

Full Length Research Paper

Anti-atherogenic effects of supplementation with vitamin B6 (Pyridoxine) in albino rats

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Elevation of serum lipids have been implicated to predispose to cardiovascular disorders which includes hypertension, stroke, atherosclerosis etc. Various studies have reported links between supplementation with vitamin B complex and reduction in oxidative stress and inflammatory reactions, two events associated with cardiovascular disorders. Data on possible direct regulation on lipid profile by vitamin B6 is very scanty. This study is therefore designed to assess role of administration of vitamin B6 in regulation of serum lipids such as total cholesterol, phospholipids, triglycerides, and high density lipoprotein cholesterol (HDL-C). Results showed that serum total cholesterol and triglyceride were significantly reduced ($p \leq 0.05$) in rats placed on vitamin B₆ supplements compared with control group. Serum phospholipids and high density lipoprotein cholesterol were significantly elevated ($p \leq 0.05$) in rats placed on vitamin B₆ supplements when compared with control group. The data showed anti-hyperlipidemic effects of vitamin B6 supplementation in rats.

Key words: Pyridoxine, lipids, cardiovascular disorder, supplement.

INTRODUCTION

Cardiovascular diseases (CVD) are caused by disorder of the heart and blood vessels and include the coronary heart disease, raised blood pressure that is, hypertension etc. Major traditional risk factors of CVD include elevated serum levels of total cholesterol, triglyceride, low density lipoprotein and reduced serum concentration of high density lipoprotein among other things. The disease is the most common cause of sudden death (Thomas et al., 1988) and is also the most common reason for death of men and women over 20 years of age (American Heart Association, 2007). According to present trends in the United States, half of healthy 40-year-old males will develop coronary artery disease (CAD) in the future, and one in three healthy 40-year-old women (Rosamond et al., 2007). Traditionally, these conditions are considered to result from lipid abnormalities, and other risk factors such as hypertension, smoking, hyperhomocysteinemia and diabetes (Niessen et al., 2003).

The conditions have caused lots of losses in both human and material resources. Among recommended

antidote to this include adjustment in lifestyle which involves nutritional modifications, slight physical exercise and in some instances use of drugs (Djousse et al., 2009).

A connection between homocysteine (a sulphur containing amino acid) and cardiovascular disease was proposed when it was observed that people with a rare hereditary condition called homocysteinuria are prone to develop severe cardiovascular disease in their teens and twenties and this was confirmed by the study of David et al. (2002). In this condition an enzyme deficiency causes homocystiene to accumulate in the blood and to be excreted in the urine. Elevations in plasma homocystiene level promote oxidative damage, inflammation and endothelial dysfunction and are independent risk factor for cardiovascular disease.

Furthermore, epidemiological studies have shown plasma homocysteine levels are inversely related to plasma levels of folate, B12 and B6- the three vitamins involved in the conversion of homocystiene to methionine and cysteine.

This study is designed to assess possible lipid-lowering effect of administration of vitamin B6 and also the protective effect of the administration towards development of cardiovascular disease.

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Table 1. Effect of vitamin B₆ supplement on total cholesterol, triglyceride, phospholipid and HDL-cholesterol.

Biochemical parameters	Control group	Vitamin B6 group
Total cholesterol	130.96 ± 19.84	109.62 ± 19.64*
Triglyceride	160.32 ± 50.15	104.93 ± 13.04*
Phospholipids	103.33 ± 28.45	137.04 ± 47.12*
HDL-cholesterol	124.03 ± 62.79	150.60 ± 84.05*

Significant at * $p < 0.05$ when compared with control values.

EXPERIMENTAL DESIGN

Experimental animals

Sixteen 14-week old albino rats with an average weight of 136 g were purchased from commercial breeder in Ilorin, Kwara State. They were kept in a well-ventilated cage in the animal house of the Department of Anatomy, Ladoke Akintola University of Technology, Nigeria. The animals have unrestricted access to clean water and were fed commercial normal feed as described by Kong et al. (2009).

They were separated into 2 groups, each group consisting of eight rats. Group one were not given vitamin B supplement (control) while group two were given a therapeutic dose of 0.2 mg/kg body weight of vitamin B6 for 28 days. All animal procedures were in strict accordance with the NIH Guide for the Care and Use of Laboratory Animals.

Sample collection

On the 28th day, the rats were sacrificed and blood samples were obtained through cardiac puncture (Yin et al., 2008). The blood was collected into appropriately labeled sample bottles and centrifuged at 4000 rev/s for 5 min (Kong et al., 2008; Deng et al., 2009). The supernatants were decanted and stored at -2°C for analyses of biochemical parameters (Wu et al., 2010).

Determination of biochemical parameters

The biochemical parameters determined included total cholesterol, triglyceride, high density lipoprotein, and phospholipid.

