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Evaluation of Nutritive Value, Functional Properties and Amino Acid Profile of *Cnidoscolus Aconitifolius* Leaf Protein Concentrates

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Abstract

Green vegetables have been recognized as a reliable source of nutrients due to their ability to provide adequate amounts of vitamins, proteins and minerals. They also served as traditional medicines for alleviating infectious and non-infectious diseases. *Cnidoscolus aconitifolius* is a vegetable with such usefulness. Global food shortages worsened by deterioration in preservation methods have led to search for alternative and reliable source of protein using feasible, cost-effective food ingredients called Leaf Protein Concentrates from plant to replace animal protein.

A fresh green sample of *Cnidoscolus aconitifolius* was harvested at the biological garden at Emmanuel Alayande University of Education. The leaves of the plant samples after authentication at the Forestry Research Institute of Nigeria were trimmed, washed with distilled water and then processed into vegetable leaf protein concentrates to analyze their nutritional properties, amino acid characteristics, mineral composition and functional properties using standard method of analysis.

The result of its nutritional composition in g/100g showed moisture; 20.89±0.02, crude fat; 9.42±0.10, fibre; 2.33± 0.05, protein; 41; 49±0.01, ash; 4.98±0.01, Nitrogen Free Extractive; 41.78±0.37. The results of the mineral content of the sample in mg/100g showed; Mg; 98.30 ±0.61, K; 91.50± 0.47, Ca; 72.60±0.73, Fe; 6.50± 0.34, Zn; 2.9±0.63, Cu; 0.2±1.61, Mn; 2.50±0.26. Pb and Se were not detected in the sample. The sample contains valuable functional properties suitable for flour production additives and some other food processing ingredients, it also contains a total of 34.6g/100g of non-essential amino acid and 36.33g/100g of essential amino acid which are dietary ingredients required for healthy living.

Keywords: Leaf protein concentrates, Functional properties, Proximate analysis, Green vegetable

Introduction

The need to change to more sustainable, healthy and medicinal sourced food ingredients has increased nowadays, especially on plant-based food products. The need arises due to an increase in the human population, a global decrease in animal-based protein foods, malnutrition, widespread deficiency diseases and persistent inflation drive in most countries all over the world. The general drive to look for alternative sources of proteins has however become the major interest of researchers and industry due to the nutritional value that can be derived from them, their techno-functional properties and their significance in the human diet [1]. Green vegetables have been considered as one of the food ingredient sources of higher sustainability. They are considered useful because of their phytochemical component which gives them high medicinal potential and due to the cheaper cost required to make them available as food.

One of the green vegetables that contains a valuable amount of protein for men in Nigeria as well as a rich source of dietary and medicinal ingredients is *Cnidoscolus aconitifolius*. The plant is an evergreen shrub, of about 6m in height with characteristics of stinging hairs, a pale trunk, alternative and simple leaves. The plants have been reported in the literature for certain medicinal activities; it has been reported to be useful for the treatment of some illnesses like alcoholism, diabetes, insomnia, scorpion stings, skin disorders and venereal diseases. It has also been useful in strengthening fingernails, darkening grey hair, improving brain function and memory [2].

Cnidoscolus aconitifolius is a green vegetable that has received the attention of numerous researchers in the literature, especially its medicinal activities phytochemical composition, anti-inflammatory activities, ethnopharmacological profile ethno botanic, bioactivity and therapeutic uses [3, 4]. However, leaf protein concentrates from green vegetables have been considered a useful and reliable source of amino acids dietary fibre, and nutritional and dietary important minerals due to a global decline in food for man and animals. It is therefore the aim of this study to evaluate the functional properties, nutritional constituents and amino acid profile of *Cnidoscolus aconitifolius* leaf protein concentrates.

Materials and Method

Sample Collection

Fresh leaves of *Cnidoscolus acinitopholis* were purchased directly from a farm near Erelu Dam adjacent to Emmanuel Alayande University of Education and identified by a botanist in the Department of Biology, Emmanuel Alayande University of Education, Oyo. Oyo State, Nigeria.

