



Journal of Nutrition and Food Security

Shahid Sadoughi University of Medical Sciences
School of Public Health
Department of Nutrition
Nutrition & Food Security Research Center



eISSN: 2476-7425

pISSN: 2476-7417

JNFS 2023; 8(3): 461-467

Website: jnfs.ssu.ac.ir

*Chaya (*Cnidoscolus Aconitifolius*) for Enhancing Food and Nutrition Security of Arid Lands of Ethiopia*

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ARTICLE INFO

ORIGINAL ARTICLE

Article history:

Received: 10 Feb 2022

Revised: 28 May 2022

Accepted: 28 May 2022

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ABSTRACT

Background: Chaya is a drought tolerant, fast growing, perennial and nutritionally rich leaf. Its nutritional values vary depending on the environment and agronomic practices. **Methods:** Proximate compositions, minerals, and phytochemicals analysis of the newly introduced Chaya leaf grown at Dire Dawa (arid land), Ethiopia, were determined using official standard methods.

Results: Moisture content, protein, crude fat, crude fiber, ash, and carbohydrate were 5.63, 23.96, 8.98, 2.87, 9.05, and 49.49%, respectively. In addition, the energy content of leaf was found 374.62 kcal/100g. Mean values of each mineral such as calcium (Ca), iron (Fe), and zinc (Zn) were 253.68, 68.02, and 4.85 mg/100g, respectively. Phytochemical analysis revealed that phenols, tannin and hydrogen cyanide contents of Chaya leaf were 1916.66, 176.53, and 102.00 mg/100g, respectively. **Conclusions:** Proximate composition, minerals, and phytochemicals make the plant valuable and a health promoting diet for communities settled in dry areas of the country. Therefore, Chaya has the potential to contribute to food and nutrition security, and health for the community suffering from malnutrition in drought prone areas.

Keywords: Food security; Malnutrition; Micronutrients; Nutritional status; Phenols

Introduction

Of the nearly 240 million people in sub-Saharan Africa, one in four people lacks adequate food for a healthy and active life (Food and Agriculture Organisation, 2010). Food prices and drought are pushing more people into poverty and hunger. It is widely recognized that climate change and variability have an impact on agro-ecosystems, affecting the productive capacities and sustainability of agriculture sectors. This can

have socio-economic implications on livelihoods and food security and nutrition. According to United Nations report, millions of children die every year due to malnutrition (Unicef, 2012). Lack of key micronutrients causes children with severe consequences on their physical and intellectual mental development.

In line with this issue, the 2005 Dietary Guideline Advisory Committee recommended an

This paper should be cited as: Alemu Gobena D, Girma Shewa A, Abera S, W/Tsadik K, Neme G, Mahamed W. *Chaya (*Cnidoscolus Aconitifolius*) for Enhancing food and Nutrition Security of Arid Lands of Ethiopia. Journal of Nutrition and Food Security (JNFS), 2023; 8(3): 461-467.*

increase in dietary intake of antioxidant vitamins (e.g. vitamins A and C) and dietary fiber from vegetables and fruits in order to reduce micronutrient malnutrition, the risk of cardiovascular disease, stroke, and cancer. The Advisory Committee recommended that fruits and vegetables intake should increase to 5-13 servings per day depending on individuals' calorie needs (Thompson and Veneman, 2005). According to the author (Naude, 2013), at least one serving of dark-green leafy vegetables could provide the daily requirements for vitamins and minerals as well as reduce the burden of nutrition-related disease.

In many developing countries, to meet the daily requirements of micronutrients in deficient population, identifying and inclusion of nutritionally rich plants in the diet is the best option (Tanumihardjo, 2008). Underutilized crops have an underexploited potential to contribute to food and nutrition security, health, income generation, and environmental services. Across most regions of Ethiopia, households report lack of/erratic rainfall as the main risk contributing to their food and nutrition insecurity and overall vulnerability. To overcome such problems, introducing, characterizing and recommending drought tolerant and nutritionally rich food sources for consumption is highly recommended for moisture stress prone areas in addition to other alternative solutions.

