



## **Antihyperlipidemic Efficiency of *Abelmoschus esculentus* (Okra) Fruits Varieties on Rats Fed High-Fat Diet**

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### **Authors' contributions**

*This work was carried out in collaboration among all authors. Authors AAN, MKA and AMW designed the study, wrote the protocol and managed the literature searches. Authors AAN and DHM performance the experiment and wrote the first draft of the manuscript. Author DHM performed the statistical analysis and managed the analyses of the study. All authors read and approved the final manuscript.*

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### **ABSTRACT**

**Background:** Okra fruits have been found to be efficient in managing hyperlipidemia. But, there are different varieties of Okra, and whether antihyperlipidemic efficiency varies with the varieties has not been reported.

**Aim of the Study:** The aim of the study was to validate antihyperlipidemic efficiencies of some Okra fruits varieties on rats fed high-fat diet.

**Methods:** The five varieties of okra fruit was each sliced, air dried and pulverized into powder then extracted with methanol (80%) using Soxhlet extractor and concentrated at 30°C in a rotary evaporator then finally air dried. Qualitative and quantitative phytochemicals, and proximate analysis were conducted on the extracts. Hyperlipidemia was induce by feeding rats with high-fat diet for 35 days, followed by treating with two selected Okra fruit varieties (*NHB-AI-B* and *Yar kolon*) which has the highest extract yield and phytochemicals for 21 days. Nine groups of five rats was

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used: groups 1-3 (hyperlipidemic rats received *NHB-AI-B* Okra fruit extract at different doses), group 4-6 (hyperlipidemic rats received *Yar kolon* Okra fruit extract at different doses), group 7 (positive control rats treated with 10mg/kg Atorvastatin), group 8 (normal control rats fed basal diet) and group 9 (negative control). Serum lipid profile were determined from each rat.

**Results:** The study confirmed the presence of some important phytochemicals like saponins, tannins, phenolics, flavonoids, fibre etc but differs in concentrations among the varieties where *Yar kolon* variety emerge highest in almost all the chemical compounds. Rat fed high-fat diet for 35 days developed hyperlipidemia as evident by the elevated triglyceride, cholesterol, LDL-C, body weight and suppressed HDL-C. When treated with extracts of *NHB-AI-B* and *Yar kolon* Okra fruits varieties, the altered lipid profile were significantly reversed toward normalcy with *Yar kolon* variety exerting the most efficient activity.

**Conclusion:** The study showed the five Okra fruit varieties possess same chemical compounds but differs in concentrations among varieties. The extracts of Okra fruits varieties exert significant antihyperlipidemic effect but in varied degrees suggesting variations in their efficiency. This, therefore calls for further study to compare more Okra varieties to determine which one is the most potent and its active agent.

**Keywords:** Antihyperlipidemic; fruits; high-fat diet; *NHB-AI-B*; okra, rats; varieties; *Yar kolon*.

## 1. INTRODUCTION

Hyperlipidemia is a group of metabolic disorders characterized by high level of lipids circulating in the blood. It is a key factor in the increase prevalence of cardiovascular diseases like atherosclerosis, stroke, coronary artery disease, myocardial infarction and cardiac sudden death [1]. Hyperlipidemia is one of the disorders for which stable treatment is yet to be discovered, and treatment with available synthetic drugs is still a challenge globally since their prolong usage are accompanied with problems like restricted efficacies and advanced side effects [2]. The challenges associated with these drugs has increases the tendency for the demand and dependence on alternative sources like natural products with few or no side effect and high efficacy.

Consumption of diets with excessive amount of fats content for a long time had shown to propagate hyperlipidemia in both human and animals [3]. High-fat diet was reported to promote elevation of serum triglyceride, total cholesterol and low density lipoprotein-cholesterol (LDL-C) which are indicators of increasing incidences of hyperlipidemia [4]. Elevation of cholesterol was reported to be due to stimulation of HMG-CoA reductase activity by the high-fat diet [5]. While, elevation of LDL-C resulted from the suppression of hepatic receptor-dependent LDL uptake by the fatty acids in the diet [6]. On the other hand, hyperlipidemia in general may be developed as a result of decreased catecholamine level accompanied by lowering of  $\beta_2$  - adrenergic

receptor function which in turn slow lipolysis of fat cells [7].

The plant '*Abelmoschus esculentus* L. (Moench)' known as ladies finger or Okra in English, is an important vegetable crop around the world [8]. In Nigeria, it is known as "*Kubewa*" (Hausa), "*O'okro*" (Igbo) and "*L'laa*" (Yoruba). It was reported that high intake of Okra led to the reduction in the risk associated to diseases like atherosclerosis and cancer [9]. Okra fibre was reported to decrease chances of cardiovascular diseases progression [10]. Research had also shown that Okra improves eye sight and help in maintaining healthy skin due to its vitamin A and beta carotene content [11].

In addition, extracts from Okra have been reported to exert antihyperlipidemic activities [11,12]. Literature survey showed that there are different varieties of Okra [13], and whether antihyperlipidemic efficiency varies with varieties has not been studied. This study therefore compared antihyperlipidemic efficiencies of two varieties (*NHB-AI-B* and *Yar kolon*) of Okra fruits commonly consumed in Bauchi State, Nigeria on rats fed high-fat diet.