Preparation of Leaf Protein Concentrates

The leaves of the sample were washed with distilled water and ground by passing it through a locally produced grinder. The pulp was collected and strained through a cotton cloth to filter off the crushed fibre. The green juice obtained by straining the pulp through a cotton cloth was heated to 90℃ by steam injection, which resulted in the coagulation of all the protein present in the pulp. The clot was then centrifuged from the remaining solution, pressed, ground, air-dried and stored in a refrigerator at 4℃ for further chemical analysis.

Proximate analysis

The proximate content of leaf concentrates of the selected plant samples was determined by various methods described by the Association of Official Analytical Chemists [5] to estimate the moisture content, crude fat, crude fiber, crude protein and ash, while the carbohydrate present in the samples was determined by subtracting the summed-up from the percentage composi-

tion of the selected sample.

Mineral Analysis

Five grams (5 g) of *Cnidoscolus aconitifolius* leaf protein concentrates were burned in a muffle furnace at 550°C for 12 hours and the resulting ash was cooled in a desiccator. The ash was dissolved in 2 mL of concentrated HCl and a few drops of concentrated HNO_3 were added. The resulting solution was evaporated to near dryness in a water bath. The contents were diluted to the mark with distilled water in a 100 ml volumetric flask. Buck Scientific Model: 210 VGP and Flame Photometer FP 902 were used to determine the concentration of various metals present in the sample after making appropriate dilutions for each element [5].

Water Absorption Capacity

The method described by Branch and Maria [6] was used to determine the Water Absorption Capacity of the leaf protein concentrates of the selected sample. 1 g of the leaf protein concentrate of *Cnidoscolus aconitifolius* was combined and mixed thoroughly with 10 mL of distilled water in a vortex centrifuge tube of known weight, for 30 minutes. The material was centrifuged for 15 minutes at 3000 rpm (25°C). The supernatant was decanted after centrifugation, while the precipitate and centrifuge tubes were reweighed. The water absorption capacity (WAC) of the leaf protein concentrates of the selected sample is calculated by weighing the amount of water absorbed per gram of the sample.

$$\frac{M_3 - M_2}{M_1}$$

Where:

W_3: the mass of the tube plus sediment,

W_2: the mass of the tube plus the dry sample, and

W_1: the mass of the dry sample

Oil Absorption Capacity

The method reported by Branch and Maria [6] was adopted with slight modifications to determine the oil absorption capacity (OAC). 1g of the leaf protein concentrate of *Cnidoscolus aconitifolius* was measured into a pre-weighed centrifuge tube, 10 mL of oil was added and vortexed for 30 minutes. The mixture was centrifuged for 15 minutes at 3000 rpm, and after resting for 30 minutes at 25°C, the centrifuge tubes and the precipitate were reweighed after the supernatant was decanted following centrifugation. The oil absorption capacity (OAC) was determined using the amount of oil absorbed per gram of sample.

Where:

$$\mathrm{OAC} = \frac{M_3 - M_2}{M_1}$$

M_3: the mass of the tube plus sediment,

M_2: the mass of the tube plus the dry sample, and

M_1: the mass of a dry sample

Determination of Foaming Capacity and Foaming Stability

Babatuyi et al. (2022) were followed to determine the foaming capacity of *Cnidoscolus aconitifolius* leaf concentrates. Five grams (5 g) of *Cnidoscolus aconitifolius* leaf concentrates were dispersed in 100 mL of distilled water, then the resulting solution was homogenized quickly for 5 minutes. The volume of foam released was recorded and the result was expressed using the formula below. Foam volume during homogenization was recorded one hour after foaming to determine foam stability as a percentage of initial foam volume.

$$\%\text{Foaming capacity} = \frac{\text{vol. after homogenisation} - \text{vol. before homogenisation}}{\text{vol. before homogenisation}} \times 100$$

