Phytochemical, Anti-nutrients and Toxicity Evaluation of Cleome gynandra and Solanum nigrum: Common Indigenous Vegetables in Zimbabwe

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Authors’ contributions

This work was carried out in collaboration between all authors. Author CZ designed the study and in collaboration with author LM wrote the protocols and supervised the work. Author CS performed the experiments and prepared the manuscript. Authors CZ and LM edited the manuscript. All authors read and approved the final manuscript.

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ABSTRACT

Aim: This study assesses the phytochemical profiles, toxicity and anti-nutrient properties of two indigenous vegetables, Cleome gynandra and Solanum nigrum.

Methodology: Aerial parts of S. nigrum and C. gynandra collected from wild populations were extracted in water and methanol solvents using the cold maceration procedure. Chemical tests to identify different phytochemicals and antinutrients were carried out using specified reagents. Toxicity of the plant extracts was tested using the Brine Shrimp (Artemia salina) lethality bioassay procedure.

Results: Phytochemical tests indicated presence of alkaloids, steroids, flavonoids, cardiac glycosides, saponins, phenols and tannins in both S. nigrum and C. gynandra. Anthraquinones were only found in C. gynandra, whereas terpenoids and coumarin glycosides were found only in...
S. nigrum. The anti-nutrient levels in the vegetables were high; tannins (3.580 and 1.820 mg/100 g), oxalates (21.560 and 24.97±0.156 g/100 g), saponins (71 and 42%), alkaloids (15.160 and 3.44%) and phytates (0.099±0.046 and 0.250±0.013%) in S. nigrum and C. gynandra respectively. The Brine Shrimp LC50 values obtained were 0.67 and 1.84 mg/ml for methanol and water extracts of S. nigrum and 1.09 and 4.9 mg/ml for C. gynandra, respectively.

**Conclusion:** The vegetables, C. gynandra and S. nigrum are rich sources of diverse phytochemicals. However, some phytochemicals are present in high levels and might act as anti-nutritional factors interfering with absorption of food. C. gynandra is safe for consumption but S. nigrum contains some toxic phytochemicals and should not be taken regularly.

**Keywords:** Solanum nigrum; Cleome gynandra; phytochemicals; anti-nutrients; toxicity.

1. **INTRODUCTION**

Indigenous leafy vegetables (ILVs) are an essential part of the diet of many Zimbabweans. ILVs refers to those plants whose leaves or aerial parts, which may include young succulent stems, flowers and young fruit, have been integrated in a community’s culture for use as food over a long time [1]. They contribute significantly to food security especially during times of drought or poor harvest [2] and are highly recommended for their health and nutritional benefits [3]. ILVs are known to have high nutritional content [3] and some have medicinal properties [1]. Some of the nutrients supplied include proteins, minerals, vitamins and fibres [4]. Leaves and young tender shoots collected from wild populations are sold in rural and urban markets. The vegetables therefore provide a source of regular income, especially for the poor and the unemployed as a basic strategy for fighting against poverty, hunger, malnutrition and under nourishment [5].

*Cleome gynandra*, *Solanum nigrum*, *Cucurbita maxima* and *Cucumis anguria* are some of the common leafy vegetables consumed by both rural and urban dwellers. The diversity of ILVs consumed varies significantly from one community to another [6]. *S. nigrum* is mainly consumed in rural areas whereas *C. gynandra* is consumed throughout the country and is now sold in urban markets.

Studies elsewhere have established that some indigenous vegetable species are potentially toxic to humans and animals. Toxicity is mainly due to secondary compounds known as phytochemicals, produced to counter predation or to gain advantage over other competing plants [7]. Phytochemicals are biologically active substances which fall into various groups which include alkaloids, oxalates, tannins, phytates, saponins, essential oils, coumarin and cardiac glycosides, anthraquinones, terpenoids and flavonoids [8]. Plant poisons may cause acute effects when ingested in high concentrations and chronic effects when accumulated [9,10]. Under stressful conditions, due to food shortages, consumption of large amounts of vegetable toxins can have negative consequences [11].

Apart from being toxic, ILVs are also known to contain anti-nutrients and plants contain a wide variety of these in their raw state. Anti-nutrients or anti-nutritional factors are compounds or substances which act to reduce nutrient intake, digestion, absorption and utilization or produce other adverse effects. These anti-nutrients are potentially harmful in that they reduce growth and healing by affecting the availability of nutrients to the body especially if present in high levels [12,13]. The major anti-nutrients include toxic amino acids, saponins, cyanogenic glycosides, tannins, phytic acid, oxalates, lectins, protease inhibitors, chlorogenic acid and amylase inhibitors. Anti-nutritional factors such as alkaloids, phytates, tannins, saponins, oxalates, have an adverse effect on health through inhibition of protein digestion, growth, iron and zinc absorption [14,15].

