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ASSESSMENT OF NUTRIENT POTENTIAL, MINERAL CONTENT AND AMINO ACID COMPOSITION OF *Thaumatococcus daniellii* LEAF PROTEIN CONCENTRATES.

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ABSTRACT

Freshly harvested Thaumatococcos daniellii, was plucked and processed for its vegetable leaf protein concentrates with a view to evaluate its proximate constituents, amino acid content and mineral composition. Proximate analysis was determined using standard analytical technique. The nutrient composition of the protein concentrates revealed; the moisture content (9.94±0.01), crude fat (6.69±0.23), crude fibre (13.06 ± 0.17), crude protein (52.07 ± 0.20), ash (15.10 ± 0.13) and Carbohydrate (1.12±0.43). The mineral content of the sample indicated that Ca, Mg, K, and Na are the most abundant minerals with the following values Na;70.6±0.42,Ca;19.70±0.28, K; 90.3±0.42, Mg; 103.9±0.76, other minerals that were present in the sample in trace concentration are Fe(2.00 ± 0.46), Zn (2.90± 1.06), Mn (2.50± 0.12), Cu (0.2± 0.58), Pb (0.1± 0.44),while selenium were not detected in the sample indicating that the leaf concentrate is fit for dietary consumption. The amino acid profile reveals favourable nutritional balance with the presence of essential and nonessential amino acids except that tryptophan which was believed to be predominant in animal protein was not detected.

KEY WORDS: *Thaumatococcus daniellii,* Leaf protein concentrates, Amino acid, Proximate analysis

RESUMO

Foi efetuada a colheita de folhas frescas de Thaumatococcus danielli com o intúito de avaliar a concentração de proteínas, aminoácidos e a composição mineral. A análise próxima foi determinada usando métodos analíticos padrão A composição nutritivas dos extratos concentrados de proteína apresentou os seguintes dados: conteúdo de umidade (9.94), gordura bruta (6.69), fibra bruta (13,06), proteína bruta (52.07), cinza (15.10) e carboidratos (1.12). A composição mineral das amostras indicou que Ca, Mg, K e Na foram os elementos mais abundantes com os seguintes valores : Na (70.6), Ca (19.70), K (90.3) e Mg (103.9). Outros elementos presentes em quantidade menores foram Fe (2.00), Zn (2.90), Mn (2.50), Cu (0.2) e Pb (0.1). Selênio não foi detectado nas amostras, indicando que os concentrados das folhas são adequados para alimentação. O perfil dos aminoácidos mostra uma balança nutricional adequada com a presença de aminoácidos essências e não-essenciais., O triptofano, que é predominante em proteína animal na foi detectado.

PALAVRAS CHAVE: *Thaumatococcus danielli,* Concentrado de proteínas das folhas, Aminoácidos, Análise Próxima

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INTRODUCTION

INTRODUCTION

Leaf protein concentrates contain protein prepared from disrupted plant cell, further processed into green chloroplastic or white cytoplasmic protein concentrates using heat coagulation. Different plant species has been used by various authors ranging from green vegetables, medicinal plant and some trees of which fruits and leaves have viable ingredient for food and other uses by man.

The need for use of leaf protein concentrates as food for man and animal arise; due to an accelerated food demand with the exponential human population growth resulting in marginal land resource availability for growing food crops especially vegetables [23]. The rapid population growth in most African countries (Nigeria inclusive) has led to serious food crises, especially among the vulnerable groups such as the weanling, pre-school children, pregnant or nursing mothers, etc. This class of people are particularly prone to dietary protein, mineral and vitamin inadequacies. The dietary inadequacies which arise mainly from the high cost of animal proteins (milk, egg and meat) have, in some developing countries resulted in kwashiorkor, marasmus, infant blindness, mortality and morbidity.

Futhermore, this ever widening food shortage cannot be alleviated by conventional agriculture alone. As an additional source of protein is required and leaf protein concentrates should be given serious attention because leaves are abundant all the year round in the tropics and many have high protein content with suitable plant material.

