Hypoglycemic Effect of *Lycopersicon esculentum* (Tomato) on Alloxan-Induced Diabetic Rats

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INTRODUCTION

Diabetes mellitus has been a key degenerative disease affecting the world’s population. *Lycopersicon esculentum* (Tomato), a fruit consumed by many and known to have certain phytochemicals was used to determine its hypoglycemic effect on alloxan induced diabetic rats. The tomato was dried, pulverized and dissolved in distilled water and administered orally to albino rats in various concentrations according to their body weight. 30 albino rats were divided into 6 groups of 5 rats each. Groups I and II served as normal and diabetic control respectively, while groups III to VI were induced with diabetes and treated with different concentrations of tomato. After 14 days of treatment with various concentrations of tomato, there was a marked decrease in blood sugar levels at all the study concentrations. The result of the lipid profile a significant increase (p<0.05) in total cholesterol (150.67±7.02 mg/dL), triglyceride (159.33±5.03 mg/dL), LDL-Cholesterol (77.53±1.83 mg/dL) and a decrease in HCL-Cholesterol (51.67±1.00 mg/dL) levels in untreated diabetic rats when compared to the normal control. Upon treatment with 200 mg/kg of tomato, there was a significant decrease (p< 0.05) in the levels of Triglyceride, total cholesterol and LDL-cholesterol and an increase in the HDL-cholesterol. These results suggest that tomato may have the ability to reduce blood sugar level and the risk of cardiovascular disease.
Regardless of the commercially available antidiabetic drugs, despite the presence of known antidiabetic drugs in the markets, diabetes and the related complication continue to be of enormous concern medically. Management of diabetes devoid of side effect is still a problem to the medical community. Therefore, it is very important to investigate plants with potent antidiabetic effect for solution.

MATERIALS AND METHODS

Reagents

The analytical grade reagents obtained from Randox Laboratories Ltd., Antrim, United Kingdom were used for the analysis. The reagents include Glucose, Total protein, total cholesterol and Triglycerides kits. Other reagents used are Choloform (BDH), Alloxan monohydrate (sigma) and methylated spirit (BDH).

Experimental animals

Forty-five (45) albino rats of both sexes were purchased from University of Jos, Animal house. The animals were brought in two weeks prior to commencing the studies so as to get use to their new environment. They were fed with Grower marsh and water in cages at the animal house of the Department of Biological Sciences, Gombe State University.

Preparation of the tomatoes powder from tomatoes fruit

Ripe *Lycopersicon esculentum* (tomato) Fruit (1500g) were procured from a nearby market in Gombe, Gombe, Nigeria and identified at the botany unit of Gombe State University. The powder was prepared from cleaned tomato, after been sliced and identified at the botany unit of Gombe state University. The powdered tomatoes were then dissolved in distilled water and administered orally as 50,100,150 and 200 mg/kg body weight per day for 14 days.

Experimental design

In this experiment, a total of 30 rats (25 diabetic and 5 normal rats) weighing between 110 to 160g were used. The rats were split into six (6) groups (5 diabetic and 1 normal rats) of 5 rats each. Group I are normal rats, Group II are diabetic rats (served as diabetic control), Groups III, IV, V and VI are diabetic rats given 50, 100, 150 and 200 mg/kg body of the extract orally for 14 days.

Induction of diabetes

Diabetes was induced by a single injection of newly made alloxan monohydrate (200 mg alloxan/ kg b.wt) dissolved in distilled water and administered intraperitoneally (IP) to rats that were denied food for at least 16 h. Blood glucose levels were taken 72 h after alloxan administration using glucometer. According to [11], rats with values of Fasting blood glucose more than 200mg/dL are to be used for the study.

Sacrificing procedure and blood sample collection

Blood samples were collected on the 14th day of the onset of diabetes. After fourteen days of treatment, the rats were not given food for 16 h and made unconscious with 10% choloform before they were sacrificed by surgical dislocation of neck and letting free flow of blood into clean containers. The blood samples were left to clot and serum separated was used to ascertain some biochemical indices.

