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Research Article

Nutritional Quality of *Nyam ngub,* Traditionally Textured Product Derived from Wild Orchid Tubers Eaten as Meat in the North West and Western Region of Cameroon

Dobgima John Fonmboh[1], Thierry Ngangmou Noumo[1], Fokunang Charles Ntungwen[2, 4], Ejoh Richard Aba[1, 3, *]

[1]Department of Nutrition, Food and Bioresource Technology, College of Technology, The University of Bamenda, Bambili, Cameroon

[2]Department of Pharmaco-toxicology and Pharmacokinetics, Faculty of Medicine and Biomedical Sciences, University of Yaoundé 1, Yaoundé, Cameroon

[3]National School of Agro-Industrial Science, University of Ngaoundere, Ngaoundere, Cameroon

[4]Laboratory for Preclinical Animal and Toxicology Research, Faculty of Medicine and Biomedical Sciences, University of Yaoundé 1, Yaoundé, Cameroon

Abstract

The knowledge gaps in the nutritional composition and quality of traditionally textured plant-based products eaten as meat is affecting the global acceptance despite the acclaimed health, environmental, ethical, religious, and social benefits. This paper aimed to prepare and evaluate the nutritional quality of *Nyam ngub* for potential valorization and vulgarization. Standard methods were used to determine the chemical composition and to evaluate the quality of protein. Protein fractions were used to estimate the solubility and individual amino acids were analysed with rapid amino acid analyser. Nutrient bio accessibility was determined firstly by calculation through the phytate: mineral ratio for iron and while the simulated *in-vitro* gastrointestinal test evaluated the protein digestibility and mineral accessibility. Results indicated that *nyam ngub* had an ash content of 13.02±1.14g/g at a moisture content of 89.56±2.43% and dry matter of 12.86±0.30%. The reducing and total sugar content were 0.8±0.02 g/1000mL and 51.42±4.26 g/1000mL respectively yielding a moderate energy supply (67.26±0.72 Kcal/mol) compared to other tubers. The crude fibre, fat and protein were respectively 6.7±0.3 (g/100g), 3.07±0.42 (g/100g) and 6.03±0.15 (g/100g). The Calcium, iron, Zinc and Copper contents were 0.01±0.00 mg/100g, 1.60 g/100g, 0.25±0.04 mg/100g and 2.87±0.00 μg/g respectively while vitamin A after conversion from β- carotene was 1.65±0.77μgRE/g and vitamin C was 5.043±0.54 mg/100g. The albumin, globulin, prolamin, and glutelin fractions were 70.51±2.48, 65.93±1.44, 16.41±3.21 and 18.46±1.35 mgBSA/100g respectively. Iron and zinc were 57.32±0.58% and 51.73±0.23% accessible while protein had the greatest digestibility in the gastric phase (74.63%) compared to 70.15% in the intestines. The essential amino acids quantified in mg/ 100g were Arg (1.39), His (0.61), Leu (2.04), Lys (1.52) Met (0.59), Phe (1.40), and Thr (1.11). Despite the limited protein content and lack of some essential amino acids, the protein of *nyam ngub* was relatively soluble and available and the micronutrients are accessible.

*Corresponding author: ejohrab62@gmail.com (Ejoh Richard Aba)

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Keywords

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1. Introduction

The ethical and culturally oriented pursuit of healthy feeding as well as environmental sustainability is reshaping global feeding habits and culture. The EAT-Lancet Commission fixed a global target by 2050 attainable through fruits, vegetables, legumes, and nuts consumption while cutting down on the quantity of added sugars and red meat; all to maintain the serenity of the environment while watching health [1]. It is unanimously accepted that in all food production and consumption patterns, meat and products are enormously environmentally anthropogenic [2]. The production of animal-based proteins requires large amounts of water (10–1000 times compared to plant proteins), land (the world's one-third of all arable land) [3], generates high levels of greenhouse gases (e.g., methane, carbon dioxide, ammonia) [4]. In addition, high levels of meat consumption are linked to heart disease [5], stroke [6], type 2 diabetes [7], obesity [8], some cancers [5], metabolic diseases, and all-cause mortality [9]. To crown it, the UN blames the inappropriate use of antimicrobials during animal production for the increased emergence of antimicrobial resistance [10]. These negative traits are envisaged to lead to a devastating 2.4 million deaths globally and total healthcare costs of 285 billion dollars by 2025 [11]. In addition, animal welfare advocates argue that the intensification of livestock production is promoting serious animal welfare concerns [12]. To achieve the recommended levels of 75% less intake of red meat by an average citizen globally and up to 90% per capita reduction for those of the western hemisphere [11], many strategies have been employed. This involved encouraging 'meatless days' or eating smaller portion sizes [13]; replacing meat with vegetables, beans, pulses, and/or nuts [14] or including plant-based meat extenders in processed meat products [15]; and finally developing structured products [16]. Though traditionally structured products like tofu, tempeh, and seitan attracted an enduring interest in Western countries [17], the development of structured products fundamentally paved the way for a new category of plant-based meat products; the meat analogues [18, 16].

