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CHEMICAL COMPOSITIONS AND EFFECTS OF AQUEOUS EXTRACT OF *CISSUS MULTISTRIATA* ON SOME BIOCHEMICAL PARAMETERS IN ALBINO RATS

Omale James[1]*, Okafor Polycarp Nnacheta[2] and Irene Ifeoma Ijeh[2]

[1]Department of Biochemistry, Kogi State University, Anyigba, Nigeria.

[2]Department of Biochemistry Michael Okpara University of Agriculture, Umudike Abia State, Nigeria

E-mail: jamesomale123@yahoo.com, Tel: +23408068291727

ABSTRACT : This study was carried out to determine the nutrient compositions of the root, young and matured leaf of *Cissus multistriata* as well as assessment of its effects on some biochemical parameter in albino rats utilizing standard methods. The crude protein content was higher in the mature leaf (21.30 ± 0.60%) than the young leaf and the root. Vitamin C content was higher in the root (32.63 ± 1.95) than other parts. The root part is richer in fat and crude fibre (34.33 ± 0.58, 17.05 ± 0.99%) respectively. The assessment of the plants extract effects on selected biochemical parameters was done by administering it to the experimental animals at regular doses of 100, 200, 300, 400, 500, 600mg/kg body weight of rats for 21 days after which the animals were anaesthesized using chloroform and their blood samples were collected via cardiac puncture and analyzed. The lipoprotein profiles decreased with increase in the dosage of the extract except the HDL-cholesterol that showed increase with the increase in dosage of the extract. This is a beneficial effect and a pointer that the plant could be used in the management of cardiovascular diseases as HDL is an anti-artherogenic agent. The total protein, albumin, and blood glucose levels increased dose-dependently. The over all result showed that the plant is a potential source of nutrients, and has capacity to serve as a dietary supplement especially in protein deficiency disease as claimed by the users and could as well be used in the management of cardiovascular diseases.

Key Words: *Cissus multistriata*, Cardiovascular diseases, Dietary supplement, Albino rats.

INTRODUCTION

Proximate analysis of leafy vegetable species in Nigeria have been conducted and it was shown that the amount of a particular nutrient is influenced by the plant genotype, climate, soil fertility, age at harvest and physiological changes during post-harvest handling[1].

Cissus multistriata is one of the medicinal herbs/vegetables used in Nigeria for the management of diverse ailments. It is estimated that more than 65% of Nigerians are not reached or cannot afford modern medicine and so rely on traditional medicine, which is based on curative plants[2]. Diseases especially those ensuing from hereditary, bacterial infections, malnourishment and accidents are increasing at an alarming rate. Malnourishment and infertility have become a large burden on Nigerians and should be given attention as many people are driven to this path daily. Majority of the victims cannot afford medical treatment. It is in this regard that alternative is sought in medicinal plants[2].

Cissus is a genus of about 350 species of tropical and subtropical, chiefly woody vines of the grape family (vitaceae). The leaves are often fleshy and somewhat succulent. They are often used as medicinal plants because they contain some bioactive compounds such as vitamins, proteins, carbohydrate, polyphenols among others. These bioactive compounds are contained in their leaves, stems, roots and barks[3,4] which make these plant species to be used medicinally in the indigenous system of medicine and in the Ayurvedic and Unani systems.

Cissus multistriata is a well known plant to the traditional medicine practitioners in Nigeria. It is called Ojekere by the Igalas, Ewebiomo in Yoruba, Ochekihiozewehi by the Ebiras in Kogi State, and Mukala by the Ibos in eastern Nigeria. It is used as medicinal plant for the management of diverse ailments in different locations. The Ebiras use the stem prepared inform of decoction as internal cleanser for new born babies while the Yorubas use the leaves for the treatment of infertility in women and stomach ailment in children. It is commonly used by the

Ibajis in Kogi State for the treatment of malnutritional diseases such as kwashiorkor and marasmus in children. Other ethnomedicinal uses of this plant include its use as cough remedy, fracture healing, management of arthritis etc.

The objective of this study was to assess the nutrient components of *cissus multistriata* and its effects on some biochemical parameters using albino rats (Rattus novergicus) as experimental animals.

MATERIALS AND METHODS

Collection and preparation of plant sample.