## 2. MATERIALS AND METHODS

### 2.1 Materials

#### 2.1.1 Chemicals

All chemicals used for the study were of analytical grade and were obtained from Sigma Aldrich, England and British Drug House (BDH),

London. Reagent kits for assaying lipid profile were obtained from Randox Lab., UK.

### 2.1.2 Experimental animals

Forty-five Wistar albino rats about 4 weeks old were procured from the Animal House, Department of Biology, Bayero University Kano, Nigeria. They were kept in cages with free access to water and feed for two weeks for acclimatization. The principles of laboratory animal care [14] and ethical guidelines for investigation of experimental pain in conscious animals [15] was observed.

### 2.1.3 Plant collection/identification

The five Okra fruits varieties: *Clemson Spinless*, *LD-88*, *NHB-AI-B*, *NHAE-47-4*, and *Yar kolon* were obtained from the Teaching and Research Farm of the Faculty of Agricultural Science and Technology, Abubakar Tafawa Balewa University, Bauchi. They were authenticated and identified with Voucher number: 1914.

## 2.2 Methods

### 2.2.1 Plant extraction

The five varieties of Okra fruit were each extracted following the method described by Doreddula et al. [16] with modification in the extraction time (12 hours). The Okra fruits were sliced, air dried at 25°C and then pulverized using pestle and mortar. The powdered form of each variety was extracted with methanol (80%) using Soxhlet extractor for 12 hours at 25°C and concentrated at 30°C in a rotary evaporator then air dried. The dried extracts were separately kept in an air-tired containers in a refrigerator at 4°C until used.

### 2.2.2 Qualitative and quantitative phytochemical of okra fruits varieties

Preliminary qualitative phytochemical tests for phenols, flavonoids, alkaloids, tannins, saponins, and glycosides was conducted using methods described by AOAC [17]. Total phenolics was measured by Meda et al. [18] using Folin-Ciocalteu reagent. Total flavonoids was determined by Singleton et al. [19] method. Tannins was determined using Folin-Denis reagent as done by Polshettiwar et al. [20]. Saponin was measured using vanillin reagent (8 %) and diosgenin as reference using Baccou et al. [21] method. A standard curve was constructed using a standard solution for each and was extrapolated to determine the unknown.

### 2.2.3 Proximate analysis of okra fruits varieties

*Determination of Moisture content:* A 2 g of Okra fruit extract was placed in the crucible and heated at 105°C until a constant weight was attained. The moisture content was calculated as loss in weight of the original sample and expressed as percentage moisture content [22].

$$(\%) \text{ Moisture} = \frac{\text{Initial weight of sample} - \text{Final weight of samples}}{\text{Initial weight of sample}} \times 100$$

Moisture=Initial weight of sample-Final weight of samples/Initial weight of sample ×100

*Determination of Ash content:* Exalt 2 g of Okra fruit extract was placed in the crucible and ignited in a muffle furnace at 550°C for 6 hours. It was then cooled in a desiccator and weighed at room temperature to get the weight of the ash using the formula [22].

$$(\%) \text{ Ash content} = \frac{\text{Weight of ash}}{\text{Initial weight of sample}} \times 100$$

*Determination of Crude fibre content:* A 5 g of Okra fruit extract was boiled in 200ml of 1.25% H<sub>2</sub>SO<sub>4</sub> for 30 minutes, and afterwards filtered with a Buchner funnel. The residue was washed with distilled water until it was acid free. It was then boiled in 200 ml of 1.25% NaOH for another 30 minutes, filtered and washed until it was also alkaline free. It was then rinsed with 10% HCl and twice with ethanol. The residue was dried at 105°C in an oven. After cooling in a desiccator, it was ignited in a muffle furnace at 550°C for 90 minutes to obtain the weight of the ash [22].

$$(\%) \text{ Crude fiber content} = \frac{\text{Weight loss on ignition}}{\text{Initial weight of sample}} \times 100$$

$$\text{Crude fiber content} = \text{Weight loss on ignition} / \text{Initial weight of sample} \times 100$$

### 2.2.4 Formulation of high-fat diet

The high-fat diet was formulated using Super starter animal feed composed of the following: maize (46%), soybean meal (18.5%), groundnut cake (15%), fishmeal (2%), wheat offal (12.45%), bone meal (2%), oyster shell (3%), salt (0.25%), premix (0.25%), methionine (0.3%), and lysine (0.25) respectively. The basal diet was 100% super starter feed while the formulated high-fat diet composed of 75% super starter feed, 5% egg yolk and 20% palm oil.

### 2.2.5 Experimental design

The extracts of the five varieties of Okra fruits were each subjected to phytochemicals and

proximate analysis where two varieties (*NHB-AI-B* and *Yar kolon*) with the highest extract yields and phytochemicals were selected and used for antihyperlipidemic efficiency study in rats. A total of forty-five rats were randomly divided into nine groups of five each. Hyperlipidemia was induced in rats by feeding them with high-fat diet as done by Karam et al. [23] with modification in the feeding duration (35 days). After the 35 days feeding exercise, blood sample were collected from their tails after immersed in water bath at 45°C for 5 min to make the blood vessel swell in order to get the desire volume (1-1.5 ml). Serum separated from blood was used to ascertain lipid fractions like TG, TC, and HDL-C which were compared with values from rats fed basal diet to make sure the success of hyperlipidemic animal model [24]. The animals were continued feeding high-fat diet alongside treatment with extracts of *NHB-AI-B* and *Yar kolon* as follows;

Group I: Hyperlipidemic rats + 250 mg/kg body wt. *NHB-AI-B* okra fruit extract variety

Group II: Hyperlipidemic rats + 500 mg/kg body wt. *NHB-AI-B* okra fruit extract variety

Group III: Hyperlipidemic rats + 750 mg/kg body wt. *NHB-AI-B* okra fruit extract variety.

