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NUTRITIVE VALUE ASSESSMENT OF SOME SELECTED TROPICAL WEEDS USING *in vitro* GAS PRODUCTION TECHNIQUE

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The nutrient value of *Amaranthus spinosus* (AMT), *Aspilia africana* (ASA), *Gomphrena celosiodes* (GOC), *Ipomea tribola* (IPT), *Luffa cylindrca* (LUF), *Plastosoma africanum* (PTA) and *Sida acuta* (SID) were evaluated using standard methods. The gas production from the fermentation of the weed samples and the proximate composition of oven dried samples were investigated. The estimated organic matter digestibility (OMD), short chain fatty acid (SCFA) meterbolizable energy (ME) were derived from the results of the proximate composition and *in vitro* gas production at 24hr of incubation. The crude protein contents (%) ranged from 8.75 (AMT) to 12.9 (SID). The crude fiber contents differed significantly ($p< 0.05$) among the investigated weed samples with the highest value obtained in GOC (14.70) and the least recorded in ASA (8.35). The ether extracts (%) were 1.65 (AMT), 2.21 (SID), 1.33 (GOC), 1.59 (IPT), 1.43 (LUF), 1.67 (PTA) and 2.12 (SID). The insoluble but degradable fraction (b,ml) was highest in GOC (38.0) followed by ASA (37.0). The estimated OMD (%), ME (MJ/Kg DM) and SCFA (mmol) ranged from 49.99 (SID) to 68.94 (ASA), 7.86 (IPT) to 12.31 (GOC), and 0.61 (SID) to 1.18 (GOC) respectively. The most abundant mineral in the investigated weeds are potassium and sodium. Phytate and oxalate contents were generally low, ranging from (2.0 to 3.0) and 1.0 to 0.3 respectively. From the results obtained in this study, GOC, ASA, IPT, LUF and SID were potential sources of protein with a correspondence high estimated OMD, ME and SCFA. Therefore it could be incorporated in complete feed mixtures in ruminant diets without major problems.

Keywords: Fermentation, *In vitro*, weeds, mineral, proximate composition.

Livestock feeds are necessary both in quantity and quality for a successful and hitch free animal production. Inadequacy of feedstuff throughout the year has been the major challenges in the livestock industry. Bamikole and Babayemi, (2004) reported that inadequate nutrition is one of the factors that generally affect livestock productivity.
Nigeria has a wide diversity of climatic zones from the humid forest zone of the south to the very dry sahel region of the North and mountainous cool belt of the plateau in the middle belt region (Akewusola *et al.*, 2007). Each of these climatic zones is richly blessed with varieties of forages, weeds, shrubs and browse plants. Despite the natural endowment with rich and green vegetation, especially in the Southern part of Nigeria, the inadequacy of feed and low animal productivity still remains a challenge. In both the Southern and Northern part of Nigeria, the livestock farmers has the issue of feed supply to contend with vis-à-vis the stress imposed on the livestock by the shortage. Such stress is often seen in the progressive loss in body weight of animals; fall in milk production, generally low productivity, and in extreme cases, loss of animals. In such extreme cases, the possible alternative is conventional feedstuff which is usually expensive in the developing. The *in vitro* gas production technique has been successfully used to evaluate feedstuffs and has thus remained a better alternative for the determination of gas production kinetics and energy value of ruminant (Isah *et al*, 2012). This is because *in vitro* gas production method is cheap, easy to handle, less expensive (Akinfemi *et al*., 2009), accurate

for the estimation of the organic matter digestibility (Makker *et al.,* 1999; Aiple *et al.*, 1996**;** Ajayi and Babayemi 2008). Tolera *et al.*, (1997) and Larbi *et al.*, (1998) affirmed that the technique is reliably useful for determining the relationship between chemical composition, incubation time and digestibility of forage.

Additionally, *in vitro* method is preferably used to predict dry matter intake (Blummel and Qrskov, 1993), digestibility (Khazaal *et al.*, 1993) and metabolisable energy (Babayemi and Bamikole, 2006) and for determining the rate or extent of gas degradation (Akinfemi *et al.*, 2009).

The use of *in vitro* gas production method has been successfully used to assess the nutritional value of few selected browse plants (Ladipo and Akinfemi, 2014) and also used to evaluate the nutritional potential of some selected weed species in North Central Nigeria (Akinfemi and Ladipo, 2014). In view of this, the study was designed to evaluate the nutritional potential of some selected tropical weeds found in Southern Nigeria.