Emulsion capacity and stability

2g of the leaf protein concentrate of *Cnidoscolus aconitifolius* and distilled water (100 ml) was blended for 30 minutes in a high--speed Panasonic blender (model VFG 24L,) at high speed (100 rpm). After the complete dispersion of the sample, a burette was used to add oil in streams of about 5ml. Blending continued until there was separation of the mixtures into two layers. Emulsification determinations were carried out at 30℃ and calculated as grams of emulsified oil by 1g of the leaf concentrates. The sample of leaf protein concentrate (4g) was dispensed in 100 ml of distilled water. 100 ml of oil was added at the rate of 12.5 revolutions per second while blending. Each sample was blended using a Panasonic blender at high speed for an additional 60 seconds before it was transferred into a 250ml graduated cylinder. Changes in the volume of the foam, oil, and aqueous layers were recorded after three hours [7, 8, 9].

Amino acid Analysis

The Amino acid leaf protein concentrate of *Cnidoscolus aconitifolius* was determined using methods described by Benitez (1989). All the known samples were dried to constant weight, defatted, hydrolyzed, evaporated in a rotary evaporator and loaded into the Applied Biosystems PTH Amino Acid Analyzer.

Defatting Selected Plant Samples

The sample leaf protein concentrates of *Cnidoscolus aconitifolius* is diluted with a 2:1 mixture of chloroform/methanol. About 500 mg of each sample was placed in an extraction hood and extracted for 15h using a Soxhlet extractor (AOAC, 2016).

Nitrogen determination

A small amount (15 mg) of the selected soil sample was weighed, packed in Whatman filter paper (No.1) and placed in a Kjeldahl digestion apparatus. Concentrated tetraoxo sulphate (vi) acid (10 ml) was added. To enhance quick digestion, a catalyst mixture (0.5g) containing sodium sulphate (Na_2SO_4), Copper sulphate ($CuSO_4$) and Selenium oxide (SeO_2) was added to the flask in a ratio of 10:5:1. Six pieces of anti-bumping granules were added. The flask was then placed in a Kjeldahl crusher for 3 hours until the liquid turned pale green. A digested sample of *Cnidoscolus aconitifolius* leaf protein concentrate was cooled and diluted with distilled water to 100 ml in a standard volumetric flask. A portion (10 ml) of the diluted solution with 10 mL of 45% sodium hydroxide was placed in a Markham flask and distilled into 10 ml of 2% boric acid containing 4 drops of bromocresol green/methyl red indicator until approximately 70 ml was dissolved and the distillate was collected. The distillate was then titrated with standardized 0.01 M hydrochloric acid to the grey endpoint. Percentage Nitrogen: $\frac{(a-b) \times 0.01 \times 14 \times V \times 100}{W \times C}$

Where: a: Titre value of the digested sample,

b: Titre value of the blank sample

V: Volume after dilution (100ml),

W: Weight of dried sample (mg)

C.: Aliquot of the sample used (10ml),

14: Nitrogen constant in mg.

Hydrolysis of the Selected Plant Sample

5g each of the defatted leaf protein concentrate of *Cnidoscolus aconitifolius* was weighed into a glass ampoule. 7ml of 6M HCl was added and oxygen was expelled by passing nitrogen into the ampoule (this is to avoid possible oxidation of some amino acids during hydrolysis e.g. methionine and cystine). The glass ampoule was then sealed with a Bunsen burner flame and put in an oven preset at 105℃± 5℃ for 22 hrs. The ampoule was allowed to cool before breaking open at the tip and the content was filtered to remove the humins. (It should be noted that tryptophan is destroyed by 6M HCl during the hydrolysis process).

The filtrate was then evaporated to dryness using a rotary evaporator. The residue was dissolved with 5ml of acetate buffer to maintain the pH in an acidic medium (pH 2.0) when stored in plastic specimen bottles, after which they were kept in the freezer.

Loading of the hydrolysate into the analyzer

60 microlitres (60µl) of the leaf protein concentrate of *Cnidoscolus aconitifolius* was loaded into the cartridge of the analyzer. The analyzer was designed to separate and analyze the free acidic, neutral and basic amino acids present in the hydrolysate.

Method of Calculating Amino Acid Values

An integrator attached to the analyzer calculates the proportion of peak area to the concentration of each amino acid.

