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R E S E A R C H A R T I C L E

Pharmacological Evaluation of *Cynodon Dactylon* Plant Extracts for Cardioprotective Activity

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A B S T R A C T

The Plant *Cynodon dactylon Family* (Graminae/Poaceae) is extensively used in clinical practice but it has various pharmacological activities have been reported. It is a very familiar plant almost available in the entire world. In ethno medicinal practices, the plant *Cynodon dactylon* used in the treatment of various diseases and has pharmacological action. Treatment with Triton-X-100 caused a significant rise in the levels of Total Cholesterol in Group-II, Group-III Group-IV Group-V Group-VI (i.e Hyperlipidemic Control, MECD 500 mg/kg, PECD 400 mg/kg, PECD 500 mg/kg, & Standard (Atorvostatin) 10mg/kg) and the levels were found to be 192.47 ± 5.05, 175.28 ± 4.43, 180.97 ± 5.21,187.86 ± 9.66, and 180.79 ±9.1, respectively. Administration of various doses of the MECD & PECD after the Induction with Triton-X-100 resulted in the decreasing of Cholesterol levels. MECD at dose of 500mg/kg were 27.1 ± 2.99 and groups treated with PECD at dose of 400mg/kg and 500mg/kg showed a dose dependant increase in the HDL-C levels 31.04 ± 4.32 and 33.15 ± 2.51 respectively. In Atorvastatin group the HDL-C was elevated to 39.18 ± 3.14. . The liver function tests involve mainly the determination of LDH and any marked necrosis of the liver cells lead to a significant rise of these enzymes in the serum. The Scavenging activity of the extract towards DPPH radicals when compared with standard ascorbic acid. IC_{50} value for standard Ascorbic acid was found to be 43.137µg/ml, where as the IC_{50}value for phenolic extract of PECD was found to be 41.024 µg/ml.

Keywords: Cardioprotective, Plant extract, pharmacological, Screening.

A R T I C L E I N F O

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C O N T E N T S

1. Introduction

India has tremendous wealth of medicinal plants and its resources which are of different kinds they grow in different climatic and ecological conditions. In ancient time India was not so advanced in therapeutic values of medicinal plants. The earliest mention of the use of medicinal plant is found in Rig-Veda [1]. The number of drugs in the olden days was not large and hence no elaborated descriptions were given with regard to their identification. Traditional knowledge of medicinal plants has always guided the search for new cures. In spite of the advent of modern high throughput drug discovery and screening techniques, traditional knowledge systems have given clues to the discovery of valuable drugs. Cynodon dactylon occurs on almost all soil types especially in fertile soil. e.g. loamy soil. It is common in disturbed areas such as gardens, roadsides, overgrazed, trampled areas, uncultivated lands, localities with high levels of nitrogen, and is often found in moist sites along rivers. It is suitable for cultivation under dry land conditions [2]. It is widely distributed in southern African countries, in biomes such as grassland, Savanna, Nama-Karoo and Fynbos.

Traditional medicinal plants are often cheaper, locally available and easily consumable, raw or as simple medicinal preparations. Phytoconstituents reported in cardioprotective plants significantly prevented the altered biochemical variation such as marker enzymes serum glutamate- pyruvate transaminase or alanine transaminase, serum glutamate oxaloacetate transaminase or aspartate transaminase, creatinephosphokinase, alkaline phosphatise, lactate dehydrogenase, lipid profile including low density lipoprotein, very low density lipoprotein, triglycerides, high density lipoprotein, total cholesterol and antioxidant parameters including Superoxide dismutase (SOD), glutathione, catalase, Glutathione peroxidise come to near normal status [17].

To evaluate the cardio protective effect of *Cynodondactylon* Methanolic Extract and Phenolic Extracts on Triton -X-100 induced hyperlipidemic rats. Collection of Plant Material *Cynodondactylon*, Preparation and Phytochemical Screening of *Cynodondactylon* Methanolic and Phenolic Extract, Toxicity Studies of Methanolic Extract of *Cynodondactylon* on wister rats, To induce hyperlipidemia by Triton X-100 in wister rats, To Investigate Antihyperlipidemic activity of *Cynodondactylon* Methanolic extract and Phenolic Extracts in Triton x-100 induced hyperlipidemic rats, To evaluate cardioprotective activity, Major complication of hyperlipidemia are atherosclerotic heart disease, heart attack and heart stroke [3], but atherosclerosis is primary cause of death. Developing countries are reliant on medicinal plants as their main source of treatment for diseases. As *Cynodondactylon have* the native habitat the production is more so it is locally available cost effective with no side effects.