Group IV: Hyperlipidemic rats + 250 mg/kg body wt. *Yar kolon* okra fruit extract variety

Group V: Hyperlipidemic rats + 500 mg/kg body wt. *Yar kolon* okra fruit extract variety

Group VI: Hyperlipidemic rats + 750 mg/kg body wt. *Yar kolon* okra fruit extract variety

Group VII: Hyperlipidemic rats + 10 mg/kg body wt. Atorvastatin (Positive control)

Group VIII: Non-hyperlipidemic rats + Distilled water (Normal control)

Group IX: Hyperlipidemic rats + Distilled water (Negative control)

The extract doses were determined from the studies by Ngoc et al. [10] and Sabitha et al. [12].

### 2.2.6 Determination of water and feed intake

Water and feed intake of the experimental animals were determined daily, water was measured using measuring cylinder before giving to the animals and after 24 hours. Feed

was also weighed using weighing balance before giving to the animals and after 24 hours.

### 2.2.7 Determination of body weight

The weight of the experimental rats was determined weekly throughout the experimental period. The weight was taken in the morning before feeding the rats by properly placing each rat in the weighing pan of the weighing scale and then the weight recorded.

### 2.2.8 Blood samples collection

Animals were sacrificed after 21 days treatment, they were anaesthetized by putting in a plastic jar saturated with chloroform vapor followed by cervical dislocation. Blood was collected in a labelled test tubes and serum removed was used for lipid profile determinations.

### 2.2.9 Lipid profile determinations

Serum triglyceride (TG) was determined using the method described by Fossati and Prencipe [25]. A 10 µl of serum was put in a dried test tube labelled as test, 10 µl of standard triglyceride was put into a test tube labelled standard, and 10 µl of distilled water was put into a test tube labelled blank. Then, 1000 µl reagent solution was added to contain in each test tube, mixed and incubated at 37°C for 5 minutes. The absorbance of the standard and test were read at 500 nm against the blank in a spectrophotometer.

Serum total cholesterol (TC) was determined using the method described by Roeschlau et al. [26]. The same procedure was applied in the determination of total cholesterol except the standard used was cholesterol standard. High density lipoprotein cholesterol (HDL-C) was measured following Lopes-Virella et al. [27] method. A 200 µl of serum was mixed with 500 µl of phosphotungastic (0.55 mmol/L) and Magnesium Chloride (25 mmol/L) and stand for 10 minutes at 25°C then centrifuged at 4000 rpm. About 100 µl of supernatant obtained was put in a dried test tube labelled as test, 100 µl of standard HDL-C was put into a test tube labelled standard, and 100 µl of distilled water was put into a test tube labelled blank. Then, 1000 µl reagent solution was added to contain in each test tube, mixed and incubated at 37°C for 5 minutes. The absorbance of the standard and test were read at 500 nm against the blank in a spectrophotometer. LDL-cholesterol and VLDL-cholesterol (VLDL-C) were determined by the

formula described by Friedewald et al. [28] as follows: LDL-cholesterol concentration (mg dLG1) =  $[TC-(HDL-C + Triglycerides / 5)]$  and VLDL-cholesterol concentration (mg dLG1) =  $[Triglycerides / 5]$ .

### 2.3 Statistical Analysis

Data from the experiments were expressed as mean  $\pm$  standard deviation (SD). Means were analyzed by ANOVA using SPSS software version 23. Significant difference was accepted at  $p < .05$ .

## 3. RESULTS

### 3.1 Yield of Methanol Extracts of Okra Fruit Varieties

The extract yield of each Okra fruit varieties after extraction with methanol (80%) is presented in

Table 1. From the result of the study, *NHB-AI-B* and *Yar kolon* had the highest yields: 58.03 g (22.85 %) and 48.24 g (17.11%) respectively.

### 3.2 Qualitative and Quantitative Phytochemicals of Methanol Extracts of Okra Fruit Varieties

Qualitative phytochemicals of methanol fruit extracts of Okra varieties is shown in Table 2 while quantitative is presented in Table 3. The study identified the presence of phytochemicals like saponins, tannins, flavonoids, and phenols in all the varieties. The quantitative phytochemical showed varied amount of the phytochemicals which differs among varieties. *Yar Kolon* variety is highest in phenolics and flavonoids (4.066 mg/g Gallic acid and  $6.7 \times 10^{-3}$  mg/g Quercetin), whereas *LD-88* is highest in saponins (164.0 mg/g Diosgenin) and tannins is highest in *Clemson spinless* (3.3 mg/g).

