PROTECTIVE EFFECT OF CUMIN (CUMINUM CYMINUM L.) SEED EXTRACT ON CARDIOVASCULAR SYSTEM, TOXICITY, AND HEMATOLOGY ON HYPERLIPIDEMIC RABBITS: AN EXPERIMENTAL STUDY

HARSHLATA CHOUGHAN*, ASHOK PUROHIT

Department of Zoology, Jai Narayan Vyas University, Jodhpur, Rajasthan, India. Email: harshlata.chouhan@gmail.com

Received: 18 May 2018, Revised and Accepted: 22 June 2018

ABSTRACT

Objective: The present study is related with the assessment of anti-atherosclerotic efficacy of 70% EtOH extract of cumin seed in diet-induced hyperlipidemic rabbits.

Methods: Rabbits were rendered hyperlipidemic by oral administration of cholesterol for 15 days. Then, the animals were treated with 70% EtOH (ethanolic extract) of cumin seed extract for 45 days (Group III). Another set of animals was treated with atorvastatin, the standard drug for 45 days. At the end of experimental period, the serum biochemical, hematological, and histological analysis of thoracic aorta was done.

Results: The cumin seed extract showed contain hypolipidemic effect by reducing plasma cholesterol, low-density lipoproteins, and triglycerides level. While toxicological studies suggest no adverse effect on renal and liver function tests, hematological parameters were also observed in a normal range. Histological analysis showed that cholesterol administration caused a narrowing of the aortal lumen while treatment with 70% EtOH and atorvastatin decreased the plaque size and restored the luminal size of the aorta to normal.

Conclusion: The present study suggests that commonly used culinary spice cumin seed possesses hypolipidemic and cardioprotective effect with a positive effect on serum biochemistry, histology, and hematology.

Keywords: Atherosclerosis, Hyperlipidemia, Cumin seed, Cardioprotective.

INTRODUCTION

Atherosclerosis underlies the leading reason of mortality and morbidity globally. Initiation of atherosclerosis involves a decade-long expansion of the arterial intima, which occurs due to an intricate interaction of circulatory factors and various cell types in the vessel wall including endothelial cells, lymphocytes, monocytes, and smooth muscle cells [1]. Cardiovascular disease (CVD) causes 1 of every 5 deaths in past years [2]. Current allopathic drug treatment for atherosclerosis involves statin (HMG-CoA reductase inhibitors), fibrates, niacin, and bile acid resins. Among these remedies, the statin is the most widely and reliably used drug as it directly lowers the serum cholesterol level by competitively inhibiting HMG-CoA reductase enzyme in the liver [3]. However, as the main end product of HMG-CoA activity is mevalonate, which acts as a precursor of many other non-steroidal isoprenoid compounds, so the inhibition of this enzyme results in pleiotropic effects and thus long-term consumption of statin causes several side effects [4]. Side effects of statin are labeled as statin-associated symptoms which include muscular damage (statin-associated muscle symptoms), rarely rhabdomyolysis, statin-induced narcotizing myopathy, diabetes mellitus, Alzheimer’s, and dementia [5]. If the condition of atherosclerosis goes more severe, the patients are suggested to undergo cardiovascular surgery. Cardiovascular surgery is an important treatment [6], but these surgical alternatives are very expensive. Hence, there is a need to find an effective, reliable drug with less or none side effects and the answer relies on herbal medicines. Studies suggest that 4 billion people (about 80% population) prefer herbal medicines for some aspects of primary health care [7]. Dietary plants and their ingredients can act effectively for the management of hyperlipidemia as they have been reported to possess lipid digestion and absorption inhibiting constituents [8]. In traditional medicinal system, aromatic plants have always been important and being used for a long time [9]. *Cuminum cyminum* Linn. is one of the important culinary, aromatic plants used as primary medicine for a long time. The present work is focused on exploring the cardioprotective and anti-atherosclerotic effect of cumin seed extract on cholesterol-fed rabbits.

METHODS

Extraction of plant material

Dried and cleaned seeds of the *C. cyminum* (Linn.) were bought from the local market. 70% of ethanolic extract was prepared with the use of Soxhlet apparatus for 24 h. The extract was treated under low pressure and temperature and then distilled to remove excess of ethanol from the extract. After complete removal of ethanol from the extract, it was dried to obtain the brown-colored sticky extract. The extract was stored in desiccation for future use in experiments.

Experimental animals

New Zealand white rabbits weighing between 1 and 1.25 kg were obtained from the certified institute. Animals were acclimatized for 10 days before the onset of the experiment. Animals were kept in clean, metallic wire gauge cages in a room with 12:12 h light-dark cycle, 20–25°C temperature, and 40–50% relative humidity and were fed with standard pellet diet and fresh green vegetables and drinking water. The experimental protocol was approved by the Institutional Animal Ethical Committee (Reg No.: 1646/GO/Re/12/CPCSEA).

Experimentation

Induction of hyperlipidemia

Rabbits were rendered hyperlipidemic by the oral dose of cholesterol powder at the dose of 500 mg/kg.b.wt./day dissolved in 5 ml of coconut oil for 15 days.
Dose regimen of atorvastatin
Atorvastatin (Atorlip-10, Cipla) was used as a standard hypolipidemic drug. It was orally administered at the dose of 0.25 mg/kg b.wt./day dissolved in 5 ml of distilled water.

Preparation of plant drug
70% of ethanolic extract (500mg/kg b.wt.) was then given to the experimental animals by mixing it in 5 ml distilled water. The dose of the extract was determined by LD₅₀ test.