Consequently, plant-based meat substitutes have attracted widespread attention and are progressively becoming a research hot spot in the food industry [19]. The global market for meat alternatives is predicted to grow from 5.37 billion dollars in 2021 to 10.80 billion dollars by 2028 [20]. Plant-based products trade generally increased by 49% from 2018 to 2020 in Europe [21], and in Spain alone, the sales of plant-based meat analogues (PBMAs) increased by 32% from 2019 to 2020 [22]. Related trends are observed in the UK, Brazil, Norway, the US, Australia, and Germany [23, 24]. Some popular PBMs available include burgers, sausages, and chicken nuggets [25]. However, the principal sources of plant-based proteins used in meat analogues currently remain soy and the wheat protein gluten, although legumes/pulses (peas, lentils, lupine, chickpea, and others), and fungi (mycoprotein, yeasts, and mushrooms) are in play [26]. To produce the prevailing plant-based meat analogues, protein is extracted from plants in the form of flours (10–20% protein), protein isolate (>80% protein), protein concentrate (55–60% protein), or protein texturate (50–70% protein) [27]; making the process technologically and cost-intensive. That notwithstanding, the strong off-flavour or beany flavour of soy-derived products due to the activity of lipoxygenases, saponins, and isoflavones [28], the allergenic effect [29], the harmful effect of cereal (wheat, rye, and barley) protein to some individuals due to gluten intolerance [16] also constitute barriers for the smooth adoption of present plant-based meat products.

Commendable efforts are being deployed to understand plant protein structuring, texturing, formulation, and fibrilization with the works of Atze Jan van der Goot of the Food Processing and Engineering Department of Wageningen University at the vanguard but the limited understanding of the meat alternative domain coupled with the scarcity of researchers investing in the sphere [30] contribute to hamper smooth success; as such, acceptance is not universal [31].

Achieving the desired texture, juiciness, organoleptic, and sensory perceptions comparable to animal products remain a major technological task since the resulting appearance, texture, and flavour of plant-based meat products remain inadequate [32]. As such, consumers portray the low sensory appeal of meat replacers as the primary barrier to acceptance [33]. To realize such attributes, the functionality of the protein(s) and/or non-protein ingredients choices as well as the implication of advanced and novel technologies [34] are highly essential. And fibrous anisotropic structure formation has been earmarked as the key factor in forming an attractive structure similar to meat products [35].

It is therefore imperative to understand what meat is and why people eat meat to propose an acceptable and convenient "alternative", "substitute" or "replacement". The consumption of plant-based diets generally was predominantly practiced based on religious and cultural edicts but the prevailing renaissance of interest in plant-based diets is tailored by the global concerns surrounding the environment, animal welfare, and human health [36]. Informed consumers today, desire minimally processed food to benefit from the acclaimed new health-promoting products [37]. However, the rhetoric directing the paradigm of meat alternatives/substitutes and/ or replacements has limited the search to “another quality protein-rich food” obtained through “a high technological process” despite consumer’s orientation to natural food. It is known that meat represents the central foods around which meals are based [38] in some societies, and the highly valued status of meat is associated with wealth, status, masculinity [39], and hospitality [40]. The perception and conception of the value/ quality of meat therefore depends on the culture, food habits, meal context, and feeding style as meat to many is an accompanier during eating which provides resistance and satiety. Some African cultures consume mushrooms, garden eggs, and young tender elephant grass shoots, and Indians take bamboo shoots while the Chinese go for grilled sweet potato even as the resistant portion to accompany the central dish. Plant-based meat products presented in this context might derive more attention and pull research from the conventional sources targeted currently towards other sources.

Plunging into the undervalued indigenous knowledge [41] to vulgarize the survival strategies of the African communities that were weakened by foreign structures and economic policies [42], this paper seeks to present *nyam ngub* as a possible traditionally texturized plant-based product that can serve the purpose. *Nyam ngub* is an endogenously processed relish eaten and accepted as meat, meat alternative, meat snack and/ meat substitute in certain localities of the North West Region of Cameroon [43], just as the *napsie* of the Bangam people of Galim in the Western Region of Cameroon [44] and the chikanda/kinaki (African polony) of the southern African countries [45]; though in the later, peanut powder in added. However, like other wild foods with little or no nutrient facts [46], the nutrient content and nutritional value/quality are scarce in the literature. The objective of this paper was to prepare and evaluate the nutritional value/quality of *nyam ngub*.

2. Methodology

2.1. Plant Material Acquisition

The wild orchid tubers were collected from Abong-Phen in Kedjom Ketingoh and its environs in the Tubah subdivision of Mezam Division in the North West Region of Cameroon. For maturity and proper tuber quality, the tubers were collected between September and November of the year 2023.

