Effect of Garlic (Allium sativum) Aqueous Extract on serum value of Glucose compared with Chromium Chloride in Male Rats

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Abstract: Garlic is probably one of the earliest known medicinal plants. Its bulbs (cloves) had been used as a cure-all in ancient Egypt and are mentioned in the Ebers Papyrus, one of the earliest treatises on medicinal plants. Chromium is a mineral which is required in trace amounts by the human body and is one of the most popular and widely used supplements. The main objective of following study was to evaluation of Garlic (Allium sativum) Aqueous Extract effects on serum value of Glucose compared with Chromium Chloride in Male Rats. In this experiment, 162 mature male rats (250 gm on the average) were acquired from Razi Serum – producing Institute of Karaj-Iran and transferred to keeping place. This design is performed as a factorial experiment 3*3 (3 level of GAE extract and 3 level of CrCl3 supplement) in the form of totally random design with 9 groups per 3 replications each containing 6 rat. At the end of fourth week, after 12 hours starvation, six rats per treatment were selected randomly from every treatment and their blood sampling was collected for biochemical traits, then serum concentrations of Glucose were determined. Results showed that garlic extract yields to decrease the Glucose. CrCl3 yields to increase in GLC at the dose of 8 mg/kg. Concomitant use of Garlic plus Supplementary CrCl3 show the decrease in blood GLC compared control group. It has been revealed that garlic and CrCl3 have the most important contents that combined use of these elements showed best effect than alone use. Also, garlic has antiglycemic effect and this is due to allicin content.

Key words: Garlic (Allium sativum), Aqueous Extract, Glucose, CrCl3, Rats.

INTRODUCTION

Garlic is known to possess a number of biologically active compounds having anticoagulant (Fukao et al., 2007), antioxidant (Banerjee et al., 2003; Lee et al., 2009), antihyperlipidemic (Gupta and Porter, 2001) and antihypertensive effects (Verma et al., 2008). Antidiabetic potential of garlic has been reported in many previous trials conducted in animal models (Marles and Farnsworth, 1995; Alarcon-Aguirala et al., 1998; Banerjee and Maulik, 2002; Anwar and Meki, 2003; Liu et al., 2005; Liu et al., 2006; Eidia et al., 2006; Jelodar et al., 2005; Lee et al., 2009). Garlic principle active agent appears to be allicin, a sulfur-containing compound that with its breakdown products give garlic its characteristic odour (Elkayam et al., 2003). Allicin is formed enzymatically from an odourless precursor, alliin, when garlic cloves are mechanically disrupted (Alpers, 2009). The probable mechanism underlying garlic hypoglycemic effects most likely is increased insulin secretion and sensitivity (Birdee and Yeh, 2010). Despite that the antidiabetic potential of garlic has been confirmed in animal studies, scientific evidence from human studies is lacking (Liu et al., 2007).

Most of the clinical studies have observed the effects of garlic on blood glucose in normal healthy individuals but not in diabetic patients, leaving no doubt that the role of garlic in the management of diabetic patients still needs to be confirmed. However, bearing in mind that garlic has been an essential part of our diet for centuries, it is taken for granted that garlic is safe in a wide range of doses. Few non-specific adverse effects were reported in clinical studies using garlic and its preparations and frequently consist of gastrointestinal discomfort and nausea. Allergic contact dermatitis has been reported in people with occupational exposure to garlic. There have also been infrequent reports of allergic conjunctivitis, rhinitis, bloating, headache, dizziness, profuse sweating and bronchospasm occurring in response to garlic inhalation or ingestion. Rarely ingestion of fresh garlic and garlic powder was reported to have synergistic effects with anticoagulants or platelet aggregation inhibitors increasing risk of haemorrhage (Banerjee and Maulik, 2002). Thus comprehensive clinical studies in diabetic patients are justified to confirm the efficacy and possible role of garlic in the management of diabetic patients (Thomson et al., 2007).

Chromium is a mineral which is required in trace amounts by the human body and is one of the most popular and widely used supplements. It is sold as a single-ingredient supplement as well as in combination.
formulas, particularly those marketed for weight loss and performance enhancement. The main objective of following study was to evaluation of Garlic (Allium sativum) Aqueous Extract effects on serum value of Glucose compared with Chromium Chloride in Male Rats.

**MATERIALS AND METHODS**

**Rats and Management:**

In this experiment, 162 mature male rats (250 gm on the average) were acquired from Razi Serum – producing Institute of Karaj-Iran and transferred to keeping place. This design is performed as a factorial experiment 3*3 (3 level of GAE extract and 3 level of CrCL3 supplement) in the form of totally random design with 9 groups per 3 replications each containing 6 rats. All of keeping cages were disinfected before performing the experiment. All of groups were kept in 12-hour light and 12-hour darkness conditions with 25-30. Temperature and free access to water and food in metal cages placed in animal husbandry of veterinary faculty of Islamic Azad University, Tabriz Branch.

