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Proximate, Phytochemical and Amino Acid Compositions of Sodom Apple (*Calotropis procera*) Leaves and Fruits

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Abstract

In light of the escalating global challenges, the exploration of unconventional edible wild plants gains significance. This study focuses on *Calotropis procera*, renowned for its medicinal and toxic properties, particularly in its leaves and fruits. Analysis of these plant parts, following the Association of Official Analytical Chemists' procedures, revealed nutritional and phytochemical compositions (dry weight). Proximate composition (%) of leaves and fruits were: Moisture (9.03, 8.37), crude protein (20.81, 19.55), crude fiber (10.20, 20.91), crude fat (3.58, 3.41), ash (17.21, 16.83), and carbohydrate (39.19, 31.04), respectively. The predominant phytochemical was phytate with values of 116.5 and 83.72 (mg/100g) for leaves and fruits. Amino acid analysis revealed total contents of 72.43 and 70.82 g/100g crude protein for leaves and fruits, with the highest concentration in both being Glu (9.45 and 9.39 g/100g cp). Essential amino acids (with His) were 38.14 g/100g cp (52.67%) for leaves and 36.81 g/100g cp (51.98%) for fruits. While these findings highlight their potential as dietary supplements, further research is essential to develop processing methods that mitigate risks. This study contributes to understanding the nutritional and ant-nutritional profile of Sodom apple, emphasizing the importance of a balanced approach to harness its benefits effectively.

Keywords: *Calotropis procera*, proximate, phytochemicals, amino acids

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Introduction

Developing nations like Nigeria rely on a variety of wild plants for food and medicine, which plays a crucial role in meeting their nutritional needs and overall well-being [1]. These wild plants serve as primary sources of medicines, food, shelters and other items used by humans every day. The diverse parts of these plants, including roots, stems, leaves, flowers, fruits and seeds, play a crucial role in meeting the dietary needs of the population [2]. Acknowledging the pivotal role of wild plants in addressing the escalating challenges posed by population growth and the demand for medicinal and food resources, a growing focus has been directed towards the exploration of unconventional edible wild plants, with particular attention to leaves and fruits [3]. *C. procera* is known for its medicinal and toxic properties and therefore stands out in this regard. The plant is an evergreen, perennial shrub belonging to the sub-family *Apocynaceae* and family *Asclepiadaceae* [4]. The word *"Calotropis"* is derived from Greek which means "beautiful," which refers to its flowers, whereas *"procera"* is a Latin word referring to the cuticular wax present on its leaves and stem [5]. It is found in most parts of the world in dry, sandy, and alkaline soils and warm climate and is more common in north central and northern Nigeria [6]. The plant exhibits chemical protective systems due to its survival in hostile environments and has various chemical components [7]. It is known with various names, such as Sodom Apple, giant swallow wart (milkweed), auricula tree, giant milkweed, madar, mudar, rooster tree, small crown flower, Sodom's milkweed, and rubber bush [4]. In Nigeria, it is locally called 'bomubomu' in Yoruba language, 'tumfafiya' in Hausa language, 'epuko' in Nupe language [6]. All parts of the plant (latex being the richest) have toxic potential, due to the presence of cardenolides (cardiac glycosides). Besides the cardenolides, other phytochemicals are also reported from the plant such as sterols, flavonoids (rutin: quercetin-3-rutinoside), coumarins, alkaloids, triterpenes, saponins, tannins, and hydrocarbons were isolated from the plant. The plant is also reported to contain resins, fatty acids, proteases, hydrocarbons, amino acids, polyphenol, and many minerals [8].

Generally, there are two species of Calotropis: *C. procera* (purplish flower) and *C. gigantea* (Whitish flower). It is usually difficult to Physically differentiate between *C. procera* and *C. gigantea* without the presence of flowers in them since flower is the main point of difference between them.

Both species can be differentiated from each other by evaluating the pH of crude latex of stem. The colour of the milky sap of Calotropis is due to the presence of calcium oxalate crystals [9, 10].