Collection and Authentification of Plant Material:

The Aerial Parts of *Cynodon dactylon* for the study were procured and authenticated

Extraction of Plant Material: The plant is grinded in to a coarse powder with the help of suitable grinder.

2. Materials and Methods

Cold Extraction: In this work the cold extraction process was done with the help of methanol. About 200gms of powdered material was taken in a clean, flat bottomed glass container and soaked in 750 ml of methanol [4]. The container with its contents were sealed and kept for period of 7 days accompanied by continuous shaking with the shaker. The whole mixture then went under a coarse filtration by a piece of a clean, white cotton wool.

Evaporation of Solvent: The filtrates (methanol extract) obtained were evaporated using Rotary evaporator in a porcelain dish. They rendered a gummy concentrate of greenish black. The extract was kept in vaccum decicator for 7 days.

Preliminary Phytochemical Screening: The *Cynodon dactylon* extract was carried out for the analysis of Alkaloids, Carbohydrates, Tannins, Saponins, Steroids, Phenols, Flavonoids. as per the standard methods [5].

Acute toxicity studies: The Acute Toxicity Studies was performed using female rats as per OECD Guideline No.423 (Short term toxicity). Male mice were selected of weight around 50 ±10 gm for main test. Single animals are dosed in sequence usually at 48 h intervals. A Dose Progression Factor of 3.2 is used. Using the default dose progression factor, doses would be selected from the sequence (1.75, 5.5, 17.5, 55, 175, 550, 1750, and 5000). However, the time intervals between dosing are determined by the onset, duration, and severity of toxic signs. Treatment of an animal at the next dose should be delayed until one is confident of survival of the previously dosed animal [6]. If the animal survives, the second animal receives a higher dose. If the first animal dies or appears moribund, the second animal receives a lower dose. The toxicological effects were observed in terms of mortality expressed as LD50. The number of animals dying or surviving during a period was noted.

Method of Induction: The systemic administration of the surfactant Triton X-100 to mice results in a biphasic elevation of plasma cholesterol and triglycerides. Hyperlipidemia was induced in Wistar albino rats by single intraperitoneal injection of freshly prepared solution of Triton-X-100 (100 mg/kg) in physiological saline solution after overnight fasting for 18 h.

Experimental Animal Protocol: Experimental rats, straved for 18 hr, were provided water *ad libitum*. The rats were divided in to six groups containing four animals in each group.

Group – I : Normal Control.(Normal saline 10ml/kg orally for 7 days)

Group – II : Hyperlipedemic control, (Triton x 100.)

Group – III : Hyperlipedemic rats treated with MEBM at dose of 500mg/kg. for 7days

Group – IV: Hyperlipedemic rats treated with PEBM at dose of 400mg/kg for 7days.

Group – V : Hyperlipedemic rats treated with PEBM at dose of 500mg/kg. for 7days.

Group – VI: Hyperlipedemic rats treated with Atorvostatin at 10 mg/kg for 7days.

All the groups recives single i.p. injection of Trition X-100 at dose of 100mg/kg, simultaneously with Group- II, Group – III, Group – IV, Group – V, Group – VI , expect Group – I (Normal control). After 72 hours of Triton X-100 injection. The Group – VI receives Atorvostatin at dose of 10 mg/kg, was prepared by suspending bulk Atorvastatin in aqueous 0.5% methyl cellulose [7] for 7 days. The Group– III, receive MEBM, at daily dose of 500mg/kg orally for 7 days and Group – IV, Group – V receives PEBM at daily dose of 400mg/kg and 500mg/kg orally for 7 days

Blood Sample Collection and Analysis: The rats are anesthetized by ether and then Blood samples were collected on 0^{th} and 8^{th} day from retro-orbital plexus of rat using micro capillary technique from rats of all the groups, and centrifuged at 3000 rpm for 15 min so as to get serum. The serum is analyzed for total cholesterol, triglycerides and HDL levels using biochemical kits [8].

