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# Nutrient and Anti-nutrient Components of Red Type Sorghum Indigenous to Ekpoma Area of Edo State as Influenced by Soaking Techniques

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### Authors' contributions

This work was carried out in collaboration between both authors. Author SOO designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Author DOO managed the analyses of the study and the literature searches. Both authors read and approved the final manuscript.

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## ABSTRACT

**Aim:** This study investigated the effects of different soaking techniques on the nutritional value and anti-nutrient substances of red type sorghum.

**Study Design:** Completely randomized design was used.

**Place and Duration of Study:** Department of Animal Science Central Laboratory, Ambrose Alli University, Ekpoma, Edo State of Nigeria between 10<sup>th</sup> and 27<sup>th</sup> of June, 2017.

**Methodology:** Raw sorghum grains were divided into five groups with the first group unsoaked (US), while others were soaked in water (SW), soaked in water/germination (SWG), soaked in wood ash (SWA) and soaked in wood ash/germination (SWAG). All the samples were analysed in triplicates for proximate and mineral compositions, fibre fractions and some anti-nutrient substances.

**Results:** The results revealed that soaking significantly ( $P \leq 0.05$ ) improved the proximate and mineral components, fibre fractions and reduced the anti-nutrient substances of the intact grains. The proximate, minerals, fibre fractions and percentage reduction of the anti-nutrient substances

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were better in SWA and SWAG samples compared to others.

**Conclusion:** Therefore, for a significant reduction of the anti-nutrient factors as well as the improvement and bioavailability of the component nutrients of red type sorghum, soaking in wood ash extract/germination is adequate.

*Keywords: Anti-nutrients; fibres; minerals; proximate; soaking; sorghum.*

## 1. INTRODUCTION

The present food insecurity in many developing countries and future challenges of feeding over nine billion people in 2050 [1] cannot be over emphasized. The demand for animal products is expected to be 58 and 70% higher in 2050 than the level in 2010 and a larger proportion of this increase will originate from developing countries [2]. Maize often constitutes the highest proportion of ingredient in diet formulation of any poultry ration and this translates to high cost of feed because of seasonality in the production and competition between man and livestock [3]. One of the major challenges of researchers in the tropics is the provision of alternative feed resources for monogastric animals [4] which can partially or completely replace these expensive conventional feed ingredients [5]. Thus, this study is focused on sorghum (*Sorghum bicolor*) L. Moench one of the most important weaning foods in low-income and high-income countries [6,7,8,9]. It is the king of cereals and is one of the important food crops in dry lands of tropical Africa, Indian and China as well as Egypt [10]. Sorghum is considered as one of the most adapted summer grain crops to drought and heat; therefore, more than 70% of cultivated areas with sorghum are located in Upper Egypt Assiut, Sohage and Fayoum. Sorghum cultivated area in Egypt is about 158,000 hectare producing 880,000 tons of grains with an average of 5.7 tons/ha [11].

The nutrient composition of sorghum indicated that it is a good source of energy, proteins, carbohydrates, vitamins and minerals including the trace elements, particularly iron and zinc, except calcium. Sorghum grain contains minerals such as phosphorus, potassium and magnesium in varying quantities [12]. It is grown for its seeds and had been limited in use due to the presence of anti-nutritional factors among which are tannins which are known to inhibit the activities of some enzymes like trypsin, amylase and lipase resulting in the formation of complexes with protein. It had been reported [13] that tannins affect the growth of animals by imparting astringent taste which affects palatability and

reduces feed consumption, form complexes with proteins and reduces its digestibility and act as enzyme inactivator. Phytates had also been known to lower bio-availability of minerals and inhibit plant food, depress growth rate, reduce feed efficiency and impair protein digestion [14]. Therefore, the reduction to safe or zero levels of the anti-nutritional factors inherent in sorghum through processing is essential in improving its nutritional quality with a view to tapping its full nutritional potentials effectively. Consequently, a 92% reduction of tannin content of a high-tannin sorghum cultivar subjected to fermentation had been reported [15], while 63 and 61% reduction for two cultivars of sorghum fermented for 14 hours were recorded [16]. Recently, [17] observed a significant improvement in the chemical and mineral compositions of fermented malted sorghum sprouts. This experiment was therefore designed to investigate the effect of different soaking techniques on some nutrient and anti-nutrient substances of red type sorghum.

## 2. MATERIALS AND METHODS

### 2.1 Experimental Site

The experiment was carried out in the Department of Animal Science Central Laboratory, Ambrose Alli University Ekpoma, Esan West Local Government Area of Edo State, Nigeria.

