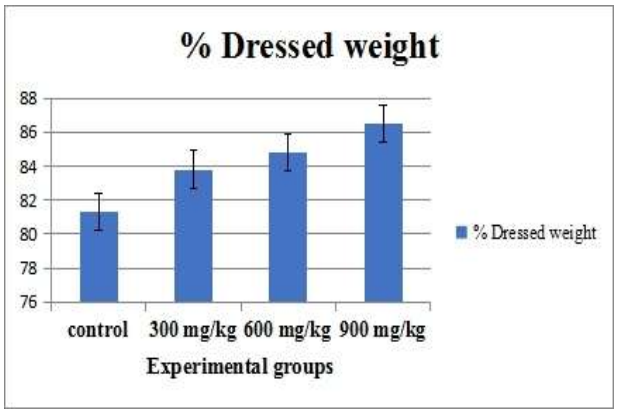

Fig. 2

Figure 2: Demonstrated increase in percentage dressed weight compared to the control (81.26±0.88) ; 300mg/kg b.w (83.51±0.59); 600 kg b.w. (84.81±0.02) and 900 kg b.w. (86.54±0.56)

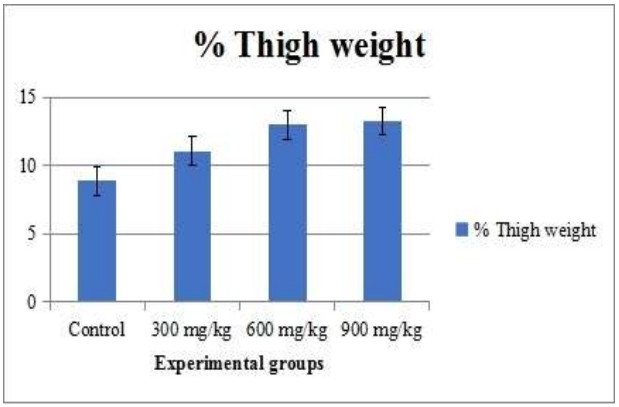

Fig. 3

Figure 3 showed thigh weight (%) increased as the doses of the extract increased from 300 mg/kg b.w.(11.08±0.01), 600 mg/kg b.w.(12.91±0.04) to 900 mg/kg b.w.(13.45±0.31) compared to the control (8.83±0.04)

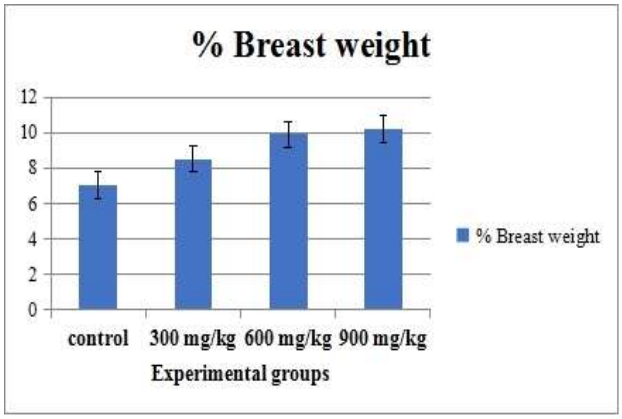

Fig. 4

Figure 4 showed breast weight (%) increased as the doses of the extract increased from 300 mg/kg b.w.(8.54±0.04), 600 mg/kg b.w.(9.93±0.05) to 900 mg/kg b.w.(10.16±0.14) compared to the control (7.07±0.04)

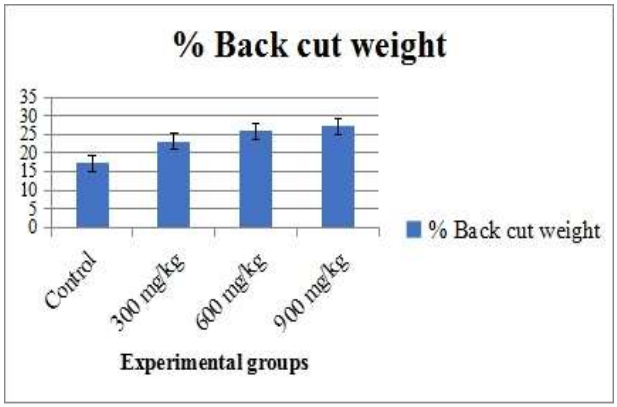

Fig. 5

Figure 5 showed back cut weight (%) increased as the doses of the extract increased from 300 mg/kg b.w.(23.26±0.14), 600 mg/kg b.w.(25.51±0.45) to 900 mg/kg b.w.(27.18±0.11) compared to the control (17.12±0.11)

