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The Effect of Feeding Varying Levels of Water Spinach (*Ipomoea aquatica*) Leaf Meal on Reproductive Tract Morphometry, Haematology and Serum Biochemical Parameters of Female Rabbits

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Abstract: A total of thirty-two (32) rabbits of mixed breeds aged 10 - 12 weeks and weighing between 900 – 1000g, were used in an experiment to determine the effect of feeding varying dietary levels of water spinach leaf meal on reproductive tract morphometry, haematology and serum biochemical parameters of female rabbits. The rabbits were randomly allocated into four dietary treatments, T1, T2, T3 and T4 containing 0%, 5%, 10% and 15% of water spinach leaf meal respectively. Feed and water were served *ad libitum* throughout the sixteen (16) weeks of the feeding trial. At the end of the feeding trial, 3 rabbits per treatment were slaughtered and their reproductive organs were carefully dissected out and separated into different components and evaluated. Some haematology and serum biochemical parameters were also evaluated. The results showed no significant (p>0.05) effect of diet on reproductive tract morphometry, except the ovarian weight. No significant (p>0.05) influences of diet were observed on all the haematological parameters studied. There was also no significant (p>0.05) effect of diet on serum biochemical indices except alanine amino transaminase. These results suggested that inclusion of up to 15% levels of water spinach leaf meal in the diets will guarantee good health and support normal reproductive processes in female rabbits.

Key words: Rabbits · Morphometry · Haematology · Serum biochemistry · Water spinach

INTRODUCTION

Reduction in the cost of feeding animals which often account for about 75% of the total cost of production has been identified by many researchers as a paramount prerequisite of ensuring the availability of affordable meat and meat products to the Nigerian populace; majority of whom are rural dwellers and who more often consume less than 10g as against recommended 35g protein per day [1]. Hence the need to source for alternative livestock feed ingredient.

Water spinach is an herbaceous trailing vine that dwells in muddy stream bank, fresh water ponds and marshes. This perennial aquatic vine is confined to the tropics and sub tropics and does not grow well when temperature is below 24°C. Water spinach can reproduce

sexually by producing one to four seeds in fruiting capsules or vegetative by stem fragmentation [2]. It is widely consumed vegetable in South East Asia. Although it is widespread in Africa, the herbs are eaten mostly in Ethiopia, Sudan and Tanzania [3, 4]. There are reports of using Ipomoea aquatica to treat diabetes [5]), abscesses and intestinal disorders [6]. Some studies have shown anti-depressant and anti-epileptic [7] activities of Ipomoea aquatica. Many feed ingredients are fed to rabbits without recourse to their health and physiological implications on the animals. Haematological and biochemical parameters are commonly used to determine these implications [8]. Haematology and serum biochemical assay of animals are suggestive of physiological disposition of the animals to their plane of nutrition [9].

Reproductive performance economically important in rabbits because of its effect on the number of offspring produced per year [10]. To maintain a good reproductive performance a clear idea about the reproductive organs of rabbits is necessary. The biometry of genital tracts of the female reveals the overall wellbeing of the animals. According to Kunbhar and co-investigators [11], knowledge of the biometrical status of female genital tract is essential to perform insemination, pregnancy diagnosis and artificial dealing with the infertility problems and its treatment. Not much has been reported on the implications of incorporating water spinach (Ipomoea aquatica) leaf meal in diets on reproductive tract morphometry, haematological and biochemical indices of female rabbits. This study was therefore designed to evaluate the effect of feeding varying levels of water spinach on reproductive tract morphometry, haematological and serum biochemical parameters of female rabbits.

MATERIALS AND METHODS

Location: The study was conducted at the rabbitry unit of the Livestock Research Farm, University of Agriculture, Makurdi, Nigeria. Makurdi is located at Latitude 7°14' North and Longitude 8°21' East and lies within the Southern guinea savannah region of Nigeria. The area is warm with temperature range of 24°C to 36°C and high temperature is experienced between late February and April. The rainfall is between 508 and 1016mm.

Test Ingredients: Water spinach leaves were collected around river Benue banks in Makurdi. The plants were shade dried, powdered and included at the levels of 0, 5, 10 and 15% in the diets labelled T1, T2, T3 and T4 respectively (Table 1). Other ingredients used include soybeans, maize offal, maize, rice offal, premix and salt. The diets were formulated to meet 16% crude protein requirement for growing rabbits. Proximate composition of water spinach is shown in Table 2.