MATERIALS AND METHODS

Sample collection

Samples of *Amaranthus spinosus* (AMT), *Aspilia africana* (ASA), *Gomphrena celosiodes* (GOC), *Ipomea tribola* (IPT), *Luffa cylindrca* (LUF), *Plastosoma africanum* (PTA) and *Sida acuta* (SID) were collected in southern Nigeria. The weeds were collected in late raining season (October) when they have attained full maturity. The samples were milled and oven-treated at 65^0C until a constant weight was obtained for dry matter determination.

In vitro gas production

Rumen fluid was obtained from three West African Dwarf female goats through suction tube before the morning feed. The animals were fed with 40% concentrate feed (40% corn, 10% wheat offal, 10% palm kernel cake, 20% groundnut cake, 5% soybean meal, 10% brewers grain, 1% common salt, 3.75% oyster shell and 0.25% fishmeal) and 60% Guinea grass. Incubation was carried out using standard methods (Menke and Steingass 1988) in 120ml calibrated syringes in three batches at 39^0C. To 200mg sample in the syringe was added 30ml inoculum that contained cheese cloth strained rumen liquor and buffer (9.8g $NaHCO_3$ + 2.77g Na_2HPO_4 + 0.57g KCL + 0.47g NaCL + 0.12g $MgSO_4$. $7H_20$ + 0.16g $CaCI_2$. $2H_20$ in a ratio (1:4 v/v) under continuous flushing with CO_2. The gas production was measured at 3, 6, 9, 12, 15, 18, 21 and 24h. The average volume of gas produced from the blanks was deducted from the volume of gas produced per sample. The gas production characteristics were estimated using the equation $Y = a + b (1 – e^{ct})$ (Órskov and McDonald, 1979), where Y = volume of gas produced at time't', a = intercept (gas produced from the soluble fraction), b = gas production from the insoluble fraction, (a+b) = final gas produced, c = gas production rate constant for the insoluble fraction (b), t = incubation time. The post incubation parameters such as metabolizable energy (ME, MJ/Kg DM), organic matter digestibility (OMD %) and short chain fatty acids (SCFA) were estimated at 24h post gas collection (Menke and Steingas 1988).

ME = 2.20 + 0.136* Gv + 0.057* CP + 0.0029*CF; OMD = 14.88 + 0.88Gv + 0.45CP +0.651XA; SCFA = 0.0239*Gv – 0.0601;

Where Gv, CP, CF and XA are net gas production (ml/200mg, DM) at 24 h incubation time crude protein, crude fibre and ash of the incubated sample respectively.

Chemical composition

DM was determined by oven drying the milled samples to a constant weight at 105^0C for 8 hours. Crude protein was determined as Kjadhal nitrogen x 6.25. Ether extracts and ash were determined according to (AOAC 1995) method. Neutral detergent fibre (NDF), Acid detergent fibre (ADF) and Acid detergent lignin (ADL) was determined using the method described by Van Soest et al. (1991).Hemicellulose was calculated as the difference between NDF and ADF while cellulose is the difference between ADF and ADL. Anti-nutritional properties like saponin were determined by gravimetric method, oxalate by permanganate titrimetric method, and tannins by Folin-Dewis spectophotometric method and cyanogens (Onwuka, 2005). The mineral elemental

constituents (Ca, Mg,Fe, Zn, K, Na, Mn, P and S) in seeds were analyzed separately, using atomic absorption spectrophotometer (Hitachi 26100 model) after acid digestion of the samples.

Statistical analysis

Data obtained were subjected to analysis of variance (ANOVA) and mean separation was by Duncan multiple range tests using Statistical Analysis System (SAS), 1999 package

RESULTS AND DISCUSSION

Presented in the Table 1 is the proximate composition (g/100g DM) and crude fiber fractions of some selected tropical weeds. All the selected tropical weeds had crude protein value (%) ranging from 8.75(*Amaranthus spinosus*) to 12.90 (*Sida acuta*). The crude protein values from the examined tropical weeds were above the critical range of 8 to 10%. Generally the CP values were below the values reported elsewhere (Isah *et al.,* 2012) for some selected browse plants. The dry matter content obtained in the present study compared favourably with earlier reports (Oduozo and Adegbola 1992 and Okoli *et al.,* 2001; 2003).