Table 1: Proximate Constituents of Cnidoscolus aconitifolius Leaf Protein Concentrates

Parameter	Value (g/100g)
Moisture	20.89±0.02
Fat	9.42±0.10
Fibre	2.33±0.05
Protein	21.09±0.01
Ash	4.98±0.01
NFE	41.78±0.37
Energy	417.86

The results of the proximate composition of the sample (leaf concentrates of *Cnidoscolus aconitifolius*) are presented in Table 1 above. The sample had 20.89±0.02g/100g of moisture content. The moisture present in this sample is slightly above 18.63±2.11g/100g reported for the leaf of *Costus afer* (Ginger lily) by Anyasor [10]. The value of moisture present in this sample implied that a proper storage method will be adopted to hinder microbial growth that could enhance easily spoilage of the sample because the reasonable moisture value suggested for most vegetables is 6% to 15% [11].

The moisture in food however determines the shelf life, handling and sustenance of food. The rate of swallowing absorption and digestion of food is also enhanced by the percentage of moisture content. A low amount of moisture in food facilitates storage facilities by preventing biochemical reactions [12]. The moisture content of this sample is slightly above the suggested mois-

ture content of vegetables in the literature, indicating leaf proper handling of the sample to prevent microbial growth.

The crude fat of the sample is 9.42±0.10g/100g the value is higher than 3.17±0.47g/100g reported for *Maerva grassifolia* leaves by Suleiman [13]. The crude fat value of the sample is however low compared to 29.68±0.01g/100g reported by Akpabo and Ikpe 2013 for *Aneilema acauinoctiable* leaves. It has been reported that green plants and vegetables are low lipid-containing samples, which is of dietary importance to people who may wish to cut down on high dietary fat [14]. indicating that the Leaf protein concentrates obtained from *Cnidoscolus aconitifolus* could provide a lower amount of lipid in the human diet which is beneficial to categories of people that want to cut down excessive intake of fat.

The crude fibre content of the leaf protein concentrates of *Cnidoscolus aconitifolus* is 2.33±0.05g/100g. The fibre content of this leaf protein concentrate sample is in the range of 2.55±0.70g/100g and 2.81±0.26g/100g reported for the leaf and root of *Maesobotrya barteri* by Etukudo and Osim 2018. The value is however lower than 33.0g/100g reported for *Cinamon* by Shunada and Mahpara 2009. Crude fibre is a powerful food ingredient that assists in the prevention of heart disease, colon cancer and diabetes. It also helps to keep the digestive system healthy and functioning properly [15, 16]. The value of crude fibre in this sample will require the utilization of a large amount of this sample to meet up with the required quantity of fibre that would perform an adequate role of dietary fibre in food.

The value of crude protein obtained for the leaf protein concentrate is 21.09±0.01g/100g. The amount of crude protein present in this sample is higher than 3.35±0.22g/100g reported for *Brysocarpus coccineus* by Suleiman *et al.,*2019 and 10.38±0.07g/100g reported for *Peperomia pellucida* by Ooi *et al.,*2012. Protein assists the human body in the formation of antibodies; it is the major source of energy for cell function and serves as an essential ingredient for the proper maintenance of the structure of blood vessels [16]. The protein content of this sample can contribute a significant amount to the daily requirement of protein in the human body. It will also be a useful ingredient to boost the protein level (formulation) of food for humans and animals.

The value of ash found in the selected sample is 4.98±0.01g/100g, the value is higher than 1.12±0.47g/100g and 0.72±0.34g/100g reported for the leaves and stem of *Alchornla codifolia* by Ngaha *et al.,*2016. The value is however low when compared with the 11.6% reported for Neem by Asuk 2015. Ash is obtained when the remnant of organic matter is converted to organic matter, it determines the value of the mineral element it would offer organisms in the diet Ayanda *et al.,* 2018. The value of ash present in this sample indicates that the sample will able to offer a moderate amount of macro minerals in the diet

Nitrogen free extractive value of the sample is 41.78±0.37g/100g. the amount of carbohydrate present in this sample is low compared with 53.1±0.46g/100g reported for *Cridax procumbens* by Runde *et al.,* 2020, it is also low compared with 54.36±0.22g/100g and 53.46g/100g reported for *Dracalena mami and Dracaena arborea* leaf respectively by Ilodibia *et al.,*2014. Carbohydrate is a good source of energy required for body and cell metabolism (Agbafor *et al.,* 2015). A high value of carbohydrates in this sample implies that they will be good sources of dietary energy. Care however has to be taken by individuals who may want to cut down on carbohydrate intake including obese and diabetic patients. Since carbohydrates produce glucose as end products.