**Table 1. Extract yield of okra fruit varieties following soxhlet extraction with methanol (80%)**

| Sample           | Weight (g) | Yield | % Yield |
|------------------|------------|-------|---------|
| Clemson Spinless | 240.00     | 38.00 | 15.83   |
| LD-88            | 255.00     | 29.00 | 11.37   |
| NHB-AI-B         | 254.00     | 58.03 | 22.85   |
| NHAE-47-4        | 285.00     | 40.36 | 14.04   |
| Yar Kolon        | 282.00     | 48.24 | 17.11   |

**Table 2. Phytochemical profile of methanol extracts of okra fruit varieties**

| Samples          | Saponins | Tannins | Flavonoids | Alkaloids | Steriods | Glycosides | Phenolics |
|------------------|----------|---------|------------|-----------|----------|------------|-----------|
| Clemson Spinless | ++       | ++      | ++         | -         | +        | ++         | +         |
| LD-88            | ++       | ++      | +          | -         | -        | +++        | ++        |
| NHB-AI-B         | ++       | ++      | ++         | -         | -        | ++         | ++        |
| NHAE-47-4        | ++       | ++      | -          | -         | -        | +          | +         |
| Yar Kolon        | +        | ++      | ++         | +         | -        | +          | +++       |

NB: + = Mild, ++ = Moderate, +++ = highly present -: Absent

**Table 3. Phytochemical content of methanol extracts of okra fruit varieties**

| Samples          | Phytochemicals            |                            |                             |                              |
|------------------|---------------------------|----------------------------|-----------------------------|------------------------------|
|                  | Saponins (mg/g diosgenin) | Tannins (mg/g tannic acid) | Flavonoids (mg/g quercetin) | Phenolics (mg/g gallic acid) |
| Clemson Spinless | 132.8                     | 3.3                        | $4.1 \times 10^{-3}$        | 1.685                        |
| LD-88            | 164.0                     | 2.1                        | $1.7 \times 10^{-3}$        | 3.432                        |
| NHB-AI-B         | 151.1                     | 2.3                        | $1.7 \times 10^{-3}$        | 3.475                        |
| NHAE-47-4        | 156.6                     | 2.4                        | $5.2 \times 10^{-3}$        | 2.142                        |
| Yar Kolon        | 115.1                     | 2.4                        | $6.7 \times 10^{-3}$        | 4.066                        |

**Table 4. Proximate analysis of methanol extracts of okra fruit varieties**

| Sample           | Moisture (%) | Ash (%)     | Fibre (%)   |
|------------------|--------------|-------------|-------------|
| Clemson Spinless | 48.72 ±0.38  | 8.48 ±0.27  | 11.83 ±0.13 |
| LD-88            | 63.47 ±0.14  | 8.76 ±0.47  | 8.62 ±0.23  |
| NHB-AI-B         | 58.34 ±0.00  | 9.68 ±0.00  | 12.51 ±0.00 |
| NHAE-47-4        | 45.80 ±0.21  | 8.10 ±0.06  | 10.27 ±0.54 |
| Yar Kolon        | 61.17±0.00   | 10.33 ±0.30 | 14.74 ±0.17 |

Values are Mean ± SD of 3 determinations

### 3.3 Proximate Analysis of Methanol Extracts of Okra Fruit Varieties

Table 4 shows the result of proximate analysis like crude fibre, ash and moisture content of the varieties of Okra fruit extracts. A significant amount of all the parameters assayed is recorded from all the okra fruit varieties, however, *Yar kolon* variety is highest in fibre (14.74 ±0.17%) followed by *NHB-AI-B* which has 12.51±0.00%.

### 3.4 Changes in Lipid Profile of Rats Fed High-Fat Diet Pre-Extract Administration

The results of changes in lipid profile of rats fed high-fat diet and those fed basal diet for a period of 35 days are presented in Table 5. A significant increase ( $p < .05$ ) in the level of triglyceride (TG) was recorded by rats fed high-fat diet compared with those fed basal diet. Elevation in serum levels of total cholesterol and LDL-cholesterol were also recorded from the same rats fed high-fat diet. In addition, the study recorded some changes in high density lipoprotein cholesterol levels in same rats fed high-fat diet compared with those fed basal diet.

### 3.5 Effect of Okra Fruit Varieties on Lipid Profile of Rats Fed High-Fat Diet

The results of lipid profile of normal control, hyperlipidemic untreated and hyperlipidemic treated rats with extracts of Okra fruits varieties are presented in Table 6. The result showed methanol extracts of *NHB-AI-B* and *Yar Kolon* Okra fruit varieties able to cause positive changes in the levels of lipid profile determined in the treated rats. Triglyceride levels were lowered from rats received Okra fruit extracts compared to the untreated rats. Extract of *Yar Kolon* Okra fruit variety showed highest percentage TG reduction (Fig. 1) as compared to *NHB-AI-B* variety. The study also noticed that cholesterol levels were lowered in rats treated

with Okra extracts of *NHB-AI-B* and *Yar Kolon* varieties. The reduction in cholesterol level by *Yar Kolon* Okra fruit variety was significant different ( $P < .05$ ) compared to the *NHB-AI-B* variety.

