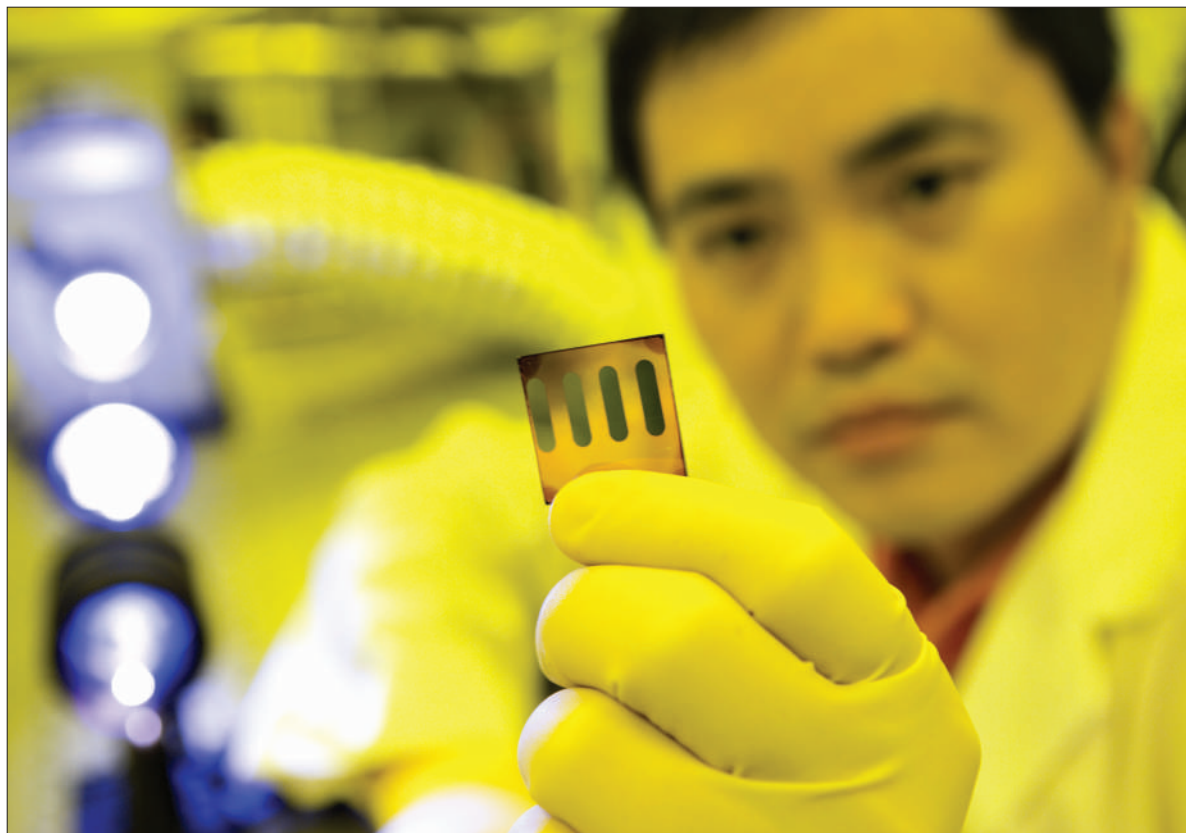




Singapore Journal of

Scientific Research

ISSN: 2010-006x

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Research Article

Nutritional and Phytochemical Evaluation of *Ageratum conyzoides*: A Neglected Edible Wild Vegetable in Ekiti State, Nigeria