Bio Chemical Assays for lipids

Estimation Procedures: Plasma Lipid Profile Estimation

Total cholesterol LDL cholesterol, HDL Cholesterol, VLDL cholesterol, Triglycerides levels were measured using commercial kits.

Estimation of Triglycerides: Determination of serum Triglycerides concentration is used to assess the possible presence of increased blood and Serum levels of triglycerides. Wave length: 546 (Green Filter), Temperature : 37 C, Reaction type: End point with standard.

Pipettle in to clean dry tube labelled Blank (B), Standard (S) and Test (T).

Assay of hematological parameters: Hemoglobin content, red blood cells (RBC), white blood cells (WBC), differential count, total count, and platelet count were assayed in the blood as per the standard methods [9].

Estimation of hemoglobin: Pipetted out 0.02 ml of serum and 5.0 ml of Drabkin's solution into a test tube. Simultaneously, a blank was set up with Drabkin's solution and distilled water. Mixed well the above tubes and allowed to stand at room temperature for 5 minutes. Measured the absorbance of test at 546 nm. Take the absorbance of cyanmethemoglobin standard was taken directly without adding working reagent against blank at 546 nm. The results were expressed as g/dl in serum [10].

Estimation of red blood cells: The whole blood was taken into the RBC pipette exactly up to the 0.5 mark and the diluting fluid was immediately drawn up to the mark 101. The pipette was rotated between the thumb and the forefinger. This gave a dilution of 1:200. The cover glass was placed in position over the ruled area using gentle pressure. The suspension was mixed thoroughly by rotating the pipette for about a minute, holding it in horizontal position, and finally shook it sidewise [11]. The fluid was expelled from the stem of the pipette and filled the chamber immediately by holding the pipette at an angle of 45° and slightly touching the tip against the edge of the cover glass. There should not be any bubbles under the cover glass. Then the red corpuscles were allowed to settle for 2 to 3 minutes. The number of RBCs was counted in 180 small squares (4 squares of 16 at each four corners and one of 16 at centre).

Estimation of white blood cells: The whole blood was taken up to the mark 0.5 in WBC pipette and diluted up to the mark 11 with WBC fluid as described in RBC counting and filled the counting chamber in the same manner. Then the cells area was allowed to settle for 3 minutes. The neubaur counting chamber was used to count the cells in the four comers and each of these 4 sq mm. areas is subdivided into 16 squares by using the low power objective and a medium ocular [12]. While counting, the cells included were those touching the lines on the left and bottom. The difference between the two squares millimeter area as thousand cells /mm blood.

Estimation of lactate dehydrogenase:

\Placed 1.0ml buffered substrate and O.lml sample into each of two tubes. Added 0.2ml water to the blank. Then to the test added 0.2ml of NAD. Mixed and incubated at 37°C for 15 mins. Exactly after 15 mins, 1.0ml of dinitrophenyl hydrazine was added to each (test and control). Left for further 15 mins. Then added 10ml of 0.4N Sodium hydroxide and the color developed was read immediately at 440 nm. A standard curve with sodium pyruvate solution with the concentration range 0.1 -l.O umole was taken. LDH activity in serum was expressed as umoles of pymvate liberated / L and in liver homogenate as nmoles of pyruvate liberated / minute / rng protein [15].

Estimation OF Creatine Kinase: The incubation mixture containing 0.75ml of double distilled water, 0.05ml of serum, 0.1ml of ATP solution, O.lml of magnesium-cysteinereagent and 0.1ml of creatine was incubated at 37°C for 20mins [13]. The tubes were centrifuged and the supernatant was used for the estimation of phosphorus. The enzyme activity is expressed as IU/L.

Estimation ofcreatine kinase-MB activity: To the test tubes added of the reagent and50filof the sample. The mixture was mixed and incubated at 37C. The absorbance wasmeasuredafter300 seconds. Two additional absorbance wastakenat1minuteinterval.The mean absorbance change/minutes (AA/min) was calculated. The change in absorbance/ minute was multiplied by factor 3376 that is equal to CK-MB.