*S. nigrum* commonly known in Zimbabwe as Musungusungu in Shona, Umsobo in Ndebele and Black nightshade in English is an annual herbaceous dicot weed in the Solanaceae family. The leaves are alternate and variable in shape from ovate-lanceolate to elliptic. Flowers are white in drooping inflorescences. Fruits are spherical black berries. It inhabits waste land, old fields, ditches and roadside [16]. Leaves of *S. nigrum* are collected from the wild and eaten as a vegetable relish after boiling [17]. *S. nigrum* has a bitter taste and is only preferred when most vegetables are not available. Stomach-aches and other side effects have been reported in cases where *S. nigrum* is eaten regularly [17].

*C. gynandra* locally known as Nyevhe in Shona, Elude in Ndebele and Cat’s whiskers in English
is an erect herbaceous annual with branched stems in the family Cleomaceae. Leaves are alternate, digitately palmate and petiolate. Inflorescences are showy, many flowered, terminal racemes. The fruits are long-stalked, dry, dehiscent siliques. The plants inhabit wasteland and arable land [18]. The tender young leaves and shoots are eaten boiled as vegetables. The leaves are also rather bitter and are often cooked with other vegetables like *Amaranthus* and *Vigna* spp [16]. The leaves and shoots are commonly preserved by drying and eaten during the dry season [19]. This plant is sold fresh in rural and urban markets during the rainy season but is now available throughout the year in dried form in some supermarkets. *C. gynandra* is becoming increasing popular in most urban markets with demand frequently surpassing supply. Small scale efforts to cultivate the vegetable are underway in a number of countries [19].

Plants consumed as vegetables should be free of toxicity or other adverse effects. *C. gynandra* and *S. nigrum* are consumed in large quantities in the rainy in Zimbabwe particularly in the rural areas. Both plants have a somewhat unpalatable bitter taste [20] leaving one to wonder whether its safe to consume these vegetables. Studies elsewhere have shown these plants or plants related to them to have a number of phytochemicals like alkaloids, tannins and saponins some of which could act as anti nutritional factors. These studies need to be extended into our region as phytochemical composition of plants is known to change with climate, soils, processing methods and genetic origin [21]. The aim of this study is to screen for phytochemicals and assess the toxicity and anti-nutrient levels of *C. gynandra* and *S. nigrum*.

2. MATERIALS AND METHODS

2.1 Plant Preparation and Extraction

*Cleome gynandra* was collected from Seke communal lands while *Solanum nigrum* was collected from Marlborough in Zimbabwe. Both plants were in flowering and fruiting stages. Plant samples were identified by a botanist in the Department of Biological Sciences, University of Zimbabwe. Voucher specimens are deposited in the University herbarium. Both fresh and dried samples were used in the experiments. Samples were dried in an oven at 45°C for 48 hrs. For extraction the cold maceration method [22] was used with two solvent systems, water and analytical grade methanol for 48 hrs at room temperature. The ground material was extracted with the solvents using the sample to solvent ratio of 1:10 (w/v). The extracts were filtered in a Buchner vacuum filter using Whatman No.1 filter paper and stored in a fridge at 4°C. Extracts were concentrated by evaporating using a rotary evaporator at 70°C under reduced pressure.

### 2.2 Phytochemical Screening of *Solanum nigrum* and *Cleome gynandra*

Concentrated crude aqueous extracts were re-dissolved using distilled water while the concentrated methanol extracts were re-dissolved using methanol to the required concentrations for bioassay analyses. The following standard tests were carried out following the procedures of [23,24].

2.3 Determination of Steroid (Liebermann Burchard Reaction)

3 ml of the extract was added with 1 ml of chloroform and a few drops of concentrated Sulphuric acid along the sides of the test tubes. Formation of a reddish brown precipitate at the bottom of the test tubes indicated the presence of steroids.

2.4 Determination of Cardiac Glycoside (Keller Kiliani Test)

3 ml of the extract was added with 1 ml of acetic acid followed by the addition of 3 ml of 10% ferric chloride and a few drops of concentrated sulphuric acid on the sides of the test tubes. A brownish ring and green blue precipitate at the bottom of the test tube confirmed the presence of cardiac glycosides.

2.5 Determination of Phenol and Tannin (Ferric Chloride Test)

Few drops of 10% ferric chloride were added with 3 ml of extract. A blue or green precipitate indicated the presence of phenols and tannins.

