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Efficiency of Garlic and Ginger Mixture on Blood Glucose Level and Lipid Profile of Alloxan Induced Diabetic Mice

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Abstract

The raw garlic (*A. sativum*) and ginger (*Z. officinale*) mixture have been used in the experiment to check any effect in lowering blood glucose and lipid profile in alloxan induced diabetic mice.For the study 25 mice were used. They were maintained at a temperature of 25 ± 1°C and relative humidity of 45 to 55% under 12-h light: 12-h dark cycle. They were fed with normal diet and water *ad libitum.* First of all diabetes was induced in mice by a single intraperitoneal injection of aqueous alloxan monohydrate (40 mg/kg, i.p.) solution. After 72 hrs animals showing serum glucose level above 180 mg/dl (diabetic) were chosen for the study. The experimental animals were randomly divided into 5 groups; having five mice in each group:

Group 1 Normal control mice

Group 2 Diabetic control mice injected with 40mg/kg b.w. of alloxan

Group 3 Diabetic mice treated with 10g/100g diet (5g+5g) of garlic and ginger mixture

Group 4 Diabetic mice treated with 20g/100g diet (10g+10g) of ginger and garlic mixture

Group 5 Diabetic mice treated with 30g/100g diet (15g+15g) of the garlic and ginger mixture.

Blood glucose level of all treated mice including normal control and diabetic control groups was checked weekly. At the end of the experiment, the mice were slaughtered and blood samples were obtained for analyzing lipid profile. From this study it was noted that the diabetic mice treated with garlic and ginger mixture showed remarkable decline in blood glucose level after 72 h compare to the diabetic control diabetic rats. Maximum blood glucose reduction was seen in G5 given 30g/100g garlic and ginger mixture this significant reduction may lead to hypoglycemia. Effect of garlic and ginger mixture on cholesterol, HDL, LDL and triglycerides

level in diabetic mice of G3, G4 and G5 was significant ($p<0.05$). Diabetic control mice showed higher level of cholesterol, LDL and triglycerides while low level of HDL. After giving 10, 20 and 30 g mixture of garlic and ginger mixture the level of cholesterol, LDL and triglycerides were decreased whereas, significant increase in HDL level was seen. Most effective dose was 30g which showed the maximum reduction in cholesterol, LDL and triglyceride level of mice of G3 but exhibited higher and normal level of HDL.

It is concluded that the consumption of garlic and ginger mixture created a beneficial hypoglycaemic and hyperlipidemic effects in diabetic mice.

Keywords

Alloxan, diabetes, blood sugar, HDL, LDL, Triglycerides, cholesterol

Introduction

Diabetes mellitus and its allied discrepancies is one of the prominent menaces of developing economics. Pakistan is at 6th position however, at the end of the year 2030; approximately 376 million people will be suffered (Wild *et al.,* 2004). Diabetes is a metabolic syndrome that steadily affects different physiological systems of the human body. It is one of the foremost causes of mortality and, if uncontrolled, can threat multi-organs system (Zakir *et al.,* 2008). Uncontrolled blood glucose is thought to be the fundamental aspect in the onset of diabetic difficulties of both type 1 and type 2 (American Association of Diabetic Educators, 2002). Most common type is Type 2 category, while Type 1 diabetes develops in early childhood. Main reasons include sedentary lifestyles, energy rich diet, lack of physical exercise and obesity (Yajnik, 2001).

Diabetes is mainly characterized by relative insufficiency in secretion of insulin or insulin action associated with hyperglycemia and malfunctioning in the metabolism of carbohydrate, lipid and protein. It may also leads to various other complications like cardiovascular disorders, oxidative stress and immune dysfunction may develop (Rana *et al.,* 2007). Cardiovascular associated risks are the prominent causes of morbidity and mortality all over the world. Increased

cholesterol level and LDL oxidation trigger events that initiate atherosclerosis (Matsuura *et al.,* 2008).

To cope with this situation a number of herbal medicines for diabetes mellitus and its allied diseases have been emerged (Alarcon-Aguilara *et al.,* 1998). Drug treatment is obligatory nevertheless, accompanied by various side effects and their effectiveness decreases with the passage of time (Zakir *et al.,* 2008). Physical exercise and diet selection is one of the significant strategies to manage diabetes and its allied complications including immune dysfunction, degenerative and cardiovascular disorder. *Allium sativum, Zingiber officinale* and their bioactive constituents hold insulinotropic properties playing significant role in maintaining β cells helpful to address the menace.

