Effect of Ethanol Extract of Lemongrass Plant (Cymbopogon Citratus), Ginger (Zingiber Officinale Rosc.), Pandan Leaf (Pandanus Amaryllifolius), and Cinnamon (Cinnamomum Burmanii) on Albumin and Blood Globulin Levels in Wistar Rats (Rattus Norvegicus) Given High Fat

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Abstract. This study aimed to determine the administration of a mixture of lemongrass ethanol extract (Cymbopogon citratus), ginger (Zingiber officinale Rosc.), pandanleaves (Pandanus amaryllifolius), and cinnamon (Cinnamomum burmanii) against high fat-induced blood albumin and globulin levels of wistar rats. The method wascarried out by dividing the mice with 7 treatment groups and 4 tests, including KO + (Groups given standard feed); KO- (Controls fed with high-fat standard an feed);K+ (Simvastatin-given controls); K1; K2; K3and K4 at a dosage of 100,200,300,and400 mg/Kg BB plant extract mixture of ginger, lemongrass, cinnamon and pandan leaves). The data was analyzed with SPSS software for windows. The results showed that giving a mixture of ethanol extracts of lemongrass plants, ginger, pandan leaves, and cinnamon affected the blood albuminand globulin levels of wistar rats.

Keywords: Albumin and Globulin; ethanol extract; mixing of : lemongrass, ginger, pandan leaves, and cinnamon

1 Introduction

Albumin is the largest protein in the blood, and has an impact on the binding and transport of endogenous and exogenous substances, such as drugs. Since the distribution of the drug in the body is bound by the albumin fraction, this happens. When albumin levels are low, the protein-bound fraction of the drug decreases, which can lead to undesirable pharmacological side effects[1]. According to hospital malnutrition have hypoalbuminemia, or blood albumin levels of less than 3.5 g/dL.[2] Globulins are blood proteins that help the immunesystem and transport steroid hormone systems, lipids, and fibrinogens, which help blood clotting and regulate circulatory function[3]. Alpha globulins, beta globulins, gamma globulins, macroglobulins, and transcobalamins are various types of globulins that operate as circulatory ions, fatty acids in the immune system, and antibodies[4]. Gamma globulin, also known as immunoglobulin playsan important role in this situation[5].

Immunity refers to the body's ability to resist the attack of the disease. When harmful organisms

ICIESC 2022, October 11, Medan, Indonesia

DOI 10.4108/eai.11-10-2022.2325484

invade the body, lymphocytes and antibodies are activated, resulting in an increase in immunity. If the body's immunity isdisturbed, germs can easily attack it[6]. Cholesterol is one of the disorders that wreak havoc on the immune system. The transport of cholesterol begins in the liver, and must be packaged into lipoproteins, such as High Density Lipoprotein(HDL), Low Density Lipoprotein (LDL), and Very Low Density Lipoprotein (VLDL), in order to be disseminated throughout the organs. During the infection phase, lipoproteins play a role in the pathophysiological process of the body's immunological response. Conversely, high LEVELS of HDL will cause the immune system to react negatively[7]. As a result, it is very important to strengthenthe immune system, which can be achieved using secondary metabolite chemicals found in plants.

Chemicals that can boost immunity can be found in medicinal plants that act as an effector on the immune system and regulate immunity. Furthermore, flavonoids have the ability to lower HMG-CoA reductase, an enzyme involved in the formation of cholesterol[8]. Like the sijukkot plant which has flavonoid compounds, it is also able to reduce cholesterol levels in high fat-induced wistar rats[9]. Cinnamon (Cinnamomum burmanii), Lemongrass (Cymbopogon citratus), Ginger (Zingiber officinale Rosc.), and pandan leaves (Pandanus amaryllifolius), are spices commonly used in Indonesia to lower cholesterol, blood sugar levels, as well as as antiviral, antifungal, and antibacterial[10]. Secondary metabolites such as tannins, phenolics, flavonoids, quinones, saponins, monoterpenes, and sesquiterpenes were found during phytochemical screening of cinnamon bark simplisia[11]. The effect of ethanol extracts of lemongrass (Cymbopogon citratus), ginger (Zingiber officinale Rosc.), pandan leaves (Pandanus amaryllifolius), and cinnamon (Cinnamomum burmanii) on cholesterol levels in this study we also reported how albumin albumin and globulin levels in male rats of wistar strain. Measurement of albumin and globulin levels was carried out by folin's phenol method. Mean differences fromeach group were analyzed statistically using Anova One Way using SPSS For Windows software.

