

GARLIC USED AS AN ALTERNATIVE MEDICINE TO CONTROL DIABETIC MELLITUS IN ALLOXAN-INDUCED MALE RABBITS

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Background: Herbal medicines are widely used because of their effectiveness, less side effects and low cost, so investigation on such agents from traditional medicinal plants has become more important in present day studies on medical sciences. Garlic is one of the most popular herbs used as an anti-diabetic agent. In present study possible anti diabetic effects of garlic were studied in alloxan-induced diabetic male rabbits, compared to normal control and diabetic control male rabbits.

Methods: The blood samples were collected every third day and anti-diabetic effects of garlic were observed every time. The serum cholesterol level and body weight were also studied. With an aqueous extract of garlic (1% solution/Kg) body weight for 30 days significantly lowered serum glucose level (38.88%) and serum cholesterol level (57%). **Results and Conclusion:** The results indicate that garlic possesses a beneficial anti-hyperglycaemic effect in alloxan-induced rabbits.

Keyword: diabetic mellitus, alloxan-induced, Herbal medicines

INTRODUCTION

Garlic (*Allium sativum*) is a member of the Liliaceae family, is one of the most popular herbs used worldwide to reduce various risk factors associated with several diseases.¹ Actually, garlic contains a variety of effective compounds that exhibit anticoagulant (anti-thrombotic)^{2,3}, antioxidant^{4,5}, antibiotic⁶, hypocholesterolaemic⁷ and hypoglycaemic as well as hypotensive activities.^{8,9} Most of the studies showed that garlic can reduce blood glucose levels in diabetic mice, rats and rabbits.¹⁰ Augusti and Sheela consistently showed that S-allyl cysteine sulphoxide, (allicin), a sulphur-containing amino acid in garlic (200 mg/kg body weight), had a potential to reduce the diabetic condition in rats almost to the same extent as did glibenclamide and insulin.^{11,12} Old garlic extract was also effective in preventing adrenal hypertrophy, hyperglycaemia and elevation of corticosterone in mice made hyperglycaemic by immobilization stress.¹³ Staba and co-workers have reported that the chemicals present in a garlic product are largely dependent on the processing conditions, such as temperature, duration of preparation, and extraction solvents used.¹⁴ In humans, the hypoglycaemic effect of garlic is not well studied while many reporters have shown a significant effect of garlic on blood glucose in normal individuals but not in diabetic patients. Thus the role of garlic in diabetes in humans is yet to be confirmed.¹⁵

The aim of the present study was to investigate the potential of an aqueous extract of raw garlic in controlling serum glucose, cholesterol, and body weight level in alloxan induced diabetic male rabbits, compared to normal control and diabetic control male rabbits.

MATERIAL AND METHODS

Extract preparation

Aqueous garlic extract was prepared from locally available garlic bulbs. The garlic bulbs were peeled on crushed ice. Then 50 g of the peeled garlic was cut into small pieces and homogenized in 70 ml of cold, sterile 0.9% NaCl in the presence of some crushed ice. The homogenisation was carried out in a blender at high speed using 30-second bursts for a total of 10 minutes. The homogenised mixture was filtered 3 times through cheesecloth, the filtrate was centrifuged at 2000 RCF for 10 minutes and the clear supernatant was diluted to 100 ml with normal saline. The concentration of this garlic preparation was considered to be 500 mg/ml on the basis of the weight of the starting material (50 g/100 ml). The aqueous extract of garlic was stored in small aliquots at 4 °C until use.

Treatment of Diabetic Rats

Male rabbits, taken from Institute of Biotechnology and Genetic Engineering, University of Sindh, Jamshoro, Pakistan, and maintained on a normal diet and filtered tap water, were used in the experiment. For baseline data, blood was drawn from all animals by marginal vein of ear and allowed to clot. Immediately, the clotted blood was centrifuged at 3,500 RPM for 30 minutes. The serum was separated and stored at -4 °C for later analysis. Fifteen male rabbits weighing 1.4–1.8 kg were taken for the present experimental purpose. After ten days of acclimation, the animals were divided into three groups. Each group was separately housed. Free access to water and standard pellet diet was provided to all rabbits. The first group (5 rabbits) was used as control and fed with standard chow and tap water. In remaining ten rabbits, diabetes mellitus was induced by alloxan monohydrate (150 mg/kg) by intraperitoneal administration. When they finish that diet, ordinary food was given to those

rabbits. After seven days, the fasting blood glucose was determined in all the groups. The rabbits with blood glucose level more than 250 mg/dl, were considered as diabetic. Diabetic rabbits were divided into two further groups. One group was fed with garlic solution and other was given simple diet. It was considered as control diabetic group.

