ANTINUTRITIONAL COMPOSITION OF Securidaca longepedunculata (VIOLET TREE)

BY

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CERTIFICATION

This is to certify that the research project reported here was concluded by HALIMAH UMAR ABACHE (0811302062) and has been approved as part of the requirements for the award of Bachelor of Science degree in biology of Usmanu Danfodiyo University, Sokoto.

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DEDICATIOTN

This research work is dedicated to my father late Umar Abache (May His Soul Rest In Peace).

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ABSTRACT

The antinutritional composition of leafs and stem bark s. *longepedunculata* were investigated using standard analytical methods. The leafs and stem bark of *S. longepedunculata* was air dried and the sample was pulverized for the determination of anti-nutritive contents. The antinutritional continents viz phytate, oxalate, nitrate and cyanide were determined in this investigation. Nitrate content was significantly ($p<0.001$) found to be abundant in both the leafs and stem bark followed by cyanide ($p<0.01$). Phytate and oxalate were found to be non-significant ($p>0.05$) in both the leafs and stem bark. In conclusion, the result of the present study shows that both the leafs and stem bark can be consumed without any restriction. However, consumption in large amount with higher level of these anti-nutrients should be avoided.

CHAPTER ONE

1.1 INTRODUCTION:

Plants commonly synthesized a range of secondary metabolites as part of their protection against attack by herbivores, insects and pathogen or as a means to survive in adverse growing conditions. If farm or domestic animals or humans consume these plants, these compounds may cause adverse physiological effects. The terms anti nutritional or natural toxicant have been widely employed or describe plant defense metabolites in the food and nutrition literature. The observed biological effects very greatly, depending upon the structures of the individual compound, which can range from high molecular weight proteins to simple amino acids and oligosaccharides (Khokhar and Apenten, 2006).

Antinutritional factors are those substances found in most food substance which are poisonous to humans or in some ways limit the nutrient availability to the body. Plant evolved these substances to protect them and prevent them from being eaten. However, if the diet is not varied, some of this toxin builds up in the body to harmful levels (Norman and Petter 1987).

Anti-nutritional factors are present in different food substances in varying amounts, defending on kind of food, mode of its propagation, chemicals used in growing crop as well as these chemicals used in storage and preservation of the food substances. These anti-nutritional factors must be inactivated or removed if values of food substances are to be fully maintained (Inuwa et.al, 20011).

The plant *Securidaca longepeduculata* is native to Africa and commonly known as Rhodesia violet, violet tree and also as ezeogwu, ipeta and uwar magunguna (mother of drugs) in Igbo, Yoruba and Hausa language in Nigeria (Agbaje and Adekoya, 2012). The plant is a semi-deciduous shrub that grows up to 12m``` tall. It is sping and much branched with an open rather straggle looking crown (owoyele et al, 2006).

The genetic name comes from a Latin word secures meaning hatched. Repairing to the shape of the met with its curved membranous wing; and long pedunculata which refers to the long peduncle. The plant is very attractive to birds, butterflies and insects especially when in flowers. The leaves are available crowded on dwarf spur branch lets which are sometimes spine tipped. They have very fine hairs when young but they lae them as they mature. Flowers are sweetly, scented, in short bunches pink to purple and are produced in early summer. They are a lot 10mm long and are each borne on a long slender stalks (Peduncle) terminals and auxiliary sprays are a lot 30-50mm long appearing with long leaves (http/www.plantzaafrica.com/plant.qrs/securidalong.htm). *Securidaca longepedunculata* is a threatened and protected species. The violet tree is used for medicinal purposes and other uses in rural areas. What make it so threatened is the fact that root are target for people using the plant, which makes it difficult for the plant to survive constant harvesting. It occurs in the Northwest and limpopes provinces of the South African and Mozambique and it is widely distributed in tropic Africa. The violet tree is found woodland and arid savannah soil. (Olaleye et al, 1998). The violet tree is the most popular of all the traditional medicinal plants in South Africa and is used for almost every conceivable ailment. Some vernacular names means "king of medicines" The roots are very poisonous, smell like wintergreen oil and are said to contain methylsalicylate. In some parts of West Africa they are used as arrow poison. The root and bark are taken orally either powdered or as infusion for treating chest complaints, inflammation, abortion, ritual suicide, tuberculosis, infertility, veneral disease and for constipation. Toothache can also be relieved by chewing the roots. Powdered roots are used to treat headache by rubbing them on the forehead. Infusions of the root are used for washing topical ulcers. In Limpopo, the venda take root for mental disorders and against children's illness during breast feeding. The vender people mix the powdered root with maize and sorghum beverages for men being sexually week. In Zimbabwe, the roots are given to people who are believed to be possessed by evil spirits. Pounded with water and salt they are used against snake bites and cough (Owoyele et al, 2006).The plant is widely used in Nigeria for treatment of many ailments. This includes the use of its root as a purgative, diuretic, diaphoretic, emetic and expectorant. The leaves extract are used in the treatment venereal disease, snake bite as an abortifacient, Haemostatic a gat and expectorant, the back is used for treating stomach problems as an anti-flammatory agent. The decoction of the seeds is giving for fever and Rheumatism (soladoye, 2006).