Garlic (*Allium sativum*) is an essential vegetable that has been widely utilized as seasoning, flavoring, culinary and in herbal remedies (Rivlin, 2001). Garlic has been exhibited to have dynamic biological actions including antithrombotic, antidiabetic, anticarcinogenic, antiatherosclerotic and a variety of other biological functions (Augusti, 1996). Scientific investigations have depicted that it contains 65% water, 30% carbohydrates along with 5 % of other bioactive components mainly sulfur containing compounds (Milner, 2001). Its important constituents are classified as; sulfur containing compounds and non sulfur containing compounds. Among these organosulphur compounds particularly cysteine sulfoxides and thiosulfinates have greater importance (Tapiero *et al.,* 2004). Allicin (diallylthiosulfinate) and S-allay cysteine are the main thiosulfinates out of which 60-80% is allicin (Lawson *et al.,* 2001). Garlic and its various preparations have potential to lower total plasma cholesterol, reduction in blood pressure and alleviation of blood glucose level (Sterling and Eagling, 2001).

Some studies confirmed anti hyperglycemic effects of garlic (Eidi *et al.,* 2006). Garlic may act on blood glucose through various mechanisms and therefore directly lowers blood glucose level by exciting glycogenisis and preventing glycogenolysis and gluconeogenisis in muscles and hepatic (Ebomoyi *et al.,* 2010). The fiber of garlic may also hamper carbohydrate absorption; thereby affecting blood glucose (Jelodar *et al.,* 2005). Antioxidant property of garlic is another possible mechanism that makes it a contender as antidiabetic agent (Queiroz *et al.,*

2009). Antioxidant action of S-allyl cysteine sulfoxide, product isolated from garlic is considered to have antiglycation properties. Different supplementations of garlic hold remarkable effect on cholesterol level, LDL cholesterol and HDL cholesterol and also contributed toward the prevention of atherosclerosis process (Lau, 2006). Garlic juice exhibits hypoglycemic and antioxidant potential to lower the renal and liver damage (El-Demerdash *et al.,* 2005).

Ginger is also very effective for lowering blood sugar, cholesterol and triglyceride levels (Bhandari *et al.,* 1998). Ginger (*Zingiber officinale*) commonly called "Adrak" belongs to family Zingiberaceae. It is used in both ways as food additives (Flavor) or as a medicine and it is valuable in treating or preventing a diversity of human health associated complications including migraine headache, hepatotoxicity, high cholesterol level, burns, peptic ulcer, nausea, vomiting and motion sickness (Robbers and Tyler, 2002). Chemical constituents of ginger are camphene, cineol, zingiberine, gingerol and β-phellandrene (Shinwari *et al.,* 2006).

Ethyl acetate extract of ginger produced significant reduction in glucose concentration and also increased the serum HDL cholesterol and significantly reduced serum total cholesterol and triglycerides; also, the extract prevented lipid peroxidation in tissues and showed a considerable lipid lowering action in diabetic rats (Goyal and Kadnur, 2006).

Acute dose of aqueous extracts of *Z. officinale* rhizome shows hypoglycaemic activity (Kalejaiye *et al.,* 2002). Ginger also plays key role in glucose removal in insulin receptive peripheral tissues, which is crucial in sustaining blood glucose homeostasis (Li *et al.,* 2012).

Researchers have found that numerous plant products exhibited unique actions against some ailment conditions in diabetic animal model and Pakistan is blessed with many of these curative plants which have been used for the treatment of various diseases. The objective of this study was to inspect the efficiency of garlic and ginger mixture in controlling elevation of blood glucose levels and lipid profile in the alloxan-induced diabetic mice.

Materials and Methods

The research work was done under the support of Institute of Rural Home Economics and National Institute of Food Science and Technology, University of Agriculture, Faisalabad.

Plant Material

The garlic (*A. sativum*) and ginger (*Z. officinale*) used for the experiment were purchased from the Ayub Agricultural Research Institute, Faisalabad.

Animal Model

For this study 25 mice were purchased from National Institute of Health, Islamabad and kept in the animal house of the National Institute of Food Science and Technology (NIFSAT), University of Agriculture, Faisalabad. They were maintained at a temperature of 25 ± 1°C and relative humidity of 45 to 55% under 12-h light: 12-h dark cycle. They were fed with normal diet and water *ad libitum*.

Induction of Diabetes Mellitus

Diabetes was induced in mice by a single intraperitoneal injection of aqueous alloxan monohydrate (40 mg/kg, i.p.) solution. After 72 hrs animals showing serum glucose level above 180 mg/dl (diabetic) were chosen for the study.