The versatile applications of *C. procera* include its use in traditional medicine for treating a spectrum of ailments, ranging from fevers and wounds to asthma and cancer [11]. It is used for coagulating cheese by Fulanis in northern Nigeria [12, 13]. The pharmacological activities of *C. procera* have been popular in the past to cure several diseases in human beings such as cold, fever, leprosy, asthma, rheumatism, eczema, indigestion, diarrhea, elephantiasis, skin diseases, and dysentery [8, 14]. The plant is described as a golden gift for humankind containing cardiotonic agents such as calotropin, calotropagenin, calotoxin, calactin, uscharin, amyrin, amyrin esters, uscharidin, coroglaucigenin, frugoside, corotoxigenin, calotropagenin, and voruscharine used in the therapeutic treatment [8]. *C. procera* holds diverse global economic potential, encompassing pharmacological applications, natural fiber source, phytoremediation, green technology utilization as nanoparticles, and alternative uses such as fodder, fuel, timber, biofuel, and ornamental value. Additionally, it serves as a coagulant in cheese production, insecticides/nematicides, and fungicides. With allelopathic activity, it aids in environmental monitoring and bioremediation. Moreover, Calotropis acts as a natural colorant for textile fabrics, and *C. procera* stands as an alternative raw material for crafting high-quality handmade paper [7, 4, 15]. *C. procera* has been declared as an invasive species in several regions of the world. It is a serious environmental weed of South America, the Caribbean Islands, Australia, the Hawaiian Islands, Mexico, Seychelles, and several Pacific Islands [16]. The plant's dispersal is facilitated by its silky floss seeds, carried by wind, floodwater, livestock, and other animals over long distances [17, 18].

The *C. procera* leaves and fruits, despite its traditional medicinal uses and its potential for yielding valuable hydrocarbons as diesel substitutes [19], there is a lack of comprehensive scientific data on the nutritional value and potential anti-nutritional factors present in this plant species. Understanding the nutritional composition, including macro and micronutrients, as well as the presence of anti-nutritional factors such as toxins, antinutrients, or allergens, is crucial for assessing the safety and potential health benefits of Sodom apple consumption. Therefore, this study was undertaken to analyze and quantify the proximate, photochemical and amino acid compositions in *C. procera* leaves and fruits, providing valuable insights into its nutritional potential and potential risks associated with its consumption.

Plate 1: *C. procera* plant (leaves and fruits)

Plate 2: *C. procera* plant as an invasive weeds across African and Asian countries [20]

Plate 3: A roadside infestation of *C. procera* [20]

Plate 4: Seeds and seed pod of *C. procera* [20]

Materials and Methods

Sample collection

The matured fresh leaves and fruits of *C. procera* were collected randomly from branches of *C. procera* tree at Federal University of Lafia main campus, Nasarawa state, Nigeria. Identification of the samples was carried out in Botany laboratory, Department of Plants Science and Biotechnology, Federal University of Lafia.

Sample treatment

The samples were brought into the laboratory, properly washed, oven-dried, cooled and blended into a fine powder using electric blender. The ground portions were put in a tight plastic container and kept in a refrigerator at about 4^0C prior to analysis.

Proximate composition

The ash, moisture, crude protein (N x 6.25), crude fat, crude fibre and carbohydrate (by difference) were determined in accordance with the standard methods of [21]. All proximate analyses of the sample flours were carried out in triplicate and reported in %. All chemicals were of Analar grade. All results were on dry weight (dw) basis.

Phytochemical analysis

The contents of oxalate, saponins, alkaloids, flavonoids, tannins, cyanide, phytate, and total phenols were determined on each of the sample flours by methods described by some workers [22].

Amino acid analysis

The amino acid analysis was by Ion Exchange Chromatography (IEC) [23] using the Technico Sequential Multisample (TSM) Amino Acid Analyzer (Technicon Instruments Corporation, New York). The period of analysis was 76 min for each sample. The gas flow rate was 0.50 $mLmin^{-1}$ at 60^{o}C with reproducibility consistent within ± 3%. The net height of each peak produced by the chart recorder of the TSM (each representing an amino acid) was measured and calculated. Amino acid values reported were the averages of two determinations. Nor–leucine was the internal standard. Tryptophan was determined after alkali (NaOH) hydrolysis by the colorimetric method.

Determination of isoelectric point (pI), quality of dietary protein and predicted protein efficiency ratio (P–PER)

The predicted isoelectric point was evaluated according to Olaofe and Akintayo [24]:

$$plm = \sum_{i=1}^{n=1} pliXi \quad ---------(1)$$

Where:

pIm = the isoelectric point of the mixture of amino acids;
Xi = the mass or mole fraction of the amino acids in the mixture.
pIi = the isoelectric point of the *i*th amino acids in the mixture;

The quality of dietary protein was measured by finding the ratio of available amino acids in the sample protein compared with the needs expressed as a ratio. Amino acid score (AAS) was then estimated by applying the formula [25]:

$$AAS = \frac{mg\ of\ amino\ acid\ in\ 1g\ of\ test\ protein}{mg\ of\ amino\ acid\ in\ reference\ protein} \times \frac{100}{1} \quad ----- (2)$$