2.6 Determination of Terpenoids (Salkowski Test)

To 3 ml of the extract, 1 ml of chloroform and a few drops of concentrated sulphuric acid was carefully added along the sides of the test tubes. A reddish brown color precipitate indicated the presence of terpenoids.
2.7 Determination of Alkaloids (Mayer’s Test)

3 ml of Mayer’s reagent was added with 300 µl of fruit extract. A pale precipitate indicated the presence of alkaloids.

2.8 Determination of Flavanoids

1 ml of 10% ammonia and 1 ml of concentrated sulphuric acid was added to 3 ml of extract. Disappearance of yellow color indicated the presence of flavonoids.

2.9 Determination of Anthraquinones (Borntrager’s Test)

1 ml of benzene and 1 ml of 10% ammonia was added to 3 ml of extract. The presence of anthraquinones was observed by the formation of pink, red or violet color below the ammonia.

2.10 Determination of Saponins (Froth Test)

2 ml of distilled water was added to 3 ml of extract in a test tube. The solution was vigorously shaken and some drops of olive oil were added. The formation of a stable foam was taken as an indication for the presence of saponins.

2.11 Determination of Coumarin Glycosides (FeCl³ test)

To 3 ml of the concentrated alcoholic extract of a drug few drops of alcoholic FeCl³ solution was added. Formation of a deep green colour, which turned yellow on addition of conc. HNO³, indicated the presence of coumarin glycosides.

2.12 Determination of Anti-nutritional Factors

2.12.1 Oxalates determination

The titration method [25] was followed in determining the oxalate content of the extracts. The details of the method are as follows: 2 g each of *S. nigrum* and *C. gynandra* extract were suspended in 190 ml of distilled water in a flask. 10 ml 6M HCl solution was added to each samples and digested at 100°C for 1 hr. The samples were allowed to cool and the volume adjusted to 250 ml with distilled water. Each sample was then filtered with number 1 Whatman filter paper and divided into two portions of 125 ml each. Thereafter four drops of methyl red indicator were added to each sample, followed by the drop wise addition of concentrated NH₄OH solution until the solution changed from pink to a yellow colour. The samples were then heated to 90°C, cooled and filtered. The filtrates were heated to 90°C and 10 ml of 5% CaCl₂ solution was added to each while stirring continuously. The following morning, the cooled samples were centrifuged at 2500 rpm for 5 min, the supernatant then decanted and the precipitates dissolved in 10 ml 20% H₂SO₄. The filtrates for each of the plants were composited and adjusted to a volume of 200 ml. 125 ml of each of the filtrates was heated to near boiling and then titrated against 0.05 M standardized KMnO₄ solution until a faint pint colour persisted for 30 sec. The oxalate content was calculated as follows:

\[
1 \text{ ml } 0.05 \text{ M KMnO}_4 = 2.2 \text{ mg oxalate}
\]

2.12.2 Phytates determination

Phytates were determined through phytic acid determination using the procedure described by [26]. 2 g of each sample were weighed into a 250 ml conical flask. Samples were soaked in 100 ml of 2% conc HCl for 3 hrs and then filtered through a double layer Whatman number 1 filter paper. 50 ml of each of the sample filtrate were placed in a 250 ml beaker and 100 ml of distilled water was added. To each sample, 10 ml of 0.3% Ammonium thiocyanate indicator solution was added. Titrated then followed with standard iron chloride solution which contained 0.00195 g iron/ml. The end point was signified by the appearance of a brownish-yellow coloration that persisted for 5 min. The percentage phytic acid was calculated as follows:

\[
\% \text{ Phytic acid} = y \times 1.19 \times 100 \text{ where, } y = \text{titre value} \times 0.00195 \text{ g}
\]

2.12.3 Saponins determination

Saponin was determined using the method of [26]. 10 g each of the ground samples of *S. nigrum* and *C. gynandra* were extracted in 20 ml of 20% with constant agitation on a shaker for 12 h at 55°C. The resultant solution was filtered and the residue re-extracted in 200 ml 20% ethanol. The solvent was evaporated under vacuum to a volume of 40 ml. Thereafter 20 ml of diethyl ether were added in a separating funnel and the mixture shaken vigorously. The aqueous layer was recovered and its pH was adjusted to
4.5 by adding NaOH. The solution was then shaken with 60 ml n-butanol, washed twice with 10 ml of 5% aqueous NaCl and evaporated to dryness in a fume cupboard to give a crude saponin which was weighed. The saponin content was calculated as follows:

\[ \% \text{ Saponin} = \left( \frac{W_{1}}{W} \right) \times 100 \]