DPPH Free radical-scavenging activity: The methanolic solution of DPPH (0.1 mM, 1 ml) was incubated with 3 ml of different concentrations of the root extract ranging from 10-100 µg/ml. Incubation was carried out at room temperature (25^0C) for 30 min. For each concentration, the assay was run in triplicate [14]. At the end of the incubation period, the optical density of each sample was determined at 517 *nm*. Ascorbic acid solution was used as a standard. EC_{50} values (concentration required to scavenge 50% of the free radicals) for both ascorbic acid and the root extract were determined. The radical scavenging activity of the tested sample was expressed as an inhibition percentage (IP) [102].

DPPH Scavenged (%) = $(A_{DPPH} – A_{test} / A_{DPPH}) \times 100$

Where,

A_{DPPH} is the absorbance of the 0.1 mM of DPPH solution and

A_{test} is the absorbance in the presence of the extract or ascorbic acid.

IC_{50} value was determined from the graph obtained using standard ascorbic acid by using the "y = mx + c" formula from the slope of the graph.

Statistical Analysis: Results are expressed as Mean ± S.D .all the results were compared with control subject one-way analysis of variance (ANOVA), followed by the dunnet t-test using Graph Pad Prism Software 6 version. P Values < 0.05 were as considered statistically significant.

3. Results and Discussion

% Yield of Methanolic Extract from Aerial Parts of *Cynodon dactylon* was found to be 34.75 % Yield value of Phenolic Extract from Aerial Parts of *Cynodon dactylon* was found to be 8.6.

Preliminary Phytochemical Screening:

Investigation revealed the presence of Alkaloid, Tannin, Saponin, Phenol in Methanolic Extract of *Cynodon dactylon* while only Phenol were present in Phenolic Extract of *Cynodon dactylon*.

Table.no 1: Preliminary Phytochemical Screening

Phytochemical	Results
Steroid	Absent
Alkaloid	Present
Tannin	Present
Carbohydrate	Absent
Phenol	Present
Flavonoid	Present
Saponin	Present

Acute toxicity studies:

As per (OECD) draft guidelines 423 adopted, Female albino rats were administered with *Cynodon dactylon* and doses was be selected in the sequence (1.75- 5000) using the default dose progression factor, for the purpose of toxicity study. Animals are observed individually at least once during the first 30 minutes after dosing, periodically during the first 24 hours and daily thereafter, for a total of 14 days,. In all the cases, no death was observed within 14 days. Overall results suggested the LD_{50} value as 5000 mg/kg. Hence therapeutic dose was calculated (i.e. 400mg/kg and 500 mg/kg) of the lethal dose for the purpose of antihyperlipidemic investigations.

Effect of *Cynodon dactylon* Extractson Serum Total Cholesterol levels:

In the Normal rats the Total Cholesterol levels were found to be 64.03 ± 1.45 on 0^{th} day respectively. Treatment with Triton-X-100 caused a significant rise in the levels of Total Cholesterol in Group-II, Group-III Group-IV Group-V Group-VI (i.e Hyperlipidemic Control, MECD 500 mg/kg, PECD 400 mg/kg, PECD 500 mg/kg, & Standard (Atorvostatin) 10mg/kg) and the levels were found to be 192.47 ± 5.05, 175.28 ± 4.43, 180.97 ± 5.21,187.86 ± 9.66, and 180.79 ±9.1, respectively. Administration of various doses of the MECD & PECD after the Induction with Triton-X-100 resulted in the decreasing of Cholesterol levels. The total cholesterol levels of groups treated with MECD at dose of 500mg/kg were 134.19 ± 3.5, and group treated with PECD at dose of 400mg/kg & 500mg/kg were 121.74 ± 7.74 and 112.97 ± 5.25 respectively. and lowering of cholesterol was dose dependent manner in PECD. In Standard (Atorvastatin) group the total cholesterol was reduced to 92.29 ± 5.63.

