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Anti-hyperlipidemic, Anti-inflammatory and Antioxidant Activities of Citrullus lanatus

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ABSTRACT

Aims: *The objective of this study was to evaluate the hypolipidemic effect, anti-inflammatory and antioxidant properties of Citrullus lanatus seed extract in cholesterol induced experimental obese mice.* ***Methods and Material:*** *Adult male Mus Musculus mice were divided into four groups. The control group (S) was fed with flour balls (100 mg/mice), group (B) was fed with cholesterol (400 mg/kg/day), group (E) was fed with cholesterol (400 mg/kg/day) plus C. lanatus seed extract (120 mg/kg/day) and the group (R) was treated with C. lanatus seed extract (120 mg/kg/day). Cholesterol, triglycerides, HDL-cholesterol, LDL-cholesterol, AST, ALT and CRP were measured with enzymatic methods. To elucidate the antioxidant property, the enzymatic antioxidants such as glutathione (GSH) and catalase, in hepatic supernatant have been determined.* ***Statistical analysis used:*** *The results were analyzed by ANOVA test and Tukey's multiple comparison tests (SPSS V.20).* ***Results:*** *The results showed that the administration of seed extract of C. lanatus significantly decreased serum cholesterol, TG and LDL-c levels, and significantly increased serum HDL-c level. Hyperlipidemic diet induced a significant rise in AST and ALT concentrations. The administration of the seed extract of C. lanatus effectively reduced cholesterol. The CRP concentrations were significantly elevated after the administration of cholesterol to mice. The results showed a significantly high decrease in CRP values in the plant group. The glutathione was reduced, and the catalase levels in plant group were significantly decreased.* ***Conclusions:*** *This study demonstrated a good anti-hyperlipidemic, anti-inflammatory and antioxidant potential of C. lanatus, which fitted well with their use in folk medicine.*

Keywords: *Hyperlipidemia, Citrullus lanatus, oxidative stress, anti-inflammatory activity.*

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1. INTRODUCTION

From the seventeen major causes of human death, cardiovascular diseases (CVD) rank the seventh. The coronary heart disease (CHD) has been the cause of mortality in 50% of people around the world (Sullivan & David 2002). The hyperlipidemia has been considered the most influential risk factor for CHD (Marzyieh et al. 2007). Moreover, the reactive oxygen species like superoxide anions, hydrogen peroxide and hydroxyl, and nitric oxide radicals, have had an important role in oxidative stress related to the pathogenesis of numerous diseases like the cardiovascular diseases (Jayaraman & Christina 2013).

The inflammation is a prominent feature of atherosclerosis (Libby et al. 2002) and it has been postulated like an acute-phase protein, and the elevation of plasma C-reactive protein may signal the underlying atherosclerotic process. Although, many epidemiological studies have shown that plasma CRP level is an excellent independent predictor of CVD in both men and women (Ridker et al. 2002) and it is an excellent marker of the rate of progression of atherosclerosis (Paul et al., 2004).

Fruits and vegetables have been recognized as natural sources of various bioactive compounds (Pennington & Fisher, 2010) which could be attributed to their phytochemical constituents such as flavonoids, anthocyanin, ascorbic acid, tocopherol, phenolic compounds, dietary fiber, and carotenoids present in fruits and vegetables (Kolawole et al. 2016).

Flavonoids are a group of polyphenolic compounds found abundantly in the plant kingdom. Interest in the possible health benefits of flavonoids and other polyphenolic compounds has increased in recent years owing to their potent antioxidant and free-radical scavenging activities (Rahman et al. 2013 a).

Watermelon (*Citrullus lanatus*, family Cucurbitaceae) is a vine-like flowering plant originally from Southern Africa (Mandel et al. 2005). It is an important vegetable crop in Africa, and can adapt to different environmental conditions (Adetutu et al. 2015).

Gill et al. evaluated the antioxidant, anti-inflammatory and analgesic potential of the *Citrullus lanatus* seeds (Gill et al. 2010). Logaraj et al. investigated that the watermelon seeds are a good source of linoleic acid (18:2 ω-6) as a major fatty acid (Logaraj, 2011). Atlaş et al. concluded that the watermelon juice keeps the liver, kidney and brain tissues safe in case of experimental CCL4 toxicity in rats, and the protective effect of watermelon juice may be because of having antioxidant

activity and inhibiting lipid peroxide formation (Altaş et al. 2011).

There have been therapeutic effects of *C. lanatus* fruit found, which have been stated to be related to its antioxidant and certain phytochemical compounds (Adetutu et al. 2015).