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Abstract

Background and Objective: *Ageratum conyzoides* is regarded to as weed and little is known about its nutritional potential among the local populace. The vegetable could help in combat malnutrition in man and as well as animal feeds supplement if properly harness. Despite this, the vegetable is quite underutilized and the need to explore the medicinal and nutritional potentials led to this study. **Materials and Methods:** The leaf and flower of *Ageratum conyzoides* were analyzed for phytochemical, proximate and mineral compositions using standard methods for chemical analysis. **Results:** The qualitative phytochemical screening results revealed the presence of active secondary metabolites which are alkaloids, tannins, total phenols, saponins, flavonoids. Steroid and cardiac glycoside are found in leaf only while terpenoids are not detected in both samples. Quantitatively, the phytochemical composition results revealed alkaloids (0.31 and 0.13 mg g⁻¹), saponins (0.13 and 0.08 mg g⁻¹), total phenol (8.46 and 6.24 mg TEA g⁻¹), tannin (3.86 and 2.83 mg TEA g⁻¹) and flavonoid (5.80 and 5.19 mg QE g⁻¹) in the leaf and flower, respectively. The results of percentage proximate estimation showed that leaf and flower of the wild vegetable have low moisture content, high contents of crude protein, ash, carbohydrate and energy as well as moderate quantity of crude fat. The vegetable was very rich in K, P, Ca, Fe, Mg and Zn and moderate amount of Cu and Mn. **Conclusion:** This wild vegetable contained bioactive phytochemicals that might support its excellent therapeutic uses in the treatment and prevention of diseases. The vegetable could equally be recommended as supplement diet considering its rich content of crude protein, essential minerals as well as high caloric value.

Key words: *Ageratum conyzoides*, phytochemical, wild vegetables, therapeutic, minerals

Citation: Oyeyemi Sunday Dele, Kayode Joshua and Olawumi Jumoke, 2019. Nutritional and phytochemical evaluation of *Ageratum conyzoides*: A neglected edible wild vegetable in Ekiti State, Nigeria. J. Sci. Res., 9: 69-76.

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Since the time immemorial, the world has been endowed completely with all its needs to survive and cater for its inhabitants. The dependent of man on plant to meet his needs on foods and drugs are increasing every day. Medicinal plants have been the major demand due to their great efficiency of herbal remedies¹. These plants whose fruits, seeds, shoots, leaves, nuts and tubers, serves as an indispensable constituents of people diets equally supply body with protein, carbohydrate, essential minerals and vitamins.

Vegetables play an important role in human nutrition. Consumptions of vegetables have been identified as one of the critical routes to good health and longevity. Studies have revealed that vegetarians are less prone to diseases, live longer, healthier and more productive lives with stronger immunity^{2,3}. Despite the fact that vegetables serve as an indispensable constituents of human diet providing the body with minerals, vitamins, fibre in addition to essential amino acid and energy⁴ a large percentage of these vegetables are exploited or completely neglected. There have been very little concerted efforts towards exploiting them for improving nutritional status of the local populace. There is need to carry out more studies on these neglected wild vegetables especially in Ekiti State where nature has endowed her greatly with these plants.

Ageratum conyzoides (Linn) belongs to the family of Asteraceae. It is an erect branching annual herb with shallow, fibrous roots. Its leaves are opposite about 20-100 mm long and 5-50 mm wide. Both leaf surfaces are sparsely hairy, rough with prominent veins and when crushed, the leaves have characteristics odour.

The plant has been used for the treatment of various ailments such as headaches, pneumonia, analgesic and gynaecological diseases⁵. In African traditional medicine, *A. conyzoides* has been used as purgative, febrifuge, anti-ulcer and wound dressing, The aqueous extract and fraction of leaves of the plants are known to be very good anti-malaria remedy⁶. The plant extract is found to have cardiovascular depressant activity as well as antispasmodic effect⁷. The decoction of infusion of the herb is given in stomach ailments such as diarrhea, dysentery and intestinal colic⁸.

The quest to create awareness on nutritional benefits and therapeutic potential of *A. conyzoides*, one of the neglected edible vegetable necessitated this present study. The study was carried out to investigate the phytochemical, proximate and mineral compositions of *A. conyzoides* in Ekiti State, Nigeria.

MATERIALS AND METHODS

Study area: The experiment was carried out at Agricultural Extension Laboratory, Faculty of Agricultural Science, Ekiti State University during the month of April, 2018 to January, 2019.

Fresh leaves and flowers of *A. conyzoides* were obtained from an uncultivated farm land in Ado Ekiti, capital city of Ekiti State, Nigeria. The town is located at latitude of 7°40'N and Longitude 5°15'E in the tropical region of south western Nigeria.

Collection of plant materials: The plant was collected, identified and authenticated in the herbarium of the Department of Plant Science and Biotechnology, Ekiti State University, Ado Ekiti. The leaves and flowers were air dried at room temperature (25-30°C) for 3 weeks. The dried samples were pulverised and stored in air tight containers for further analysis.