Glucose estimation

Fasting blood sugar was estimated at 1st, 7th and the 14th days, the animals were not fed for 16 h before collecting their blood samples. Glucose oxidase method was employed to ascertain the level of glucose after fasting [12]. The test, standard and blank test tubes were labeled, and the appropriate amount of the reagents and samples were poured into the test tubes at room temperature. The content of the tubes was shaken and kept at 20-25 °C for 10 min. Absorbance of sample and standard were measured within 60 min against reagent blank at a wavelength of 510 nm in a spectrophotometer. Glucose concentration was calculated using the expression below

\[
\text{Glucose concentration} = \frac{A_{\text{sample}}}{A_{\text{standard}}} \times \text{Standard concentration}
\]

Serum lipid profile estimation

Triglycerides

The triglyceride levels were assayed using the method described by [13]. Briefly, Reagents and samples kept at room temperature were poured into test tubes labeled test, standard and blank, 10µL of serum and standard were added to the right test tubes and then 1000 µL of the reagent was poured to all test tubes. The content of the tubes was left at room temperature for 20min. After 60min, the spectrophotometer reading was obtained at 500nm for the test and standard. Triglyceride level was calculated using the expression below

\[
\text{Triglyceride concentration} = \frac{A_{\text{sample}}}{A_{\text{standard}}} \times 533\text{mg/dL}
\]

Total cholesterol

This was estimated by the enzymatic endpoint manual method of [12] using the RANDOX(R) kit for cholesterol estimation. 10µL of test and standard were poured into an already labeled test tubes followed by 1000 µL of the reagent. After mixing for 10min at room temperature, the absorbance at 500 nm was taken within 60min. Total cholesterol was calculated using the expression below

\[
\text{Concentration of cholesterol in sample} = \frac{A_{\text{sample}}}{A_{\text{standard}}} \times 200\text{mg/dL}
\]

Determination of HDL-cholesterol

After precipitation of other fractions with magnesium chloride, the HDL- cholesterol content to the supernatant fluid is estimated as explained for total cholesterol using method of [12].

LDL- cholesterol estimation

This is estimated using the equation [13].

\[
C_{\text{LDL}} = C_{\text{TC}} - (C_{\text{HDLC}} + \frac{TG}{5})
\]

\[
C = \text{Concentration}
\]

\[
C_{\text{TC}} = \text{Total cholesterol}
\]

\[
TG = \text{Triglyceride}
\]

\[
C_{\text{HDLC}} = \text{High density lipoprotein cholesterol}
\]

Determination of total proteins

Total protein concentration was determined using Biuret methods [14]. Into test tubes arranged and labeled as test, standard and blank, 0.02ml of test and standard were poured into the right tube; then 1ml of reagent was added to all test tubes. The content of the tubes was mixed and kept for half an hour at room temperature. The absorbance of the sample was measured at a wavelength of 546nm against blank within 60 min in a spectrophotometer. Total protein concentration was calculated using the expression below

\[
\text{Calculation: Total Protein Concentration} = \frac{A_{\text{sample}}}{A_{\text{standard}}} \times 190\text{g/l}
\]
RESULTS AND DISCUSSION

The results of fasting blood sugar for the 1st, 7th, and 14th days are shown in Table 1. This result shows that there was no substantial difference (p > 0.05) in fasting blood glucose levels in the test groups in comparison with the diabetic control group (202.33 ± 7.07 mg/dL) on day one of the experiment. On the 7th day, there was a marked difference (p < 0.05) in fasting blood glucose level in the treatment groups compared to the values in normal (68.67 ± 3.80 mg/dL) and diabetic (200.53 ± 5.50 mg/dL) control groups. On the last day of the second week, as seen in Table 1, there was significant decrease (p < 0.05) in fasting blood glucose levels in treatment groups III, IV, V and VI compared to the diabetic control group (198.21 ± 6.10 mg/dL). Again there was significant decline (p < 0.05) in fasting blood glucose level in treatment groups in comparison to the normal control. As in Table 1, there was significant decrease (p < 0.05) in fasting blood glucose level in treatment groups III, IV, V and VI compared to the diabetic control group (198.21 ± 6.10 mg/dL). Again there was significant difference (p < 0.05) compared with the normal control. As presented in Table 1, there was significant decrease (p < 0.05) in triglyceride in the treatment groups compared with the diabetic control, and all treatment groups displayed a reasonable difference (p < 0.05) compared with the normal control.