Biochemical Assays

For analysis the blood samples were collected every sixth day of experiment, the blood was collected in the centrifuge tube and allowed to clot. After centrifuging at 20,000 rpm for 10 min, the supernatant (serum) was collected and the level of serum glucose and cholesterol were determined by the respective standard assay kit (Merck Germany) and body weight was measured by mechanical balance.

RESULTS AND DISCUSSION

The present study was undertaken to study the anti-diabetic activity of aqueous extract of garlic in alloxan induced diabetic male rabbits in order to check the scientific effect on serum glucose, cholesterol as well as on body weight in the treatment of diabetes. Administration of alloxan (150 mg/kg) led to about 3-fold elevation of fasting blood glucose levels, which was maintained over a period of 4 weeks. One month of daily treatment with aqueous extract of garlic caused a significant fall in elevated blood glucose levels from 300 to 216 mg/dl (38.88%) in diabetic rabbit (Figure-1). Vehicle control animals were found to be stable in their glucose level. It is well established that alloxan administration to experimental rabbits selectively causes pancreatic β cell-membrane disruption and cytotoxicity after its intracellular accumulation.¹⁶ The anti-hyperglycaemic activity caused by garlic extract is due to the presence of Flavonoids and sulphur containing compounds in garlic.¹⁷ Figure-2 shows the effect on garlic on serum cholesterol level and it is clearly observed that the serum cholesterol level was increased after the injection of alloxan (57%) in diabetic rabbits but after consistently treating with aqueous extract of garlic with normal diet, the shooting rate of serum cholesterol was 25% in non treated animals where as it was 13.6% in treated animals. It is 12% less than untreated normal control and alloxan induced hyperglycaemic level. It is also reported that this is due to antioxidant property of the garlic which decrease the free radicals and resulting in lipid oxidation inhibition.⁸ Normal rabbits gained weight significantly throughout the experimental period, while both the control diabetic and garlic-treated diabetic animals had significantly lower body weights when compared to normal animals (Figure-3). However, garlic-treated diabetic rabbits maintained their initial weights during the 4 week

treatment period although at the end of the experiment their body weights were significantly less than those of normal rabbits. In contrast, the control diabetic rats showed significant weight loss when compared to both the normal rabbits and the garlic-treated diabetic rabbits at the end of the 4-week experiment.

Garlic has Hypoglycaemic and Hypolipidemic effect on diabetic rabbits but further extensive investigations including histopathology and blood insulin level determination are needed to confirm and clear the mechanisms involved in hypoglycaemic effect of garlic in diabetic mellitus. Reduced blood glucose and cholesterol level observed in the present study agree with the reports showing the hypoglycaemic effect of garlic and effect of insulin on atherosclerosis and dislipidemia respectively. Our results confirmed that raw garlic has significant hypoglycaemic, hypocholesterolaemic and hypolipidaemic effects. Therefore, the present study reinforces the findings of previous studies that garlic had a significant effect in reducing blood glucose.^{14,15,18-20}

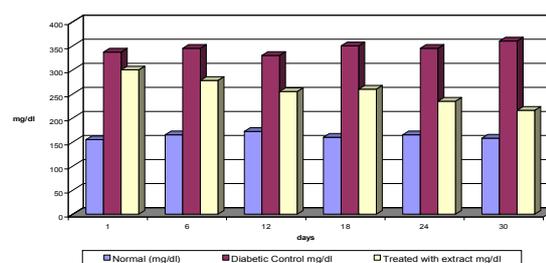


Figure-1: Effect of garlic extract on serum glucose level in alloxan induced diabetic male rabbits

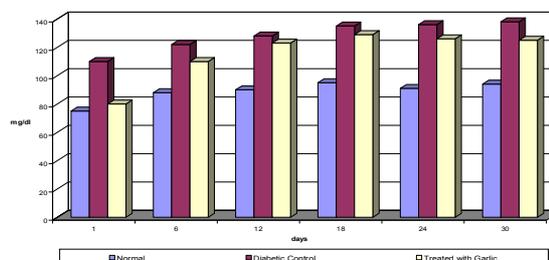


Figure-2: Effect of garlic extract on serum cholesterol level in alloxan induced diabetic male rabbits

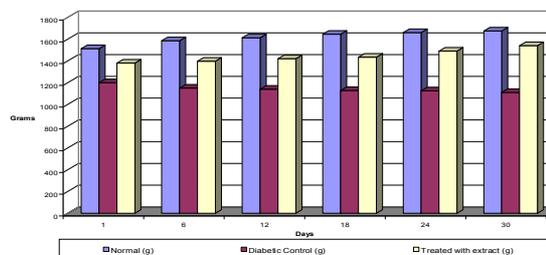


Figure-3: Effect of garlic extract on body weight in alloxan induced diabetic male rabbits

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