Qualitative phytochemical screening of extracts: The crude extracts of the samples were tested for the presence of phytochemicals like tannins, flavonoids, alkaloids, saponins, steroids, phenols, cardiac glycoside and terpenoids according to AOAC⁹.

Qualitative analysis of the phytochemicals

Test for tannins: A portion (1 g) of dried powdered sample of the leaf and flower each was taken and boiled in 200 mL of water in a separate test tube and then filtered. A few drops of 0.1% ferric chloride were added and the solution was observed for brownish green colouration.

Test for saponins: One gram of the powdered leaf and flower each was boiled in 20 mL of distilled water in a water bath and filtered 5 mL of the filtrate was mixed with 5 mL of distilled water and shaken vigorously. The formation of stable foam was taken as an indication for presence of saponins.

Test for alkaloids: Three milliliters of 1% HCL was taken and added to 3 mL of aqueous extract of the leaf and flower each and stirred on a steam bath and filtered. Mayer's reagent (potassium mercuric iodide) was then added to the filtrate. Formation of yellow coloured precipitate shows the presence of alkaloids.

Test for flavonoids: To 1 mL of aqueous extract of the leaf and flower, 5 mL of 10% dilute ammonia solution was added. This is followed by addition of few drops of concentrated H₂SO₄. A yellow coloration observed showed the presence of flavonoids.

Test for phlobatanins: A portion (2 mL) of aqueous extract was added to 1% aqueous hydrochloric acid and then boiled. Formation of red precipitate confirmed the presence of phlobatanins.

Test for phenols: In a test tube 1 mL of the crude extract of the leaf and flower each was taken and dissolved in 5 mL of alcohol. To 1 mL of the above solution is added one drop of neutral ferric (5%) solution. Formation of an intense blue colour indicates the presence of phenols.

Test for terpenoids: A portion (5 mL) of the leaf and flower aqueous extract each was mixed with 2 mL of chloroform and 3 mL of concentrated H_2SO_4 was carefully added to form a layer. A reddish brown colouration of the interface was formed to show positive results for the presence of terpenoids¹⁰.

Test for steroids: Two millimeters of acetic anhydride were added to 0.5 g of the leaf and flower each with 2 mL of concentration H_2SO_4 acid. Presence of steroids is noted by the changing of colour from violet to blue^{10,11}.

Test for cardiac glycosides (Keller-Kiliani test): A portion (1 g) of the powdered sample of the leaf and flower each was dissolved in 5 mL of distilled water and 2 mL of glacial acetic acid solution containing one drop of ferric chloride solution. This was underplayed with 1 mL of concentrated H_2SO_4 . A brown ring at the interface indicated the presence of deoxysugar of cardenolides. A violet ring appeared below the ring while in the acetic acid layer a greenish ring was formed just above the brown ring and gradually spread throughout this layer.

Quantitative analysis of the phytochemicals: The quantitative estimations of the phytochemicals present in the leaf and flower extracts were determined using the standard methods¹¹⁻¹⁴.

Proximate composition: The proximate composition of the wild vegetable was carried out according to the AOAC procedures to determine the moisture content, crude protein and ash content¹⁵. The moisture content of the leaf and flower of the vegetable were determined by heating 5 g of the sample each in a crucible inside an oven at temperature of 105°C to a constant weight. Total ash was determined by weighing 5 g of each sample in a crucible and ignited in a muffle furnace at 550°C for 6 h, cooled and reweighed. Crude protein was determined using the micro-Kjeldahl

method by multiplying the nitrogen content of the sample each¹⁶ by 6.25. Carbohydrate content was determined according to Onwuka¹⁷. Available carbohydrate was calculated as follows:

$$\text{Available carbohydrate (\%)} = 100 - (\text{Moisture (\%)} + \text{ash} + \text{protein} + \text{fiber})$$

Determination of the mineral composition: Elemental analysis was carried out using atomic absorption spectrophotometer (Buck Scientific Model-210 VGP) for sodium, potassium, manganese, magnesium, iron, calcium, zinc and copper while phosphorus was determined calorimetrically¹⁸.

Determination of energy: The estimated energy values in the leaf and flower in Kilocalorie (Kcal/100 g) was determined adding the multiplied values for crude protein, crude lipid and carbohydrate respectively, using the factor (4, 9 and 4 Kcal) as:

$$\text{Energy value (Kcal/100 g)} = (\text{Crude protein} \times 4) + (\text{Crude fat} \times 9) + (\text{Total carbohydrate} \times 4)$$

Statistical analysis of data: All experiments were carried out in triplicates and the data expressed as Mean $\pm$ SD.

