The Effects of Cymbopogon citratus (Lemon grass) on the Blood Sugar Level, Lipid Profiles and Hormonal Profiles of Wistar Albino Rats

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Abstract
The present study was undertaken to investigate the effect of extracts of Cymbopogon citratus on the blood sugar level, lipid profiles and hormonal profiles of normal rats. Oral administration of ethanolic and aqueous extract of C. citratus at a doses of 200 mg/kg body weight, for a period of 30 days, caused a significant (p<0.05) reduction in blood glucose levels. Effect on hormonal profile (TSH, T3, and T4) was also determined, and was found to be significantly higher in all the administered groups when compared with control. Lipid profiles levels; Total cholesterol, triglycerides, high density lipoprotein-cholesterol and low density lipoprotein-cholesterol were significantly (p>0.05) higher for all treated rats as compared against control. Findings in this study showed that this plant has hypoglycemic properties and did not exert oxidative damage to the heart and the various hormonal profiles as well as its relative safety and possible use for weight gain.

Keywords: medicinal plants; Blood glucose; Cymbopogon citratus hypoglycaemic oxidative damage

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Introduction

*Cymbopogon citratus* commonly called lemon grass is an aromatic, perennial grass belonging to the family grimeae [1]. It is a tropical plant, grown as an ornamental in many temperate areas with a height of about 1.8m and its leaves 1.9cm wide covered with a whitish bloom [2]. In certain medications, it is used for mental illness. It is an antifungal, antitoxicant and deodorizing agent. In combination with other herbs, it has large use as cure for Malaria [2].

One of the main constituents of the many different species of lemongrass (genus *Cymbopogon*) is citral (3,7-dimethyl-2,6-octadieni-1-al) [3,4]. Lemongrass oil has been found to contain up to 75-85% citral [5]. Lemongrass also contains z-citral, borneol, estragole, methyleugenol, geranyl acetate (3,7-dimethyl-2,6-octadieni-1-ol acetate), geraniol (some species higher in this compound than citral), beta-myrcene (MYR, 7-methyl-3-methylene-1,6 octadiene), limonene, piperitone, citronellal, carene-2, alpha-terpineole, pinene, farnesol, proximadiol, and (+)-cymbodiaetylal [6]. The volatile oil from the roots contains 56.67% longifolene-(V4) and 20.03% selina-6-en-4-ol [7]. In particular, a study of *Cymbopogon martini* isolated fatty acids, common sterols, and 16-hydroxypentacos-14(z)-enoic acid [8].

The reactive oxygen species produced in cells include hydrogen peroxide (H$_2$O$_2$), hypochlorous acid (HClO), and free radicals such as the hydroxyl radical (·OH) and the superoxide anion (O$_2^-$) [9]. The hydroxyl radical is particularly unstable and will react rapidly and non-specifically with most biological molecules. These oxidants can damage cells by starting chemical chain reactions such as lipid peroxidation, or by oxidizing DNA or proteins [10].

Triglycerides, as major components of very-low-density lipoprotein (VLDL) and chylomicrons, play an important role in metabolism as energy sources and transporters of dietary fat. They contain more than twice as much energy (approximately 9 kcal/g or 38 kJ/g) as carbohydrates (approximately 4 kcal/g or 17 kJ/g) [11]. In the human body, high levels of triglycerides in the bloodstream have been linked to atherosclerosis and, by extension, the risk of heart disease and stroke [12]. Cholesterol is an organic molecule. It is a sterol (or modified steroid) [13], and an essential structural component of animal cell membranes that is required to establish proper membrane permeability and fluidity. Cholesterol is thus considered within the class of lipid molecules. In addition to its importance within cells, cholesterol also serves as a precursor for the biosynthesis of steroid hormones, bile acids, and vitamin D. Cholesterol is the principal sterol synthesized by animals. All kinds of cells in animals can produce it. In vertebrates, the hepatic cells typically produce greater amounts than other cells [14].

The purpose of this study, therefore, is to carry out the effects of *cymbopogon citratus* (lemon grass) on the blood sugar level, lipid profiles and hormonal profiles using wistar albino rats.

Materials and methods

*Cymbopogon citratus* was harvested and collected freshly from a native farms and authenticated in Environmental Biology Laboratory, Department of Science Laboratory Technology, Rufus Giwa
Polytechnic, Owo.

