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Research Article

EFFECTIVENESS OF *CYPERUS ROTUNDUS* IN COUNTERING ATHEROGENIC DIET INDUCED CHANGES IN LIPID PROFILE AND METABOLISM BY NORMALISING THE PLASMA ADIPONECTIN CONCENTRATION

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ABSTRACT

Obesity-associated with Cardio vascular properties is rapidly increasing throughout the world. It is generally recognized that natural products with a long history of safety can modulate obesity. To investigate the development of obesity in response to an atherogenic diet and to estimate the effect of *Cyperus rotundus Linn* on bodyweight, body temperature, loco motor activity, organ and fat pad weights, plasma Adiponectin, serum lipid profiles, renal, hepatic, cardiac function markers, ECG, Heart rate and Blood pressure in wistar rats. Results showed that feeding of atherogenic diet significantly increased final body weight, fat pad weights, glucose, triglycerides, total cholesterol, & LDL concentration compared with controls, while significantly decreasing HDL and Plasma Adiponectin; meanwhile treatment with *Cyperusr otundus* significantly normalized the glucose, lipid profile. And Plasma Adiponectin, Serum ALT, AST, urea, uric acid, creatinine, LDH, CK-NAC, and CK-MB were significantly higher in an atherogenic diet group compared with normal controls; and administration of *Cyperus rotundus* significantly lessened the effect of the atherogenic diet. BP increased in an atherogenic diet group in comparison with the control group. The treatment with *Cyperus rotundus* normalized the condition. Atherogenic diet induced obesity associated with a disturbed lipid profile (TGs, TC, LDL, HDL), renal (Urea, Uric acid, Creatinine), liver (AST, ALT) and cardiac biomarkers (LDH, CK-NAC, CK-MB), Plasma Adiponectin and cardiac parameters (ECG, BP, Heart rate); this may have implications for the progress of obesity related problems. Treatment with *Cyperus rotundus* extract improved obesity and its associated metabolic problems in different degrees. Moreover *Cyperus rotundus* linn might be a safe drug on the organs whose functions were examined, as a way to surmount the obesity state; and it has a distinct anti-obesity effect.

Key words: Atherogenic diet, Lipid Profile, Renal Profile, Adiponectin, Cardiac biomarkers.

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INTRODUCTION

Obesity is a global problem and it is a chronic, relapsing, stigmatized neuro-chemical disease and it is more prevalent in developing/developed countries, such as socioeconomic changes have taken place. Prevalence of obesity in Indian population is 20% in adults and 10% in

children in Punjab state of India (Srivastava N *et al.,* 2007). International Obesity Task Force (IOTF) estimated that there are over 300 million people worldwide who would be classified as clinically obese having a body mass index (BMI) (Paul P *et al.,* 2006). A continuously rising trend in childhood and adolescence has been noted in several studies, associated with rise in obesity rates presage a dire future for these children as complication of blindness, heart disease, renal failure, amputation and compromised quality of life (Chen H, 2006). It has been well established that nutrition plays an important role in the etiology of hyperlipidemias, atherosclerosis and other coronary heart disease (CHD) complications like myocardial infarction (Shekar HS *et al.,* 2017). The etiology and pathogenicity of coronary heart diseases in the casual relationship between the development of atherosclerosis, elevated plasma lipid percentage cholesterol levels in blood and plasma, genetic makeup, endocrinological aberration, immunologic and autonomic factors, blood flow and coagulation(Angeles Z *et al.,* 1999).

A recent survey, carried out by world health organization (WHO), indicates that CHD alone accounts for more than half of the total mortalities associated with cardiovascular diseases (Raja C *et al*., 1996). Several well-recognized risk factors contribute to the development of CHD; include hypertension, smoking, diabetes, hyperlipidemia, current cigarette smoking, and a family history of premature CHD. In the last decade, however, cholesterol has emerged as an independent risk factor for the development of CHD in the elderly population (Sanjay B et al., 2000).

To reduce the rate of mortality, it is therapeutically recommended to undergo diet or/and drug therapy to lower lipid levels within the normal range. Therapy of hyperlipidemia merits the consideration in the established atherosclerotic state. Allopathic hyperlipidemia drugs are available at large in the market but the side effects and contraindications of these drugs have marred their popularity (Scott M *et al.,* 1999). The herbal hypolipidemics have gained importance to fill the lacunae created by the allopathic drugs.A number of plants

have been found to be useful in hyperlipidemia. Condiments like garlic, onion and coriander, adraka used in day to day preparation of food in Indian kitchens have been identified as hypolipidemics in Ayurveda (Annon, 1999). And few of allopathic drugs areAjamoda, Banana, and Meshashringi, Lemon grass oil, Kala isabgol, Tulsi, guggul, Bhumiamalkietc. From the literature available the herb *Cyperus rotundus Linn* has found to show various degrees of antihypertensive, anti-obesity, anti hyperlipidemic activities besides many other beneficial pharmacological properties of interest. It is also known fact that adulthood obesity depicts many co-morbid conditions such as hypertension, diabetes, and hyperlipidemia.

Cyperus rotundus Linn. (Family Cyperaceae), commonly known as *mustaka*, is a pestiferous perennial weed with dark green glabrous culms, arising from a system of underground tubers found throughout India (Anonymous, 1999; Nadkarni KM. and Nadkarni AK, 1996). The tubers are useful as infusion or as a soup in fever, diarrhea, dysentery, dyspepsia, vomiting and cholera. Fresh tubers are applied on the breast in the form of paste or plaster as galactagogu. The acetone and ethanol extracts of tubers were found to possess anti-bacterial activity (Puratchikody A *et al*., 2001). It is one of the plants mentioned in the literature having claims of activity against liver disorders (Pan Q and Lix, 1985). The tubers of the plant are used as anthelmintic, antihistaminic, antiemetic, antipyretic, hypotensive, smooth-muscle relaxant and emmenagogue in uterine complaint (Nadkarni KM and Nadkarni AK, 1996). The plant has also been reported to have antimalarial, tranquillizing, hepato-protective against carbon tetrachloride induced liver damage, lipolytic action and reduced obesity by releasing enhanced concentration of biogenic amines from nerve terminals of the brain, which suppressed the appetite Centre (Gupta AK, 2003). The plant has also been reported to have antimalarial; tranquillizing action as well as hepato-protective action against carbon tetrachloride induced liver damage. It is said to have lipolytic action and also property that helps reduce obesity by releasing enhanced concentration of biogenic amines from nerve terminals of the brain that suppress the appetite centre[15]. It is also reported to have anti-inflammatory activity (Anonymous, 1992). It contains a wide variety of phytoconstituents that are useful in the treatment of different ailments and includes sesquiterpene 4a-, 5a-, oxidoeudesm-11-en-3a-ol, cyperene-1 (a tricyclic sesquiterpene), cyperene-2 (a bicyclic sesquesterpene hydrocarbon), cyperenone, and a-cyperone, mustakone (a new sesquesterpene ketone), β -selinene, sugetriol triacetate (a new sesquesterpenoid), sugenol (sesquesterpenicketol) (Rastogi RP and Mehrotra BN, 1969); the essential oil including copadiene, epoxyguaienerotundone, cyperenol, cyperolone, eugenol, cyperol, isocyperol, a-and β -rotunol, kobusone, isokobusone, d-cadinene and calamenone; a flavonol glycoside, rhamnetin 3-O-rhamnosyl-1-4rhamnopyranoside and β –sitosterol.

Hence, it is envisaged to undertake to study effects of aqueous extracts of tubers of *Cyperus rotundus Linn*on hyperlipidemia and on metabolic indicators of CVS.

MATERIALS AND METHODS

Chemicals, Drugs, Plant material and Preparation of the extract

The aqueous extract of *Cyperus rotundus* was obtained from PUKHRAJ HERBALS, 15-16, Industrial area, Mandasaur (M.P). Batch No: NGMT-01/DEC 09.Cholesterol from SD Fine-chem limited; Cholic acid and Lard oil from LOBA CHEMIE obtained. CK-MB, CK-NAC, lactate dehydrogenase (LDH-P), Serum Glutamate Oxaloacetate Transaminase (SGOT), and Serum Glutamate Pyruvate Transaminase (SGPT), Urea, Uric acid, Creatinine, Cholesterol, Triglyceride, HDL and Glucose were procured from Preicugent, Maharashtra.

Dose Selection

In the present study two doses of the aqueous extracts of tubers of *Cyperus rotundus Linn*., were selected one being the higher dose (50mg /kg body wt) the other being the lower dose(30mg /kg body wt) (Malhotra SC, 1996). The doses of extracts were administered by per oral route in morning throughout the study period.