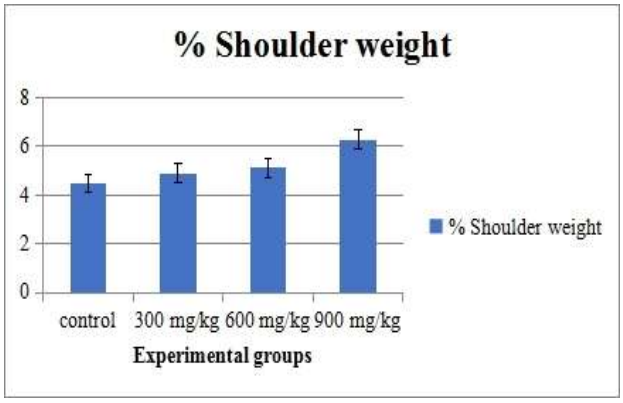

Fig. 6

Figure 6 shoulder weight (%) increased as the doses of the extract increased from 300 mg/kg b.w.(4.93±0.05), 600 mg/kg b.w.(5.21±0.09) to 900 mg/kg b.w.(6.27±0.02) compared to the control (4.60±0.20)

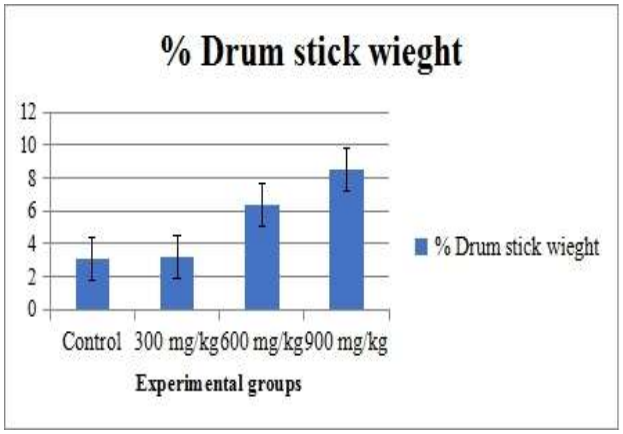

Fig. 7

Figure 7 shows drum stick weight (%) increased as the doses of the extract increased from 300 mg/kg b.w.(3.29±0.10), 600 mg/kg b.w.(6.35±0.04) to 900 mg/kg b.w.(8.51±0.10) compared to the control (3.18±0.05)

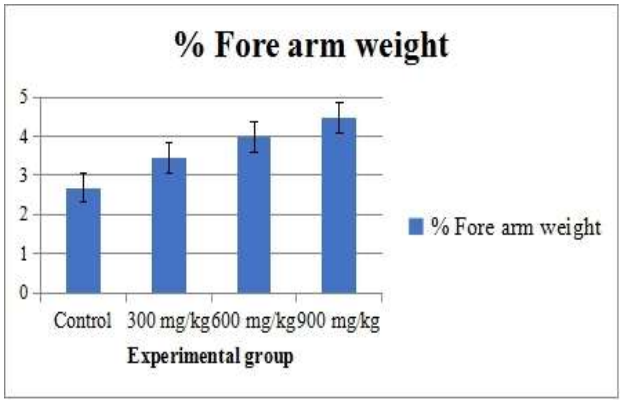

Fig. 8

Figure 8 shows fore arm weight (%) increased as the doses of the extract increased from 300 mg/kg b.w.(3.49±0.06), 600 mg/kg b.w.(3.79±0.32) to 900 mg/kg b.w.(3.79±0.32) compared to the control (2.72±0.07)

