ISSN: 2536-7153 SJMLS

Sokoto Journal of Medical Laboratory Science 2024; 9(3): 308 - 315

SJMLS - 9(3) - 032

Effect of Aqueous Extract of Allium cepa (Onion) on Ethanol-Induced Dyslipidemia and Inflammation in Albino Rats

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https://dx.doi.org/10.4314/sokjmls.v9i3.32

Abstract

Alcohol consumption has increased over the years due to social activities. However, chronic alcohol consumption is a known risk factor for several diseases, including dyslipidemia. This study evaluated the effect of Allium cepa (onion) aqueous extract on ethanol-induced dyslipidemia and inflammation in Albino Rats. Twenty-five Rats, weighing 130-180g, were randomly divided into five groups of five. The rats were acclimatized for 14 days with free access to standard feed and water. Group I served as the negative control, while Group II was the positive control, receiving 50% ethanol at a dose of 0.5ml/100g. Groups III to V received 50% ethanol followed by oral administration of 200mg/kg, 400mg/kg, and 600mg/kg of Allium cepa extract, respectively, once daily for 30 days. The rats were then anesthetized with chloroform, and blood samples were collected to analyze C-reactive protein (CRP) and lipid profiles, including triglycerides (TG), total cholesterol (T. Chol), and HDL, with LDL calculated using Friedewald's equation. Data analysis was performed using SPSS version 24, with $p<0.05$ considered statistically significant. There was a significant decrease in CRP ($p = 0.000035$), T. Chol ($P=0.010$), and LDL ($P=0.038$) in the treatment groups compared to the positive control group, but there were no significant differences in HDL ($P=0.179$) and TG ($P=0.899$) across all groups. These findings suggest that Allium cepa extract may ameliorate ethanol-induced dyslipidemia and inflammation. Further studies are recommended.

Keywords: Alcohol consumption, dyslipidaemia, inflammation, Allium cepa, Anti-inflammatory effects, metabolic syndrome, cardiovascular diseases

Introduction

Recent years have witnessed a significant rise in alcohol consumption worldwide, a trend that has alarming public health implications. Excessive alcohol intake is closely linked to the development of numerous health disorders, including liver cirrhosis, cardiovascular diseases, pancreatitis, and various forms of cancer (WHO, 2018). Among these, alcohol-induced dyslipidaemia and inflammation are particularly concerning due to their roles in the progression of metabolic syndrome and chronic inflammatory conditions (Rehm *et al*., 2009; Gao and Bataller, 2011).

Excess alcohol consumption prompts the increased production of acetaldehyde and reactive oxygen species (ROS) during alcohol metabolism. These byproducts deplete glutathione (GSH), trigger ROS-mediated hepatocellular apoptosis (Koch *et al*., 2004). It also encourages the movement of nuclear factor erythroid 2-related factor 2 (Nrf2) into the nucleus and enhances its activity. The Nrf2 plays a role in regulating protective genes responsible for antioxidant enzymes like glutathione peroxidase (GPX), thereby aiding in the reduction of hepatic oxidative stress (Kono *et al*., 2000; Yan *et al*., 2010). Furthermore, ethanol triggers the activation of the hepatic nuclear factor kappa (NF-κB) pathway, leading to the subsequent release of inflammatory molecules, such as interleukin (IL)-6 and tumour necrosis factor (TNF)-alpha (Szuster-Ciesielska *et al*., 2013; Zima and Kalousová, 2015).

Dyslipidaemia which is characterized by abnormal levels of lipids in the blood, is a major

risk factor for cardiovascular diseases (CVDs) (Gylling *et al*., 2014). Chronic alcohol consumption disrupts lipid metabolism, leading to elevated triglycerides and low-density lipoprotein (LDL) cholesterol, and reduced high-density lipoprotein (HDL) cholesterol (Hagiwara *et al*., 2014). Additionally, ethanol intake triggers systemic inflammation, contributing to atherosclerosis and other inflammatory diseases (Bala *et al*., 2014).

In the quest for alternative therapies, medicinal herbs have gained attention for their potential to mitigate the adverse effects due to alcohol intake. *Allium cepa*, commonly known as onion, is one such herb that has been extensively studied for its medicinal properties. They contribute their unique taste to various dishes such as stews, roasts, soups, and salads, and are even prepared as a cooked vegetable (Marrelli *et al*., 2022). Onion extract is rich in flavonoids, particularly quercetin, which exhibit potent antioxidant, anti-inflammatory, and lipid-lowering effects (Griffiths *et al*., 2002). Research suggests that these bioactive compounds in onion can ameliorate ethanol-induced dyslipidaemia and inflammation, making it a promising natural remedy (Kim *et al*., 2000; Gorinstein *et al*., 2010). Hence, this study is necessary, as it seeks to provide insights into natural interventions that could complement existing treatments for alcohol-related health disorders. This study aims to evaluate the effect of aqueous extract of *Allium cepa* (onion) on ethanol-induced dyslipidaemia and inflammation in Albino Rats.