Thaumatococcus danielli is a plant species from Africa, known for being the natural source of thaumatin, an intensely sweet protein which is of interest in development of sweeteners widely used for different industrial and domestic purposes. The plant has a number of uses besides flavouring. The leaves of this plant has found application in wrapping food because of

the thaumatin which the leaves gives food, the petiole is used to weave mats and as tools used for building materials. The entire leaf is also used for roofing while the leaf sap is used as antidote against venoms and bites, leaf and root's sap are used as sedative and for treating sanity. *Thaumatococcus danielli* is an economic plant with versatile uses especially in southern Nigeria [12].

However, protein are of prime importance to health and are often deficient in the diets of people in developing countries especially those in vulnerable groups such as nursing mothers, expectant mothers, weanlings and pre-school children. In addition, deficiencies in protein observed in the diet of people in some developing countries may result in serious health problems, this is due to the fact that protein from animal sources are very expensive and are becoming in adequate to cope with teaming population year after year [15].

As a result of in adequacies of protein from animal sources coupled with population explosion leading to malnutrition and wide spread deficiency diseases, Nutritionist are researching on suitability of some plants and green vegetables that has promising values as a means of replacing proteins from animal sources.

The search through the literature about the economic importance of *Thaumatococcus daniellii* reveals numerous benefit derivable from the leaves, saps, stem and root of this plant by various researcher. Despite the fact that the anti-nutrient components (terpenoids, steroids, saponins, phlobanins and tannins) for this leaves, fruit and roots has been determined to be very low [35]. But, the leaf protein concentrates of this plant has not been given prominent attention. It is the objective of this paper therefore to determine the nutrition potential, mineral content and amino acid composition of the leaf protein concentrates of *Thaumatococcos daniellii*.

MATERIALS AND METHODS

Preparation of Sample; Fresh Broad, large sized leaves of *Thaumatococcus daniellii* were obtained from abandoned farm land in Iware town of Oyo State Nigeria. The leaves were washed with distilled water and pulped by passing it through the locally produced mincer (technically referred to as cell rupture). The pulp was collected and strained through a cotton cloth followed by screw press. The green juice obtained from straining the pulp through the cotton cloth, was heated between 85^0C 90^0C by steam injection, which resulted in coagulation of all the protein present within the pulp. The coagulum was then centrifuge from the rest of the solution, pressed, pulverized and air-dried prior chemical analysis.

Proximate analysis: The proximate analysis of the air dried sample of *Thaumatococcus daniellii* was determined by the official method of the Association of Official and Analytical Chemists [9]. To determine moisture content, crude protein, crude fat and crude fibre while Nitrogen free extract (NFE) was calculated by difference.

Analysis of mineral content; Five grams (5g) of the sample was ashed in a muffle finance at 550^0C for 12 hours the resulting ash was cooled in a desiccator. The ash was dissolved in 2ml of concentrated HCl and few drops of concentrated HNO_3 were added, the resulting solution was evaporated almost to dryness in water bath. The content was diluted to the mark level in 100ml volumetric flask with distilled water. Bulk Scientific Atomic Absorption Spectrophotometer was used to determine each metals reported for the sample after the appropriate dilutions were made for each element.

Amino Acid analysis: The amino acids were determined by using modified method [36], by loading the sample into Technicon Sequential Multi-sample Amino-acid analyser after the sample has been defatted using

40% petroleum ether, followed by hydrolysis using 6M HCl and evaporated in rotary evaporator.

Results and discussion

Table1: Proximate composition of *Thaumatococcus daniellii* (g/100g)

PARAMETER	VALUE (g/100g)
Moisture	9.94±0.01
Ash	15.10 ± 0.13
Crude fat	6.69±0.23
Crude protein	52.07 ± 0.20
Crude Fibre	13.06 ± 0.17
NFE	1.12±0.43

The results of proximate composition of *Thaumatococcus danielli* leaf protein concentrates are contained in Table 1. The sample contained 9.94±0.01g/100g. Moisture content. The moisture content of food determines the keeping quality and influences the rate of food absorption and digestion. The value reported for this sample indicated that the sample is less prone to deterioration.