1.2 JUSTIFICATION OF THE STUDY

Securidaca longepedunculata is widely used in Nigeria for treatment of many ailments. However, despite the frequent use of this plant for the treatment of various ailments there is currently no study have been reported so far on the antinutritional composition of this medicinal plant.

1.3 AIMS AND OBJECTIVES

The aim of this study was to determine the anti nutritional composition of *Securidaca longepedunculata* and the specific objective of the study were to;

i. Determine the content of oxalate in the plant extract.

ii. Evaluate the composition of cyanide in the sample.

iii. Composition of nitrate in the sample.

iv. Evalute the content of phytic.

CHAPTER TWO

2.1 LITERATURE REVIEW

Afrikaans (Krinkhout); Amharic (es a manahi); Arabic (Saggat, alali); Benba (mupapi); English (violet tree, fiber tree, Rhodesia violet); Hausa (uwar magunguna, sanya); lozy (mwinda) Nyanja (mwinda, mpulaka); shana (mufufu); Swahili (muteya, mzigi); Tigrigna (shotora); tonga (njefu, bwazi, mufufuma); voolllof (fouf); yoruba (ipeta)

2.1.1 DISTRIBUTION

Native: Angola, Benin Botswana, Burundi, Cameroon, Chad, Coted'ivoire, Democratic Republic of Congo, Eritrea, Ethiopia, Gambia, Ghana, Guinea, Kenya, Malawi, Mali, Mozambique, Namibia, Niger, Nigeria, Rwanda, Senegal, Sierra Leone, south Arica, Sudan, Tanzania, Uganda, Zambia, Zimbabwe.

2.1.2 DESCRIPTION

Securidaca longepedunculata is a semi-deciduous shrubs or small tree that grows to 12m tall, with an often flattened or slightly fluted bole. It is spiny and much branched with an open, rather straggly looking crown.

Leaves alternate or clustered on dwarf, lateral branchlets simply variable in size and shape, broadly oblong to narrowly elliptic, 1-5X, 0.50-.2cm with very fine hairs when young but losing these by maturity; apex rounded.

Image of *Securidaca longepedunculata* leafs

Flowers rather small, about 10mm long, pink to lilac or purple, sweetly scented, on long slender stalks produced in beautiful profusion in terminal axilllary sprays 3-5cm long, appearing with the very young leaves, bisexual, sepals 5, unequal, the lateral 2 being petalloid, large and wing like; petals 3 free, the medium petal hooked; stamens 8, joined to form a split tube.

Image of *Securidaca longepedunculata* flower

Fruit is more or less a round nut, somewhat heavily veined, occasionally smooth, bearing a single oblong rather curved, membranous wing up to 4cm long; purplish-green when mature.(orwa et al,2009).

Image of *Securidaca longepedunculata* fruits

2.2 ETHNOPHARMACOLOGICAL USES

Many studies have been carried out on the ethnopharmacological uses of different part of *S.longepedunculata* research has shown that the aqueous root bark extract of S longepedunculata posses cytotoxic and antiproliferative activity on ehrlich ascites carcinoma cells in swiss albino mice at IC_{50} of 67mg/ml (Lawal et al, 2013).