### 2.2 Sources of Raw Materials

The red type sorghum (*Sorghum bicolor*) L. Moench grains were purchased from the open market in Benin City and Ekpoma, while the wood ash was obtained from Ambrose Alli University dormitory kitchen.

### 2.3 Preparation of the Wood Ash Extract

Wood ash of 2 kg weight were dissolved in 10litres of water and allowed to stay for 48 hours. The wood ash solution was then filtered through cotton cloth to obtain the wood ash extract and

the pH was adjusted to 8.5 by using digital hand pH meter [18]. The pH meter (TOA pH Meter HM-7B, Tokyo, Japan) was standardized using buffer solutions of acidic and basic values of 4.01 and 9.08 at room temperature. The adjustment of the pH was done by adding 2 mole of hydrochloric acid in a ratio of 2 moles/2 L of extract to reduce the alkalinity of the medium. The electrode was rinsed with distilled water before taking measurements. The wood ash extract was homogenized by stirring to achieve uniformity and the pH readings were taken by dipping the electrode into the extract and measurements taken from the displayed screen as soon as the readings stabilized.

## **2.4 Processing of Raw Sorghum Grains**

The sorghum grains were divided into five parts. Part 1 was unsoaked/raw, while parts 2, 3, 4 and 5 were subjected to soaking in water, soaking in water/germination, soaking in wood ash extract and soaking in wood ash extract/germination respectively.

### **2.4.1 Soaking in water**

Two (2) kg of raw sorghum were soaked in water for 48 hours after which the seeds were sieved from the water and sundried on jute mat for 5 days at atmospheric temperature of between 26 – 29°C to prevent fermentation and achieve a moisture content of 12% and stored in plastic containers.

### **2.4.2 Soaking in water/germination**

The same quantity of sorghum seeds were soaked in water for 48 hours. Thereafter, the grains were sieved from the solution and spread on jute mat and allowed to germinate for five days. The grains were covered with a layer of fresh plantain leaves to preserve moisture. Thereafter, the malted seeds were sundried for 5 days at atmospheric temperature of between 26 – 29°C to prevent fermentation and achieve a moisture content of 12% and stored in plastic containers.

### **2.4.3 Soaking in wood ash extract**

Same quantity (2 kg) of sorghum seeds were soaked in wood ash extract for 48 hours, the seeds were then sieved from the extract and sundried on jute mat for 5 days at atmospheric temperature of between 26 – 29°C to prevent fermentation and achieve a moisture content of 12% and stored in plastic containers.

### **2.4.4 Soaking in wood ash extract/germination**

Two (2) kg of raw sorghum seeds were soaked in wood ash extract for 48 hours after which the seeds were sieved from the solution and spread on jute mat and allowed for germination for 5 days. During this period the soaked grains were covered with fresh plantain leaves to preserve moisture. Thereafter, the malted seeds were sundried for five days at atmospheric temperature of between 26 – 29°C to prevent fermentation and achieve a moisture content of 12% and stored in plastic containers. Thereafter, all the samples were ground in a hammer mill to pass through a sieve of 0.05 mm and stored in a polythene bag before they were used for laboratory analyses.

## **2.5 Chemical and Mineral Analyses**

The proximate compositions of the raw and processed samples were analyzed in triplicates using standard procedures [19]. The fibre fractions which included: neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin (ADL) were determined [20]. Cellulose was calculated as the difference between ADF and ADL, while hemicellulose was calculated as the difference between NDF and ADL. Quantitative determination of tannin, oxalate and phytate contents were determined [21,22]. The minerals: calcium (Ca), phosphorous (P), potassium (K), magnesium (Mg), sodium (Na), copper (Cu), zinc (Zn), iron (Fe) and manganese (Mn) were determined with the atomic absorption spectrophotometer model 420 [23]. Phosphorus in the digest was estimated with vanadomolybdate solution and the colour so developed was read with spectrophotometer at 420 m/u. The concentration of K was estimated with a flame photometer.

## **2.6 Statistical Analysis**

Data generated from the study were subjected to a one-way analysis of variance (ANOVA) and significant treatment means were compared using Duncan's multiple range test with SPSS package [24,25].

