

Influence of the Ethnoveterinary Plant-*Spondias mombin* L. on Partial Daily Milk Yield (PDM), Haematology and Serum Biochemistry of Lactating West African Dwarf (WAD) Ewes

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Abstract: Twelve lactating West African Dwarf (WAD) ewes averaging 14 kg in weight and age between 2-3 years were used in 12-weeks study to determine the partial daily milk yield (PDM), haematological and serum biochemical parameters of ewes fed the ethnoveterinary and ethnomedical plant *Spondias mombin*. The lactating ewes were assigned to 2 treatment groups of 6 ewes per group, designated as T1 and T2, respectively. The ewes in T1 were the treated group fed *Spondias mombin* L. post partum while those in T2 served as a control and were not fed *Spondias mombin*. Haematological parameters measured include the Packed Cell Volume (PCV) and total white blood cell count while serum biochemical parameters were liver enzyme assay (including Aspartate Transaminase, (AST); alanine amino transferase, (ALT) and Alkaline Phosphatase (ALP) as well as total and conjugated bilirubin. Kidney function tests highlighting urea and creatinine concentrations as well as potassium, sodium, bicarbonate and chlorine ion concentrations were conducted on the control and treated groups. Results showed that *S. mombin* increased the PDM yield of the lactating ewes in the treatment group over their counterparts in the control group in all the 12 weeks of lactation. These increases were significant ($p < 0.05$) in the 3rd and 8th weeks. Weeks 3 and 8 PDM values were 61.66 ; 35.44 and 40.31 g; 21.28 g for the treated and control groups, respectively. *Spondias mombin* did not significantly ($p > 0.05$) influence PCV and WBC values within experimental groups. Also, liver enzymes (ALT, AST and ALP) as well as total and conjugated bilirubin were also not significantly ($p > 0.05$) affected by *S. mombin*. However, creatinine concentration was significantly higher ($p < 0.05$) in the treated group (0.73 mg dL^{-1}) than the control group (0.65 mg dL^{-1}). These results suggest that *S. mombin* could be useful in improving on the milk production of our WAD ewes; without any adverse effects on their general health and performance.

Key words: WAD ewes, PDM, *Spondias mombin*, haematology, serum biochemistry, lactation

INTRODUCTION

Spondias mombin L. (hog plum, ichikara) is an ethnoveterinary and ethnomedical plant that is used in the treatment of dystokia and retained placenta in humans and livestock (Okafor and Lamb, 1994). It is also used as an aphrodisiac in man (Burkill, 1985). It is highly relished by livestock especially sheep (Sosa *et al.*, 2004). Phytochemical analyses show that it contains tannins, alkaloids, including pyrolidines and 2-monosubstituted piperidines; proanthocyanidins, glycosides, saponins, but no cardiac glycosides (Edeoga and Eriata, 2001).

The contractile action of *S. mombin* on the uterine muscle that has led to its use in cases of dystokia and retained placenta can be compared to that of Oxytocin (OT) on uterine muscles during labour and on the

myoepithelial cells of the alveoli during milk ejection. Oxytocin is a neurohypophysial nonapeptide hormone named after the “quick birth” which it causes due to its uterotonic activity (Gimpl and Fahrenholz, 2001). Thus, the “quick birth” action of *S. mombin* that has led the rural farmers to use it in cases of dystokia and retained placenta could be an indication that it is a phyto-genic source of OT; or better still, contains a potent agonist of the oxytocin receptor site.

Apart from its effect on the uterine muscles, another classical role assigned to oxytocin is milk ejection from the mammary gland (Hafez, 1987; Bruickmaier and Blum, 1998; Gimpl and Fahrenholz, 2001; Waugh and Grant, 2001; Macuhova *et al.*, 2004). Milk ejection- a neuroendocrine reflex (Macuhova *et al.*, 2004) is triggered when there is suckling of the teat. This generates sensory impulses that

are transmitted from the teat to the spinal cord and then to the secretory oxytocinergic neurons in the hypothalamus. These are the magnocellular neurons in the hypothalamic paraventricular and supraoptic nuclei; and they display a synchronized high frequency bursting activity consisting of a brief (3-4) high frequency discharge of action potential, leading to oxytocin release (Gimpl and Fahrenholz, 2001). Oxytocin then causes contraction of myoepithelial cells surrounding the alveoli, forcing milk stored in the alveoli into the mammary ducts and gland cistern (Lefcourt and Akers, 1983; Waugh and Grant, 2001). Studies in OT deficient mice showed that it is the only hormone responsible for efficient milk ejection.