**Preparation of plant extract**

The fresh plant was washed, chopped into pieces and air-dried at room temperature. The dried plant part was milled into powder and weighed. The Plant powder was divided into two groups. One portion was soaked in 90% absolute ethanol to obtain the ethanolic extract and the other group in distilled water to obtain the aqueous form separately in a container for 72 hours with intermittent shaking. Then, it was filtered through a muslin clothe and later Whatman No. 1 filter paper.

The resulting filtrate was evaporated under reduced pressure using a rotary evaporator and there after freeze dried to get powder form of both ethanolic and aqueous extracts. The yield was stored in a refrigerator (4°C) till when needed [15].

**Experimental animal**

Male albino rats (Wistar strain) weighing between 109-170g, purchased from the central animal house of University of Ibadan were used for the study.

**Acclimatization:** 15 days prior to dosing.

**Identification of animals:** By cage number.

**Diet:** Pelleted feed

**Water:** Potable drinking water

**Housing & Environment:** 4 animals each in a group

**Determination of the weight of animals**

The weights of the animals were weighed using an electronic weighing balance every 7 days to verify and quantitate the change in weight over the period of administration.

**Animal ethics**

All of the animals received humane care according to the criteria outline in the Guide for the Care and the Use of Laboratory Animals prepared by the National Academy Science and published by the National Institute of Health (USA). The ethic regulations have been followed in accordance with national and institutional guidelines for the protection of animals’ welfare during experiments.

**Experimental design**

To carry out this study, twelve male Albino rats were randomly, equally divided and assigned to either control or experimental groups. The control group received 2ml distilled water while the experimental rats group received oral doses of 200mg/kg for both the ethanolic and aqueous extracts of *Cymbopogon citratus* dissolved in 2ml distilled water through a stainless steel intra-gastric intubation and administered for 30 days. Here, the blood glucose levels and hormonal, were determined and recorded.

**Chemicals and reagents preparation**

All chemicals were of an analytical grade and are supplied from sigma chemical co. USA. Distilled water was used in all biochemical assays.

**Blood glucose determination**

The blood of the rat was drawn from the tail and measured the blood glucose levels by using Accuchek glucometer. The body weights of the rats were also recorded.
Hormonal Assays
Blood samples were collected in glass tube from retro-orbital puncture to obtain haemolysis free clear serum for the analysis of hormonal assays [16].

Lipid profile assay
Serum was used to assay for the following parameters: total cholesterol [17], triglycerides [18], high density lipoprotein cholesterol [19], very low density lipoprotein cholesterol (triglycerides/5) and low density lipoprotein cholesterol [20].

Blood sample preparation
At the end of the experiment, rats were fasted for 12 to 14 h. Blood were collected by cardiac puncture from the rats at fasting state after being anesthetized with chloroform. The blood samples were collected in plain tubes, allowed to coagulate at room temperature and centrifuged at 3500 rpm for 15 min at room temperature for separation of serum. The clear, non-haemolysed supernatant was separated using clean dry Pasteur pipette and stored at -20°C. Serum was used to assay for the lipid profile levels of the rats.

Statistical analysis
The experimental results were expressed as the mean ± S.E.M. Statistical significance of difference in parameters amongst groups was determined by One way ANOVA followed by Duncan’s multiple range test. P<0.05 was considered to be significant.

Results

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>Ethanolic extract</th>
<th>Aqueous extract</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Initial weight</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(g)</td>
<td>115 ± 1.5</td>
<td>130 ± 1.71⁹</td>
<td>135 ± 1.50⁶</td>
</tr>
<tr>
<td><strong>Final weight</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(g)</td>
<td>129 ± 1.7</td>
<td>150 ± 1.50⁹</td>
<td>149 ± 1.70⁶</td>
</tr>
</tbody>
</table>

Values are expressed as means ± SEM of four independent experiments. Means in the same column not sharing the same letter(s) are significantly different (p < 0.05)
Table 2 Effects of oral administration of ethanolic and aqueous extracts of Cymbopogon citratus on Serum glucose in normal rats.

