

# Evaluation of Serum Lipid Profile and Oxidative Stress Markers of Wistar Rats Treated with Extract of *Momordica Charantia*

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## Abstract

This study was carried out to investigate the antioxidant and serum lipid effects of the leaf extract of *Momordica charantia* on male wistar rats. The animals were divided into three (3) groups of six (6) rats each. Group one (1) served as control and received distilled water. Group two (2) and group three (3) were treated with 200mg/kg bw and 400mg/kg bw of the hydromethanol extract respectively. Extract was orally administered once daily for 30 days. The results obtained showed that the extract caused a significant increase in the catalase and glutathione enzyme activity and a significant reduction of malondialdehyde level. There were no significant alterations of low density lipoprotein cholesterol (LDL-c), high density lipoprotein cholesterol (HDL-c), triglyceride and total cholesterol concentrations. The lipid ratios like the Castelli's Risk Index I and II were not affected. This study showed that, the leaf extract of *Momordica charantia* demonstrated anti-oxidant effects but did not influence risk ratios for developing cardiovascular diseases in wistar rats.

**Keywords:** *Momordica charantia*, antioxidant, serum lipids, hydromethanol, wistar rats.

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## INTRODUCTION

There has been a tremendous increase in the population of people using medicinal plants for prevention and treatment of various illnesses around the world. This practice is deep rooted in developing countries where a large proportion of the people depend on these natural products to improve their health and well-being. In Africa, these plants are easily accessible and cost effective making them very useful resource in basic health care [1]. *Momordica charantia* (*M.charantia*) which is also referred to as bitter melon or bitter gourd is native to the tropics but also a subtropical plant with an edible fruit, and widely grown in Africa, China, Southern and South eastern Asia, etc. In Bangladesh, *M. charantia* is one of the most popular cucurbitaceous vegetable as a result of its nutritive and medicinal value [2]. Several experimental studies carried out with *M. charantia* has documented numerous pharmacological actions of this plant. The findings in some studies showed that, *M. charantia* has antibacterial [3], anticancer [4] and antihypertensive [5] effects. The anti-diabetic effects of this plant was evaluated in alloxan induced diabetic animal models and was found to demonstrate antihyperglycemic effects [6]. In addition, complications are commonly

encountered in diabetes leading to irreversible structural and functional changes in various organs [7]. *M. charantia* has shown encouraging results in prevention and delay in progression of diabetic complications such as nephropathy, neuropathy and insulin resistance in experimental animals [8, 9]. Insulin resistance is another important feature of diabetes that is linked to dyslipidemia (Abnormal lipid profile) and atherosclerosis which contribute significantly to morbidity and early mortality [10]. The lipid profile [(total cholesterol (TC), High density lipoprotein cholesterol (HDL-c), Triglyceride (TG) and Low density lipoprotein cholesterol (LDL-c)] analysed after a 12 to 14 hour fast is of great value in identifying cardiovascular risk factors as indicated in the findings from a study which showed that abnormal serum lipids play important role in the development of atherosclerosis in later life [11]. In recent times, research studies has also indicated the involvement of oxidative stress in a number of disease processes, including cardiovascular disease (CVD), atherosclerosis, arthritis, diabetes, cancer etc. [12, 13]. Reactive oxygen and nitrogen species (RO/NS) produced during oxidative stress cause damage to cellular biomolecules, however, several defense

systems within the cells act to protect the cell and prevent uncontrolled RO/NS increase. Thus, oxidative stress markers are important tools to assess the redox states in biological systems, the presence and progression of disease, and the health-promoting effects of antioxidants in humans. The objective for carrying out this study is to investigate the serum lipid and antioxidant effects of the leaves of *M. charantia*.

## MATERIALS AND METHODS

### Animal models

Adult male wistar rats used for this study were bred in the animal house of the Faculty of Basic Medical Sciences, University of Port Harcourt, Nigeria. They were placed in standard cages and acclimatized in two weeks while maintaining them in environmental conditions with proper ventilation, 12 hour light and 12 hour dark cycle and free access to food and water. Generally, the procedures conformed to the established principles for the care and use of laboratory animals published by the National Institute of Health, USA [14]. Appropriate institutional approval was obtained for this study.