Following extracts administration to various rats' groups, LDL-cholesterol and VLDL-cholesterol levels were observed to have been reduced compared with the value from the untreated rats. Also rats treated with methanol extracts of *NHB-AI-B* and *Yar Kolon* Okra fruit varieties had a slight elevation of HDL-cholesterol while untreated rats recorded reduction in HDL-C. The percentage difference between values of lipid profile before and after treatment with extracts is shown Fig. 1. The result showed extracts and standard drug reduced lipid profile in a varied degrees where rats received *Yar kolon* variety had the highest percentage reduction of lipid profile.

### 3.6 Effect of Okra Fruit Varieties on Water and Feed Intake of Rats Fed High-Fat Diet

The water intake of all rats in the various groups was in the range of 30.00±0.01 ml/day/rat before the extracts administration. During extracts administration, the water intake were elevated at the average of 50.00±0.20 ml/day/rat. The study recorded a little or no significant changes in the levels of feed intake (18.39±0.12g/day/rat) by rats from all the various groups pre/post-administration of the methanol extracts of the two okra fruit varieties.

### 3.7 Effect of Okra Fruit Varieties on Body Weight of Rats Fed High-Fat Diet

The change in body weight of rats during experimental study is presented in Table 7. The results showed body weight of rats at day 0 (baseline), after 35 days feeding with high-fat diet, and the weight change following 21 days treatment with Okra extracts. The result showed

rapid increase in weight by rats fed high-fats diet pre-extract treatment compare with those that fed basal diet. When treated with extracts particularly the *Yar kolon* variety, the study observed insignificant changes in the weight of those rats which are not significantly different ( $P>.05$ ) from their weight after the 35 days feeding with high-fat diet.

#### 4. DISCUSSION

Hyperlipidemia is a major factor associated with the development of atherosclerosis and subsequent cardiovascular diseases and stroke [1]. It is a major health problem to both the affluent and non-affluent populations of every societies [29]. To date, stable treatment of hyperlipidemia is yet to be discovered, and prolong usage of available synthetic drugs are accompanied with problems like restricted efficacies and advanced side effects [2]. Research toward finding safer, inexpensive, and effective agents to combat the disorder has been encouraged [30].

In this study, antihyperlipidemic efficiency of two varieties (NHB-AI-B and *Yar kolon*) of Okra fruits commonly consumed in Bauchi State, Nigeria was investigated on rats fed high-fat diet. After feeding rats with high-fat diet for a period of 35 days, the study observed elevation in serum lipid components like triglyceride, cholesterol, low density lipoprotein-cholesterol, very low density lipoprotein-cholesterol, and high density lipoprotein-cholesterol level. This is in-line with the findings from a similar study conducted by Kumar et al. [31] where rats were fed high-fat diet for same period. When rats were treated with different doses of methanol extracts from *NHB-AI-B* and *Yar kolon* Okra fruit varieties and standard drug for 21 days period, the altered lipid- components were reversed toward normalcy by a varied degrees placing *Yar kolon* Okra fruit variety as promising candidate. This may be attributable to its high fibre and phytochemicals content, attesting to the fact that Okra fibre plays a major role in antihyperlipidemic activity [10,12]. With this finding, one may suggest the possibility that antihyperlipidemic efficiency of Okra varies with varieties.

The alternations in the measured lipid components; triglyceride, cholesterol, low density lipoproteincholesterol and high density lipoprotein-cholesterol in rats fed high-fat diet in the study agrees with the report from a study

conducted by Karam et al. [22], where it was found that diet rich in fats could induces hyperlipidermia in animals' when consumed for long-time period. The ability for *Abelmoschus esculentus* variety; *Yar kolon* and NHB-AI-B fruit extracts to lower the elevated triglyceride, cholesterol, low density lipoprotein-cholesterol and very low density lipoprotein-cholesterol levels in rats fed high-fat diet has been experimentally demonstrated with plant extracts as reported by several studies [10,12]. In one of the earlier study, high intake of Okra products was found to reduce risk associated with a number of chronic diseases like atherosclerosis and cancer [9].

The high levels of triglycerides from rats fed high-fat diet in the study is an indication of fats deposition which resulted in the increase accumulation of the lipid. Literature survey shown that intake of high-fat diet promotes deposition of fats in tissues like the adipose tissue which may alters lipid metabolism [32,33]. Declined in serum triglyceride level after rats were treated with extracts from *Yar kolon* and NHB-AI-B Okra fruit varieties is an indication that they contain components that are extractable with methanol and likely retaining their antihypertriglyceridemic properties. The findings of this study are in tandem with the antihypertriglyceridemic activity of Okra extract reported in a study conducted by Esan et al. [34].

The reduction in levels of cholesterol by the two varieties of Okra fruit extracts in the study could in part be probably attributed to their fibre content. It was reported that Okra contain fibre which plays vital role in supressing lipid fractions like cholesterol leading to the amelioration of risk associated with the development of cardiovascular disease [14]. The varied degrees in reduction of cholesterol by the two selected varieties of Okra extracts; *Yar kolon* and NHB-AI-B fruit could be a reflection of their fibre content. *Yar kolon* had the highest fibre content and this might explain why it perform better with about 54% cholesterol reduction. Antihyper cholesterolemic effect of Okra fruit extract had been reported in a recent study by Djamil et al. [35].