In this context, the aim of this research was to determine the effects of the consumption of watermelon seeds' extract against hypercholesterolemia induced by high cholesterol intake in mice and also to assess its antioxidant and anti-inflammatory potentials.

2. MATERIALS AND METHODS

2.1. Plant material

The seeds of *C. lanatus* were collected from Constantine (East of Algeria). They were mechanically grinded and converted into the coarse powder, then the extract was prepared by simple maceration process. The coarse seed powder was weighed, and adequate distill water was added. After the completion of duration of maceration, they were filtered using whatmann filter, then the filtrates were collected in order to be used for treatment preparations.

2.2. Experimental animal and diets

The experiments were performed on 28 adult male *Mus Musculus* mice (2.5- 3 months old) which were obtained from animal center of Pasteur's Institute (Algiers–Algeria), weighing (14-18g). They were housed in hanging transparent plastic cages (55 × 33× 19 cm) in the animal room of Faculty of Sciences (University Mentouri Brothers, Constantine 1-Algeria) and maintained under the controlled conditions of humidity; 12-h light and 12-h dark cycle, and a constant temperature of 22±3°c. Food and water were freely available in the home cages. The food was provided in the form of dry pellets (SARL Production Locale, Bouzareah, Algiers-Algeria). The litter was renewed every 3 days.

After the adaptative period, the animals were assigned into four groups of similar mean of body weights, and were fed for 3 w* with control and experimental diets.

The control group (S) was fed with white flour (100 mg/mice), group (B) was fed with cholesterol (400mg/kg/day), group (E) was fed with cholesterol (400mg/kg/day) plus *C. lanatus* seed extract (120m g/kg/day), and the group (R) was treated with *C. lanatus* seed extract (120mg/kg/day). The experimental diets were given white flour.

The experiments were carried out in strict compliance based on ethical principles, and provided by the Committee for the Purpose of Control and Supervision of Experiments on the Animal (CPCSEA).

2.3. Biochemical investigation

After 3 w* of the treatment, the blood samples were collected after fasting, from the retro orbital plexus into EDTA tubes by using glass capillaries. Plasma was obtained by low speed (2000 rpm) centrifugation for biochemical analysis.

Total cholesterol, TG, HDL-Cholesterol, LDL-Cholesterol and Aspartate Transaminase _AST and Alanine Transaminase _ALT concentrations, assays were done by enzymatic kits.

The plasma CRP values were measured by the immunoturbidimetric method. All parameters were analysed by Auto analyser Cobas integra 400 plus analyzer (Roche).

The livers were removed from all animals, and washed with ice cold saline. Small pieces of liver tissues was collected and kept in 10% formalin solution for conducting histopathological examinations.

2.4. Preparation of the homogenate

The liver was excised, and rinsed in ice- cold saline. The weight of 0,5g liver was homogenized in 2ml of TBS (Tris 50 mM, NaCl 150 mM, pH 7.4) at 4°C, in a Potter–Elvehjem homogenizer with a Teflon pestle at 600 rpm for 3 min. The homogenate was then centrifuged at 9000 g for 15 min at 4°C. The supernatant was collected as tissue homogenate, that was used to determine protein concentrations, reduced glutathione and the catalase activities.

2.5. Protein quantification

The total soluble protein in the supernatant was measured the method of Bradford (Bradford 1976) by using bovine serum albumin as standard protein.

2.6. Determination of reduced glutathione (GSH)

The decreased glutathione content in the liver was estimated spectrophotometrically by using 5,5′-dithiobis-2 nitrobenzoic acid) (DTNB) as a coloring reagent, according to the method of Weckbeker (Rahman 2006).

2.7. Determination of catalase (CAT)

Tissue CAT activity was determined according to Aebi's method (Aebi 1974). The principle of the assay was based on the determination of H_2O_2 decomposition rate at 240 nm.

2.8. Statistical analysis

The results were analyzed for differences between the groups across dietary treatments by one way ANOVA test and Tukey's multiple comparison tests (SPSS version 20), P value*<0.05 was considered as statistically significant.

3. RESULTS

3.1. Effect of cholesterol and the extract of *C. lanatus* seed on the serum lipid profile

The results of the effect of administration of cholesterol and the extract of *C. lanatus* seed on lipid profile (table 1) showed that, the total cholesterol concentration of mice in group (B) (1,67+0,15 mmol/L) was significantly higher ($p<0.05$) compared to the other groups.

Also, the administration of cholesterol caused a significantly high elevation (P=0,000) of triglyceride concentration in group (B) (1,81±0,30 mmol/L).