Experimental Design

Atherogenicdiet induced obesity

Experimental study was carried out using adult Male Wistar rats weighing between 150-170g. The animals were housed in polypropylene cages of dimension 16"×9"×7". The cages were maintained under clean and hygienic conditions. Animals were acclimatized to light and temperature with a 12h-12h dark-light cycle; the rats were fed with commercial pelleted rat feed and water *ad libitum.*The approval of Institutional Animal Ethics Committee (IAEC/NCP/40/10) was taken prior to the commencement of the studyin accordance with animal experimentation and care guidelines provided by IAEC/CPCSEA.

Animals were divided into 5 groups each group consisting of 6 animals.

Group 01: Control (fed with normal pellets chows)

Group 02: Extract (plant extract (50mg/kg b.w.*p.o.*) with normal pellets)

Group 03: Atherogenic diet

Group 04: Atherogenic diet + extract (High dose 50mg/kg b.w.*p.o.*)

Group 05: Atherogenic diet + extract (Low dose 30mg/kg b.w.*p.o.*)

Composition of the Atherogenic diet

1% Cholesterol (Sd fine-chem limited), 0.5% Cholic acid (LOBA CHEMIE), 5% Lard oil (Open Market). These diets were provided in addition to normal pellet chow (Jiao S *et al*., 1991).

Treatment Protocol

Induction of obesity in the experimental animals was carried out by feeding the animals with the atherogenic diet.

The Aqueous extract of *Cyperus rotundus* was administered orally in a dose of 50mg/kg,*p.o.* (High dose), 30mg/kg,*p.o*. (Low dose) daily for the entire study period. 24hrs before the sacrifice of the study animals, they were kept on fast but they had access to water.The blood samples were collected in eppendroffs tubes by puncturing the retro orbital plexusfor biochemical determinations. Then experimental animals were sacrificed and the aorta, coronary artery, liver and the kidney were collected and were kept in 15% V/V formalin solution for histopathological examination.

PARAMETERS

Normal parameters

The body weight (gm.) was recorded on day 1 and then on alternate days during the study period using an electronic balance. The body temperature was recorded for all groups of animals on day 1 and on day 39 using rectal digital tele-thermometer (yellow springs instruments Co. inc., USA) before and after drug administration at 30.60, 90, 120, and 180 min with a contact time of 1 minute.

Loco motor activity was recorded on day 40 using open field behavior test apparatus and 30 min after the administration of extracts of *Cyperousrotundus* tubers to the treatment groups. The apparatus consisted of a circular wooden arena of 75 cm diameter and wall with a height of 25 cm. Open field test was performed by placing the rat in the center circle and recording the Ambulatory activity, the frequency of Rearing and Grooming for a 5 min test period.

Biochemical parameters

Bio chemical parameters like Total Cholesterol, Triglyceride level, HDLwere analyzed by using auto biochemistry analyzer manufactured by ROBONIK PVT.LTD, Mumbai.

Organ and fat pad weights

The animals were sacrificed by cervical dislocation and then different organs (kidney, liver, heart, and spleen) and fat pads (mesenteric, left and right perirenaland uterine fat pads) were removed and weighed.

Haemodynamic parameter

Haemodynamic parameters like Blood pressure, Heart rate; ECG was monitored on day 40. For monitoring blood pressure a cannula was inserted into the left common carotid artery of 6 animals per group and connected to a pressure transducer. Changes in electrical activity of the myocardium were detected by the electrocardiogram (ECG) in lead II (INCO NIVIQURE DIGITAL ECG SYSTEM). All data obtained for each module of the system were analyzed with computer software AmbECG Ver.52 (INCO SOFTWARE).

Metabolic Indictors

LDH, CK-NAC, CK-MB, AST, ALT, Urea, Uric acid, Creatinine investigations were carried out by usingAuto biochemistry analyzer and centrifuge.

Plasma adiponectin

The plasma adiponectin was analyzed by using the ELISA kit; it is a sandwich enzyme immunoassay for the in vitro quantitative measurement of rat ADP in plasma.

Statistical Analysis

All results are expressed as mean±SEM. Statistical analysis was performed using the Graph pad prism 5, Graph pad software. One-way ANOVA followed by Dunnett's test was performed.

RESULTS

AD group showed a significant (P<0.001) increase in body weight (g) when compared to control group. *CR* (50mg/kg) extract group showed a decrease in body weight (g) non-significantly when compared to control group. *CR* (50mg/kg) treated group showed a significant decrease (P<0.001) *CR* (30mg/kg) treated group showed a significant decrease (P<0.01) in body weight when compared to AD group.

The animals treated with CR extract 50 mg/kg body wt, AD + CR extract at two dose levels (30 mg/kg, 50 mg/kg) showed significant rise in body temperature as compared with to animals in control and AD fed group respectively.

AD group showed a significant decrease (P<0.01) in ambulation activity when compared to control group.*CR* (50mg/kg) extract group showed a non-significant increase in ambulation activity when compared to control group.*CR* (50mg/kg) treated group showed a significant increase (P<0.001) in ambulation activity when compared to AD group.*CR* (30mg/kg) treated group showed a significant increase (P<0.001) in ambulation activity when compared to AD group.

AD group showed a significant increase (P<0.001) in rearing activity when compared to control group.*CR* (50mg/kg) extract group showed a significant decrease (P<0.05) in rearing activity when compared to control group.*CR* (50mg/kg) treated group showed a significant decrease (P<0.001) in rearing activity when compared to AD group. *Cyperus rotundus* (30mg/kg) treated group showed a significantdecrease (P<0.05) in rearing activity when compared to AD group.

AD group showed a significant decrease (P<0.05) in grooming activity when compared to control group.*CR* (50mg/kg) extract group showed a significant increase (P<0.05) in grooming activity when compared to control group.*CR* (50mg/kg) treated group showed a significant increase (P<0.001) in grooming activity when compared to AD group.*CR* (30mg/kg) treated group showed a significant increase (P<0.01) in grooming activity when compared to AD group.

AD group showed a significant (P<0.001) increase in organ weight of spleen, liver, right and left kidney and significant (P<0.05) increase heart weight when compared to control group.*CR* (50mg/kg) extract group showed a decrease in organ weight of spleen, right and left kidney, liver non significantly and no changes observed in organ weight of heart when compared to control group.*CR* (50mg/kg) treated group showed a significant decrease (P<0.001) in organ weight of spleen, liver, left kidney and significant decrease (P<0.05) in organ weight of right kidney, heart when compared to AD group.*CR* (30mg/kg) treated group showed a decrease in organ weight of spleen, heart, liver, right and left kidney which is not significant, when compared to AD group.

AD group showed a significant (P<0.001) increase in fat pad weight of mesenteric, left and right Perirenal weight when compared to control group.*CR* (50mg/kg) extract group showed a significant (P<0.05) decrease in fat pad weight of Perirenal and significant (P<0.01) decrease mesenteric weight non-significantly when compared to control group.*CR* (50mg/kg) treated group showed a significant decrease (P<0.001) in fat pad weight of mesenteric, left and right perirenal when compared to AD group.*CR* (30mg/kg) treated group showed a significant decrease (P<0.001) in fat pad weight of left and right Perirenal and significant (P<0.05) decrease mesenteric fat pad when compared to AD group.

AD group showed a significant (P<0.001) increase in serum glucose, total cholesterol, triglyceride LDL level when compared to control group.AD group showed a significant (P<0.05) decrease in serum HDL-C level when compared to control group.*CR*(50mg/kg) extract showed a decrease in serum total cholesterol and HDL-C which is not significant and increased glucose and triglyceride level non-significantly when compared to control.*CR*(50mg/kg) treated group showed a significant (P<0.001) decrease in total cholesterol, LDL and significant (P<0.01) decrease in glucose and significant (P<0.05) decrease in triglyceride level when compared to AD group.*CR* (50mg/kg) group showed a significant (P<0.01) increase in serum HDL-C level, when compared to AD group.*CR* (30mg/kg) group showed a significant (P<0.01) decrease in serum total cholesterol and decreased glucose and triglyceride level non-significantly and significant (P<0.001) decrease when compared to AD group.*CR* (30mg/kg) treated group showed an increase in HDL-C level, which is not significant when compared to AD group.