2 Material and Method

2.1 Tools

The tools used in this study were beaker glass 200, 100, 50 ml, stirring rod,measuring flask 50 ml, micropipette, drip pipette, injection syringe 3 ml, 3 ml bending syringe, analytical balance, test tube, tube rack, measuring cup 10 ml, spray bottle, animal cage and maintenance, blender, flour sieve, scissors, knives,paraffin tubs, plastic jars, hot plates, evaporation devices vacuum rotary evaporators (Heidolph), drying ovens, Funnel bunchers, Glass jars, Reflux, distillation, funnels, funnels and UV VIS spectrophotometers.

2.2 Material

Lemongrass plants (Cymbopogon citratus), ginger (Zingiber officinale Rosc.), pandan leaves (Pandanus amaryllifolius), and cinnamon (Cinnamomum burmanii) Ethanol 96%, Na2SO4 22.5%, Simvastatin, chloroform, NaOH, Na- CMC (Carboxy Methyl Cellulose), filter paper, folin's phenol reagents, aluminum foil, tissues, and aquades. The test animals used in this study were obtained from the animal house of the Faculty of Mathematics and Natural Sciences, Medan State University.Method. This research started from sampling in Sukarame village, Pematang Tanah JavaVillage, Tanah Java District, North Sumatra Province.

2.3 Sample Preparation

Sample preparation was carried out, as much as ginger, lemongrass, pandan leaves, and cinnamon, each of which was 5kg, 5kg, 3kg, and 1/2kg of the plant then washed thoroughly with water. Plants are dried using an oven with a temperature of 40-500 C for 6-8 hours (H After drying the leaves are grinded toincrease the surface area so that plant powder can be extracted as much as possible, with the hope that the metabolite content of the sample is not damaged.It is then sifted through a 60 mesh sieve after blending.

2.4 Rats Cage Preparation

The test animals used in this study were male rats of the Wistar Strain which were in the range of 2-3 months and the weight of mice ±200grams.

2.5 Plants Extraction

The dry powder from the plant was weighed ±200grams for Ginger and Lemongrass and 50 grams for cinnamon and pandan leaves, respectively. Then it is put into a container and extracted using 96% ethanol by the maceration method. The maceration procedure is repeated three times for a total of 3x24 hours, and filtrates and residues are obtained using a Buchner filter. To make theextract, the filtrate is thickened using a vacuum rotary evaporator. To prevent the extract from clumping, the extract obtained is stored in a glass jar, wrapped in aluminum foil, and stored in a cooler.

2.6 Treatment of Test Animals

In this study, an experimental design in the form of a Complete RandomizedDesign was used. An experiment was conducted on 28 wistar male rats aged 2- 3 months with a body weight of 100 - 200grams were kept in healthy conditions.Adaptation was carried out for 7 days, after which high-fat feeding was carried out consisting of 30grams of quail egg yolk, PTU (Propyltiourasil) 100 mg 0.1%, and water in 1L for 21 days.

Blood draw is carried out from the eyes of rats and then put into a blood collection tube, then the serum is taken by inserting 2 mL of blood into a clean and dry tube then allowed to stand for 15 minutes. Then for 15 minutes, it was centrifuged at a speed of 3000 rpm. The serum is immediately removed with a pipette and placed in another clean and dry tube. This is a clear light yellow coating on the top. The test animals were divided into 7 treatment groups, each treatment consisted of 3 mice. The treatment groups included: Group K0 (+), (negative control) which is rats that are only given standard feed. Group K0 (-),(Negative control), that is, rats fed with standard feed and high-fat feed. K+ group, that is, rats that were given the drug simvastatin orally.