Studies also shown that the leave, stem and roots of *S.longepedunculata* posses mulluscicidal activities at LC_{50} value ranged from 0.15-0.60ppm and LC_{90} value ranged from 0.80-6.90ppm for both ethanoic and methanoic extract (Olofintoye et al, 2010).

The antimicrobial activities of volatile oil obtained from the roots back of *S.longepedunculata* shown to have spectrum of activities against *Escherichia coli* and *Candida albicans* (Alitonon et al, 2012).

Others ethnopharmacological uses includes purgative (Olajide et al, 1998), diuretic, diaphoretic, emetic and expectorant (Gill, 1992). The leaves extracts are used in the treatment of venereal diseases, snake bites as an abortifacient, haemostatic agent and expectorant (Olaleye et, al 1998; Gill 1992). The back is used for treating stomach problems and as an anti-inflammatory agent. The decoction of the seed is given for fever and rheumatism (Gill 1992), insecticidal (Afful et al, 2012; Jayasekara et al, 2005).

2.3 CONSTITUENTS

In the roots the flavonoids 1,7-dimethoxy-2-hydroxyxanthone and 1,4-dihydroxy-7-methoxy-xanthone, the saponine aglycones presenegin, elymclavine, sinapic acid, 3,4,5-o-caffeoyl-quinic acid, securinine, methylsalicylate and different monosaccharides like glucose and fructose were found. The sugers result from hydrolysis of saponines (Sapeika, 1944) .

From the ethyl acetate fraction further xanthones were isolated. Some of them showed moderate antimicrobial activity (Smith et al, 1979). The seed oil contains of fatty acids and triaclglycerols with unusual structures, so seven chromatographically distinct groups of triacylglycerols in two series. One series represents monoaceto-triacylglycerols and the other one normal triacylglycerols with only long chain fatty acids (Meli et al, 2007).

2.4 ANTINUTRITIONAL FACTORS

Antinutritional factors of any food substances can be defined as a compound which acts to reduce utilization and or food intake (Osagie Eka, 1998). Any substances that prevents absorption of nutrient by altering its nature or binding to it in the body is termed anti-nutritional factors found in the world in different plants and all of them have one effect or the other. The following are example of antinutritional factors found in plants namely: trypsin inhibitor, saponins, glutanins, goitregens, lectins, mineral binding substances, tannins, oxalate, cyanide, phytate and so on (Liener, 1994). The knowledge of substance naturally present in plants that we use as food may be useful in several ways.

2.4.1 PHYTATE

Phytate is the salt of phytic acid. This is also responsible for the storage of phosphorous in plant tissues. This is a hostitol with phosphate groups. It has high binding affinity for minerals such as Zn, ca, Na, Mg, k, Fe. It forms insoluble complexes with them thus making them unavailable by preventing their absorption by the intestine. This process contributes to mineral deficiencies.

2.4.2 OXALATE

There are naturally occurring substances found in plants, animals and in humans. In chemical terms, oxalates belong to a group of molecules called organic acids, and are routinely made by plants, animals and humans. Our bodies always contain oxalates, and our cells routinely convert other substances into oxalates. In addition to the oxalates that are made inside of our body, oxalates can arrive at our body from the outside from certain foods that contain them.

There are found in leafy vegetables, fruits, nuts and whole grains. They prevent the body from absorbing some of the ca, and others minerals in food. They are found highest in wheat, beans and spinach.

2.4.3 CYANIDE

Cyanide are chemical compound that contains the cyano group, it is highly toxic- cyanide is found in a number of plants, In plants cyanide is usually bound to sugar molecules in the form of cyanogenic glycosides and defend the plant against herbivores.

Cyanide is a plant and rapid acting as phyxiant which prevent tissue utilization of oxygen by inhibition of the cellular respiratory enzymes, cytochrome oxidase. Inhalation or ingestion of cyanide produces reactions with a few seconds and death within minutes.

2.4.4 NITRATE

Nitrate is a naturally occurring form of nitrogen. The formation of nitrate is an integral part of the nitrogen cycle in our environment. In moderate ant it is harmless but in high ants it is toxic plants use nitrate from the soil to satisfy nutrients requirement and may accumulate nitrate in their leaves and stems.