However, HDL-c concentration in groups (B) (0,86±0,12 mmol/L) and (R) (0,77±0,08 mmol/L) was significantly different (P=0,000) compared to the groups (S) (1,09±0,04 mmol/L) and (E) (0,95±0,06 mmol/L); respectively.

Furthermore, LDL-c concentration in group (B) (0,53±0,42 mmol/L) statistically showed no significant difference ($p>0.05$) when compared to the group (S) (0,28±0,11 mmol/L).

The administration of the seed extract of *C. lanatus* seed, caused a significant reduction ($p<0.05$) in the values of total cholesterol, triglyceride and LDL-c in groups (E) and as (R); respectively compared to group B. However, the extract of *C. lanatus* seed caused a significant elevation in the values of HDL-c cholesterol in groups (E) (0,48±0,20 mmol/L) and (R) (0,22±0,06 mmol/L); respectively ($p<0.05$).

Table 1. Effect of cholesterol and the extract of *C. lanatus* seed on lipid profile of mice.

Treatment	Total cholesterol (mmol/L)	Triglyceride (mmol/L)	High Density Lipoprotein (mmol/L)	Low Density Lipoprotein (mmol/L)
(S) Control group: white flour (100mg/mice)	1,5+0,31	1,73±0,12	1,09±0,04***	0,28±0,11
(B) : cholesterol (400mg/kg/day)	1,67+0,15 *	1,81±0,30***	0,86±0,12***	0,53±0,42 **
(E) :cholesterol (400mg/kg/day) +*C. lanatus* seed extract (120m g/kg/day)	1,47+0,13*	1,15±0,16	0,95±0,06*	0,48±0,20
(R) :*C. lanatus* seed extract (120mg/kg/day)	1,4+0,07*	0,84±0,08	0,77±0,08*	0,22±0,06

n = 7; *p<0.05, **p<0.01 and ***p=0.000; compared with the control (*one way ANOVA; LSD post hoc test*); values represented in Mean ± SEM.

3.2. Effect of cholesterol and the extract of *C. lanatus* seed on the CRP values

The present data showed that, at the third week, there was a significantly high difference in the means for the plasma CRP concentrations between the groups (P=0,000). Fig.1 showed that the plasma CRP concentration in group (B) achieved higher levels, and the average was (4,36 ±0,06µmol/L), which was very higher than that of the control group (S) (0,11±0,06) and group (E) (3,72±1,04) (P=0,000). However, the treatment with the extract of *C. lanatus* seed in the group (R) (2.55±0,84) decreased very significantly compared to the groups of (B) and (E); respectively (P=0,000).

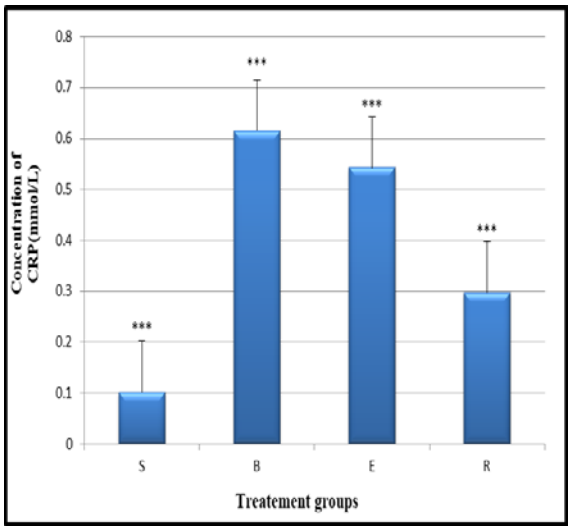

Fig. 1. The interaction of cholesterol and the extract of *C. lanatus* seed on the plasma CRP in mice during 21 days of treatment, Values are the means ± SEM (n=7); *p<0.05 , **p<0.01 and ***p=0.000

3.3. Effects of cholesterol and the extract of *C. lanatus* seed on the assayed liver enzymes

The effects of the extract of *C. lanatus* seed on the assayed liver enzymes, (ALT) and (AST) has been shown in (table 2).

The administration of cholesterol resulted in a significant (*P*<0.05) increase in both the serum levels of ALT and AST in group (B) when compared with the control group (S) (table 2).

However, the treatment with the extract of *C. lanatus* seed caused a very high significant (p=0.000) reduction in the levels of AST in group (R) (101,80±3,12 IU/L) compared to the control group (S) (88,37±21,26 IU/L).

Also, the results indicated that the animals treated with the extract of *C. lanatus* seed, group (R) (94,66±9,93 lU/L) showed a significantly high (p=0.000) decrease in the activity of ALT compared to the control group (S) (28,35±4,86 lU/L).

