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Original Research Article

The effects of aqueous ginger extract on aluminium chloride ($AlCl_3$) induced alteration in lipid profile of male wister rats

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ABSTRACT

Background: This study was aimed at investigating the effects of aluminium chloride ($AlCl_3$) in altering the serum lipid profiles and ways to reduce its effect using two different doses of ginger extract 500mg/kg and 1000mg/kg body weight in male wister rats.
Methods: The rats were randomly divided into 4 groups consisting of 5 animals in each group. Groups II, III and IV received $AlCl_3$ 100mg/kg bodyweight single dose, Groups III and IV receiving an additional daily oral single dose of ginger plant extract through a stomach tube. All animals were fasted before the treatment. All rats were weighed before the start of the experiment and at the end of the experiment. The blood was collected firstly at the beginning of the experiment, then on the 45th day. The collected blood was left to clot then centrifuged at 3500 rpm for 5 min. The serum was separated and stored at - 80°C for later analyses.
Results: This study shows that a single dose of 100mg/kg aluminium chloride causes a rise in total body weight, TC (total cholesterol), LDL (low density lipoproteins) and TG (triglycerides) levels in the rat, and aqueous Zingiber officinal (ginger) extract reduces this rise in TC, LDL and TG levels in the rats.
Conclusions: Ginger was effective in lowering serum cholesterol levels levels in the ginger treated rats to almost normal value. These results indicate that treatment with aqueous extract of ginger may be effective in lowering lipid levels in AlCl3 induced hyperlipidemia in rats.

Keywords: Aluminium chloride, Hyperlipidemia, Zingiber officinal

INTRODUCTION

Aluminium is the third most abundant element in the earth's crust. Aluminium has many uses, mainly in the form of alloys in packaging, building, construction, transportation and electrical applications. Over 95% of beer and carbonated drinks are packaged in aluminium cans. Cooking in aluminium utensils results in consumption of aluminium along with food. Human exposure to aluminium comes from food and drinking water as well as from pharmaceuticals. The normal average daily intake is 1 to 10 mg for adults.[1] Aluminium content in majority of naturally derived products does not exceed 10 mg/kg (usually 0.1-1mg/kg). This element is consumed by humans mainly through cereals, cheese and salt. Herbs, spices and tea have a naturally high content of aluminium.[2] Aluminium is poorly absorbed following either oral or inhalation exposure and, are essentially not absorbed through the skin. In plasma 80 to 90% of aluminium binds to transferrin, iron-transport proteins for which there are receptors in many body tissues. Aluminium is removed from blood by the kidneys and excreted in urine. High level of Al in diet has led to an increase in the deposition of this metal in tissues such as heart, kidneys, brain and liver which may lead to cardiotoxicity, nephrotoxicity, neutrotoxicity and hepatic dysfunctions.[3] There were indications that aluminium

could induce toxic manifestation such as osteomalacia, gastrointestinal toxicity and Alzheimer's disease.[4,5]

Zingiber officinal is one of the Zingiberaceae family.[6] Z. officinal is commonly called ginger. Ginger rhizome is widely used as a spice worldwide. It has been used as a medicine in Asian, Indian and Arabic herbal traditions.[7] Major active components of Z. officinale are thought to be volatile oils, and phenol compounds such as gingerols, Zingbrene and shogaols.[8,9] Extensive research has been done to clarify ginger's influence on lipid profile; HDLC, LDL-C, TC and Triglycerides (TG).[10] In our knowledge no research was conducted about ginger's effect on aluminium induced altered lipid profile hence this study was conducted.

This study was conducted to investigate the effects of aluminium chloride (AlCl3) in altering the serum lipid profiles and ways to reduce its effect using two different doses of ginger extract 500mg/kg and 1000mg/kg body weight in male wister rats.

METHODS

Chemicals

- Aluminium Chloride was purchased from Sd fine Chem Ltd. From Mumbai.
- Ginger was obtained from the local market.
- Diagnostic kits for the estimation of TC, triglyceride and HDL-C were obtained from Coral Ltd., Goa.