Effect of *Cynodon dactylon* Extractson Serum Triglyceride levels: In the Normal rats the Triglycerides levels were to found be 82.66 ± 2.46 on 0^{th} day respectively. Induction of hyperlipidemia resulted in significantly raised in Triglyceride levels in Group-II, Group-III Group-IV Group-V Group-VI (i.e Hyperlipidemic Control, MECD 500 mg/kg, PECD 400 mg/kg, PECD 500 mg/kg, & Standard Atorvostatin 10mg/kg). and the levels were found to be 168.9 ± 5.28, 136.43 ± 7.74, 138.46 ± 1.61, 144.11 ± 7.12, and 148.78 ± 10.23, respectively. The triglyceride values of hyperlipidemic rats treated with MECD at dose of 500mg/kg were found to be 117.57 ± 5.25 and PECD at dose of 400mg/kg and 500mg/kg were 107.93 ± 6.67 and 103.55 ± 4.2. Administration of various doses of the PECD was able to produce a dose dependant decrease in the triglyceride levels and lowering of triglycerides was dose dependent manner in PECD. In Standard (Atorvastatin) group the triglycerides was reduced to 102.26 ± 7.68.

Effect of *Cynodon dactylon* Extracts on Serum LDL-C levels: In the Normal rats the LDL-C levels were to found be 8.45 ± 3.43 on 0^{th} day respectively. Treatment with Triton-X-100 caused a significant rise in the levels of LDL-C in Group-II, Group-III, Group-IV, Group-V, Group-VI (i.e Hyperlipidemic Control, MECD 500 mg/kg, PECD 400 mg/kg, PECD 500 mg/kg, & Standard Atorvostatin 10mg/kg) and the levels were found to be 136.82 ± 7.00, 122.7 ± 10.93, 132.3 ± 5.05, 139.8 ± 3.44, 130.52 ± 7.98. Administration of various doses of the MECD & PECD after the induction of Titon-X-100 resulted in the decreasing of LDL-C levels. The LDL-C levels of groups treated with MECD at dose of 500mg/kg were 83.58 ± 5.26, and Groups treated with PECD at dose of 400mg/kg & 500mg/kg were 69.11 ± 10.51 and 59.1 ± 6.89 respectively. and lowering of LDL-C was dose dependent manner in PECD. In Standard (Atorvastatin) group the LDL-C was reduced to 32.91 ± 7.61. The reduction in LDL-C level by MECD and PECD was significant at ($p<0.01$).

Effect of *Cynodon dactylon* Extracts on Serum VLDL-C levels: The VLDL-C levels in Normal rats at 0^{th} were found to be 16.5 ± 0.5. Administration of Triton-X-100 resulted in a rise in VLDL-C levels. Treatment with Triton-X-100 caused a significant rise in the levels of VLDL-C in Group-II, Group-III, Group-IV, Group-V, Group-VI (i.e Hyperlipidemic Control, MECD 500 mg/kg, PECD 400 mg/kg, PECD 500 mg/kg, & Standard Atorvostatin 10mg/kg) and the levels were found to be 33.79 ± 1.05, 27.28 ± 1.54, 27.69 ± 0.32, 28.97 ± 1.4229.75 ± 2.05. Administration of various doses of the MECD & PECD after the Induction with Titon-X-100 resulted in the

decreasing of VLDL-C levels. The VLDL-C levels of groups treated with MECD at dose of 500mg/kg were 23.51 ± 1.05, and group treated with PECD at dose of 400mg/kg & 500mg/kg were 21.58 ± 1.33 and 20.71 ± 0.84 respectively. and lowering of vldl-c was dose dependent manner in PETP. In Standard (Atorvastatin) group the VLDL-C was reduced to 20.44 ± 1.53. The reduction in cholesterol level by MECD and PECD was significant at ($p<0.05$).

Effect of *Cynodon dactylon* on Serum HDL-C levels: The HDL-C levels in normal rats at 0^{th} were found to be 38.91 ± 2.33. Treatment with Triton-X-100 caused a significant fall in the levels of HDL-C in Group-II, Group-III, Group-IV, Group-V, and Group-VI (i.e Hyperlipidemic Control, MECD 500 mg/kg, PECD 400 mg/kg, PECD 500 mg/kg, & Standard Atorvostatin 10mg/kg). And the levels were found to be 21.86±2.74, 25.3 ± 4.94, 20.98 ± 0.48, 19.01 ±4.29 and 20.53±0.93. Whereas groups treated with MECD at dose of 500mg/kg were 27.1 ± 2.99 and groups treated with PECD at dose of 400mg/kg and 500mg/kg showed a dose dependant increase in the HDL-C levels 31.04 ± 4.32 and 33.15 ± 2.51 respectively. In Atorvastatin group the HDL-C was elevated to 39.18 ± 3.14.