**Preparation of garlic extract (GAE) and CrCL3 supplement:**

Fresh garlic aqueous was used in this experiment, and garlic aqueous extract was obtained through soxhlet apparatus in combination with deionized distilled water within 6 hours in two successive days with temperature of 30 (to prevent elements and materials of garlic aqueous from decomposition). Then, the extract was placed in incubator in order to be concentrated. Certain concentrations of garlic aqueous extract were dissolved in pure water and became reachable by rat on a daily basis. Crcl3 supplement was acquired (Merck-Germany) and after measuring certain rate by digital scale was given to rat on a daily basis. It should be mentioned that onion extract was give as gavage (gastro – oral) and Crcl3 compleiment was dissolved in water in certain amount and it was added to feed after steeping and powdering of pellets, then the feed was mixed, ground and dried, and obtained pellets was given to animal. Moreover, during the first week of experiment, all groups consumed basal diet in order to adapt with breeding environment conditions; then basal diet, basal diet + 60 mg/rat/day fresh GAE, basal diet + 120 mg/rat/day fresh GAE, basal diet + 4 mg/ kg diet Crcl3, basal diet + 8 mg/ kg diet Crcl3, basal diet + 60 mg/ rat/day fresh GAE+ 4 mg/ kg diet Crcl3, basal diet + 60 mg/ rat/day fresh GAE+ 8 mg/ kg diet Crcl3, basal diet + 120 mg/ rat/day fresh GAE+ 4 mg/ kg diet CrCl3 and 120 mg/ rat/day fresh GAE+ 8 mg/ kg diet CrCl3, respectively, were given to 1st group, 2nd group, 3rd group,4th group,5th group,6th group, 7th group, 8th group, and 9th group, within 4 weeks on a daily basis.

**Determination Of The Biochemical Traits:**

At the end of fourth week, after 12 hours starvation, six rats per treatment (2 rats per replicate or cage) were selected randomly from every treatment and their blood sampling was collected for biochemical traits, then serum concentration of Glucose, were determined.

**Statistical Analysis:**

Data were subjected to a one-way analysis of variance using the General Linear Models (GLM), and the statistical analysis system (SAS) User’s guide. The result of the Analysis of variance according to the model is,

\[
Y_{ijk} = \mu + \alpha_i + \beta_j + (\alpha \beta)_{ij} + e_{ijk}
\]

Where,
- \(Y_{ijk}\) = All dependent variable
- \(\mu\) = Overall mean
- \(\alpha_i\) = The fixed effect of GAE levels ( \(i = 1, 2, 3\) )
- \(\beta_j\) = The fixed effect of CrCL3 levels ( \(j = 1, 2, 3\) )
- \(e_{ijk}\) = The effect of experimental error

When significant difference among the means was found, means were separated using Duncan’s multiple range tests.

**Results:**

By comparison of the treatment groups (Table 1), observed that garlic extract yields to decrease the Glucose. CrCl3 yields to increase in GLC at the dose of 8 mg/kg.

Concomitant use of Garlic plus Supplementary CrCl3 show the decrease in blood GLC compared control group.
Table 1: Comparison of data obtained from analyzing of serum values of GLC.

<table>
<thead>
<tr>
<th>Garlic</th>
<th>GLC</th>
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<tbody>
<tr>
<td>0 mg /kg (control)</td>
<td>170.66</td>
</tr>
<tr>
<td>60 mg /kg</td>
<td>158.76</td>
</tr>
<tr>
<td>120 mg /kg</td>
<td>156.5</td>
</tr>
<tr>
<td>P</td>
<td>0.14</td>
</tr>
<tr>
<td>SEM</td>
<td>6.71</td>
</tr>
<tr>
<td>Supplementary CrCl₃</td>
<td></td>
</tr>
<tr>
<td>0 (control)</td>
<td>156.06</td>
</tr>
<tr>
<td>4 mg /kg</td>
<td>172.36</td>
</tr>
<tr>
<td>8 mg /kg</td>
<td>159.06</td>
</tr>
<tr>
<td>P</td>
<td>0.04</td>
</tr>
<tr>
<td>SEM</td>
<td>6.71</td>
</tr>
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</table>

Interaction

<table>
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<tr>
<th>Garlic</th>
<th>Supplementary CrCl₃</th>
<th>GLC</th>
</tr>
</thead>
<tbody>
<tr>
<td>0 mg /kg</td>
<td>0 (control)</td>
<td>175.17</td>
</tr>
<tr>
<td>4 mg /kg</td>
<td>168.00</td>
<td></td>
</tr>
<tr>
<td>8 mg /kg</td>
<td>163.25</td>
<td></td>
</tr>
<tr>
<td>60 mg /kg</td>
<td>0 (control)</td>
<td>155.25</td>
</tr>
<tr>
<td>4 mg /kg</td>
<td>178.00</td>
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<td>8 mg /kg</td>
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<td>P</td>
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</tr>
<tr>
<td>SEM</td>
<td>8.33</td>
<td></td>
</tr>
</tbody>
</table>

