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Effect of Steamed Cake Substituted with Okra (Abelmoschus esculentus) Flour on Total Cholesterol and LDL Levels in Hypercholesterolemic Rats

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#### Abstract

Hypercholesterolemia is a condition characterized by elevated total cholesterol levels above 200 mg/dL, which serves as a primary risk factor for cardiovascular disease. Efforts to control cholesterol levels can be achieved through the consumption of functional foods high in fiber, one of which is steamed cake made with okra flour substitution. This product contains 5.34 grams of fiber per 100 grams, thereby meeting the claim as a fiber source. This study aims to analyze the effect of okra cake administration on total cholesterol and LDL levels in male Wistar rats with a hypercholesterolemia model. The study design used was a true experimental design with a pretest-posttest control group. A total of 18 rats were divided into three groups: negative control (K-), positive control (K+), and treatment (P), each consisting of six rats. Hypercholesterolemia induction was performed for 21 days using a high-fat diet (HFD). The treatment group was then given okra cake at a dose of 8.4 g/rat/day for 14 days. The results showed that total cholesterol levels in the treatment group decreased significantly (p=0.018), while in the positive control group, there was a significant increase (p=0.000). For LDL levels, no significant differences were found in each group before and after treatment (p>0.05), but there were significant differences between the K+ and P groups (p=0.027), as well as significant differences in cholesterol levels between the K+ and P groups (p=0.016). Conclusion: Steamed cake with okra flour substitution has the potential to significantly reduce total cholesterol and LDL levels in hypercholesterolemia rats.

Keywords: Hypercholesterolemia; Okra Cake; Total Cholesterol; LDL; Functional Food

#### INTRODUCTION

Hypercholesterolemia is one of the major risk factors for cardiovascular disease, which remains the leading cause of death globally. Elevated levels of total cholesterol and low-density lipoprotein (LDL) in the blood play a crucial role in the formation of atherosclerotic plaques, which can trigger cardiovascular disorders. This condition is defined as total cholesterol levels exceeding 200 mg/dL. The American Heart Association (AHA) hypercholesterolemia as a condition in which both

total and LDL cholesterol levels exceed normal values.

Data from the Survei Kesehatan Indonesia (SKI) in 2023 showed that the prevalence of high total cholesterol in the population aged over 15 years reached a significant level, with the 55-64 age group having the highest prevalence at 21.2%. Meanwhile, high LDL levels were most prevalent in the 65-74 age range at 14.5%. In general, the prevalence of hypercholesterolemia increases with age, which was 14.7% in the 25-34 age group and increased to 29.2% in the 55-64 age group (SKI 2023).

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One of the non-pharmacological approaches to treating hypercholesterolemia is the consumption of functional foods made from okra. Okra is known to have health benefits (Dantas, et al., 2021). Okra is known to have fiber of 3.2 gr/100 g. Okra fiber is higher than broccoli (2.8 gr/100 g) and cabbage (2 gr/100 g). Soluble dietary fibers are a type of fiber that can reduce cholesterol, including modulating the gut microbiome. Another study mentioned that increasing the rate of bile acid excretion leads to a decrease in LDL levels (Bakr and Farag 2023).

Processing okra into flour has been shown to increase its fiber content and bioactive compounds. Okra flour contains about 14.21 grams of fiber and 202 ppm flavonoids per 100 grams, and has advantages in shelf life (Febriyatna and Widiyawati 2018). One of the product innovations based on okra flour is realized in the form of steamed sponge cake. Steamed cake with okra flour substitution as an alternative high-fiber snack. This product contains 5.34 grams of fiber per 100 grams, so it can meet claims as a source of fiber and meet about 17.8% of daily fiber needs (Kumalasari 2020). Several studies have shown that fiber consumption contributes to lowering total and LDL cholesterol levels (Sinulingga 2020: Fairudz and Nisa 2015).

However, despite the growing evidence on okra's potential lipid-lowering effects, studies on the use of okra flour as a functional ingredient in commonly consumed foods particularly steamed sponge cake remain limited. Most previous research has focused on okra extracts or isolated bioactive compounds rather than its food-based applications. Furthermore, the effect of okra flour-based foods on lipid profiles, especially in experimental animal models of hypercholesterolemia, has not been thoroughly investigated. Therefore, this study aimed to analyze the effect of okra-based steamed sponge cake on total and LDL cholesterol levels in male Wistar rats induced with a hypercholesterolemia model.

#### **METHOD**

This study is a pure experimental study (True Experimental) with a pretest-posttest with control group design. The experimental animals used were 18 male Wistar strain rats, divided into three groups, namely the negative control group (K-), the positive control group (K+), and the treatment group (P), each consisting of six rats. The allocation of rats into groups was done randomly using simple randomization with the help of a random number generator, while data analysis was conducted in a single-blind manner to minimize potential bias during assessment. The K+ and P groups were induced with a high-fat diet (HFD) consisting of 2 grams of beef fat, 1 gram of quail egg yolk, and 2 grams of butter, administered via gavage for 21 days. After the induction period, the P group was given an intervention in the form of steamed cake with 8.4 grams of okra flour substitution per rat per day for 14 days. This research has obtained ethical approval from Polytechnic with number Jember State 1273/PL17.4/PG/2023.