However, the DM differed from those reported by Mecha and Adegbola (1980) (14.48 – 55.22%) and Bellow (2003) (8.32 – 40.0%). The differences in the dry matter contents of the selected weeds with the earlier results reported could be due to the processing methods adopted, period of establishment (wet or dry season) or harvesting of the forage plant (Ajayi, 2012).

Previous studies (Akinfemi and Ladipo, 2014), reported a crude protein range of 8.17 to 18.80% for some selected tropical weeds in North Central Nigeria. The CP values reported for *Amaranthus spinosus* (8.75%), *Gomphrena celosiodes* (9.63%) and *Plastosoma Africana* (9.75%) is comparable with values reported elsewhere (Akinfemi and Ladipo 2014) for *Acroleras zizanoides* (8.17%), *Gomphrena celosiodes* (9.21%) and *Mariscus alternifolius* (9.58%). Wide variations were observed in the cell wall constituents (NDF, ADF and ADF) with significant variation among the weeds investigated. The NDF and ADF contents were highest in *Ipomea tribola* while the highest ADL content was recorded for *Plastosoma Africana*. Stage of growth, species or variety are some of the reasons reported for variations in cell wall constituents (Von Keyserlingk et al 1996; Agbagbla – Dohnami *et al.,* 2001; Promkof and Wanapat 2004), others (Mupagwa *et al.*, 1997) indicated that method of drying and growth methods could be responsible for such variations. Whereas, Thu and Preston (1999) indicated soil types as a possible reason responsible for variation in the chemical composition obtained in the present study.

The value obtained for PDMI in the present study showed an inherent potential that can improve the performance characteristics of small ruminates in free range in Western

Table 1: Proximate Composition (g/100gDm) and Crude Fiber Fractions of Seven Selected Tropical Weeds

Parameters	AMT	ASA	GOC	IPT	LUF	PTA	SID	SEM
CP	8.75^{e}	11.76^{b}	9.63^{d}	10.50^{c}	10.65^{c}	9.75^{d}	12.59^{a}	0.06
CF	11.50^{e}	8.35^{e}	14.70^{a}	12.67^{b}	9.22^{d}	12.50^{b}	8.47^{e}	0.07
Fat	1.65^{c}	2.21^{a}	1.33^{f}	1.59^{d}	1.43^{e}	1.67^{c}	2.12^{b}	0.01
Ash	11.25^{ab}	8.00^{b}	11.00^{ab}	8.50^{ab}	8.10^{b}	12.70^{a}	7.01^{b}	0.75
NDF	14.68^{d}	10.33^{f}	17.90^{b}	19.52^{a}	14.92^{d}	16.65^{c}	11.19^{e}	0.07
ADF	13.48^{d}	9.43^{f}	16.29^{b}	18.56^{a}	13.28^{d}	15.59^{c}	10.54^{e}	0.12
ADL	12.30^{cd}	8.45^{e}	15.36^{bc}	17.29^{ab}	12.31^{dc}	19.46^{a}	9.65^{de}	0.61

$^{a\text{ - }f}$Means on the same row with different superscripts are significantly varied ($P < 0.05$)

SEM = Standard error of mean, AMT = *Amaranthus spinosus*, ASA = *Aspilia africana,* GOC *Gomphrena celosiodes*, LUF = *Luffa cylindrical*, IPT = *Ipomea tribola*, PTA = *Plastosoma africanum*, SID = *Sida acuta.*

Table 2: Major minerals (mg/100gDM) and trace minerals (ppm) composition of Seven selected weeds

Parameters	AMT	ASA	GOC	IPT	LUF	PTA	SID	SEM
Major Minerals								
Calcium	68.40^{d}	69.05^{c}	57.95^{f}	60.8^{e}	72.10^{b}	73.20^{a}	68.80^{cd}	0.09
Sodium	208.25^{d}	260.65^{a}	235.25^{b}	221.50^{c}	224.70^{c}	235.14^{b}	236.72^{b}	1.43
Potassium	346.13^{b}	373.43^{c}	270.63^{c}	225.93^{d}	348.15^{b}	346.37^{b}	354.6^{a}	0.71
Phosphorus	6.81^{a}	4.38^{cd}	4.37^{cd}	3.95^{d}	5.84^{b}	5.12^{bc}	4.62^{c}	0.14
Trace Minerals								
Iron	4.23^{e}	3.95^{f}	4.64^{b}	3.91^{f}	4.69^{a}	4.29^{d}	4.56^{c}	0.01
Zinc	5.12^{d}	4.35^{d}	4.67^{b}	4.24^{e}	4.54^{c}	3.82^{g}	3.95^{f}	0.006