The predicted protein efficiency ratio (P–PER) of the seed sample was calculated from their amino acid composition based on the equation developed by Alsmeyer *et al.* [26] as stated thus;

P–PER = –0.468 + 0.454 (Leu) – 0.105 (Tyr) - - - (3)

Statistical Analysis of the Samples

The fatty acid values were obtained by multiplying crude fat value of each sample with a factor of 0.8 (i.e. crude fat × 0.8 = corresponding to fatty acids value). The energy values were calculated by adding up the carbohydrates × 17 kJ, crude protein × 17 kJ and crude fat × 37 kJ for each of the samples [27, 28]. Errors of three determinations were computed as standard deviation (SD) for the proximate composition by using MS Excel Spread Sheet. The mean, standard deviation and coefficient of variation (%) for variability test on the fish samples were also analyzed.

Results and Discussion

The result of proximate composition is reported in Table 1. The study showed that the leaves of *C. procera* boast a slightly higher crude protein percentage (20.81%) compared to the fruits (19.55%). These figures align with the findings of [29], who reported even higher value for leaves (26.69%) and lower value for fruits (14.48%). However, the stark contrast emerges when compared with the study by [30], where a mere 1.16% crude protein was reported, underscoring the variability in protein content within the same plant species. Ajiboso *et al.* [6] highlighted a substantial protein content of 28.53% in *C. procera* leaves, indicating their value for growth and tissue repair. Comparisons with well-known vegetables like cabbage and lettuce, as per USDA data, revealed that *C. procera's* leaves stand out as a rich source of protein. With a protein content of 12.8% in cabbage and 14% in lettuce, the leaves of *C. procera* offer a competitive alternative in the pursuit of addressing protein energy malnutrition.

Fat is a lipid component, and the energy derived from it could be utilized for body maintenance. The quantity of crude fat in the leaves (3.58%) was higher than that of the fruits (3.41 %). Ogundola *et al*. [29] observed 21.7% fat in the leaves and 6.29% in the fruit of *C. procera*. This shows that the oil in this present study is relatively lower than the values obtained by [29]. Ajiboso *et al.* [6] reported a high lipid content of 20.42% in *C. procera* leaves, indicating a substantial amount of oil.

The ash content in plant material serves as an indicator of mineral element levels, reflecting potential nutritional and environmental variations. In the current investigation, the ash content analysis revealed a higher level in the leaves (17.21%) compared to the fruits (16.83%).

This aligns with the anticipated notion that mineral elements are more concentrated in leaves, making them a potentially richer source of inorganic elements than fruits [31]. This finding is consistent with the study of Ogundola *et al*. [29], where the ash content in leaves (5.32%) surpassed that in fruits (3.69%). However, it's noteworthy that the present study reported considerably higher ash content compared to Ogundola *et al*. [29]. This discrepancy may be attributed to the distinct geographical locations where the plants were cultivated, suggesting a potential influence of environmental factors on mineral accumulation. Adding to the context, [32] reported the total ash content of *C. procera* as 18.3±0.8 mg/g. This reference provides an additional benchmark for ash content in *C. procera*, contributing to the broader understanding of the species' mineral composition. The crude fiber analysis revealed a notable disparity between the fruit (20.91%) and leaf (10.20%) components of *C. procera*. This finding is consistent with Ogundola *et al*. [29] research, which reported higher fiber levels in the fruits (15.73%) compared to the leaves (7.54%). Adding to this body of knowledge, Rehman *et al*. [33] recent investigation delved into specific extracts of *C. procera*, demonstrating higher crude fiber content (21.3 mg) in leaf extracts compared to flower extracts (18.5 mg), while the root extract records lower levels (10.5 mg). Expanding the scope to *Calotropis gigantea*, [33] note higher crude fiber in leaf extracts (23.4 mg) compared to flowers (23.1 mg), with the root extract registering a lower content (23 mg). The nuanced examination of different plant parts adds depth to our understanding of the variability in crude fiber content within the *Calotropis genus*. Fiber is characterized as a nutraceutical for its health benefits, plays a crucial role despite being indigestible by humans. It provides essential roughage that aids digestion and, notably, can influence glucose absorption and insulin secretion, holding potential benefits for diabetic patients [34]. Incorporating ground seeds into low-fiber flour meals emerges as a practical strategy to prevent constipation. The broader health implications are significant, as a high-fiber diet has been linked to lower cholesterol levels, reduced risks of various cancers and bowel diseases, and an overall improvement in general health and well-being [29, 35].