Haematological Parameters:

Alterations in blood parameters may be due to changes in cellular integrity and membrane permeability of cells or even due to exposure to toxic chemicals (Hoffbr and and Petrit 1997). Results of the haematological studies data showed that all the hematological parameters for the control rats were not significantly different ($P<0.05$) from those treated with the phenolic extract of Cynodon dactylon. As far as the haematological and biochemical parameters are concerned, no index of significant alteration in relation to control appeared, after 14days of treatment.

Marker enzymes namely (LDH) were determined after administration of MECD and PECD did not produce any deleterious alteration in the levels of LDH in both serum and liver of extract treated rats compared to control rats. It was found that most of the parameters were slightly changed with respect to control group rats but remain within the normal range. This indicates insignificant adverse effect of MECD and PECD on liver function.

The significant ($P<0.05$) rise observed in the activities of diagnostic marker enzyme namely CK and CK-MB in the serum of Doxorubicin administered rats as compared to that of control rats is an indication of the severity of the necrotic damage to the myocardial membrane. MECD and PECD pretreatment at all the three doses significantly reduced (PO.05) the activities of the marker enzymes as compared to the rats treated with DOX alone. This reduction in enzyme activities could be due to its action on maintaining membrane integrity. There by restricting the leakage of this enzyme. The activities of both CK and CK-MB in heart tissues were decreased on DOX treatment where as it reverted back towards normalcy on treatment with phenolic extracts of MECD and PECD at all the selected concentrations.

***In vitro* anti oxidant Activity:**

DPPH free radical scavenging activity:

DPPH is a relatively stable free radical which when encounters proton donors' such as antioxidants, the radicals get quenched and absorbance gets reduced. Results indicated definite scavenging activity of the extract towards DPPH radicals when compared with standard ascorbic acid. IC_{50} value for standard Ascorbic acid was found to be 43.137µg/ml, where as the IC_{50}value for phenolic extract of *PECD* was found to be 41.024 µg/ml.

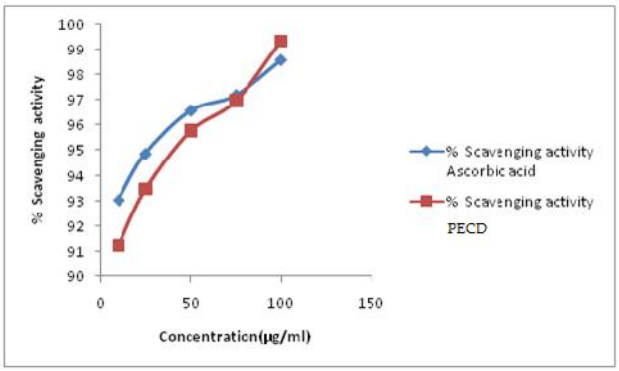

Figure 8: Anti oxidant activity of PECD& ascorbic acid

Discussion: The present study was designed to investigate the cardioprotective activity of *Cynodon dactylon* extract in Triton X-100 induced hyperlipidemic rats. Phytochemical Investigation revealed the presence of Alkaloid, Tannin, Saponin, Phenol in Methanolic Extract of *Cynodon dactylon* while only Phenol were present in Phenolic Extract of *Cynodon dactylon* %Yield value of Methanolic Extract from Aerial Parts of *Cynodon dactylon* was found to be 34.75 % Yield value of Phenolic Extract from Aerial Parts of *Cynodon dactylon* was found to be 8.6 %. Administration of Triton-X-100 (100mg/kg) to all the fasted rats caused an elevation of TC,TG, VLDL and LDL and reduction in HDL levels. After 72 hrs of induction of Triton X-100 results in hyperlipidemia which is compared with normal control group .which results in significantly increased serum lipid levels in hyperlipidemic group.

The change in lipid levels in group number III to VI, were comparable with group of Hyperlipidemic control (i.e Triton X-100 ,Group- II) . The Standard group (i.e Atorvostatin group) significantly lowers the serum lipid level ($P<0.001$). The results of the study clearly indicate that MECD Extract and PECD Extract at a dose of 500 mg/kg & 400 mg/kg significantly lowered serum lipid levels ($P<0.01$). PECD Extract at a dose of 500 mg/kg significantly lowered serum lipid levels, ($P<0.001$) i.e. antihyperlipidemic activity which was found to be more effective in higher dose of PECD as compared to MECD and lower dose of PECD when administered orally in triton induced hyperlipidemic models.