## **3. RESULTS AND DISCUSSION**

### **3.1 Proximate Composition of Differently Soaked Red Type Sorghum Grains**

The proximate compositions of the unsoaked/raw and soaked samples of the red type

sorghum seeds (Table 1) revealed that soaking significantly ( $P \leq 0.05$ ) affected the dry matter (DM), crude protein (CP), ash, ether extract and nitrogen free extract (NFE), but crude fibre (CF) was not affected ( $P \geq 0.05$ ). Percentage DM was highest ( $P \leq 0.05$ ) in the raw sample (89.66%) followed by SW (88.51%), while the values (87.65 and 87.84%) recorded in SWAG and SWA samples were ( $P \geq 0.05$ ) similar and least in SWG (87.05%). The crude protein content of the intact seeds was increased ( $P \leq 0.05$ ) by the soaking techniques with the highest value (11.52%) obtained in SWAG. SWG and SWA had similar ( $P \geq 0.05$ ) values of 11.42 and 11.35% respectively, followed by SW (10.42%) and lowest value was recorded in US (10.14%) sample. The high values recorded in the processed samples particularly the germinated ones could be due to the reduction of nitrates in the intact seeds into plant protein or nitrate compounds during germination. Besides, it might also be as a result of the effect of fermentation on the soaked grains due to the growth of microflora in the soaking medium. This observation contradicts [26] the report that fermentation process did not significantly change the total protein content and amino acid composition of substrate in African yam bean. However, the present report is in consonance with [12,27] who found out that the crude protein of whole sorghum grain ranged from 7.0 to 15.0% or 10.3 to 14.9%. Although, soaking techniques did not affect ( $P \geq 0.05$ ) the fibre content of the raw seeds but there was a numerical reduction of the fibre value from 3.63% in US to 2.74% in SWA. This observation might be due to the degradation of fibre initiated by endogenous enzymes during germination [28]. Higher ( $P \leq 0.05$ ) content of ash was recorded in SWAG (2.11%) followed by SWA (1.85%), SWG (1.03%), US (1.03%) and lowest in SW (0.90%). The ether extract of the raw seeds was progressively reduced ( $P \leq 0.05$ )

from 10.06% in US to 8.18% in SWAG. The NFE of US (64.54%) and SW (64.74%) were statistically ( $P \geq 0.05$ ) the same but differed ( $P \leq 0.05$ ) from those of SWAG (62.91%), SWA (62.57%) and SWG (62.31%).

### 3.2 Mineral Contents of Differently Soaked Red Type Sorghum Grains

Mineral contents of the samples (Table 2) showed significant ( $P \leq 0.05$ ) increase in the levels of Mn, Cu, Ca, P and decrease in the levels of Zn and Fe by the soaking techniques. While the values recorded for Mg and Na increased ( $P \leq 0.05$ ) in SW and SWA but decreased in SWG and SWAG samples. Similarly, K level was higher ( $P \leq 0.05$ ) in SWAG compared to others. The significant increases in the values of Mn, Cu, Ca and P in the processed samples compared to the US may be ascribed to the degradation of phytate, oxalate and tannin which bound with the minerals thereby making them unavailable in the intact seeds. These observations lend support from the report [29] that there was an increase in the mineral content of yellow maize after fermentation of the soaked seeds in water. However, the values recorded in this study were slightly varied from a previous report [17] which perhaps may be due to the fact that the sorghum grains were from different sources thereby leading to varietal differences.

### 3.3 Fibre Fractions of Differently Soaked Red Type Sorghum Grains

The results of the fibre fractions (Table 3) showed that NDF, ADF and ADL were markedly ( $P \leq 0.05$ ) reduced by the soaking techniques and this was more evident in SWA sample with 4.92 and 39.21 respectively for NDF and ADF as well as in SWAG with 18.92 for ADL.

**Table 1. Proximate composition of red type sorghum as affected by soaking techniques**

Proximate (%)	Soaking techniques					SEM $\pm$
	US	SW	SWG	SWA	SWAG	
Dry Matter	89.66 <sup>a</sup>	88.57 <sup>b</sup>	87.05 <sup>d</sup>	87.84 <sup>c</sup>	87.65 <sup>c</sup>	0.03
Ash	1.03 <sup>c</sup>	0.90 <sup>d</sup>	1.03 <sup>c</sup>	1.85 <sup>b</sup>	2.11 <sup>a</sup>	0.01
Crude Protein	10.42 <sup>c</sup>	10.14 <sup>d</sup>	11.42 <sup>b</sup>	11.35 <sup>b</sup>	11.57 <sup>a</sup>	0.03
Ether Extract	10.06 <sup>a</sup>	9.66 <sup>b</sup>	9.05 <sup>c</sup>	8.85 <sup>c</sup>	8.18 <sup>d</sup>	0.03
Crude Fibre	3.63	3.08	3.23	2.74	2.89	0.32
NFE	64.54 <sup>a</sup>	64.74 <sup>a</sup>	62.31 <sup>d</sup>	62.57 <sup>c</sup>	62.91 <sup>b</sup>	0.09