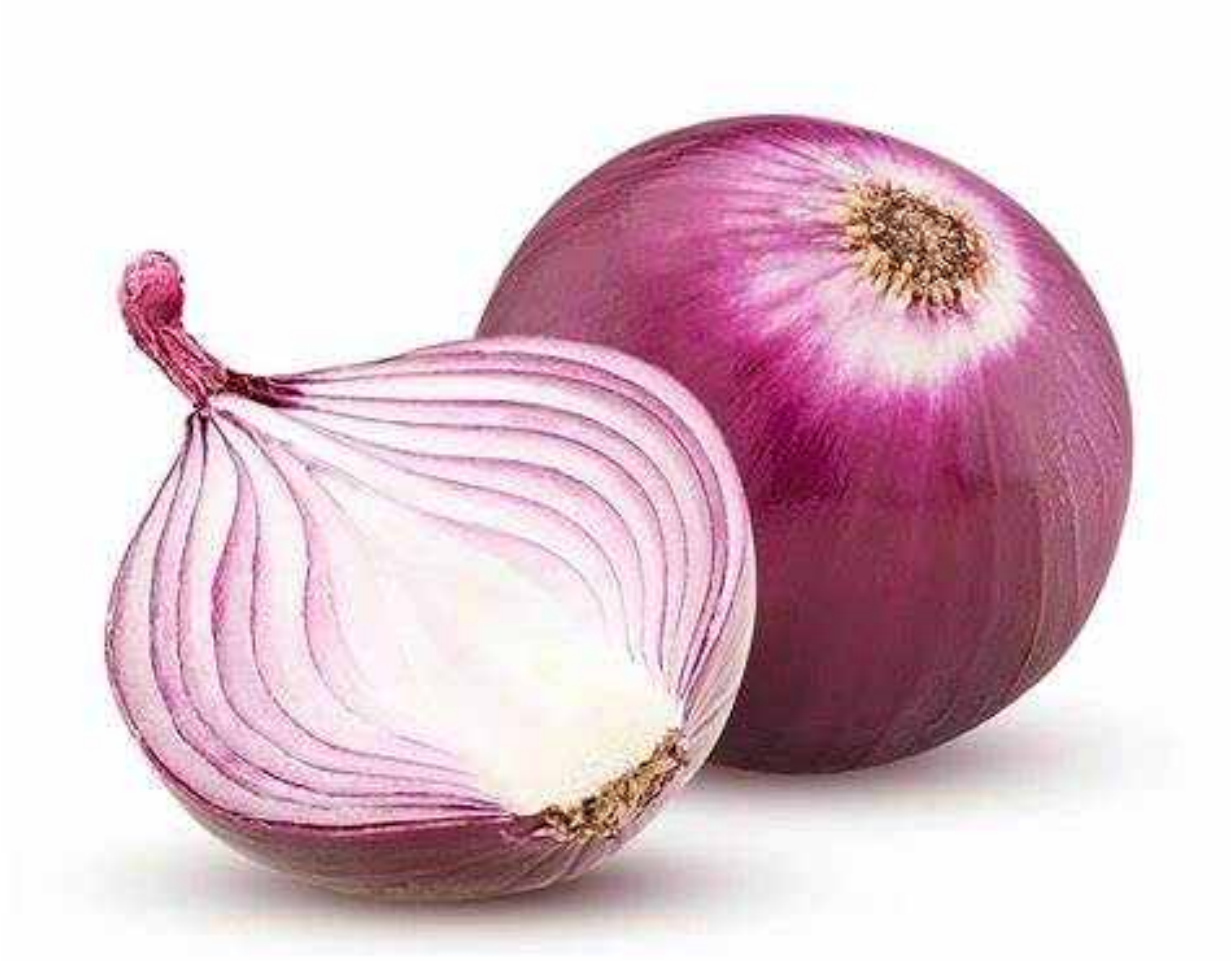

Figure 1: Bulbs of *Allium cepa* (Onion)

Materials and Methods

Experimental animals

For the study, twenty-five albino rats, each weighing between 150 and 200 grams, were randomly selected. These animals were sourced from the Department of Anatomy at the College of Medical Sciences, Rivers State University. They were transported in well-ventilated wire cages to the Animal House in the Department of Animal and Environmental Sciences at Rivers State University, Port Harcourt. The rats were maintained on a 12-hour light/dark cycle and given unlimited access to solid poultry chow and water. Prior to the start of the experiment, the rats were allowed a two-week acclimatization period.