<table>
<thead>
<tr>
<th>Group</th>
<th>Day 0(mg/dl)</th>
<th>Day 7(mg/dl)</th>
<th>Day 14 (mg/dl)</th>
<th>Day 21(mg/dl)</th>
<th>Day 28(mg/dl)</th>
<th>Day 30(mg/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>91.5 ± 7.78</td>
<td>85.3 ± 3.78</td>
<td>87.0 ± 2.82</td>
<td>95.33 ± 4.93</td>
<td>97.7 ± 2.08</td>
<td>101.0 ± 4.00</td>
</tr>
<tr>
<td>Ethanolic extract</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(200mg/kg)</td>
<td>104.0 ± 8.41</td>
<td>94.5 ± 4.95</td>
<td>75.0 ± 10.61</td>
<td>80.0 ± 2.28</td>
<td>74.5 ± 9.19</td>
<td>79.48 ± 7.78</td>
</tr>
<tr>
<td>Aqueous extract</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(200mg/kg)</td>
<td>103.3 ± 6.8</td>
<td>98.5 ± 16.2</td>
<td>89.10 ± 2.83</td>
<td>84.50 ± 2.83</td>
<td>79.0 ± 2.12</td>
<td>81.0 ± 10.61</td>
</tr>
</tbody>
</table>

Values are expressed as means ± SEM of four independent experiments. Means in the same column not sharing the same letter(s) are significantly different (p < 0.05)

Blood sugar level of the plant was determined and no significant (p<0.05) difference was observed rats in control and the ethanolic extract treated rats. Administration of single dose at 200mg/kg of the plant ethanolic and aqueous extracts did not show any reduction in blood glucose level in normal rats with single dose administration study on day ‘7’. There was a significant increase in levels at intervals of 0 and 7 days.

Result showed that administration of both ethanolic and aqueous extracts of Cymbopogon citratus at a dose of 200 mg/kg body weight for a period of 30 days to the test animals caused a steady decrease in their blood glucose level. The significant (p<0.05) decrease in the blood glucose of the test animals as compared to the control is a reflection of the hypoglycemic effect of the plant.

Table 3 Effects of oral administration of ethanolic and aqueous extracts of Cymbopogon citratus on Serum lipid profiles in normal rats.

<table>
<thead>
<tr>
<th>Group</th>
<th>TotalCholesterol (mmol/l)</th>
<th>Triglycerides (mmol/l)</th>
<th>HDL (mmol/l)</th>
<th>LDL (mmol/l)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>30.54 ± 2.79</td>
<td>29.21 ± 6.09</td>
<td>36.18 ± 3.01</td>
<td>15.60 ± 1.34</td>
</tr>
<tr>
<td>Ethanolic extract</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(200mg/kg)</td>
<td>28.82 ± 4.83</td>
<td>19.89 ± 3.99</td>
<td>42.03 ± 0.27</td>
<td>14.91 ± 3.42</td>
</tr>
<tr>
<td>Aqueous extract</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(200mg/kg)</td>
<td>29.22 ± 2.41</td>
<td>21.05 ± 2.82</td>
<td>41.90 ± 0.63</td>
<td>16.11 ± 0.42</td>
</tr>
</tbody>
</table>

Values are expressed as means ± SEM of four independent experiments. Means in the same column not sharing the same letter(s) are significantly different (p < 0.05)

HDL- High Density Lipoprotein, LDL- Low Density Lipoprotein
The lipid profiles of rats treated with both ethanolic and aqueous extracts of lemon grass was also observed. The Total cholesterol, Triglycerides, High density lipoprotein (HDL) and low density lipoprotein (LDL) levels of the ethanolic extracts were observed to be significantly lowered when compared with the aqueous extract and the control normal rats. The results of the study also showed that the level of the LDL-Cholesterol in both ethanolic and aqueous extracts were significantly (p<0.05) lowered when compared with the control group and the level of the HDL-Cholesterol in the treated groups. Thus, blood serum cholesterol level was found to be down regulated in this study.

Table 4 Effects of oral administration of ethanolic and aqueous extracts of lemon grass (Cymbopogon citratus) on Hormonal profiles in normal rats.