CHAPTER THREE

3.1 METHODOLOGY AND PROCEDURE

3.2 Collection of samples

The stem bark and leaves of *Securidata Longependunculata* was collected around Besse Local Government of kebbi State. It was identified in botany unit Usman Danfodiyo University, Sokoto and was deposited at the herbarium for reference.

3.2.1 Preparation of sample

The stem bark and leafs of *Securidata Longependunculata* was air dried under the sun, the samples was pulverized into fine powder form and weighed for the analysis.

3.3 ANTINUTRITIONAL FACTORS DETERMINATION

3.3.1 Determination of phytate

Procedure: Two grams (2g) of sample was soaked in 100cm^3 of 2% HCl for hours and filtered. 5cm^3 of NH_4SCN and 53.3cm^3 of distilled water were mixed together and titrate against 0.01M standard $FeCl_3$. Solution containing 0.019g Fe/cm^3 until a brownish yellow colour persisted for 5mins. Phytin phosphorus (1cm^3 = Fe = 1.19mg phytin phosphorous) was determined and phytate content was calculated by multiplying the value of phytin phosphorous by 3.55.

3.3.2 Determination of oxalate (Day and Underwood, 1986)

Principle:

Oxalate is precipitated as calcium-oxalate; the concentration is determined by filtration with potassium permanganate which gives a faint pink colour as end point.

Procedure:

To 2g of the sample, 75ml of 15% H_2SO_4 was added. The solution was carefully stirred intermittently with a magnetic stirrer for 1hr and filtered using whattmann no.1 filter paper . A 25ml of the filtrate was then collected and titrated against 0.1N $KMnO_4$ solution till a faint pink colour appeared that had persisted for 30secs, $1cm^3$ of $KMn0_4$ = 0.00450g oxalic acid.

3.3.3 Determination of cyanide (Wang and filled)

Principle:

Cyanide standard curve was prepared by the alkaline picrate method. This is based on the reaction between HCN and alkaline picrate to give a characteristic orange colour of whose absorbance could be measured at 490nm.

Procedure:

To 0.5g of the grounded sample, 50mls of distilled water was added in a conical flask and shaken for an hour. The mixture was filtered using cotton wool. To 1ml of the filtrate, 4mls of alkaline picrate solution was added to the test tube corked. The test tubes were incubated in a water bath at 95^0C for 5minutes. The test tubes were cooled and measured at 490nm against a reagent blank. The amount of cyanide was then extrapolated from the cyanide standard calibration curve.

3.3.4 Determination of nitrate

Principle:

This method was described by IITA(1988), and was adopted in which 100mg of the sample was weighted into a $15cm^3$ centrifuge tube and $10cm^3$ of distilled water added. The content was incubated in water bath at 45^0C for 1hr, cooled and centrifuge at 5000 revolution per minutes. The clear supernatant was into a clean test tube stopper and stored in a refrigerator prior to nitrate analysis.

Procedure:

Nitrate stock solution (100ppm) was prepared by dissolving KNO_3 (1.63g) With distilled water in a $100cm^3$ volumetric flask up to the mark. To prepare series of standard solutions of 0, 1, 2, 3, 4, and 5ppm, 0.2, 0.4, 0.6, 0.8, $1.0cm^3$ of the stock solution were added to six $20cm^3$ volumetric flask. Similarly $0.2cm^3$ of the extract was put into another $20cm^3$ volumetric flask. To the flasks $0.8cm^3$ of 5% salicylic acid-sulfuric acid reagent was added and mixed thoroughly. The contents were allowed to stand for 20min and followed by the addition of 2M NaOH solution (to raise the Ph to above 12) to the mark. The contents were cooled at room temperature and its absorbance measured at 410nm with spectrophotometer. The calibration curve was plotted from which the concentration of nitrate (x) in the samples was extrapolated. Nitrate content in the sample was calculated using equation 2.20

CHAPTER FOUR

4.1 RESULT

The result of antinutritive content of *S. longepedunculata* leafs and stem bark was presented in the table 1 and 2.