The moisture content in the leaves (9.03%) of *C. procera* exceeded that of the fruits (8.39%). Similar value (10.92%.) was reported for the moisture content of aqueous leaf extract of *C. procera* [6]. In contrast, Ogundola *et al*. [29] found lower leaves moisture content (8.11%) compared to the fruit (9.53%). *C. procera's* moisture content, both in leaves and fruits, was notably lower than conventional leafy vegetables of 55.76 ± 0.05 to 91.83 ± 0.04 g/100 g moisture [36]. Moisture content is among the most vital factors considered in food processing, preservation, and storage [37]. Ajiboso *et al*. [6] concluded that the low moisture content in *C. procera's* aqueous leaf extract indicates low perishability, hindering microorganism growth and potentially ensuring a longer shelf-life.

The current study reveals a carbohydrate content of 39.19 and 31.04% in *C. procera* leaves and fruits, respectively. Contrasting findings from Ogundola *et al*. [29] showed 30.64% in leaves and 50.29% in fruits. Ajiboso *et al*. [6] found 24.13% carbohydrate content, and Al–Snafi [38] recorded a 50% variation range in *C. procera* flower carbohydrates between summer (9.45 g%) and winter (18.46 g%). These diverse carbohydrate levels, crucial for energy, underscore *C. procera*'s potential as a significant dietary energy source.

The calculated fatty acids indicated higher level in *C. procera* leaves (2.86%) compared to fruits (2.73%). This disparity emphasizes the richer fatty acid content in the leaves. In terms of caloric value, *C. procera* leaves exhibited a higher value of 1152.46 kJ/100g compared to fruits at 986.246 kJ/100 g and this shows that both samples have energy concentrations favourably compare to cereal. This discrepancy underscores the greater energy content in the leaves. The CV% varied from 1.12% in ash content to 34.38% in crude fibre.

Table 1: Proximate composition (%) of Sodom apple (*C. procera*) leaves and fruits

Parameter	*C. procera* Leaves	*C. procera* Fruits	Mean	SD	CV(%)
Crude protein	20.81±0.02	19.55±0.11	20.18	0.63	3.12
Crude fat	3.58±0.08	3.41±0.01	3.50	0.09	2.57
Ash	17.21±0.09	16.83±0.07	17.02	0.19	1.12
Crude fibre	10.20±0.07	20.91±0.09	15.56	5.35	34.38
Moisture	9.03±0.01	8.37±0.21	8.7	0.33	3.79
Carbohydrate	39.19±0.17	31.04±0.14	35.12	4.08	11.62
Fatty acid*	2.86	2.73	2.80	0.22	7.86
Energy (kJ/100g)[a]	1152.46	986.2	1069.33	83.13	7.77

All values are the mean ± standard deviation of three determinations expressed in dry weight basis; *Calculated fatty acids; [a]Calculated metabolizable energy; SD = Standard Deviation; CV = Coefficient of Variation

The result of phytochemical is presented in Table 2. *C. procera* leaves and fruits contained oxalate levels of 12.18% and 11.1%, respectively, which are lower than those in *Solanum nigrum* (5.81 mg/100 g) and *Anetum africanmum* (20.9 mg/100g) [39]. Despite its lower oxalate content, the broader utilization of tropical plants like *C. procera* in food is limited by anti-nutrients universally present in them, affecting nutrient bioavailability [40]. Oxalates causes irritation and swelling in the mouth and throat, and phytate as inhibiting digestive enzymes are significant anti-nutrients. Moreover, the oxalate values for *C. procera* exceed those in soybean and pigeon pea [41], *Daucus carota* and *Cucumis sativus* [42], as well as red kidney bean and black turtle bean [43]. The concentrations of phytate of *C. procera* leaves and fruits were 116 and 83.72 mg/100g, respectively. Phytate and oxalates form poorly soluble compounds with metallic ions, affecting their bioavailability. While the presence of phytate is crucial, high concentrations negatively impact digestion [44]. The anti-inflammatory, antioxidant, anti-allergic, antiviral, and anti-carcinogenic properties found in the secondary metabolites of *C. procera* indicate its potential health benefits [45].