The plant samples were collected from Ega in Idah Local Government Area, Kogi State, Nigeria during rainy season when the plant thrives well. Dirts were removed by rinsing the samples properly in clean water. They were air dried for two weeks and then pulverized using motorized blender.

Extraction Procedure

A portion (200g) of the powdered plant sample was soaked in 250ml of water for 24hours, filtered and the filtrate evaporated to give off water. The solid concentrate of the extract was stored in vials.

Chemical analysis

A portion (1g) of the powdered samples were taken and reduced to ash in a furnace. The residue was mixed with concentrated nitric acid and further ashed. The residue was dissolved in water and made up to 100ml. The elements present and their concentrations were determined with an atomic absorption/flame emission spectrophotometer (AA680 Shimadzu).

Moisture content

The moisture of the plant samples were determined following the method described by A.O.A.C[5] .Total ash content was determined by furnace incineration using the method of James[6]. Crude protein, crude fibre and fat contents were determined using the methods described by Pearson[7].

The vitamin C content was estimated as described by A.O.A.C[5]. The percentage carbohydrate was determined by difference utilizing the relationship described by Udoh and Ogun wale[8].

Experimental animals

The animals selected for this study were albino rats (Rattus novergicus). They were obtained from National Veterinary Research Institute, Vom, Plateau State, Nigeria. Twenty eight (28) young albino rats were used for the research. Their body weight ranged between 153.47 to 233.43g. The rats were divided into seven groups of four rats ach. These were six experimental groups and a control.

Administration of crude aqueous extract

A regular doses of 100, 200, 300, 400, 500 and 600mg/kg body weight of rats were administered orally twice daily for 21 days in treatment groups 1,2,3,4,5 and 6 respectively. The were fed with commercial rat feeds. Rats in the control group were given the commercial rat feeds only and water ad libitum.

Blood sample analysis

At the end of the experimental period, rats in all the groups were anaesthesized, dissected and bleed via cardiac puncture. Their blood samples were collected into heparinized sample bottles to prevent coagulation and later centrifuged for 20 minutes at 3000rpm to get the plasma which was taken up with Pasteur pipette into set of labeled sample bottles and were refrigerated until used. Total plasma cholesterol was determined by the method described by Allain[9] and Steele[10]. The method of Fossah and Principe[11] was followed in the determination of plasma triglycerides. The determinations of high density lipoprotein (HDL) was carried out following the method of Burstein[12] and Grove[13] .Low density lipoprotein (LDL) and very low density lipoprotein (VLDL) were estimated using the method of Friedwald[14]. Plasma albumin concentration was determined according to the modified Bromocresol green method of McPherson and Everad[15].

Total plasma protein determination was carried out by Biuret method[16]. The plasma glucose was measured using the method described by Trinder[17].

RESULTS AND DISCUSSION

The results presented in Table 1 indicate that the plant sample contain important mineral elements. Calcium was highest in the matured leaves (0.66 $\pm$ 0.01ppm). Sodium and potassium were abundant in the roots (0.79 $\pm$ 0.03ppm and 0.57 $\pm$ 0.01ppm) respectively than any other part of the plant. The concentration of zinc was more in the young leaves. The results of nutrient composition analysis is as presented on Table 2. The quantity of ash and crude fibre obtained were highest in the root. Similarly fat and carbohydrate were most abundant in the root as well. The crude protein did not follow this trend but was highest in the matured leaves (21.30 $\pm$ 0.60%). The root of the plant contain more vitamin C than the leaves.

The ash of a biological material is an analytical term of the inorganic residue that remains after the organic matters has been burnt away. It is not usually the same as the inorganic matter present in the original plant since there may be losses due to volatilization or interaction between the constituents [18]. The value is useful in assessing the quality of soluble edible materials. The importance is that it gives an idea of the amount of mineral elements present in a sample. This is evident in the results obtained for elemental analysis, it revealed that the plant is rich in some important mineral elements. Calcium for instance is an essential nutrient required for critical biological functions such as nerve function, blood congulation, structural supporting of the skeleton, cell adhesiveness, mitosis and muscle contract[19]. It is partly responsible for solidity as well as movement of the body.