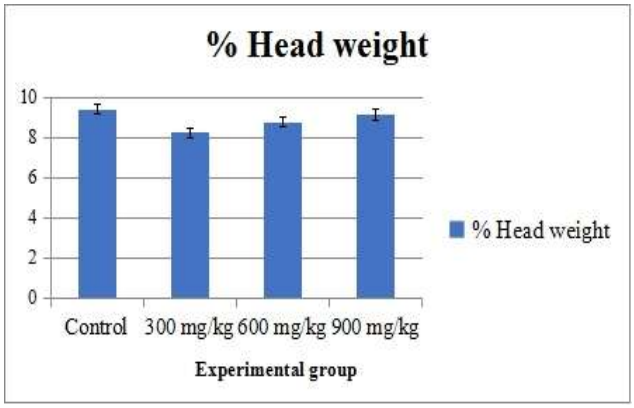

Fig. 9

Figure 9 shows head weight (%) increased as the doses of the extract increased from 300 mg/kg b.w.(8.95±0.64), 600 mg/kg b.w.(9.22±0.47) to 900 mg/kg b.w.(8.77±0.55) compared to the control (8.84±0.22) though a slight decrease at 900 mg/ kg b.w.

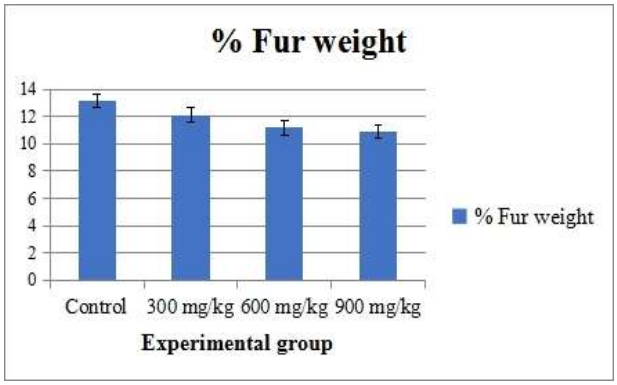

Fig. 10

Figure 10 shows fur weight (%) progressive decrease as the doses of the extract increased from 300 mg/kg b.w. (12.13±0.02), 600 mg/kg b.w.(11.11±0.05) to 900 mg/kg b.w.(10.84±0.03) compared to the control (13.14±0.06)
The result as presented in Figures 2-10, showed a consistent increase in relative weight of the weighed parts as the doses of the plant extract increased. All the cut parts weighed heavier ($P<0.05$), in the extract groups, than their respective weights obtained from the control group. This significant increase in the cut parts supported the significant weight gain recorded in the extract groups compared with the control. In contrast, the fur weight significantly reduced in the treated groups compared to the control.

Influence of *P. maximum* crude extract on relative organ weight

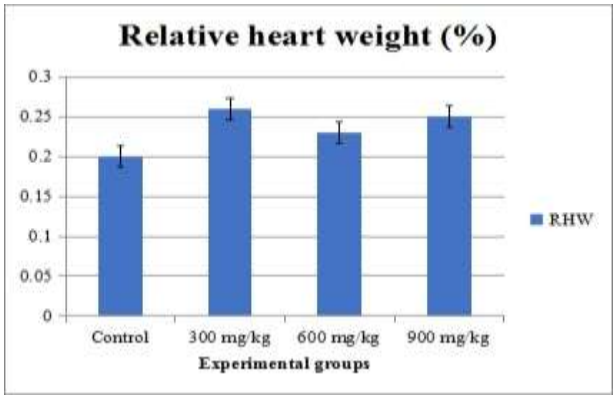

Fig. 11

Figure 11 shows that relative heart weight (%) increased as the doses of the extract increased from 300 mg/kg b.w. (0.22±0.02), 600 mg/kg b.w.(0.23±0.00) to 900 mg/kg b.w.(0.23±0.02) compared to the control

(0.22±0.03). There was a significant increase in the sizes of the heart relative to the animal body weight in the group administered the extract at the various doses in this study. This could mean that the extract has influence on the heart muscle cells

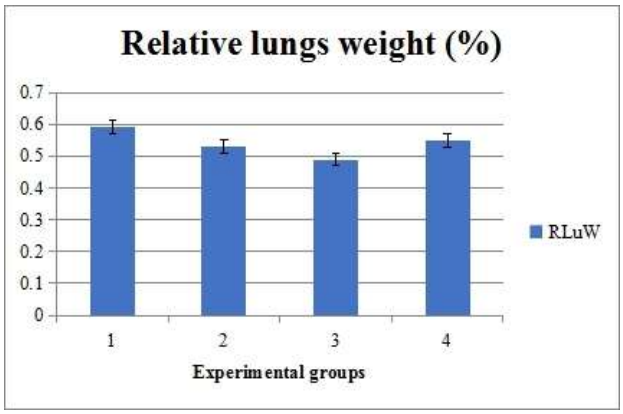

Fig. 12

Figure 12 shows that relative lungs weight (%) increased as the doses of the extract increased from 300 mg/kg b.w.(0.54±0.01), 600 mg/kg b.w.(0.53±0.03) to 900 mg/kg b.w.(0.55±0.02) compared to the control (0.56±0.02). The lungs in the extract groups were not significantly reduced compared with the control group.