Nevertheless, the antioxidant property of phytate comes at a cost, inhibiting mineral absorption and reducing zinc and iron bioavailability [46]. Saponin values in *C. procera* leaves and fruits were 0.94 and 0.71 mg/100g, respectively. While saponins are widespread in plants, they exhibit diverse effects on animal metabolism and health. Some of these effects include bloating in ruminants, reduced nutrient absorption, decreased liver cholesterol, altered growth rate, and hindrance of nutrient absorption in the small intestine. Despite their generally beneficial nature, certain saponins can be poisonous, causing skin rashes if ingested. On the positive side, specific saponins contribute to controlling cholesterol levels [9, 47]. Tannin concentrations in *C. procera* leaves and fruits were 96.7 and 44.99 mg/100g, respectively. With antibacterial potential, tannins react with proteins, forming stable water-soluble compounds that damage bacterial cell membranes [47]. However, they cause inactivation of digestive enzymes and decrease protein digestibility in animals [48]. Tannins, generally found in woody plants, exist in hydrolyzed or condensed forms, binding to proteins, starches, cellulose, and minerals. This binding property is effectively utilized in the leather industry for tanning processes [9]. The *C. procera* leaves and fruits were found to contain 5.59 and 3.15 mg/100g of alkaloids, and 1.37 and 1.22 mg/100g of flavonoids, respectively. Alkaloids and flavonoids, possessing antioxidant properties, provide health benefits by acting against cancer and heart disease. Cyanide levels in *C. procera* leaves and fruits were observed to be 10.73 and 9.28 mg/100g, respectively, exceeding the permissible limit of 200 mg/kg fresh weight for vegetables or forages [49]. Whereas, phytate amounts in *C. procera* were 116.5 ± 0.19 and 83.72 ± 0.37 for leaves and fruits, respectively. Cyanogens, glycosides releasing cyanide, can cause energy deprivation and death. Total phenol concentrations were detected as 5.51 and 3.5 mg/100g, respectively. Phenolic compounds are crucial antioxidants in plant extracts, play a vital role in inactivating hydroxide and peroxide radicals, preventing cytotoxicity and cancer. Plant extracts containing antimicrobial compounds present a viable alternative to antibiotics, which have developed resistance in recent years [15].

Table 2: **Phytochemical content of Sodom apple (*C. procera)* leaves and fruits**

Parameter	*C. procera* Leaves	*C. procera* Fruits	Mean	SD	CV(%)
Oxalate (%)	12.18±0.09	11.10±0.09	11.64	0.54	4.64
Saponins (%)	0.94±0.01	0.71±0.01	0.83	0.12	14.46
Alkaloids(%)	5.59±0.02	3.15±0.05	4.37	1.22	27.92
Flavonoids (%)	1.37±0.04	1.22±0.02	1.30	0.08	6.15
Tannins(mg/100g)	96.7±0.08	44.99±0.07	70.85	25.86	36.50
Cyanide(mg/100g)	10.73±0.13	9.28±0.11	10.01	0.73	7.29
Phytate (mg/100g)	116.5±0.19	83.72±0.37	100.15	16.43	16.45
Total Phenol (%)	5.51±0.02	3.50±0.08	4.51	1.01	22.39

All values are the mean± standard deviation of three determinations expressed in dry weight basis; SD = Standard Deviation; CV = Coefficient of Variance

Table 3: Amino acid composition (g/100 g crude protein of Sodom apple (*C. procera)* leaves and fruits

Parameter	*C. procera* Leaves	*C. procera* Fruits	Mean	SD	CV (%)
Leucine (Leu)[e]	8.13	7.82	7.98	0.16	2.01
Lysine (Lys)[e]	4.02	3.63	3.83	0.20	5.22
Isoleucine (Ile)[e]	3.61	4.03	3.82	0.21	5.50
Phenylalanine (Phe)[e]	4.22	3.90	4.06	0.16	3.94
Tryptophan (Trp)[e]	0.76	0.84	0.80	0.04	5.00
Valine (Val)[e]	3.85	3.65	3.75	0.10	2.67
Methionine (Met)	1.12	0.91	1.02	0.11	10.78
Proline (Pro)	3.37	3.55	3.46	0.09	2.60
Arginine (Arg)[e]	5.01	4.73	4.87	0.14	2.87
Tyrosine (Tyr)	2.94	3.10	3.02	0.09	2.98
Histidine (His)[e]	2.16	1.82	1.99	1.38	69.35
Cystine (Cys)	1.13	1.03	1.08	0.05	4.63
Alanine (Ala)	4.20	5.04	4.62	0.42	9.09
Glutamic acid (Glu)	9.45	9.39	9.42	0.03	0.32
Glycine (Gly)	2.96	3.02	2.99	0.03	1.00
Threonine (Thr)[e]	3.01	2.86	2.94	0.03	1.02
Serine (Ser)	3.61	3.30	3.46	0.16	4.62
Aspartic acid (Asp)	8.88	8.22	8.55	0.33	3.86
P-PER	2.91	2.75	2.83	0.08	2.83
Leu/Ile	2.25	1.94	2.10	0.19	9.05