Table 1. Fasting blood sugar on the 1st, 7th, and 14th days.

<table>
<thead>
<tr>
<th>Group (mg/kg)</th>
<th>Dosage (mg/kg)</th>
<th>1st day</th>
<th>7th day</th>
<th>14th day</th>
</tr>
</thead>
<tbody>
<tr>
<td>I Normal control</td>
<td>.....</td>
<td>67.5±6.03</td>
<td>68.67±3.80</td>
<td>68.33±3.51</td>
</tr>
<tr>
<td>II Diabetic control</td>
<td>.....</td>
<td>202.33±7.07*</td>
<td>200.53±5.5*</td>
<td>198.21±6.10*</td>
</tr>
<tr>
<td>III</td>
<td>50</td>
<td>198.00±7.64*</td>
<td>148.67±3.21**</td>
<td>135.33±4.30**</td>
</tr>
<tr>
<td>IV</td>
<td>100</td>
<td>199.76±1.52*</td>
<td>141.67±3.72**</td>
<td>125.30±5.03**</td>
</tr>
<tr>
<td>V</td>
<td>150</td>
<td>200.33±2.00**</td>
<td>141.67±1.52**</td>
<td>118.00±8.06**</td>
</tr>
<tr>
<td>VI</td>
<td>200</td>
<td>200.33±2.51*</td>
<td>136.33±4.16**</td>
<td>108.01±5.5**</td>
</tr>
</tbody>
</table>

Note: The values in the Table are mean ± standard deviation of fasting blood sugar determination n=5. A single asterisk shows a significant difference from the normal control (p < 0.05) while double asterisks shows a significant difference from the diabetic control (p < 0.05).

Table 2. Effect of Lycopersicon esculentum (tomato) administration on Serum lipid profile and total protein in diabetic rats.

<table>
<thead>
<tr>
<th>Grp. Dosage</th>
<th>TG (mg/dL)</th>
<th>TC (mg/dL)</th>
<th>HDL-CH (mg/dL)</th>
<th>LDL-CH (mg/dL)</th>
<th>TP (g/dL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>I Normal control</td>
<td>106.33±4.50</td>
<td>61.23±3.10</td>
<td>63.00±5.19</td>
<td>30.88±2.50</td>
<td>6.57±0.74</td>
</tr>
<tr>
<td>II Diabetic control</td>
<td>159.33±5.03*</td>
<td>150.67±7.02*</td>
<td>51.67±1.00**</td>
<td>77.53±1.83**</td>
<td>9.00±0.50*</td>
</tr>
<tr>
<td>III</td>
<td>50</td>
<td>131.33±3.33**</td>
<td>90.00±2.17**</td>
<td>63.00±1.00**</td>
<td>50.33±5.30**</td>
</tr>
<tr>
<td>IV</td>
<td>100</td>
<td>122.00±3.60**</td>
<td>86.67±7.64**</td>
<td>59.67±1.53**</td>
<td>43.13±4.00**</td>
</tr>
<tr>
<td>V</td>
<td>150</td>
<td>115.33±5.50*</td>
<td>70.53±9.97**</td>
<td>58.67±1.33*</td>
<td>42.56±9.77**</td>
</tr>
<tr>
<td>VI</td>
<td>200</td>
<td>107.33±2.51*</td>
<td>67.40±2.35**</td>
<td>58.00±5.19**</td>
<td>35.85±3.33**</td>
</tr>
</tbody>
</table>

Note: The values in the Table are mean ± standard deviation of serum lipid profile and total protein determination n=5. A single asterisk shows a significant difference from the normal control (p < 0.05) while double asterisks shows a significant difference from the diabetic control (p < 0.05). TG= Triglyceride; TC= Total cholesterol; HDL-CH= High density lipoprotein cholesterol; LDL-CH= Low density lipoprotein cholesterol; TP= Total protein.