Chaya (*Cnidoscolus aconitifolius*) belongs to the Euphorbiaceae family. It is a fast-growing, drought-tolerant, nutritionally rich, perennial shrub, typically reaching 6 m in height. It is cultivated for producing abundant dark-green leaves. Chaya has been used for food since pre-Columbian times, and is still eaten regularly, especially in Central America and Southern Mexico. Tender stem tips are cut and leaves are picked as needed, and are used immediately or stored for few days. Leaves and even petioles and stems, can be dried and ground to make a storable product. Although drying helps reduce hydrogen cyanide (HCN) content, this powder should still be cooked before consumption (Berkelaar, 2006)

Chaya leaves are an excellent source of a number of essential nutrients for a healthy, balanced diet relatively higher than other green leafy vegetables such as spinach, Chinese cabbage, and amaranth. The leaves are very high in protein, calcium (Ca), iron (Fe), and vitamins A and C. Amino acids in Chaya are well balanced, which is important for those who have a diet low in protein and for children and pregnant or nursing mothers. Young leaves and the thick, tender stem tips are cut and boiled as "spinach" (Berkelaar, 2006). The blood glucose lowering potential of Chaya plant extract was justified by the strong presence of flavonoids in its phytochemical analysis (Hussain and Marouf, 2013, Vinayagam and Xu, 2015). Various *in vitro* studies have revealed the free radical scavenging potential of Chaya (Oloyede *et al.*, 2010).

Moreover, it could enhance food and nutrition security of moisture stress areas in Ethiopia due to its potential drought resistance, fast growing, available throughout the year, and nutritionally rich. There are considerable variations among the nutritional values of Chaya, depending on factors like environment and agronomic practices. Therefore, this study was conducted with the aim of characterizing and introducing multipurpose, drought tolerant, and nutritionally rich vegetable crop, Chaya, to eastern Ethiopia as a new food.

Materials and Methods

Sample source: Young Chaya leaves (*Cnidoscolus aconitifolius*) were collected at the age of two months after planting from Dire Dawa Agricultural Research Station of Haramaya University (Tony farm). Dire Dawa is located between $09^{\circ} 28.1$ N and $41^{\circ} 38.1$ E. The altitude of the station is 1116 m.a.s.l. and the mean annual temperature ranges from 19 to 31.5°C . The temperature is generally high with the monthly mean maximum temperature ranging from 28.1°C in December and to 34.6°C June. Dire Dawa enjoys bimodal type of rainfall with April as a peak for the scanty rainfall and July for the heavy

rains. The mean annual rain fall in the study area is 550 mm representing low-land (semi-arid) climate condition.

Sample preparation: Chaya leaves obtained from Dire Dawa research center were transported to the Department of Food Science and Postharvest Technology Laboratory at Haramaya University. The sorted leaves were thoroughly washed and carefully handled. Leaves were dried in an oven at 105°C for 2 h and milled to powder prior to further analysis. Some portion of fresh Chaya leaves was stored in cold place (5 °C) until further analysis.

Proximate composition: The proximate compositions such as moisture content, crude protein, crude fat, ash content, crude fiber content and total carbohydrate (by difference) of Chaya leaves were determined according to Association of Official Analytical Chemists methods (American Association of Cereal Chemists (AACC), 2000). Energy value (kcal/100g) was calculated as:

$$(\% \text{ protein} \times 4) + (\% \text{ fat} \times 9) + (\% \text{ carbohydrate} \times 4)$$

Minerals analysis: Ca, Fe, and Zn content were determined by atomic absorption spectrophotometer (American Association of Cereal Chemists (AACC), 2000).