Groups of K1, K2, K3 and K4 rats were given plant ethanol extracts of 100 mg, 200mg, 300mg and 400mg/kg BB, respectively. Intra-oral administration of simvastatin 4.5 mg/kgBB for 30 days the treatment of 7 groups of rats remained fed and drinking. On day 30 all rats measured albumin and globulin levels Measurement of blood albumin and globulin levels was performed by Folin's Phenol method. Mean differences from each group were analyzed statistically using Anova One Way using SPSS For Windows software.

2.7 Blood Draw for Level Testing

For blood draws, it starts by making the rat faint and then blood is drawn using spoits on the eyes. After that it is allowed to stand for ± 1 hour until a separate serum is visible. Then a centrifuge is carried out on the serum. After that, measurements of albumin and globulin levels are carried out.

2.8 Measurement of Albumin and Globulin Levels

Measurements are carried out by Folin's Phenol method with the following steps:

Table 1. Rat Albumin and Globulin Measurement

Material	Standard (Tube 1)	Albumin (Tube 2)	Globulin (Tube 3)
Filtrate Albumin	-	5ml	-
Standar	4ml	-	-
Aquadest	25ml	25ml	-
NaOH 5N	2ml	2ml	2ml
Folin’s Fenol	2ml	3ml	3ml

3 Results and Discussion

3.1 Results

The maceration process is carried out by mixing the plant simplicia powder withethanol solvent and allowed to stand for 3x24 hours with three repetitions whilestirring. Then filtration is carried out using vacuum to separate the filtrate with theamendment. The ethanol extraction process from plant powder produces a viscous extract through a solvent separation process with a Randemen rotary evaporator obtained as follows:

Table 2. Plant Extract Amendments

Plant	Amendments (%)
Ginger	8,9
Lemongrass	16
Sweet skin	13
Pandan leaves	29

Rat Weight Measurement. Measurement of the body weight of rats is carried out once a week. On the 30th day after the experiment was carried out the last weight measurement. Measurements showed that mice fed a diet high in fat, simvastatin, and plant extract doses experienced weight gain as follows table.

Table 3. Rat Weight Loss Before and After Treatment

Treatment	N	Initial weight	Weight After Treatment
KO(+)	4	135±2,8,32	16,25±30,31
KO(-)	4	1321,25±5,31	161,25±8,53
K+	4	119,25±2,21	143,25±1,25
K1	4	145,25±8,84	179,75±14,31
K2	4	150,75±3,3	203,75±5,67
K3	4	140,5±3,69	175,75±12,6
K4	4	137,5±7,55	168±2,3

Information
KO(+) : The group to which the standard feed is given KO(-) : The group given standard feed and high-fat feed
K+ : The group given standard feed, high-fat feed, and simvastatin
K1, K2, K3 and K4 : The group given standard feed, high-fat feed and plant extract doses of 100mg/kg BB, 200mg/kg BB, 300mg/kgBB/400kgBB

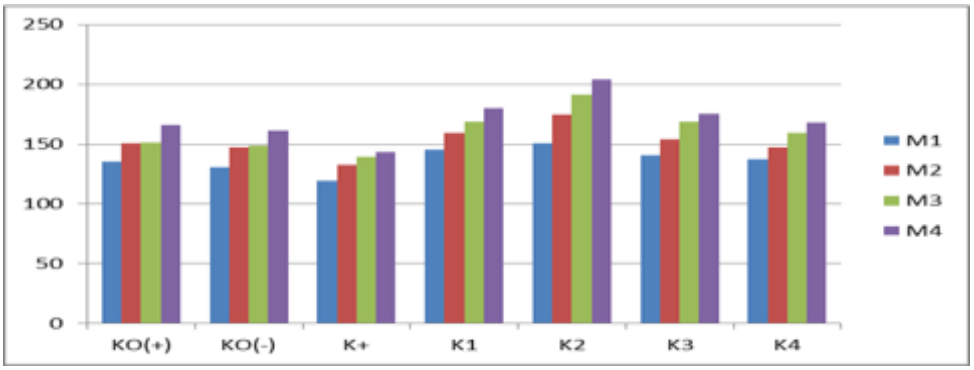

Figure 1. Rat weight gain increased weekly by giving plant extracts in 4doses and high-fat feeding

Measurement of Blood Albumin and Globulin Levels in Rats. The results of serum blood analysis in rats that have been given high-fat feed and extracts of lemongrass, ginger, pandan leaves and cinnamon are shown in the following table:

