

Cardioprotective effect of Kolaviron (*Garcinia kola* seed extract) in cholesterol-fed rats.

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ABSTRACT.

Flavonoids – a group of polyphenolic substances are naturally present in vegetables, fruits, seeds and beverages such as tea and wine. Studies have shown that flavonoid intake is inversely correlated with mortality from coronary heart diseases and myocardial infarction. The effect of kolaviron (a flavonoid complex) extracted from *Garcinia kola* seeds on the organ weights (lungs, kidneys, heart, spleen and liver) of rats administered with cholesterol, five times a week, for eight consecutive weeks was investigated.

The results revealed that cholesterol administration at a dose of 30mg/day for eight consecutive weeks caused a significant increase ($p < 0.001$) in relative heart weights of the cholesterol-fed rats when compared with the control. However, co-treatment with kolaviron at doses 100 and 200mg/kg significantly ($p < 0.001$) reduced the cholesterol induced enlargement of the heart. This is a pointer to the cardioprotective potential of kolaviron; and thus suggests a possible use as a dietary supplement for the prevention and management of coronary heart diseases.

KEY WORDS: Flavonoids, kolaviron, *Garcinia kola*, cholesterol, coronary heart diseases (CHD).

INTRODUCTION:

Coronary heart diseases had continued to be a major cause of mortality in the United States, Europe and much of Asia despite changes in life style and the introduction of lipid lowering drugs (Brannwald, 1997). The developing countries are not totally spared as a result of the influence of the western culture and dietary habit. However, a long standing tenet of nutrition holds that people with diets rich in fruits and vegetables enjoy better health than people eating few fruits and vegetables. Consequently, research has sought for the components or compounds present in these fruits and vegetables which are responsible for this apparent health benefit. Besides, since the discovery of the French paradox; that is, the low cardiovascular mortality rate observed in Mediterranean populations in association with red wine consumption (Formica and Regelson, 1995); Research in the field of flavonoids which are present in red wine has been on the increase. Furthermore, epidemiological studies suggest a protective role of dietary flavonoids against coronary heart diseases (De Groot and Rauhen, 1998), (Knekt et al, 1996). More than four thousand varieties of these flavonoids have been identified, many of which are responsible for the attractive colours of flowers, fruits and leaves (De Groot and Rauhen, 1998). The widespread distribution of these flavonoids, their variety and their relative low toxicity compared to other active plant compounds mean that many animals, including humans ingest significant quantities in their diet.

Several other beneficial properties of flavonoids have since been ascertained. They are most commonly known for their antioxidant activity, reduction of oxidative stress and inhibition of low density lipoproteins (LDL) oxidation (Hanasaki et al, 1994). They also act as vasodilators of blood vessels, anti inflammatory and anti tumour agents (Knekt et al, 1997).

Kolaviron is a biflavonoid complex extracted from the seeds of *Garcinia kola* heckel which is commonly known as bitter kola. Bitter kola is a highly valued ingredient in African traditional medicine (Ayensu, 1978). Kolaviron has been shown to exhibit many pharmacological effects such as anti oxidant (Farombi et al, 2000), anti atherogenic (Adaramoye et al, 2005), anti hepatotoxic (Iwu et al, 1987), (Farombi et al, 2000), anti diabetic (Iwu et al, 1990) etc.

In the present study, the cardioprotective potential of this extract was investigated in rats rendered hypercholesterolemic by the administration of dietary cholesterol for eight consecutive weeks.

MATERIALS AND METHODS:

PLANT MATERIALS:

Garcinia kola seeds were obtained locally in Ibadan, Nigeria. A total of 3kg of peeled seeds were sliced, pulverized with electric blender and then air dried in the laboratory. Extraction of kolaviron was achieved using

the method of Iwu, *et al* 1990. Briefly, powdered seeds were extracted with light petroleum ether in a soxhlet extractor. The defatted dried marc was repacked and then extracted with methanol. The extract was concentrated and diluted to twice its volume with distilled water and extracted with ethylacetate (6x250ml). The concentrated ethylacetate fraction gave a yellow solid known as kolaviron. The extract was prepared into two concentrations (100 and 200mg/kg) using olive oil as a vehicle.

REPARTITION OF ANIMAL GROUPS:

Twenty male albino rats (wister strain), weighing between 130 -200g were used. The animals were fed on normal laboratory chow, purchased from Ladokun feeds Ibadan. Animals were given access to food and water ad libitum. They were distributed randomly into five groups of four animals each.

Group A served as the control group and received olive oil. Rats in group B (positive control) received kolaviron 100mg/kg. Those in group C received cholesterol only (hypercholesterolemic animals). Rats in group D and E were treated orally with kolaviron at 100 and 200mg/kg respectively and were simultaneously administered cholesterol. Olive oil served as the vehicle for kolaviron and cholesterol.

METHODS:

Dietary cholesterol was administered orally at a dose of 30mg/0.3ml per animal (Bhandari and Sharma, 1999). Kolaviron was administered at doses 100 and 200mg/kg (Iwu, 1985); (Farombi et al, 2000). Kolaviron and cholesterol were administered five times a week for a period of eight consecutive weeks.

CHEMICALS: All reagents used were of analytical grade and the purest quality available.

COLLECTION OF SAMPLES:

In the 8th week, the rats were fasted for about 12 hours prior to sacrifice. They were sacrificed by cervical dislocation; the thoracic region was cut open and the tissues to be used (lungs, kidneys, heart, spleen and the liver) were rapidly excised and immediately placed in ice-cold 0.25M sucrose to wash off the excess blood and cool off the tissue. The tissues were then blotted dry and weighed.

STATISTICAL ANALYSIS: Results were expressed as mean \pm SD (n =4). One way analysis of variance (ANOVA) was used for the data analysis. Duncan's multiple range tests at $p < 0.001$.

