



Phytochemical Screening and Haematological Parameters of Aqueous and Ethanolic Leaf Extracts of *Momordica charantia* on Experimental Albino Rats

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Abstract

Momordica charantia is a species of *momordica* belonging to the cucurbitaceae family. Qualitative phytochemical analysis of *Momordica charantia* confirms the presence of alkaloid, carbohydrate, flavonoid, glycoside, saponins, terpenoids, steroids, tannins and phenol. The effects of aqueous and ethanol extracts of *Momordica charantia* on haematological parameters were investigated. In this study, twenty five (25) albino rats were divided into 5 groups of 5 rats each. The control group was given normal feed and water/ saline (orally) while the test groups were also given orally; 200mg/kg body weight and 400mg/kg body weight of *Momordica charantia* aqueous and ethanol leaves extracts. The administration lasted for 21 days. At the end of the 21 days of experimental period, the rats were sacrificed and blood sample was collected into anticoagulant (EDTA) bottle for haematological analysis (RBC WBC, Hb PCV and Platelets). The results showed that the value WBC, RBC, Hb, PCV and platelets of 200mg/kg body weight aqueous (4.11 ± 2.00 , 5.58 ± 0.15 , 93.80 ± 7.79 , 29.77 ± 2.57 , 286.80 ± 9.10^b) and ethanol (6.16 ± 0.71 , 6.28 ± 0.54 , 104.60 ± 7.93 , 34.23 ± 2.76 , 256.30 ± 9.87^b) significantly decreased ($P < 0.05$) when compared to the normal control group (7.22 ± 2.09 , 6.54 ± 0.63 , 112.60 ± 11.44 , 35.38 ± 3.50 , 265.20 ± 45.18^b). The groups treated 400mg/kg body weight of aqueous extracts (3.14 ± 0.47 , 5.30 ± 0.66 , 86.20 ± 9.99 , 27.36 ± 3.63 , 296.60 ± 5.95^b) and ethanol (3.59 ± 1.08 , 5.65 ± 0.71 , 93.80 ± 1.11 , 30.41 ± 4.04 , 182.80 ± 18.75^a) showed significance difference ($P < 0.05$) when compared to normal control respectively. The results suggested that incessant consumption of the leaves of the plant may not be advisable.

Keywords: Phytochemical; haematological parameters; *Momordica charantia*; aqueous and ethanolic extracts

Introduction

Plants play a prominent role in maintenance of human health and have been used as medicine, since ancient times. According to the estimation of World Health Organization (WHO) 1995, plant extracts are used as medicines in traditional therapies by 80% of the World's population (Baker *et al.*, 1995) and more than 30% of the plant species have been used for medicinal purposes (Joy *et al.*, 1998). The use of plants as sources of drugs, vegetables and foods cannot be underestimated. Virtually all plants are medicinal hence they serve as raw materials for synthetic drugs (Sofowora, 1993). The medicinal value of these plants lies in some chemical substances that produce a definite physiological action on the human body (Antony *et al.*, 2013) Therefore, the analysis of these bioactive constituents would help in

determining various biological activities of plants. These bioactive substances which can be present in all plant parts include terpenoids, steroids, saponins, tannins, flavonoids, alkaloids (Sofowora, 1993). The medicinal plants of Africa accounts for nearly two third of the total plants species used in modern system of medicine and in rural areas as tea, extracts. Herbal drugs are widely prescribed, even when their biological ingredients are not known, due to their effectiveness, fewer side effects and low cost (Kumar *et al.*, 2009; Ajayi *et al.*, 2011). The rational design of novel drugs from traditional medicine obtained from plant offers new prospects in modern health care (Manjamalai *et al.*, 2010).

Medicinal plants have been identified and used throughout human history. Plants have the ability to

synthesize a wide variety of chemical compounds that are used to perform important biological functions, and to defend them against attack from predators such as insects, fungi and herbivorous mammals. Chemical compounds in plants mediate their effects on the human body through processes identical to those already well understood for the chemical compounds in conventional drugs; thus herbal medicines do not differ greatly from conventional drugs in terms of how they work. The conventional medicine is more than the herbal medicine in terms of their standards and purity (Lai & Roy, 2004).