Sample Size

The experimental animals were randomly divided into 5 groups; having five mice in each group and mice of each group were placed separately by making five sections in each cage:

Group 1 Normal control mice

Group 2 Diabetic control mice injected with 40mg/kg b.w. of alloxan

Group 3 Diabetic mice treated with 10g/100g diet (5g+5g) of garlic and ginger mixture

Group 4 Diabetic mice treated with 20g/100g diet (10g+10g) of ginger and garlic mixture

Group 5 Diabetic mice treated with 30g/100g diet (15g+15g) of the garlic and ginger mixture.

Blood glucose level of all treated mice including normal control and diabetic control groups was checked weekly. At the end of the experiment, the mice were slaughtered and blood samples were obtained for analyzing lipid profile.

Analytical Methods

Blood glucose was determined by glucose glucometer Accuchek Active®. Plasma total cholesterol was checked using the CHOD-PAP method determined by Stockbridge *et al.* (1989), HDL-cholesterol was estimated by HDL Cholesterol Precipitant method (Assmann, 1979) while LDL-cholesterol and triglycerides were determined by methods as described by McNamara *et al*. (1990) and Annoni *et al.* (1982) respectively.

Statistical Analysis

All the data obtained were statistically analyzed for mean and standard deviation. Statistical analysis was carried out using SPSS (Version 17). Comparison of means among groups was performed via one way Analysis of Variance (ANOVA) and value was taken as statistically different when p value was less than 0.05.

Results and Discussion

From this study it was noted that the diabetic mice treated with garlic and ginger mixture showed remarkable decline in blood glucose level after 72 h compare to the diabetic control diabetic rats. Maximum blood glucose reduction was seen in G5 given 30g/100g garlic and ginger mixture this significant reduction may lead to hypoglycemia (Table. 1). This reduction may be due to garlic and ginger intake caused β cells of pancreas to produce more insulin, which caused the conversion of glucose into glycogen that resulted in decreased blood glucose level (Guyton and Hall, 2000).

Results of this study were resembled with data obtained by Ahmed and Sharma, (1997) as they studied the effect of garlic and ginger mixture in reducing blood glucose in Wister rats and found the significant decline in the level of blood glucose and elevation in insulin level. Helal *et al.* (2012) also explored that rats having fatty liver pre and post treated with ginger depicted the

reduced serum glucose level. Ginger treatment given for 30 days to streptozotocin induced diabetic rats showed significant decrease in increased blood sugar level (Ramudu *et al.,* 2011).

A garlic study conducted by Jhonson *et al.* (2012) on alloxan induced diabetic rats gave same result as serum glucose level was decreased in diabetic rats after giving garlic extract for 3 weeks to pre treated rats and for 2 weeks to post treated diabetic rats. The dose of 300mg/kg body weight of an aqueous extract of garlic was significant in lowering the blood sugar level in alloxan induced diabetic rats (Ozougwu, 2011). Ashour *et al.* (2011) resulted that orally given 200mg garlic oil according to per kg body weight exhibited drastic increase in serum insulin level.

Contrary findings were reached by Jain *et al.* (1993), Mansell *et al.* (2002) and Mohammad *et al.* (2006) who concluded that garlic had no effect on blood sugar.

Effect of garlic and ginger mixture on cholesterol, HDL, LDL and triglycerides level in diabetic mice of G3, G4 and G5 was significant ($p<0.05$). Diabetic control mice showed higher level of cholesterol, LDL and triglycerides while low level of HDL. After giving 10, 20 and 30 g mixture of garlic and ginger mixture the level of cholesterol, LDL and triglycerides were decreased significantly whereas, significant increase in HDL level was seen (Table 2).

Most effective dose was 30g which showed the maximum reduction in cholesterol, LDL and triglyceride level of mice of G3 but exhibited higher and normal level of HDL.

The results of this parameter resembled with results of study conducted by Ahmed and Sharma, (1997) and Ademola *et al* ,(2009) who revealed that rats diet containing combination of ginger plus garlic for 4 weeks significantly decreased total cholesterol and increased HDL showing that mixture of these are more effective in reducing total lipids.

Mice exhibited decreased serum cholesterol, triglycerides and LDL level while increased HDL level when fed on ginger including diet (Hussein, 2012). Comparative results depicted that ginger has better although not significant preventive effect on systolic blood pressure and garlic has better preventive effect on lipid levels (Sanghal *et al.* 2012). Sterling and Eagling (2001) found that garlic has beneficial effects in reducing total plasma cholesterol and lowering blood pressure.