2.2. Sample Preparation

Nyam ngub was prepared by washing the tubers thoroughly with running tap water and allowing them to drain. The tubers (100 g) were then crushed into 3 mm and 7 mm diameters using a meat mincer (Hendi Food Service equipment, Steenoven 213911 TX Rhenen, item/kod: 210864, voltage: 230V/50Hz, serial number: 18100263, 2018). Water was added and mixed properly in a mixer (Russell Hobbs model: 25930, 220-2240V, 50/60 Hz, 1000W, 29119X0) after which wood ash extract prepared from selected tree trunks was introduced and properly homogenized. The resulting marsh was allowed for 25 minutes to mature at room temperature and a laboratory meat filler (Hendi Food Service Equipment, Steenoven, 21, 3911 TX Rhenen, item/kod: 282137, serial number: 181031100, 2018) was used to fill the mix in an artificial sausage casing. An electronic warm water bath was then set at ebullition and the packaged product was cooked for 40 minutes. The water bath was put off and the sausage casing and content cooled to room temperature. The cooked product was then dried at 45 ^{0}C for 4 days in an oven (Huanghua Faithfull desktop constant temperature; Model: WHL-25AB, 600W, 220V, 50Hz, SN:20210101120010). The dried sample was grind and sieved to obtain fine powder which was preserved for subsequent analysis. Figure 1 indicates the procedure.

Figure 1. *Flow diagram for Nyam ngub production.*

2.3. Proximate Analysis

The protein content (total nitrogen x 6.25), moisture, ash, fat, and fibre were determined according to methods 955.04,925.09, 923.03, 920.39, and 991.41 respectively of AOAC, [47]. Reducing sugars and total sugars were determined by a modification of the Phenol-Sulfuric acid of Dubois *et al*. [48] where the reducing sugars were extracted by boiling 5 g of the sample in 50 mL of boiling water and the resulting mixture was finely ground in a mortar with a pestle, triturated, filtered with a Whatman Nº 1 filter paper, and the volume completed to the 50 mL mark with hot water. The total sugars were extracted by 0.5 g of the sample in 0.5 mL of 12N hydrochloric acid and allowed at room temperature for 1 hour and boiling thereafter for 2 hours in an electronic thermostat water thank for three usages (Model: HH-W21Cr42 II, 37-2000 ± 0.5 ^{0}C, 220V, 50 Hz, 825 W, 05/2010). Distilled water (5.5 mL), 10 mL of 70% ethanol (Fischer chemical 99.8+%, MDL: 3568, Product code: 12498740, MW:46.069), 0.5 mL of Zinc acetate dihydrate (2 g/100 mL; Kermel 99.0%, CAS N0: 0532-83889090; EC N0:022-28546966; MW:219.50) and 0.5 mL of potassium hexacyanoferrate III (10.6 g/ 100 mL; 99.5%; CAS: 13746-66-2; MW:329.25) were added, properly homogenized and allowed to interact for 10 minutes and the mixture filtered with Whatman filter paper N^{0} 1 and the volume completed to the 50 mL mark.

2.4. Mineral Content

The concentrations of minerals Ca, Cu, Fe, K, Mg, Mn, Na, and Zn were determined using flame atomic absorption spectroscopy described by Prapasri *et al.* [49].

2.5. Determination of Vitamins

The vitamin A content was deduced from conventional conversion factors as 12 µg of carotenoid corresponding to 1 µg of Provitamin A according to Wolff, [50] while the 2,6-dichloroindophenol titrimetric method described by Harris and Ray, [51] was used to determine the vitamin C content.

2.6. Evaluation of Antinutrient content

The total phenol, phytate, and oxalate contents were quantified following the methods described by Amorim *et al.* [52], Akalu and Geleta, [53], and Inuwa, [54] respectively.

2.7. Evaluation of Nutritional Quality/Value

The sequential extraction of the proteins was carried out according to the methods of AACC, [55] and the protein content in the supernatants was quantified by the method of Bradford, [56] while the amino acid profile was determined by the amino acid analyser method outlined by Prapasri *et al.* [49]. The quality of the protein was then evaluated through the determination of the amino acid score, essential amino acid score, proportion of total essential amino acids to total amino acid, predicted biological value, nutritional index according to FAO, [57]; FAO/WHO, [58]; Chavan, [59]; Mune *et al.* [60], and Ijarotimi and Keshinro [61] respectively.

2.8. Evaluation of Bioaccessibility/Digestibility

The molar ratios of phy: Fe and phy: Zn were calculated to estimate the bioavailability of iron and zinc, respectively, and to give an indication of the inhibitory effects of antinutrients particularly phytates on the minerals. Phytate: mineral is commonly used as a simple method to estimate the bioavailability of Ca, Fe, and Zn in the presence of phytate. Since inhibition due to phytate on both non-heme iron and zinc absorption is dose-dependent in humans, low phytate: mineral molar ratio corresponds to high theoretical bioavailability [62].