AD group showed a significant (P<0.001) increase in serum LDH, CK-NAC, CK-MB, AST, ALT, Urea, Uric acid and Creatinine levels when compared to control group. *CR* (50mg/kg) extract showed an increase in serum LDH, CK-NAC, CK-MB which is not significant when compared to control group. *CR* (50mg/kg) extract

showed a decrease serum AST, ALT, Urea, Uric acid and Creatinine levels which is not significant when compared to control group. *CR* (50mg/kg) treated group showed a significant (P<0.001) decrease in serum CK-MB, AST and Urea when compared to AR group. *CR* (50mg/kg) treated group showed a significant (P<0.01) decrease in serum LDH, CK-NAC and Uric acid when compared to AD group. *CR* (50mg/kg) treated group showed a significant (P<0.05) decrease in serum ALT and Creatinine levels when compared to AD group. CR (30mg/kg) treated group showed a significant (P<0.01) decrease in serum CK-MB and significant (P<0.05) decrease in serum CK-NAC and Urea level when compared to AD group. *CR* (30mg/kg) treated group showed a decrease in serum LDH, ALT, Uric acid, Creatinine which is not significant when compared to AD group. *CR* (50mg/kg) extract showed a decrease serum AST, ALT, Urea, Uric acid and Creatinine levels which is not significant when compared to control group. *CR* (50mg/kg) treated group showed a significant (P<0.001) decrease in serum CK-MB, AST and Urea when compared to AD group. *CR* (50mg/kg) treated group showed a significant (P<0.01) decrease in serum LDH, CK-NAC and Uric acid when compared to AD group. *CR* (50mg/kg) treated group showed a significant (P<0.05) decrease in serum ALT and Creatinine levels when compared to AD group. CR (30mg/kg) treated group showed a significant (P<0.01) decrease in serum CK-MB and significant (P<0.05) decrease in serum CK-NAC and Urea level when compared to AD group. *CR* (30mg/kg) treated group showed a decrease in serum LDH, ALT, Uric acid, Creatinine which is not significant when compared to AD group.

AD group showed a significant (P<0.001) increase in Mean arterial blood pressure when compared to control group.*CR (*50mg/kg) Extract group showed a significant (P<0.01) decrease in Mean arterial blood pressure when compared to control group.*CR (*50mg/kg) treated group showed a significant (P<0.001) decrease in Mean arterial blood pressure when compared to AD group.*CR (*30mg/kg) group showed a significant (P<0.001) decrease in Mean arterial blood pressure when compared to AD group.

AD group showed an increase in R-R Interval, Q-T Interval non-significantly and significant (P<0.001) increase in Heart rate when compared to Control group. CR Extract (50/kg) group showed a decrease in R-R Interval, Q-T Interval & Heart rate non-significantly when compared to control group.*CR* (50mg/kg) treated group showed a significant (P<0.05) decrease in R-R interval and significant (P<0.01) decrease in Q-T interval and significant (P<0.001) decrease in Heart rate when compared to AD group.*CR*(30mg/kg) treated group showed a significant (P<0.001) decrease in Heart rate and decreased Q-T intervals non-significantly and no changes observed in R-R interval when compared to AD group.

AD group showed a significant decrease (P<0.01) in plasma adiponectin value when compared to control group.*CR* (50mg/kg) extract group showed a significant (P<0.001) increase in plasma adiponectin value when compared to control group.*CR* (50mg/kg) treated group showed a significant increase (P<0.001) in plasma adiponectin value when compared to AD group.*CR* (30mg/kg) treated group showed a significant increase (P<0.01) in plasma adiponectin value when compared to AD group.

HISTOPATHOLOGICAL STUDIES

Liver

In control group, Section studied from the liver showed parenchyma with intact architecture. Most of the perivenular hepatocytes and periportal hepatocytes appear normal. Within the hepatic parenchyma are seen few scattered mononuclear inflammatory cells [Fig.2, arrow].

In atherogenic diet group Section studied from liver showed parenchyma with partially effaced architecture. Most of the hepatocytes show apoptotic changes (Short-arrow, Fig.3), while some show cytoplasmic vacuolations (Long-arrow, Fig.4). Most of the central veins (Long-arrow, Fig.3) and sinusoids are dilated and congested (Short-arrow, Fig.4).

In aqueous extract of tubers of CR, (30mg/kg b.w) Section of liver showed parenchyma with intact architecture. Some of the hepatocytes show regenerative changes (Arrow, Fig.5), while few show regenerative changes (Arrow, Fig.5). Most of the central veins are dilated and congested (Arrow, Fig.6). There are seen scattered mononuclear inflammatory infiltrations within parenchyma.

In aqueous extract of tubers of CR,(50mg/kg b.w) Section of liver showed intact architecture. Few of the central veins (Short-Arrow, Fig.7) and sinusoids show congestion (Long-Arrow, Fig.7). Some of the hepatocytes show regenerative changes (Arrow, Fig.8). There are seen few mononuclear inflammatory infiltration within parenchyma.

KIDNEY

In control group, Section studied from kidney showed renal parenchyma with intact normal architecture. The renal glomeruli [Fig.10, arrow], tubules and blood vessels appear unremarkable. The interstitium shows scattered mononuclear inflammatory infiltration [Fig.9, arrow].

In atherogenic diet group, section studied from kidney showed: Architecture-Intact, Glomerulus-Diffuse hypercellularity due to extravasation of erythrocytes. Also seen are congested capillaries in the glomerulus [Fig.11, Short-arrow]. Tubules-Few tubules show hydropic degeneration, Blood Vessels-Dilated and Congested [Fig.11, Long-arrow]. Interstitium-Aggregates of foamy

macrophages [Fig.12, Long-arrow], mononuclear inflammatory infiltration [Fig.12, Short-arrow].

In aqueous extract of tubers of CR, (30mg/kg b.w)Section studied from kidney showed: Architecture-Intact, Glomerulus-Focal segmental hypercellularity due to extravasation of erythrocytes [Fig.13, arrow], Tubules-Few tubules show hydropic degeneration, Blood Vessels - Dilated and Congested [Fig.14, arrow], Interstitium-Scattered mononuclear inflammatory infiltration.

In aqueous extract of tubers of CR, (50mg/kg b.w) Section studied from kidney showed: Architecture-Intact, Glomerulus-The mesangium and capillaries appear within normal limits, Tubules-Normal, Blood Vessels-Few are dilated and congested, Interstitium-Few scattered mononuclear inflammatory infiltration.

Artery

In control group, the layers of artery appear intact, The tunica intima, tunica media and tunica adventitia appear within normal limits, The Tunica intima - media thickness - 30µm.

In atherogenic diet group, the layers of artery appear intact except for focal disruption of the endothelium. Within the tunica media and beneath the intima are seen few closely packed foam cells (Fig.19, arrow) and scattered lipid containing elongated smooth muscle cells. The tunica intima - media thickness - 36µm.

In aqueous extract of tubers of CR, (30mg/kg b.w) the layers of artery appear intact except for focal disruption of the endothelium. Within the tunica media and beneath the intima are seen scattered lipid containing elongated smooth muscle cells (Fig.21, arrow) and some lymphocytes. The tunica intima - media thickness – 32.25µm.

In aqueous extract of tubers of CR, (50mg/kg b.w) the layers of artery appear intact. The tunica intima, tunica media and tunica adventitia appear within normal limits. The tunica intima-media thickness-28.5µm.

Heart

InControl groupSection studied from the myocardium shows cardiac muscle with intervening fibrovascularseptae. The cardiocyte arrangement appears almost intact [Fig.25& 26, arrow].

In Atherogenic diet groupSection studied from the myocardium shows cardiac muscle with intervening fibrovascularseptae. Most of the cardiocytes appear arranged haphazardly [Fig.27, arrow]. Most of the cardiocytes contain cytoplasmic vacuolations (? Fat deposition) of varying sizes [Fig.28, arrow].

In aqueous extract of tubers of CR, (30mg/kg b.w) Section studied from the myocardium shows cardiac muscle with intervening fibrovascularseptae. Most of the cardiocytes appear arranged intact [Fig.29, arrow]. Some of the cardiocytes contain cytoplasmic vacuolations (? Fat deposition) of varying sizes [Fig.30, arrow].

In aqueous extract of tubers of CR, (50mg/kg b.w)Section studied from the myocardium shows cardiac muscle with intervening fibrovascularseptae. The cardiocyte arrangement appears intact [Fig.31 &32, arrow].

In plain CR Extract (50mg/kgb.w) Section studied from the myocardium shows cardiac muscle with intervening fibrovascularseptae. The cardiocyte arrangement appears almost intact [Fig.33 & 34, arrow].