Calcium aids digestion and promotes good growth and vigor by helping to regulate metabolism. Sodium regulates blood volume, acid-base balances, nerve and muscle function and ATP hydrolyzing activity in skeletal muscle[20]. This could justify the ethnomedicinal use of this plant in the management of kwashiorkor where minerals and vitamins are deficient.

Zinc is higher in the young leaves and the root than the matured leaves. Zinc is an essential element for humans. In young men deficiency causes staunted growth, anemia, enlarged liver and spleen[20]. The potassium content of the plant sample is higher in the root (0.57 $\pm$ 0.01ppm) followed by the matured leaves. Potassium is an essential dietary mineral that is also known as an electrolyte. Normal function of the body depends highly on the regulation of potassium concentration both inside and outside of cells[20]. This plant therefore has a lot of nutritional potentials to be harnessed.

As presented on Table 2, the root contains more fibre than the leaves. Fibre has some nutritional significance as it absorbs some toxic substances as well reduce occurrence of colonic cancer[20] . Regarding fats, the mature leaves showed a lower percentage of lipid composition (18.00 $\pm$ 1.73%) more lipids are found in the root (34.33 $\pm$ 0.58%) than the leaves. Lipids when combined with proteins form part of biological membrane that serve to contain and compartmentalize cells[21]. The crude protein composition is higher in the natured leaves than the young leaves and root. The importance of proteins cannot be over emphasized. Structurally and functionally they are the most diverse and dynamic of molecules and play key roles in nearly all biological process. Proteins are complex macromolecules with exquisite specificity, each is a specialized player in the orchestrated activity of the cell. The presence of protein in this plant sample could justify its use in the management of kwashiorkor – a protein deficiency disease. The root is richer in vitamin C (32.6 $\pm$ 1.95mg) than the leaves as presented in Table 2, even though the difference is not statistically significant ($P > 0.05$).

Vitamin C is a water soluble antioxidant. As a water-soluble antioxidant it is in a unique position to scavenge aqueous peroxyl radicals before these destructive substance have a chance to damage lipids. It works synergistically with vitamin E and glutathione peroxidase to stop free radical chain reactions. It also helps the body to absorb iron and to breakdown of histamine, the inflammatory components of many allergic reactions[22] . This plant therefore has potential to serve as a source of natural antioxidant.

The results of the effect of *Cissus multistriata* leaf extract on some biochemical parameters are as presented in Table 3. The results show that the cholesterol levels decreased with increase in the dosage of the extract. Group six (6) for instance which received the highest dosage of the extract had 80.52 $\pm$ 9.60mg/dl of cholesterol in their plasma. This is lower when compared with group 1 (85.80 $\pm$ 6.60mg/dl) and control (96.60 $\pm$ 7.43mg/dl).

The decrease in the mean cholesterol level when compared with the control was significant ($P < 0.05$). Similarly, such decrease was observed with the lipoprotein levels except high density lipoproteins which increased dose-dependently. Group 6 which received the highest dosage of the extract had 64.00 $\pm$ 7.43mg/dl of HDL.

This is higher when compared with group 1 (45.00 $\pm$ 19.98mg/dl) and the control (40.80 $\pm$ 1.12mg/dl). The difference in the mean values between each test group and the control was statistically significant ($P < 0.05$). This increase was dose-dependent and is beneficial as HDL – cholesterol participates in reverse cholesterol transport (RCT). Higher density lipoproteins are lipoproteins that participate in the clearance of cholesterol from the circulation back to the liver for synthesis of bile salts and acids and their subsequent excretion, a process known as 'reverse cholesterol transport'. Therefore, HDL is said to be anti-artherogenic since it associates with the surface macrophages and promotes the removal of intracellular cholesterol.

HDL also protects against the oxidation of lipids that activates arterogenic processes[23]. This then means that *Cissus multistriata* leaf extract could have stimulated HDL – cholesterol synthesis and decreased the level of other lipoprotein profiles, thus it could be used in the management of cardiovascular diseases and artherosclerosis as it decreased the levels of the major risk factors. The plasma glucose level like other biochemical parameters measured increased dose-dependently, this shows that the extract may not be useful as dietary therapy for the management of diabetes as claimed by herbal doctors.

