

Effects of Aqueous Stem Bark Extract of *Cissus Populnea* on Serum Lipid Profile in Normal and Diabetic Induced Rats

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Abstract:

The study form four weeks experimental protocol involving intragastric administration of 100mg/Kg body weight of aqueous stem bark extract of *Cissus populnea* in normal and alloxan induced diabetic albino rats. In the diabetes induced group there is a significant increase ($P < 0.05$) in the serum level of cholesterol (2.80 ± 0.46 mmol/L 1.83 ± 0.08 mmol/L). The result also revealed a significant increase in the serum level of HDL cholesterol (1.51 ± 0.08 mmol/L from 1.08 ± 0.05 mmol/L). and of triglyceride (1.52 ± 0.10 mmol/L from 0.78 ± 0.01 mmol/L). However, no significant differences was observed in the serum level LDL cholesterol (1.30 ± 0.01 mmol/L and 1.30 ± 0.12 mmol/L). In the diabetic group administered with the extract there was a decrease in the level of cholesterol (1.83 ± 0.08 mmol/L to 1.06 ± 0.15 mmol/L). All other three lipid parameters increased significantly ($P < 0.05$)

Key words: *Cissus populnea*, lipid profile, cholesterol, Triglyceride, HDL cholesterol.

Introduction

There is wide spread and continuous search for biologically interesting natural products in the flora, fauna and microbial organisms in the developing world. this has led to revelation of a number of interesting alkaloids, antibiotics, terpenoids and flavonoids among others. With the recent breakthroughs in the field of science and engineering highly precise analytical equipment have been developed that makes it easy to analyze the molecular structure of many natural products. However, studies on the bioactivity of identified natural products on key biochemical parameters in experimental animals in health and disease

conditions are noble, recommended and encouraged by the World Health Organization (WHO).

Cissus populnea, the threaded part of the stem bark of which is used in making soup in most homes, especially in north central Nigeria. it is believed that it contains biological principles that help lower the risk of coronary heart diseases such as atherosclerosis and other related diseases. the main purpose of this study is to examine the possible hypolipidaemic activity of the stem bark aqueous extract in normal and alloxan induced albino rats.

Materials and Methods

Experimental Plant Material.

Cissus populnea (Family Ampelidaceae) locally called “okoho” purchased fresh from the open market in Jos was harvested from the wild in Tilden Fulani (Longitude 9⁰ Latitude 10⁰) on the out skirts of Jos, Nigeria in winter (Tyler et al, 1976). It was cleaned by scrapping the powdery ash coloured bark with a knife. This was then pounded lightly with a large pestle to expose the strips of thread-like part containing the mucilagenous substance. These were separated from the inner wooden core and then room dried (Tyler et al, 1976). The dried threaded strips were then pounded in a mortar to get a clear brownish ground *Cissus*. This was stored in a glass stoppered brown bottle until use.

Procedure with rats

The experimental animals, which were acclimatized for one week, were reweighed and divided into four groups of nine rats each. The four groups and the treatments they received were as follows: -

Group I: Normal Control group on unrestricted but measured standard diet and water ad libitum for 4 weeks,

Group II: Normal rats on unrestricted but measured standard diet and water ad- libitum and intubated with 10mg/ml/100g body weight aqueous extract of *Cissus* stem bark for 4 weeks.

Group III: Diabetic control on unrestricted but measured standard diet and water ad -libitum for 4 weeks.

Group IV: Diabetic rats on unrestricted but measured diet, water ad- libitum and intubated with 10mg/ml/100g body weight aqueous extract of *Cissus* stem bark for 4 weeks.

Diabetes Induction Test Result

The intraperitoneal injection in a single dose of 17.5mg/100g body weight of alloxan monohydrate produced a fasting average blood glucose concentration of 189mg/dl \pm 5.2 for groups III and IV after 48 hours. This result is consistent with the establishment of hyperglycemia and as a corollary diabetes mellitus.

Estimation of Serum Cholesterol

This was determined by enzymatic method by the procedure of Allain et al (1974). cholesteryl esters in serum are hydrolyzed by cholesterol ester hydrolase. All of the cholesterol is then oxidized by cholesterol oxidase to the corresponding ketone, with a shift in the location of double bond. The H₂O₂ generated by the oxidation is decomposed by horseradish peroxidase in the presence of 4-aminoantipyrine and phenol to yield a quinoneimine dye. The absorbance of the dye, measured at 500nm, is proportional to the cholesterol concentration. All of the enzymes and chromogen are contained in a single, buffered reagent.

