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Hypocholesterolemia Effect of Guazuma Ulmifolia Lamk on Rats Model Hyperlipidemic and Liver Histopathological Picture

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The number of herbal medicines in Indonesia as cholesterol drugs is still lacking. Guazuma ulmifolia Lamk. is one of the plants suspected of having an anticholesterol effect. The test design is pre and post test with control group design. A total of 30 rats were divided into 5 groups, with each group consisting of 6 rats. Aquadest as a negative control group and simvastatin 0.72mg/200 bw orally is a positive control group. Groups III, IV, and V were treated the test extract with 3 dose levels. A mouse model of hyperlipidemia was performed with Triton-X. Treatment was carried out for 7 days and cholesterol is measured. Liver organs was taken for histopathological examination. The ethanolic extract of the ethyl acetate fraction of Guazuma ulmifolia Lamk was able to reduce total cholesterol in rats induced by a high cholesterol diet. From the histopathological examination, the ethanol extract of the ethyl acetate fraction of Guazuma ulmifolia Lamk improved liver histoPA. The ethanolic extract of the ethyl acetate fraction of Guazuma ulmifolia Lamk has potencies to develop as hypocholesterolemia agent.

Keywords: Guazuma ulmifolia Lamk; Herbal Medicine; hypocholesterolemia Agent.

Dyslipidemia is one of factor that contribute heart diseases. Based on data from the basic health research in 2018, the incidence of heart and blood vessel disease is increasing from year to year. The prevalence of heart diseases in Indonesia in 2018 is 19,6%. It is estimated that 15 out of 1000 people (1.5%) or around 2,784,064 individuals in Indonesia suffer heart disease[1].

Several medicinal plants from Indonesia in preclinical study showed the effect of lowering cholesterol, including *Guazuma ulmifolia* Lamk[2,3]. The aim of this research are to determine of hypocholesterolemia effect of ethanolic extract of the ethyl acetate fraction of *Guazuma ulmifolia* Lamk on rats induced by a high cholesterol diet.

METHOD

Ethical clearance

This research has been accredited by ethic committee medical faculty of Universitas Muhammadiyah Surakarta with no: 3064/A.2/KEPK-FKUMS/VII/2021.

Extract preparation

The test plants were obtained from Tawang mangu (Central of Java Indonesia) dried under indirect sunlight. The dried simplicial was powdered and extracted by 96% ethanol followed by fractionation with ethyl acetate. The process is as follows: Simplicial powder was macerated by 96% ethanol for 3 days and filtered to get filtrate.

The simplicial was re-macerated once. The second filtrate was mixed with the first filtrate and was evaporated to obtain a pure extract. This pure extract was soaked in ethyl acetate for 24 hours and evaporated to obtain the ethyl acetate fraction. Several concentration series were prepared by adding Dimetil sulfoksida (DMSO) solvent.

Anti-cholesterol test

A number 30 male rats (Rattus norvegicus), 200±20 gbw (referred research by Usdiana *et al*,)[4], were randomly divided into 5 groups, with each group consisting of 6 rats. All rats were fed by high fat diet referred research by muhtadi et al,[5]. Aquest as a negative control group and simvastatin 0.72mg/200 BW orally was a positive control group. Groups III, IV, and V were treated by ethyl acetate fraction of *Guazuma ulmifolia* Lamk as dosage of 250; 500 and 1000 mg/kgbw. On the first day, all groups were induced by Triton X at a dose of 20 mg/200g BW by intraperitoneally. On the second day all rats were fed by a high cholesterol diet containing goat fat. On this day, all rats were treated according to their groups, namely: group I (negative control) was Aquest, group II (positive control) was simvastatin at dose of 0.72mg/200 BW

Table 1. Cholesterol levels in various groups during administration of the ethyl acetate fraction of Guazuma ulmifolia Lamk for 15 days

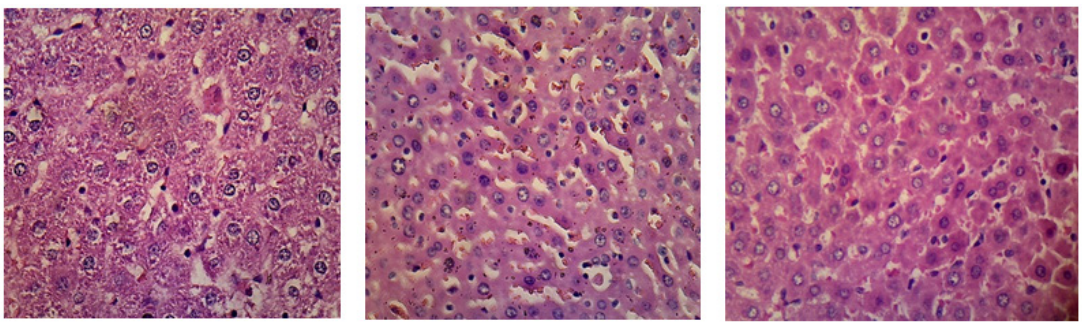
Group	Cholesterol level			
	Day 0	Day 1	Day 10	Day 15
Positive control (simvastatin)	172+48,73	162,75+27,65	205,75+106,64*	134,25+42,63*
Negative control – (*aqueous*)	154,4+44,76	168,6+30,07	272,33+41,12	145,12+86,95
GUL at dose 250mg/kgbw	117±29,44	231,2±59,68	185,2±37,0*	159±38,60
GUL at dose 500mg/kgbw	104,4±4,15	225,25±82,81	258,5±135,05**	201,5±31,81**
GUL at dose 1000mg/kgbw	110,5±14,84	208,25±100,13	182,81±28,90	168±50,02

GUL= *Guazuma ulmifolia* Lamk; *= significantly different from the negative control; **=not significantly different from the positive control.

