Original Research Article

Effects of Ethanolic Leaf Extract of Gongronema Latifolium on Liver Enzymes and Lipid Profile in Albino Wistar Rats

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ABSTRACT

The effects of ethanolic leaf extract of Gongronema latifolium (GL) on liver enzymes and lipid profile was investigated in normal wistar rats. A total of thirty (30) wistar rats were divided into 6 groups of 5 rats each. Groups I and II served as control and received water for 14 and 28 days respectively. Groups III and IV were administered with GL (300mg/kg) orally for 14 and 28 days respectively. Groups V and VI were administered with GL (600mg/kg) orally for 14 and 28 days respectively. Blood samples were collected through cardiac puncture and analyzed for liver enzymes and lipid parameters. The liver were also harvested for histological analysis. The results showed significant (P<0.05) increase in the level of alanine transaminase (ALT) for 300mg/kg when compared to the control on day 14. However, there was no significant increase in the levels of aspartate transaminase (AST), alkaline phosphatase (ALP), total protein and albumin for the test groups when compared to the control. The serum levels of total cholesterol, triglyceride, low density lipoprotein - cholesterol (LDL-C) and the atherogenic index decreased significantly (P<0.05) when compared to the control. High density lipoprotein – cholesterol (HDL-C) level significantly increased in a dose dependent manner. There was a significant (P<0.05) reduction observed in body weight of treated rats. Results of this study suggest that ethanolic leaf extracts of G. latifolium possess lipid lowering (hypolipidaemic), and a possible hepatotoxic potential, particularly at higher concentration. Thus, consumption of G. latifolium should be done with caution especially in conditions of hepatic diseases.

Keywords: Gongronema latifolium, ethanolic, liver function, lipid profile, hypolipidaemic and hepatotoxic.

INTRODUCTION

The liver is one of the most important organs in the body; playing an essential role in various physiological and biochemical processes through its numerous enzymatic cascades. Liver enzymes catalyze and regulate most biochemical reactions in the human body; from replication of DNA by DNA polymerases to metabolism of xenobiotics. [1]

The functional integrity of the liver can be assessed by measuring the serum levels of some biochemical markers:
ALT (alanine aminotransferase), ALP (alkaline phosphatase) and AST (aspartate aminotransferase). The liver also functions in the synthesis of lipids; [2] lipids play a critical role in almost all aspects of biological life. Lipids are structural components of cells and are involved in several metabolic and hormonal pathways. [3] Lipid and lipoprotein abnormalities are well known risk factors for heart disease; elevated levels of triacylglycerols (TG), cholesterol, and low density lipoprotein-cholesterol (LDL-C) are documented risk factors for atherogenesis. [4] The blood level of high density lipoprotein-cholesterol (HDL-C) in contrast bears an inverse relationship with the risk of atherosclerosis and coronary heart disease: the higher the level, the lesser the risk. [5] Diet and genetic factors both play a major role in regulating cholesterol and triglycerides levels in the plasma. [5,6] High levels of cholesterol, particularly LDL-cholesterol, are mainly responsible for hypercholesterolemia. [8] Hypercholesterolemia is known to be associated with oxidative stress related to increased lipid peroxidation. [7] Increased generation of oxidized LDL-C by the liver is a major factor in the vascular damage associated with high cholesterol levels. [8]

In view of the importance of the liver to the body and the adverse effects of synthetic lipid-lowering drugs, [4] the quest for natural products with lipid-lowering potential and minimal toxicity profile continues to be importance. Plants have been the companions of man since time immemorial and form the basis of useful drugs with toxicity screening of medicinal plants, therefore presents avenue for the discovery of new drugs.

Gongronema latifolium is a tropical rainforest plant belonging to the Asclepiadaceae family. [9] G. latifolium is a perennial edible plant with soft and pliable stem. It is widely used in the West African sub region for a number of medicinal and nutritional purposes. It is a shrub, with milky or less often a clear latex.

It is commonly used as a spice and is known by various local names across southern, Nigeria. [10,11] G. latifolium has a bitter taste.

The phytochemical constituents of Gongronema latifolium leaf were investigated. [12] The result of the study reported the presence of alkaloid, saponin, inulin and the absence of tannin. [13] Also reported the presence of glycosides in G. latifolium leaf extract.

The aim of this study, therefore, is to determine the effect of the ethanolic leaf extract of Gongronema latifolium on liver enzymes and lipid profile in male albino wistar rats.