Animals

In this study, Wistar albino adult male rats weighing 200-250gm was collected from central animal house and were housed in polypropylene cages in a room where the congenial temperature 27°C±1°C and 12 hrs light and dark cycles maintained. The animals were allowed to acclimatize to the environment for 7 days and supplied with standard pellet diet and water ad libitum. All animals were handled according to the guidelines approved by the CPCSCA.

Preparation of plant extracts

The fresh rhizomes of ginger were obtained from a local market in Chennai. The raw extract was prepared according to the method used by Elshater et al.[11] Plant parts were separated and washed with distilled water, dried and then grind using blender. Each 30g powdered plant material was extracted by refluxing with 100ml of hot water for two weeks at room temperature with shaking at 150 rpm. The extract was filtered through White canvas and filter paper. The mixture was filtered, then the filtrate was centrifuged and the clear supernatant fraction was separated. The concentration was considered to have 1g/ml based on the weight of the starting material. The extracts were purified by filtration through 0.22µm filter units and kept at -20°C until use.

Experimental design

The animals were divided into four groups and each group consisted of five animals.

- *Group I:* Control. (administered normal saline based on weight of animal)
- *Group II:* Aluminium chloride 100 mg/kg bodyweight.
- *Group III:* Aluminium chloride 100 mg/kg bodyweight single dose+ aqueous ginger extract 500mg/kg bodyweight.
- *Group IV:* Aluminium chloride 100 mg/kg bodyweight single dose + aqueous ginger extract 1000mg/kg bodyweight.

The rats were randomly divided into 4 groups consisting of 5 animals in each group. Groups II, III and IV received $AlCl_3$100 mg/kg bodyweight single dose, Groups III and IV receiving an additional daily oral single dose of ginger plant extract through a stomach tube. All animals were fasted before the treatment. All rats were weighed before the start of the experiment and at the end of the experiment.

Collection of blood samples

Blood samples were collected in clean plain tubes from eye vein of all animals two times and the serum was analyzed in laboratory to determine the serum levels of blood glucose and their biochemical parameters. The blood was collected firstly at the beginning of the experiment, then on the 45th day. The collected blood was left to clot then centrifuged at 3500 rpm for 5 min. The serum was separated and stored at - 80°C for later analyses.

Biochemical study

Estimation of lipid profile

Total cholesterol (TC), HDL-Cholesterol and Triglycerides (TG) levels were estimated spectrophotometrically using commercial diagnostic kits (Coral Ltd., Goa). The calculation of LDL-Cholesterol and VLDL-Cholesterol concentrations in plasma were performed according to the method of Arcol following two equations.

- VLDL-C concentration (mg/dl) = Triglycerides / 5
- LDL-C concentration (mg/dl) = total cholesterol concentration - (VLDL-C +HDL-C)

Statistical analyses

Data for all groups were expressed as mean ± standard deviation. Statistical analyses were performed using one-way analysis of variance (ANOVA) and student t-test. Differences were considered to be statistically significant when P value was <0.05. All statistical analyses were performed with SPSS software.

RESULTS

The body weight at the experiment showed a significant ($p<0.05$) rise in group II ($AlCl_3$ 100mg) (Table 1).

Table 1: Body weight in grams of the rats on day one treatment and at the end of treatment.

Groups	Base Line (day 1)	On 45th day	P value
I	272.40±3.87	274.0±3.67	0.188
II	280.2±5.26	310.4±8.02	0.0005*
III	260.0±7.12	264.8±6.92	0.098
IV	279.0±6.02	282.6±8.82	0.206

*p <0.05 = Significant

Table 2 shows a highly significant ($p<0.001$) rise in the total serum cholesterol TC in group II and a highly significant reduction of TC in group IV ($AlCl_3$ + 1000mg/kg body weight of aqueous ginger extract).

Table 2: Total serum cholesterol in mg/dl of the rats before commencement of treatment and at the end of 45 days.

Groups	Baseline (day 1)	On 45th day	P value
I	71.40±10.52	74.12±8.56	0.16773
II	73.40±6.44	88.25±5.39	0.00176*
III	71.80±4.65	65.02±3.92	0.4986
IV	74.12±6.13	54.96±4.54	0.00111*

*p <0.05 = Significant

HDL cholesterol does not show any significant changes in any of the study groups as shown in Table 3.