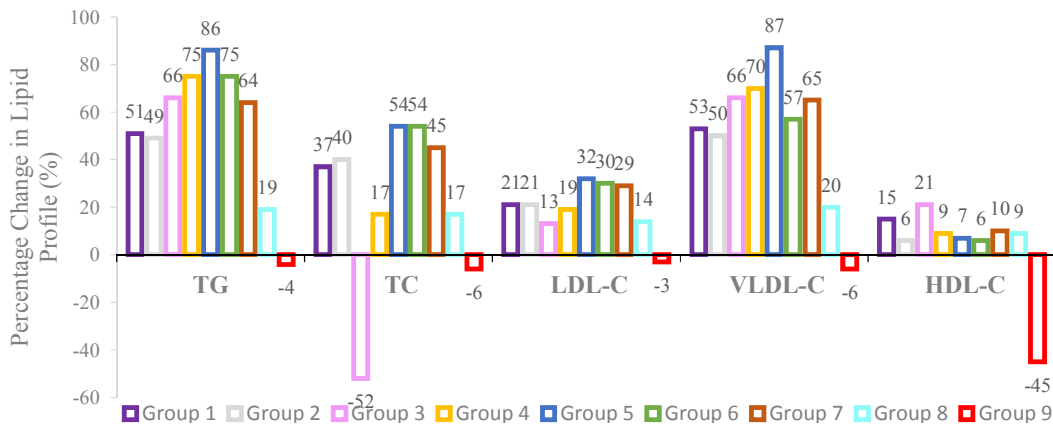
The hypothetic mechanisms of antihypercholesterolemic effect of Okra plant extract have been postulated by some scholars. A report by Esan et al. [34] indicates that Okra extract altered HMG-CoA activity thereby

ameliorating hypercholesterolemia. Another researcher pointed out that reduction in cholesterol levels by Okra products resulted from a decreased LDL-C [36]. Decreased LDL-C levels of rats fed high-fat diet after treatment with extracts of Okra fruit varieties particularly the *Yar kolon* variety might have in part contributed to their cholesterol reductions in a similar manner as suggested by Poorva and Sunita [37]. A reduction in serum LDL cholesterol levels by extract from Okra seeds have been reported in a study conducted with hypercholesterolemic mice [34].

High density lipoprotein (HDL) was reported to aid in the movement of cholesterol from arterial walls to the liver for degradation. Retardation in the process may increase risk associated with the developing of atherosclerosis [37]. In the present study, decreased levels of HDL-C recorded in rats fed high-fat diet pre-extract treatment is an indication of failure in the cholesterol translocation process. Treating rats fed high-fat diet with different doses of *Yar kolon* and *NHB-AI-B* fruit extracts, the HDL-C level was slightly elevated. Increased level of HDL-C after treatment have been reported by several studies where it was found to correlate with a decreased in cholesterol level due to either increasing cholesterol excretion or decreasing cholesterol absorption alongside LDL-C reduction [38,39].

Relating this scenario to the present study, it could be stated that the elevated HDL-C in rats that received Okra fruit varieties might have contributed in slowing or eliminating the progression of hyperlipidemia and its related disorders as reported by several earlier studies [40,41]

Literature survey shows that the growth rate of animals is influenced by species, individuals, sex, age, feeding, and diet consumed [42]. The present study had observed changes in both growth and body weights gain throughout the experimental period by rats fed high-fat diet and those that fed basal diet. Study have shown that weight gain is caused by an increase in the fat deposition in the adipose tissue, especially those under the skin and the abdominal cavity [33]. Continuous feeding rats with high-fat diet might have contributed to their increasing body weight. However, reduction in body weight gain by rats received extracts particularly the *Yar kolon* Okra variety might be due to loss of body lipid as evidence by the high percentage reduction in their lipid profile. Going by this, one may stipulate that increased weight of rats received *NHB-AI-B* variety may be a reflection of their low lipid profile reduction. However, Okra powder was reported to have promoted body weight gain in high-fat diet-induced diabetic rats [43].



**Fig. 1. Percentage difference in lipid profile before and after treatment of rats fed high-fat diet with extracts of okra fruit varieties**

Group 1, 2 and 3 = Hyperlipidemic rats + *NHB-AI-B* (250, 500 and 750mg/kg Body wt.).  
 Group 4, 5, and 6 = Hyperlipidemic rats + *Yar Kolon* (250, 500 and 750mg/kg Body wt.)  
 Group 7 = Positive Control, Group 8 = Normal Control, and Group 9 = Negative Control