Table 3: Secondary metabolite (%) of seven selected weeds

Parameters	AMT	ASA	GOC	IPT	LUF	PTA	SID	SEM
Tannin	58^{a}	39^{f}	50^{c}	43^{e}	47^{d}	45^{d}	54^{b}	0.016
Saponin	71^{a}	48^{e}	53^{d}	53^{d}	61^{b}	56^{c}	48^{e}	0.003
Phytate	2^{a}	3^{b}	2^{c}	2^{e}	2^{ed}	3^{a}	3^{b}	0.003
Oxalate	1^{d}	2^{bc}	2^{c}	1^{d}	1^{b}	3^{a}	2^{b}	0.001

Table 4: *In vitro* gas production characteristics and estimated organic matter digestibility ($^{0}/_{0}$), metabolisable energy (ME, MJ/Kg DM). Short Chain fatty acid (SCFA mmol), and fermentation of the insoluble but degradable fraction (b, ml)

Parameters	AMT	ASA	GOC	IPT	LUF	PTA	SID	SEM
OMD ($^{0}/_{0}$)	29.00^{e}	37.00^{b}	38.00^{a}	21.00^{f}	36.00^{c}	30.0^{a}	21.00^{f}	0.05
ME (MJ/KgDM)	58.15^{c}	68.94^{b}	72.60^{a}	50.92^{a}	66.73^{b}	61.32^{c}	49.99^{d}	0.67
SCFA (mmol)	9.20^{e}	11.69^{b}	12.31^{a}	7.86^{f}	11.31^{c}	9.59^{d}	7.67^{f}	0.06
b (ml)	0.80^{e}	1.11^{b}	1.18^{a}	0.63^{f}	1.06^{c}	0.85^{d}	0.61^{f}	0.01

part of Nigeria (Isha *et al*., 2012). The mineral composition of the investigated weed samples are depicted in Table 1. The values reported in the present study for potassium is below the (8 g/kg) recommended for lactating goats (NRC, 1981), similarly they were also lower than the maintenance requirement of 5g/kg for non – lactating goats (NRC 1981).

The most available mineral in the investigated weed samples is potassium (225. 93 mg/kg to 354.60 mg/kg). This observation is consistent with earlier reports (Afolabi *et al.*, 1985; Olaofe and Sanni, 1980) which indicated that potassium is the dominant mineral in Nigeria Agricultural products. The potassium contents obtained in the present study is generally low when compared with the values reported by Ilelaboye and Pikuda, (2009) for *Jatropha curcass*, *Trichosanthe cucumerina* and *Citrilus vulgaris*. The result obtained for sodium differed significantly among the treatments. The values observed is consistent with the observation of researcher (Chamberlian, 1955) who reported that tropical crops carry subnormal concentration of sodium, which they attributed to content of sodium in the soil.

The values of calcium (mg/ kg) obtained ranged from 57.95 (G*omphrena celosiodes*) to 73.30 (*Plastosoma africana*). Calcium plays an essential role in strengthening the tissues and bones of the body. In the present study, the calcium obtained is higher than those reported (Babayemi, 2006), for *Enterolobium cyclocarpum* but lower than those reported for lesser known crop seeds (Ilelaboye and Pikuda, 2009). However, the values were comparable to those reported for

four Dominant weeds (Akinfemi and Mako, 2012). Generally, the major minerals were within the range values previously reported (Mcdowell, 1985). The values were adequate to meet the requirement for growth, reproduction and milk in West Afircan sheep and goats (Babayemi, 2006). However the calcium and phosphorous ratio were not within the approval 1:1 to 2:1 range recommended (McDowell, 1985). Iron, manganese and zinc were extremely deficient in the present study. This therefore requires mineral fortification in the form of either salt lick or diet inclusions.