Preparation of 50% percent ethanol

A stock solution of absolute ethanol was obtained from a chemical shop on Hospital Road in Port Harcourt, Nigeria. To prepare a 50% ethanol solution, 50 milliliters (50 mL) of absolute ethanol was measured into a mixing beaker, and then 50 milliliters (50 mL) of distilled water was added and gently mixed.

Preparation of onions extract

Fresh onion bulbs were purchased from the local Mile 3 market in Port Harcourt, Nigeria. Among the three local onion varieties identified at the National Institute of Horticultural Research (NIHORT) in Ibadan, the Kano Red variety was chosen due to its high antioxidant potential and pungency (Denton and Ojeifo, 1990). Then, sixty grams (60g) of the bulbs were carefully cleaned, blended, and homogenized in 100 mL of distilled water for one hour. The homogenate was then filtered three times using Whatman filter paper No. 1 to produce a crude extract with a concentration of 60g/100ml or 6000mg/ml. Additionally, concentrations of 400mg/ml and 200mg/ml of the extract were prepared from the crude extract using the appropriate dilution formula:

$C_1V_1 = C_2V_2$

Where C_1 = Concentration of crude extract

C_2 = Concentration of desired extract

V_1 = Volume of crude extract

V_2 = Volume of desired extract

The qualitative phytochemical analysis of the *Allium cepa* was also conducted.

Dose determination

The 50% ethanol solution was administered at a dosage of 0.5ml/100g of body weight, following the method of Lodh *et al*. (2014). Each rat was weighed individually, and 0.5ml of the 50% ethanol was administered for every 100g of its weight. For instance, a rat weighing 168.9g received 0.84ml of the 50% ethanol solution, which is shown as follows:

If 100g of rat was administered with 0.5ml

Then, 168.9g of rat will be administered with:

$0.5/100 \times 168.9 = 0.84$ml

Acute toxicity study (ethanol)

The Fixed Dose procedure, as outlined by OECD (2001), was employed, where three rats were orally administered 50% ethanol at a dose of 0.5ml/100g of body weight for two weeks.

Acute toxicity study (Allium cepa)

The acute toxicity study of the *Allium cepa* extract was conducted using Locke's method, as described by (Ibama *et al*., 2021). Nine rats were divided into three groups of three rats each. Each group was orally administered the aqueous extract of *Allium cepa* at doses of 400mg/kg, 800mg/kg, and 1600mg/kg, respectively. The animals were then observed for 48 hours for any signs of toxicity or death.

Experimental design

After a 14-day acclimatization period, the rats were divided into five groups of five rats each. Group 1 received only rat pellets and water *ad libitum* for 30 days and served as the negative control. Group 2 was administered 50% ethanol orally at a dose of 0.5 ml/100g once daily for 30 days, serving as the positive control. Groups 3, 4, and 5 were administered 50% ethanol orally at a dose of 0.5 ml/100g, followed by 1.0 ml of aqueous extract of *Allium cepa* (onion) at doses of 200 mg/kg, 400 mg/kg, and 600 mg/kg, respectively, one hour later, once daily for 30 days.

Blood specimen collection

At the end of the 30th day of the experimental study, animals in each group were fasted overnight. They were then anaesthetized using cotton wool soaked with chloroform in a jar, after which blood samples were collected via cardiac puncture. Blood samples, amounting to 4 ml

each, were aseptically collected into plain bottles using sterile 5 ml syringes. The samples were centrifuged at 3000 rpm for 5 minutes, separating the serum, which was then transferred into another plain bottle.

Sample analysis

The serum samples were analyzed for C-reactive protein and lipid profile, including triglycerides, total cholesterol, HDL, and LDL (calculated using Friedewald□s equation). Absorbances for each parameter in the samples were measured using the UV1720 UV-Vis Spectrophotometer (Shanghai Yoke Instrument Co., Ltd., China).

Statistical analysis

The analysis results were presented as Mean ± Standard Deviation and processed using Statistical Package for the Social Sciences (SPSS) version 26 (IBM Corp., Armonk, NY, USA). Mean and standard deviation values for different parameters between test and control groups were compared using one-way ANOVA and Tukey tests. Statistical significance was set at a 95% confidence interval ($p < 0.05$).