[e] = Essential amino acid, P–PER = Predicted Protein Efficiency Ratio, SD = Standard Deviation, Leu/Ile = Leucine/Isoleucine ratio, CV = Coefficient of Variation

The result of amino acid composition of the samples of *C. procera* leaves and fruits is shown in Table 3. The most abundant and non essential amino acids (NEAA) in both samples of *C. procera* leaves and fruits were glutamic acid with concentrations of 9.45 and 9.39 g/100 g crude protein (cp) respectively. The second is aspartic acid with concentrations of 8.88 g/100g cp and 8.22 g/100 g cp respectively. Tyrosine has a concentration of 2.94 and 3.10 g/100g cp in the leaves and fruits samples of *C. procera.* Tyrosine is a NEAA and serves as a precursor of some hormones like the thyroid hormones and the brown pigment melanine formed in hair, eyes, and tanned skin [50].

Leucine has the most abundant essential amino acid (EAA) with the concentrations of 8.13 g/100 g cp in leaves and 7.82 g/100 g cp in fruits of *C. procera*. This result is consistent with the findings of Aremu *et al.* [51] on *M. charantia* with concentration of 7.20 g/100 g cp. Arginine which is also an EAA in the sample and is responsible for growth in children has concentrations of 5.01 and 4.73 g/100 g cp of *C. procera*'s leaves and fruits respectively. This is consistent with the findings of Aremu *et al.* [51] on *M. charantia* and *D mespiliformis* with concentrations of 5.33 and 3.01 g/100g cp for both samples.

The lysine content is 4.02 g/100g cp for leaves and 3.63 g/100g cp for the fruits. Both samples are less than the 6.3 g/100 g content of the reference egg protein [51]. Phenylalanine has the concentration of 4.22 g/100g cp in the leaves and 3.90 g/100g cp in the fruits. The phenylalanine content here is higher than reported by Aremu *et al.* [51] (3.30 and 2.13 g/100 g cp). Phenylalanine is the precursor of some hormones and pigment melanin in hair, eyes and tanned skin. Phenylketonuria is the commonest inborn error of metabolism successfully treated by diet.

The absence of enzymes in the liver blocks the normal metabolism of phenylalanine and the brain is irreversibly damaged unless a diet low phenylalanine is given in the first few weeks of life [51]. Isoleucine (3.61 and 4.03 g/100 g cp) is an essential amino acid for both old and young. It is comparable to that of Aremu *et al*. [51] which recorded (4.12 and 2.30 g/100 g cp). Maple syrup urine disease is an inborn error of metabolism in which brain damage and early death can be avoided by diet low in isoleucine and two other essential amino acids, valine and leucine [51]. Proline has a concentration of (3.37 and 3.55 g/100g cp) for *C. procera*'s leave and fruits samples. Threonine has concentrations of 3.01 and 2.86 g/100g cp leaves and fruits samples of *C. procera.* Cystine was found to be (1.13 and 1.03 g/100g cp) which is consistent with that obtained by [51] as seen in the concentrations of 1.09 and 1.33 g/100g cp in both samples of *M. charantia* fruits and pulp of *D. mespiliformis*.

Tryptophan contained (0.76 and 0.84 g/100g) for the leaves and fruits of *C. procera*, respectively. This is comparable with the values reported by Aremu *et al*. [51], where the concentrations were (0.42 and 0.72 g/100 g) for both samples of *M. charantia* fruits and pulp of *D. mespiliformis*, respectively. This concentration was found to be the least concentrated amino acids. Tryptophan is one of the biochemically active amino acids which plays a significant role in the protein and enzyme syntheses, cognition and neurohormonal regulation [52]. The predicted protein efficiency ratio (P–PER) values in this report were 2.91 g/100g cp and 2.75 g/100g cp for the leaves and fruits of *C. procera* respectively. These values are higher than those reported for *M. charantia* fruits (2.55) and pulp of *D. mespiliformis* (1.09) [51], higher than some legume flours/concentrates: *Prosopis africana*, *Kerstingella geocarpa* and *Vigna subterranean* (1.03) [53, 54, 55]. Above all, P-PER is one of the quality parameters used for protein evaluation and the *C. procera*'s P-PER values for this investigation satisfy [55, 56] requirements. The Leu/Ile values from the study were 2.25 g/100g cp and 1.94 g/100g cp for the leaves and fruits respectively. These values are higher than that obtained by [51] that is 1.75 g/100 g cp for *M. charantia* and 1.57 g/100 g cp for *D. mespiliformis*, respectively. The nutritive value of a protein depends primarily on the capacity to satisfy the needs for nitrogen and essential amino acids [57]. The CV% varied from 1.00 in Gly to 69.35 in His.