<table>
<thead>
<tr>
<th>Group</th>
<th>TSH (µIU/ml)</th>
<th>T3 (ng/ml)</th>
<th>T4 (nmol/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>1.17 ± 0.11</td>
<td>0.69 ± 0.064</td>
<td>35.31 ± 1.07</td>
</tr>
<tr>
<td>Ethanol extract (200mg/kg)</td>
<td>2.66 ± 0.44</td>
<td>1.01 ± 0.00</td>
<td>34.93 ± 1.62</td>
</tr>
<tr>
<td>Aqueous extract (200mg/kg)</td>
<td>2.19 ± 0.065</td>
<td>0.93 ± 0.025</td>
<td>35.89 ± 0.62</td>
</tr>
</tbody>
</table>

Values are expressed as means ± SEM of four independent experiments. Means in the same column not sharing the same letter(s) are significantly different (p < 0.05)

TSH - Thyroid-stimulating hormone (thyrotropin), T3- Triiodothyronine, T4 – thyroxine

The results of the hormonal assays showed that there is a significant (p>0.05) increase in the level of the Thyroid stimulating hormones (TSH) in the ethanolic extract treated rats and aqueous extract treated rats when compared with the normal control. In the same vein, the level of Triiodothyronine (T3) in both the ethanolic and aqueous extracts treated rats was significantly (p<0.05) higher as compared with the control. And no significant (p<0.05) difference was observed in thyroxine (T4) level of both the ethanolic and aqueous extracts of C. citratus when compared with the normal control animal.

Discussion

Many indigenous medicinal plants have been reported by various authors to have hypoglycemic effects [21]. Some of these hypoglycemic medicinal plants have been shown to significantly reduce blood glucose concentration in normal and diabetic animals. These plants, for example C. citratus, tend to participate in the tight regulation of blood glucose levels as a part of metabolic homeostasis.

The result in table 2 showed that administration of both ethanolic and aqueous extracts of C. citratus at a dose of 200 mg/kg body weight for a period of 30 days to the test animals caused a steady decrease in their blood glucose level. The significant decrease in the blood glucose of the test animals as compared to the control is a reflection of the hypoglycemic effect of the plant.

Recent Phytochemical studies have shown that the hypoglycaemic effect of any plants is due to presence of tannins and polyphenols singly or in combination having anti-oxidant property [22]. Tannic acid possesses glucose transport-stimulatory and adipocyte differentiation-inhibitory activity and induces
GLUT 4 translocation [23], while polyphenols inhibit α-glucosidase enzyme from the intestine and initiate release of insulin from the beta cells of pancreas [24]. Based on increased insulin levels in rats treated with C. citratus extracts, it can be suggested that the possible mechanism of action of both ethanolic and aqueous extract from C. citratus could be related to anti-oxidant activity that aids to recovery from impaired glucose metabolism through release of insulin from the pancreas.

In diabetes, elevation of blood glucose is a consequence of increased hepatic glucose output along with reduced peripheral glucose utilization. Insulin deficiency is clearly associated with a change in hepatic metabolism [25]. Moreover, a reduction in insulin-mediated glucose uptake caused by decreasing gene expression of GLUT-4 has been reported in skeletal muscle, a major site for glucose disposal, in diabetic rats [26].

Mohammad et al., [27] has earlier reported the hypoglycemic activity of aqueous extract of G. lucidum in Wistar rats. Over 400 medicinal plants are available globally for the medication of diabetes mellitus, with a few having been subjected to scientific authentication to ascertain their effectiveness as anti-diabetic agents [15]. Substances with hypoglycemic properties would be effective in the management of diabetes mellitus [21].