Table 1. Effect of CR Tuber Extract on Body Weight

Groups→	Control	Atherogenic diet (AD)	CR Extract (50mg/kg)	AD+CR (50mg/kg)	AD+CR (30mg/kg)
% Increased in Body weight.→	36.487±0.540	85.220±9.297^{a***}	29.000±5.864$^{a\ ns}$	45.533±0.481^{b***}	54.493±1.471^{b**}

Table 2. Effect of CR Tuber Extract on Body Temperature

Groups	0 day	15th day	30th day	39th day
Control	36.173	36.333	36.177	36.263
Atherogenic diet (AD)	36.2	36.14	36.02	35.89
CR Extract	36.193	36.217	37.107	37.087
AD+CR (50mg/kg)	36.16	36.21	36.853	37.41
AD+CR (30mg/kg)	36.18	36.217	36.717	37.21

Table 3. Effect of CR Tuber Extract on Locomotor Activity

Groups	Ambulation Mean ± SEM	Rearing Mean ± SEM	Grooming Mean ± SEM
Control	67.567±1.157	15.800±0.208	6.933±0.088
Atherogenic diet(AD)	57.700±0.839^{a**}	18.633±0.406^{a***}	6.233±0.176^{a*}
CR Extract (50mg/kg)	73.317±1.187$^{a\ ns}$	14.567±0.819^{a*}	7.300±0.115^{a*}
AD + CR (50mg/kg)	78.100±2.346^{b***}	16.300±0.115^{b***}	9.33±0.260^{b***}
AD + CR (30mg/kg)	74.600±1.617^{b***}	17.633±0.088^{b**}	7.400±0.058^{b***}

Table 4. Effect of CR Tuber Extract on Organ Weight

Groups	Spleen(g) Mean ± SEM	Liver(g) Mean ± SEM	Right kidney(g) Mean ± SEM	Left Kidney (g) Mean ± SEM	Heart (g) Mean ± SEM
Control	0.380±0.006	2.577±0.029	0.340±0.006	0.327±0.009	0.380±0.006
Atherogenic diet (AD)	0.450±0.006^{a***}	3.147±0.026^{a***}	0.457±0.020^{a***}	0.467±0.012^{a***}	0.423±0.009^{a*}
CR Extract(50mg/kg)	0.370±0.006^{ans}	2.563±0.024^{ans}	0.337±0.009^{ans}	0.323±0.018^{ans}	0.380±0.006^{ans}
AD + CR(50mg/kg)	0.377±0.012^{b***}	2.587±0.012^{b***}	0.397±0.009^{b*}	0.360±0.017^{b***}	0.377±0.009^{b*}
AD + CR(30mg/kg)	0.407±0.009^{b*}	2.820±0.017^{b***}	0.410±0.12^{bns}	0.407±0.009^{b*}	0.393±0.012^{bns}

Table 5. Effect of CR Tuber Extract on Fat Pad Weight

Groups	Left Perirenal(g) Mean ± SEM	Right Perirenal(g) Mean ± SEM	Mesenteric (g) Mean ± SEM
Control	0.483±0.009	0.410±0.012	0.640±0.006
Atherogenic diet (AD)	0.750±0.012^{a***}	0.730±0.023^{a***}	0.920±0.029^{a***}
CR Extract (50mg/kg)	0.407±0.027^{a*}	0.337±0.018^{a*}	0.527±0.009^{a**}
AD + CR (50mg/kg)	0.520±0.012^{b***}	0.550±0.012^{b***}	0.733±0.009^{b***}
AD + CR (30mg/kg)	0.613±0.015^{b***}	0.607±0.012^{b***}	0.810±0.012^{b**}

Results are expressed as mean ± SEM. (n=6): a=compared with vehicle control: b=compared with Atherogenic diet: CR=*Cyperus rotundus:* Data was analyzed by One way ANOVA followed by Dunnett's multiple comparison tests: *P<0.05, **P<0.01, ***P<0.001, ns=non significant.

Table 6.1. Effect of CR tubers extraction biochemical determinations

Groups	Mean ± SEM				
	Glucose (mg/dL)	Total Cholesterol (mg/dL)	Triglycerides (mg/dL)	HDL-C (mg/dL)	LDL-C (mg/dL)
Control	64.33±1.468	79.30±3.09	77.373±0.609	44.917±0.795	19.343±0.056
Atherogenic diet (AD)	107.013±2.99^{a***}	166.67±2.88 ^{a***}	160.67±2.963 ^{a***}	39.567±1.658^{a*}	94.567±0.560^{a***}
CR Extract (50mg/kg)	67.48±2.11$^{a\ ns}$	79.217±2.32^{a} ^{ns}	75.157±1.248 $^{a\ ns}$	43.453±0.522 $^{a\ ns}$	20.667±0.239$^{a\ ns}$
AD + CR (50mg/kg)	94.553±0.635 ^{b**}	111.653±5.67 ^{b***}	150.210±2.919 ^{b*}	46.650±1.137^{b**}	34.457±0.601^{b***}
AD + CR (30mg/kg)	102.440±0.668$^{b\ ns}$	141.75±5.69 ^{b**}	155.840±0.380$^{b\ ns}$	40.083±1.228$^{b\ ns}$	70.670±0.271^{b***}

Table 6.2. Effect of CR tubers extraction biochemical determinations

Groups	LDH (u/l)	CK-NAC (u/l)	CK-MB (u/l)	AST (u/l)	ALT (u/l)	Urea (mg/dL)	Uric acid (mg/dL)	Creatinine (mg/dL)
Control	233.230± 4.050	239.067 ±3.753	140.753 ±2.427	36.683 ±0.970	25.290 ±0.194	33.623± 1.840	1.727± 0.050	0.560± 0.025
AD	384.003± 5.717 ^{a***}	367.347 ±7.324 ^{a***}	254.533 ±4.694^{a***}	67.550 ±1.862 ^{a***}	45.203 ±0.609^{a***}	48.023±0.965 ^{a***}	4.820± 0.211 ^{a***}	1.403± 0.078 ^{a***}
CR (50mg/kg)	235.833± 3.83 $^{a\ ns}$	244.973 ±5.565$^{a\ ns}$	145.833 ±4.910 $^{a\ ns}$	23.483 ±3.074^{a**}	24.557 ±0.673$^{a\ ns}$	32.747±1.758 $^{a\ ns}$	1.683± 0.035 $^{a\ ns}$	0.557± 0.042 $^{a\ ns}$
AD + CR (50mg/kg)	359.833± 3.667 ^{b**}	334.80 ±4.972 ^{b**}	225.167 ±4.256^{b***}	51.317 ±1.262 $^{b\ ***}$	42.267 ±0.540 $^{b\ *}$	35.970± 0.950 ^{b***}	3.513± 0.145 ^{b**}	1.130± 0.104 ^{b*}
AD + CR (30mg/kg)	371.167± 3.383 $^{b\ ns}$	342.513 ±2.079 ^{b*}	237.833 ±0.882 ^{b**}	56.920 ±2.168 $^{b\ *}$	44.487 ±0.604^{bns}	42.567± 0.706 ^{b*}	4.410± 0.358 $^{b\ ns}$	1.210± 0.010 $^{b\ ns}$

Table 7. Effect of CR Tubers Extract on Blood Pressure

Groups	BP
Control	91.067±0.384
Atherogenic diet (AD)	115.100± 1.940^{a***}
CR Extract (50mg/kg)	83.833±0.906^{a**}
AD+CR (50mg/kg)	98.400±0.462^{b***}
AD+CR (30mg/kg)	105.833±0.845^{b***}

Table 8. Effect of CR tubers extract on ECG and heart rate

ECG Intervals	Control	Atherogenic diet(AD)	CR Extract (50mg/kg)	AD+CR (50mg/kg)	AD+CR (30mg/kg)
R-R Interval (mSec)	437.000 ±0.577	439.000 ±0.577$^{a\ ns}$	436.333 ±0.533$^{a\ ns}$	438.000 ±0.577^{bns}	438.667 ±0.677$^{b\ ns}$
Q-T Interval (mSec)	137.000±0.577	139.000 ±0.577$^{a\ ns}$	136.667 ±0.333$^{a\ ns}$	138.000 ±0.577$^{b\ ns}$	138.000 1.155$^{b\ ns}$
Heart rate(bpm)	268.000±0.577	320.000 ±3.215^{a***}	255.333 ±0.333^{a**}	283.667 ±1.856^{b***}	304.333 ±2.906^{b**}

Table 9. Effect of CR Tubers Extract on Plasma Adiponectin Value

Groups	BP
Control	91.067±0.384
Atherogenic diet (AD)	115.100± 1.940^{a***}
CR Extract (50mg/kg)	83.833±0.906^{a**}
AD+CR (50mg/kg)	98.400±0.462^{b***}
AD+CR (30mg/kg)	105.833±0.845^{b***}

Fig 1. [H&E, x400]Fig.2 [H&E, x400]