RESULTS

The leaf and flower of the *A. conyzoides* were investigated for active secondary metabolites. The results obtained revealed the presence of alkaloids, tannins, saponins, flavonoids and total phenols in the leaf and flower while cardiac glycosides and steroids were present in the leaf but not detected in the flower. Terpenoids and phlobatanins were absent in both the leaf and the flower of the sample investigated (Table 1). Results of the quantitative phytochemical estimation are presented in Table 2. The results revealed that the leaf has higher amount of saponins, tannins flavonoids and total phenol as compared to the flower.

Table 1: Qualitative phytochemical screening of the leaf and flower of *A. conyzoides*

Phytochemicals	Leaf	Flower
Alkaloids	+	+
Tannins	+	+
Phenols	+	+
Saponins	+	+
Flavonoids	+	-
Terpenoids	-	-
Phlobatanins	-	-
Steroids	+	-
Cardiac glycosides	+	-

+: Presence, -: Absence

Table 2: Quantitative phytochemical estimation of the leaf and flower of *A. conyzoides*

Phytochemicals	Leaf	Flower
Alkaloids (mg g ⁻¹)	0.31±0.01	0.13±0.01
Saponins (mg g ⁻¹)	0.13±0.01	0.08±0.01
Total phenol (mg TAE g ⁻¹)	8.46±0.04	6.24±0.01
Tannins (mg TAE g ⁻¹)	3.86±0.04	2.83±0.01
Flavonoids (mg QE g ⁻¹)	5.80±0.05	5.19±0.01

Values are expressed as Means±SD, n = 3

Table 3: Percentage proximate composition of the leaf and flower of *A. conyzoides*

Proximate/composition	Leaf	Flower
Moisture content	11.74±0.06	11.76±0.04
Crude protein	16.84±0.09	8.74±0.06
Crude fat	6.24±0.04	4.10±0.01
Ash content	12.70±0.05	10.31±0.01
Crude fiber	6.90±0.05	8.17±0.05
Carbohydrate	45.65±0.01	56.90±0.01
Energy (Kcal/100 g)	306.12±0.05	299.46±0.01

Values are expressed as Means±SD, n = 3

Table 4: Mineral composition (mg/100 g) of the leaf and flower of *A. conyzoides*

Minerals	Leaf	Flower
Sodium	54.15±0.05	38.35±0.25
Potassium	243.55±0.05	132.60±0.20
Phosphorus	30.33±0.07	21.43±0.21
Calcium	51.80±0.20	88.41±0.01
Copper	5.06±0.04	0.95±0.03
Magnesium	52.55±0.05	45.16±0.06
Zinc	11.86±0.04	8.18±0.01
Iron	42.30±0.20	98.80±0.01
Manganese	7.43±0.03	45.16±0.06

Values are expressed as Means±SD, n = 3

The result of the proximate composition of the leaf and the flower of *A. conyzoides* as presented in Table 3 revealed low moisture content (11.74 and 11.76%), moderate amount of crude fat (6.24 and 4.10%) and crude fiber (6.90 and 8.17%). The percentage crude protein content was high (16.84 and 8.74%) with relatively high carbohydrate (45.65 and 56.90%) and the overall estimated energy values are 306.12 and 299.46 Kcal/100 g, respectively.

The result for the mineral compositions of the leaf and flower is shown in Table 4. Potassium content (243.55 and 132.60 mg/100 g) was highest compared to other minerals assayed. The leaf has highest values of Na (54.55 mg/100 g), Mg (52.55 mg/100 g), P (30.33 mg/100 g) and Mn (7.43 mg/100 g), while the Ca value (51.80 mg/100 g) and Fe (98.80 mg/100 g) were the highest in the flower as compared to the leaf of the wild vegetable.