Data presented in Table 3 are the contents (%) of Tannins, Saponins, phytate and Oxalete of some selected tropical weeds. The contents of tannins (%) ranged from 39 to 58, saponin (%) 48 to 71, phytate (%) 2 to 3 and oxalate (%) 1 to 2. The variation in tannin contents observed in this study suggests differences in the nutritional quality of the weeds investigated. The tannin levels obtained were low when compared with those reported elsewhere (Ayo – Enwerem *et al.,* 2009) but higher than those reported by Okwu and Josiah (2006). Tannin concentration in plants increased as the season progressed and attributed it to factors such as rainfall, temperature and humidity which may be peculiar to different locations (Arthur *et al.,* 1992) others (Singh 1984: Oduguwa *et al.*, 1998) attribute the differences to season of planting, the stage at which these plants specie were harvested and differences in assay methodology of tannin (Reed, 1995). These reasons may have influenced the tannin level in this study, and it is tolerable to ruminant. The oxalate content of the selected weeds is within the range of 0.1 – 25 mg/kg reported in trees, shrubs, herbs and grasses (Onwuka, 1996). The oxalate content obtained in our present study is within the range of 2.36 to 3.24% reported for four selected weeds (Aknifemi and Mako, 2012)

The phytate range of 0.02 to 0.03 observed in the present study is within the range of 82.03 – 245 mg/g recorded for trees and 65.05 – 293.62 mg/g obtained for shrubs (Onwuka, 1996) in Southwest Nigeria. However (Akinfemi and Mako, 2012) reported higher phytate level of 1.18 to 1.34% for tropical weeds. Phytate interferes with the utilization of mineral elements by forming compounds with anions and protein (Akinmutimi and Abasiekong, 1997).

The saponin content of the weed was generally low, compared with results obtained elsewhere (Gestetner *et al.*, 1996). The low sapomin content recorded in this study may not likely affect their nutritional potentials to any significant extent.

Shown in Table 4 are the results of *in vitro* gas production characteristics and estimated ME, SCFA, OMD and PDMI. Fermentation of the soluble but degradable fraction (b,ml) ranged from 21 to 38. The values recorded were significantly different. The highest fermentation of the insoluble fraction (b) was observed in GOC followed by ASA, possibly influenced by the carbohydrate fraction readily available to the microbial population (Chumpawadee *et al.,* 2007). Others (Deaville and Givens 2001) have also reported that kinetics of gas production could be affected by carbohydrate fractions. The fastest rate of gas production (c ml/h) which was recorded in GOC, suggest that the soluble carbohydrate fraction were readily available to rumen microbial population (Chumpawadee *et al.,* 2007). This observation is constituent with earlier reports (Akinfemi and Mako, 2012). Slower rates, however, obtained in AMT and PTA was so because of unavailability of the feed samples to rumen microbes.

The estimated OMD (%) ranged from 49.99 to 72.60 with the highest value recorded for GOC. High estimated OMD were observed of high soluble carbohydrate in them. This suggests that the microbes in the rumen and animal have high nutrient uptake (chumpawadee, 2007). Estimated ME (MJ/ kg DM) differed significantly among the treatments. Menke and Steingass (1998) and Chenost et al, (1997) reported that the production of ME is more accurate when based on gas and chemical constituents measurements as compared to calculations based on chemical constituents only. There is a positive correlation between ME calculated from *in vitro* gas production together with CP and fat content with ME value of conventional feed measured *in vivo* (Menke and Steingas, 1998).

Variations observed in the gas production characteristics reported in this study may be traceable to differences in the proximate composition. Negative correction has been reported between NDF and ADF and the rate and extent of gas production Nsahlai *et al.*, 1994 and Larbi *et al.*, 1998.

The SCFA varied significantly among the different weeds investigated. The gas production from different classes of feeds incubated *in vitro* in buffered rumen fluid was closely related to the production of SCFA which was based on carbohydrate fermentation (Blummel *et al.*, 1990). Since the highest values recorded in SCFA were observed in GOC, LUF and ASA, this suggests more energy availability to the animal. This observation is consistent with previous studies (Akinfemi and Mako 2012; Akinfemi and Ladipo, 2014).

As the PDMI value increases, the NDF of the selected weeds decreases, so as the percentage of NDF increase in forage, animals consume more (Schwedes, 1994). This notwithstanding, supplementation at 2 to 3% body weight has been suggested for optimum weight in small ruminant (Osuhur *et al.*, 1991)

CONCLUSION

The *in vitro* gas production method of feed evaluation can be effectively used to determine the nutritive value of tropical weeds and to identify difference among their potential digestibility and also estimate their energy contents. From the results obtained in the present study, *Gomphrena celosiodes, Amaranthus spinosus and Luffa cylindrical* could be interesting alternative animal feed sources and valuable in the ruminant feed. It could be used in combination with other feed sources for ruminant production.

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