According to the European Food Safety Authority (EFSA), a phy: Zn molar ratio below 5 corresponds to high absorption efficiency, 5–15 is moderate, and above 15 is low bioavailability [63]. For iron, the phy: Fe molar ratio should be below 1, or preferably below 0.4, to significantly improve non-heme iron absorption in plant-based meals with no iron absorption stimulating factors suggested by Hurrell and Egli is recommended by FAO/INFOODS/IZINCG [64].

The in vitro digestion was also simulated according to Brodkorb *et al.* [65] while iron and zinc bio-accessibility were determined after in vitro digestion following Brodkorb *et al*. [65] and Minekus *et al.* [66]. However, pancreatin and bile salt were respectively added to realize a final enzyme activity of 100 U/mL protease and a concentration of 2 mM in the final mixture which reduced the iron and zinc concentration in the digesta to concentrations that have previously been indicated to be sufficient in the determination of iron and zinc bio-accessibility [67].

2.9. Statistical Analysis

The results obtained were analysed on Statgraphics, (Centurion XVI, 2011). The means of triplicate analysis were subjected to a one-way ANOVA at $p \leq 0.05$ and where there were significant differences, the means were separated by the Duncan Multiple Range test.

3. Results and Discussion

3.1. Proximate Analysis

Life sustenance relies intricately on proper nutrient supply since they are essential for physical growth, maintenance of normal body function, and good health [68]. Table 1 presents the proximate composition of dried *nyam ngub* powder.

Table 1. *Proximate composition of nyam ngub.*

Nutrient	*Nyam ngub*
Moisture content (%)	89.56±2.43
Dry matter (%)	12.86 ± 0.30
Ash (g/100g)	13.02 ± 1.14
Reducing sugars (g/1000mL)	0.80 ± 0.02
Total sugars (g/1000mL)	51.42 ± 4.26
Crude protein (g/100g)	6.03 ± 0.15
Crude fibre (g/100g)	6.7 ± 0.30
Crude fat (g/100g)	3.07 ± 0.42
Vitamin A (μg RE/g)	1.65 ± 0.77
Vitamin C (mg/g)	5.43 ± 0.54
Energy value (Kcal/100g)	67.26 ± 0.72
Total phenol (g/100g)	*NA
Phytate (g/100g)	*NA
Oxalate (g/100g)	*NA

Values are presented as mean of 3 repetitions±standard deviation

*Total phenols, phytate and oxalate were not found at all in the tubers. This could be due to the varieties used, period of harvest and most importantly place of tuber collection.

Food moisture content is an index to determine water activity and food stability [69]. High moisture foods are more susceptible to microbial contamination while foods with low moisture content can be stored for a long time and suitable for processing in food industries [70]. The moisture content of *nyam ngub* powder was 12.86 ± 0.30% which was within the recommended range of (12-15)% for shelf-stable products. The moisture content of *nyam ngub* was comparable to the 14.81±0.88% reported for *nyam ngub* powders [43]. The difference in tuber type, powder age after preparation, and area of tuber collection could account for such disparity. Latunde-Dada *et al.* [71] reported relatively lower moisture content values on wet basis for plant-based burger products (PBBPs) produced from pumpkin, beetroot, mycoprotein, red cabbage, jackfruit, potato, soy, and mushroom when raw, ranging from 50.5% to 77.9% respectively, and 47.8% and 75.2% respectively when cooked. Also, the moisture content of plant-based pork products (PBPPs) prepared from different proportions of low-moisture TVP, pigment solution (0.50% soy sauce solution), high-moisture TVP, wheat gluten (1.00%), curdlan gum (2.50%), sweet potato starch (10.00%), sunflower oil (10.00%), TG enzyme (1.00%), pork flavoring powder (0.20%), and water was lower than *nyam ngub* ranging from (56.12 - 63.31) g/100 g [72]. Bakhsh *et al.* [25] reported the moisture contents of plant-based patties prepared from textured vegetable protein (TVP), Shiitake mushroom, isolate soy protein (ISP), isolate wheat protein (IWP), tapioca starch, fats, salts, seasoning, methylcellulose, garlic powder, molasses, natural pigment, and ice to range from (59.23±0.35-64.35±0.25)%.

Ash refers to the inorganic residue that endures either ignition or complete oxidation of organic matter in a foodstuff. The ash content represents the mineral part of a meal and provides an idea of the mineral salts input [73]. The ash content of *nyam ngub* was 13.02 ± 1.14 g/100g dw, significantly different from 8.98 g/100g dw reported by Dobgima *et al.* [43]. The period of harvest, maturity of the tubers, and the inclusion of different varieties as well as the variation in soil composition of the collecting spots could account for the difference. The ash content of the PBPPs reported by Liu *et al.* [72] ranged from 0.84±0.02 – 1.96±0.05 g/100 g while those of the plant-based patties ranged from (1.71±0.03-3.30±0.05)% [25] and was significantly lower than *nyam ngub*.