The value of moisture content is higher than those reported for *Amaranthus hybridus* (7.6±0.6g/100g) and *Telfairia occidentalis* (6.6± 0.6g/100g) leaf protein concentrates [6]. The value is lower than 10.67± 0.03g/100g reported for dried leaf of *Thaumatococcus daniellii* [34]. The low moisture content of the samples means that there is a concentration of solutes and decreased ability to perishability [18].

The total ash content of *Thaumatococcus daniellii* leaf protein concentrates is 15.10±0.13g/100g. the value is lower than 17.21±0.03g/100g reported dried leaf of the same sample [27] but the

higher than 11.60% and 11.37% reported for the ash content of food is a useful index two varieties of *Ipomea batatas* leaf samples respectively to express the total mineral content of plant tissue [31], [20]. A high ash content means that the mineral content of food is also high.

According to [20], the most abundant mineral elements in plants are potassium, calcium, magnesium, iron, phosphorus, sulphur and Nitrogen. This means that sample with high ash content (high total mineral content of the plant) are good in treating or proven thing malnourishment. The result obtained for the crude ash content of this indigenous leaf protein concentrates is in agreement with those reported in literature for some green leafy vegetables [41] and [8].

Crude fat content is 6.69±0.23g/100g. The value fall in range with 6.80± 0.1g/100g and 6.81±0.49g/100g reported for *Solanum microcarpon* and *Cochorus olitorius* respectively [1]. Fat in food determines the amount of energy available dietary fats function in the increase of palatability of food by absorbing and retaining flavours. In addition, a diet providing 1-2% of fat is said to provide caloric energy sufficient to human beings [13], [26].

The Crude fibre value of *Thaumatococcus danielli* leaf protein concentrate is 13.06±0.17g/100g the values is higher than 1.7g/100g and 1.6g/100g reported [4] for *Amaranthus hybridus* and *Telfaira occidentalis* respectively. The value however is lower than 28.6g/100g reported for *Amaranthus cruentus* [30]. It has been reported [5] that non starch vegetable are the richest sources of dietary fibre and it is helpful in the treatment of diseases such as obesity, diabetes and gastro-intestinal disorders [33]. This makes *Thaumatococcus danielli* more favourable to be consumed as food or food ingredients since high fibre content of foods help

in digestion, prevention of constipation and prevention of colon cancer [37].

Crude Protein

Crude protein content of the samples is 52.07±0.20. The value is higher than 24.85g/100g reported for sweet potatoe leaf protein concentrate [6]. The value is also higher than 21.06± 0.12g/100g reported for dried leaf of *Thaumatococcus danielli* [34]. The value is also higher than crude protein levels reported for lentil, cowpea and pigeon pea which are highly recommended as substitute for animal protein [24]. Protein in food is required for component of every living tissue. This reported value indicates that *Thaumatococcus danielli* is a good source of protein.

Nitrogen free Extract (Crude Carbohydrate); Crude carbohydrate content of this sample is very low (1.12±0.43g/100g). Compared to 37.27±1.14g/100g reported for dried leaf of the sample [34]. The value is also low compared to 23.58±3.64g/100g reported for *Vernonia amygdalina* [35]. The low carbohydrate content of *Thaumatococcus danielli* leaf protein concentrates means that it is more suitable for those who want to cut down on carbohydrate intake and for the obese who need less carbohydrate in their diet. This is because excess glucose, which is the sub unit of carbohydrate [27], in the body is converted to fat, which in the end leads to obesity [40]. The leaf protein concentrates will also be good for diabetics who need less sugar or glucose in their diet [14].

Table 2. Concentration of mineral element in *Thaumatoccocus danielli* leaf protein concentrates (mg/100g).