Where

- \( W \) = Weight of sample used
- \( W_{1} \) = Weight of saponin extract

2.12.4 Alkaloids determination

Alkaloids were quantitatively estimated using the method by [23]. 5 grams of the powdered sample was extracted in 200 ml of 10% acetic acid in ethanol in a 250 ml beaker. Samples were incubated for 4 h at room temperature, then filtered and the filtrate concentrated on a water bath to one-quarter of the original volume. The extract was then precipitated by the addition of drops of concentrated ammonium hydroxide and allowed to settle. The precipitate was washed with dilute ammonium hydroxide and then filtered. The residue which comprised of the alkaloid, was dried and weighed. The alkaloid content was determined using the formula:

\[ \text{alkaloid (\%)} = \frac{\text{final weight of sample}}{\text{initial weight of extract}} \times 100. \]

2.12.5 Tannin determination

2.12.5.1 Preparation of the standard solution

Tannins were determined following the procedure of [27]. Tannic acid (100 g) was dissolved in 100 ml distilled water to prepare the standard tannic acid solution. The working solution was prepared by diluting 5 ml stock solution to 100 ml with distilled water. Each ml contained 50 mg of tannic acid

2.12.5.2 Preparation of the working solution

10 ml of the standard solution was made up to 100 ml distilled water. 1 -10 ml aliquots were put in clear test tubes. 4 ml of 0.1 M ferric chloride was added to each tube. Each tube was made up to 10 ml with distilled water. All the reagents in each tube were mixed well and kept undisturbed for about 10 minutes and absorbance read on spectrophotometer at 395 nm against a blank. The standard curve was obtained by plotting concentration versus absorbance.

2.12.5.3 Extraction of tannin

500 mg of the plant sample was weighed and transferred to a 50 ml flask. Then 50 ml of distilled water was added and the mixture was stirred for 1 hr. The sample was filtered into a 50 ml volumetric flask and made up to the mark with same distilled water. 10 ml of the filtered sample was pipette out into test tube and then mixed with 4 ml of 0.1 M ferric chloride. The absorbance was measured using a spectrophotometer at 395 nm within 10 min. The tannin content was calculated from the tannic acid standard curve. The calculation of the tannin content was done following (27) as follows:

\[ C = \left( \frac{C_{1} \times V}{N} \right) \]

Where

- \( C \) = total content of the tannin in mg/g.
- \( C_{1} \) = the concentration of tannic acid established from calibration curve in mg/ml.
- \( V \) = volume of the extract in ml.
- \( N \) = the weight of the plant extract in mg.

2.13 Brine Shrimp Lethality Assay

2.13.1 Sample preparation

Different concentrations of analytical methanol and aqueous extracts (1; 0.5; 0.25; 0.125 and 0.0625 mg/ml) were prepared. Solution A was prepared by dissolving 0.1 g of each extract in 10 ml of 1% DMSO (solution A). Solution B was prepared by diluting 5 ml of solution A to 10 ml with 1% DMSO. Solutions C, D and E were prepared using a serial dilution factor of 2. This was done for both sample extracts and potassium dichromate which was used as the reference toxicant (standard). The negative control was prepared using only artificial sea water [28]

2.13.2 Hatching the shrimp

About 1 g of Artemia salina were hatched using brine shrimps cysts in a glass vessel (2500 ml), filled with sterile artificial sea water (Prepared using sea salt (20 g/l) and adjusted to pH 8.2 using 0.05M NaOH) under constant aeration for 24 hrs. After 24 hrs of incubation at room temperature (25-29°C), under continuous illumination of fluorescence lamp newly hatching, active nauplii free from egg shells were harvested using a glass capillary from the
brighter portion of the hatching chamber and used for the assay [29].

2.13.3 Bioassay

The assay system was prepared with 5 ml of filtered seawater containing 50 µl of the chosen extract concentration and 10 newly hatched brine shrimps in a glass vial. The vials were then incubated at room temperature for 24 hrs. Each concentration was replicated three times. The setup was allowed to stand for 24 hrs, after which the number of living shrimps were counted under a hand magnifying lens. The percentage death at each dose and control were determined. In cases where control deaths occurred, the data was corrected using the following formula by [24]: % death = [(test-control)/100-control] x 100. After 24 hrs the LC50 values were determined, based on the percentage mortality, using probit analysis in Excel-XL STATS 2015.

2.14 Data Analysis

Means and standard deviations were calculated using Microsoft Excel 2010. Toxicity data were analysed using probit analysis as described by Finney in Excel-XL STATS 2015. Percentage (%) mortalities were transformed to probits and plotted against log concentration. Trend lines were fitted and LC50 were calculated from linear regression equations and 95% confidence intervals were determined.