Increase in the protein profiles observed could be contributory to management of kwashiorkor using this plant by herbal practitioners. Kwsashiorkor is protein deficiency disease.

In conclusion, the results of this study showed that the plant *Cissus multistriata* has the potential as dietary supplement for the management of nutrition related diseases such as kwashiorkor and could be used in the management of cardiovascular diseases. Also *Cissus multistriata* has been shown by this work to be rich in nutrient, most of which are resident in the leaves than the root with the exception of the mineral elements which are found in higher proportion in the roots. Based on the results obtained in this work, *Cissus multistriata* can be integrated into Nigerian folk medicine and food drugs.

Table 1: Shows elemental constituents of Cissus multistriata

S/N	PLANT PARTS	ELEMENTS (ppm) CALCIUM	SODIUM	POTASSIUM	ZINC
1.	Young leaves	0.45 ± 0.02	0.22 ± 0.02	0.15 ± 0.01	0.97± 0.01
2.	Matured leaves	0.66 ± 0.01	0.15 ± 0.02	0.24 ± 0.02	0.87 ± 0.01
3.	Root	0.49 ± 0.04	0.79 ± 0.03	0.57 ± 0.01	0.88 ± 0.02

Mean ± S.D. of three determination

Table 2: Nutrient composition of the leaf and root of Cissus multistriata

S/N	Plant Sample parts	Moisture %	Ash %	Crude Fibre %	Fat %	Carbohy – drate %	Crude Protein %	Vitamin C(mg)
1.	Young leaves	229.33 ±13.43	17.67 ± 0.58	7.53± 67	30.67 ± 1.15	0.71 ± 0.08	18.77 ± 0.65	27.53 ± 1.10
2.	Matured leaves	198.00± 17.78	23.00 ± 1.00	13.70±1.53	18.00± 1.73	1.62 ± 0.06	21.30 ± 0.60	30.70 ± 1.90
3.	Root	59.67 ±18.34	24.33 ± 2.52	17.05± 0.99	34.33± 0.58	0.85 ±0.05	17.30 ± 0.60	32.63 ± 1.95

Mean ± S.D. of three determinations

Table 3: Shows the Effects of *Cissus multistriata* Leaf Extract on some Biochemical Parameters in Albino Rats

Group	Dose mg /kg	Total plasma Cholesterol mg/dl	HDL mg/dl	Trig mg/dl	LDL mg/dl	VLDL mg/dl	Total plasma Protein mg/dl	Glucose mg/dl	Albumin mg/dl
Control	-	96.60 ± 7.43	40.80 ± 1.12	69.00 ± 8.89	42.00 ± 3.65	13.80 ± 0.76	5.90 ± 0.08	46 ± 2.94	1.50 ± 0.02
1	100	85.80 ± 6.60	45.00 ± 19.98	65.50 ± 7.13	27.70 ± 2.33	13.10 ± 0.36	5.91 ± 0.08	46 ± 2.94	1.50 ± 0.02
2	200	85.20 ± 6.02	55.00 ± 2.47	63.50 ± 8.44	17.52 ± 4.99	12.70 ± 0.23	6.30 ± 0.21	94 ± 0.81	2.00 ± 0.08
3	300	84.48 ± 3.36	60.00 ± 3.75	62.80 ± 4.26	11.92 ± 3.05	12.56 ± 0.59	6.30 ± 0.08	106±2.16	2.10 ± 0.14
4	400	83.96 ± 8.34	62.00 ± 10.21	61.12 ± 6.20	9.74 ± 0.84	12.20 ± 0.29	6.40 ± 0.08	112±2.16	2.50 ± 0.08
5	500	81.81 ± 11.94	63.80 ± 7.44	60.09 ± 6.69	5.99 ± 1.07	12.02 ± 0.05	7.10 ± 0.21	135±0.81	3.10 ± 0.08
6	600	80.52 ± 9.60	64.00 ± 7.43	59.92 ± 3.77	4.54 ± 0.59	11.98 ± 0.83	7.50 ± 0.08	186±2.16	3.20 ± 0.01

Mean ± S.D. of four determinations.
HDL = Higher density lipoprotein, Trig = Triglycerides, LDL = Low density lipoproteins
VLDL = Very low density lipoproteins.

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