Mineral Element	Concentration (mg/100g)
Fe	2.00± 0.46
Zn	2.90± 1.06
Mg	103.9 ± 0.76
K	90.3±0.42
Na	70.6± 0.42
Pb	0.1 ± 0.44
Mn	2.50± 0.12
Ca	1.70 ± 0.28
Se	0.1 ± 0.97
Cu	0.2 ± 0.58

The results of mineral analysis of *Thaumatococcus daniellii* were presented in Table 2.

The concentration of Iron in this sample is 2.00±0.46mg/100g. The recommended dietary allowance of iron in adult and children is 10mg per day while female adult is 15mg per day. The value obtained for this samples is lower than the recommended dietary allowance. The value is however higher than 0.01±0.60 reported for dried leaf of *Thaumtococcus daniellii* by shalom et al 2014, but fell in range with 2.3±0.42 reported for *Vernonia amygdalina* leaf protein concentrates [35]. Iron in food is required for blood (heamoglobin) formation [2].

Zinc concentration is 2.90±1.06mg/100g. The value is higher compared to 0.02mg/100g reported for *Diospyrus mespilliformis* [39], but lower when compared to 6.85±1.00 reported [16] for *Amaranthus cruenthus*. The recommended daily allowance of Zinc is 12-15mg per day [29]. This indicate that *Thaumatococcus daniellii*. Leaf protein concentrate is a poor source of dietary Zinc. Zinc plays a vital role in gene expression regulation of cellular growth and participates as a cofactor of enzymes responsible metabolism of carbohydrates, proteins and nuclear acid [19].

Magnesium concentration is 103.9±0.76mg/100g. Magnesium is very important in calcium metabolism in bones and also involved in prevention of circulation diseases, it help in regulating blood pressure and release of insulin [38]. The recommended daily allowance of magnesium for adult is 350mg/day while dietary recommendation for children is 170mg/day [29]. *Thaumatococcus daniellii* leaf protein concentrates can contribute 30% to recommended daily allowance.

Potassium concentration is 90.3±042mg/100g, high amount of potassium in the body was reported to increase iron utilization [3] and beneficial to people taking diuretics to control hypertension and suffer from excessive excretion of potassium through body fluid [11]. The value obtained for Thaumatococcus daniellii is lower than 220.0±7.8mg/100g reported for *Cassia siamea* leaves [28]. The recommended daily allowance of potassium is 2000mg for adults [29]. *Thaumatococcus Daniellii* can contribute4.5% to dietary allowance.

Sodium concentration is 70.6±0.42mg/100g. Sodium is important sources of electrolytes within the body. The recommended daily allowance of sodium is 500mg for adult [29]. *Thaumatococcus danielli* leaf protein concentrates can contribute 14.12% of recommended daily allowance. This is an indication that this sample is suitable for hypertensive patient.

Calcium concentration in this sample is 197.0±0.28mg/100g. Calcium containing substances is required by children, pregnant and lactating women for bones and teeth development. The recommended daily allowance of calcium is 800mg per day for children and adult. The value obtained for this sample is lower than recommended daily allowance but higher than 68mg/100g and 124mg/100g reported for pigeon pea and lima bean leaves respectively [10]. Calcium plays other roles in the body apart from skeletal development (cell membrane integrity, regulation of ion transport, control of muscle action, transmission of nerve impulses, blood clothing and co-factor for several enzymes) and as a result, foods that are high in calcium are needed in the body. However, it must be noted that the choice of calcium richen foods must be done with care because approximately 85% of kidney stones are composed predominantly of calcium compounds. The most common cause of calcium stone formation is excess calcium in the urine (hypercalciuria). Excess calcium is normally removed from the blood by the kidneys and excreted in the urine. In hypercalciuria, excess calcium builds up in the kidneys and urine, where it combines with other waste products to form stone. Calcium stone can also be formed through low levels of citrate, high levels of oxalate and uric acids; and inadequate urinary volume [25].