Lipids are generally substances that are soluble in ether, chloroform, or other organic solvents but are sparingly soluble in water. The terms lipids, fats, and oils are often used interchangeably. The low-fat content of *nyam ngub* may make it safe for consumption by people in areas where obesity remains a severe risk to people's health and lives [74]. The fat content of *nyam ngub* was 3.07 ± 0.42 g/ 100g and was significantly higher than 2.41±0.31g/100g reported by Dobgima *et al.* [43]. The discovery and inclusion of other tuber species in the preparation and extraction method employed could account for the difference. The fat content of PBPPs (4.14±0.16 -6.54±0.24) g/100g was relatively higher than that of *nyam ngub* [85] and this was due to the 10% sunflower oil added in the formulation. This is verified by the fat contents of certain conventional meat analogue products already on the market like Beyond burger (15.93 g/100g), Impossible burger (12.39 g/100g), Tofurky ham roast with glaze (5.56 g/100g) and Quorn brand chicken nuggets (8.47 g/100g) [75] as well as plant-based patties (29.2± 4.3-31.8±2.1) g/100g [25] which are considerably higher than that of *nyam ngub*.

Protein contributes to the structural and functional activities of cells, as well as the regulation of metabolic activities in all living organisms [76]. A sufficient protein intake in the diet increases the calorific value of the food, which indicates a nutritionally balanced diet [69]. The total protein content of *nyam ngub* was 6.03 ± 0.15 mg/100g significantly higher than the 2.59±0.35 mg/ 100g reported by Dobgima *et al.* [43]. The additional varieties of the tubers involved and the sensitivity of the Kjehdahle method compared to the Dumas method used in 2020 could account for the difference. The total protein content of *nyam ngub*, though within the range of (6.04±0.28-18.22±0.93) mg/ 100g reported for PBPPs [72] was relatively lower. This is consistent with the protein content of some available meat analogue products like Beyond burger (17.70 g/100g), Impossible burger (16.81 g/100g), Tofurky ham roast with glaze (20.37 g/100g) and Quorn brand chicken nuggets (10.17 g/100g) and plant-based patties ranged from 2.02±0.05 – 21.01±0,56% [25]. The relatively higher protein content for the available plant-based products could come from the input of the soy or texturized proteins used in the formulation of the other products.

Carbohydrate is a key component of the proximate composition of food that gives energy to the body and is essential to the structure and operation of cellular mechanisms [77]. Generally, roots and tubers have been reported to be high in carbohydrates and suitably serve as energy staple foods [78]. The total and reducing sugar content of *nyam ngub* was 51.42 ± 4.26 g/1000mL was 0.8 ± 0.02 g/1000mL respectively. The total sugar content was significantly higher significantly higher than 13.08 ± 0.33 g/100mL while the reducing sugar was comparable to 1.05 ± 0.03g/ 100mL presented in previous work [43]. The different varieties of tubers used and the modification of the sugar extraction method can explain this trend.

Dietary fibre of food sustains the intestinal microflora healthy and lowers the risk of colon cancer and cardiovascular disease [79]. Increased fibre intake in the diet enhances the digestion and absorption process in the large intestinal, which helps to prevent constipation and manage blood sugar levels [77]. The crude fibre content of *nyam ngub* was 6.7 ± 0.3g/100g significantly greater than 4.88±0.17g/ 100g reported previously [43]. The fibre content of *nyam ngub* is greater than the 1.96 ± 0.35-5.02 ± 0.44 g/100g for PBPPs evaluated by Liu *et al*. [72]. The trend was the same with burger products like Beyond burger (1.77 g/100g), Impossible burger (2.65 g/100g), Tofurky ham roast with glaze (0.93 g/100g), and Quorn brand chicken nuggets (5.93 g/100g) [75] which were lover in fibre than *nyam ngub*.

3.2. Vitamin Analysis

Vitamins are defined as relatively low-molecular-weight compounds that humans, and for that matter, any living organism that depends on organic matter as a source of nutrients, require small quantities for normal metabolism. Vitamin analysis of food is critical to determine animal and human nutritional requirements. Besides that, accurate food composition information is essential to determine dietary intakes to assess diet adequacy and improve human nutrition worldwide. The vitamin A and C content of *nyam ngub* was 1.65 ± 0.77 μg/g and 5.43 ± 0.54 mg/100g respectively and were comparable to the respective values 1.27±0.12 μg/g and 5.93±0.77 mg/100g obtained by Dobgima *et al.* [43].