The value of dietary energy present in the sample is 417.84kcal/100g, The amount of energy that this sample would offer is greater than 348.64kcal/100g reported for *Maerua crassifolia* by Aletan and Kwazo 2019.

Table 2: Mineral Content of *Cnidoscolus aconitifolius* Leaf Protein Concentrates

Minerals	Value (mg/100g)
Mg	98.30±0.61
K	91.50±0.47

Na	76.40±0.21
Ca	72.60±0.73
Fe	6.50±0.34
Zn	2.9±0.63
Cu	0.2±0.16
Mn	2.5±0.26

Results of the mineral composition of the leaf protein concentrates of *Cnidoscolus aconitifolius* are reported in Table 2. The concentration of magnesium found in the selected sample is 98.30±0.61mg/100g. The value is higher than 33.5±0.54mg/100g, 22.20±0.37mg/100g and 30.11±0.19mg/100g reported for the leaf, stem and root of *Ipomea ivolucrata* by Oku *et al.,* 2017. Magnesium is one of the macro minerals required for the prevention of circulatory and heart-related diseases. It also assists in the utilization of calcium and its metabolism in men [17]. The value of magnesium concentration in this sample may be one of the reasons accounting for the use of *Cnidoscolus aconitifolius* in the treatment of high blood pressure.

The concentration of potassium determined for the leaf protein concentrates of the selected sample is 91.50±0.47mg/100g. The value of potassium in this sample is higher than 37.13mg/100g reported for *Cochorus oliforius* by Oloye *et al.,*2014 but low compared with 438.00±1.29mg/100g, 114.00±1.17mg/100g and 96.00±0.16mg/100g reported for the leaf, stem and root of *Milletia aboensis* by Oku *et al.,*2017. The recommended daily intake of potassium is 3500mg per day [18]. This implies that *Cnidoscolus acnitifolius* would contribute 2.61% of the recommended daily requirement. Potassium assists in a large number of biological processes in the human body, it assists in the regulation of osmotic pressure, the acid-base balance of the body fluid, the conduction of nerve impulses around the body and muscular movement coordination [19]. The deficiency of potassium always results in mental confusion muscular weakness and abnormal renal function. The level of potassium in this sample could prevent potassium-deficient diseases.

The value of sodium present in the leaf protein concentrates of the selected sample is 76.40±0.23mg/100g. The presence of sodium in adequate amounts in food assists in acid-base balance regulation, transport of metabolites and maintenance of blood pressure [20]. The amount of sodium present in this sample is adequate to furnish man with the dietary roles listed above the amount of sodium present in this sample is higher than 29.48±0.25mg/100g reported for *Urena lobata* stem by Njoku *et al.,*2009. The recommended daily allowance of sodium is 500mg per day for an adult. This Indicates that *Cnidosclous aconitifolius* leaf protein concentrates can contribute 15.28% of the recommended dietary allowance to the human diet

The concentration of iron present in the selected sample is 6.50±0.34mg/100g. The iron content present in the sample is high compared with 0.39±0.12mg/100g, 0.27±0.61mg/100g and 0.19±0.45mg/100g reported for the leaf, root and stem of *Maesoborya barteri* respectively by Etukudo and Osim 2018. Iron assists in the formation of haemoglobin, tendon and ligaments in the body. Deficiency of iron results in anaemia tiredness and weakness, the value of iron in this sample means that a small quantity of *Cnidoscolus aconitifolius* leaf protein concentrates will be required to provide 8.5% of the recommended daily allowance.

