American Journal of Biomedical Research, 2016, Vol. 4, No. 2, 38-41
Available online at http://pubs.sciepub.com/ajbr/4/2/2

DOI:10.12691/ajbr-4-2-2

Modulatory Effects of *Ricinus Communis* Leaf Extract on Cadmium Chloride-Induced Hyperlipidemia and Pancytopenia in Rats

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Abstract Cadmium (Cd) and its compounds are ubiquitous environmental toxins capable of inducing different types of toxicity in animals and human. *Ricinus communis* (castor bean) is a known medicinal plant with enormous health benefits. This study investigated the protective effects of ethanolic leaf extract of *Ricinus communis* on hyperlipidemia and pancytopenia induced by cadmium chloride in Wistar albino rats. Twenty five (25) adult male rats were divided into 5 groups of 5 rats each. Group A received distilled water, group B, C, D and E were administered 5mg/kg body weight $CdCl_2$, group C, D and E were treated with 250, 500 and 1000 mg/kg bw respectively of *Ricinus communis* leaf extract for 14 days while rats in group B were left untreated. Results obtained showed that administration of cadmium caused significant reduction in cellular elements of the blood (pancytopenia) as well as increase in the level of serum total cholesterol, triglycerides, LDL-cholesterol and coronary heart disease risk ratio while it decrease the level of HDL-cholesterol. Treatment of rats with graded dose of *Ricinus communis* leaf extract significantly ameliorate the adverse effects of cadmium as it boosted cellular elements of the blood while it also reduced serum total cholesterol, triglycerides, LDL-cholesterol, coronary heart disease risk ratio as well as elevation of HDL-cholesterol level in the blood. We conclude that leaf extract of *Ricinus communis* has modulatory effects on cadmium induced pancytopenia and hyperlipidemia in rats.

Keywords: *pancytopenia, hyperlipidemia, cadmium chloride, cholesterol, Ricinus communis*

Cite This Article: Olu Israel OYEWOLE, Mojisola Oluwakemi SHOREMI, and Johnson Olaleye OLADELE, "Modulatory Effects of *Ricinus Communis* Leaf Extract on Cadmium Chloride-Induced Hyperlipidemia and Pancytopenia in Rats." *American Journal of Biomedical Research*, vol. 4, no. 2 (2016): 38-41. doi: 10.12691/ajbr-4-2-2.

1. Introduction

Environmental contaminants and xenobiotics play vital role in the etiology and pathogenesis of many disease conditions including anaemia, diabetes, neurodegenerative diseases and cardiovascular disorders. Cadmium (Cd) and its compounds are ubiquitous environmental toxins which can get in contact with man through industrial exposure and via food, particularly through leafy vegetables, grains and cereals. Cd and its compounds are toxic to several tissues, acute and chronic exposure to these compounds has been reported to induce hepatotoxicity and nephrotoxicity [1]. There are myriad evidences supporting the carcinogenicity of Cd and its compounds in human [2].

The act of using plants as medicines predates written human history. These medicines took the form of crude drugs such as tinctures, teas, poultices, powders and other herbal formulations [3]. Among these medicinal plants is Castor bean (*Ricinus communis* L.), a member of Euphorbiaceae family and an important medicinal plant of Africa [4]. *Ricinus communis* leaf is used traditionally as laxative, analgesic, antipyretic, cardiac tonic, anti asthamatic and also as purgative [5].

Sufficient data from previous experimental studies has proven the antimicrobial [6], antioxidant [7], anti-inflammatory [8], anti-tumour [9], anti-nociceptive [10] and insecticidal [11] activities of *Ricinus communis* leaf. In the present study, we investigated the protective effects of *Ricinus communis* leaf extract on cadmium chloride-induced hyperlipidemia and pancytopenia in male Wistar albino rats.

2. Materials and Methods

2.1. Chemicals/ Reagents

Lipid profile kits used (total cholesterol, triglyceride, HDL-cholesterol and LDL-cholesterol) are products of Randox Laboratories Limited, United Kingdom. All other chemicals are of analytical grade and were obtained from Analar BDH Limited, Poole, England.

2.2. Collection of Plant Material and Preparation of Crude Extract

Fresh leaves of *Ricinus communis* were collected at Isale Aro Area, Osogbo, Nigeria. The samples were air dried for 2 months after which it was pulverized in to

powdery form using industrial laboratory grinder. Extraction of the phytochemicals was done by dissolving 700g of the powder in 4.2litres of 98% absolute ethanol for 14 days after which the extract was filtered using a white moslin cloth. Crude extract was obtained by filtration followed by evaporation of the solvent in a rotatory evaporator at 80°C. The paste was weighed and used to prepare stock solution and different doses of the extract.