3.3. Mineral Analysis

Minerals have significant roles in the body to execute different metabolic functions from building strong bones to transmitting nerve impulses for a healthy and long life [80]. Minerals constitute the micronutrients and are necessary for physiological and biochemical processes by which the human body acquires, assimilates, and utilizes food to maintain health and activity [81]. Minerals are thus essential in human nutrition supporting the maintenance of acid-base balance, the response of nerves to physiological stimulation and blood clotting [70], and ensuring adequate immune competence and cognitive development [68]. Table 2 presents the mineral content of *nyam ngub*.

Table 2. *Mineral content of nyam ngub.*

Minerals	*Nyam ngub*
Ca (mg/100g)	0.10 ± 0.00
Mg (mg/100g)	0.15 ± 0.01
K (mg/100g)	6.34 ± 0.25
P (mg/100g)	0.17 ± 0.00
N (mg/100g)	0.96 ± 0.02
Na (mg/100g)	54.90 ± 0.97
Zn (mg/100g)	0.25 ± 0.04
Mn (mg/100g)	0.07 ± 0.00
Fe (mg/100g)	1.60 ± 0.04
Cu (ug/g)	2.87 ± 0.00

Values are presented as mean of 3 repetitions±standard deviation

Calcium is an indispensable mineral in human nutrition involved in blood clotting, muscle contraction, neurological functioning, bone and teeth maintenance, and enzyme metabolic processes [82]. The calcium of *nyam ngub* was significantly lower than those of burgers produced from pumpkin, beetroot, mycoprotein, Red cabbage, Jackfruit, Potato, Soy and Mushroom which amount respectively to 134±0.93 mg/100g, 156±1.38 mg/100g, 260±7.01 mg/100g, 93.4±1.39 mg/100g, 31.7±0.10 mg/100g, 57.9±0.92 mg/100g, 150±1.32 mg/100g, 46.2±0.28 mg/100g [71] while PBPPs had calcium values ranging from 15.24±1.99-48.57±3.58 mg/100g [72].

Iron is a basic part of the formation of blood haemoglobin and helps in the transportation of oxygen in the body. Iron deficiency is the most common mineral insufficiency worldwide and is estimated to affect two billion people globally [83] with more than 90% of the affected populations living in developing countries. In children, impaired cognitive development [84], increased susceptibility to infections, increased fragility, poor physical growth, decreased appetite, reduced mental performance, retardation, psychomotor development and congestive cardiac failure [85] are diagnosed. In adolescents, impaired physical and mental development, while for pregnant women it is associated with multiple adverse outcomes for both mother and infant, including increased risk of maternal mortality and low birth weight [86]. In adults, it reduces the capacity for physical work and mental work [87]. The iron content of *nyam ngub* was comparable to 1.60 ± 0.84 mg/100g reported by Dobgima *et al.* [43] and aligns with those reported for PBPPs of 1.22 ± 0.14-3.90 ± 0.35 mg/ 100g [72] while Latunde-Dada *et al*. [71] indicated that burgers from pumpkin, beetroot, mycoprotein, Red cabbage, Jackfruit, Potato, Soy and Mushroom had iron contents of 3.36 ± 0.03 mg/ 100g, 3.39 ± 0.06 mg/ 100g, 1.13 ± 0.04 mg/ 100g, 1.13 ± 0.04 mg/ 100g, 1.27± 0.03 mg/ 100g, 1.99 ± 0.08 mg/ 100g, 1.45 ± 0.03 mg/ 100g, 2.90 ± 0.02 mg/ 100g respectively.

Zinc plays a key role to develop the brain and bone and also helps the metabolism of carbohydrates, proteins, vitamin A and nucleic acid biosynthesis processes [88]. Zinc indispensable component of a host of enzymes that are crucial for optimal metabolism and body function [89]. Zinc deficiency is common in populations that consume diets with a low content of animal products and a high consumption of vegetables and cereal crops. Zinc deficiency in humans is estimated to be 31% (range is 4%–73%) with a high prevalence of (37%–62%) found in Southern and Central African regions [90]. Prasad *et al*. [89] also reported zinc as an anti-inflammatory and antioxidant agent and that it also functions in cell-mediated immune processes. Zinc deficiency causes stunted growth in children [91], as well as morbidity from diarrhoea, pneumonia and malaria [92]. In childhood, zinc ensures optimal physical growth as well as neuro-behavioral and brain development [93]. The zinc content of *nyam ngub* (0.25 ± 0.04 mg/100g) was relatively lower compared to plant-based burgers from pumpkin, beetroot, mycoprotein, Red cabbage, Jackfruit, Potato, Soy, and Mushroom with values of 1.10 ± 0.02 mg/100g, 1.67 ± 0.03 mg/100g, 7.15 ± 0.08 mg/100g, 1.14 ± 0.04 mg/100g, 0.89 ± 0.02 mg/100g, 0.91 ± 0.03 mg/100g, 1.14 ± 0.01 mg/100g and 2.24 ± 0.08 mg/100g [71] respectively.