The WAD ewe is a poor milker, kept exclusively for meat, as the ewe's milk yield barely suffices to feed the lambs. Hence, lamb mortality is high in this breed (Charray *et al.*, 1992). On the other hand, the rural farmer may not have easy access to genetic improvement of these WAD ewes via cross breeding and concentrate for feeding lactating ewes is scarce and expensive. This study seeks to investigate the use of *S. mombin* on our trypanotolerant WAD ewes to improve on their milk yield. The blood profile of lactating ewe helps to determine the level of safety of *S. mombin* on the lactating WAD ewe.

MATERIALS AND METHODS

Location of study: The experiment was conducted at the Sheep and Goat Unit of the Teaching and Research farm of Michael Okpara University of Agriculture, Umudike, near Umuahia, Abia State of Nigeria. Umudike bears the co-ordinate of 05° 28' North and lies at an altitude of 122 m above sea level. It is located within the tropical rain forest zone and an annual rainfall of about 2177 mm characterizes the environment. The relative humidity ranges from 50-90%. The monthly environmental temperature ranges from 17-36°C. The warmest month of the year is March, with an average temperature of 22-30°C. The meteorological data were collected from the Meteorological Station of the National Root Crop Research Institute Umudike, Abia state Nigeria.

Management of experimental animals: Twelve mature WAD ewes, randomly sampled from the University Research flock, weighing 12-16 kg and aged 2-3 years, were used for the study. The experimental animals were quarantined in the quarantine house for a period of 4 weeks during which they were treated against ecto and endoparasites and equally vaccinated against Pestes de Petit Ruminante (PPR). The ewes were then transferred to breeding pens where they were housed singly. Forage was cut and fed to the ewes every morning and fresh

Table 1: Proximate composition of *Spondias mombin*

Nutrient	Composition (%)
Ash	4.20
Ether Extract	7.05
Moisture content	75.40
Crude protein	8.50
Crude fibre	13.45

water supplied *ad libitum*. The ewes were also placed on a supplementary concentrate diet (1kg/ewe/day) made up of maize offal, wheat offal, groundnut cake, palm kernel cake, bone meal and table salt. The crude protein content of the concentrate diet was approximately 13%.

A proven ram was introduced into the flock for breeding following detection of estrus. Pregnancy diagnosis was done by linear measurement of changes in udder size (Akomas *et al.*, 2006) and non-return to heat. Following pregnancy confirmation, the ewes were divided into two groups; control and treatment groups, designated T₁ and T₂, respectively with each group made up of 6 ewes.

After lambing, *Spondias mombin* were freshly cut and introduced to the ewes in the treatment group, at the rate of 1kg per ewe per day, in addition to the concentrate and forage previously fed. The proximate composition of *S. mombin* is presented in Table 1. *Spondias mombin* was given to the T₂ ewes the night before the milking after the remnants of the forage given in the morning had been removed.

Milk collection and sampling: Milk collection commenced after a week of lambing. The lambs were separated from their mothers the night before the milking (12 h) after they were fed with artificial milk and the ewes were milked by hand the following morning. The milk obtained was defined as the Partial Daily Milk yield (PDM), which is a combination of milk off-take and 12 h milk yield. Milk samples collected were weighed using an electronic balance and cent-o-gram manual weighing balance. Thereafter, the volumes of the milk samples were read off in a calibrated cylinder (in mL). The milk samples were then fed back to the lambs whose mothers were milked. Milk collection and sampling continued three times per week until twelve weeks of lactation.

Blood sampling and analysis: The ewes were bled through the jugular vein at 6 weeks post partum using a hypodermic syringe and sterile needle. Ten mL of blood was collected per ewe and 5 mL was deposited into different plastic tubes containing ethylenediaminetetraacetate (EDTA) for hematology. The remaining 5 mL was allowed to stand and clot in the hypodermic syringe at room temperature within 3 h. Sera were then collected and used for biochemical analysis;

including liver and kidney function tests. Tests for liver function done were serum bilirubin (Total and Conjugated), ALP, AST and ALT. Tests for kidney function done include serum Na⁺, K⁺, Cl⁻, HCO³⁻, urea and creatinine concentrations.