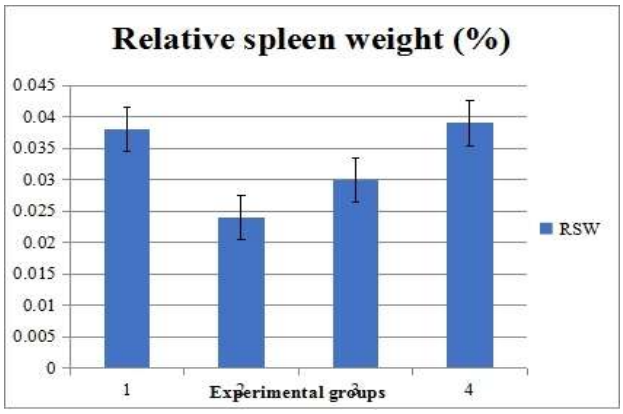

Fig. 13

Figure 13 shows that relative spleen weight (%) increased as the doses of the extract increased from 300 mg/kg b.w.(0.03±0.00), 600 mg/kg b.w.(0.15±0.21) to 900 mg/kg b.w.(0.15±0.21) compared to the control (0.03±0.00). The increase was not statistically significant.

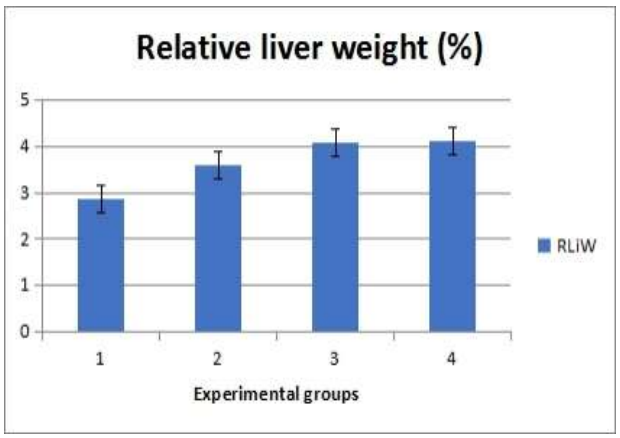

Fig. 14

Figure 14 shows that relative liver weight (%) increased as the doses of the extract increased from 300 mg/kg b.w.(3.61±0.01), 600 mg/kg b.w.(3.37±0.53) to 900 mg/kg b.w.(3.47±0.55) compared to the control (3.28±0.49). The increase was not statistically significant. There was a significant effect of the plant extract on the relative liver weights. The size of the liver organ was increased as the extract doses were increased. This could be attributed to increase metabolic activity of the liver at increased doses.

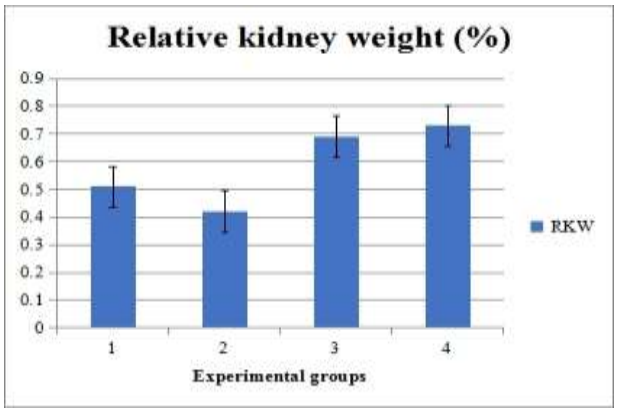

Fig. 15

Figure 15 shows that relative kidney weight (%) increased as the doses of the extract increased from 300 mg/kg b.w.(0.55±0.03), 600 mg/kg b.w.(0.55±0.02) to 900 mg/kg b.w.(0.55±0.02) compared to the control (0.56±0.04).There was a significant change in the relative kidney weight of the treated rabbits compared with the control. The observed increased kidney weight could be as result of increase activity of the organ in excretory function.