Table 4: Concentrations of essential, non–essential, neutral, sulphur, aromatic amino acids, etc. (g/100 g crude protein) of Sodom apple (*C. procera)* leaves and fruits

Amino acid description	*C.procera* Leaves	*C. procera* Fruits	Mean	SD	CV (%)
Total amino acids (TAA)	72.43	70.82	71.63	0.01	0.01
%Total amino Acids	100	100	100	0.00	0.00
Total non–essential amino acids (TNEAA)	34.29	34.01	34.15	0.14	0.41
% TNEAA	47.34	48.02	47.68	0.34	0.71
Total essential amino acids (TEAA)					
With histidine	38.14	36.81	37.48	0.67	1.79
Without histidine	35.98	34.99	35.49	0.56	1.58
% TEAA					
With histidine	52.67	51.98	52.33	0.35	0.67
Without histidine	49.68	49.41	49.55	0.14	0.28
Essential aliphatic amino acids (EAAA)	29.37	29.72	29.55	0.17	0.58
Essential aromatic amino acids (EArAA)	13.45	13.21	13.33	0.12	0.90
Total neutral amino acids (TNAA)	29.37	29.72	29.55	0.17	0.58
% TNAA	40.55	41.97	41.26	0.71	1.72
Total acidic amino acids (TAAA)	18.33	17.16	17.75	0.56	3.15
% TAAA	25.31	24.23	24.77	0.54	2.13
Total basic amino acids (TBAA)	11.19	10.18	10.69	0.51	4.77
% TBAA	15.45	14.37	14.91	0.54	3.62
Total sulphur amino acids (TSAA)	2.25	1.94	2.10	0.16	7.62
% Cystine in TSAA	50.22	53.09	51.66	1.43	2.77

SD = Standard deviation; CV = Coefficient of variation

Table 4 depicts the essential, non–essential, acidic, neutral and sulphur containing amino acids. The total essential amino acids (TEAA) (with His) of *C. procera* was 38.14 g/100 g cp (leaves) and 36.81 g/100 g cp (fruits). TEA without His is 35.98 g/100 g cp and 34.99 g/100 g cp respectively. Aremu *et al*. [51] reported 22.93 g/100 g cp for *Daucus carota L*. and 30.11 g/100 g cp for *Cucumis sativus L.* which agree with the current studies. Aja *et al*. [58] recorded for the leaves of *S. aethiopicum*, *A. hybridus* and *T. occidentalis* with 40.75, 37.21 and 38.71 g/100g respectively, these values are higher than those of the current study on *C. procera.* This is less than that of some Nigeria legume protein concentrates: lima bean (44.88 g/100 g cp), pigeon pea (48.11 g/100 g cp), and African yam bean (48.28 g/100 g cp) reported by Oshodi *et al*. [59] and tiger nut (41.21 g/100 g cp) by Aremu *et al*. [60].

Nevertheless, the TEAA contents (%) of *C. procera* (leaves and fruits) in this report are well above the 39% considered to be adequate for ideal protein food for infants, 26% for children and 11% for adults [55]. The concentrations of total sulphur amino acids (TSAA) were 2.25 and 1.94 g/100 g cp in *C. procera'*s leaves and fruits and are lower than the 5.8 g/100 g cp recommended for infants [73]. Similar result was reported in the works of Aremu *et al.* [51]. The values of essential aromatic acids (EArAA) are 13.45 and 13.21 g/ 100g cp for leaves and fruits respectively. These values are higher than the ideal range suggested for infant protein of 6.8 – 11.8 g/100 g cp as prescribed in FAO/ WHO/UNU [55]. Lowervalues were reported by Aremu *et al.* [51].

The total acidic amino acids (TAAA) 18.33 and 17.16 g/100g cp and that of the %TAAA is 25.31 and 24.23 g/100g cp for leaves and fruits of *C. procera* plants respectively. Lower values were reported by [51]. The total basic amino acids (TBAA) are 11.19 and 10.18 g/100g cp. That of the %TBAA is 15.45 and 14.37 g/100g cp respectively. The result showed clearly that the TAAA and %TAAA values are greater than the TBAA and %TBAA values thus implying that their protein is probably acidic in nature. This result is consistent with that reported by Oshodi *et al.* [50].