Table 4. Rat Blood Albumin and Globulin Levels After Treatment

Group	Albumin	Globulin
K0(+)	4,56±0,11	4,1±0,11
K0(-)	5,52±0,55	3,8±0,22
K+	4,72±0,26	3,4±0,24
K1	5,5±0,36	3,5±0,24
K2	5,52±0,20	3,8±0,05
K3	5,75±0,31	4,0±0,21
K4	5,96±0,56	4,3±0,11

Blood Albumin And Globulin Levels In Wistar Rats (Rattus Norvegicus) GivenEthanol Extract Of Lemongrass Plant, Ginger, Pandan Leaves And Cinnamon.

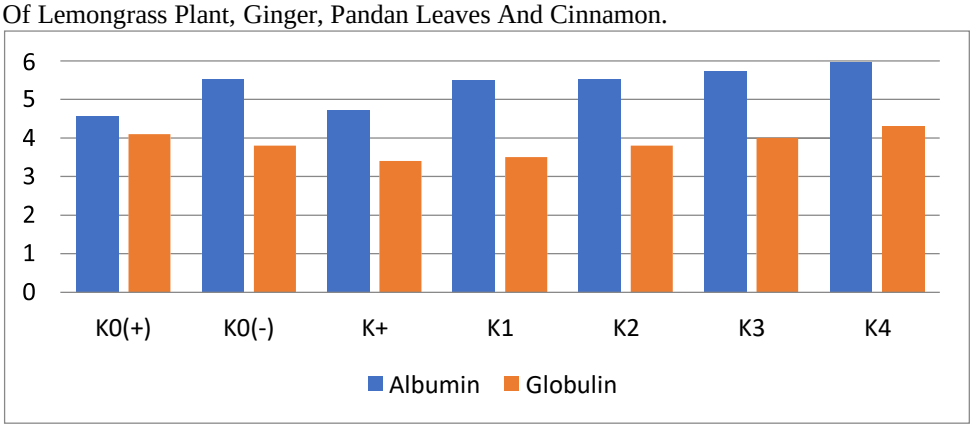

Figure 2. Blood Albumin And Globulin Levels In Wistar Rats (Rattus Norvegicus) Given Ethanol Extract Of Lemongrass Plant, Ginger, Pandan Leaves And Cinnamon

3.2 Discussion

Plant Extraction. Plants of ginger, cinnamon, pandan leaves and lemongrass are extracted by the maceration method using ethanol solvent. The use of ethanol is based on itspolar properties so that it can be used to identify the presence of flavonoid compounds. Extraction by maceration with ethanol solvent is good enough to obtain plant extracts[12]. The maceration process is carried out by mixing the plantsimplicia powder with ethanol solvent and allowed to stand for 3x24 hours withthree repetitions while stirring. Then filtration is carried out using vacuum to separate the filtrate from the amendment. The ethanol extraction process from plant powders produces a viscous extract through a solvent separation process.

Feeding. The feed given to rodents is standard feed and high-fat feed. This is in accordance with the parameters tested, namely cholesterol test and blood globulin albumin test. The standard feed given is 552 sp feed and the high-fat feed given contains quail egg yolk, animal fat with a ratio of 1:5 and propyltiourasil (PTU) 0.1%. The administration of propyltiouracyl aims toinhibit the formation of thyroid hormones so that it can reduce hormone levels in the body. The manufacture of high-fat feed was carried out by adding 50gr ofpuyub egg yolk, 10 ml of animal fat and 0.06gr of PTU then in czech as much as 2 ml to the experimental rats.