Copper is used for the synthesis of haemoglobin, proper iron metabolism, and maintenance of blood vessels [94]. Copper plays a role in the production of haemoglobin, myelin, and melanin and it also keeps the thyroid gland functioning normally [95]. Copper can act as both an antioxidant and a pro-oxidant. As an antioxidant, Cu scavenges or neutralizes free radicals and may moderate or support the prevention of some of the damage they cause [96]. As a pro-oxidant, sometimes copper promotes free radical damage which contributes to the development of Alzheimer's disease [97]. The copper content of *nyam ngub* was 2.87 ± 0.00 µg/g.

3.4. *Nyam ngub* Food Value/Quality

Protein fraction

Solubility is a complex biophysical property of globular proteins often crucial for proper functioning [98]. Solubility, as a thermodynamic process, represents the most important functional characteristic of proteins and denotes the protein concentration in a saturated solution at equilibrium with a solid phase (crystalline or amorphous) under a given pH, temperature, and in the presence of a solvent, salts, and other additives [99]. Consequently, the upshots of a suitable solvent (often water) is a soluble and high-protein food in addition to enhancing the conditions to express the functional properties (emulsifying, stabilizing, foaming, and gelation), which is difficult to attain with insoluble proteins [100]). Table 3 presents the protein fractions of *nyam ngub* powder.

Table 3. *Protein fractions of nyam ngub (mgBSAE/g).*

Fractions	*Nyam ngub*
Albumin (Water)	70.51 ± 2.48
Globulin (0.1 M Na_2HPO_4)	65.93 ± 1.44
Prolamin (2-Propanol)	16.41 ± 3.21
Glutelin (NaOH)	18.46 ± 1.35

Values are presented as mean of 3 repetitions±standard deviation.

Amino acid profile

Different amino acids are involved in the formation of proteins and making the nutritional quality of a protein-dependent on the content, proportion, and availability of the constitutive amino acids and represents a measure of the efficiency with which the body can utilize the protein [101]. Table 4 presents the amino acid found in *nyam ngub*.

Table 4. *Amino acid profile of nyam ngub (mg/100g).*

Amino acid	FAO/WHO, 1991	*Nyam ngub*
	Essential	
Arg		1.39
His	1.9	0.61
Ile	2.8	
Leu	6.6	2.04
Lys	5.8	1.52
Met		0.59
Phe		1.40
Thr		1.11
	Non -essential	
Ala		1.62
Asp		6.20
Glu		5.95
Gly		1.34

Amino acid	FAO/WHO, 1991	*Nyam ngub*
Pro		1.24
Ser		1.29

The protein quality of *nyam ngub* was established based on the amino acid profile (Table 4). *Nyam ngub* possesses seven essential amino acids and six nonessential amino acids with Ile completely absent and Met and His limiting with the least values below a unit. Table 5 presents the different quality predictors used to evaluate the protein quality and hence the food value of *nyam ngub*. The content of SAAs (Met + Cys) was 0.59 g/100g relatively lower than the required reference pattern (22–28 mg/g protein) set by WHO, [102] for different age groups. The AAAs (Phe + Tyr) was 1.40 mg/100g lower than the 38–46 mg/g protein) of amino acids requirement suggested by WHO, [102] for different age groups except for the ideal infant (52 mg/g protein) requirement. The Leu/Ile ratio was 2.04. Excess Leu content in foods interferes with the utilization of Ile and Lys [103]. Since high leucine in the diet impairs tryptophan and niacin metabolism creating an amino acid imbalance that leads to pellagra, a high Leu/Ile ratio is indicative of a food's contribution towards pellagra [104].

Generally, BCAAs play a role in normal growth and/ or function at the cellular and organ levels [107] intervening significantly during protein synthesis, glucose homeostasis, and anti-obesity reactions besides nutrient-sensitive signaling pathways [106] through the accumulation and that of related metabolites may create negative effects [107].

According to Ijarotimi and Keshinro, [60], EAAI can be used as a rapid tool to evaluate the protein quality of food formulations where an EAAI value > 95 indicates a superior protein source, 86 < EAAI ≤ 95 indicates a good protein source, 76 < EAAI ≤ 86 indicates a usable protein source, and EAAI ≤ 75 indicates an unsuitable protein source [72].

According to Kowalczewski *et al.* [108], PER can be used to assess the nutritional value of a protein in which a value greater than two indicates the high quality of the protein. The BV can indicate the degree of utilization of the protein in food after digestion and absorption. Although the protein quality by score appears to be of inferior quality, the biological value of 129.80 was close to animal products.

Table 5*. Calculation of protein value.*

Parameter	Value
TAA	8.66
TEAA	26.28
TNEAA	17.62
BCAA	2.04
SAA	0.59
AAA	1.40
TEAA/TAA	0.33
EAAS	27.18
EAAI	129.80
PPE	0.46
PBV	129.78

Assessment of Biodigestibility and Bioaccessibility

The amount of nutrients released from the food matrix and accessible for absorption represents the bio-accessibility of the nutrient [109] while the ability of the body to digest and absorb minerals from the dietary food is termed bioavailability [110]. Table 6 indicates the value for bio-digestibility and bio-accessibility of nutrients from *nyam ngub*.