**Table 5. Changes in lipid profile of rats fed high-fat diet pre-extract administration**

|         | Animal Grouping      |                      |                     |                      |                      |                      |                     |                   |                      |
|---------|----------------------|----------------------|---------------------|----------------------|----------------------|----------------------|---------------------|-------------------|----------------------|
|         | Group 1              | Group 2              | Group 3             | Group 4              | Group 5              | Group 6              | Group 7             | Group 8           | Group 9              |
| TG      | 180.9                | 221.5                | 179.2               | 187.0                | 233.8                | 223.5                | 201.7               | 124.9             | 254.4                |
| (mg/dL) | ±9.5 <sup>b</sup>    | ±55.1 <sup>bc</sup>  | ±13.4 <sup>b</sup>  | ±10.6 <sup>bc</sup>  | ±39.7 <sup>bcd</sup> | ±27.2 <sup>bc</sup>  | ±10.5 <sup>bc</sup> | ±3.5 <sup>a</sup> | ±23.7 <sup>bcd</sup> |
| TC      | 337.7                | 391.3                | 285.6               | 400.3                | 424.9                | 394.1                | 344.4               | 157.2             | 364.7                |
| (mg/dL) | ±39.9 <sup>bcd</sup> | ±81.7 <sup>bcd</sup> | ±59.3 <sup>bc</sup> | ±36.5 <sup>bcd</sup> | ±56.8 <sup>bcd</sup> | ±45.4 <sup>bcd</sup> | ±39.0 <sup>cd</sup> | ±2.8 <sup>a</sup> | ±26.7 <sup>bcd</sup> |
| LDL-C   | 19.8                 | 28.2                 | 16.8                | 26.0                 | 25.6                 | 23.3                 | 17.8                | 7.0               | 31.0                 |
| (mg/dL) | ±3.7 <sup>b</sup>    | ±3.8 <sup>b</sup>    | ±11.4 <sup>b</sup>  | ±6.8 <sup>b</sup>    | ±16.5 <sup>b</sup>   | ±10.6 <sup>b</sup>   | ±5.4 <sup>b</sup>   | ±1.6 <sup>a</sup> | ±6.7 <sup>b</sup>    |
| VLDL-C  | 36.2                 | 44.3                 | 35.8                | 37.4                 | 46.8                 | 44.7                 | 40.3                | 25.8              | 50.9                 |
| (mg/dL) | ±1.9 <sup>b</sup>    | ±11.0 <sup>bc</sup>  | ±2.7 <sup>b</sup>   | ±2.1 <sup>bc</sup>   | ±7.9 <sup>bcd</sup>  | ±5.4 <sup>bcd</sup>  | ±2.1 <sup>bc</sup>  | ±0.7 <sup>a</sup> | ±4.7 <sup>bcd</sup>  |
| HDL-C   | 57.8                 | 48.7                 | 42.4                | 73.1                 | 43.2                 | 44.3                 | 44.6                | 43.1              | 55.3                 |
| (mg/dL) | ±33.2 <sup>bc</sup>  | ±8.1 <sup>ab</sup>   | ±4.3 <sup>a</sup>   | ±28.1 <sup>bcd</sup> | ±14.3 <sup>a</sup>   | ±8.8 <sup>a</sup>    | ±6.4 <sup>a</sup>   | ±4.5 <sup>a</sup> | ±4.5 <sup>bc</sup>   |

Values are Mean ± SD of 5 determinations. Values with different superscript across the rows are significantly different (P<.05)

Group 1, 2 and 3 =Hyperlipidemic rats + NHB-AI-B (250, 500 and 750mg/kg Body wt.).

Group 4, 5, and 6 = Hyperlipidemic rats + Yar Kolon (250, 500 and 750mg/kg Body wt.)

Group 7 = Positive Control, Group 8 = Normal Control, and Group 9 = Negative Control

**Table 6. Effect of okra fruit varieties (NHB-AI-B and Yar kolon) on lipid profile in rats fed high-fat diet**

|         | Animal Grouping      |                      |                      |                      |                     |                    |                      |                     |                      |
|---------|----------------------|----------------------|----------------------|----------------------|---------------------|--------------------|----------------------|---------------------|----------------------|
|         | Group 1              | Group 2              | Group 3              | Group 4              | Group 5             | Group 6            | Group 7              | Group 8             | Group 9              |
| TG      | 89.5                 | 113.8                | 61.2                 | 46.3                 | 32.5                | 56.4               | 73.3                 | 101.7               | 266.0                |
| (mg/dL) | ±4.9 <sup>bcd</sup>  | ±2.7 <sup>bcd</sup>  | ±6.9 <sup>bc</sup>   | ±7.2 <sup>b</sup>    | ±9.5 <sup>a</sup>   | ±9.9 <sup>b</sup>  | ±10.2 <sup>bcd</sup> | ±8.6 <sup>bcd</sup> | ±15.5 <sup>bcd</sup> |
| TC      | 213.1                | 235.8                | 434.9                | 322.0                | 194.9               | 183.6              | 183.4                | 131.5               | 387.3                |
| (mg/dL) | ±29.0 <sup>bcd</sup> | ±36.6 <sup>bcd</sup> | ±80.5 <sup>bcd</sup> | ±80.2 <sup>bcd</sup> | ±25.6 <sup>bc</sup> | ±10.2 <sup>b</sup> | ±11.8 <sup>b</sup>   | ±7.1 <sup>a</sup>   | ±27.5 <sup>bcd</sup> |
| LDL-C   | 15.2                 | 22.7                 | 13.1                 | 21.1                 | 17.1                | 16.0               | 12.6                 | 6.1                 | 32.6                 |
| (mg/dL) | ±1.1 <sup>ab</sup>   | ±7.6 <sup>ab</sup>   | ±15.3 <sup>ab</sup>  | ±1.4 <sup>ab</sup>   | ±6.9 <sup>ab</sup>  | ±3.8 <sup>ab</sup> | ±4.1 <sup>ab</sup>   | ±2.4 <sup>a</sup>   | ±6.1 <sup>bc</sup>   |
| VLDL-C  | 17.9                 | 22.2                 | 12.2                 | 11.3                 | 6.5                 | 11.3               | 14.5                 | 20.7                | 53.2                 |
| (mg/dL) | ±1.0 <sup>bcd</sup>  | ±0.5 <sup>bcd</sup>  | ±1.4 <sup>b</sup>    | ±1.4 <sup>b</sup>    | ±1.9 <sup>a</sup>   | ±2.0 <sup>b</sup>  | ±2.0 <sup>bc</sup>   | ±1.7 <sup>bcd</sup> | ±3.1 <sup>bcd</sup>  |
| HDL-C   | 67.5                 | 51.5                 | 53.2                 | 80.2                 | 46.9                | 47.2               | 48.1                 | 47.3                | 38.2                 |
| (mg/dL) | ±8.9 <sup>bc</sup>   | ±6.7 <sup>bc</sup>   | ±2.3 <sup>bc</sup>   | ±4.3 <sup>bcd</sup>  | ±4.4 <sup>b</sup>   | ±3.8 <sup>b</sup>  | ±3.5 <sup>b</sup>    | ±5.3 <sup>b</sup>   | ±2.1 <sup>a</sup>    |