Determination of total cholesterol

Total cholesterol was determined using enzymatic method described by Allain et al. (1974). Cholesterol esterase hydrolyses cholesterol esters to free cholesterol. The free cholesterol produced is oxidized by cholesterol oxidase to cholesten-4-ene-3-one with simultaneous production of hydrogen peroxide which couples with 4-aminoantipyrine and phenol in the presence of peroxidase to yield chromogen with maximum absorption at wavelength 510 nm. The colour intensity is proportional to the cholesterol concentration.

Determination of triglycerides

Triglyceride was determined using enzymatic method described by Buccolo and David (1973). Triglycerides are hydrolyzed by lipases to yield glycerol and fatty acids. The glycerol produced is oxidized

to dihydroxyacetone phosphate with the production of hydrogen peroxide which couples with 4-aminophenazone and 4-chlorophenol to produce a chromogen referred to as quinoneimine. The reaction is catalyzed by peroxidase. The degree of absorbance of the chromogen is directly proportional to the concentration of triglyceride measured at 505 nm.

Determination of high density lipoprotein

The precipitation method by Assmann et al. (1983) was used to determine HDL-cholesterol. The addition of phosphotungstic acid in the presence of magnesium ions precipitates quantitatively low density lipoprotein, very low density lipoprotein and chylomicron fractions from whole plasma, leaving the HDL fraction in the supernate. The cholesterol in the HDL which remains in the supernatant after centrifugation is estimated using the enzymatic method of Allain et al. (1974).

Statistical analysis

Quantitative data were presented as mean ± SD. Triglyceride, total cholesterol, phospholipids and high density lipoprotein between the two groups were compared using student's 't' test. A value of $p < 0.05$ was considered statistically significant.

RESULT

Table 1 shows the mean plasma concentrations of selected biochemical parameters (total cholesterol, triglyceride, phospholipids, HDL-cholesterol) in different experimental groups that is, control and group given vitamin B6.

The mean concentrations of total cholesterol and triglyceride were significantly decreased in group given vitamin B6 as compared with the values in the control group, while there were increases in the mean concentrations of phospholipids and HDL-cholesterol when compared with the values obtained in the control group.

DISCUSSION

Cholesterol plays a major role in human heart health and high cholesterol is a leading risk factor for the

development of human cardiovascular disease. It was observed that there was a significant decrease in the level of cholesterol in animals that were administered with vitamin B6 as compared to control animals. High plasma cholesterol has been linked with hyperlipidemia which is associated with increased risk of cardiovascular disease (Naito, 1984). Studies have proved that low cholesterol is a desirable level in the body for normal and proper function (Zuhani et al., 2010).

Triglyceride is a fat in the blood stream and high levels of triglycerides has been linked to atherosclerosis' (hardening of arteries) and by implication the risk of heart disease and stroke. It was observed that there was a significant decrease in the level of triglycerides in animals that were administered with vitamin B6 as compared with control animals. Studies have shown that low concentration of triglyceride is normal for the body (Zuhani et al., 2010). High triglyceride level does indicate a defect in the system and recent evidence strongly suggests that high serum concentration is significantly associated with cardiovascular disease (Altan et al., 2006).

High density lipoprotein-cholesterol (HDL-C) is referred to as the good cholesterol because of its relevance to the cardiovascular system. HDL-C helps to remove extra cholesterol from the body. Study had shown that when HDL-C is higher, there is lower chance of heart disease and that higher level of HDL-C predicts longevity (Zuliani et al., 2010). It was observed that there was a significant increase in the levels of HDL-C in animals that were placed on vitamin B6 compared to control animals. The observation is consistent with the findings of Balch (2006), who observed significantly elevated serum HDL-cholesterol in vitamin B6 administered rats.

Phospholipids are compound lipids that participate in the lipoprotein complexes which are thought to constitute the matrix of cell walls and membranes, the myelin sheath, and of such structure as mitochondria. It was observed that there was a significant increase in the level of phospholipids in animals that were administered with vitamin B6 compared to control animals. For the future study, the factional amino acids, such as arginine also should be considered to be used by combined with B₆. Recently, leading findings told us that arginine can enhance blood flow, antioxidant activity, protein synthesis, immune cell proliferation, and intestinal development, thereby improve animal health (Tan et al., 2009; Yin and Tan, 2010; Yao et al., 2008, 2011; Geng et al., 2011).

From this study, it was observed that the supplementation with vitamin B6 resulted in an increased serum concentration of HDL-cholesterol and phospholipids and a reduction in the serum level of total cholesterol and triglyceride. High levels of HDL-cholesterol and low levels of total cholesterol and triglyceride are indications of a good cardiovascular health. It may be suggested that administration of vitamin B6 can be encouraged as this may help in reducing the rate of

accumulation of total cholesterol and triglyceride, which is the two major factors of atherogenicity.

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