### Preparation of Plant extract

The leaves of *M. charantia* were obtained from Choba community in Obio Akpor Local Government Area of Rivers State, Nigeria. The plant materials were authenticated by the taxonomist before use. The leaves were washed to remove any dirt on it and later dried. The dried leaves were blended to fine powder and extraction was carried out using the soxhlet apparatus with hydromethanol (1:4) as solvent, at 60 – 70°C. The solution was filtered after 24 hours and the filtrate concentrated under reduced pressure of 60°C to a semi solid form using the rotary evaporator. The extract was preserved in a refrigerator at 4°C. Serial dilutions of the extracts were made to obtain concentration of 200mg per ml of solution for animal oral treatment.

### Experimental design/procedure

This study was designed to investigate the antioxidant and lipid effects of the leaf extract of *M. charantia*. Male wistar rats were divided into 3 groups of 6 rats each. Group one (1) which served as control

received distilled water. Group 2 received 200mg/kg bw and group 3 received 400mg/kg bw of the hydromethanol leaf extract of *M. charantia*. The extracts were administered orally as single daily doses throughout the period of the experiment using appropriate animal feeding tube. Administration lasted for 30 days. The rats were sacrificed under chloroform anaesthesia, after 24hours of last administered dose.

For estimation of serum lipid profile [Total cholesterol (TC), Triglyceride, High density lipoprotein-cholesterol (HDL-c)], blood was collected through cardiac puncture into appropriate sample tubes. After collection, the samples were centrifuged for twenty minutes in a serologic manual centrifuge. Then, 1 mL aliquots of serum were removed and used to perform the biochemical assay. In analysing biochemical parameters, commercial Labtest Diagnostic kits were used, with standard techniques based on enzymatic and colorimetric methods, spectrophotometry, in accordance to the manufacturer's recommendations. The concentrations were determined by use of an automatic biochemical analyzer. The determination of Low density lipoprotein-cholesterol (LDL-c) was calculated using the Friedewald formula. The plasma was used for the estimation of oxidative stress biomarkers in accordance with standard methods [15-20].

## STATISTICAL ANALYSIS

For the statistical analysis, SPSS software, version 20.0 was used. We calculated the Mean and standard error of mean (SEM). In the comparison of the mean level of serum lipids and plasma antioxidants between the control and test groups, we used the analysis of variance (ANOVA) and then post hoc test least significant difference. For all crosses whose probability (*P*) value was less than 0.05 were considered statistically significant.

## RESULT

The result for the study is presented in tables 1-4.

**Table-1: Mean level of Gluthathione and Superoxide dismutase activity.**

Groups/ Extracts (mg/kg)	Oxidative stress markers			
	Gluthathione (µg/min/mg.protein)	% Change	SOD (Ug/mg.protein)	% Change
Control	0.88±0.04	0	0.12±0.01	0
200	0.91±0.11	3.41	0.13±0.01	8.33
400	1.37±0.16*	55.68	0.14±0.01	16.66

Values expressed as Mean ± SEM. n=6. Significant at [\*(P<0.05)] when compared to control group

**Table-2: Mean level of Malondialdehyde and Catalase activity**

Groups/ Extracts (mg/kg)	Oxidative stress markers			
	MDA (Umol/mg.protein)	% Change	Catalase (Units/mg.protein)	% Change
Control	0.72±0.02	0	3.85±0.55	0
200	0.63±0.01*	-12.50	3.56±0.37	-7.53
400	0.68±0.03	-5.56	7.08±0.83*	83.90

Values expressed as Mean ± SEM. n=6. Significant at [\* (P<0.05)] when compared to control group

**Table-3: Effect of leaf extract of *M.charantia* on the serum lipid profile**

Groups/ Extracts (mg/kg)	Serum lipid profile (mg/dl)			
	Total cholesterol	Triglyceride	LDL-c	HDL-c
Control	3.20±0.21	1.10±0.08	1.29±0.06	1.33±0.08
200	3.33±0.32	0.89±0.08	1.25±0.08	1.55±0.17
400	4.13±0.66	1.11±0.15	1.19±0.17	1.35±0.07

Values expressed as Mean ± SEM. n=6.