MECD Extract having very low hypolipidemic activity. PECD Extracts showed a dose dependant decrease in the levels of cholesterol, Triglyceride, LDL-C and VLDL-C level. Among three groups (i.e. group number III-V), Group number- V reduced the elevated lipid levels more

significantly than the other Groups.(P<0.001). Flavonoids have exhibited a variety of pharmacological activities, including the antiatherogenesis and antioxidant effect[48]. Thus the present result strongly suggests that the hypolipidemic activity of this medicinal plant could be attributed to the presence of Tannis, Phenols, Flavonoids. in the Extracts. The changes in Hb content, RBC, platelets, and differential count of WBC due to Doxorubicin administration were found to be reversed in the rats pretreated with different doses of ethanolic extracts which indicates that ethanolic extracts possessed protective action on the haemopoietic system.

The effect of the extract on the levels of cardiac marker enzymes CK, CK-MB, troponin-T, AST, ALT, LDH, and ALP in both serum and heart was assessed. In case of marker enzymes, DOX was showing significant increase in the activities of CK, CK-MB, troponin-T, AST, ALT, LDH and ALP (important markers of myocardial infarction) in serum with subsequent decrease activities of CK, AST, ALT, and LDH in heart. In heart tissue, the levels of CK-MB and ALP were found to be increased, in DOX induced rats when compared with control rats. Increase in the activities of these enzymes in serum could be due to the leakage of these enzymes from the heart as a result of DOX-induced necrosis. The changes in the activities of these enzymes were reverted back to near normal in the rats pretreated with different concentrations of ethanolic extracts

4. Conclusion

Natural products extracts of therapeutic relevance are of paramount importance as reservoirs of structural and chemical diversity. A recent review on national pharmacopoeias from several countries reveals at least 120 distinct chemical substances from different plants that have utility as life saving drugs. This has been achieved through chemical and pharmacological screening of only 6% of the total plants pecies. Untapped, hidden wealth in the flora needs to be unearthed and explored to cure diseases like heart disease, cancer, diabetes, AIDS etc. Most countries face high and increasing rates of cardiovascular disease. Combating these cardiovascular diseases is of a paramount importance today. Ischemic heart disease leading to myocardial infarction (MI) is a major clinical concern and remains as a clinical challenge and a problem of great importance despite considerable advances in therapy and management that have been made over the pastthreedecades. MI continues to beamajorpublic health problem, not only in western countries but also increasingly in developing countries and makes significant contribution to the mortality statistics. The results concluded that PEAG (500 mg/kg) have definite antihyperlipidemic activity in Triton X-100 induced hyperlipidemic model and which is equipotent activity when compared with Atorvastatin treated groups. And also proved to be cardio protective in the respective concentrations further studies on this extract may lead to identify the possible mechanism of action and isolation of active principle from the same.

Table 2: Lipids Levels Obtained on 8th Day (After Treatment)

GROUPS	TC	TG	HDL	LDL	VLDL
Normal Control	64.03 ± 1.45	82.66 ± 2.46	38.91 ± 2.33	8.45 ± 3.43	16.53 ± 0.49
Hyperlipidemic Control	192.47 ± 5.05	168.9±5.28	21.86±2.74	136.82±7.00	33.79±1.05
MECD 500mg/kg	134.19 ± 3.5*	117.57 ± 5.25*	27.1 ± 2.99***	83.58± 5.26*	23.51 ± 1.05***
PECD 400mg/kg.	121.74 ±7.74*	107.93 ± 6.67*	31.04 ± 4.32**	69.11± 10.51***	21.58 ± 1.33***
PECD 500mg/kg.	112.97 ±5.25*	103.55 ± 4.2*	33.15 ± 2.51**	59.1± 6.89*	20.71 ± 0.84***
Standard Atrovastatin 10mg/kg	92.29 ±5.63*	102.26 ± 7.68*	39.18 ± 3.14**	32.91± 7.61*	20.44 ± 1.53**