The concentration of zinc present in the selected sample is 2.9±0.63mg/100g. The value of zinc in this sample is lower than 12.59±0.25mg/100g reported for *Peperomia pellucida* by Ooi *et al.,*2012. The value is however in the range of 2.6mg reported for *Cinammon* by Shumaila and Mahpara (2009). Zinc assists in tissue growth and bone formation. enhances brain function, growth of the foetus, child development. and quick healing of the wound (Ogundola *et al.,*2018). The value of copper concentration determined for *Cnidoscolus aconitifolius* is 0.2±0.16mg/100g, the value of copper in this sample appears to be the least among all the minerals analyzed in the sample. The value is higher than 0.17±0.05mg/100g reported for *Emilia sonchifolia* by

Morshad *et al.,*2021. It is however lower than 0.87±0.08mg/100g reported for *Crocus sativa* by Kizil *et al.,*2010. Copper is required for the absorption of iron, the movement of iron from tissues to plasma. Copper is also required for growth, bone formation and formation of myelin sheath in the nervous system.

The concentration of manganese is 2.5±0.26mg/100g in leaf protein concentrates of *Cnidocolus aconitifolius*. The value is high when compared with 0.23mg/100g and 0.20mg/100g reported for ginger and Guajara leaves respectively by Paul *et al.,*2018. Manganese acts as a constituent of enzymes, it is one of the micro minerals which excessive intake would lead to gradual mental loss, coma, confusion convulsion and even death (Elmostafa *et al.,*2014). 2-5mg of manganese is recommended for adult males and females. Therefore, excessive intake of this sample should be avoided due to the high amount of manganese it contains. Excessive accumulation of manganese in the body is not good for healthy living. However, selenium and lead minerals were not detected in the sample.

The results of the functional properties of the leaf protein concentrates of *Cnidoscolus aconitifolius* are presented in Table 3. The water absorption capacity of the sample is 588.72±0.6g/g. The value is high compared with 140±12.25g/g, 192.0±10.95 and 196.0±10.65g/g reported for white flour, rice flour and green gram flour respectively by Chandra and Smasher 2024. The value is also high compared with 71.67±0.58 and 78.00±0.00 reported for *Musa acuminate and Musa balblsiana* respectively by Mosuro *et al.,*2023.

The water absorption capacity of a food sample determines the amount of water it can absorb to swell, improving yield and consistency during food processing. It is also a critical function of protein in food products. The observed high water absorption capacity may be due to a high value of carbohydrates present in the sample. Good water absorption capacity indicates that the sample would be useful as an ingredient in food systems such as bakery products to enhance dough formation.

Table 3: Functional Properties of *Cnidoscolus aconitifolius* Leaf Protein Concentrates

Functional properties	Value g/g
Water Absorption Capacity	588.71±0.6
Oil Absorption Capacity	295.21±1.0
Foaming Capacity	6.00±0.41
Foaming Stability	2.00±0.1
Emulsion Capacity	30.00±0.1
Emulsion Stability	40.44±0.1

The oil absorption capacity determined for the sample is 295.21±1.0g/g. The value is high compared with 195.00±3.00g/g and 200.00±2.0g/g reported for oven-dried and fluidized bed-dried unripe plantain flour respectively by Ariola *et al.,*2016. The oil absorption capacity of a food sample is important because it is an indication that the food sample will help to retain flavor and mouth feel. The high value of oil absorption capacity of this sample implies that it will be useful in food formation like whipped toppings, sausages, Chiffon desserts and sponge cakes. In addition, the ability of leaf protein concentrates of this sample to bind with oil makes it useful in a food system where optimal oil absorption is desired.

The emulsion capacity of the *Cnidoscolus aconitifolius* is 30.00±0.13g/g, 46.2±3.3g/g and 49.7±3.3g/g reported for chickpea, lentil and red lentil flours by Badia-Olmos *et al.,*2023. This implies that the tendency of the sample to promote dispersion of the food phases in which they are not well dissolved is high and the tendency of the sample to undergo changes that will affect its physical appearance is low and can therefore make the sample useful as a fat stabilizer in confectionaries.