DISCUSSION

In the present study, attempts were made to assess the phytochemical profile, proximate and mineral composition of *A. conyzoides*, an important underutilized wild leafy vegetable. The results of the phytochemical investigation

revealed the presence of medically active constituents of *A. conyzoides*, an important underutilized wild leafy vegetable. Both the leaf and the flower were rich in active secondary metabolites having medicinal properties. The leaf was found to be richer in phytochemical characters as compared to the flower part. The results showed the presence of flavonoids in the leaf but absent in the flower. The results correlated with the previous works of Amadi *et al.*¹⁹ who carried out phytochemical profiling and biological study of the leaf and flower of *A. conyzoides* in Nigeria. However, the findings differ with the absence of glycosides and steroids in the flower of plant sample investigated. Phytochemicals act in numerous ways to help the body in fighting diseases and health challenges. The consumption of active secondary metabolites enhances reduction in the emergence of degenerating diseases²⁰.

Several health benefits that have been attributed to *A. conyzoides* might be as a result of their rich biologically active chemical substances. Presence of varieties of chemical compounds impart significant amount of biological activities in this underutilized leafy vegetable. Plants and herbs that contain tannins have been reported to possess beneficial effects for treating intestinal disorder, healing of wounds and inflammation of mucus membrane²¹. Saponins are known to have anti-fungal, anti-tumours and anti-viral activities, diuretic, analgesic and healing of wound properties²². The presence of tannins and saponins support the healing and analgesic properties of *A. conyzoides*. This vegetable also contains flavonoids, which are phenolic compounds one of the largest and most important groups of secondary metabolites and bioactive compound in plants^{23,24}. Flavonoids have been reported to show antibacterial, anti-inflammatory, anti-allergic and antiviral activity²⁵.

Phenols are considered to have anti-microbial properties which in turn could make the plant effective in the treatment of typhoid fever and bacterial infections²⁶. The previous experimental reports⁶ corroborated the acclaimed herbal use of this vegetable as a very good anti-malaria remedy. Alkaloids are one of the most effective therapeutic bioactive substances in plants. They are noted for their analgesic, antispasmodic and anti-bacterial properties²⁷. Steroids are known for their antimicrobial and anti-inflammatory activities²⁷. Steroids equally regulate carbohydrate and protein metabolism, increase muscles and bone synthesis and are also associated with hormonal control in women²⁸. The vegetable contained cardiac glycosides which are increased without increasing the amount of oxygen in the heart muscle²⁹. Cardiac glycosides are very effective stimulant for cardiac failure and heart related diseases³⁰. *A. conyzoides* plant has been documented to have cardiovascular depressant activity³¹.

Several health benefits have been attributed to *A. conyzoides* leaf and flower as a result of their rich biologically active chemical substances³². However, high concentrations of these phytochemicals have been reported to be harmful to the body³³. Albeit this fear may be allayed as these multiple constituents usually act synergistically. In addition, soaking, steaming or boiling reduces their levels in plant extracts³⁴. The concentrations of the phytochemicals assayed in this study were however within the safe limit and they may not cause any hazard to the body.

The results of ash content in the study were close to the results obtained for five wild vegetables from Bangladesh³⁵ and higher than the values of *Myrianthus arboreus* and *Sparganophorus sparganophora*, underutilized vegetables in Nigeria³⁶. The high ash content shows the richness of food in terms of mineral composition. The leaf and flower of *A. conyzoides* vegetable investigated were rich in minerals and would provide an appreciable quantity of mineral elements in our diet. The crude fiber contents in this study were higher than the values reported for *Andrographis paniculata* leaves (1.28%) and *Boerhaavia diffusa* leaves (1.98%)¹ but lower than the values recorded for *Amaranthus hybridus* (10.07%), *Launaea taraxacifolia* (20.10%) and *Celestia argentea* (22.03%)³⁷. Fiber plays important role in lowering the body cholesterol level thereby reduce the risks of cardiovascular disease³⁸, as well as aids digestion³⁹. The crude fiber contents of the vegetables were moderately high and can contribute significantly to dietary intakes.

The crude protein values of the investigated wild vegetable leaf and flower were higher than the value reported for *Kedrostis africana* an edible wild plant in South Africa⁴⁰ but comparatively lower than 18.08% reported for *Adansonia digitata*⁴¹. It is generally recommended that plant food providing more than 12% of its calorific value from protein is a good source of protein. Protein is an important component of human diet needed for the growth of children as well as for constant replacement of worn out tissues⁴². The crude protein values in this study indicated that the *A. conyzoides* could provide a good source of protein to human diet. The crude fat content in this study was high compared to the values reported for some common leafy vegetables in Nigeria⁴³. However, the values were lower compared to 7.02%, 14.82% and 9.12% reported for *Amaranthus dubius*, *Pennisetum purpureum* and *Vernonia amygdalina*⁴⁴.