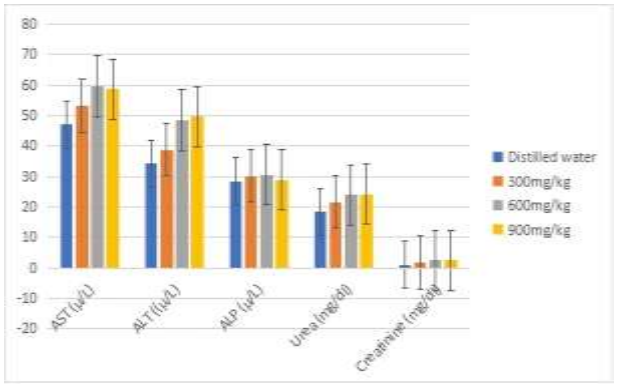

Fig. 16: Effect of *P. maximum* on serum biochemical markers.

The graph in Figure 16 represents the value of serum biochemistry of AST, ALT, ALP, Urea and Creatinine and represented as Mean ± S.E at (P<0.05). Treatment was 300, 600 and 900 mg/kg b.w. compared to control administered distilled water.

Figure 16 shows elevated **AST (µ/L)** as the doses of the extract increased from 300 mg/kg b.w.(53.22±0.08), 600 mg/kg b.w.(59.63±0.30,) to 900 mg/kg b.w.(58.72±0.40) compared to the control (47.06±0.58). *[Reference range: 32-84 µ/L]*

ALT (µ/L) was elevated as the doses of the extract increased from 300 mg/kg b.w.(38.81±0.85), 600 mg/kg b.w.(48.56±0.34) to 900 mg/kg b.w.(49.69±0.35) compared to the control (34.30±0.56). *[Reference range: 30-58 µ/L]*

ALP (µ/L) was elevated as the doses of the extract increased from 300 mg/kg b.w.(30.19±0.56), 600 mg/kg b.w.(30.19±0.56) to 900 mg/kg b.w.(28.96±0.61) compared to the control (28.56±0.87). *[Reference range: 0-500 µ/L]*

Urea (mg/dl) was elevated as the doses of the extract increased from 300 mg/kg b.w.(21.55±0.62), 600 mg/kg b.w.(23.95±0.84) to 900 mg/kg b.w.(24.18±0.63) compared to the control (18.28±0.03). *[Reference range: 10.7 – 2.0 mg/dl]*

Creatinine (mg/dl) was elevated as the doses of the extract increased from 300 mg/kg b.w.(1.73±0.04), 600 mg/kg b.w.(2.55±0.03) to 900 mg/kg b.w.(2.46±0.02) compared to the control (1.06±0.07). *[Reference range: 0.3 – 0.5 mg/dl]*

All the biochemical markers in the graph (Fig 16) showed no significant difference at p<0.05 and all values fall within the normal reference range.

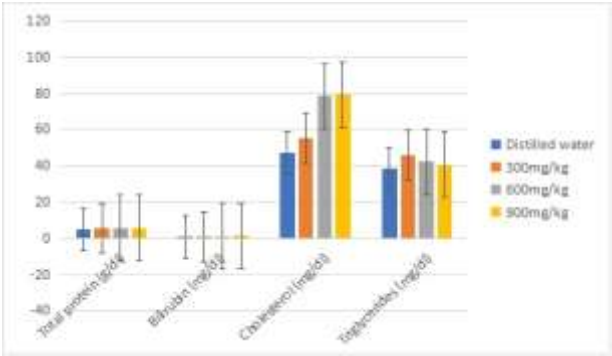

Fig. 17: Shows Total protein (g/dl) Bilirubin (mg/dl) Cholesterol (mg/dl) Triglycerides (mg/dl).

The graph in Figure 17 represents the value of Total protein (g/dl) Bilirubin (mg/dl) Cholesterol (mg/dl) Triglycerides (mg/dl) and represented as Mean ± S.E at (P<0.05). Treatment was 300, 600 and 900 mg/kg b.w. compared to control administered distilled water.