All the data are expressed as MEAN ± S.D (n=4),*P = < 0.001, **P = < 0.01, ***P = < 0.05. vs GROUP .**IITC**: Total Cholesterol ; **TG**: Triglycerides ; **HDL-C** : High Density Lipoprotein cholesterol; **LDL-C** : Low Density Lipoprotein-cholesterol ; **VLDL-C** : Very Low Density Lipoprotein ; **MECD:** Methanolic Extract of *Cynodon dactylon* .; **PECD:** Phenolic Extract of *Cynodon dactylon.*

Table 2: Effect of phenolic extracts on haematological profilein bloodofcontrol and experimental rats

Parameters	Group I	Group II	Group III	Group IV	Group V	Group VI
Hemoglobin (g%)	10.99± 0.22	13. 8 ±0.6	11. 92 ±0.09	12.32 ±0.09	12.3±0.04	11.82±0.07
RBC (X10%il)	3.83±0.31	4.60 ±0.09	3.97±0.10	4.09 ±0.20	4.01±0.14	3.93±0.1
PCV	32.21±0.1 l	41.4±0.10	35.62 ±0.09	36.93 ±0.06	35.1±0.6	34.5±0.03
MCV (fl)	84.04±1.04	90.01 ±1.01	89.02 ±0.98	87.08 ±0.17	89.01 ±0.03	86.40 ± 0.99
MCHC (g/dl)	32.62±0.25	33.30 ±0.30	34.32 ±0.42	34.20 ±0.01	34.82 ±0.41	33.00 ±0.50
MCH (Pg)	28.00±0.06	30.00 ±0.02	31.00±0.2	28.00 ±0.6	29.2 ± 0.4	29.1±0.2

PLT (k/ul)	4.48±0.88	15.17±1.99	7.82±0.15	7.75±0.21	4.25 ± 0.03	3.21±0.04

Values are expressed by mean ± SD (n =6)

Table 3: Estimation of blood pressure and heart rate

Sl.No	Groups	Blood Pressure(mmHg)		Heart Rate/Min
		Mean-BP	Systolic-BP	
I	Normal Control	122±0.68	121±0.38	335±0.84
II	Hyperlipidemic Control	149±0.35	151±0.74	275±0.68
III	MECD 500mg/kg	142±0.68	141±0.65	282±0.41
IV	PECD 400mg/kg.	140±0.68	139±0.29	321±0.64
v	PECD 500mg/kg.	132±0.87	130±0.15	325±0.68

Table 4: Activities of liver marker enzymes in serum and liver of control and experimental rats

Sl.No	Groups	LDH	
		Serum	Liver
I	Normal Control	73.58 ±0.68	6.12 ±0.75
II	Hyperlipidemic Control	73.70 ±0.50	6.35 ± 0.84
III	MECD 500mg/kg	73.61± 0.39	6.29±0.81
IV	PECD 400mg/kg.	73.66± 0.38	6.00 ± 0.8
V	PECD 500mg/kg.	73.69± 0.40	6.23 ± 0.74

Table 5: Activities of creatinekinase (CK), creatinekinase MB (CK-MB) in serum and heart of control and experimental rats

Parameters	CK		CK-MB	
	Serum	Heart	Serum	Heart
Normal Control	250.79 ±2.27	181.85 ± 1.14	41.54 ±0.82	106.29 ±2. 45
Hyperlipidemic Control	490.83 ±2.59	1 14.00 ± 1.14	103.97±0.82	62.15 ± 1.96*
MECD 500mg/kg	433.45 ±2.14	145.92 ± 1.57	91.12±0.42	75.58 ± 1.85
PECD 400mg/kg.	363.00 ±2.88	159.09 ±0.93	76.02 ±0.27	81.68±0.93
PECD 500mg/kg	325.08±2.68	165.67 ±1.40	58.54 ±0.67	86.47 ± 1.83

Table No 7: Showing % scavenging activity of Ascorbic acid and PECD

Concentration (µg/ml)	% Scavenging activity	
	Ascorbic acid	PECD
10	93.05	91.23
25	94.87	93.45
50	96.58	95.78
75	97.2	96.98
100	98.58	99.34
IC_{50} (µg/ml)	**43.137µg/ml**	**41.024 µg/ml**

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