Results

Qualitative phytochemical analysis

The findings of the qualitative phytochemical analysis of the aqueous extract of *Allium cepa*, is presented in Table 1. It indicates that the extract contains alkaloids, flavonoids, cardiac glycosides, phenols, saponins, terpenes, and steroids.

Table 1:Result of the Qualitative Phytochemical Analysis

Phytochemical compounds	Status
Alkaloids	+
Flavonoids	+
Saponin	+
Cardiac glycosides	+
Terpenes	+
Steroids	+
Phenols	+

Table 2: Toxicity study of Allium cepa extract

The results of the toxicity study for *Allium cepa* extract is displayed in Table 2. It indicates that administration of the aqueous extract at doses of 400 mg/kg, 800 mg/kg, and 1600 mg/kg did not produce any signs of toxicity or cause mortality after 48 hours of observation.

Table 2: Results of Toxicity Study of *Allium cepa* Extract

Dose (mg/kg)	Observation
400	No mortality
800	No mortality
1600	No mortality

Toxicity Study of 50% Ethanol

The results of the toxicity study on 50% ethanol, as presented in Table 3 show that oral administration of 0.5 ml/100g once daily for two weeks did not result in any signs of toxicity or mortality.

Table 3: Results of Toxicity Study of 50% Ethanol

Dose	Observation
0.5ml/100g	No mortality

Comparison of the Levels of Lipid Profile and C-reactive Protein of the Control and Test Groups

The comparison of lipid profiles and serum C-reactive protein (CRP) levels across groups I, II, III, IV, and V is detailed in Table 4. CRP Levels: Group II (positive control) had significantly higher CRP levels (14.30±0.97 mg/L) compared to group I (7.64±2.12 mg/L, p=0.000035). Groups III, IV, and V showed significantly lower CRP levels compared to group II, but no significant difference was found among groups I, III, IV, and V. For total Cholesterol, the mean total cholesterol of Group II (2.55±0.28 mmol/L) was significantly higher than that of group I (1.77±0.48 mmol/L, p=0.010). Groups III, IV, and V had significantly lower total cholesterol levels compared to group II, with no significant differences among groups I, III, IV, and V. HDL Levels: No significant differences in HDL levels were found among the groups (p=0.179). LDL Levels: Group II had significantly higher LDL levels (1.40±0.40 mmol/L) compared to group I (0.78±0.30 mmol/L, p=0.038). Groups III and IV had lower LDL levels than group II but higher than groups I and V, with no significant difference between groups III and IV. Triglycerides: No significant differences were observed in triglyceride levels among the groups (p=0.898).

Table 4: Mean Levels of the Lipid Profile Parameters of Control and Test Groups

Groups	CRP (mg/L)	T. Chol. (mmol/L)	HDL (mmol/L)	LDL (mmol/L)	TG (mmol/L)
Group I (NC)	7.64±2.12^{b}	1.77±0.48^{b}	0.92±0.35	0.78±0.30^{a}	0.45±.039
Group II (PC)	14.30±0.97^{a}	2.55±0.28^{a}	0.82±0.14	1.40±0.40^{d}	0.37±0.16
Group III (200mg/Kg)	10.48±1.50^{b}	2.18±0.38^{b}	1.06±0.72	1.01±0.41	0.33±0.29
Group IV (400mg/Kg	10.78±2.16^{b}	1.64±0.46^{b}	0.57±0.19	1.02±0.52	0.43±0.30
Group V (600mg/Kg)	8.06±1.49^{b}	1.71±0.42^{b}	0.53±0.20	0.54±0.37^{b}	0.31±0.07
F-value	12.201	4.426	1.749	3.121	0.263
***P* -value**	0.000035	0.010	0.179	0.038	0.898
Remark	**S**	**S**	**NS**	**S**	**NS**

KEY: *CRP =C-reactive protein, T. Chol = Total cholesterol, HDL = High density lipoprotein, LDL = Low density lipoprotein, TG = Triglyceride. Values with different superscripts are significantly different at P <0.05, S = Significant, NS = Not significant*

Discussion

This study aimed to evaluate the therapeutic effects of aqueous *Allium cepa* extract on ethanol-induced dyslipidemia and inflammation in Albino Rats. The phytochemical analysis of *Allium cepa* extract revealed the presence of alkaloids, flavonoids, cardiac glycosides, phenols, saponins, terpenes, and steroids, aligning with the findings of Gazuwa *et al*. (2013). Phytochemicals in onions play a crucial role in promoting health due to their antioxidant, anti-inflammatory, and cardioprotective properties. These compounds, such as flavonoids, saponins, and phenolics, help reduce oxidative stress and inflammation, contributing to overall cardiovascular health and disease prevention (Zhao *et al*., 2021).