Condensed tannin: Condensed tannin was analyzed using the modified Vanillin-HCl methanol method adopted from (Price *et al.*, 1987, Shi *et al.*, 2004) us. The Vanillin-HCl reagent was prepared by mixing equal volume of 8% concentrated HCl in methanol and 1% Vanillin in methanol. The solution of the reagent was mixed just prior to use. About 0.2 g of the powder sample was placed in small conical flask. Then, 10 ml of 1% concentrated HCl was added to methanol. The conical flask was capped and continuously shaken for 20 min and then centrifuged at 2500 rpm for 5 min. About 1 mL of the supernatant was pipetted into a test tube containing 5 mL of Vanillin-HCl reagent. Absorbance at 450 nm was read on

spectrophotometer after 20 min of incubation at 30°C. A blank sample was also analyzed and its absorbance was subtracted from sample absorbance. A standard calibration curve was also prepared from catechin (1 mg/ml). Tannins content was expressed as catechin equivalent as follows:

$$\text{Tannin (\%)} = \frac{C \times 10}{200} \times 100$$

where C = Concentration corresponding to the optical density; 10 = Volume of the extract (ml); 200 = Sample weight (mg).

Hydrogen cyanide content: Cyanogenic glycoside was determined using alkaline picrate method. Ground sample (5.0 g) was weighed and dissolved in 50 cm³ distilled water. The cyanide extraction was allowed to stay overnight and then filtered (Inuwa *et al.*, 2011).

Preparation of cyanide standard calibration curve: Different concentrations of potassium cyanide (KCN) solution ranging from 0.1 to 1.0 mg/mL cyanide were prepared. One milliliter of the sample filtrate and standard cyanide solution in test tubes and 4 ml of alkaline picrate solution was added and incubated in water bath for 15 min. After color development, the absorbance was read with spectrophotometer at 490 nm against blank containing only 1 ml distilled water and 4 cm³ alkaline picrate solution. The cyanide content was extrapolated from the cyanide standard curve.

Calculation

$$\text{Cyanogenic glycoside } \left(\frac{\text{mg}}{100\text{g}} \right) = \frac{C \text{ (mg)}}{\text{Sample weight (mg)}} * 10$$

Where C (mg) = Concentration of cyanide content read on the graph.

Total phenol content: Total phenolic content was determined according to Folin-Ciocalteu method (Upadhyay *et al.*, 2010). Two hundred milligrams of the sample was extracted with 4 ml of acidified methanol (HCl/Methanol/water,

1:80:10 V/V/V) at room temperature (25 °C) for 2 h. Aliquot of the extract (200 µl) was added to 1.5 ml freshly diluted (10 fold) Folin-Ciocalteu reagent. The mixture was allowed to equilibrate for 5 min and then mixed with 1.5 ml of sodium carbonate solution (60 g/l). After incubation at room temperature (25 °C) for 90 min, the absorbance of mixture was read at 725 nm (6505 uv/vis spectrophotometer, Model 6505, U.K, GENWAY). Acidified methanol was used as a blank. A series of six standard solutions (0 ppm, 5 ppm, 10 ppm, 15 ppm, 20 ppm, 25 ppm, 30 ppm, and 35 ppm) were prepared from the stock solution of standard Gallic acid. The amount of total phenol was estimated from the calibrated curve as Gallic acid equivalent (GAE) in milligram per kilogram of sample. Total phenolic content was calculated by the following formula:

$$\text{Total phenol} \left(\frac{\text{mg}}{100\text{g}} \right) = \frac{(\mu\text{g/mL}) \times Df \times 100}{\text{Sample mass (db)}}$$

where $\mu\text{g/mL}$ is the absorbance reading concentration, Df is dilution factor

Data analysis: Proximate composition analysis was done in triplicate for each component and descriptive statistic was used to report values as a mean and standard deviation (mean \pm SD). For data handling and processing, R-studio statistical package was used.

Results

The proximate compositions of Chaya leaf powder are presented in **Table 1**. The analysis showed that moisture content, protein, crude fat, crude fiber, ash, and carbohydrate were 5.63, 23.96, 8.98, 2.87, 9.05, and 49.49%, respectively. In addition, the energy content of the leaf was found 374.62 kcal/100g.