Table 1: Ingredient and nutrient composition (%) of the experimental diets

Ingredients	Diets				
	T ₁	T ₂	T ₃	T ₄	
Maize	29.95	29.95	29.00	30.95	
Full fat soya bean	28.00	27.50	26.55	26.00	
Maize bran	24.00	19.50	15.00	15.00	
Rice offal	15.00	15.00	15.00	15.00	
Water spinach leaf meal	0.00	5.00	10.00	15.00	
Premix*	0.25	0.25	0.25	0.25	
Salt	0.30	0.30	0.30	0.30	
Total	100.00	100.00	100.00	100.00	
Calculated values (%)					
Crude protein	18.28	18.28	18.28	18.04	
Crude fibre	9.01	9.46	9.79	10.14	
Dry matter	93.40	93.45	93.51	93.77	
Calcium	0.55	0.54	0.54	0.53	
Phosphorus	0.65	0.63	0.62	0.60	

T1=0% water spinach meal T2=5% water spinach meal T3=10% water spinach meal T4=15% water spinach meal

Table 2: Proximate composition of water spinach leaf meal

Measurements	%
Moisture	86.62
Crude protein	20.75
Crude fibre	10.59
Ether extract	18.67
Ash	10.87

^{*0.25}kg of Mineral/Vitamin Premix contained the following: Vitamin A 1,800IU, Vitamin D 250IU, Vitamin E 8,000IU, Vitamin K 750mg, B₁ 750mg, B₂ 1000 mg, B₆ 800mg, B₁₂ 25mg Folic 300mg, Niacin 5000mg, Pantothenate 3000mg, Biotin 25mg, Choline 160g, Thyroxine 300mg, Copper 0.4g, Iron 4g, Manganese 5.5g, Iodine 0.2g, Zinc 5g, Cobalt 0.15g, Selenium 0.15g.

Experimental Animals and Management: A total of thirty-two growing rabbits of mixed breeds, aged between 10 and 12 weeks with average weight of 995.75g were used for the study. The rabbits were randomly assigned to four (4) treatments with six (6) rabbits per treatment and kept under standard management conditions. Each rabbit served as replicate. The rabbits were weighed individually at the beginning of the study and weekly thereafter. The experimental animals were sacrificed at the end of the feeding trial that lasted for sixteen (16) weeks.

Reproductive Tract Morphometry: At the end of the sixteen (16) weeks of feeding trial, 3 does were randomly selected from each experimental diet, starved for 12 hours and thereafter sacrificed by stunning and decapitation. The reproductive tract was carefully removed *intoto*, trimmed free of fats and adhering connective tissues and subjected to morphometric analysis as previously described [12]. The weights of the various components of the reproductive tract namely, vagina, uterine horns and oviducts were taken using highly sensitive digital balance. Their linear measurements were also taken with the aid of calibrated ruler.

Blood Sampling: Five (5) millilitres blood samples were collected from the sacrificed bucks at slaughter from the jugular vein and allowing free flow of blood into ethylene diamine tetra acetic acid (EDTA) treated bottles. This was used to determine the haematological indices. The haematological indices evaluated were haemoglobin (Hb) concentration, packed cell volume (PCV), red blood cell

(RBC) counts, white blood cell (WBC) counts and leucocytes differential counts, mean corpuscular haemoglobin (MCH), mean corpuscular volume (MCV) and mean corpuscular haemoglobin concentration (MCHC) as previously described [13]. Another 5 millilitres was collected in the bottles without anti-coagulant and used to determine the serum biochemical indices. The biochemical indices determined were alanine amino transaminase, alkaline phosphatase, total protein, albumin, glucose, total cholesterol, creatinine, conjugated bilirubin, total bilirubin and urea.

Statistical Analysis: Data obtained were subjected to analyses of variance using GenStat (Release 4.24) statistical package [13]. Significant differences between treatment means were separated using Duncan's Multiple Range Test of the same statistical package.

RESULTS AND DISCUSSION

The results of the reproductive tract morphometry of female rabbits fed graded levels of water spinach are presented in Table 3. The results showed that water spinach significantly (P>0.05) influenced paired ovarian weight. The values ranged between 0.11±0.02-0.15±0.02g were comparable to values ranged between 0.11±0.04-0.29±0.04g reported by Bitto and co-workers [12] who fed female rabbits with diets containing up to 30% pawpaw peel meal. The paired oviduct weight was not significantly (p>0.05) influenced by the treatment diets. The paired values obtained ranged between 0.20±0.03-0.41±0.04g.