RESULTS:

Table 1: Effect of kolaviron (kv) on body weight, organ weight and the relative weight of visceral organs in cholesterol – fed rats.

Group	Initial body weight(g)	Final body weight(g)	Change in weight(g)	Lung weight(g)	Kidney weight(g)	Heart weight(g)	Liver weight(g)	Relative heart weight(g)	Relative liver weight(g)
control	163.0 \pm 5.00	257.3 \pm 11.4	94.3 \pm 19.2	1.2 \pm 0.3	1.2 \pm 0.1	0.7 \pm 0.1	5.7 \pm 0.7	0.29 \pm 0.01	2.2 \pm 0.1
Kv100mg/kg	176.3 \pm 4.15	237.5 \pm 12.5	61.2 \pm 21.8	1.6 \pm 0.3	1.2 \pm 0.3	0.7 \pm 0.1	5.7 \pm 0.6	0.29 \pm 0.01	2.4 \pm 0.1
Chol.	135.0 \pm 5.10	205.0 \pm 12.0	70.0 \pm 14.3	1.3 \pm 0.1	1.2 \pm 0.2	0.7 \pm 0.1	6.0 \pm 1.4	0.32 \pm 0.01	3.0 \pm 0.1
Chol. +kv100mg/kg	150.0 \pm 2.0	228.8 \pm 17.4	78.8 \pm 16.1	1.2 \pm 0.3	1.2 \pm 0.2	0.7 \pm 0.1	6.3 \pm 1.0	0.30 \pm 0.01	2.7 \pm 0.1
Chol. +kv200mg/kg	146.0 \pm 1.50	233.8 \pm 15.2	87.8 \pm 15.2	1.5 \pm 0.5	1.4 \pm 0.1	0.7 \pm 0.1	7.1 \pm 1.1	0.28 \pm 0.01	3.0 \pm 0.1

Data are the mean \pm SD (n=4). $P < 0.001$ compared with control. Chol. Cholesterol, kv kolaviron.

Table 1 depicts the effect of kolaviron (kv) on body weight, visceral organ weights and relative organ weight of cholesterol – fed rats.

Cholesterol administration at a dose of 30mg/day for eight consecutive weeks caused a significant increase ($p < 0.001$) in the relative heart weight of the hypercholesterolemic animals (group C) when compared with the control group. Co-treatment with kolaviron (groups D and E) significantly ($p < 0.001$) reduced the cholesterol induced enlargement of the heart. This reversal also seemed to be dose dependent. However, the increase in the relative weight of the liver caused by cholesterol administration was not significantly ($p < 0.001$) reversed following co-treatment with kolaviron at the doses of 100 and 200mg/kg. There was no significant effect produced by kolaviron administration at the same doses on the other visceral organs.

DISCUSSION:

Flavonoids – a group of natural substances with variable phenolic structures, are abundantly present in fruits, vegetables, grains, bark, roots, stems, flowers, tea and wine. In vitro experimental studies have shown that flavonoids possess anti-inflammatory, anti-allergic, anti-viral and anti-carcinogenic properties (Middleton, 1998).

Kolaviron – a complex mixture of biflavonoids, benzophenones and xanthenes (Waterman and Hussein, 1983) has been demonstrated to be anti-hepatotoxic (Akintonwa and Essien, 1990), anti-inflammatory (Igboko, 1987), anti-oxidant (Farombi et al, 2000) etc.

Dietary anti-oxidants have been observed to increase low density lipoprotein oxidative resistance in vitro (Esterbauer et al, 1991), (Reaven et al, 1993). It is postulated that ingesting antioxidants and minimizing free radical exposure may reduce low density lipoprotein's contribution to atherosclerosis and hence coronary heart diseases (Esterbauer et al, 1991).

In the present study, cholesterol administration produced a significant increase in the relative heart weight of the hypercholesterolemic animals. This could be as a result of cholesterol deposition on the arterial walls of these animals, leading to increased resistance to blood flow. Thus the heart in an effort to pump blood through the narrow arteries becomes enlarged and hence the increased weight.

However, the co-treatment with kolaviron at 100 and 200mg/kg reversed this increase in the relative heart weight of the pre-treated animals. It has been reported (Hammon, 1996) that flavonoids present in hawthorn berries have a favourable effect on blood pressure by its dilation of the blood vessels resulting in reduced peripheral resistance and increased coronary circulation. Hence hawthorn berries are utilized as cardiotonics, coronary remedies and antihypertensives (Grainger, 1994). Kolaviron probably working in the same way prevented the cholesterol induced enlargement of the heart, indicating that regular intake of kolaviron can protect against heart defects and failures.

This result conforms to epidemiological studies by (Degroot and Rauen, 1998) which suggests a protective role of dietary flavonoids against coronary heart diseases. The result also agrees with the report of Hertog et al, 1995 which established an inverse relationship between flavonoid intake and mortality due to coronary heart diseases.

Besides, garcinia biflavonoids have been found to inhibit lipid peroxidation in vivo (Farombi et al, 2000), (Adaramoye et al, 2005). Lipid peroxidation has been implicated in many pathological conditions that include coronary heart diseases and cancer (Mora et al, 1990).

CONCLUSION:

The present study revealed the cardioprotective influence of kolaviron on cholesterol-fed rats. Studies have shown that the biflavonoids of garcinia kola are pharmacologically active with several pharmacokinetic advantages over simple monomeric flavonoids. They survive first pass metabolism which inactivates most flavonoids (Iwu, 1986). Garcinia kola seeds therefore could be a potent preventive agent for coronary heart diseases.

However, further work needs to be done to ascertain the long term effect of kolaviron on these visceral organs.

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