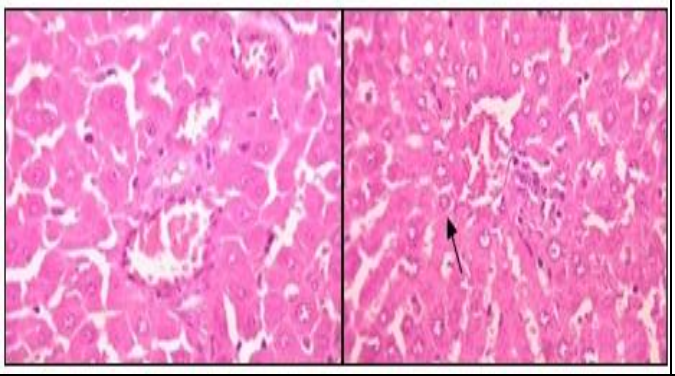


Fig 3 and 4. H&E, x400

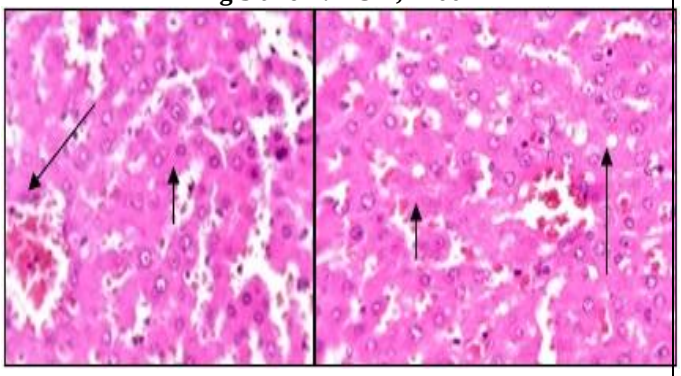


Fig 5, 6. H&E, x400

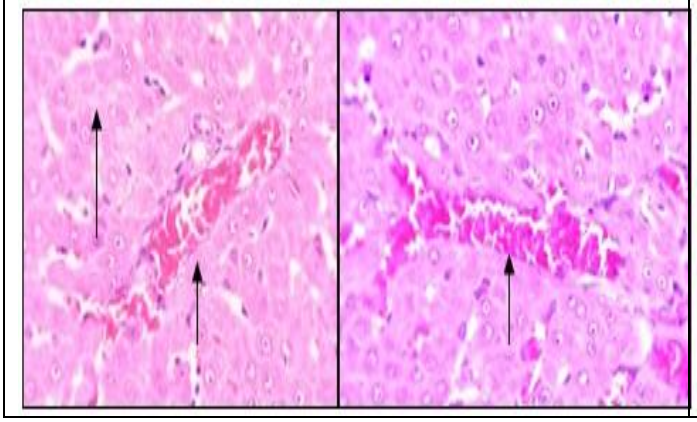


Fig 7, 8. H&E, x400

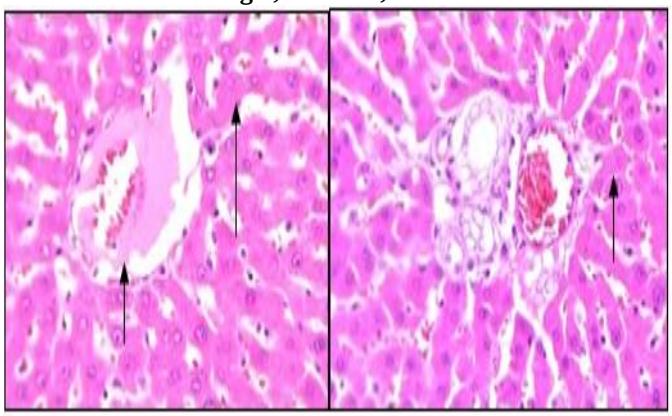


Fig 9, 10. H&E, x100

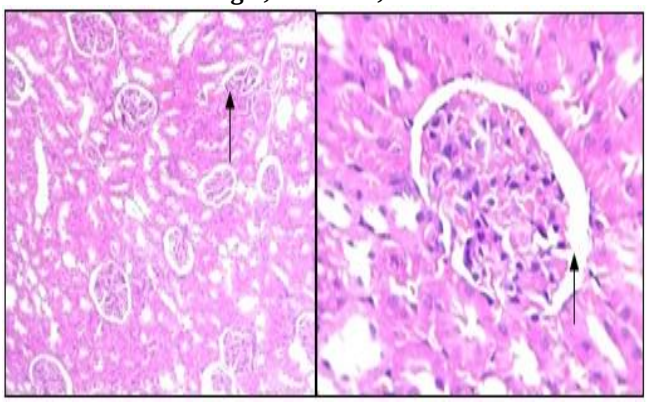


Fig 11, 12. H&E, x100

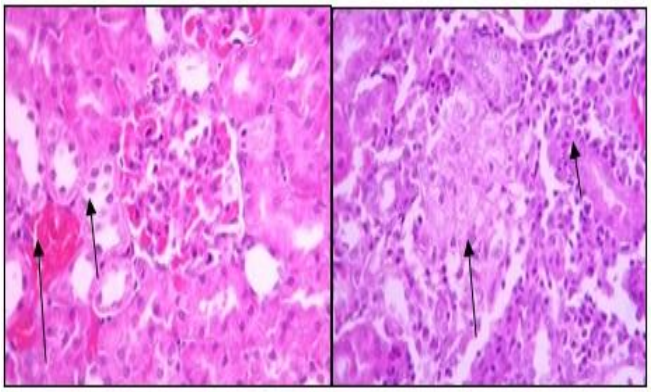


Fig 13, 14. H&E, x100

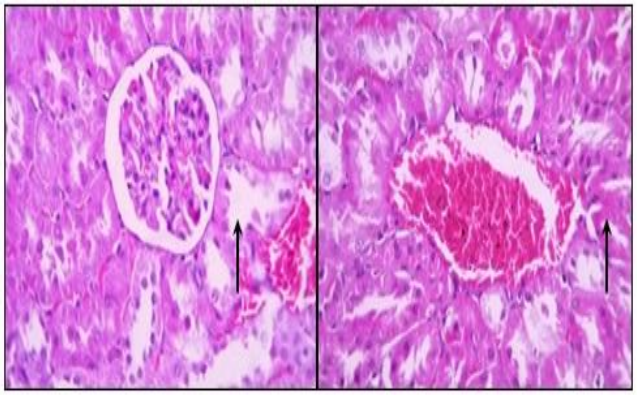
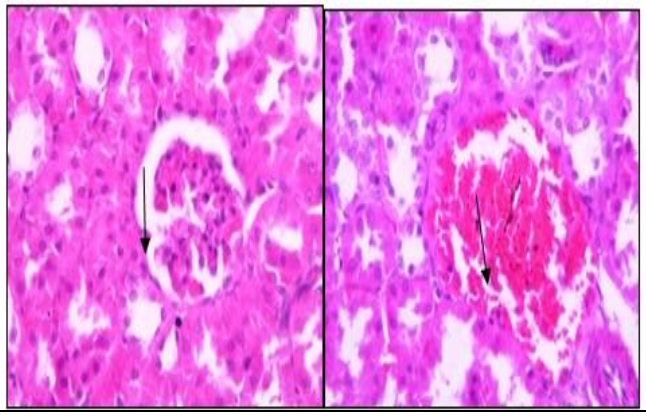


Fig 15, 16. H&E, x100



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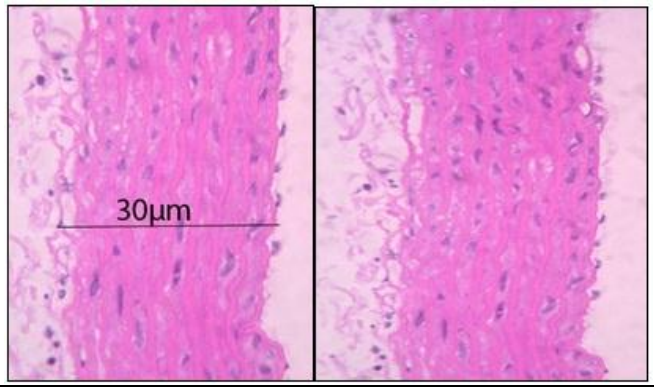

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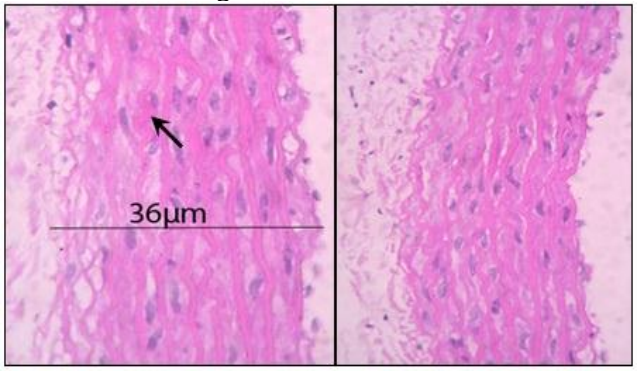