Table 5: Amino acids scores of Sodom apple (*C. procera*) leaves and fruits based on FAO/WHO standards

EAA	PAAESP g/100g protein	*C. procera* leaves		*C. procera* fruits	
		EAAC	AAS	EAAC	AAS
Ile	4.0	3.61	0.90	4.03	1.01
Leu	7.0	8.13	1.15	7.82	1.12
Lys	5.5	4.02	0.73	3.63	0.66
Met + Cys (TSAA)	3.5	5.06	1.45	3.36	0.96
Phe + Tyr	6.0	7.16	1.19	7.0	1.17
Thr	4.0	3.01	0.75	2.86	0.72
Trp	1.0	0.76	0.76	0.84	0.84
Val	5.0	3.85	0.77	3.65	0.73
Total	**36.0**	**37.02**	**9.12**	**34.61**	**8.63**

EAA = Essential amino acids; PAAESP = Provisional amino acids (Egg) scoring pattern; EAAC = Essential amino acid composition; AAS = Amino acids score

The essential amino acid (EAA) scores of the samples based on (FAO/WHO) scoring pattern are presented in Table 5. Considering the scores of the two samples, the results showed that leaves will supply more essential amino acids than fruits. The (Ile) score in leaves (0.90) is lower than that of fruits (1.01), however the scores in both leaves and fruits of *C. procera* suggest that they are good sources of isoleucine. Isoleucine is important for protein synthesis and energy production also important for muscle metabolism, immune function, and energy regulation. (Leu) score values are almost the same in both leaves and fruits (1.15 and 1.12). Leucine is involved in muscle protein synthesis and plays a key role in regulating muscle mass, muscle growth, and maintenance. The scores imply that the leaves and fruits of *C. procera* are rich in leucine. Lysine is essential for growth, tissue repair, and the production of enzymes, hormones, collagen formation, proper calcium absorption and antibodies. The score values in leaves and fruits (0.73 and 0.66), of *Calotropis procera* may not be a significant source of (Lys) compared to other essential amino acids. (Met + Cys) are sulfur-containing amino acids that are important for protein synthesis, antioxidant defense, and liver function and various metabolic processes. The score of (1.45 and 0.96) suggests that *C. procera* leaves and fruits are relatively good source of these amino acids. (Phe + Tyr) are involved in the production of neurotransmitters, hormones and protein formation. The score of (1.19 and 1.17) indicates that *C. procera* leaves and fruits contain a moderate amount of these amino acids. Thr is necessary for the synthesis of proteins and the formation of collagen, elastin, antibody production and nutrient absorption. The score of (0.76 and 0.72) suggests that *C. procera* leaves and fruits provide less amount of threonine. Trp is a precursor for serotonin and melatonin, which regulate mood and sleep. The score of (0.74 and 0.84) indicates that *C. procera* leaves and fruits contain a moderate amount of tryptophan. Val is involved in muscle metabolism, tissue repair, energy production and the maintenance of nitrogen balance. The score for both samples are almost the same (0.72 and 0.73) suggests that the valine content in *C. procera* leaves and fruits may be relatively lower compared to other essential amino acids.

Overall, the essential amino acids score indicates that the leaves and fruits of *C. procera* are a good source of several essential amino acids, including Ile, Leu, (Met + Cys), (Phe + Tyr), Thr, Trp, and Val. However, the contents of (Lys, Thr, Trp and Val) and (Lys) in leaves and fruits, respectively appear to be relatively lower and dietary formula based on these food samples will require supplementation of these essential amino acids. It's important to note that this discussion is based on the provided amino acid scores and does not take into account other nutritional factors or potential toxicity or allergenicity or phytochemical components that may be present in *C. procera* leaves and fruits. The limiting amino acids (LAA) for both the leaves and fruits of *C. procera* was Lys (0.73 and 0.66), respectively.

Conclusion

Sodom apple's leaves and fruits exhibited nutritional richness, highlighting their potential as dietary supplements. However, the presence of some anti-nutritional components, notably high oxalates and cyanides, necessitates caution in consumption. Further research is crucial for developing processing methods that mitigate risks. This study enhances our understanding of Sodom apple's nutritional and anti-nutritional profile, emphasizing the need for a balanced approach to harness its benefits effectively.

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