The main mineral contents of Chaya leaf are shown in **Table 2**. Mean values of each mineral such as Ca, Fe, and Zn were 253.68, 68.02, and 4.85 mg/100g, respectively. The phytochemical content of fresh Chaya leaf is presented in **Table 3**. The phytochemical analysis revealed that phenols, tannin, and hydrogen cyanide contents of the Chaya leaf were 1916.66, 176.53, and 102.00 mg/100g, respectively.

Table 1. Proximate compositions and energy of Chaya leave powder.

Proximate analysis	Concentration (%)
Moisture content	5.63 \pm 0.315 ^a
Protein	23.96 \pm 0.13
Crude fat	8.98 \pm 0.08
Crude fiber	2.87 \pm 0.10
Total ash	9.05 \pm 0.06
Carbohydrate	49.49 \pm 0.20
Energy(kcal/100g)	374.62 \pm 4.57

^a: Mean \pm SE, n=3.

Table 2. Mineral content of Chaya leaf.

Minerals	Concentration (mg/100g)
Ca	253.68 \pm 30.92 ^a
Fe	68.02 \pm 0.11
Zn	4.85 \pm 0.18

^a: Mean \pm SE, n=3.

Table 3. Phytochemical content of fresh Chaya leaf.

Phytochemical	Concentration (mg/100g)
Tanins	176.533 \pm 0.25
Total Phenol	1916.66 \pm 1.53
Hydrogen Cyanide	102.007 \pm 1.83

^a: Mean \pm SE, n=3.

Discussion

The moisture content of the leaf powder in this research was found to be 5.63 \pm 0.315%, which is far lower than the moisture content of dried Chaya leaf reported in different studies. The value ranges between 8.73 \pm 0.01 and 12.73 \pm 0.76% (Kuri-GarcÃa and GuzmÃ, 2017, Otitoju *et al.*, 2014), and in this study, it is marginally close to 6.88 \pm 0.82 and 7.64 \pm 0.3% of moringa leaf (Iyaka *et al.*, 2014). The lower moisture content in the leaf would reduce the growth of microorganisms and rate of chemical reaction, thereby increasing their storage life (Iyaka *et al.*, 2014).

The result of this study shows that Chaya leaf is a good source of protein (23.96%). This value is in line with the protein value (24.13%) reported by

Otitoju (Otitoju *et al.*, 2014) but the value obtained in this research was higher than 20.59% and 18.73% for the same plant (Adanlawo and Elekofehinti, 2012, Akindahunsi and Salawu, 2005). This value was also similar with *Moringa oleifera* leave grown in Ethiopia (24.31%) (Mikore and Mulugeta, 2017). Protein is an important component of human diet needed for the growth of children, adults, and infants as well as for constant replacement of worn out tissues (Obahigbon and Erhabor, 2010).

The crude fat (8.98%), recorded in this study, was significantly higher than 8.05% (Otitoju *et al.*, 2014) but lower than 6.73% in Chaya leaf (Adanlawo and Elekofehinti, 2012). This difference could be due to the variation in climate and agronomic practices during Chaya growing. Comparing this value with crude fat content of *Moringa oleifera* leaf grown in Ethiopia, Chaya leaf in this study had higher value than 6.73% and 7.35% (Melesse *et al.*, 2011). However, fat content of Chaya leaf obtained in this study was lower than fat content of kale (*Brassica oleracea*) which was 11.91% (Iyaka *et al.*, 2014). According to studies, food types with low fat content are reported to reduce the level of cholesterol and obesity and no lipid accumulation that cause arteriosclerosis and aging (Antia *et al.*, 2006). Crude fiber content (2.87%) obtained in this study was lower than 4.73% (Otitoju *et al.*, 2014) and 9.68% in Chaya leaf (Adanlawo and Elekofehinti, 2012). Epidemiological studies have shown that high dietary fiber intake helps to prevent or treat hyperlipidemia, cardiovascular disease, hypertension, obesity, certain cancers, gastrointestinal disorders, and diabetes (Ijarotimi *et al.*, 2013). Crude fiber content in the leaves was low but can still make significant contribution to dietary intakes. Since fiber lowers body cholesterol level, it could decrease the risk of cardiovascular diseases (Hanif *et al.*, 2006).