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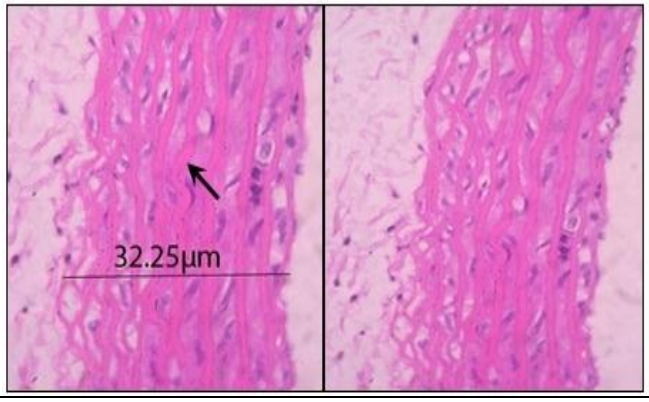

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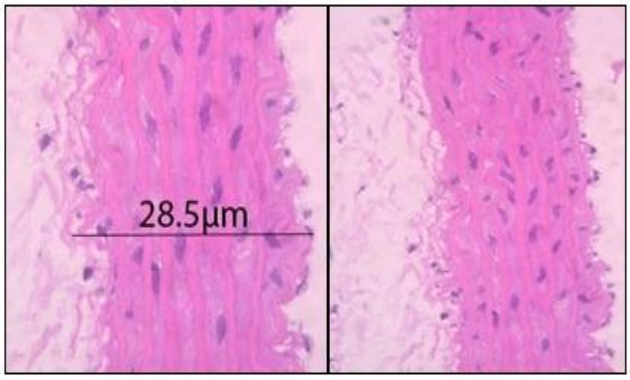

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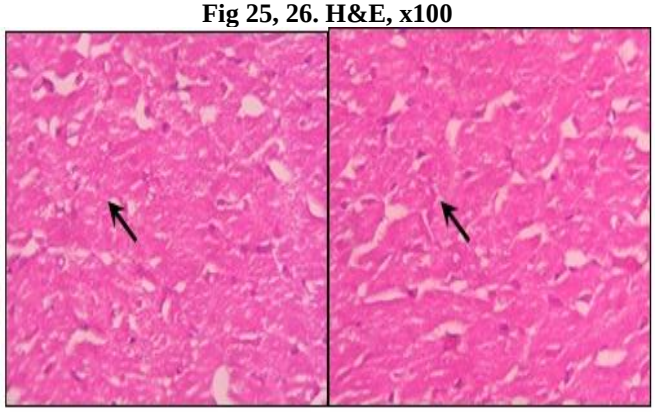
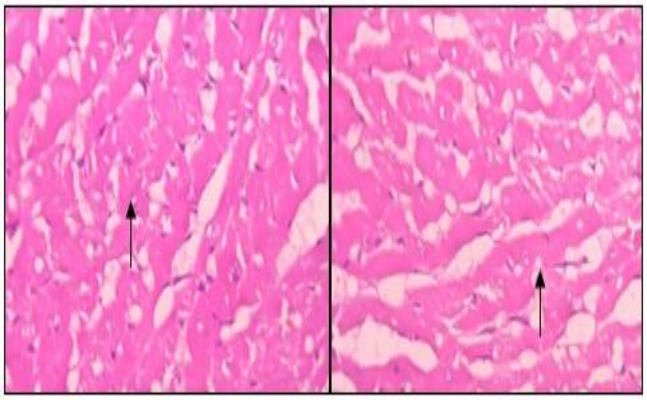


Fig 27, 28. H&E, x100



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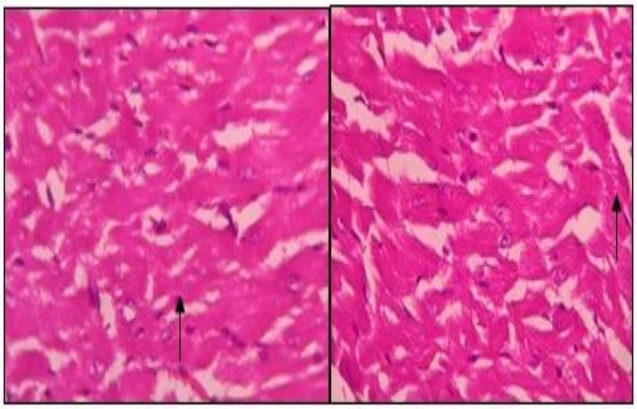


Fig 31, 32. H&E, x100

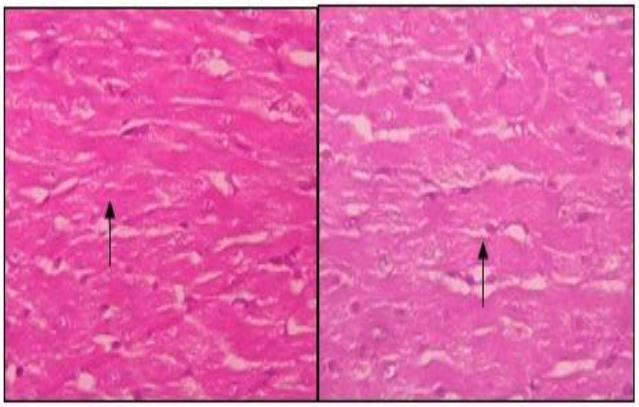
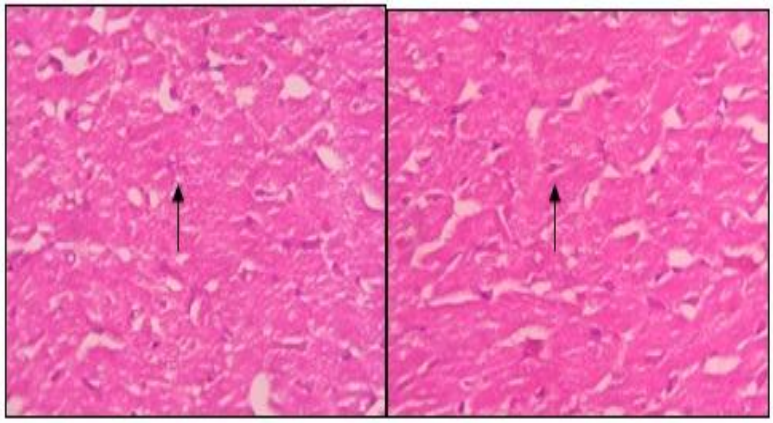


Fig 33, 34. H&E, x100



DISCUSSION

In the present study, the anti-obesity and cardio vascular properties of aqueous extracts of *Cyperus rotundus* tubers was studied using atherogenic diet fed animal model of obesity as they have been reported to bear close resemblance to human obesity. Atherogenic diets have been previously reported to increase energy intake and cause obesity in humans as well as animals. Further, the composition and variety of atherogenic diet also exert synergistic effects on the development of obesity.

Both obesity and the cardiovascular associated properties are common most health problems that affect millions of people. In recent decades, the association between obesity and co-morbid cardiovascular changes has been actively investigated from an epidemiological standpoint and from a basic research standpoint. And common pathogenic links have been proposed. Recent epidemiologic studies show that body fat itself excises a risk factor for cardiac function. Accumulating evidence suggests that metabolic syndrome like obesity and the individual components of the metabolic syndrome such as

increased triglycerides, and reduced high-density lipoprotein cholesterol are heightened risk factors for cardiovascular properties.

Obesity is considered to be a disorder of energy balance, occurring when energy expenditure is no longer in equilibrium with daily energy intake, so as to ensure body weight homeostasis. Although the etiology of obesity is complex, dietary factors, particularly the consumption of an atherogenic diet, is considered a risk factor for its development(Xu RY *et al.,* 2008). The current results showed that body weight increased significantly (P<0.001) in the atherogenic diet group when compared with the control group**.** Consumption of the atherogenic diet led to obesity because it facilitates the development of a positive energy balance leading to an increase in visceral fat deposition; this led to abdominal obesity in particular. Moreover, it was found that an atherogenic diet (high fat diet) feeding is accompanied by molecular adaptations that favor fat storage in muscle rather than oxidation(Hinderling VB *et al.,* 2005). The very fact in appreciated in the present study , because of the fact that the experimental animal consumed considerable more food that is the atherogenic diet than the control group animals throughout the experiment. In the current study, rats fed atherogenic diet consumed considerably more food than the control rats throughout the experiment. So their caloric intake was increased and they showed a large increase in perirenal visceral adipose tissue mass, suggesting that the excess energy led to the buildup of adiposity.