The emulsion stability of the *Cnidoscolus aconitifolius* leaf protein concentrates is 40.44±0.1g/g. The value is lower than 47.7±2.1g/g reported for the emulsion stability of white beans by Chandra and Samsher 2013. Food or food ingredients with substantive emulsion stability of 25g/g have been recognized as an emulsifiers that would enhance the stability of food by increasing the kinetic stability. This implies that the leaf protein concentrates of the *Cnidoscolus aconitifolius* will be useful ingredients in pharmaceutical industries for the preparation of emulsions that could be useful in the preparation of creams and drugs like polysorbate Hasmnadi *et al.,*2020.

The foaming capacity of the leaf protein concentrate of *Cnidoscolus aconitifolius* is 6.00±0.41g/g. The value is low compared to 13.6±4.8g/g reported for Amaranth and 34.3±10.0g/g reported for Quinoa by Badia-Olmos 2023. Foaming capacity is the measure of the size of the interfacial area created by protein when forms are formed (Zhu *et al.,*2017). The low value of the foaming capacity of the sample may be due to a decrease in surface tension of the air and water interface, leading to the absorption of soluble protein molecules thereby promoting hydrophobic interactions.

The foaming stability of the leaf protein concentrate of *Cnidoscolus aconitifolius* is 2.00±0.1g/g. The value is low compared to 4.9±2.8g/g reported for Amaranth by Badia Olmos 2013. Food samples with reliable foam stability enhance an increase in protein concentration due to protein-protein interaction at the air-water interface which promotes the formation of a viscoelastic multiplayer film that offers resistance to the coalescence of bubbles. The value of the foaming stability of this sample is lower than its tendency to perform the characteristics listed above.

Table 4: Essential Amino Acid of *Cnidoscolus aconitifolius* Leaf Protein Concentrates

Essential Amino Acid	Value(mg/100g)
Leucine	7.19
Isoleucine	3.78
Lysine	4.61
Methionine	1.06
Phenylalanine	4.95
Threonine	3.01
Valine	3.24
Histidine	2.87
Tyrosine	3.32
Selenocystein	2.30

Table 5: Non-Essential Amino Acid of *Cnidoscolus aconitifolius* Leaf Protein Concentrates

Non-Essential Amino Acid	Value (mg/100g)
Alanine	2.50
Arginine	5.59
Aspartic acid	8.80
Cysteine	1.00
Glutamic acid	10.92
Glycine	1.12
Proline	2.25

Serine	2.45
TAA (Total Amino Acid)	70.96
TNEA (Total Non-Essential Amino Acid)	34.63
TEAA (Total Essential Amino Acid)	36.33
%TNAA	48.80
%TEAA	51.20

The values of the essential amino acid content obtained for the leaf protein concentrates of *Cnidoscolus aconitifolius* were reported in Table 4 while non-essential amino acids were reported in Table 5. Out of eighteen total amino acids determined for the sample, ten were essential amino acids while the remaining eight were non-essential amino acids. Leucine is one of the essential amino acids found in the leaf concentrate of the sample. The concentration of leucine present is 7.10mg/100g. The value is low compared with 20.76±0.58mg/100g reported for *Rhizphora mangle L.* by Moran Palacio *et al.*, 2014. It is also low compared with 34.9±0.65mg/100g reported for *Angelica sylvestris L.* by Budniak *et al.,* 2022. Leucine is a branched-chain amino acid, that assists in muscle repair, and protein synthesis; it facilitates quick wound restoration and an increase in hormone production Campbell.

The value of isoleucine determined for the sample is 3.78mg/100g. The value is in the range of 3.44mg/100g reported for *Indigofera astragaline* leaves by Itodo *et al.*,2010. Isoleucine enhances the formation of haemoglobin, and enhances the regulation of electrical signals. It enhances immune features.