The ash content of $9.05 \pm 0.058\%$ of Chaya leaf powder obtained in this study was significantly higher than the result of the study by Otitoju *et al.* (8.10%) (Otitoju *et al.*, 2014). This value was lower than ash content values of 16.40 ± 0.5 and

11.17% in *Telfairia occidentalis* and kale, respectively (Iyaka *et al.*, 2014). The ash content of a leaf is an index of total mineral content indicating Chaya leaf is a good source of minerals.

Carbohydrate content of Chaya leaf was about 49.49% which was higher than values of 42.69% and 35.75% reported in other studies (Iyaka *et al.*, 2014). According to this study, Chaya leaf had moderately higher carbohydrate value than *Moringa olifera* leaf (40.00%) grown in Ethiopia. Carbohydrates are pivotal nutrients required for adequate diet (Hanif *et al.*, 2006). The higher carbohydrate and protein content of Chaya leaf makes it a good source of balanced diet.

The energy value of Chaya leaf was obtained 374.62 kcal/100g which was significantly higher than the energy value of other edible leaves such as *Brassica Oleracea* (319.80 kcal/100 g), *Moringa oleifera* (363.60 kcal/100 g), and *Telfairia occidentalis* (354.20 kcal/100 g) (Iyaka *et al.*, 2014).

Minerals analysis of Chaya leaf indicated that Ca, Fe, and Zn contents were 253.68 mg/100 g, 68.02 mg/100 g, and 4.85 mg/100 g, which were comparable with the values obtained by (Adanlawo and Elekofehinti, 2012)). Ca, which is present in a relatively in high concentration, is essential in the formation of bone and teeth, as a cofactor for enzymes and a component of ATP, DNA, RNA, and cell membranes, respectively. The minerals present relatively in low concentration of Fe and Zn perform various important functions in the body, like the formation of hemoglobin, growth, and sexual maturation, as cofactor for enzymes and so many other functions (Gafar *et al.*, 2011).

In this study, Chaya leaf has shown to have high content of total phenol (1916.66 mg/100 g) compared to the results reported in Nigera (Orji *et al.*, 2016). The presence of high amounts of phenol is a clear indication that the Chaya leaf can be exploited in pharmaceuticals for the treatment of many diseases. Phenol makes the plant a potential cancer therapy, since phenols are well known for the enormous ability to combat cancer (Adeniran *et al.*, 2013). The blood glucose lowering potential of Chaya plant extract was justified by the strong

presence of flavonoids in its phytochemical analysis (Chen *et al.*, 2018). The tannin content of Chaya leaf in this study was 176.53 mg/100 g. There is experimental evidence supporting the free radical scavenging capacity of phytochemicals including tannin (Mordi and Akanji, 2012, Rodrigues *et al.*, 2005).

The level of hydrogen cyanide was 102.00 mg/100 g. Cyanide levels decrease during cooking to below the allowable levels established for dry beans, peas, and nuts (0.025 mg HCN/g), leaving no residual HCN in the cooking water.

Conclusions

Consuming adequate amounts of Chaya leaf could help a lot to meet humans' nutritional needs for normal body growth, infants, and protection against diseases caused by malnutrition. Chaya leaf would provide a substantial advantage providing nutrients including health-promoting phytochemicals such as phenolic. It is also rich in protein (23.96 ± 0.13) and minerals such as Ca (253.68 mg/100 g) Fe (68.02 ± 0.11), and Zn (4.857 ± 0.18). If properly harnessed, Chaya plant will present a nutritional rich food source that is cheap, available throughout the year, and drought tolerant. It can help communities like Ethiopia to tackle problems of malnutrition. In general, it will enhance food and nutrition security of community suffering from malnutrition in drought prone areas.

Acknowledgement

Thanks are owed to Haramaya University and Research Directorate Office. The authors also thank the editorial board and an anonymous reviewer for their assistance.

Conflicts of interest

The authors declare that there is no conflict of interest.

Authors' contribution

All authors contributed to data gathering, laboratory analyzing, writing the manuscript, and on final revision of the manuscript.

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