Administering 50% ethanol increased total cholesterol, low-density lipoprotein (LDL), and

C-reactive protein (CRP) levels. These increases likely result from the oxidative stress-inducing properties of ethanol, leading to dyslipidaemia and inflammation. This observation concurs with Ferreira-Borges *et al*. (2015), who reported similar lipid profile changes in ethanol-treated albino rats, and Albert *et al.* (2003), who noted elevated CRP levels in heavy alcohol consumers. According to Ye *et al*. (2023), ethanol consumption disrupts lipid metabolism, leading to elevated total cholesterol, LDL, and triglycerides, and decreased HDL, primarily due to oxidative stress, inflammation, and impaired liver function, and that this dyslipidemia significantly increases the risk of cardiovascular diseases.

The *Allium cepa* extract significantly decreased CRP levels across different doses, highlighting its anti-inflammatory properties. Elevated CRP in group II (positive control) was due to ethanol-induced inflammation, while the reduction in CRP in groups III (200mg/kg), IV (400mg/kg), and V (600mg/kg) may be attributed to the anti-inflammatory effects of *Allium cepa*, supported by the studies of de-Groot and Rauben (1998) and Kim *et al.* (2012). These studies demonstrated the role of flavonoids and quercetin in *Allium cepa* in reducing inflammation.

The extract also significantly lowered total cholesterol in treatment groups compared to the ethanol group, indicating an antihypercholesterolemic effect likely due to the phytochemicals in *Allium cepa*. This result is consistent with Lee *et al.* (2008), who found that *Allium cepa* extract significantly reduced total cholesterol in rats with diet-induced dyslipidaemia.

A notable decrease in LDL was observed in group V (600mg/kg) compared to groups I and II, suggesting that a higher dose of *Allium cepa* extract effectively lowers serum LDL levels. This finding aligns with Lee *et al*. (2008), who reported a significant reduction in LDL with *Allium cepa* treatment in dyslipidemia rats.

No significant differences in high-density lipoprotein (HDL) levels were observed among the treatment, positive control, and negative control groups. This result supports the findings of Lu *et al*. (2015), who also reported no significant change in HDL levels with *Allium cepa* treatment in ethanol-induced dyslipidaemic rats. However, it contradicts Mattiuzzi *et al*. (2020), who observed a significant HDL decrease with *Allium cepa* extract, possibly due to variations in ethanol administration duration and dosage between their work and the present study.

Lastly, there were no significant differences in triglyceride levels between the treatment groups and control groups. This outcome disagrees with Mattiuzzi *et al.* (2020), who reported a significant triglyceride reduction after *Allium cepa* extract administration. The discrepancy may be due to differences in ethanol administration duration and dosage. However, it agrees with Lu *et al*. (2015), who found no significant change in triglyceride levels in alcohol-induced dyslipidemic rats treated with Allium cepa extract.

Conclusion

The aqueous extract of *Allium cepa* demonstrated significant anti-inflammatory and anti-hypercholesterolemic effects on ethanol-induced dyslipidaemia Albino Rats. This was evidenced by a reduction in serum C-reactive protein, total cholesterol, and low-density lipoprotein levels. All treatment doses of the extract effectively lowered serum C-reactive protein and total cholesterol levels. However, a decrease in serum low-density lipoprotein levels was only observed with the highest dose of the extract. This implies that the aqueous extract of *Allium cepa* has potential therapeutic benefits for managing inflammation and dyslipidaemia, particularly those induced by ethanol. The ability of the extract to reduce serum C-reactive protein and total cholesterol levels across all doses suggests a consistent anti-inflammatory and cholesterol-lowering effect. However, the requirement of a higher dose to decrease low-density lipoprotein levels indicates that the efficacy of the extract on different lipid parameters may vary and that higher concentrations might be necessary to achieve comprehensive improvements of lipid profile.

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Citation: Onengiyeofori Ibama, Godselect Chibuihe Elekwachi, Arit Okechukwu Nwogu, Ubong Friday Akpannanah. Effect of Aqueous Extract of *Allium cepa* (Onion) on Ethanol-Induced Dyslipidemia and Inflammation in Albino Rats .***Sokoto Journal of Medical Laboratory Science*; 9(3): 308 – 315.** https://dx.doi.org/10.4314/sokjmls.v9i3.32