MATERIALS AND METHODS
Collection and identification of plant materials
Fresh leaf samples of Gongronema latifolium obtained from a local Market in Port Harcourt, Nigeria, were identified and authenticated by Dr. N. L. Edwin–Wosu, of the Department of Plant Science and Biotechnology, University of Port Harcourt, Nigeria.
Preparation of ethanolic leaf extracts of Gongronema latifolium
The leaves were washed in tap water to remove dirt, and dried at room temperature (26°C) over a period of 3 weeks. The dried leaves were milled to fine powder using a manual engine grinder and 500g of the leaves obtained. This quantity was soaked in 400ml of ethanol (80% v/v) for 48 hours. The solution obtained was then filtered with Whatman No. 1 filter paper to separate the filtrate from the residue. The extract was concentrated under reduced pressure in a vacuum at 45°C using a rotary evaporator (Searl Instruments Ltd., England). The yield
of the crude ethanolic extract of *G. latifolium* leaves obtained weighed 63.1g. The extract was stored in a refrigerator at 4°C before use for the study.

**Experimental animals**

Thirty (30) male albino wistar rats weighing between (160 and 270g) were used for this study. The animals were kept in the animal house, Department of Human Physiology, University of Port Harcourt, Nigeria, in spacious and well ventilated cages at room temperature of 28 ± 1°C and under natural day / light cycles. They were allowed to acclimatize for 14 days and had free access to feed and water *ad libitum*.

**Experimental design**

The rats were randomly distributed into six (6) groups (I - VI) of 5 each. Groups I and II served as the Control and received tap water for 14 and 28 days respectively. Groups III and IV received 300mg/kg body weight of ethanolic leaf extract of *G. latifolium* for 14 and 28 days respectively. Groups V and VI received 600mg/kg body weight of ethanolic leaf extract of *G. latifolium* for 14 and 28 days respectively. The extracts were administered to the animals orally once daily between (9 and 10am).

The doses of 300mg/kg and 600 mg/kg for *Gongronema latifolium* were used based on previous data of LD₅₀ value of *G. latifolium* leaves of 1050mg/kg. [14]

The animals were sacrificed at the end of the administration. Blood was collected by cardiac puncture into lithium heparin bottles, for liver function and lipid profile evaluation. The following parameters were determined: total protein, albumin, alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), total cholesterol, high density lipoprotein cholesterol (HDL-C) and triglycerides (TG) using commercial kits obtained from Randox Laboratories, UK in a Mindray auto–analyzer (Model: BS 800M).

LDL- cholesterol (LDL-C) was calculated using the formula of. [15]

LDL-C (mg/dl) = (Total cholesterol – HDL) – (Triglyceride ÷ 5). [16]

While Atherogenic index (AI) = (log (TG ÷HDL-C)) was determined as described by. [17]

**Statistical analysis**

Data were expressed as Mean ± SEM and data were analyzed using one way analysis of variance (ANOVA) difference was considered significant at p value less than 0.05.

**RESULTS**

Tables 1 and 2 shows values of liver enzymes, total protein and albumin following 14 and 28 days of administration of ethanolic leaf extract of *G. latifolium* respectively.

Following 14 days of administration, there was a significant (p<0.05) increase in values of ALT at low dose (Grp III). A dose dependent non – significant reduction was observed in values of ALP. The values of AST increased non- significantly in a dose dependent manner. There were no effect on the serum levels of total protein and albumin.

However, following 28 days, there was a non–significant increase in the values of ALT and AST in a dose dependent manner.

Non – significant reduction in values of ALP was also observed. Similarly, there were no effects on the serum levels of total protein and albumin.

Effect of ethanolic leaf extracts of *G. latifolium* on the lipid profile following 14 and 28 days of administration are as shown in Tables 3 and 4 respectively.
Table 1: Values of liver enzymes, total protein and albumin following 14 days of the administration of ethanolic leaf extracts of *Gongronema latifolium* (GL) in albino wistar rats.