2.3. Experimental Design and Animal Grouping

Twenty five male Wistar albino rats (average weight 150g) were used for this experiment. They were obtained and housed in the Central Animal House, Osun State University, Osogbo Nigeria. The rats were kept in ventilated cage at optimum temperature, 12hrs light / dark cycle and fed with rat pellet and water *ad libitum*. The experiment was carried out in accordance with existing guidelines for the care of laboratory animals [12]. The rats were acclimatized for 2 weeks before administration commenced. Rats were sorted into five (5) different groups containing five (5) rats each. Group A received distilled water, group B, C, D and E were administered 5mg/kg body weight $CdCl_2$, group C, D and E were treated with 250, 500 and 1000 mg/kg bw respectively of *Ricinus communis* leaf extract for 14 days while rats in group B were left untreated. The average weight of each group were taken and recorded daily. Administration of extract was done using the oral canula.

2.4. Collection of Whole Blood and Serum

Rats were sacrificed by cutting through the jugular vein and whole blood for haematological analysis collected into labeled EDTA bottles to prevent clotting. Serum for lipid profile analysis was collected into plain bottles and allowed to clot after which it was centrifuged at 4000rpm for 30mins. The serum obtained was stored in a refrigerator at -4°C.

2.5. Estimation of Haematological Parameters

Haematological parameters including haemoglogbin concentration (Hb), red blood cell count (RBC), white blood cell count (WBC), packed cell volume (PCV), mean cell haemoglobin concentration (MCHC), mean cell volume (MCV), lymphocyte, platelet counts and reticulocyte count were measured in the whole blood using the automated multiparameter blood analyzer SYSMEX KX21 as earlier described [13]. Fifty microlitres of blood samples were introduced into the equipment and it automatically employ the differences in characteristics possessed by each of the blood components to distinguish and estimate them.

2.6. Measurement of Serum Lipid Profile

Total cholesterol, triglyceride, HDL-cholesterol and LDL-cholesterol were measured in the serum of individual rats using the appropriate methods. Total cholesterol was determined by the enzymatic endpoint method [14]. Triglycerides was assayed using the GPO-PAP method [15] while precipitant method [16] was used in the measurement of HDL cholesterol. LDL cholesterol was estimated using the procedure earlier described [17]. Coronary heart disease risk ratio (CHD risk ratio) was obtained by calculating the ratio of concentrations of total cholesterol to HDL-cholesterol. Measurement of concentrations were done by the use of Camspec M106 UV spectrophotometer (Ohaus Corporation Pine Brook USA).

2.7. Statistical Analysis

Data were expressed as mean ± standard deviation (mean ± SD) and analyzed using one-way analysis of variance (ANOVA) with the aid of SPSS 12.0 computer software package (SPSS Inc; Chicago, U.S.A). Student's t-test was employed for comparison between two sets of data and differences at $P<0.05$ were considered significant.

3. Results

Table 1 show the haematological indices in rats administered cadmium chloride and *Ricinus communis* leaf extract. Rats administered cadmium without treatment recorded significant reduction in haematological indices (PCV, RBC, WBC, Hb, platelets, MCV and MCHC) which were all boosted upon treatment with the extract. Reticulocyte and lymphocyte counts in treated and untreated groups (groups B-E) were however not significantly different from that of the control group (group A).

Serum lipid profile in the experimental animals is shown in Table 2. Cadmium chloride administration caused significant elevation in total cholesterol, triglycerides, LDL-cholesterol and coronary heart disease risk ratio while it also reduced HDL-cholesterol in the rats. These anomalies were however annulled in rats treated with graded dose of *Ricinus communis* leaf extract.

Table 1. Haematological indices in rats administered cadmium chloride and *Ricinus communis* leaf extract

Parameters	Control	Cd Only	Cd +250 mg/kg bw RCE	Cd +500 mg/kg bw RCE	Cd +1000 mg/kg bw RCE
Hb Conc (g/dl)	13.22±2.17	9.68±1.76*	11.50±2.06	11.25±1.89	12.39±2.00
PCV (%)	26.90±3.60	19.80±2.56*	25.10±3.00	24.80±2.78	25.00±3.02
RBC (10^{12}/L)	6.12±0.16	4.33±0.43*	5.88±0.31	6.00±0.28	5.96±0.22
WBC (10^9/L)	12.35±2.18	9.04±1.69*	12.41±2.22	11.87±1.89	12.26±2.07
Platelet (10^9/L)	551.5±12.55	408.5±15.32*	543.5±20.47	536.4±18.38	549.5±12.40
MCV (fl)	68.55±5.24	53.77±4.72*	67.22±6.01	68.01±6.44	66.35±5.42
MCHC (g/dl)	36.23±3.12	28.82±2.27*	34.46±2.82	35.91±3.34	33.65±3.20
Lymphocyte (%)	21.32±3.60	22.16±2.55	22.90±3.06	21.84±2.88	24.16±2.25
Reticulocytes (%)	13.53±2.76	13.56±1.60	12.88±2.02	13.24±1.75	13.44±2.07

Values are mean ± SD; n=5. *Significantly different from normal control group at $p<0.05$.
Cd=Cadmium chloride, RCE=Ricinus communis extract.