Experimental design
The experimental period was of 60 days and divided into four groups (n=5).
Group I: Intact control
Group II: Hyperlipidemic control
Group III: 70% EtOH of cumin seed treatment group
Group IV: Atorvastatin (Atorlip-10) treatment group.

Assessment of hematology
Blood was collected by direct cardiac puncture at the end of experimental period. The collected blood was stored in EDTA vials at −20°C. Hematological assessments of hemoglobin, TRBC, hematocrit, mean corpuscular volume, mean cell hemoglobin, mean corpuscular hemoglobin concentration, red cell distribution width, total leucocyte count, platelet count, plateletcrit, mean platelet volume, and platelet distribution width were examined using hematologic analyzer through standard methods [10].

Assessment of serum biochemistry
At the end of experimental period, animals were sacrificed under prolonged anesthesia and direct cardiac puncture. Serum was separated by centrifugation of blood at 3000 rpm for 15 min and stored at −20°C for further need. Serum samples were analyzed using Biochm Auto-analyzer RX-50 (Microlab Instrument) for total cholesterol (TC), high-density lipoprotein-cholesterol (HDL-C), and triglycerides (TGs) using commercial diagnostic kits (Siemens Healthcare Diagnostics, USA). Low-density lipoprotein (LDL-C) and very-LDL (VLDL-C) were calculated by Friedewald’s formula [11]. TC/HDL and LDL/HDL were also calculated as ischemic indices to assess the atherogenic risk. The toxicity profile was also performed by standard kits [12].

Histopathology of the aorta
The thoracic aorta was cut (2–3 cm length) and excised from the heart. The tissue was cleaned in 0.9% saline and kept in 10% formalin fixative. The processed aorta was then ultra sectioned (5–6 μm thickness). Sectioned tissues were stained with hematoxylin and eosin stain and examined under the light microscope for histopathological observations.

Statistical analysis
All biochemical parameters were expressed as the mean±standard error of the mean. One-way ANOVA was analyzed using Tukey’s multiple comparison tests. GraphPad Prism-7 was used for data analysis, and graphical representation was made using MS Excel-2007. Level of significance was expressed as p<0.05.

RESULTS
Effect on lipid profile
A drastic increase in serum TC was observed when high cholesterol diet was administered to the animals, and this increase in TC was decreased significantly 85% (p<0.01) in animals treated with 70% EtOH of cumin seed extract. While highly significant (p<0.001) increase in HDL-C concentration was observed in the hyperlipidemic group, this level came back down when treated with 70% EtOH cumin seed extract and statin. Similarly, in the case of LDL-C, 96%, in VLDL-C, 39.61%, and in TG, 64.08% reduction were observed in cumin seed extract-treated groups which are comparable with statin treatment. CHOL/HDL and HDL/LDL ratio of both 70% EtOH cumin seed extract and statin-treated groups were comparable with the control group. Total lipid concentration also showed high significant (83.69%) increase in case of the hyperlipidemic group, while other two groups showed significant decrease, and 74.78% in cumin extract treated and 81.88% in atorvastatin-treated groups were observed.

Effect on serum biochemistry
Serum biochemical analysis suggests that there is a considerable increase in the level of blood sugar in the hyperlipidemic group while cumin seed extract and atorvastatin-treated animals showed almost normal values near to the control group. Some of the other parameters of the liver functional test and the renal function test did not show any considerable variation throughout the experimental period in all the four groups.

Effect on hematology
Observations suggest that no noticeable variation was observed in hematological parameters. All the parameters were near the control values.

Effect on aortic plaque formation (Figs. 1 and 2, Tables 1 and 2)
Histopathological study of ascending aorta shows plaque formation with the presence of foam cells, collagen, and lipid deposition causing thickened intimal lining, resulting in narrowing of the arterial lumen.
while treatment with 70% EtOH extract of cumin seed showed a significant reduction in plaque size and restoratin of arterial wall integrity. These findings were comparable to atorvastatin-treated groups.

**DISCUSSION**

Diet with a high level of cholesterol and fat has shown to promote the progression of atherosclerosis [13,14]. Major risk factors for coronary heart disease include hypercholesterolemia, hypertension, cigarette smoking, diabetes, and high cholesterol-rich and fat diet, which eventually result in impairment of endothelial function which further develops atherosclerosis [13,15]. It is well established that hyperlipidemia is one of the major risk factors in CVD, including atherosclerosis. Managing hyperlipidemia as a way to prevent atherosclerosis is a common activity in the treatment of this disease [16]. Current treatment therapies treat the disease either by lowering the cholesterol production through inhibition of HMG-CoA reductase enzyme in liver (like statins) or by inhibition of cholesterol absorption in the small intestine or the drug may have some anti-inflammatory and antiplatelet aggregation activity which stops plaque progression.

The present study proves that the plant cumin seed has the anti-inflammatory and antiplatelet aggregation activity which stops plaque progression. Administration of cumin seed caused a reduction in serum TC and also shows hypoglycemic effects, and similar findings were observed in the present study also. These findings are supported by another study in which administration of cumin seed powder capsule was reported to lower down the serum cholesterol as well as serum insulin level in diabetes Type-II suffering patients [25].

**ACKNOWLEDGMENT**

We would like to acknowledge Head, Department of zoology, Jai Narain Vyas University, Jodhpur, for providing all the necessary amenities.

**AUTHORS’ CONTRIBUTIONS**

The complete research work was suggested and mentored by Prof. Ashok Purohit. All the experimental work was performed by Harshlata Chouhan. Authors drafted and approved the final manuscript.

**CONFICTS OF INTEREST**

The author declares no conflict of interest.
REFERENCES