**Table-4: Mean values of the lipid ratio**

Groups/ Extracts (mg/kg)	Lipid ratio (mg/dl)					
	TG/ HDL-c	TC/ HDL-c	LDL-c/ HDL-c	LDL-c/ TC	LDL-c/ TG	HDL-c/ LDL-c
Control	0.83±0.06	2.41±0.13	0.99±0.08	0.42±0.04	1.22±0.14	1.05±0.08
200	0.63±0.11	2.29±0.31	0.91±0.19	0.40±0.05	1.46±0.15	1.28±0.19
400	0.82±0.09	3.04±0.40	0.91±0.17	0.32±0.07	1.15±0.22	1.44±0.43

Values expressed as Mean ± SEM. n=6.

## DISCUSSION

The assessment of antioxidant properties of a plant extract is an interesting and useful task, as it is particularly concerned with finding potential new sources of natural antioxidants that acts as functional food products. In this study, there was a significant ( $p<0.05$ ) increase in the activity of glutathione reductase enzyme (Table 1). The superoxide dismutase enzyme activity was not significantly affected but the catalase (Table 2) enzyme activity was increased for the group treated with the higher dose (400mg/kg) of *Momordica charantia* extract. This can be observed in the result when the measured enzyme activity in the test group is compared to control. The increase in these enzyme activities implies that leaf extract of *Momordica charantia* are capable of boosting the antioxidant production in the experimental animals.

The Malondialdehyde (MDA) level was significantly ( $p<0.05$ ) reduced in the group treated with 200mg/kg of extract (Table 2) when test groups were compared to control. The lower dose (200mg/kg) and higher dose (400mg/kg) of *Momordica charantia* extract caused a 12.50% and 5.56% reductions in MDA level, respectively. MDA is a product of lipid peroxidation. Lipid peroxidation is considered a useful indicator of reactive oxygen species-mediated damage to cell membranes. MDA is an important end-product of peroxidation of polyunsaturated fatty acids and is frequently used in determination of oxidative stress conditions [21]. Oxidative stress often results from an imbalance between the production of reactive oxygen

species and ability of the antioxidant systems to detoxify them.

The antioxidant property of many plants has been attributed to the presence of certain phenolic compounds such as flavonoids and phenolic acids which are reportedly main anti-oxidative compounds of fruits and vegetables. Phenols, flavonoids and tannins are good antioxidants useful in prevention or control of oxidative stress and related disorders [22, 23]. Although, we did not carry out a phytochemical analysis of the extract, but these compounds were discovered in the leaf extract of *Momordica charantia* in an earlier study [24], and may be responsible for its antioxidant actions. The ability of *Momordica charantia* to inhibit lipid peroxidation may be due to a free radical scavenging action which prevented further peroxidation and possible damage to cell.

In this study, there was no significant ( $p<0.05$ ) change in the level of serum total cholesterol, LDL-c, HDL-c and triglyceride concentrations, when test groups were compared to control. The findings in this study do not agree with the report showing that extract of *Momordica charantia* reduced triglyceride and LDL-c while increasing HDL-c in animal models [25]. The differences in findings may be due to the different methods of extraction used.

The lipid profile is usually considered in the evaluation of dyslipidemia. Interestingly, the extract neither affected the LDL-c whose elevation in serum is considered atherogenic, nor causes an increase in HDL-

c which is regarded as cardioprotective [26, 27]. Although, in a human study [28] on the effects of *Momordica charantia* on lipid profile, there was a reported significant decrease in LDL-c but a non-significant change in TC, HDL-c and triglyceride concentrations.

There was no significant ( $p < 0.05$ ) alterations of the Castelli's risk index-I (CRI-I) [TC/HDL-c] and Castelli's risk index-II (CRI-II) [LDL-c/HDL-c] (Table 4) as well as other cardiovascular risk indices in this study. But a high CRI-I and CRI-II has been associated with higher risk of Ishaemic heart disease. These lipid ratios are often used as an alternative diagnostic tool to predict risk of developing cardiovascular disease [29], even when conventional lipid parameters appear normal.

## CONCLUSION

The leaf extract of *Momordica charantia* has the potential to inhibit lipid peroxidation due to the reduction in production of malondialdehyde. The extract did not exert significant effects on the lipid parameters and lipid ratios such as the Castelli's Risk Index I and II.

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