Total protein (g/dl) was elevated as the doses of the extract increased from 300 mg/kg b.w.(5.46±0.13), 600 mg/kg b.w.(5.66±0.16) to 900 mg/kg b.w.(5.73±0.20) compared to the control (4.64±0.25).Mild hyperproteinaemia *[Reference range: 5.6 -7.6 mg/dl]*

Bilirubin (mg/dl) was elevated as the doses of the extract increased from 300 mg/kg b.w.(1.07±0.01), 600 mg/kg b.w.(1.19±0.05) to 900 mg/kg b.w.(1.33±0.05) compared to the control (0.99±0.03). Mild hyperbilirubinaemia *[Reference range: 0.04 – 0.2 mg/dl]*

Cholesterol (mg/dl) was elevated as the doses of the extract increased from 300 mg/kg b.w.(55.02±0.82), 600 mg/kg b.w.(78.46±1.39) to 900 mg/kg b.w.(79.23±0.54) compared to the control (47.47±1.59). Moderate hypercholesteraemia *[Reference range: 37 – 95 mg/dl]*

Triglycerides (mg/dl) was slightly elevated but was reducing as the doses of the extract increased from 300 mg/kg b.w.(45.89±1.95), 600 mg/kg b.w.(42.33±0.88) to 900 mg/kg b.w.(40.69±1.45) compared to the control (38.25±1.29). *[Reference range: 27 – 160 mg/dl]*

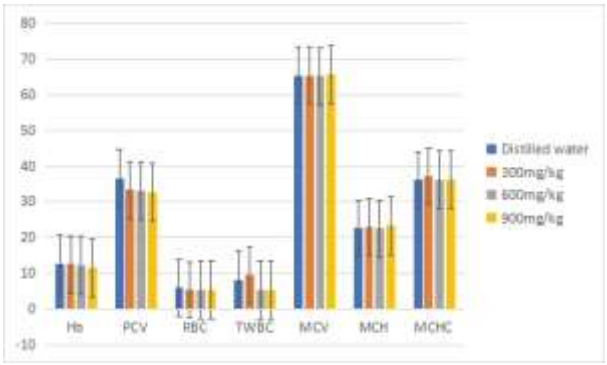

Fig. 18: Effect of *P. maximum* on haematology parameters.

The effect of extract on the haematology profile as presented in Fig 18 showed no alteration in the mean levels of Haemoglobin (g/dl), 300 mg/kg b.w. 12.53±0.29; 600 mg/kg b.w. 12.53±0.29; 900 mg/kg b.w. 11.63±0.31 compared to the control 12.76±0.64

Packed Cell Volume (%) levels of the experimental rabbits decreased, compared with the control, in Fig 18. 300 mg/kg b.w. 33.33±1.20; 600 mg/kg b.w. 33.00±0.57; 900 mg/kg b.w. 32.66±0.88 compared to the control 36.66±0.33. Values are within normal reference range. *[Reference range: 38.5 – 52 %]*

Red Blood Cell ($\times 10^6 mm^3$) levels of the experimental rabbits changed, compared with the control, in Fig 18. 300 mg/kg b.w. 5.43±0.19; 600 mg/kg b.w. 5.38±0.10; 900 mg/kg b.w. 5.46±0.11 compared to the control 5.96±0.04. Values are within normal reference range. *[Reference range: 7.62 –9.9 $\times 10^6 mm^3$]*

Total White Blood Cell ($\times 10^3 mm^3$) TWBC levels of the experimental rabbits increased, compared with the control, in Fig 18. 300 mg/kg b.w. 9.53±0.24; 600 mg/kg b.w. 12.83±0.16; 900 mg/kg b.w. 13.10±0.47 compared to the control 8.23±0.50. Values are within normal reference range. *[Reference range: 1.98 – 11.06$\times 10^3 mm^3$]*

Mean Corpuscular Volume (fl) level did not change compared with the control, in Fig 18. 300 mg/kg b.w. 65.40±0.01; 600 mg/kg b.w. 65.19±0.19; 900 mg/kg b.w. 65.60±0.30 compared to the control 65.37±0.47. Values are within normal reference range indicating normocytic

state of the Red blood cells. *[Reference range: 46.3 – 56.2 fl]*

Mean Corpuscular Haemoglobin (pg) level did not change compared with the control, in Fig 18. 300 mg/kg b.w. 23.03±0.43; 600 mg/kg b.w.22.54±0.24; 900 mg/kg b.w. 23.39±0.30 compared to the control 22.54±0.05. Values are within normal reference range indicating normocytic state of the Red blood cells. *[Reference range: 16.3 – 19.5 pg]*

Mean Corpuscular Haemoglobin Concentration (g/dl) level did not change compared with the control, in Fig 18. 300 mg/kg b.w. 37.23±0.79; 600 mg/kg b.w.36.25±0.31; 900 mg/kg b.w. 36.35±0.05 compared to the control 36.19±0.31. Values are within normal reference range indicating normochromic state of the Red blood cells**.** *[Reference range:31 – 38.5 g/dl]*

Therefore *P. maximun* at various doses in this research showed normocytic normochromic state of the red blood cells.