3. RESULTS AND DISCUSSION

3.1 Qualitative Phytochemical Screening

The vegetables S. nigrum and C. gynandra contain a wide variety of secondary metabolites including alkaloids, steroids, flavonoids, cardiac glycosides, saponins, phenols and tannins (Table 1). Similar studies from Kenya [30] and Mexico [31] observed similar phytochemicals. Alkaloids, tannins, flavonoids, and phenolic compounds are known to be the most biologically active phytochemicals [32] and the observation of all these in S. nigrum and C. gynandra shows that the vegetables are rich sources of phytochemicals. In humans phytochemicals are reported to promote good health and prevent disease development [33]. This is due to their antimicrobial, antioxidant, anti-inflammatory, antiviral, immune system stimulant and detoxification activities [33].

3.2 Quantitative Test of Anti-nutritional Factors

S. nigrum and C. gynandra contain high levels of anti-nutritional factors (Table 2). Alkaloids, saponins and tannins were higher in Solanum nigrum whereas oxalates and phytates were higher in C. gynandra. Antinutrients are reported to reduce the bioavailability of proteins and essential nutrients and also to cause toxicity [34]. Saponins were found in the two vegetables and their characteristic bitter taste is most likely due to presence of saponin. The consumption of raw and undercooked vegetables therefore exposes the individual to high levels of saponins which are known to affect the digestive processes by disrupting the epithelial function, creating pores in the cell surface, producing increased permeability (i.e. leaky gut) and allowing substances to enter the bloodstream [34].

High concentrations of alkaloids have been observed in the two plants but more so in S. nigrum. The alkaloid content in S. nigrum is due to the alkaloid solanine, a neurotoxic glycoalkaloid found in species of the family Solanaceae [35]. Solanine is produced as a natural defense agent by plants, and is known to possess antimicrobial, pesticidal, nematicidal and molluscicidal properties [7]. Solanine is not destroyed by normal cooking temperatures as it only decomposes at 243°C [36]. However it is water soluble and boiling for long periods will reduce the solanine levels [36]. S. nigrum is therefore not a recommended for regular diet. Glycoalkaloids cause toxicity by disrupting cell membranes causing gastrointestinal disturbances like vomiting, diarrhoea and abdominal pain [15]. Alkaloids also interfere with the body's ability to regulate the enzyme acetylcholine, responsible for conducting nerve impulses, leading to symptoms such as sweating, vomiting, diarrhoea and bronchospasm [37].

The tannin content of the vegetables was also high (Table 2), implying that the leaves have astringent properties. The presence of even a low amount of tannin is not desirable from a nutritional point of view. Tannins are water soluble phenolic compounds that chelate Fe and Zn limiting their absorption and they also interfere with digestion by exhibiting anti-trypsin and anti-amylase activity. They are not easily or completely destroyed by heat due to their high molecular weight but are preferred due to their anticancer activities [38].
Oxalates and phytates reduce bioavailability of metallic ions such as Mg, Zn, Fe and Cd by chelating with them to form poorly soluble compounds not readily absorbed from the gastrointestinal tract [39]. Oxalates also combine with calcium to form calcium oxalate crystals which are deposited as urinary stones that are associated with the renal tubule blockages [40].

The Brine shrimp lethality assay is one of the tests that is useful for toxicity screening of plant extracts, insecticides, heavy metals, food additives and pharmaceutical compounds because it is rapid and also inexpensive [29]. The methanol extracts of both vegetables had lower LC$_{50}$ values than the aqueous extracts (Table 3). In assessing the toxicity of plant extracts [29] considered any extracts with LC$_{50}$ values less than 1 mg/ml to be toxic. Using this benchmark, only one extract the S. nigrum (methanol) with an LC$_{50}$ of 0.67 is toxic. A comparable LC$_{50}$ value of 0.63 mg/ml was obtained in a similar study on S. nigrum in Bangladesh [41]. C. gynandra is considered safe for consumption according to this assay.

Figs. 1 and 2 show probit analyses for the brine shrimp mortality data. There was an increase in mortality with increasing extract concentrations. The lethality rates (probits) were strongly and positively correlated to the extract concentrations, as shown by the correlation coefficient (R$^2$) values which ranged from 0.858-0.981. There was a difference in both the aqueous and methanol extracts of both S. nigrum and C. gynandra compared to the control. The respective regression lines gave LC$_{50}$ values ranging from 0.308-4.887 mg/ml, the highest LC$_{50}$ was the methanol extract of S. nigrum (0.673 mg/ml) and the lowest was the aqueous extract of C. gynandra (4.887 mg/ml) as also shown in Table 3.