Estimation of Serum HDL - Cholesterol

Low density lipoproteins (LDL and VLDL) and chylomicron fractions are precipitated quantitatively by the addition of phosphotungstic acid in the presence of magnesium ions. After

centrifugation, the cholesterol concentration in the HDL fraction, which remains in the supernatant, is determined by the enzymatic method of Allain et al, (1974).

Estimation of Serum LDL- Cholesterol

Low-density lipoproteins (LDL) are precipitated by heparin at their isoelectric point (pH 5.04) After centrifugation the high-density lipoprotein (HDL) and the very low-density lipoprotein (VLDL) remain in the supernatant. LDL cholesterol can then be determined by the enzymatic method of Weiland and Seidel (1983).

Estimation of Serum triglycerides

This was determined by the enzymatic method as described by Wahlefeld (1974).

Estimation of Total Protein

This was determined by the biuret method of Tietz (1970) in serum, kidney and liver. The method is based on the fact that polypeptide and protein peptide bonds produce, when reacted with Cu^{2+} ions in an alkaline medium a complex compound of violet colour, whose colour intensity is proportional to the protein concentration in the medium.

Estimation of Serum albumin

The procedure was based on the bromocresol green method as described by Silverman *et al* (1986). The procedure is based on the principle that bromocresol green, an anionic dye, binds tightly to albumin when added to serum, and the complex absorbs light much more intensely at pH 4.20 and 628nm, than does the unbound dye. The

increase in light absorption is directly proportional to the albumin concentration.

Statistical analysis

The data from this study were statistically analyzed to test for any significant difference within and between the test groups and control groups. The results obtained are tabulated and presented in table 1 below. Statistical analysis was performed using the Mann-Whitney two-sample test. A p value of <0.05 was considered statistically significant.

Discussion

The result of the serum lipid analysis in four weeks alloxan induced diabetes mellitus suggests that diabetes causes an increase in the serum level of total cholesterol and triglycerides. Increases were also observed in the serum levels of LDL- and HDL-cholesterols (Table 3.4). Earlier studies by Stamler et al (1975), Fuller (1985); O'meara et al (1991) and Nikkila (1984) have shown that hyperlipidaemia, due to the increases in the above lipids are a common feature in both diabetic and hypertensive patients. Hyperglycaemia in diabetics may trigger an increase in activities of some lipogenic enzymes. Increases in the level of NADH, induced by the derangements associated with diabetes, stimulates fatty acids and glycerol phosphate synthesis and inhibit fatty acid oxidation (Bobboi et al, 1999) Cissus stem bark intubations caused highly significant decreases in the serum levels of total cholesterol, triglycerides, HDL- and LDL-cholesterol (table3.4) in non-diabetic and diabetic control groups. This agrees

with the findings of Milgate and Roberts (1995) and Cheeke (1971) who reported that some of these phytochemicals form complexes with cholesterol and bile in the gastrointestinal tract leading to reduced blood concentration of cholesterol. These complexes also interfere with entero-hepatic circulation inducing the formation of bile from cholesterol. Tannins, flavonoids, anthraquinones, and others such as arginine that are present in *Cissus* stem bark may be responsible for the hypolipidaemia observed in the groups treated with the extract. The increase by about the same extent of the levels of HDL- and LDL-cholesterol, however, as a result of *Cissus* stem bark administration does not portray a beneficial property of the plant. This is because the benefits of hypolipidaemic properties (low risks of coronary heart disease) of *Cissus* stem bark is assumed to manifest as containing substance(s) that lower the levels of total and LDL-cholesterols, and raising the levels of HDL-cholesterol (Lipid hypothesis). Nevertheless in spite of statistical and experimental ambiguities, professionals who deal with patients have interpreted epidemiological results as demonstrating that hypertriglyceridaemia signals the presence of increased coronary risk (Nikkila et al, 1978; Schonfeld, 1983). However, among individuals with significant atherosclerotic coronary disease and survivors of myocardial infarction, a sizeable proportion are found to have one or another form of hyperlipoproteinemia and lesion severity is related with lipoprotein levels (Schonfeld, 1983).

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