Table 3: Serum high density lipoproteins in mg/dl of the rats before commencement of treatment and at the end of 45 days.

Groups	Baseline (day 1)	On 45th day	P value
I	25.10±1.66	24.82±1.92	0.63585
II	23.42±2.01	20.14±6.54	0.83627
III	24.42±1.42	23.54±1.94	0.8161
IV	23.40±3.10	22.54±3.62	0.6883

Table 4: Serum low density lipoproteins in mg/dl of the rats before commencement of treatment and at the end of 45 days.

Groups	Baseline (day 1)	On 45th day	P value
I	57.63±5.81	56.21±4.18	0.75511
II	55.34±2.68	72.86±6.177	0.00188*
III	61.76±5.54	62.40±3.18	0.338
IV	63.06±5.74	51.69±4.17	0.03405*

*p <0.05 = Significant

Animals in group II showed a significant ($p<0.05$) rise in low density lipoproteins LDL, whereas group IV showed a significant ($p<0.05$) reduction as shown in Table 4.

There was a highly significant ($p<0.001$) rise of triglycerides in group II and a significant ($p<0.05$) reduction in both groups III and IV as seen in Table 5.

Table 5: Serum triglycerides in mg/dl of the rats before commencement of treatment and at the end of 45 days.

Groups	Baseline (day 1)	On 45th day	P value
I	97.70±2.09	93.11±4.35	0.96114
II	88.32±2.143	110.0±3.77	0.00011*
III	94.28±2.83	88.50±2.01	0.00514*
IV	89.54±5.10	76.71±2.76	0.00246*

*p <0.05 = Significant

DISCUSSION

Aluminium (Al) is known to be toxic to humans and animals. Its toxicity results to generation of reactive oxygen species which leads to oxidative damage of biomolecules in an organism.[12]

A lipid dystrophy which could be lipotoxic or non-lipotoxic could play a role in the pathogenesis or progression of a plethora of diseases. The present study investigated the effects of a natural substance, like ginger against the harmful effects of $AlCl_3$ in rats. The efficacy of ginger extract may be due to the presence of (ZT) compound that was isolated from ginger, which lowered plasma cholesterol levels in rats and mice by blocking cholesterol biosynthesis.[13] These results are compatible with the results of existing research done, wherein ginger was orally administered to rabbits on a high cholesterol diet, to cause reduction in atherogenesis and lipid levels, by disrupting the absorption of cholesterol from gastrointestinal tract.[14] Ginger's effect may also be due to the pharmacological action of ginger which elevates the activity of hepatic cholesterol-7α-hydroxylase as it is the rate-limiting enzyme in the biosynthesis of bile acids and stimulates the conversion of cholesterol to bile acids.[15] Moreover, ginger antihypercholesterolemic effect may be due to the inhibition of cellular cholesterol synthesis this may be due to the presence of niacin in ginger, niacin causes increased clearance of VLDL, lower TG levels, increase hepatic uptake of LDL, and inhibition of cholesterogenesis.[16-19] Aqueous ginger infusion 5% yielded nearly the same antioxidant activity toward lipid peroxidation as did the synthetic antioxidant butylhydroxyanizole, this may be due to the essential oil content.[20] Also this antioxidant activity may be due to the high polyphenols content and the presence of polyphenolic flavonoids prevents coronary artery disease by reducing plasma cholesterol levels or by inhibition LDL oxidation.[21,22] The main antioxidant active principles in ginger are the polyphenolic compounds which called

gingerols and some related phenolic ketone derivatives.[17] The effect of ginger could also be due to the inhibition or scavenging radicals of rat body in different degrees, or by increasing the antioxidative defense mechanisms of liver cells.[23,24] When ginger was added to the animal diet, a considerable increase in the pancreatic and intestine lipase occurred; lipase is the other key factor that plays a vital role in fat digestion, the previous activity of ginger may be responsible for the TG reduction effect. these findings suggest that ginger may have therapeutic potential as antihypercholesterolaemic agent in itself.[25] This may be a novel finding, because it shows ginger's effect on lipid profile in rats which have been given $AlCl_3$ has not been studied yet, in our knowledge, and compare between two doses of ginger action on models of rats.

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