Table 2. Effect of cholesterol and the extract of *C. lanatus* seed on AST and ALT levels

Treatment	AST (lU/L)	ALT (IU/L)
(S) Control group: white flour (100mg/mice)	88,37± 21,26	28,35± 4,86
(B) : cholesterol (400mg/kg/day)	121,06± 14,85 *	101,58± 22,76 *
(E):cholesterol (400mg/kg/day) + *C. lanatus* seed extract (120m g/kg/day)	106,9± 25***	95,57± 10,14***
(R) : *C. lanatus* seed extract (120mg/kg/day)	101,80± 3,12***	94,66± 9,93***

n = 7; *p<0.05, **p<0.01 and ***p=0.000; compared with the control (*one way ANOVA; LSD post hoc test*); values represented in Mean ± SEM

3.4. Effect of cholesterol and the extract of *C. lanatus* seed on oxidative stress parameters of mice

As shown in (fig.2), there was a significantly high (p=0.000) decrease in the GSH concentration of group (B) (7,99±0,27 n mol/mg protein) compared with the normal control group (S) (12,7±0,29 n mol/mg protein). However, the concentration of reduced GSH increased very highly in group (R) (10.88±0.89 n mol/mg protein) (P=0.000), and significantly in group (E) (9.09±0.75 n mol/mg protein) (P<0.05). On the other hand, the concentration of catalase decreased significantly (p<0.05) in group (B) (60.72±0.26 m mol/mg protein) compared to the control group (S) (85.64±0.3 m mol/mg protein). While, a significant (p<0.05) increase was observed in the catalase level of group (R) (79.22±0.05 m mol/mg protein), but it was not significant in the group (E) (72.06±0.25 m mol/mg protein) (P>0.05) (fig. 3).

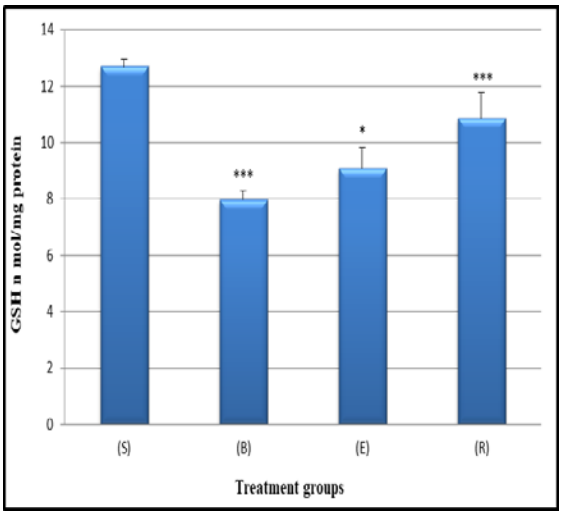

Fig. 2. The interaction of cholesterol and the extract of *C .lanatus* seed on the reduced glutathione GSH in mice during 21 days of treatment, Values are the means ± SEM (n=7); *p<0.05, **p<0.01 and ***p=0.000.

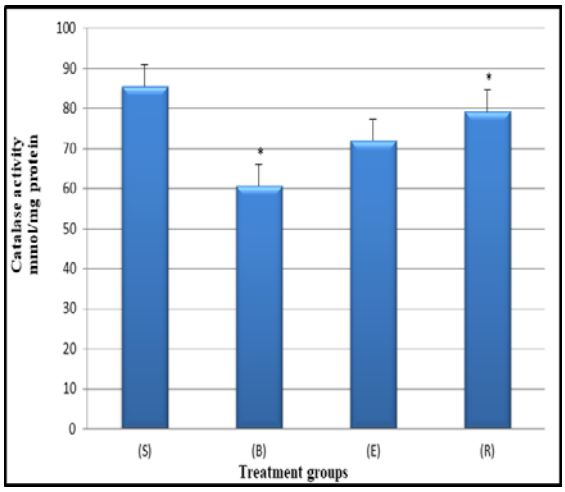

Fig. 3. The interaction of cholesterol and the extract of *C. lanatus* seed on Catalase activity in mice during 21 days of treatment, Values are the means ± SEM (n=7); *p<0.05, **p<0.01 and ***p=0.000.

4. DISCUSSION

The present study determined the effects of the extract of *C. lanatus* seed on lipid profile parameters, CRP, AST, ALT and some oxidative stress parameters in normal and obese mice. This was with the view to determine the anti-hyperlipidemic potentials of the extract of *C. lanatus* seed.