Lysine concentration in the leaf protein concentrate of *Cnidoscolus aconitifolius* is 4.61mg/100g. The value is higher than 0.16mg/100g reported for the ethanolic leaf extract of *Rauwolfia vomitora* by Ugwu *et al.*,2019. Lysine is an essential amino acid that assists in the formation of collagen and tissue repair in the human body. Lysine performs a crucial role in energy maintenance, the building of muscle protein and tissue repairs. It also assists in the production of enzymes and hormones used to reduce the incidence of herpes infections.

The methionine content of the selected leaf protein concentrates is 1.06mg/100g, while the value of phenylalanine is 4.95mg/100g. the values of these two essential amino acids are higher than 0.77mg/100g and 3.68mg/100g reported by Itodo 2010 respectively for *Indigofera astragalina leaves*. Methionine is required for body detoxification, metabolism and absorption of zinc and selenium minerals in the body Budniak *et al.,* (2022). Phenylalanine is however a precursor to the neurotransmitter and norepinephrine, it is also needed for mental alertness proper functioning of the thyroid gland and blood vessel formation.

The value of threonine in the sample is 3.01mg/100g, valine is 3.24mg/100g. The value of methionine in this sample is lower than 15.4±0.18mg/100g reported for *Angelica sylvestris* by Budniak *et al.,* 2022, the value of valine is also low compared with 92.1±0.98mg/100g reported by same author for the same sample. Threonine forms the major part of structural proteins like elastin and collagen which assist proper development of skin and connective tissue. Threonine is also helpful in fat metabolism and immune function. The recommended daily allowance of valine and threonine is 26mg and 10.4mg respectively.

Valine however is an amino acid required for the growth of mammary glands and ovaries. It assists in the biosynthesis of protein, regulation of the blood sugar levels and provides the body with energy. Too much valine in the human body leads to diabetes due to the formation of insulin resistance. The value of valine in this sample is not beyond the amount that the human body could utilize optimally. The concentration of histidine is 2.87mg/100g while tyrosine is 3.32mg/100g. the values of the two essential amino acids are higher than 0.550 and 0.639mg/100g reported for *Rauwolfa vomitoria* by Ugwu Okechukwu 2019. Histidine is an important amino acid required for proper tissue growth and repair. It aids formation and storage of glycogen in the

liver. It provides relief for rheumatoid arthritis and orthopedic problem.

Tyrosine, however, assists in the regeneration of red and white blood cells, it is considered helpful in overcoming depression, memory improvement and mental alertness. The concentration of selenocysteine in the sample of 2.30mg/100g. It is an amino acid that assists in the utilization of selenium. It also helps to create positive feelings and a sense of mental energy or alertness. It is also required for the proper functioning of the thyroid gland and the flow of blood vessels.

A total number of eight amino acids were found in the leaf protein concentrates of the *Cnidoscolus aconitifolius*. The value of non-essential amino acids ranged from 10.92mg/100g in glutamic acid to 1.00mg/100g in cysteine. Glutamic acid and glycine participate in the synthesis of glutathione increasing the antioxidant of the plant. Aspartic acid (8.80mg/100g) is the value of the sample it is also one of the amino acids that is present in high concentration and likewise the arginine (5.59mg/100g). the non-essential amino acids also play an important role in human nutrition and metabolism. They are helpful in the maintenance of intestinal integrity and health, they are helpful in the formation of healthy nails, skin and hair.

The values of the amino acid profile present in the leaf protein concentrates of *Cnidoscolus aconitifolius* indicate that some medical issues due to malnutrition and insufficient consumption of crucial amino acids such as marasmus and kwashiorkor could be avoided through utilization of enough quantity of this sample of food ingredients.

Conclusion

Leaf protein concentrates of *Cnidoscolus aconitifolius* could be used as a food ingredient due to the presence of reliable values of nutritive, parameters, dietary minerals, functional properties and valuable amino acids. This is because when human diets lack one or more reliable nutrients, the symptoms experienced are similar to those of vitamin deficiencies such as eye infection, poor muscle tone, low blood pressure anaemia, weight loss, slow healing of wounds and low immunity. Thus sufficient amount of this sample could assist in overcoming these dietary problems. Therefore, these vegetable leaf protein concentrates could be processed and included in food formulations to boost the level of man and animal nutrition power.

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