Values are Mean ± SD of 5 determinations. Values with different superscript across the rows are significantly different (P<.05)

Group 1, 2 and 3 = Hyperlipidemic rats + NHB-AI-B (250, 500 and 750mg/kg Body wt.).

Group 4, 5, and 6 = Hyperlipidemic rats + Yar Kolon (250, 500 and 750mg/kg Body wt.)

Group 7=PositiveControl, Group 8 = Normal Control, and Group 9 = Negative Control

**Table 7. Effects of okra fruit varieties (NHB-AI-B and Yar kolon) on body weights of rats fed high-fat diet**

|                      | Animal Grouping             |                              |                              |                             |                             |                             |                             |                              |                              |
|----------------------|-----------------------------|------------------------------|------------------------------|-----------------------------|-----------------------------|-----------------------------|-----------------------------|------------------------------|------------------------------|
|                      | Group 1                     | Group 2                      | Group 3                      | Group 4                     | Group 5                     | Group 6                     | Group 7                     | Group 8                      | Group 9                      |
| Initial Body wt. (g) | 101.1<br>±16.7 <sup>a</sup> | 135.1<br>±33.0 <sup>a</sup>  | 145.9<br>±50.3 <sup>a</sup>  | 127.8<br>±62.5 <sup>a</sup> | 119.5<br>±18.7 <sup>a</sup> | 126.7<br>±30.0 <sup>a</sup> | 104.0<br>±6.2 <sup>a</sup>  | 100.6<br>±29.9 <sup>a</sup>  | 116.6<br>±16.7 <sup>a</sup>  |
| Median Body wt. (g)  | 139.5<br>±15.7 <sup>b</sup> | 147.3<br>±29.2 <sup>b</sup>  | 161.0<br>±49.7 <sup>b</sup>  | 198.4<br>±20.3 <sup>b</sup> | 157.3<br>±24.3 <sup>b</sup> | 161.3<br>±25.5 <sup>b</sup> | 113.5<br>±13.0 <sup>a</sup> | 116.1<br>±23.6 <sup>b</sup>  | 141.6<br>±11.7 <sup>b</sup>  |
| Final Body wt.(g)    | 162.8 <sup>bc</sup><br>±4.9 | 206.6<br>±17.0 <sup>bc</sup> | 211.6<br>±42.0 <sup>bc</sup> | 172.2<br>±27.5 <sup>b</sup> | 165.9<br>±13.1 <sup>b</sup> | 158.2<br>±18.4 <sup>b</sup> | 143.8<br>±17.2 <sup>b</sup> | 187.6<br>±33.6 <sup>bc</sup> | 156.2<br>±15.3 <sup>bc</sup> |

Values are Mean ± SD of 5 determinations. Values with different superscript along the columns are significantly different (P<.05)

Initial body weight = Baseline weight at day 0, Median body weight =Weight after feeding high-fat days for 35 days period, Final body weight = Weight change following extracts administration. Group 1, 2 and 3 = Hyperlipidemic rats + NHB-AI-B (250, 500 and 750mg/kg Body wt.), Group 4, 5, and 6 = Hyperlipidemic rats + Yar Kolon (250, 500 and 750mg/kg Body wt.), Group 7 = Positive Control, Group 8 = Normal Control, and Group 9 = Negative Control

## 5. CONCLUSION

The study showed the five Okra fruit varieties possess same chemical compounds but differs in concentrations among varieties. The extracts of Okra fruits (*NHB-AI-B* and *Yar kolon*) varieties exert significant antihyperlipidemic effect but in varied degrees suggesting variations in their efficiency. This, therefore calls for further study to compare more Okra varieties to determine which one is the most potent and its active agent.

## DISCLAIMER

The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

## ETHICAL APPROVAL

Authors hereby declare that the experiment was performance in accordance with "Principles of laboratory animal care" (NIH publication No: 85-23, revised 1985) and ethical guidelines for investigation of experimental pain in conscious animals (Zimmermann, 1983) were observed.

## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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