Before the time cholesterol was deposited in the plaques that hardens and narrow arteries, it must be oxidized by free radicals. Several epidemiological studies have indicated an inverse relationship between HDL-cholesterol levels in serum and the incidence and prevalence of coronary heart disease (Georgina et al. 2011).

The results of this study showed that the administration of cholesterol (400mg/kg/day) to the experimental animals for a period of 3 w* caused a significant increase in the concentration of total cholesterol, triglyceride and LDL-c. Also, the administration of cholesterol caused a significant reduction in the values of HDL-c.

This result could be attributed to hepatotoxicity caused by the administration of 400mg/kg/day of cholesterol.

Hypertriglyceridemia and hypercholesterolemia have probably been resulted due to the increased synthesis and secretion of lipoproteins because of hepatic hyperlipogenese (Khemmar et al. 2012).

In addition, the extract of *C. lanatus* seed was found to significantly reduce cholesterol, triglyceride and LDL-c concentration, and significantly increase the HDL-c concentration in the obese treated mice.

In accordance with (Mehta & Gelfand 2014), the consumption of *C. lanatus* led to the reduced body weight gain, decreased plasma cholesterol, triglyceride and LDL-c concentrations which was in agreement with the findings of this research work.

These effects can be explained by the presence of some inhibitory factors of digestibility in C.*lanatus* decreasing intestinal absorption of lipids by promoting their fecal elimination. Moreover, the mean levels of plasma lipids remained relatively low in rats which were fed diets with *C. lanatus*, which was rich in minor components such as hydrocarbons, mainly squalenes, α- tocopherol and phytosterols (Khemmar et al. 2012). Many and various studies have shown that these substances exerted beneficial effects (Chan et al. 1996).

The observed decrease in serum triglyceride concentration in the water melon-fed group (R) with *C. lanatus* seed extract (120mg/kg/day) also explained the positive benefits of the extract of *C. lanatus* seed on serum lipids.

High density lipoprotein (HDL-c) acted as a powerful endogenous defense mechanism against atherogenesis. Apolipoprotein A-1 is a central component of HDL-c that led to the formation of HDL-c *in vivo*.

It has been observed that apolipoprotein A-1 transgenic expression resulted in the reduction of lesion formation in apolipoprotein-E knockout mice (Alex & Adekunle 2016).

LDL-c is a lipoprotein that transports cholesterol and triglyceride from the liver to peripheral tissues. It enables fat and cholesterol to move within the water blood solution of the blood stream. LDL-c is often called bad cholesterol; hence low levels are beneficial (Georgina et al. 2011).

In group (R), there was a significant decrease in the levels of LDL-c, this might be due to the fact that *C. lanatus* contains citrulline, which is an amino acid produced in the body from glutamate; citrulline is used in the body to make arginine which produces the nitric oxide that is vital in maintaining the vessels including L-arginine and nitric oxide dilates vessels, hence, aid the kidney to function better, also lower the concentration of LDL-c (Onyeso et al. 2016).

Arginine increases the oxidation of fats and glucose, reduces hyperglycemia, improves dyslipidemia, and reduces the fat mass in obese diabetic animals (Fu et al. 2005). Citrulline is converted into arginine, thus highlights the potential for exploring the effects of the consumption of watermelon on regulating the metabolism of energy substrates, improves the cardiovascular and immunological functions, and avoids the increased oxidative stress (Massa et al. 2016).

Group B (cholesterol) showed a very high increase in the level of plasma CRP compared to the control group (S). This result could reflect the initiation of an inflammatory process.

In group (E), it was noticed that the extract of *C. lanatus* seed lowered the plasma CRP level because of its confirmed anti-inflammatory effect, and this was due to the presence of established polyphenolic compounds such as tannin and flavonoids (Benmebarek et al. 2013). Flavonoids like fisetin and quercetin have been shown to inhibit the oxidative modification of LDL-c by macrophages. The activation of NF-κB is critical for the production of pro-inflammatory cytokines (Benmebarek et al. 2014).

Considering the findings of the current examination, the oral administration of *C. lanatus* seed resulted in anti inflammatory and analgesic actions which might caused by the free radical scavenging activity (Gill et al. 2010).

The metabolism of cholesterol takes place in the liver. Consequently, the metabolism of cholesterol in the liver was affected by cholesterol extra induced hepatotoxicity (Mehta & Gelfand 2014).

The biochemical quantification of activities of liver enzymes such as AST and ALT has been used as biomarkers of liver damage (Adeyemi et al. 2018). In particular, the increased AST and ALT levels have been the indicatives of liver damage (Adebayo et al. 2014). They are located in hepatic cells and are released after cell damage (Himmerich et al. 2001). The rise in levels of ALT is always accompanied by the elevation in the level of AST, which plays a role in the conversion of amino acid to keto acid (Haturvedi et al. 2014).