Toxicity is attributed to phytochemical compounds like saponins and alkaloids [42,43]. The high toxicity of S. nigrum may be due to the presence of higher levels of alkaloids and saponins (Table 2) than in C. gynandra and also because of the presence of terpenoids and coumarin glycosides which were absent in C. gynandra. S. nigrum is reported to be highly toxic due to the presence of solanine and its concentration increases in leaves as the plant matures [7]. The LC$_{50}$ values of S. nigrum and C. gynandra were higher than the LC$_{50}$ value of the positive control (Table 3) meaning that the extracts were not as toxic as the control.
Table 1. Phytochemical screening results of the aqueous and methanol extracts of *Solanum nigrum* and *Cleome gynandra*

| Phytochemical          | Treatment |  |  |  |  |
|------------------------|-----------|---|---|---|---|
|                        | Aqueous   |  |  |  |  |
|                        | *C. gynandra* | S. nigrum |  |  |  |
| Alkaloids              | +         | +  | ++ | ++ | ++ |
| Steroids               | ++        | _  | +  | ++ | ++ |
| Flavonoids             | _         | +  | ++ | ++ | ++ |
| Terpenoids             | _         | +  | _  | +  | ++ |
| Anthraquinones         | +         | _  | ++ | _  | _  |
| Saponins               | ++        | ++ | +  | _  | _  |
| Phenols and tannins    | +         | ++ | ++ | ++ | ++ |
| Coumarin glycosides    | _         | _  | _  | _  | _  |

* - absent, + present, ++ abundantly present

Table 2. Anti-nutritional factors present in *S. nigrum* and *C. gynandra* methanol extracts

| Samples         | Tannins mg/100 g | Alkaloids % | Oxalates g/100 g | Phytates % | Saponins % |
|-----------------|------------------|-------------|------------------|------------|------------|
| *Solanum nigrum* | 3.580            | 15.160      | 21.560           | 0.099±0.046| 7.1        |
| *Cleome gynandra*| 1.820            | 3.464       | 24.971±0.156     | 0.250±0.015| 4.2        |

Table 3. The effects of *S. nigrum* and *C. gynandra* extracts on Brine shrimp nauplii (n=3 for each concentration level)

| Samples         | Extracts     | Mortalities in probits after 24 hrs | LC<sub>50</sub> mg/ml | 95% confidence interval (Limits) |
|-----------------|--------------|-------------------------------------|------------------------|----------------------------------|
|                 |              | 1 mg/ml | 0.5 mg/ml | 0.25 mg/ml | 0.125 mg/ml | 0.0625 mg/l | Lower | Upper       |
| *C. gynandra*   | Methanol     | 4.77     | 4.61      | 4.42       | 3.25       | 3.25        | 1.09±1.12 | -0.023 | 3.09       |
| *C. gynandra*   | Water        | 4.42     | 4.50      | 4.19       | 3.92       | 3.92        | 4.9±35.02 | 30.12  | 39.9       |
| *S. nigrum*     | Methanol     | 5.23     | 4.77      | 4.61       | 4.16       | 3.92        | 0.67±1.24 | -0.566 | 1.914      |
| *S. nigrum*     | Water        | 4.69     | 4.69      | 4.42       | 4.05       | 3.87        | 1.84±8.31 | 6.473  | 10.147     |
| Potassium dichromate | _  | _      | 5.50      | 4.59       | 4.42       | 3.72        | 0.31±0.22 | 0.088  | 0.528      |
Fig. 2. Toxicity effect of potassium dichromate and the methanol extracts of *S. nigrum* and *C. gynandra* on Brine shrimps nauplii

*C. gynandra* with LC$_{50}$ higher than 1 mg/ml is safe for consumption when properly cooked and *S. nigrum* is toxic since its LC$_{50}$ value is less than 1 mg/ml. The Solanum species studied by [31] were much more toxic (LC$_{50}$ <1 ug/ml. Major differences with results presented here could be due to the fact that [31] used a different solvent ethyl acetate to the methanol used in this study. Differences in phytochemical composition and concentrations could also be due to possible effects of genetic origin, climate, soil or processing methods [36].