<table>
<thead>
<tr>
<th>PARAMETERS</th>
<th>GROUP I (CONTROL)</th>
<th>GROUP III (300mg/kg)</th>
<th>GROUP V (600mg/kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>ALT (U/L)</td>
<td>60.63 ± 5.27</td>
<td>131.28 ± 31.35*</td>
<td>104.65 ± 12.66</td>
</tr>
<tr>
<td>ALP (U/L)</td>
<td>295.97 ± 18.58</td>
<td>278.80 ± 44.62</td>
<td>190.25 ± 33.06</td>
</tr>
<tr>
<td>AST (U/L)</td>
<td>314.53 ± 28.17</td>
<td>339.20 ± 89.22</td>
<td>379.88 ± 75.11</td>
</tr>
<tr>
<td>TOTAL PROTEIN (g/L)</td>
<td>6.20 ± 1.33</td>
<td>5.93 ± 2.99</td>
<td>6.83 ± 1.73</td>
</tr>
<tr>
<td>ALBUMIN (g/L)</td>
<td>28.67 ± 0.57</td>
<td>27.90 ± 1.90</td>
<td>30.95 ± 1.25</td>
</tr>
</tbody>
</table>

Values are expressed as mean ± SEM; * Significant at P<0.05

Following 14 days of administration, there was no effect on the serum levels of total cholesterol. A significant (p<0.05) reduction in values of triglyceride was observed at 300mg/kg. Non - significant increase in HDL - C was observed. A dose dependent non - significant reduction in values of LDL - C was observed. There was a significant (p<0.05) reduction in the atherogenic index at dose of 300mg/kg b. w.

Following 28 days of administration, a significant (P<0.05) reduction was observed in the values of total cholesterol. There was a significant (p<0.05) reduction in serum level of triglyceride at low dose. A dose dependent significant increase in values of HDL - C significant (p<0.05) at high dose was observed. There was a significant (p<0.05) reduction in values of LDL - C. There was significant (p<0.05) reduction in atherogenic index in a dose dependent manner.

Table 2: Values of liver enzymes, total protein and albumin levels following 28 day of administration of the ethanolic leaf extracts of *G. latifolium* (GL) in albino wistar rats.

<table>
<thead>
<tr>
<th>PARAMETERS</th>
<th>GROUP II (CONTROL)</th>
<th>GROUP IV (300mg/kg)</th>
<th>GROUP VI (600mg/kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>ALT (U/L)</td>
<td>64.33 ± 3.28</td>
<td>75.78 ± 9.61</td>
<td>110.10 ± 9.09</td>
</tr>
<tr>
<td>ALP (U/L)</td>
<td>292.30 ± 19.76</td>
<td>203.48 ± 20.03</td>
<td>265.43 ± 45.01</td>
</tr>
<tr>
<td>AST (U/L)</td>
<td>318.87 ± 25.33</td>
<td>410.68 ± 59.78</td>
<td>509.43 ± 62.34</td>
</tr>
<tr>
<td>TOTAL PROTEIN (g/L)</td>
<td>6.04 ± 0.68</td>
<td>5.84 ± 3.04</td>
<td>6.27 ± 8.19</td>
</tr>
<tr>
<td>ALBUMIN (g/L)</td>
<td>29.13 ± 0.32</td>
<td>31.73 ± 2.15</td>
<td>31.53 ± 4.66</td>
</tr>
</tbody>
</table>

Values are expressed as MEAN ± SEM; n=5.

Table 3: Effects of ethanolic leaf extracts of *G. latifolium* (GL) on lipid profile following 14 days of administration in albino wistar rats.

<table>
<thead>
<tr>
<th>PARAMETERS</th>
<th>GROUP I (CONTROL)</th>
<th>GROUP III (300mg/kg)</th>
<th>GROUP V (600mg/kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total Cholesterol (mg/dl)</td>
<td>32.55 ± 2.02</td>
<td>31.05 ± 1.69</td>
<td>31.68 ± 2.13</td>
</tr>
<tr>
<td>Triglyceride (mg/dl)</td>
<td>27.27 ± 0.65</td>
<td>17.44 ± 2.51*</td>
<td>27.81 ± 2.06</td>
</tr>
<tr>
<td>HDL - C (mg/dl)</td>
<td>14.67 ± 2.34</td>
<td>17.98 ± 1.21</td>
<td>17.55 ± 1.51</td>
</tr>
<tr>
<td>LDL - C (mg/dl)</td>
<td>12.43 ± 3.56</td>
<td>9.95 ± 0.68</td>
<td>8.57 ± 1.57</td>
</tr>
<tr>
<td>Atherogenic Index = log (TG/HDL-C)</td>
<td>0.28 ± 0.78</td>
<td>-0.03 ± 0.07*</td>
<td>0.20 ± 0.06</td>
</tr>
</tbody>
</table>

Values are expressed as MEAN ± SEM; n=5; * Significant at P<0.05

Table 4: Effects of ethanolic leaf extracts of *G. latifolium* (GL) on lipid profile following 84 days of administration in albino wistar rats.