The results showed a significant increase in ALT and AST levels in the cholesterol-treated group (B) homogenate. The elevated levels of these liver marker enzymes were the indications of cell membrane damage in the hepatocytes leading to the loss of functional integrity of the liver (Onyeso et al. 2016).

The results of this study revealed that the administration of the extract of *C. lanatus* seed caused very high ($p=0.000$) decrease on AST and ALT level in mice groups treated with the extract (120m g/kg/day) compared to the untreated groups.

This result was in line with the work reported by (Rahman et al. 2013 b) that the plant *C.lanatus* lowered the serum level of transaminases, and that the plant had a hepatoprotective function.

The hepatoprotective property of this plant might be due to the presence of flavonoids in their individual state (Gupta & Misra 2006).

The antioxidant properties of plants must be evaluated by several methods because these plants contain complex phytochemicals. In this case, tests of the antioxidant activity whose mechanisms of action were different and complementary were necessary (Boudjouref et al., 2018).

The activity levels of antioxidant enzymes such as catalase and glutathione peroxidase have often been employed as biomarkers of oxidative stress in animals (Adeyemi et al. 2018).

Oral administration of extract of *C.lanatus* seed at doses of (120 mg/kg/day) showed a very high ($P<0.01$) decrease in catalase and glutathione concentrations compared with the cholesterol exposed mice (400 mg/kg/day) as shown in (table 2).

This might suggest that the consumption of high amount of watermelon might protect against lipid peroxidation. And as (Pellegrini & Porrini 2000) stated, eating foods rich in lycopene, beta carotene and vitamin C might not inhibit the activity of other antioxidant enzymes in plasma.

An increase in the level of lipid peroxidation in hepatotoxic mice suggested that there was an increased generation of free radicals. Cellular radical scavenging systems included the enzymes such as catalase (CAT) which removes hydrogen peroxide.

Therefore, the reduction in the activity of this enzyme (CAT) resulted in a number of deleterious effects due to the accumulation of hydrogen peroxide (Ehrhart & Zeevalk 2003).

Glutathione (GSH) is the abundant thiol compound present in mammalian cells and plays an important role to scavenge free radicals in the first line of anti-oxidant defense system. Glutathione is a natural antioxidant which donates one electron to hydrogen peroxide and is formed in the process of oxidized glutathione. The reaction is catalyzed by glutathione peroxidase. Subsequently, the oxidized glutathione (GSSG) is reduced to GSH via NADPH-dependent reduction by glutathione reductase. The depletion of glutathione results in the inhibition of glutathione peroxidase activity and thus enhancement in the lipid peroxidation activities. The antioxidant role of glutathione is both direct and indirect in that it stimulates other endogenous antioxidants (Haturvedi et al. 2014).

This phenomenon might indicate that the supplementation of antioxidant in watermelon has successfully increased the levels of GSH in liver tissues.

The obtained results of the current study were in agreement with the study done by (Saada et al. 2010) which demonstrated that the pretreatment with lycopene, which is abundant in watermelon, remarkably improved the oxidant/antioxidant status and decreased the oxidative damage.

Watermelon showed a good antioxidant activity in vitro. In addition, the hepatic-protective effect of water-melon on oxidative stress in mice has been demonstrated (Oyenihi et al. 2016; Adebayo et al. 2014).

Also, the study of (Kumar et al. 2010) suggested that an increased intake of antioxidants appeared to be protective in cardiovascular diseases.

5. CONCLUSION

In conclusion, the present study reported that the administration of the extract of *C. lanatus* seed caused a reduction in the concentration of cholesterol, triglyceride, LDL-c and increased HDL-c concentration with associated reduction in the concentration of CRP, AST and ALT concentrations in male *M. musculus* mice.

It also reported that *C. lanatus* seed contains some nutrients possibly antioxidants which were capable of suppressing oxidative stress.

This study suggested that the extract of *C. lanatus* seed might be a good antioxidant as its supplementation decreased lipid peroxidation.

However, the phytoconstituents which cause the antioxidant activity of the extract have not been determined clearly. Hence, more examinations are needed to be conducted to identify the mechanism of the antioxidant activity of this plant.

Conflict of Interest

The authors declared no conflict of interest.