Tabel 2. *Score* of histopathology of liver

–	Liver Score
Positive control (simvastatin)	1,27±0,41*
Negative control (Aquest)	3,12±0,74
GUL at dose 250mg/kgbw	1,6±0,22*
GUL at dose 500mg/kgbw	3,5±0,72
GUL at dose 1000mg/kgbw	3,8±0,44

Note: GUL= *Guazuma ulmifolia* Lamk); *significantly different from the negative control.



A B C

A. Negative Control; B. *Parenchymatous degeneration* C. Hydropic degeneration

Fig. 1. Histopathological of liver

po. Groups III, IV, and V were treated by fraction for 7 days. On day 0 and the end of treatment (day 7) all rats were measured for cholesterol levels.

Histopathological examination

At the end of the observation, mice were sacrificed by cervical decapitation. The liver was taken and stored in formalin for histopathological examination

RESULTS

Measurement of total cholesterol levels was carried out on day 0; 1; 10 and 15. The results of reducing total cholesterol levels can be seen in the tables 1 below.

From the tables 2 and figure 1, *Guazuma ulmifolia* Lamk at a dose of 250 mg/kgbw can reduce cholesterol levels in male Wistar rats. *Guazuma ulmifolia* Lamk at a dose of 250mg/bw improve liver histopathological profile.

This study is in line with research by Prahastuti *et al*., that found the ethanol extract of *Guazuma ulmifolia* Lamk at dose of 30 mg/rat for 42 days was able to reduce rat cholesterol levels with a percentage reduction of 7.35%[6]. The leaf extract of *Guazuma ulmifolia* Lamk at dose of 25 mg kg/bw was effective to improve the lipid profile of dyslipidemic rats[2]. The combination of aqueous extract of *Guazuma ulmifolia* Lamk leaf at dose of 25mg/kgbw and *Curcuma xanthorrhiza* Roxb. at dose of 12.5mg/kgbw for 7 days reduced total cholesterol and LDL significantly[3].

Leaf extract of *Guazuma ulmifolia* Lamk contains flavonoids, phenols, tannin and antosianin[2]. Flavonoids are thought to improve blood lipid profiles in mice dyslipidemia[7,8]. These flavonoids are suspected potential as an antioxidant[9]. Flavonoids can capture free radicals and prevent the process lipid peroxidation in microsomes and liposomes[10]. In vitro, flavonoids work as 3-hydroxy-3-methylglutaryl CoA reductase (HMGCoA reductase) inhibitor, (an enzyme involved in the formation of cholesterol) resulting in decreased intracellular cholesterol synthesis. This causes a decrease in the formation of chylomicrons and chylomicron remnants that reach the liver. This mechanism will stimulate the synthesis of LDL receptors which causes a decrease in LDL levels[11].

Phenol and flavonoid compounds were allegedly having activity antioxidants.[12] In addition to flavonoids, phenolic components others such as tannins are known to have antioxidants[13.14]. An increase in the number of free radicals can cause breakdown of nucleic acids, proteins and lipid membrane so that it can cause liver damage. The heart plays an important role in transport and metabolism of fat including the production of bile for excretion of cholesterol. The liver has system that synthesize and oxidizes fatty acids, converts fatty acids into bile acids and role in lipoprotein metabolism. Liver damage and toxic effects can be interfering with metabolism and excretion cholesterol from the body resulting in an increase in cholesterol levels. The presence of flavonoids that act as anti-oxidants will prevent this effect[15]. Research by Septiana et al., 2021 found that total phenolic can increase antioxidant activity levels[16].

The other mechanisms were: 1). Flavonoids are thought to inhibit the action of the lipase enzyme. Hydrolyzed fat in the body into saturated fatty acids was catalyzed by lipase. fatty acids are converted to acetyl CoA which is a precursor of cholesterol. The presence of flavonoids will suppress the increase in the number of precursors lead to increased levels of cholesterol in the blood[15]. 2). Flavonoids are thought to inhibit the action of HMG-CoA reductase resulting in reduced mevalonate formation from HMG-CoA. Flavonoids can bind cholesterol in the lumen of the duodenum and jejunum [17,18].

Guazuma ulmifolia Lambk leaves are thought to contain compounds tannins and phytosterols that can inhibit sterol regulatory binding element (SRE) on SRE binding (a proteins involved in the process LDL receptor gene transcription). Inhibition of this bond, will reduce the activity of the enzyme 3-hydroxy-3-methylglutaryl CoA reductase (HMGCoA reductase) which reduce cholesterol synthesis[19]. Tannins in ethanolic extract of *Guazuma ulmifolia* inhibits the action of the HMG-CoA, enzyme reductase thereby lowering LDL levels [20].

Ethyl acetate fraction of ethanolic extract of *Guazuma ulmifolia* Lamk at dose of 250 mg/kgbw improved liver histopathological profile. This is thought by antioxidant activity of this extract.

CONCLUSION

The administration of ethyl acetate fraction of ethanolic extract of *Guazuma ulmifolia* Lamk at dose of 250 and 500 mg/kgbw was able to reduce total cholesterol in rats induced by a high cholesterol diet, and improved liver histopathological profile on dose of 250 mg/kgbw. The presumed mechanism is through by antioxidant pathway.

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Conflict of Interest

No conflict of interest is declared.

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