Table 2. Serum lipid profile in rats administered *Ricinus communis* leaf extract and cadmium chloride

Parameters (mg/dl)	Control	Cd Only	Cd +250 mg/kg bw RCE	Cd +500 mg/kg bw RCE	Cd +1000 mg/kg bw RCE
Total Cholesterol	138.66±9.33	160.43±8.64*	142.21±9.71	148.32±8.87	139.72±8.31
Triglycerides	91.58±6.46	112.38±7.50*	98.63±6.34	93.45±5.98	94.75±7.33
LDL Cholesterol	76.45±4.58	89.62±6.04*	81.33±4.63	78.85±5.33	80.37±4.76
HDL-Cholesterol	55.21±3.89	45.37±2.41*	51.42±3.27	57.50±4.11	53.56±4.05
CHD risk ratio	2.51±0.28	3.54±0.43	2.77±0.31	2.58±0.26	2.61±0.33

Values are mean ± SD; n=5. *Significantly different from normal control group at
Cd=Cadmium chloride, RCE = Ricinus communis extract.

4. Discussion

Results obtained in this study (Table 1) indicate that exposure of rats to cadmium chloride resulted into pancytopenia, a medical condition characterized by general reduction in blood cellular components. This reduction in blood cellular components could be as a result of toxicity and stress induced by cadmium on haematopoiesis in the rats. Previous studies have implicated acute stress in animals as a factor in the pathogenesis of blood toxicity/diseases [18]. The reduction in cellular components by cadmium could also be attributed to diminished production, redistribution from peripheral blood into the tissues or rapid destruction of blood cells [19].

Administration of *Ricinus communis* leaf extract in this study significantly increased the PCV, RBC, WBC, Hb, platelets, MCV and MCHC values which suggests a net beneficial effect on erythropoiesis. This protective role elicited by *Ricinus communis* may be due to its bone regeneration activity as previously reported [20,21]. The observe increase in MCV and MCHC is synonymous with red blood cell hydration while the observed increase in total WBC count by the extract suggests that it might be immunoprotective. However, the significant increase in platelets count might however promote vaso-occlusion in the blood vessel of the rats administered the extract [22]. Taken together, these results suggested that *Ricinus communis* possess heamoprotective properties.

The observed significant increase ($P<0.05$) in the level of serum total cholesterol, triglycerides, LDL, coronary heart disease risk ratio and a significant decrease ($P<0.05$) in the level of HDL in the serum of cadmium treated rats (group B) as compared to normal control (group A) is an evidence of hyperlipidemia. This could be due to the peroxidation of membranes and alteration of cellular structure by cadmium chloride. High levels of LDL-cholesterol promote health problems and cardiovascular disease, they are often called “bad cholesterol” as opposed to HDL particles, which are referred to as “good cholesterol” or “healthy cholesterol” [23,24]. HDL particles are able to remove cholesterol from within the artery and transport it back to the liver for excretion or re-utilization [25]. Those with higher levels of HDL-cholesterol seem to have fewer problems with cardiovascular diseases, while those with low HDL cholesterol levels have increased rates of heart disease [26]. *Ricinus communis* leaf extract demonstrated hypolipidemic activity as it significantly reduced triglyceride, cholesterol, LDL, coronary heart disease risk ratio and increased HDL in rats serum presenting the herb as a candidate drug in the treatment of hyperlipidemia and cardiovascular diseases [27]. This might explain the traditional use of the leaf extract as a natural remedy against heart diseases in the West Africa Sub Region. The hypolipidemic and blood boosting activities of the leaf can be ascribed to its phytochemical constituents which are tannins, phlobatannins, flavonoids, steroids, terpenoids, saponins and cardiac glycosides as earlier reported [28,29]. Some of these phytochemicals have been reported to have positive physiological actions on haematopoiesis and lipid metabolisim in animals and human [30].

5. Conclusion

Results obtained in this study clearly demonstrate that *Ricinus communis* leaf extract has positive modulatory effects on hyperlipidemia and pancytopenia as it annulled dyslipidemia and haematoxicity caused by cadmium in rats. The usefulness of *Ricinus communis* leaf as traditional remedy for boosting blood and treatment of cardiovascular diseases is hereby justified in this study.

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