Estimated mineral bio-accessibility

Generally, total phenol, oxalate, and phytate were not found in *nyam ngub* at the qualitative and quantitative evaluations. This indicates that there was no absorptive hindrance from the known inhibitors of accessibility and availability of nutrients. The total phenol content declared for the wild edible tubers of the North West region was 333.01±3.32 mg/ 100g [43]. The change in the tuber collection area could be advanced to explain the absence of the total phenol in the 2023 tubers. The ability of phytate to form insoluble complexes with calcium decreases the absorption, and then the phytate: calcium molar ratio is used as an indicator for bioavailability [111]. The critical molar ratio of phytate: Ca of < 0.24 is the threshold for good calcium bioavailability [70].

The phytate: Fe molar ratio of nyam ngub (0:1.6) was lower than the critical limits of 1 [112] or preferably 0.4 suggested as limits for adequate bioavailability of iron without stimulating factors [113]; which implies the bioavailability of Fe was high and the absorption was not inhibited by phytate.

Similarly, the phytate: Zn molar ratio is considered an indicator of zinc bioavailability [70]. According to EFSA, a phy: Zn molar ratio below 5 corresponds to high absorption efficiency, 5–15 is moderate, and >15 is low bioavailability [114]. This implies the phytate: Zn ratio (0:0.25) if nyam ngub indicated high bioavailability and was not likely to be affected by phytate [70].

Protein digestibility

The extent of protein digestibility was used to evaluate the bioavailability of dietary proteins. The digestion of *nyam ngub* proteins was relatively higher in the gastric phase (74.63%) than in the intestinal phase (70.15%) while the oral phase was minimum (7.76%) (Table 6). The protein digestibility of plant-based pork and beef at the gastric phase was

39.8% and 41% respectively while at the gastrointestinal phase it was 51% and 61% respectively [115]. However, Wehrmaker *et al.* [116] reported higher protein digestibility for meat analogue produced from wheat gluten and faba bean concentrate (78%) when raw and 72.8, 96% when cooked while that based on wheat gluten and pea protein isolate was 77.9% when raw and 80.1% cooked and meat analogue based on wheat gluten and soy protein isolate was 81.9% when raw and 83.2%).

Table 6. *Assessment of protein digestibility and Fe/Zn accessibility.*

Minerals		**Protein**		
Accessibility (%)		**Digestibility (%)**		
Iron	**Zn**	**Oral**	**Gastric**	**Intestinal**
57.32± 0.58	51.73±0.23	7.76	74.63	70.15

In Vitro Simulated Gastrointestinal Digestion of Samples

Mineral accessibility

The iron and zinc accessibility for burgers produced from pumpkin, beetroot, red cabbage, jackfruits, potato, soy, and mushroom in percentages were respectively 20.3, 8; 17.8,1; 19,7;6.9,6.7; 17.6,11; 31.5, 4; 26,14 [83] while the iron and zinc of *nyam ngub* were 57.32% and 51.73% accessible (table 6).

4. Conclusion

The work aimed to evaluate the nutritional quality of *nyam ngub* (an endogenously processed plant-based relish from wild edible orchid tubers eaten as a meat snack, alternative, or substitute in certain localities of the North West and Western Regions of Cameroon). The results indicated that *nyam ngub* has quality and/ or valuable nutrients as well as an appreciable hardness and colour comparatively to the existing plant-based meat products.

Although *nyam ngub* was low in protein (6.03 mg/ 100g), seven essential amino acids constitute the protein with Ile(0 mg), Met(0.59 mg) and His(0.61) representing the limiting amino acids having the smallest values, lower than unit. The proteins of *nyam ngub* were relatively soluble with the albumin and globulin fractions above 50% while highly digestible at the gastric phase (74.63%) yielding a biological value, comparable to that of animal products (129.80). Furthermore, the minerals were readily available with Iron having 57.32±0.58% and Zinc 51.73±0.23%.

Nyam ngub could therefore a potential traditionally texturized product which if well processed and introduced to consumers, based on its qualities especially nutritional and the potential to be chewy and tender, can be a suitable plan-based meat alternative since it is already accepted by some indigenous populations.

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Abbreviations

PBMA	Plant-Based Meat Analogue
AOAC	American Organization of Analytical Chemists
EFSA	European Food Safety Authority
FAO	Food and Agricultural Organization
WHO	World Health Organization
BCAA	Branched Chain Amino Acids
EAAI	Essential Amino Acid Index

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Data described in the manuscript, code book, and analytic code will be made available upon request pending approval.

Conflicts of Interest

The authors declare no conflicts of interest.

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