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REFERENCES

1. Adebayo AH, Song FH, Liu XT et al. (2014) Citrullus lanatus extract reverses oxidative and haematological dysfuntion in carbon tetrachloride induced liver damaged rats. Inter J Pharmacol 10 (4): 218-224.
2. Adebayo AH, Yakubu OF, Balogun TM (2014) Protective properties of Citrullus lanatus on carbon tetrachloride induced liver damage in rats. Eur J Med Plants 4 (8): 979-989.
3. Adetutu A, Olorunnisola OS, Owoade OA (2015) Nutritive values and antioxidant activity of Citrullus lanatus fruit extract. Food Nutr Sci 6 (11): 1056-1064.
4. Adeyemi JA, Arowolo OK, Olawuyi ST et al. (2018) Effect of co-administration of Green Tea (Camellia sinensis) on Clove-(Syzygium Aromaticum) Induced Hepatotoxicity and Oxidative Stress in Wistar Rats. Indian J Physiol Pharmacol 62 (2): 195-201.
5. Aebi H (1974) Catalase in Methods of Enzymatic Analysis. In: Bergmayer, H.U., Ed., Chemie, 2nd Edition, Vol. 2, FRG, Weinheim, 673-684.
6. Alex OD, Adekunle AA (2016) Effect of methanolic extract of Citrulus lanatus seed on lipid profile and oxidative stress in acetaminophen intoxicated rats. Adv Biomed Pharm 3 (2): 87-93.
7. Altaş S, Kizil G, Kizil M, Ketani A, Haris PI (2011) Protective effect of Diyarbakır watermelon juice on carbon tetrachloride-induced toxicity in rats. Food Chem Toxicol 49 (9): 2433-2438.
8. Benmebarek A, Zerizer S, Laggoune S, Kabouche Z (2013) Immunostimulatory activity of Stachys mialhesi de Noé. Allergy, Asthma Clin Immunol 9 (1): 2.
9. Benmebarek A, Zerizer S, Lakhal H, Kabouche Z (2014) Biphasic dose response effect of Stachys ocymastrum on the reticuloendothelial system phagocytic activity. Intern J Pharm Pharm Sci 6 (2): 534-537.
10. Boudjouref M, Belhattab R, Bouteghrine S (2018) Antioxidant Activity and Phenolic Content of Artemisia Campestris from Two Regions of Algeria. World Journal of Environmental Biosciences 7 (2) : 61-66.
11. Bradford MA (1976) Rapid and sensitive method for the quantities of microgram quantities of protein utilizing the principle of protein-dye binding. Anal Biochem 72 (1-2): 248-254.
12. Chan P, Tomlinson B, Lee CB, Lee YS (1996) Effectiveness and safety of low-dose pravastatin and squalene, alone and in combination, in elderly patients with hypercholesterolemia. J Clin Pharmacol 36 (5): 422-427.
13. Ehrhart J, Zeevalk GD (2003) Cooperative interaction between ascorbate and glutathione during mitochondrial impairment in mesencephalic cultures. J Neurochem 86 (6): 1487-1497.
14. Fu WJ, Haynes TE, Kohli R et al. (2005) Dietary L-arginine supplementation reduces fat mass in Zucker diabetic fatty rats. J Nutr 135 (4): 714–721.
15. Georgina EO, Kingsley O, Esosa US, Njoya KH, Amegor OF, Okonkwo CA (2011) Comparative evaluation of antioxidant effects of watermelon and orange, and their effects on some serum lipid profile of Wistar albino rats. Inter J Nutr Metab 3 (8): 97-102.
16. Gill NS, Bansal RK, Manju G, Shailja, S, Arunachalam M, Manoj B (2010) Evaluation of antioxidant, anti-inflammatory and analgesic potential of Citrullus lanatus seed extract in rodent model. Internet J Nutr Wellness 9. The Internet Journal of Nutrition and Wellness. 2009 Volume 9 Number 2. unpaginated ref.25.
17. Gupta AK, Misra N (2006) Hepatoprotective activity of aqueous ethanolic extract of Chamomile capitula in paracetamol intoxicated albino rats. Am J Pharmacol Toxicol 1 (1): 17-20.
18. Haturvedi P, Pipedi TM, Tumed A (2014) Supplementation with watermelon renders protection against toxicity induced by paracetamol in albino rats: the mutual and fine interaction of antioxidants prevented the cellular damage. Int J Food Agric Vet Sci 4(1): 102-111.
19. Himmerich H, Anghelescu I, Klawe C, Szegedi A (2001) Vitamin B12 and hepatic enzyme serum levels correlate in male alcohol-dependent patients. Alcohol 36 (1): 26-28.
20. Jayaraman R, Christina AJM (2013) Citrullus colocynthis attenuates hyperlipidemia and hyperglycemia through its anti-oxidant property against hyperlipidemic and diabetic animal models. Der Pharmacia Sinica 4 (1): 60-66.
21. Khemmar L, Amamou F, Yazit M, Didi A, Daoudi CS (2012) Anti-obesity and anti-hyperlipidemic effect of Citrullus colocynthis oil in the offspring of obese rats. Ann Biol Res 3 (5): 2486-2490.
22. Kolawole TA, Ojeka SO, Dapper DV (2016) Effect of methanolic extract of Citrullus lanatus seed on lipid profile and oxidative stress in acetaminophen intoxicated rats. Adv Biomed Pharm 3 (2): 87-93.
23. Kumar SV, Saritha G, Fareedullah Md (2010) Role of antioxidants and oxidative stress in cardiovascular diseases. Ann Biol Res 1 (3): 158-173.
24. Libby P, Ridker PM, Maseri A (2002) Inflammation and atherosclerosis. Circulation 105 (9): 868-874.
25. Logaraj TV (2011) Watermelon Citrullus lanatus (Thunb) Matsumura and Nakai seed oils and their use in health. Nuts and seeds in health and disease prevention. (1st edtn), Cambridge, Massachusetts (USA): Academic Press p 1149-1157.
26. Mandel H, Levy N, Izkovitch S, Korman SH (2005) Elevated plasma citrulline and arginine due to consumption of Citrullus vulgaris. J Inherited Metab Dis 28 (4): 467-472.
27. Massa NML, Silva AS, Oliveira CVC, Darlene CP, Carlos VS, Maria CR (2016) Supplementation with watermelon extract reduces total cholesterol and LDL