Table 1. Antinutritive contents of leafs and stem bark of *S. Longepedunculata*

Plant Material	oxalate	phytate	nitrate	cyanide
leafs	–	–	++	+
stem bark	–	–	++	+

+ present , – absent , ++ higher .

Table 2. Antinutritive contents of leafs and stem bark of *S. Longepedunculata*

Plant Material	oxalate	phytate	nitrate	cyanide
leafs	0.0108 ± 0.009^{ns}	3.802 ± 0.423^{ns}	3.733±0.058***	0.044±0.003**
stem bark	0.007 ± 0.005^{ns}	7.886 ± 1.062^{ns}	2.9±0.100***	0.063±0.003**

Values are expressed as mean ± standard deviation, **p<0.01, ***p<0.001, ns: non-significant.

Result from this study revealed that both the leafs and stem bark of S. Longepedunculata contain the antinutrients investigation, These include oxalate, phytate nitrate and cyanide. However, nitrate was the most abundant antinutritive factor in the leafs and stem bark followed by cyanide having significant difference (p<0.001) and (p<0.01) respectively. Phytate and oxalate present in both leafs and stem bark of *Securidata longepedunculata* were found to be non-significant (p>0.05).

CHAPTER FIVE

5.1 DISCUSSION

To avoid predation, plants synthesized a range of low and high molecular weight compounds. These secondary metabolites play a role in defense against herbivores, insects, pathogens, or adverse growing conditions. Many of these compounds may be labeled as antinutrients in the human diet due to their occurrence in fresh foods and processed foodstuffs (Khokhur and Apenten, 2006). Such antinutrients found in human diet include oxalate, phytate, nitrate as well as cyanide.

According to Ladeji (2005) oxalate can bind to calcium present in food therapy rendering calcium unavailability for normal physiological and biochemical role such as the maintenance of strong bones, teeth, cofactor in enzymatic reaction, nerve impulse, transmission and as clotting factor in the blood. The calcium oxalate which is insoluble may also precipitate around soft tissues such as the kidney, causing kidney stones (Oke 1969). Though, lost of calcium leads to degeneration of bones, teeth and impairment of blood clotting process (Badifu and Okeke, 1992). The present finding therefore indicate that oxalate is safe for human consumption due to limited amount detected.

According to Oke (1968) a phytate diet of 1.6% over long period decrease the bioavailability of mineral elements in mono gastric animals. Phytic acid can bind to mineral elements such as calcium, zinc, manganese, iron and magnesium to form complexes that are indigestible, thereby decreasing the bioavailability of these elements for absorption (Erdman 1979). Phytate is also associated with nutritional diseases such as rickets and osteomalacia in children and adult respectively (Makkar and becker 1998). Nevertheless, phytate was known to be anticarcinogen that protects against colon cancer and it is known to be a potent antioxidant that inhibits Fenton reactions eading to lipid peroxidation and inhibition of polyphenol oxidase (Umar, 2005).

Studies have indicated that nitrates generally cause methaemoglobinaemia in young infants but not in adult (fytianos and zarogiannis, 1999). Too much of nitrate in the body prevents the cells from being able to use oxygen which they used in order to survive, that is, it interferes in cellular respiration. However, when reduced to nitric oxide it plays an important role in the body as it provide host defense against numerous micro organisms (Benjamin, 2000).

Consumption of high levels of cyanide is associated with a serious health problem, spastic paraparetis known as Konzo , in Nigeria a neurological disease known as tropical ataxis neuropathy (TAN) was also linked to consumption of high level of cyanide in cassava based diet (Hassan and Umar , 2004). Only plants with more than 200mg of cyanide equivalent per 100mg fresh weight are considered dangerous. (Betancur –Ancona et al., 2008). However, the presence of cyanide found in the plant extract shows that both the leaves and stem bark are not safe for consumption as far as cyanide is concerned.

CONCLUSION

In conclusion, the antinutritional analysis of the leafs and stem bark of *Securidata longepedunculata* commonly consumed for various ailments showed that both the leafs and stem bark contained traceable amount of nitrate and cyanide having lower quantity of oxalate and phytate. Hence they can be consumed without any restriction. However, consumption in large amounts with higher level of these antinutrients should be avoided.

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