The carbohydrate contents were relatively higher than 22.20 and 17.14% reported for roots and leaves of *Boerhaavia diffusa* investigated¹. However, these values were lower than those reported for *Dryopteris filix-mas* (52.78%),

Corchorus capsularis (60.21%) and *Hymphasa stellate* (76.34%)³⁵. The high carbohydrate content makes the plant a potentially good source of energy and could be incorporated into human diets to enrich the energy content of diets. The estimated energy content obtained in this study were higher when compared to the values reported for *Lasianthera africana* (287.62 Kcal/100 g) and *Heinsia crinita* (293.52 Kcal/100 g)⁴⁵ but favourably compared to the range of 298.61-338.65 Kcal/100 g for some accessions of *Colocasia esculenta* leaves⁴⁶. The low overall estimated nutritive values of *A. conyzoides* leaf and flower make the vegetable to be a low energy food source which in turn may be very helpful in overweight and obesity management programme.

The results of the essential minerals K, Na, Ca, P, Mg and trace minerals in this study showed that the underutilized vegetables investigated is rich in macro and micro elements needed in human diets for overall mental and physical wellbeing. Potassium was the most abundant mineral in the leaf and the flower part of the plant investigated. Potassium helps in regulating heartbeat, neurotransmission and water balance of the body⁴⁶. In this study, the Na/K ratio = 0.45 which implies that the ratio is good and adequate. Hence, it could be recommended in diets of hypertensive patients.

The calcium contents were moderately high. The calcium is an important macro-nutrient for the growth and maintenance of teeth, bone and muscle. Similarly, phosphorus is very important in the formation of strong bone and teeth, normal nerve and muscle action, heart function and cell metabolism⁴⁷. A food source is considered as good sources of calcium and phosphorus if the Ca/P ratio is above 1 and poor sources if the ratio⁴⁸ is <0.5. The Ca/P in this study is above 1, therefore the vegetable could be considered as valuable sources of calcium and phosphorus.

Iron (Fe) is essential in the diet for the formation of haemoglobin, normal functioning of the central nervous system and in the metabolism of carbohydrate, protein and fats⁴⁹. The RDA of iron is 10 mg/100 g for adult and children and 15 mg/100 g for female adult/day⁵⁰. In fact, the vegetable flower could contribute 6 times more than the RDA of 18 mg/day needed by adults. *A. conyzoides* could be recommended in human diets for reducing anaemia mostly found among children, pregnant and menstruating women. Zinc is an essential element in the nutrition of man and it plays an important physiological role such as normal growth, behavioural response, bone formation and wound healing⁵¹. The RDA for zinc is 4-14 mg/day hence, the values obtained in this study fall within the recommended daily requirement for children and adults.

Copper is needed in the formation of haemoglobin and also in many different enzyme activities such as enzyme that catalyses the oxidation of Fe ion⁵². The Cu levels in this present study were below the permissive limit (4 mg/100 g) in the flower and slightly above this limit in the leaf as recommended by FAO/WHO⁵³. High concentration of Cu can lead to diarrhea, epigastric pain and discomfort, hypotension and vomiting⁵⁴. Magnesium is required as co-factor in many enzymes, protein and nucleic acids synthesis in the human body. It has been reported that magnesium may help to support mineral bone density in aged people⁵⁵.

CONCLUSION

This study has elucidated the phytochemical profile of the leaf and the flower of *A. conyzoides*. Presence of the varied and appreciable concentrations of active secondary metabolites in the vegetable could be attributed to its acclaimed therapeutic property in traditional medicine. The results further established the rich nutritional value of the vegetable as good sources of protein, fiber, carbohydrate and essential macro and micro elements needed for human health. Hence, the vegetable could be recommended as food supplement for both man and animal. Further study on the toxicity of the vegetable parts is however required to ascertain their possible adverse effects.

SIGNIFICANCE STATEMENT

Ageratum conyzoides contained bioactive phytochemicals that support its therapeutic utilization in the treatment and prevention of diseases.

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