In Mubi community, *M. charantia* leaves and seeds are traditionally used as medicine to cure diseases such as hypertension, diabetes, menstrual problems and malaria fever. Its many varieties differ substantially in the shape and bitterness of the fruit Bitter mole originated in the 14th century (Bagchi, 2005, Liquid extracts of the seed are used in treatment of hypertension.

Material and Method

Sample collection and Authentication

Fully matured dark green leaves of *M. Charantia* were collected in and around the vicinity of Mubi North Local Government, Adamawa State, Nigeria in the month of July, 2015. Plant species was authenticated in the State Ministry of Forestry, Mubi Adamawa State.

Sample preparation

The plant leaves were thoroughly washed with tap water and rinsed with distilled water to remove dust and other unwanted materials accumulated on the leaves from their natural environment. The dust free leaves were shade dried and pounded/pulverised to powdered form. using pestle and mortar

Extraction of Sample with Aqueous and Ethanol

The powdered leaves (100g) were weighed and soaked in 350mL of ethanol in a volumetric flask. The flask with instantaneous shaking was corked and left to stand for 48hrs at room temperature. After 48hrs, the mixture was filtered using Watman Filter Paper No. 1. The filtrate was concentrated using Rotary evaporator to dryness (Evans and Trease, 1996).

The powdered leaves (100g) were weighed and soaked in 350mL of water in a volumetric flask. The flask with instantaneous shaking was corked and left to stand for 48hrs at room temperature. After 48hrs, the mixture was filtered using Watman Filter Paper No. 1, the filtrate was concentrated using water bath at 38°C to dryness (Evans, and Trease, 1996).

Phytochemical Screening

The leaves extracts were subjected to preliminary phytochemical screening using methods described by (Evans and Trease 1996) Various qualitative chemical tests were conducted for detection of alkaloids, phytosterols, terpenoids, phenols, steroids, flavonoids, tannins, carbohydrates, glycosides and saponins.

Alkaloid detection Test

Meyer's test using Meyer's reagent: 2mL of plant extract, 2mL of concentrated HCl were added. Then 3 drops of Mayer's reagent was added by side of the tube, a white-creamy or yellow coloured precipitate indicated a positive result.

Test for glycosides

Extracts were hydrolysed with dil. HCl and then subjected to test for glycosides.

Modified Borntrager's Test: extracts were treated 5% ferric chloride solution and immersed in boiling water for about 5 minutes. The mixture was cooled and extracted with equal volumes of benzene. The benzene layer was separated and treated with ammonia solution. Formation of rose-pink colour in ammonical layer indicated the presence of glycosides.

Test for Steroids

To 1mL of plant extract, 1mL chloroform and 3 drops of conc. H₂SO₄ were added to form a layer. A reddish-brown interface showed the presence of steroids.

Phenols detection test

Ferric chloride test: 1mL of plant extract was added to 2mL of distilled water followed by 5 drops of 10% ferric chloride. Formation of bluish black colour indicated the presence of phenols.

Test for flavonoids

Alkaline reagent test: 1mL of plant extract, 1mL of 1N of aqueous NaOH were added and observed. Formation of intense yellow colour, which became colourless on addition of dilute acid, indicated the presence of flavonoids.

Test for Tannins

Gelatin test: to the extract 1% of gelatine solution containing 10% NaCl was added. Formation of white precipitate was observed which indicated the presence of tannins.

Test for Saponins

Froth test: 1mL of plant extract was diluted with 20mL distilled water and the mixture shaken vigorously in graduated cylinder for 15 minutes and then observed on standing for stable froth. Formation of 1cm layer of the froth indicated the presence of saponins.

Experimental Design

A total of 25 albino rats were used. The experiment lasted for 21 days. The rats were divided into 5 groups

The result of the phytochemical screening of aqueous and ethanol extract of *M. Charantia* revealed the

of 5 rats each to study the effect of the treated and none treated thus;

Group 1. Control: The rats in this group were given normal feed and water/saline.

Group 2. This group was given 200mg/kg body weight of the ethanol extracts of *M. charantia* orally using orogastric tube.

Group 3. This group was given 400mg/kg body weight of the ethanol extracts of *M. charantia* orally using orogastric tube.