In animal models of obesity, there will be decrease in diet induced thermogenesis, due to decrease in sympathetic activation of brown adipose tissue. The main factor for this decrease in diet induced thermogenesis is the neurotransmitter NPY synthesized throughout the brain. NPY causes increase in food intake within 10-15 min, combined with a reduction in thermogenesis on its endogenous release. NPY inhibits thermogenesis by reducing sympathetic activation of brown adipose tissue. The basal body temperature recording noted under Table No: 2 indicate a significant (P<0.001) changes in the thermogenesis levels in experimental animals which indicate the fact that aqueous extract of *Cyperus rotundus* tubers influence the energy balance and shift the balance towards increased break down of fats and mobilization of fats from adipocyte tissue. The above fat can be substantiated from the results obtained in the study on locomotor activity of the experimental animals. The data in the Table No: 3 indicate that there is a good ambulatory activity observed in the group of the experimental animals treated with the aqueous extract of *Cyperus rotundus* tubers.

It is well known that, obesity is associated with increased adipose tissue accumulation in the body. It is reported that feeding with an atherogenic diet in wistar rats leads to increase in weight of body organs such as liver, heart, spleen and both kidneys. It is also reported that atherogenic diet induces substantial increase in deposition of fat in the mesenteric, perirenal and uterine region in wistar rats. The above facts can be verified by the data provided in Table No: 4 and 5 experimental animals fed with atherogenic diet showed increase in the weight of the organs namely liver, heart, spleen and kidneys and also a significant increase in the weight of the mesenteric, perirenal fat pads. The study also showed that aqueous extract of *Cyperus rotundus* tubers at the dose levels studied, significantly exerts effect on weight gain of the organs viz. liver, heart, spleen and kidneys and greatly influenced the fat deposition process in the fat pads and infact significantly reduced the fat accumulation in mesenteric, perirenal and uterine fatty tissues. This fact again establishes that the aqueous extracts of *Cyperus rotundus*tubers has a definite influence in body fat metabolism.

It is well known that hyperlipedemia is the leading risk factor for atherosclerosis. Epidemiological investigation revealeda positive correlation between the severe degree of atherosclerosis and the concentration of plasma cholesterol as well as LDL (Yaling WU *et al.,* 2009). . In the present study, an atherogenic diet resulted in dyslipidemic changes as illustrated by increasing triglycerides, total cholesterol and low density lipoprotein (LDL) and a decrease in serum level of high density lipoprotein (HDL) in the present investigation the results obtained are tabulated vide Table No: 6 showed the animals treated with aqueous extract of tubers of *Cyperus rotundus* decrease in the lipid component such as TGs, TC, LDL and a significant increase in the lipid component HDL. The study suggests that the aqueous extract of tubers of *Cyperus rotundus* shifts the disturbed lipid profile to the normal thereby counters the effects that would have been followed as secondary effects such as Hypertension, Atherosclerosis, stroke(Woo MN *et al.,* 2008).Components of *Cyperus rotundus* have hypocholesterolemic activity, *Cyperus rotundus* tubers might possess lipophilic properties such as sesquiterpenoids or sterols have lipid-lowering effects, resulting in depression of lipid accumulation. It consequently has anti-atherosclerotic properties (Lemaure B *et al.,* 2007).

The obese rats showed a significant increase in the activity of serum LDH, CK-NAC and CK-MB when compared to the normal rats(Amat SFC *et al.,* 2008). Reduced muscle mitochondrial content function with increasing obesity would lower the total cellular ATP yield, which would result most notably in increased mitochondrial volume, and increased glycolytic enzymes necessitating increased activity of creatine kinase, as this enzyme is responsible for rapidly transferring high-energy phosphate groups from the site of production to the site of use(Janssen E *et al.,* 2003). The increased blood levels of total cholesterol, LDL, VLDL as well as lowered levels of

HDL in atherogenic diet rat have been identified in the development of hypercholestremia, which is one of the risk factors for CAD (Duarte J *et al.,* 2000) Administration of *Cyperus rotundus* produces a significant decrease in the activity of CK-NAC, CK-MB and LDH (Table 8). Our findings showed that obese rats treated with the *Cyperus rotundus* exhibited significant decreases in LDH, CK-NAC and CK MB activity, The *Cyperus rotundus* content could prevent the development of atherosclerosis through regulating vascular inflammatory processes in rats fed with an atherogenic diet.

The obese rats showed a highly significant increase in the concentration of serum urea, uric acid creatinine, compared with the control group (Table 6) (Katzung BG, 1998). Abnormal renal function is mainly associated with diabetic nephropathy. The pathophysiology involves glucose that binds irreversibly to proteins in the kidney circulation to form advanced glycosylation end products that can form complexes that contribute to renal damage by stimulation of fibrotic growth factors (Mao et al., 2000). Atherogenic diet induces alteration of renal lipid metabolism by an imbalance between lipogenesis and lipolysis in the kidney, as well as systemic metabolic abnormalities and subsequent renal lipid accumulation leading to renal injury(Ferreira M *et al.,* 2000; Guyton AC, 1996). The oral administration of *Cyperus rotundus* (Table 6) shows that serum concentration of urea, uric acid and Creatinine were significantly decreased. The effect of *Cyperus rotundus* on renal lipid metabolism could serve as a new therapeutic approach, as it counters the renal changes associated with metabolic syndrome(Ouwens DM *et al.,* 2005; Fielding BA and Frayn KN, 2000). Hence, *Cyperus rotundus*has beneficial effects on renal function. Although there is still debate on the significance of uricacid as a risk factor for cardiovascular disease, many physicians do consider elevated uric acid to be a component of the metabolic syndrome. There is little support for an independent causal role for serum uric acid in the development of CHD. However, uric acid may provide useful prognostic information in subjects with hypertensive vascular disease, suggesting that the influence of uric acid on CHD is explained by the secondary association of it with other risk factors such as dyslipidemia, hyperinsulinaemia and obesity (Cindik N *et al.,* 2005). The oral administration of *Cyperus rotundus* extract (Table 8) clarified that serum concentration of urea,uricacid and Creatinine were significantly decreased. The *Cyperus rotundus* therapy retains the balance between lipogenesis and lipolysis in the kidney to counteract the obesity-associated renal damage.

The current data showed a significant increase in the activity of enzymes AST and ALT in the obese compared with control rats (Table 6). Liver is bombarded by the free fatty acids (FFA) that pour out of the adipose tissue into the portal blood. This can directly cause inflammation within the liver cells, which then release further pro-inflammatory cytokines, leading to more hepatocyte injury and affecting the integrity of liver cells(Rao NK *et al.,* 2006). The present results demonstrate that the *Cyperus rotundus* showed a significant decrease in the activity of both AST and ALT and showing a hepatic protective action.

It is generally accepted that arterial blood pressure depends on cardiac output and vascular resistance. The physiological processes related to vascular function differ greatly between resistance arteries and conduit arteries. Large arteries, such as the aorta, are considered as passive conduits with the principle function of conducting and distributing cardiac output to various tissues. Resistance arteries, but not conduit arteries, are important for the regulation of both regional blood flow and vascular resistance. The aim of the present work was to study the effects of C. *rotundu*s on blood pressure. For that purpose, we have investigated the effects of C. *rotundus* on blood pressure parameters in the anaesthetized obese wistar rats. Aqueous extract of C. *rotundu*s caused a decrease in mean arterial blood pressure in anaesthetized wistar rats. At the dose of 50mg/kg the mean arterial blood pressure showed a statistically significant ($P<0.001$) decrease in BP when compared to atherogenic diet group.

On the basis of this study it is suggested that C. *rotundu*s is probably acting both centrally and peripherally to produce changes in blood pressure by altering the peripheral resistance and cardiac mechanics**.** Therefore it can be concluded that C. *rotundu*s is an active drug both physiologically and pharmacologically.

In ECG there is no significant changes occurred in both R-R and Q-T interval in atherogenic diet group experimental animals when compared to control group, increased heart rate was observed in diet group when compared to control group. The present results demonstrate that the aqueous extract of tubers of *Cyperus rotundus* showed no effect on R-R, Q-T interval and decrease effect of heart rate non-significantly when compared to atherogenic diet group.