cholesterol in adults with dyslipidemia under the influence of the MTHFR C677T polymorphism. J Am Coll Nutr 35 (6): 514-520.

28. Mehta NN, Gelfand JM (2014) High-density lipoprotein cholesterol function improves after successful treatment of psoriasis: a step forward in the right direction. J Invest Dermatol 134 (3): 592-595.
29. Onyeso GI, Nkpaa KW, Onokohwomo JN, Nwaeme OVA (2016) Coadministration of caffeine and hydromethanolic fraction of Citrullus lanatus seeds extracts improved heamatological and lipid profile of wistar albino rats. J Adv Med Pharm Sci 5 (2): 1-7.
30. Oyenihi OR, Afolabi BA, Oyenihi AB, Ogunmokun OJ, Oguntibeju OO (2016) Hepato-and neuro-protective effects of watermelon juice on acute ethanol-induced oxidative stress in rats. Toxicol Rep 3: 288-294.
31. Paul A, KO WS, Lan L et al. (2004) C-reactive protein accelerates the progression of atherosclerosis in apolipoprotein E–deficient mice. Circulation 109 (5): 647-655.
32. Pellegrini NP, Porrini M (2000) Tomato consumption does not affect the total antioxidant capacity of plasma. Nutrition 16 (4): 268-271.
33. Pennington JAT, Fisher RA (2010) Food component profiles for fruit and vegetable subgroups. J Food Compos Anal 23 (5): 411-418.
34. Rahman H, Kode A, Biswas SK (2006) Assay for quantitative determination of glutathione and glutathione disulfide levels using enzymatic recycling method. Nat Protoc 1 (6): 3159-3165.
35. Rahman H, Manjula K, Anoosha T .a (2013) In-vitro antioxidant activity of Citrullus lanatus seed extracts. Asian J Pharm Clin Res 6 (3): 152-157.
36. Rahman H, Manjula K, Anoosha T, Nagaveni K, Eswaraiah CM, Bardalai D .b (2013) In-vitro antioxidant activity of Citrullus lanatus seed extracts. Asian J Pharm Clin Res 6 (3): 152-157.
37. Ridker PM, Rifai N, Rose L, Buring JE, Cook NR (2002) Comparison of C-reactive protein and low-density lipoprotein cholesterol levels in the prediction of first cardiovascular events. N Engl J Med 347 (20): 1557-1565.
38. Saada HN, Rezk RG, Eltahawy NA (2010) Lycopene protects the structure of the small intestine against gamma-radiation-induced oxidative stress. Phytother Res 24 (2): 204-208.
39. Sullivan D, David R (2002) Screening for cardiovascular disease with cholesterol. Clin Chim Acta 315 (1-2): 49-60.
40. Zamani,M. Rahimi, O. Mahdavi, R. Nikbakhsh, M. (2007) Assessment of anti-hyperlipidemic effect of Citrullus colocynthis. Rev Bras Farmacogn 17 (4): 492-496.