Discussion and Conclusion:

Many of the trials evaluating the hypoglycemic effects of garlic were done in animal models. Garlic was reported to be effective in reducing blood glucose in streptozocin-induced as well as alloxan induced diabetic rats and mice (Mathew and Augusti, 1973; Swanston-Flatt et al., 1990; Kasuga et al., 1999; Ohaeri, 2001). It was also reported that ingestion of garlic juice and aqueous garlic extracts resulted in better utilization of glucose in glucose tolerance test performed in diabetic animal models (Jain and Vyas, 1973; Jain and Vyas, 1975; Jalal et al., 2007). Two of the previous studies reported that allicin, a sulfur containing amino acid in garlic has a potential to reduce diabetic condition in rat almost to the same extent as did glibenclamide and insulin (Sheela and Augusti, 1992; Sheela et al., 1995). Although many of the previous trials in animal models showed significant effects of garlic on glycemic control, hypoglycemic effect of garlic in human is not well studied. All human studies (Kiesewetter et al., 1991; Jain et al., 1993; Ali and Thomson, 1995; Bordia et al., 1998; Zhang et al., 2001) apart from two (Afkhami-Ardekani et al., 2006; Sobenin et al., 2008), has showed the effect of garlic on blood glucose level in normal healthy individuals but not in diabetic patients. Hassan et al., 2009 worked on In vivo evidence of hepato- and reno-protective effect of garlic oil against sodium nitrite-induced oxidative stress and showed that NaNO2 treatment for a period of three months induced a significant increase in serum levels of glucose, aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), bilirubin, urea and creatinine as well as hepatic AST and ALT. Our data indicate that garlic is a phytoantioxidant with powerful chemopreventive properties against chemically-induced oxidative stress.

In one study by Pei et al., 2006 it has been revealed that chromium group demonstrated a lower FPG and fasting insulin (-38.1 +/- 9.2 vs 63 +/- 8.5 mg/dL and -1.7 +/- 0.2 vs 1.9 +/- 0.3 microU/mL, respectively; P < .05), especially in male patients (-41 +/- 9.2 vs 85 +/- 11.7 mg/dL and -2.7 +/- 0.2 vs 3.1 +/- 0.3 microU/mL, respectively; P < .01), at the end of the study. Lower glycosylated hemoglobin was observed in chromium-treated male patients (-1.1 +/- 0.5 vs 0.7 +/- 0.2; P < .05). However, there were no significant changes in other metabolic parameters (lipid profiles including total cholesterol, triglyceride, low-density lipoprotein cholesterol, and high-density lipoprotein cholesterol), except improvement of insulin resistance (homeostasis model assessment for insulin resistance and insulin sensitivity index from frequently sampled intravenous glucose tolerance test) observed in male patients (-2.1 +/- 1.1 vs -0.41 +/- 1.12 and 0.18 +/- 0.11 vs -0.15 +/- 0.2, respectively; P < .05). There were no adverse events in both groups, except for mild complaints in the chromium group on constipation (5%) and flatulence (5%). Intake of milk powder containing 400 microg/d of chromium for 16 weeks in subjects with type 2 diabetes mellitus resulted in lowering of FPG, fasting insulin, and improvement of metabolic control in male patients.

Jelodar et al., 2005 showed that the results of them study indicate that only garlic was able to reduce blood glucose significantly compared with the control group (P<0.05). In the control positive group all the mentioned morphometric factors were significantly changed in comparison with the control negative (normal health) group, but the same did not show significant change between treated and untreated diabetics.
In one other study by Shinde Urmila et al., 2004 demonstrated that treatment with chromium chloride significantly improved the impaired glucose tolerance and insulin sensitivity of both STZ diabetic and nSTZ diabetic rats without any change in basal or glucose stimulated insulin response indicating insulin-sensitizing action of chromium. CC treatment also significantly improved deranged lipid metabolism. CC per se did not produce any effect in vitro, however, significantly increased insulin stimulated glucose uptake in C2C12 myoblasts and differentiation of 3T3-L1 preadipocytes into mature adipocytes supporting the in vivo insulin-sensitizing action of chromium. This study shows that CC exhibited significant anti-diabetic potential in chemically-induced diabetes in rats, the mechanism of which appears to be potentiation of insulin actions at the target tissues leading to improved peripheral insulin sensitivity. By comparison of above mentioned literatures it has been revealed that garlic and CrCl₃ have the most important contents that combined use of these elements showed best effect than alone use. Also, garlic has antiglycemic effect and this is due to allicin content.

REFERENCES