Group 4: This group was given 200mg/kg body weight of the aqueous extracts of *M. charantia* orally using orogastric tube.

Group 5: This group was given 400mg/kg body weight of the extracts of aqueous extracts of *M. charantia* orally using orogastric tube.

Collection of Blood Sample

At the end of 21 days of experimental period, the rats were sacrificed by anaesthetics and blood sample was collected into anticoagulant (EDTA) bottle for haematological analysis (RBC WBC, Hb, PCV and Platelets).

Results

presence of alkaloids, flavonoids, glycoside, saponin,, steroids, tannins, terpenoids, and phenol.

Table 1: Phytochemical analysis of *M. Charantia* leaves extracts

Chemical Constituents	Aqueous	Ethanol
Alkaloids	+	+
Flavonoids	+	+
Glycoside	+	+
Saponin,	+	+
Steroids,	+	+
Tannins	+	+
Terpenoids,	+	+
Phenol.	+	+

+ Sign shows detection level of the phytochemicals present in extracts.

Table 2: Effect of aqueous and ethanol extract on haematological parameters

Group	WBC($\times 10^3/\text{mm}^3$)	RBC($\times 10^6/\text{mm}^3$)	Hb (g/dl)	PCV (%)	PLATELETS(mL)
Normal	7.22± 2.09	6.54±0.63	112.60±11.44	35.38±3.50	265.20±45.18 ^b
200mg/kg bwt. aqueous	4.11±2.00	5.58±0.15	93.80±7.79	29.77±2.57	286.80±9.10 ^b
200mg/kg bwt. Ethanol	6.16±0.71	6.28±0.54	104.60±7.93	34.23±2.76	256.30±9.87 ^b
400mg/kg bwt. Aqueous	3.14±0.47	5.30±0.66	86.20±9.99	27.36±3.63	296.60±5.95 ^b
400mg/kg bwt. Ethanol	3.59±1.08	5.65±0.71	93.80±1.11	30.41±4.04	182.80±18.75 ^a

The values are mean± SEM for five replicate.

WBC, White Blood Count, RBC, Red Blood Count Hb, Haemoglobin PCV Pack Cell Volume respectively

Discussion

The qualitative phytochemical analysis of aqueous and ethanolic leaf extracts of *Momordica charantia* revealed the presence of alkaloid, flavonoid, glycoside, saponins, steroid, tannins, terpenoids and phenols. Phytochemicals are plant secondary metabolite that have been reported to have many biological and therapeutic uses, so the presence of these phytochemicals is expected to make the plant a potential for many medicinal uses table 1 (Vishnu *et al.*, 2013 and Narender *et al.*, 2012).

The study of haematological status is one of the important ways for diagnosis of the root cause of disease. There were significant reductions ($p < 0.05$) in WBC, RBC, Hb PCV and platelets in rats administered *M. charantia* aqueous and ethanol extracts (Tables 2). This implies that *M. charantia* extracts could cause disturbances in osmoregulatory system of the blood cells and/or oxidative injury to the cell membrane. The extracts could suppress the activity of haemopoietic system that is responsible for the RBC production in bone marrow. The reduction may also have occurred due to lysis of blood cells (Sule *et al.*, 2012).

This is an indication that the extract could affect erythropoiesis in animals. The observed decrease in RBC count, Hb and PCV may therefore be assumed to be associated with retarded haemopoiesis, destruction and shrinkage of RBC. Also, the oxygen-carrying capacity of the blood and amount of oxygen delivered to the tissues could be affected due to the reduction of RBC and Hb which are responsible in

transferring respiratory gases (De Gruchy, 1996). Haemoglobin observed a decrease and consistent with previous studies of anemia by Horiguchi, (2007) and Dhanapakiam and Ramasamy (2001). It was evidenced that the anemia was caused not only by increased destruction of erythrocytes (Kori-Siakpere *et al.*, 2009) but also by decrease in the synthesis and release of erythrocytes into the blood circulation (Vinodhini and Narayanan, 2008).

Conclusion

It was concluded that the oral administration has a mild effect on haematological indices therefore; the incessant consumption of the leaves is not advisable, since it will cause a reduction of WBC, RBC Hb PCV and platelets.

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