Manganese concentration is 2.5± 0.88mg/100g. Manganese is required for regulation of blood sugar level and is involves in production of energy and cell reproduction. It also supports the immune system. The proportion of manganese require in body is small. Therefore, the reported value for this sample cannot cause any health problem that could arise from excess manganese in food. Copper concentration is 0.2±0.88mg/100g. Copper is require in the body for enzymes production and biological transfer of election. The concentration of copper in this sample fall below

the recommended dietary allowance of 3mg per day for adult and 2mg per day for children [29]. *Thaumatococcus daniellii* leaf protein concentrate can contribute 6.67% and 10% respectively. Lead and Selenlum were also present in this sample in concentration of 0.1±0.44mg/100g and 0.1±0.97 respectively too much Lead and Selenium in food are not good, they can lead to metal poisoning.

However, the proportion of these two metals in *Thaumatococcus daniellii* are not present in the concentration that could impair health, its consumption is safe.

The amino acid concentration of *Thaumatococcus daniellii* leaf protein concentrated were presented in Table 3. Glutamic acid has the highest value (10.23g) follow by aspartic acid (9.12) while Cysteine is present in the least quantity. Norleucine, Tryptophan and Selenocystine are the limiting amino acid. These limiting amino acid is expected because the sample under investigation is plant sample and these four amino acids are common in animal protein. The recommended daily allowance of Aspartic acid is 21.6g for males and 20.0g for females. These values were higher than those reported for *Thamatococcus daniellii* leaf in order to meet up with the recommended daily allowance significant quantity of this leaf concentrates would be consumed.

However, the values of other amino acid reported for this sample are generally favourable when compared with the recommended dietary allowance [17] (Table 6). From this table, the recommended value for lysine is 5.80 while 4.03g was observed, for this leaf protein concentrates. The recommended value for Threonine is 3.40g while 5.00g was reported. Summarily, *Thaumatococcus daniellii* leaf protein concentrates appears to be better source of other amino acid when compared with the reference table. The total essential amino acid value for *Thaummatococcus daniellii* is 42.44g/100g while the non-essential amino acid is 44.36g/100g (Table 4 and Table 5 respectively).

Table 3: Amino acid content of *Thaumatococcus daniellii* in g/100g

Amino Acid	Concentration
Lysine	4.03
Histidine	2.30
Ammonia	ND
Arginine	5.61
Aspartic acid	9.12
Threonine	5.00
Serine	3.41
Glutamic acid	10.23
Proline	3.66
Glycine	6.88
Alanine	4.39
Cystine	1.06
Valine	5.86
Methloline	2.03
Isoleucine	6.22
Leucine	7.87
Norleucine	ND
Tyrosine	3.65
Phenylalanine	5.48
Tryptophan	ND
Selenocysteine	ND

Note; ND means not detected.

Table 4: Essential amino acid present in *Thaummatococcus daniellii.*

Essential amino acid	g/100g
Lysine	4.03
Histidine	2.30
Threonine	5.00
Valine	5.86
Methioline	2.03
Isoleucine	6.22
Leucine	7.87
Tyrosine	3.65
Phenylalanine	5.48
Tryptophain	ND
TOTAL	42.44

Table 5: Non-essential amino acid present in *Thaummatococcus daniellii.*

Non-essential amino acid	g/100g
Alanine	4.39
Arginine	5.61
Aspartic acid	9.12
Cysteine	1.06
Glutamic acid	10.23
Glycine	6.88
Proline	3.66
Serine	3.41
Selenocysteine	ND
Norileucine	ND
Ammonia	ND
TOTAL	44.36

Table 6: FAO/WHO/UNU reference value of amino acid.

Amino acid	Reference value
Lysine	5.80
Methionine + Cysteine	2.50
Threonine	3.40
Tryptophan	1.00
Valine	3.50
Leucine	6.60
Isoleucine	2.80
Phenylalanine + Tyrosine	6.30
TOTAL	31.90

CONCLUSION

The leaf protein concentrates of *Thaummatococcus daniellii* revealed nutritional, mineral and amino acid content of considerable interest in addition to the local use for wrapping proceed foods, it can be proceed in to viable food ingredients or used as vegetable for man and animal. The functional properties of this leaf protein concentrates should also be explored.

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