Effect of Dosage on Rat Weight Loss. This study used 28 white male rats (Rattus Norvegicus) with a division into7 experimental groups with 4 male rats each. Before the treatment, adaptation was carried out for 7 days then high-fat feed was given for 14 days and then given a dose of plant extract for 14 days. On the 14th day after being given the plant extract, a blood draw was then measured blood albumin and globulin levelsin rats.
Rats' weight at the beginning and end of treatment tended to increase. The first data test with the Shapiro-Wilk normality test with a value of sig>0.05 or H0 was received. Therefore, the initial and final rat body weight data after the treatment were normally distributed. Then data analysis was carried out on the Test of Homogenity of Variances obtained an initial body weight sig value of 0.012 and a final body weight sig of 0.083. Therefore the initial body weight group obtained sig= 0.012 < 0.05 which showed inhomogeneous data and the final body weight

group obtained sig= 0.083>0.05 indicating homogeneous dataor H0 was received. This suggests that dosing in experimental animals had an effect on rat weight gain. This is because one of the extract contents is the gingerplant which contains protein, carbohydrates and energy so that experimental ratsexperienced weight gain[13].

Effect of Dosage on Rat Albumin and Globulin Levels. With more than 60% of the total protein in plasma being albumin, it is the most common protein in human plasma. The binding and transport of endogenous and exogenous molecules, including drugs, is influenced by albumin levels. This is possible because the albumin fraction has an impact on how the drug is distributed throughout the body. When albumin levels are low,the protein-bound fraction of the drug decreases, which can lead to undesirable pharmacological side effects[1]. Globulins are blood proteins that help the immunesystem by transporting steroid hormones, lipids, and the fibrinogen system in thebloodstream and aiding in the regulation of blood circulation[14].
The results of the analysis test using the Shapiro-Wilk normality test showedthat the entire data was normally distributed. Based on the analysis of albumin data obtained, the sig value in the Test of Homogenity of Variances was 0.044. Based on a sig value greater than 0.05, then H0 is rejected. The data from the anova analysis showed f count of 11.229 and f table (0.05) of 2.57 where obtained f count was greater than f table. It can be concluded that the dosing of ethanol extract of lemongrass plants, ginger, pandan leaves, and cinnamon has an effect on increasing blood albumin levels in rats. Analysis of globulin data using Test of Homogenity of Variances obtained sig data of 0.021 < 0.05, H0 was rejected. The anova analysis data showed a calculated f of 11.367 and f of the table (0.05) of 2.57. Where the calculated f value is greater than the f table which shows that ethanol extracts of lemongrass, ginger, pandan leaves and cinnamon affect the stability of blood globulin levels in rats.
Albumin levels in the group that were only given standard feed had normalalbumin levels, then albumin decreased significantly in the treatment group of mice fed high-fat feed. This is because rats have experienced hypercholesterolemia. This decrease is because lipids are involved in the regulation of cytokine levels which causes plasma leakage and extravascular plasma release which results in a decrease in albumin levels[7]. In the administration of dose variations of plant ethanol extract, albumin levels tend toincrease a significant increase was shown at a dose of 400 mg / Kg BB, which was 4.3 g / dL. Normal globulin levels in mice ranged from 2.4-3.9 g/d[15]. Fromthe data showed globulins in mice in each experimental group were above normal. So it can be said that the administration of extracts of lemongrass, ginger, pandan leaves and cinnamon can increase or stabilize globulin levels in the blood.

4 Conclusions and Suggestions

4.1 Conclusions

Based on the research that has been carried out, it can be concluded that: The administration of ethanol extracts of lemongrass plants (Cymbopogonciratus) ginger (Zingeber officinale osc.), pandan leaves (Pandanusamarylifolius), and cinnamon (Cinnamomum burmanii) on albumin and bloodglobulin levels in wistar rats has an influence in raising albumin levels andstabilizing globulin levels above normal. The effective dose of plant ethanol extract in increasing albumin and globulin levels is a dose of 400mg/Kg BB.

4.2 Suggestion

It is recommended that further studies be carried out with more variations in concentration and the period of time carried out to obtain a more effective dose in raising blood albumin and globulin levels using lemongrass plants (Cymbopogon ciratus), ginger (Zingeber officinale Rosc.), pandan leaves (Pandanus amarylifolius), and cinnamon (Cinnamomum burmanii). Given that Carbohydrate metabolism is closely related to diabetes mellitus disease [16]. It is necessary to study the antidiabetic proliferation of these plant mixtures against the inhibition of the enzyme amylase invitro and by using experimental animalsinvivo.

Aknowledgment. I would like to thank the State University of Medan, who funded this research witha contract No: 0229/UN 33.8/PPKM/PD/2022

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