Means in the same row with varying superscripts differ significantly ( $P \leq 0.05$ )

SEM: Standard error of mean, US: Unsoaked sample, SW: soaked in water, SWG: soaked in water/germination, SWA: soaked in wood ash extract, SWAG: soaked in wood ash extract/germination

**Table 2. Mineral content of red type sorghum as affected by soaking techniques**

Minerals	Soaking techniques					SEM ±
	US	SW	SWG	SWA	SWAG	
Mn (%)	10.89 <sup>c</sup>	11.58 <sup>bc</sup>	12.34 <sup>b</sup>	12.15 <sup>b</sup>	18.37 <sup>a</sup>	0.34
Cu (ppm)	4.08 <sup>e</sup>	5.30 <sup>c</sup>	4.53 <sup>d</sup>	7.43 <sup>b</sup>	8.51 <sup>a</sup>	0.20
Zn (%)	21.32 <sup>a</sup>	19.67 <sup>a</sup>	19.58 <sup>a</sup>	14.28 <sup>b</sup>	20.56 <sup>a</sup>	0.92
Fe (ppm)	62.56 <sup>a</sup>	17.86 <sup>e</sup>	25.42 <sup>d</sup>	30.35 <sup>c</sup>	35.80 <sup>b</sup>	1.37
Mg (%)	128.79 <sup>bc</sup>	144.69 <sup>a</sup>	122.39 <sup>bc</sup>	135.13 <sup>ab</sup>	114.68 <sup>d</sup>	3.71
Ca (%)	59.64 <sup>e</sup>	126.13 <sup>a</sup>	99.03 <sup>d</sup>	105.98 <sup>c</sup>	114.89 <sup>b</sup>	0.50
Na (%)	649.46 <sup>c</sup>	711.82 <sup>b</sup>	639.70 <sup>c</sup>	805.98 <sup>a</sup>	511.47 <sup>d</sup>	6.02
K (%)	1629.39 <sup>b</sup>	1373.38 <sup>d</sup>	1333.63 <sup>e</sup>	1521.61 <sup>c</sup>	1942.10 <sup>a</sup>	7.75
P (%)	232.35 <sup>d</sup>	560.07 <sup>b</sup>	340.73 <sup>c</sup>	783.15 <sup>a</sup>	768.46 <sup>a</sup>	11.75

Means in the same row with varying superscripts differ significantly ( $P \leq 0.05$ )

SEM: Standard error of mean, US: Unsoaked sample, SW: soaked in water, SWG: soaked in water/germination, SWA: soaked in wood ash extract, SWAG: soaked in wood ash extract/germination

Similarly, the cellulose contents of the raw samples were significantly ( $P \leq 0.05$ ) reduced by the soaking methods with the lowest value recorded in SWA sample (20.29) compared to other soaked samples. On the contrary, highest ( $P \leq 0.05$ ) value (38.12) for hemicellulose was obtained in SWG sample compared to others. While, the values recorded for US (36.30) and SWAG (35.57) samples were similar, followed by SW (34.99) and least (31.50) in SWA sample. The observed reduction in NDF, ADF, ADL, hemicellulose and cellulose by the soaking techniques can be adduced to the activities of some fibre degrading microorganisms such as *Fibrobacter succinogenes* and *Bacillus* species [30]. The present study is in consonance with the report [31] that a significant reduction in the fibre fractions of sweet orange fruit peels soaked in water was recorded.

### 3.4 Anti-nutrient Substances of Differently Soaked Red Type Sorghum Grains

The phytates, oxalates, tannins and trypsin inhibitors activities of raw sorghum grains were influenced ( $P \leq 0.05$ ) by the soaking techniques (Table 4). SWAG significantly