Table 3: Morphometric	abaracteristics of the	raproductive treet	of famala rabbita	fed water chine	h laaf maal
Table 5. Morbiometric	Characteristics of the	reproductive tract	of female fabbles	icu water spina	JII ICAI IIICAI

	Diets				
Parameters	T1	T2	T3	T4	
Body weight (g)	1473.75±101.30	1510.59±92.32	1496.09±87.20	1500.63±66.32	
Paired ovary weight (g)	0.11 ± 0.02^{b}	0.14±0.01 ^a	0.15 ± 0.02^a	0.16 ± 0.01^a	
Paired oviducts weight (g)	0.33 ± 0.06	0.41 ± 0.04	0.34±0.05	0.30 ± 0.03	
Paired uterine horn weight (g)	1.63±0.07	1.83±0.32	1.89±0.17	1.71±0.11	
Cervix weight (g)	1.40 ± 0.20	1.59±0.08	1.36±0.09	1.58 ± 0.12	
Vagina weight (g)	1.56±0.34	1.54±0.24	1.67±0.25	1.51±0.09	
Ovary length(cm)	2.10±0.12	2.14±0.10	2.20±0.15	2.43 ± 0.30	
Oviduct length(cm)	10.73±0.12	11.93±1.05	9.30±0.56	11.20±0.66	
Uterine horn length(cm)	11.73±0.77	11.84±0.29	12.50±0.59	12.48±0.32	
Cervix length(cm)	1.40±0.21	1.63±0.09	1.63±0.21	1.70±0.25	
Vestibule length(cm)	4.44±0.15	5.17±0.32	5.27±0.34	4.77±0.38	
Vagina length(cm)	4.60±0.10	4.87±0.47	4.83±0.22	4.70±0.35	

^{ab} means in the same row with different superscripts are significantly different (p<0.05) T1=0% water spinach meal T2=5%, water spinach leaf meal, T3=10% water spinach meal, T4=15% water spinach leaf meal

Table 4: Haematology of female rabbit fed water spinach leaf meal

	Diets	Diets				
Parameters	T1	T2	Т3	T4	Los	
Packed cell volume (%)	36.60±0.88	37.00±1.53	38.00±1.70	34.00±2.00	ns	
Red blood cells (x1012/l)	6.10±0.15	6.14±0.24	6.30±0.29	5.47±0.25	ns	
Haemoglobin (g/dl)	12.10±0.29	12.03±0.32	12.30±0.45	11.00±0.50	ns	
MCV (fl)	60.11±0.22	60.32±0.18	60.30±0.02	58.52±0.14	ns	
MCH (pg)	19.84 ± 0.16	19.64±0.28	19.55±0.39	20.35±0.47	ns	
MCHC (g/dl)	33.00 ± 0.15	32.55±0.54	32.41±0.65	32.43±0.57	ns	
White blood cells (x109/l)	8.08 ± 0.32	6.60 ± 0.42	7.20±0.57	6.63±0.25	ns	
Lymphocytes (%)	50.00±1.53	49.00±1.30	48.00±1.16	49.60±0.25	ns	
Neutrophils (%)	44.67±1.30	46.67±1.45	46.66±1.20	44.66±0.88	ns	
Monocytes (%)	3.30±0.30	2.67±0.30	3.00±1.58	3.30 ± 0.30	ns	
Eosinophils (%)	1.60±0.34	1.30 ± 0.26	1.67±0.30	1.67±0.52	ns	
Basophils (%)	0.30 ± 0.01	0.36 ± 0.02	0.46 ± 0.02	0.50 ± 0.03	ns	

T1=0% water spinach meal T2=5% water spinach meal Los=Level of significance T3=10% water spinach meal T4=15% water spinach meal, ns = not significant (P> 0.05), MCHC=Mean Corpuscular Haemoglobin Concentration (g/dl)