4. CONCLUSION AND RECOMMENDATION

*C. gynandra* and *S. nigrum* contain varying amounts of phytochemicals and anti-nutrients such as alkaloids, saponins, oxalates, tannins and phytates. *C. gynandra* is safe for consumption, but *S. nigrum* contains high levels of toxic phytochemicals and should not be a part of the regular diet. Some of these phytochemicals may interfere with food absorption in the body but this aspect needs further study as the present study has not addressed this. These two vegetables, however, need to be properly cooked to reduce the amounts of phytochemicals. An evaluation of the best cooking method also needs to be carried out.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Ogoye-Ndegwa C, Aagaard-Hansen J. Traditional gathering of wild vegetables among the Luo of western Kenya-a nutritional anthropology project. Journal of Ecology, Food and Nutrition. 2003;8:69-89.

2. Chweya JA, Mnzava NA. Cat’s whiskers. *Cleome gynandra* L. Promoting the conservation and use of underutilized and neglected crops. 11. International Plant Genetic Resources Institute. Rome. Italy. 2011:15-54.

3. Otoito O, Akpanabiatu MI, Otoito GTO, Ndem JI, Uwah AF, Akpanyung EO, Ekanem JT. Heavy metal contamination of green leafy vegetable gardens in Itam Road construction site in Uyo, Nigeria. Research Journal Environmental and Earth Sciences. 2012;4:371-375.
4. Mohammed MI, Sharif N. Mineral composition of some leafy vegetables consumed in Kano. Nigeria Journal of Basic and Applied Science. 2011;19:208-213.

5. Nkafamiya II, Osimeahan SA, Modibbo UU, Aminu A. Nutritional status of non-conventional leafy vegetables, *Ficus asperifolia* and *Ficus sycomorus*. African Journal of Food Science. 2010;4(3):104-108.

6. Mavengahama S. The contribution of indigenous vegetables to food security and nutrition within selected sites in S. A. Stellenbosch University. 2013;20-27.

7. Atanu FO, Ebiloma UG, Ajayi EL. A review of the pharmacological aspects of *Solanum nigrum* Linn. Biotechnology and Molecular Biology Review. 2011;6(1):1-7.

8. Pai V, Chanu TR, Chakraborty R, Raju B, Lobo R, Ballal M. Preliminary phytochemical screening in *Cleome gynandra* L. Asian Journal of Plant Science Researches. 2011;1(2):57-62.

9. Kofi-Tsekpo WM. Pharmaceutical Applications of Ethnobotany: Conservation and utilization of indigenous medicinal plants and wild relatives of food crops. United Nations Educational. Scientific and Cultural Organisation (UNESCO). Nairobi. 1997:57-63.

10. Pfander FA. Colour atlas of poisonous plants: A handbook for pharmacists, Doctors, Toxicologists and Biologists. Wolfe Publishing Limited, London. 1984;10-222.

11. Johns T, Kokwaro JO. Food plants of the Luo of Siaya District, Kenya. Economic Botany. 1991;45:103-113.

12. Abara AE. Tannin Content of *Dioscorea bulbofera*. Journal of the Chemical Society. 2003;28:55-56.

13. Agbaire PO, Emoyan OO. Nutritional and antinutritional levels of some local vegetables from Delta State, Nigeria. African Journal of Food Science. 2012;6:8-11.

14. Liener IE, Kcade ML. Protease inhibitor. In Liener I.E. (edition). Toxic constituents of plants foodstuffs. Academic Press, New York. 1980:43-92.

15. Larsson M, Rossande- Hulthen L, Sandstrom B, Sandberg A. Improved iron and zinc absorption from breakfast meals containing malted oats with reduced phytate content. British Journal of Nutrition. 1996;76:677-688.

16. Heever E, Venter SL. Nutritional and medicinal properties of *Cleome gynandra*. Acta Hortic. 2007;752:127-130

17. Jansen PCM. *Solanum nigrum* L. In: Schmelzer GH, Gurib-Fakim A (Ed.). PROTA (Plant Resources of Tropical Africa, Wageningen, Netherlands. 2008; 528-530.

18. Mishra SS, Moharan SK, Dash MR. Review on *Cleome gynandra*. International Journal of Research Pharmaceutical Chemistry. 2011;1(3):2231-2781.

19. Mnzava NA, Chigumira NF. *Cleome gynandra* L. In; Grubben GJH & Denton OA (Ed.). PROTA (Plant Resources of Tropical Africa, Wageningen, Netherlands; 2004.

20. Machakaire V, Turner AD, Chivinge, OA. Agronomic and nutrition studies of two indigenous vegetables in Zimbabwe: *Cleome gynandra* (shona=nyeve, ndebele=ulude) and *Corchorus tridens* (shona=derere, ndebele=idelele). Acta Horticulturae. 1998;513:145-152.

21. Soetan KO, Oyewole OE. The need for adequate processing to reduce the antinutritional factors in plants used as human foods and animal feeds: A review. African Journal of Food Science. 2009;3(9):223-232.