Total WBC count was done using improved Neubauer counting chamber and packed cell volume was determined using the microhaematocrit device. Serum urea and Creatinine concentrations were analyzed spectrophotometrically using Randox® kit. Sodium and Potassium ion concentrations were determined by flame photometry. Bicarbonate and Chlorine ion concentrations were estimated using appropriate indicators. Liver enzymes: Alanine amino transferase, ALT (SGPT), Aspartate amino transferase, AST (SGOT) and Alkaline Phosphatase were determined spectrophotometrically using Randox ® test kits.

Statistical analysis: Data generated were analysed using the independent Student's t-test. Mean and standard error of difference were calculated for the 2 treatment groups.

RESULTS AND DISCUSSION

The PDM yield of ewes fed *S. mombin* was and significantly higher (p<0.05) than that of the control group at weeks 3 and 8 of lactation (Table 2). At week 3, PDM yield was 61.66 g for the ewes in the treated group, while those in the control group had a PDM yield of 40.31 g. At week 8, the treated group had a PDM yield of 35.44 g and that of control was 21.88 g. The other weeks of lactation showed no significant differences (p>0.05). This is evident in the lactation curves of both groups (Fig. 1).

These higher values in the PDM yield observed in the treated ewes could be indicative of the oxytocin-like effect (agonist effect) of *S. mombin* on the myoepithelial cells of the mammary gland. The result obtained is in

Table 2: Weekly milk yield (g) of the lactating ewes

Lactation week	Control mean	Treated mean	SED	Significance
1	55.45	82.26	14.92	ns
2	45.96	64.71	12.86	ns
3	40.31	61.66	9.47	*
4	42.96	58.57	11.09	ns
5	36.07	50.44	7.53	ns
6	29.43	33.58	6.35	ns
7	24.19	28.58	6.21	ns
8	21.28	35.44	5.48	*
9	24.52	29.39	8.44	ns
10	22.79	29.78	7.65	ns
11	21.69	25.57	6.51	ns
12	20.63	26.43	6.64	ns

NS = Not Significant (p<0.05), * = Significantly different (p<0.05), SED = Standard Error of Difference

consonance with that of previous studies on the effect of exogenous OT administration on milk yield. However, Bruickmaier (2003) reported that oxytocin treatment might have no effect on milk yield of cows. Other authors have reported a stimulatory effect of oxytocin in milk yield in dairy cows (Ballou *et al.*, 1993; Knight, 1994).

This seeming disparity or disagreement may not be unconnected to the mechanism of down regulation or desensitization of receptor molecules as a result constant stimulation by agonists (Gimpl and Fahrenholz, 2001). Several mechanisms have been proposed to explain the effect of administration of OT or its analogs on milk yield. One possibility is that OT may directly affect productivity or maintenance of mammary epithelial cells (Ballou *et al.*, 1993; Lollivier *et al.*, 2001). However, a previous study indicated that the galactopoietic effect of OT is due to increased efficacy of milk removal (Knight, 1994).

The results of the haematological and serum biochemical tests are presented in Table 3. There was no

Table 3: Haematological and blood biochemical values of control and treated ewes

Parameters	Control mean	Treated mean	SED	Significance
PCV (%)	44.33	43.50	3.04	ns
Total WBC (10 ³ µL ⁻¹)	7.38	7.23	1.05	ns
Liver function tests				
Total bilirubin (g dL ⁻¹)	0.73	0.73	0.05	ns
Conjugated bilirubin (g dL ⁻¹)	0.15	0.22	0.03	ns
AST (SGOT) IU L ⁻¹	10.67	10.50	0.31	ns
ALT (SGPT) IU L ⁻¹	7.33	7.50	0.65	ns
ALP IU L ⁻¹	49.67	49.67	1.51	ns
Kidney function tests				
Urea (mg dL ⁻¹)	31.33	29.33	1.92	ns
Creatinine (mg dL ⁻¹)	0.73	0.65	0.03	*
Sodium ion (mmol L ⁻¹)	139.67	139.33	0.60	ns
Potassium ion (mmol L ⁻¹)	4.07	4.03	0.05	ns
Chlorine ion (mmol L ⁻¹)	98.83	98.67	0.78	ns
Bicarbonate ion (g dL ⁻¹)	26.33	25.83	0.45	ns

NS = Not Significant (p<0.05), * = Significantly different (p<0.05), SED = Standard error of difference