<table>
<thead>
<tr>
<th>PARAMETERS</th>
<th>GROUP II (CONTROL)</th>
<th>GROUP IV (300mg/kg)</th>
<th>GROUP VI (600mg/kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total Cholesterol (mg/dl)</td>
<td>34.53 ± 2.66</td>
<td>22.32 ± 0.89*</td>
<td>25.29 ± 2.18*</td>
</tr>
<tr>
<td>Triglyceride (mg/dl)</td>
<td>25.99 ± 3.42</td>
<td>19.36 ± 1.69*</td>
<td>20.20 ± 1.06</td>
</tr>
<tr>
<td>HDL - C (mg/dl)</td>
<td>11.60 ± 0.42</td>
<td>15.37 ± 0.97</td>
<td>17.10 ± 1.77*</td>
</tr>
<tr>
<td>LDL - C (mg/dl)</td>
<td>17.73 ± 3.33</td>
<td>3.08 ± 1.11*</td>
<td>4.15 ± 0.91*</td>
</tr>
<tr>
<td>Atherogenic Index = log (TG/HDL-C)</td>
<td>0.34 ± 0.04</td>
<td>0.10 ± 0.03*</td>
<td>0.08 ± 0.05*</td>
</tr>
</tbody>
</table>

Values are expressed as MEAN ± SEM; n=5; * Significant at P<0.05
**DISCUSSION**

Effects of ethanolic leaf extract of *Gongronema latifolium* on liver enzymes and lipid profile in albino wistar rats were investigated. This study revealed a significant (P<0.05) increase in ALT values at low dose. This finding is consistent with the report of. The study also shows non-significant changes in ALP and AST values following 14 days of administration and non-significant changes in ALP, ALT and AST following 28 days of administration.

Assay for liver enzymes: ALT, AST and ALP are important in assessing optimal liver function. Increase in the level of liver enzymes in the plasma is an indication of liver dysfunction. The increase in ALT observed from this study may be a clear demonstration of cellular leakage and loss of functionality of membrane integrity of hepatocytes. ALT is a good indicator of liver dysfunction and this further substantiates the possible hepatoprotective effects of administration of the plant extract especially at low dose. The effect of the plant at low dose following 14 days of administration may be attributed to the action of glycosides and phytosteroids (constituent of leaves extracts of *G. latifolium*), which has been shown to inhibit antioxidant activities in hepatocytes.

The result revealed non – significant changes in values of total protein and albumin following 14 and 28 days of administration. This could be attributed to low protein content of *G. latifolium*. Lipid profile (which involves levels of total cholesterol, HDL, LDL and triglycerol) which serves as diagnostic indices in conditions such as coronary heart disease, atherosclerosis, chronic obstructive jaundice and hepatitis. Hyperlipidemia is one of the risk factors for coronary heart disease while cholesterol is the major lipid constituent of atherosclerotic plaque.

The results from this study showed that the administration of *G. latifolium* leaf extract led to a significant decrease in values of triglyceride, total cholesterol, LDL-C and atherogenic index and a significant increase in values of HDL-C, showing a potential protective role against cardiovascular diseases. This effect may be due to inhibition of cholesterol absorption by saponin (a major constituent of *Gongronema latifolium*). These may also be due to saponin mediated interference of lipid metabolic pathways by the plant component (saponin) reducing plasma and post-mitochondrial fraction cholesterol, triglyceride, LDL-C and increasing plasma HDL-C levels or favoring the redistribution of cholesterol among the lipoprotein molecules. These effects indicate that *Gongronema latifolium* could be beneficial in preventing lipid abnormalities which may arise in certain metabolic disorders.

**CONCLUSION**

In conclusion, the plant possesses hypolipidaemic effects which may reduce the risk of cardiovascular diseases. The plant may also have hepatotoxic potentials particularly at high concentrations owing to the elevation of hepatic enzymes levels. Further studies on the possible mechanisms of the above effects of the plant are recommended.

**Competing Interests:** The authors have declared that no competing interests exist.

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