22. Tiwari P, Kumar B, Kaur M, Kaur G, Kaur H. Phytochemical screening and extraction: A review. Internationale Pharmaceutica Scienza. 2011;1:90-104.

23. Harborne JB. Phytochemical methods: A guide to modern technique of plant analysis. 3rd edn. Springer (India) Private Limited. New Delhi. 2005;40-137.

24. Sofowora A. Medicinal plants and traditional medicine in Africa. John Wiley and Sons Ltd. Nigeria. 1982;33-34.

25. Inuwa HM, Aina VO, Gabi B, Aimola I, Toyin A. Comparative determination of antinutritional factors in groundnut oil and palm oil. Advanced Journal of Food Science and Technology. 2011;3(4):275-279.

26. Akaneme Fl, Igata D, Okafor H, Anyabenechi O. Breeding for nutritional quality for *Corchorus olitorius*, *Annona muricata* and *Pantacanthera macrophylla* 1:a study of their nutritional contents. African Journal of Agricultural Research. 2014;9(14):1107-1112.

27. Omoruyi BE, Bradley G, Afolayan AJ. Antioxidant and phytochemical properties of *Carpobrotus edulis* (L.) bolus leaf used
for the management of common infections in HIV/AIDS patients in Eastern Cape Province. BMC Complementary and Alternative Medicine. 2012;12:215-241.

28. Latha LY, Sasidharan S, Zuraini Z, Suryani S, Shirley L, Sangetha S, Davaselvi M. Antimicrobial activities and toxicity of crude extract of the Psophocarpus tetragonolobus pods. Afr. J. Trad. CAM. 2007;4(1):59–63.

29. Meyer BN, Ferrigni NR, Putnam JE, Jacobsen LB, Nicholas DE, Mclaughlin JL. Brine shrimp: A convenient general bioassay for active plant constituents. Journal of Medical Plant Research. 1982;45:31–34.

30. Mibei EK, Ojijo NKO, Karanja SM. Kinyua JK. Phytochemical and antioxidant analysis of methanolic extracts of four African indigenous leaf vegetables. Annual Review of Food Science and Technology. 2012;13(42):37-41.

31. Peralta S., Aarland RC, Campos MG, Jimenez ME, José Alberto ME. Toxicity analysis, phytochemical and pharmacological study of the plant known as Mora Herb, collected at the Environmental Education Centre of Yautlica (CEA-Yautlica). Asian Journal of Plant Sciences. 2013;12(4):159-164.

32. Anderson GD. Are tannins a double edge sword in biology and health? Trends in Food Science Technology. 2004;4:168-175.

33. Chukwuebuka E, Chinenyе JL. Biological functions and anti-nutritional effects of phytochemicals in living system. Journal of Pharmacy and Biological Sciences. 2015;10(2):10-19.

34. Johnson IT, Gee JM, Price K, Curl C, Fenwick GR. Influence of saponins on gut permeability and active nutrients transport in vitro. Journal of Nutrition. 1986;11:2270-7.

35. Lin HM, Tseng HC, Wong CJ. Induction of autophagy and apoptosis by the extract of Solanum nigrum Linn in HepG2 cells. Journal of Agriculture and Food Chemistry, 2007;55:3620-3628.

36. FAO 1990 Roots, tubers, plantains and bananas in human nutrition. Agriculture and Consumer protection.

37. Cantwell M. A review of important facts about potato glycoalkaloids. Perishables Handling Newsletter. 1996;87:26-27.

38. Price KR, John IT, Frederick GR. The chemical and biological significant of saponins in foods and feeding stuffs. Food Science and Nutrition. 1987;26:27-90.

39. Plaami S. Myoinositol phosphates: Analysis, content in foods and effect in nutrition. Lebensmittel-wissenschaff and Technology. 1997;3(7):633-647.

40. Lorenz EC, Michet CJ, Milliner DS, Lieske JC. Update on oxalate crystal disease. Curr Rheumatol Rep. 2013;15(7):340-348.

41. Karmakar UK, Tarafder UK, Shill MC. Biological Investigations of Dried Fruit of Solanum nigrum Linn. S. J. Pharm. Sci. 2010;3(1):38-45.

42. Musa AA. Cytotoxicity activity and phytochemical screening of Cochlospermum tinctorum Perr Ex A. Rich Rhizome. J Appl Pharm Sci. 2012;2(07):155-159. DOI: 10.7324/JAPS.2012.2723

43. Olaleye MT. Cytotoxicity and antibacterial activity of methanolic extract of Hibiscus sabdarifla. J Med Plants Res. 2007;1(1):9-13.

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