Table 5: Serum biochemistry of female rabbits fed water spinach leaf meal

	Diets				
Parameters	T1	T2	T3	T4	
Alkaline amino transaminase (U/l)	47.00b±4.30	48.30 ^b ±0.88	49.00±0.58	53.67a±1.20	
Alkaline phosphatase (U/l)	98.67±1.20	98.00±1.16	102.00±2.31	100.30±1.20	
Total protein (g/ 100ml)	7.67±0.35	7.80 ± 0.46	8.07±0.02	7.97±0.32	
Albumin (g/ 100ml)	3.53±0.24	3.47±0.29	4.00±0.12	3.77±0.19	
Glucose (mg/100ml)	101.60±1.45	100.00±1.53	102.30±2.19	100.03±1.20	
Total cholesterol (mg/100ml)	0.79±0.04	0.77 ± 0.03	0.84 ± 0.03	0.80 ± 0.04	
Creatinine (mg/100ml)	0.79±0.04	0.77 ± 0.03	0.84 ± 0.03	0.80 ± 0.04	
Conjugated bil. (mg/100ml)	0.39±0.04	0.30 ± 0.02	0.38 ± 0.02	0.34 ± 0.03	
Total bilirubin (mg/100ml)	0.68±0.21	0.77 ± 0.04	0.81 ± 0.02	0.77±0.04	
Urea (mg/100ml)	51.30±1.76	49.60±1.45	51.30±2.40	51.66±0.76	

abmeans in the same row with different superscripts are significantly different (p<0.05) T1=0% water spinach meal T2=5% water spinach meal Los=Level of significance T3=10% water spinach meal T4=15% water spinach meal

The values were greater than the paired values reported by Bitto and co-workers [12]. The paired uterine horn, cervixes and vagina weights were also not significantly (p>0.05) influenced by dietary treatments. The values obtained were generally higher than the corresponding values of 0.55±0.07-1.33±0.34, 0.25±0.04-1.09±0.41 and 0.36±0.08-0.93±0.22g for paired uterine horn, cervixes and vagina weights as previously by reported Bitto and coworkers [12]. The results showed no significant (p<0.05) influence of diet on the reproductive tract organ lengths. The values obtained for oviduct, uterine horns and cervixes lengths were higher than values of 6.00±0.79- 9.20 ± 0.60 , $6.40\pm2.13-10.83\pm0.34$ and $0.87\pm0.19-1.37\pm0.19$ cm for oviducts, uterine horns and cervixes lengths previously reported Bitto and co-workers [12] who fed female rabbits with diets containing up to 30% pawpaw

peel meal. These differences may be attributed to the age and weights of the rabbits used in the experiments.

The results of haematology of female rabbits fed graded levels of water spinach are shown in Table 4.

Packed cell volume (PCV), haemoglobin (Hb), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC) mean corpuscular volume (MCV), red blood cell (RBC) counts were not significantly (P>0.05) different across the treatments. The values obtained were within the normal ranges reported for rabbits [14]. These reports showed that the normal ranges of value for rabbits are as follows: PCV: 33-50%, Hb: 9.4-17.4 g/dl, RBC: 3.8-7.9x106/mm³, MCH: 18-24pg, MCHC: 27-34% and MCV: 50-75fl. It has been reported that when the haematological values fall within the normal range for rabbits [15], it implies that the diets did not have

adverse effects on haematological indices during the feeding trial, but when the values fall below the normal range, it is an indication of anaemia [16].

White blood cell (WBC) count and leucocyte differential counts were not significantly (p>0.05) influenced by dietary treatments. The mean values obtained for WBC ranged between 6.60±0.42-8.08±0.32 x10°/l and were within the normal ranges of value of 5.20-10.6x10°/L reported for adult female rabbits [17]. Values obtained in this study therefore fall within the normal range as higher WBC count may suggest resistance or prevalence disease condition [18]. White blood cell count below the normal range is an indication of allergic conditions, anaphylactic shock and certain parasitism, while elevated values indicate the existence of recent infection, usually with bacteria [19].

The result of serum biochemistry of female rabbits fed graded levels of water spinach is presented in Table 5. Alanine amino transaminase (ALT) value significantly (p<0.05) increased at T4. ALT values found in this study were within the normal range of 25-65 IU/l as reported in clinically healthy rabbits [19]. The increase cannot be attributed to any cause since values fall within the normal range. No significant (p>0.05) difference was observed for Alkaline Phosphatase (AP) values since they are within the normal range of between 98.00 -102.00 [19] stated that in rabbits, there is a wide variation between laboratory reference ranges for AP value.

Similarly, no significant (P>0.05) differences were observed amongst the treatment for total protein, glucose, total cholesterol, conjugated and total bilirubin, creatinine and urea. The non significant (p>0.005) values obtained in this study suggest nutritional adequacy of the diets for rabbits.

CONCLUSION

Based on the results obtained from this study, it was concluded that the inclusion of up to 15% levels of water spinach leaf meal in diets will guarantee good health and support normal reproductive processes in female rabbits.

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