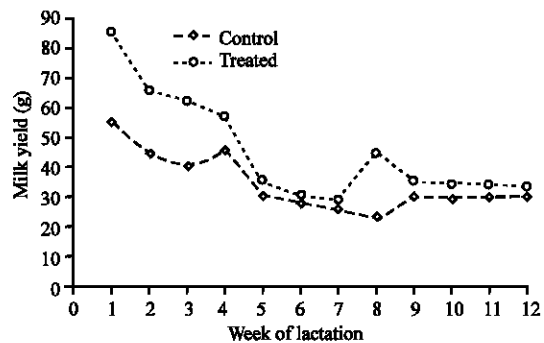


Fig. 1: Weekly milk yield (g) of the lactating ewes in two groups

significant difference in the mean PCV values of ewes in both groups. Mean PCV value was 43.50 and 44.30% for the treated and control groups, respectively. These values are at variance with the value of 35.5±1.0% reported by Taiwo and Ogunsami (2003). This disparity may not be unconnected to the difference in the environment and other physiological factors of these WAD ewes, particularly lactation. These variations in PCV values suggest that WAD sheep have the tendency for Compensatory Accelerated Production (CAP) of PCV in case of infection (Daramola *et al.*, 2005). Compensatory accelerated production has been shown to return PCV to normal following an infection (Dargie and Allonby, 1975).

Total WBC counts for both groups were not significantly different ($p>0.05$). Ewes in the treated group had a total WBC count of $7.20 \times 10^3/\mu\text{L}$ while those in the control group had $7.38 \times 10^3/\mu\text{L}$. The values obtained from this study are considerably lower than the value of $11.7 \times 10^3/\mu\text{L}$ reported by Taiwo and Ogunsami (2003) for WAD sheep in South-Western Nigeria. Such disparities in WBC count of the same breed of sheep, but in different eco-zones may not be unconnected to the prevalence of such diseases like pestes des petit ruminante and trypanosomiasis in the South-Eastern zone. The leucocytosis seen could be the basis for the adaptation of these ewes to this eco-zone; hence they are termed "trypanotolerant".

The liver enzymes AST, ALT and ALP showed no significant difference ($p>0.05$) for both groups of ewes. AST values were 10.67 and 10.50 IU L⁻¹ for control and treated groups, ALT values were 7.33 and 7.50 IU L⁻¹ while ALP values were 49.67 and 49.67 IU L⁻¹ for the control and treated groups, respectively. Analyses of these enzyme activities give valuable diagnostic information for a number of disease conditions. ALT and AST assay are important in the diagnosis of liver damage caused by drug toxicity or harmful chemicals (Nelson and Cox, 2005). ALP assay is useful in the diagnosis of obstructive liver diseases (Murray *et al.*, 2003). The liver is the organ involved in the detoxification of xenobiotics and other harmful chemicals that gain entrance into the body (Murray *et al.*, 2003). Thus, very toxic xenobiotics put a lot of stress on the liver, hence an increase in liver enzyme activity. In this regard, *S. mombin* can be considered safe for the lactating ewe, as there were no significant differences in the liver enzyme activity of both groups of ewes.

The tests for kidney function were not significantly difference ($p>0.05$) for urea, sodium, potassium, chlorine and bicarbonate ions. However, creatinine concentration in the sera of ewes fed *Spondias mombin* was significantly lower ($p<0.05$) than that of ewes in the control group. The values were 0.65 mg dL⁻¹ for the

treated ewes and 0.73 mg dL⁻¹ for the control. These values lie within the range reported by Taiwo and Ogunsami (2003) which is 0.70 mg dL⁻¹. Creatinine is a waste product of metabolism (Waugh and Grant, 2001). Specifically, it is a biosynthetic product of the amino acids arginine and glycine, with methionine, in the form of S-adenosylmethionine acting as the methyl donor (Nelson and Cox, 2005). The low creatinine level, therefore, could be an indication of a poor utilization of the crude protein in the diets of the ewes fed *S. mombin*. This could be as a result of anti nutritional factors (ANFs) present in *S. mombin*. These ANFs like the tannins may form protein-tannin complexes that inhibit enzymatic digestion of protein to form amino acids, leading to a reduction in protein digestibility (Apata and Ologhobo, 1998). Hence reduced amino acid production and reduction in creatinine levels in these ewes.

CONCLUSION

Spondias mombin L. (Hog plum), can be used by local farmers to improve on the milk yield of the lactating WAD ewes. It could be said to have a wide safety margin for the lactating ewe. However, its safety can be improved upon by finding the best methods of reducing any anti-nutritional factor that it may contain to ensure its maximum utilization by the ewes.

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