The parameters observed were total cholesterol and LDL. Total and LDL cholesterol examinations were conducted after adaptation (T0), after induction (Pretest), and after intervention (Posttest). Total and LDL cholesterol examinations used rat blood samples. Total and LDL cholesterol data were statistically analyzed using IBM SPSS Statistics 26 software.

#### RESULT AND DISCUSSION

The adaptation period of rats in this study lasted for 10 days. Total cholesterol and LDL cholesterol of rats during the adaptation period (T0) (Table 1).

Table 1. Total Cholesterol (TC) and LDL on adaptation period (T0) between groups

Variable	Groups	n	Mean		
			$(mg/dl\pm SD)$	P-Value	
TC	K-	6	$51.83 \pm 6.21$		
	K+	6	$51.67 \pm 8.23$	0.987	
	P	6	$51.17 \pm 8.06$		
LDL	K-	6	$16.33 \pm 5.57$		
	K+	6	$18.17 \pm 5.38$	0.858	
	P	6	$17.00 \pm 6.36$		

Table 2 presents the baseline (pretest) condition of total cholesterol (TC) and LDL levels in rats during the adaptation period, prior to the intervention. During this period, the rats were given only standard rat bio feed and drinking water ad libitum. Statistical analysis showed no significant differences in total cholesterol or LDL levels among the negative control (K-), positive control (K+), and treatment (P) groups (p > 0.05). These findings indicate that all groups had relatively homogeneous baseline conditions, ensuring that any subsequent changes in cholesterol levels could be attributed to the intervention rather than to initial physiological differences among the groups.

Table 2. Total Cholesterol (TC) and LDL pretestposttest in each groups

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Variable	Groups	n	Mean (m	P- value	
			Pretest	Posttest	
TC	K-	6	$59.50 \pm 4.93$	$63.83 \pm 6.43$	0.366
	K+	6	$80.17 \pm 4.79$	$85.33 \pm 5.43$	0.000*
	P	6	$75.67 \pm 4.76$	$69.17 \pm 5.38$	0.018*
LDL	K-	6	$26.33 \pm 3.01$	$29.33 \pm 3.67$	0.226
	K+	6	$31.00\pm1.90$	$34.00 \pm 4.86$	0.168
	P	6	$30.33\pm1.21$	$25.83 \pm 5.04$	0.074

Note: (\*) significantly different

The pretest-posttest total cholesterol levels in the K+ group showed a significant increase (p=0.000). This condition was due to the HFD of beef tallow, quail egg yolk, and butter.

The treatment group (P) in the pre-post test showed a significant decrease in total cholesterol levels (p = 0.018) (Table 2). This reduction was attributed to the substitution of steamed sponge cake with okra flour, which is rich in bioactive components. Okra flour contains approximately 14.21% dietary fiber and 202 ppm flavonoids, both of which contribute to lipid regulation. Previous studies have reported that okra flour can effectively lower total cholesterol levels (Febriyatna and Widiyawati 2017). At the molecular level, dietary fiber functions as an antihyperlipidemic agent by modulating several lipid metabolism pathways, including HMG-CoA reductase activity, LDL receptor expression, regulation, and the MAPK signaling pathway (Nie and Luo, 2021).

For LDL cholesterol levels, no statistically significant changes were observed among the three however, the treatment groups; group demonstrated a decreasing trend, from  $30.33 \pm 1.21$ mg/dl to 25.83  $\pm$  5.04 mg/dl (p = 0.074) (Table 2). Okra is recognized as a nutrient-dense natural food rich in fiber and phytochemicals, with potential nutraceutical benefits for maintaining lipid balance and overall health. Its bioactive compounds particularly fiber, flavonoids, and polysaccharides - play an essential role in reducing oxidative stress and improving lipid metabolism, thereby contributing to the prevention of chronic diseases (Elkhalifa et al., 2021; Nikpayam et al., 2021).

Table 3. Difference in Total Cholesterol (TC) and LDL pretest-posttest

Variable	Groups	n	Δ	P-value
TC	K-	6	4.33ª	
	K+	6	$5.16^{a}$	0.016*
	P	6	-6.5 <sup>b</sup>	
LDL	K-	6	3ª	
	K+	6	3 <sup>a</sup>	0.027*
	P	6	-4.5 <sup>b</sup>	

Note: (\*) significantly different

The results indicated a significant difference in total cholesterol and LDL levels between the treatment group (P) and the control groups (K- and K+). The treatment group exhibited a decrease of 6.5 mg/dL in total cholesterol and 4.5 mg/dL in LDL levels compared with the control groups. These findings are consistent with the study by Algita et al. (2020), which demonstrated that administration of okra extract at a dose of 100 mg/kg body weight significantly reduced LDL, triglyceride, and total cholesterol levels. Similarly Majd et al (2018), eported that okra supplementation effectively decreased LDL and total cholesterol concentrations in mice induced with a high-fat diet (HFD) and streptozotocin (STZ).

The hypocholesterolemic effect of okra is primarily attributed to its high pectin content, a soluble fiber known to inhibit fat absorption and modulate bile acid metabolism in the gastrointestinal tract. Through these mechanisms, okra helps reduce circulating cholesterol levels and prevent lipid accumulation in body tissues (Soma Das, *et al.*, 2019).

### **CONCLUSION**

Steamed cake with okra flour substitution was able to significantly reduce total cholesterol and LDL in hypercholesterolemic rats.

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