The lipid profiles: Total cholesterol, Triglycerides, High density lipoprotein (HDL) and low density lipoprotein (LDL) levels of the ethanolic extracts as indicated in table 3 were observed to be significantly lowered when compared with the aqueous extract and the control normal rats. The results of the study also showed that the level of the LDL-Cholesterol in both ethanolic and aqueous extracts were significantly (p<0.05) lowered when compared with the control group and the level of the HDL-Cholesterol in the treated groups. Thus, blood serum cholesterol level was found to be down regulated in this study. It is known that high blood cholesterol levels and hyper-lipidemia can be the consequence and frequently associated with diabetes [28,29]. The concomitant protein stabilization and the elevation in the serum cholesterol levels are considered an added value of this plant protective mechanism. These events and together with the hypoglycemic properties of this plant indicated that it can be considered as a preventive factor for long-term complications of diabetes. These findings are in agreement with other cholesterol modulating effects of several other plants [30]. The reduction of this lipid profile in rats after treatment can be attributed to their promotion in utilization glucose reflected by a decrease in blood glucose and elevated insulin levels and hence depressed mobilization of fat [30]. Elevated blood triglyceride and cholesterol are some of the diabetic indicators and significant decreases of these parameters obtained in this study (Table 3) were indicative of the potentials of extracts of C. citratus to reduce plasma levels of these indicators to asymptomatic limit. These reductions of both cholesterol and triglyceride concentrations in the ethanolic extracts treated normal rats could be beneficial in preventing diabetic complications and improving lipid metabolism in diabetics [31]. Hypercholesterolemia plays an important role in the initiation and progression of atherosclerosis and is known to have a positive correlation with cardiovascular disease, largely depending on the oxidation of LDL, the main cholesterol carrier in plasma [32]. Flavonoid and saponin contents of extracts of T. scleroxylon may have also contributed to the reduction in the blood levels of triglyceride and cholesterol in experimental rats as has been widely reported by Prohp and Onoagbe [33].

Increase in LDL (bad cholesterol) or lethally dangerous lipoprotein carry a lot of health risks as its accumulation could occlude major arteries causing myocardial infarction, atherosclerosis, heart attack, stroke and even high blood pressure. Thus, both extracts significantly reduced LDL.

Avci et al., [34] reported an increase in HDL in male Swiss albino mice by aqueous and ethanol extracts of Agrostemma githago, Potentilla reptans, Thymbra spicata, Urtica dioica and Viscum album. HDL carries cholesterol and cholesterol esters from the peripheral tissues and cells to the liver, where
cholesterol is metabolised into bile acids. This pathway plays a very important role in reducing cholesterol levels in the blood and peripheral tissues and in inhibiting atherosclerotic plaque formation in the aorta [35,36]. The increased levels of HDL (good cholesterol) concentrations obtained in this study were indicative of potentials of extracts to protect against heart diseases.

For the hormonal assays, Uboh et al., [37] reported that exposure to gasoline vapours increased serum FSH, LH and testosterone in male rats and decreased serum FSH, LH and estradiol and progesterone in female rats. Testicular androgens (testosterone) are responsible for the growth and development of male urogenital system and the accessory sex organs. The prostate gland tissue is also known to be a testosterone dependent organ and when the testosterone level decreases as a result from an increasingly pronounced metabolic syndrome [38,39], the growth-stimulating effect on the prostate gland by other aberrations might possibly be reduced. Niu et al., [40](2003) also stated that the hormonal anatomical and functional growth of the prostate is mainly controlled by androgens. The hormonal hypothesis seems to be one of the most important hypotheses in prostatic cancer etiology, and efforts are continuing to improve the understanding of androgen action in prostatic cancer [41]. Although evidence from epidemiological studies of an association between circulating levels of androgens and prostatic cancer risk has been inconsistent, the traditional view that higher testosterone represents a risk factor for prostatic cancer appears to have little evidentiary support [42].

The increasing trend of blood hormones Thyroid stimulating Hormones and testosterone on oral administration of Cymbopogon citratus (table 4) was novel since no works has earlier reported these. Raji et al., [43] found the mean serum testosterone level of rats treated with 400 mg/kg of the Morinda lucida leaf extract for 4 and 13 weeks significantly increased (p < 0.01) compared with the controls. He also stated that the extract caused an increase in the weight of the testes, which was accompanied by an increase in the serum levels of testosterone. Similar changes have been reported with the extract of Zingiber officinale and Pentadiplendra brazzeana in rats [44]. Others have reported testicular weight reduction accompanied by decreased serum testosterone levels in male rats treated with the extracts of Quassia amara [45], Azadirachta indica [28] and gossypol, a phenolic compound extracted from the cotton seed[45]. The MEPH treatment may lead to carcinogenesis. Thus, it can be inferred that C. citratus extracts has no negative effects on the various hormonal profiles tested for.

**Conclusion**

In conclusion, *Cymbopogon citratus* are recommended cardiac glycoside and the cardiac glycosides serves as defence mechanisms against cardiovascular disease and digestive problems. Thus, *Cymbopogon citratus* (Lemon grass) whole plant materials are recommended to be taken because it has many beneficial effects in human health (Ozcan et al., 2009).

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