Garlic consists of many organosulpher compounds like allin, allicin, s-allyl cysteine and ajoene (Agarwal, 1996). Most of these compounds inhibit the cholesterol production from hepatocytes (Gebhardt and Beck, 1996). The above mentioned components inhibit the crucial enzyme (3-hydroxy-3-methyl-glutaryl-coenzyme A, HMG-coA, reductase) required in the cholesterol biosynthesis (Ferri *et al.,* 2003; Liu and Yeh, 2002). Garlic-derived compounds may also inhibit other enzymes in cholesterol biosynthesis pathway, like sterol 4-alpha-methyl oxidase (Liu and Yeh, 2002).

The results of this study were in favour of Bhandari *et al.* (2005) and Andallu *et al.* (2003). It has been suggested that ginger chemical components like (E)-8β, 17-epoxylabd-12-ene-15, 16 diol inhibits the biosynthesis of cholesterol (Tanabe *et al.,* 1993). Another mechanism of cholesterol reduction is that supplementation of ginger enhances the activity of LDH receptors which in turn increase the elimination of LDL from plasma thus causing reduction in plasma cholesterol level (Ness *et al.,*1996).

The results of present study were not in consensus with Sambaiah and Srinivasan (1991) who recorded no change in cholesterol content of rats fed ginger for 56 days. Carrijo *et al.* (2005) reported that garlic powered did not alter the serum levels of cholesterol and triacylglycerol's after feeding broiler chicks so, the results of this research are contradictory to present findings. Bordia *et al.* (1997) also found that 4g ginger powder for 3 months did not show any significant difference in lipid levels.

This difference in results may be due to different feeding periods and quantity of mixture fed to mice.

Conclusion

It is concluded that the consumption of garlic and ginger mixture created a beneficial hypoglycaemic and hyperlipidemic effects in diabetic mice.

Abbreviations

HDL: high density lipoproteins; LDL: low density lipoproteins; CHOD: cholesterol oxidase; PAP: phenol + aminophenazone

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Authors' contributions

NA carried out the design of study. SS participated in the experimental coordination including draft to manuscript. AM conducted and controlled the whole research and analyzed the biochemical features of the study. SK helped out in reviewing and arranging the article. MH designed the study, performed the statistical analysis and finalized the manuscript. All authors read and approved the final manuscript.

Conflict of Interest:

There is no conflict of interests regarding the publication of this article.

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Table 1. Effect of garlic and ginger mixture on blood glucose level of alloxan induced diabetic mice

Duration	**Groups**				
	G1 (Control)	**G2 (Diabetic)**	**G3 (g+G* /10g)**	**G4 (g+G /20g)**	**G5 (g+G /30g)**
Zero time	98.80±14.08^{b}	186.80±14.97^{a}	196.40±15.52^{a}	204.40±20.81^{a}	190.00±18.09^{a}
1st Week	95.00±10.81^{b}	204.00±14.38^{a}	188.40±14.64^{a}	186.00±13.91^{a}	174.00±13.54^{a}
2nd Week	98.00±8.60^{b}	193.00±12.41^{a}	178.00±10.26^{a}	180.40±11.18^{a}	168.00±8.74^{ab}
3rd Week	97.00±9.94^{c}	197.00±13.93^{a}	171.00±9.02^{ab}	169.00±11.64^{ab}	156.00±10.25^{b}
4th Week	102.00±11.02^{c}	202.10±16.77^{a}	161.00±11.55^{b}	156.00±10.48^{b}	143.00±10.48^{c}

Table 2. Effect of garlic and ginger mixture on lipid profile of alloxan induced diabetic mice

Parameters	Groups				
	G1 (Control)	**G2 (Diabetic)**	**G3 (g+G /10g)**	**G4 (g+G /20g)**	**G5 (g+G /30g)**
Cholesterol (mg/dl)	151.40±9.79^{b}	215.00±21.99^{a}	187.00±11.07$^{a b}$	176.60±11.07$^{a b}$	160.00±8.55^{b}
HDL (mg/dl)	47.80±6.79^{a}	21.80±3.64^{b}	29.60±5.64^{b}	37.20±5.78^{ab}	49.60±5.68^{a}
LDL (mg/dl)	119.40±27.28^{b}	198.80±14.41^{a}	184.80±9.61^{a}	173.00±13.69^{a}	160.80±11.26$^{a b}$
Triglycerides (mg/dl)	100.6±21.62^{b}	196.60±22.37^{a}	179.80±15.29^{a}	169.20±15.49^{a}	157.40±12.62^{a}

Means sharing different superscripts in rows differ significantly ($P<0.05$).

* g+G = garlic + Ginger mixture