reduced the phytate level of the raw seeds by 19.91%, followed by 17.29, 15.91 and 5.60% reductions in SWG, SWA and SW samples respectively. On the contrary, the highest ( $P \leq 0.05$ ) percentage reduction of oxalate was recorded in SWA sample (60.39%) followed by SW, SWG and SWAG samples with respective values of 53.96, 20.13 and 9.74%. Although, the tannin level of the intact seeds was not affected ( $P \geq 0.05$ ) by SW (0.00%) and SWG (3.23%) but SWA (32.26%) and SWAG (32.26%) significantly ( $P \leq 0.05$ ) reduced it. The non-significant effect of SW and SWG on the tannin content of the raw seeds contradict the reports [32,33] that soaking and fermentation processes reduced the tannin content of sorghum grains. However, the 32.26 and 32.21% reduction of tannin content in SWA and SWAG respectively were lower than the 84.50% reduction of tannin level in malted white sorghum [34]. The reduction in tannin contents in the soaked samples might be due to the leaching out of the polyphenols into the soaked medium since polyphenolic compounds are water soluble and mostly located in the seed coat. Trypsin inhibitor activity of the raw sample was reduced ( $P \leq 0.05$ ) by 77.36, 77.36, 69.81 and 43.40% in SWAG, SWA, SWG and SW respectively.

**Table 3. Fibre fractions of red type sorghum as affected by soaking techniques**

Fibre fraction	Soaking techniques					SEM ±
	US	SW	SWG	SWA	SWAG	
NDF	56.63 <sup>b</sup>	55.12 <sup>c</sup>	56.84 <sup>a</sup>	49.92 <sup>e</sup>	53.13 <sup>d</sup>	0.03
ADF	49.51 <sup>a</sup>	47.38 <sup>b</sup>	45.92 <sup>c</sup>	39.21 <sup>e</sup>	45.80 <sup>d</sup>	0.01
ADL	20.33 <sup>a</sup>	20.13 <sup>a</sup>	18.72 <sup>b</sup>	18.92 <sup>b</sup>	17.56 <sup>c</sup>	0.05
Hemicellulose	4.13 <sup>ab</sup>	7.74 <sup>ab</sup>	7.35 <sup>a</sup>	10.71 <sup>a</sup>	10.92 <sup>ab</sup>	1.88
Cellulose	29.18 <sup>a</sup>	27.25 <sup>d</sup>	27.38 <sup>c</sup>	20.30 <sup>e</sup>	28.25 <sup>b</sup>	0.03

Means in the same row with varying superscripts differ significantly ( $P \leq 0.05$ )

SEM: Standard error of mean, US: Unsoaked sample, SW: soaked in water, SWG: soaked in water/germination, SWA: soaked in wood ash extract, SWAG: soaked in wood ash extract/germination

**Table 4. Anti-nutrient substances of red type sorghum as affected by soaking techniques**

Components	Soaking techniques					SEM ±
	US	SW	SWG	SWA	SWAG	
Phytate (mg/100 g)	59.7 <sup>a</sup>	56.42 <sup>b</sup>	49.47 <sup>d</sup>	50.25 <sup>c</sup>	48.21 <sup>e</sup>	0.03
Oxalate (mg/g)	1.54 <sup>a</sup>	0.71 <sup>d</sup>	1.23 <sup>c</sup>	0.61 <sup>e</sup>	1.39 <sup>b</sup>	0.02
Tannin (mg/100 g)	0.31 <sup>a</sup>	0.31 <sup>a</sup>	0.30 <sup>a</sup>	0.21 <sup>b</sup>	0.21 <sup>b</sup>	0.01
Trypsin (mg/g)	0.53 <sup>a</sup>	0.30 <sup>b</sup>	0.16 <sup>c</sup>	0.12 <sup>d</sup>	0.12 <sup>d</sup>	0.01

Means in the same row with varying superscripts differ significantly ( $P \leq 0.05$ )

SEM: Standard error of mean, US: Unsoaked sample, SW: soaked in water, SWG: soaked in water/germination, SWA: soaked in wood ash extract, SWAG: soaked in wood ash extract/germination

The percentage reduction of trypsin inhibitor activities in the intact seeds by SWAG, SWA, SWG and SW were higher than the 36.5% recorded in malted white sorghum grains [34]. Generally, the reduction in the anti-nutrient substances in the intact seeds by the soaking techniques might be as a result of the leaching of these anti-nutrient substances into the soaking medium which conformed with the previous study [35] that soaking cowpea in both acidic and alkaline solutions led to decrease in phytic acid.

#### 4. CONCLUSION

The results from this study revealed that soaking methods significantly improved the nutrients and reduced the levels of anti-nutrient substances in the intact red type sorghum grains. However, these observations were more evident in sorghum grains subjected to soaking in wood ash extract/germination (SWAG). Therefore, to enhance bioavailability and utilization of nutrients in red type sorghum grains, soaking in wood ash extract/ germination is recommended.

#### COMPETING INTERESTS

Authors have declared that no competing interests exist.

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