Histopathological studies of vital organs such as Liver, Heart, Artery & Kidney was conducted. The results found are highly encouraging. The regenerative changes were observed in the group of animals treated with the aqueous extracts of *Cyperus rotundus* tubers alone and in presence co-morbid conditions also. The tissue elements lost due to the induction of disease condition namely obesity, the cardiovascular co-morbidity were regenerated and restored in the treated animals.Atherosclerosis (also known as arteriosclerotic vascular disease or ASVD) is a condition in which an artery wall thickens as a result of the accumulation of fatty materials such as cholesterol. It is a syndrome affecting arterialblood vessels, a chronic inflammatory response in the walls of arteries, caused largely by the accumulation of macrophagewhite blood

cells and promoted by low-density lipoproteins (plasma proteins that carry cholesterol and triglycerides) without adequate removal of fats and cholesterol from the macrophages by functional high density lipoproteins (HDL), Early atherogenesis is characterized by the adherence of blood circulating monocytes to the vascular bed lining, the endothelium, followed by their migration to the sub-endothelial space, and further activation into monocyte-derived macrophages. The primary documented driver of this process is oxidized Lipoprotein particles within the wall, beneath the endothelial cells, though upper normal or elevated concentrations of blood glucose also plays a major role and not all factors are fully understood. Fatty streaks may appear and disappear.

Low Density Lipoprotein particles in blood plasma, when they invade the endothelium become oxidized creates a risk for cardiovascular disease. A complex set of biochemical reactions regulates the oxidation of LDL, chiefly stimulated by presence of enzymes, e.g. Lp-LpA2 and free radicals in the endothelium or blood vessel lining.

The initial damage to the blood vessel wall results in an inflammatory response. Monocytes (a type of white blood cell) enter the artery wall from the bloodstream, with platelets adhering to the area of insult. This may be promoted by redox signaling induction of factors such as VCAM-1, which recruit circulating monocytes. The monocytes differentiate into macrophages, which ingest oxidizedLDL, slowly turning into large foam cells – so-described because of their changed appearance resulting from the numerous internal cytoplasmic vesicles and resulting high lipid content. Under the microscope, the lesion now appears as a fatty streak. Foam cells eventually die, and further propagate the inflammatory process. There is also smooth muscle proliferation and migration from tunica media to intima responding to cytokines secreted by damaged endothelial cells. This would cause the formation of a fibrous capsule covering the fatty streak. The layer of artery and cardiocyte arrangement appears intact.

The study showed that the extracts of *Cyperousrotundus* significantly counters the atherogenic diet induced cardiovascular properties and improves the lipid profile in the experimental animals. Adiponectin is a protein hormone that modulates a number of metabolic processes, including glucose regulation and fatty acid catabolism. Adiponectin is exclusively secreted from adipose tissue into the bloodstream and is very abundant in plasma relative to many hormones. Levels of the hormone are inversely correlated with body fat percentage in adults,while the association in infants and young children is less clear. The hormone plays a role in the suppression of the metabolic derangements that may result in type 2 diabetes, obesity, atherosclerosis, non-alcoholic fatty liver disease(NAFLD) and an independent risk factor for metabolic syndrome. Adiponectin is secreted into the blood stream where it accounts for approximately 0.01% of all plasma protein at around 5-10 μg/mL. Plasma concentrations reveal a sexual dimorphism, with females having higher levels than males. Levels of adiponectin are reduced in diabetics compared to non-diabetics. Weight reduction significantly increases circulating levels.

In normal humans, its expression is restricted to adipose tissue. Plasma adiponectin levels are negatively correlated with the body mass index (BMI), fasting plasma glucose, fasting insulin, insulin resistance, and triglycerides. It is an anti-inflammatory adipocytokine that modulates insulin effects. The administration of adiponectin to mice decreased the plasma glucose, free fatty acid (FFA) and triglyceride levels, and hepatic glucose production. Adiponectin was first characterised in mice as a transcript over expressed in preadipocytes (precursors of fat cells) differentiating into adipocytes.

The human homologue was identified as the most abundant transcript in adipose tissue. Contrary to expectations, despite being produced in adipose tissue, adiponectin was found to be decreased in obesity. This downregulation has not been fully explained. The gene was localised to chromosome 3q27, a region highlighted as affecting genetic susceptibility to type 2 diabetes and obesity. Supplementation by differing forms of adiponectin was able to improve insulin control, blood glucose and triglyceride levels in mouse models.

Berberine, an herbal folk medicine, has been shown to increase adiponectin expression which partly explains its beneficial effects on metabolic disturbances. To gain further insights into metabolic determinants of adiponectinemia in animals, it would be useful to examine the relationship between the plasma adiponectin concentration and direct measures of adiposity, body fat distribution, lipid profile & CVS parameters. In the present study, we examined the effect of the tuber extracts of *Cyperus rotundus* and the relationship between the plasma adiponectin concentration and adiposity, body fat distribution, lipid profile, renal profile, liver enzymes, CVS parameters, histopathology of Liver & a major artery and heart, in atherogenic diet fed wister rats.

The results obtained showed significant correlation between adiponectemia and adiposity, body fat distribution, in atherogenic diet fed obese animals. The preliminary investigation of effects of extracts of *Cyperus rotundus* showed to improve the overall picture of metabolism especially the fatty acid metabolism. The plasma adiponectin levels were found to be significantly decreased in AD (obese) group ($p<0.001$), and a significant increase was observed in AD+CR (50mg/kg) (obese+treated) groups ($p<0.001$), AD+CR (30mg/kg) (obese+treated) group ($p<0.01$) when compared to control group.

To understand the results obtained and also to have an insight into the mechanism, the AMPK loop was investigated to find two important hormones namely adiponectin and leptin to be involved in the regulation of AMPK pathway. 5'AMP-activated protein kinase or AMPK or 5'adenosine monophosphate-activated protein kinase is an enzyme that plays a role in cellular energy homeostasis. It consists of three proteins (subunits) that together make a functional enzyme, conserved from yeast to humans. It is expressed in a number of tissues, including the liver, brain, and skeletal muscle. The net effect of AMPK activation is stimulation of hepatic fatty acid oxidation.

And ketogenesis, inhibition of cholesterol synthes is, lipogenesis, and triglyceride synthesis, inhibition of adipocyte lipolysis and lipogenesis, stimulation of skeletal muscle fatty acid oxidation and muscle glucose uptake, and modulation of insulin secretion by pancreatic beta-cells. A similar study showed that much like leptin, adiponectin also stimulates the oxidation of fatty acids via the AMPK pathway, and that it also stimulates the uptake of glucose in skeletal muscle.

Our findings suggest the extracts of CR were responsible in improvisation of the plasma adiponectin levels. Adiponectin is a cytokine (cell-cell signaling protein) acts through the AMPK signaling pathway. The results obtained in our study, on weight gain/loss, lipid profile, renal profile, liver enzymes, cardiac biomarkers, body temperature, serum glucose level, plasma adiponectin estimations by Elisa, indicate the fact that the extracts of CR improve adiponectin levels and thus interfere the AMPK dependent metabolic pathway. The facts also corroborate the reported findings that increased BMI, increased visceral body fat distribution, increased triglycerides, increased free fatty acid levels, increased LDL-C, decreased HDL-C, and CVS parameters are negatively correlated with plasma adiponectin levels.

The results obtained in our study of weight gain/loss , locomotor activity, body temperature, weight of vital organs, fat distribution, lipid profile, renal profile, liver enzymes, cardiac biomarkers, glucose, BP, ECG, Heart rate, histopathology & the plasma adiponectin concentration, in the treated and untreated experimental animals indicate that properties claimed & reported for the plant extracts were found to be true primarily and our study also indicate the active principles present in the extracts of *Cyperus rotundus* interfere the AMPK pathway and improve the overall health. However, further extensive investigation is needed to understand the overall mechanisms through which the active principles present in the plant body act and bring about the metabolic changes.

Thus the investigation undertaken to study the effect of aqueous extracts of *Cyperus rotundus* tubers were very effective in countering the atherogenic fed obesity and obesity induced co-morbid condition such as cardiovascular diseases. The results also indicate aqueous extracts of *Cyperus rotundus* tubers affect the total lipid metabolism and favour the correction of disturbed lipid profile. Further studies are needed to explore the underlying mechanisms.

CONCLUSION

The investigation undertaken to study the effects of extracts of *Cyperus rotundus* were found to be very effective in countering atherogenic diet induced changes in lipid profile /metabolic disturbances. The extracts were effective in improving the lipid profile, liver function, kidney function and CVS parameters and in countering atherogenic diet induced changes in tissue elements of Liver and blood vessels. The plasma Adiponectin concentrations were found to be increased in animal groups treated with extracts of *Cyperus rotundus* and were negatively correlated in atherogenic diet fed obese animals and positively correlated in animals with lean body mass. Since, the extracts were found to increase the plasma Adiponectin concentration in the experimental animals; they may exert effects through AMPK pathway. Further studies are needed to explore the underlying mechanisms.

CONFLICT OF INTEREST

There is no conflict of interest associated with the authors of this paper.

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