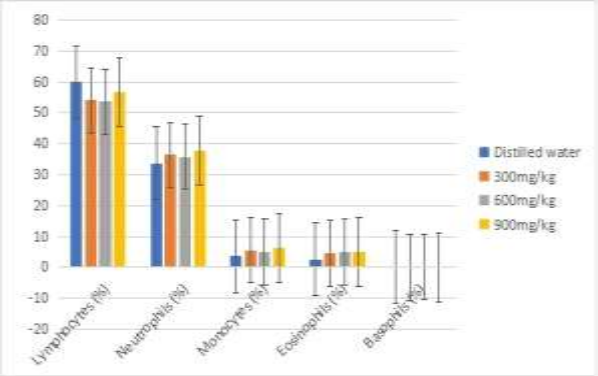

Fig. 19: Effect of *P. maximum* on differential white blood cells.

Lymphocytes (%) level decreased compared with the control, in Fig 19. 300 mg/kg b.w. 56.66±0.88; 600 mg/kg b.w.54.00±0.57; 900 mg/kg b.w. 53.66±0.33 compared to the control 60.00±1.15. There was mild lymphopaenia though the value was within normal reference range. *[Reference range: 44.7 – 87.1 %]*

Neutrophils (%) level increased compared with the control, in Fig 19. 300 mg/kg b.w. 36.33±1.20; 600 mg/kg b.w.35.83±0.16; 900 mg/kg b.w. 37.90±0.45 compared to the control 33.66±1.45. There was mild neutrophilia though the value was within normal reference range. *[Reference range: 9 – 49.3 %]*

Monocytes (%) level increased compared with the control, in Fig 19. 300 mg/kg b.w. 5.56±0.29; 600 mg/kg b.w.5.03±0.03; 900 mg/kg b.w. 6.06±0.07 compared to the control 3.66±0.33. There was moderate monocytosis though the value was within normal reference range. *[Reference range: 1 – 3.3 %]*

Eosinophils (%) level increased compared with the control, in Fig 19. 300 mg/kg b.w. 4.63±0.31; 600 mg/kg b.w.4.93±0.06; 900 mg/kg b.w. 4.93±0.06 compared to the control 2.60±0.30. There was moderate eosinophilia though the value was within normal reference range. *[Reference range: 0 – 4.4 %]*

Basophils (%) No cell count change recorded. 0.00±0.00 0.00±0.00 0.00±0.00 0.00±0.00. *[Reference range: 0 – 0.6 %]*

DISCUSSION

P. maximum is by far the most cultivated and fast growing, mainly used in cut and carry system for dairy and beef cattle feeding. It is often considered as one of the best species for beef production Van Soest, (1994). Investigations into the contributions of plant nutrition in animal production and the management of disease conditions have occupied research focus lately. Rabbits are known as 'hind gut fermenters' meaning that they have an organ called caecum which functions much like the rumen of a cow (Mc Donald *et al*, 2002; Boonman, 1993). The effect of *P. maximum* on weight gain, carcass quality and relative organ weight of male rabbit were determined in this research. The animal mean body weights of the extract groups increased as the dose increased. The dressed weight, thigh weight, breast, shoulder, back cut, drum stick and the weight of forearm relative to the live weights of the experimental rabbits were significantly increased in a dose dependent manner, with the percentage weights of these cut parts of the rabbit administered 900 mg/kg. However, the percentage fur weight was significantly decreased across the extract groups, as the doses were decreased compared with the percentage fur weight of the rabbits in control group. There was no significant difference in the percentage fur weight of the experimental rabbits. *Panicum maximum* caused a significant weight gain in the rabbit model. The biochemical and haematological results showed no toxic effect. This grass is good for ranching and grazing for weight gain in animals.

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