DISCUSSION

This study checked the action of tomato in reducing the level of high blood sugar caused by alloxan in albino rats. The normal control group had normal fasting blood glucose level while the diabetic control group had constant hyperglycaemia all through the experiment. In other words, it has been shown in this study that the albino rats that were confirmed diabetic and not treated with tomato had hyperglycaemia that lasted throughout the experiment while on the other hand, diabetic animals treated with tomato showed reduced fasting blood glucose levels on the 7th and 14th day (the last day of the experimental duration) as shown in Table 1. Although there was significant difference (p < 0.05) between the normal control and treatment groups, oral administration of tomatoes extract highly reduced the fasting blood glucose level in diabetic rats at 7th and 14th days compared to diabetic control group (Group II). This revelation maybe as a result of the phytochemical constituents as well as caroteneoids and chromium. Carotenoids and chromium have been shown to have affect glucose metabolism and insulin secretion [15-18]. Similarly, it has been reported that intake of carotenoid rich fruit protects against hyperglycemic induced damage in diabetes mellitus [17,19,20].

This study, oral administration of tomato even at 50mg/kg can potentiate effect of lowering fasting blood sugar though this study could not elucidate the mechanism by which tomatoes lowers blood glucose levels, it may be probably due to their phytochemical constituents, especially carotenoids and chromium [15]. It was also reported that tomatoes are rich in fibre [21]. Fibre in tomato lowers gastric emptying and therefore absorption of sugar into the bloodstream, positively impacting diabetes.

In this study, oral administration of tomato in untreated diabetic rats, this proved the results of other investigator that said untreated diabetes is associated with impairment in lipid metabolism [22]. It was also reported that hypercholesterolemia in induced-diabetic rats results from increased intestinal absorption and synthesis of cholesterol [23] has been demonstrated in this study where the diabetic untreated rats had high serum cholesterol.

However, animals receiving oral administration of tomatoes have significantly lower level of serum cholesterol. Tomatoes are rich in fibre and fibre helps in transforming the nature of what is contained in the gastrointestinal tract and this affect how other nutrients and chemicals are absorbed [24]. Soluble fibre binds to bile acids in the small intestine and lowers its absorption This in turns lowers cholesterol levels in the blood [25]. However, the fall in serum total cholesterol was less than that of the normal control group and the fall was proportionate to the increase in the dose of tomatoes extract, with the highest reduction observed in the highest dose (200 mg/kg).

The diabetic group has the lowest HDL-cholesterol levels compared with all the treatment groups. Some constituents of tomatoes like lycopene and vitamin E are powerful antioxidant. The constituent’s vitamin E and lycopene in tomato are known to prevent LDL oxidation effectively [26]. There was also significant increase in serum triglyceride levels in the diabetic control compared with the treatment groups, this is in accordance with fact that untreated diabetes is associated with increased in serum triglyceride levels [27]. However, in the treatment groups
there was significant decrease in the serum levels of triglycerides, although levels are higher than that of the normal control group.

The diabetic control had the highest level of LDL-Cholesterol, while the other treated groups and the normal control groups showed a decreased levels of serum LDL-cholesterol, but the levels in all the treatment groups were significantly (p<0.05) different from the normal control. Research shows that lycopene lowers the oxidation of LDL-cholesterol [28]. This study shows that in the diabetic control there was a significant increase in the serum total protein compared with both the normal control and the treatment groups. A previous study has shown that one of the complications of untreated diabetes is the increase in total serum protein [29].

CONCLUSION
Tomatoes reduced fasting blood sugar, LDL-cholesterol, total cholesterol and triglycerides levels in alloxan induced diabetic rats. It also increased the levels of HDL-cholesterol. Tomatoes may have hypoglycaemic effects and may also reduce the risk of cardiovascular diseases by decreasing the levels of serum triglyceride, total cholesterol and LDL cholesterol. Further studies should be carried out on its toxicity and the actual component of tomato that have the hypoglycaemic property.

REFERENCE