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Test Antioxidant Activity Permot extracts (Passiflora foetida.L) With Parameters MDA (Malondialdehyde) and SGPT Using rat (Rattus Norvegicus) Strain Wistar induced by CCl₄

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Abstract: Activity Test AntioxidantPermotextracts (Passiflorafoetida.L) With Parameters MDA (Malondi aldehyde) Using rat (RattusNorvegicus) Strain Wistar induced by CCl₄ has been conducted. Antioxidants are compounds electron donor (electron donor) or reductant, that are able to scavenge free radicals. The purpose of this research was to determine the antioxidant activity and the effective concentration of Permot extracts (Passiflorafoetida.L). Method of Research is eksperimental study. 18 rats divided into 6 groups. Normal Group, Control group which were administrated by intraperitonial with CCl₄ dose 1,0 ml/kgwb and Na-CMC. Group treatment by Vitamin C, Group Treatment by Permot extracts dose 100 mg/kgwb, 200 mg/kgwb and 400 mg/kgwb. The treatment of the all groups was conducted for 7 days. MDA (Malondialdehyde) was measured by spectrophotometer UV-Vis with a wavelength of 532 nm and SGPT measured by human analyzer. The analysis data between the all groups using One Way Anova, and continue by using Duncan comparison test after vitamin C and permot extract administration among the treatment groups was MDA level and SGPT. The results of research are permotekstracts has antioksidant activity in hepatic rat and effective dose is 400 mg/kgwb.

Key words: MDA (Malondialdehvde), SGPT, Permot (Passiflorafoetida), Antioksidant, CCl₄, hepatic rats.

I. Introduction

Degenerative disease generally occurs due to damage to the cell, tissue fat, protein, immune system , and DNA caused by various factors both naturally occurring, exposed to radiation, or by chemical substances which are carcinogenic. There are various theories that can explain the cause of degenerative diseases, one of which is the theory that free radical reactions. According to this theory due to the onset of degenerative diseases caused by hydroxyl radicals in biochemical mechanisms that occur in the body¹.

One way of preventing the formation of free radicals is to use nutrients that can act as antioxidants such as vitamin E, carotene, vitamin C, as well as other drugs that can capture these radicals. However, synthetic antioxidants often raise concerns about the side effects resulting in long-term use. Bioactive compounds that can be used as an antioxidant phenols are compounds such as flavonoids oligoresveratrol, and phenolic acids¹

Today a large number of drugs developed from a variety of medicinal plants that have therapeutic potential privileges and fewer side effects². One of the plants that contain medicinal compounds that permot (Passiflorafoetida L). The main component of this plant contain alkaloids, phenols, flavonoids and components

cyanogenic glycosides³ and passifloricin, polyketida and alpha- pyrones⁴ which contained bioactive compounds that can be used as an antioxidant .

Based on the description above, this study uses the parameter MDA (malondialdehyde) and SGPT (serum glutamic pyruvic transaminase) to test the antioxidant activity of permot (Passiflorafoetida L.) in rats, in order to add a reference the use of antioxidants from plants to prevent degenerative diseases.

II. Research Method

Material and Tools

 $\label{eq:matrix} Materials are Distilled water , ascorbic acid , 10 \% trichloroacetic acid (TCA) , 0.67 \% tiobarbiturat acid (TBA), 10 \% EDTA , ethanol, permot (Passiflorafoetida L), 1 N HCl , malondialdehyde (MDA), sodium sulfate 1mol / L. Tools are UV - Vis spectrophotometer (Thermoo , Russia), ice bath , cyclo mixer, a set of centrifuges , human analyzer for measured SGPT, syringe, the water bath .$

Preparation of extracts

Herbapermot extracted by maceration at room temperature using ethanol for 3×24 hours. Furthermore filtered, the residue was added ethanol and macerated again until the perfect extraction. The filtrate obtained was collected and concentrated by vacuum evaporator, so that the ethanol extract obtained thick

Standarisation of Ekstract permot

Standardization was done by determining the parameters of non -specific (determination of water content, ash content, the determination of the metal boundary) and Determination spesifik (organoleptic parameters, including shape, color, taste and smell)

Preparation of animal experiments

18 rats strain wistar, 2-3 months with a body weight of 100-200 g imported from Airlanggauniversity. Before treatment, the test animals adapted for 1 week with standard feed and drinking water ad libtium .and have eticalclereance in medical of faculty Hasanuuddin University.

Treatment of Animal experiments

The rats were divided into six groups. Induction by carbon tetrachloride (CCl4) at a dose of 1.0 mL / kg intraperitoneally in 5 groups except the normal group and left for 1 x 24. The treatment groups were given Na.CMC 1 % w / v, were given the extract at a dose of 100 mg / kg, 200 mg / kg and 400 mg / kg. vitamin C were given a dose of 100 mg / 70kg BB for 7 days. Furthermore, blood rat was taken from each treatment group for measurement of MDA (Malondialdehyde) and SGPT (Serum Glutamic Pyruvic transaminase/ alanine transaminase).

Measured of MDA and SGPT

Treatment group	Average
Normal Group	42,644±17,356
Control Group	84,023 ±17,695
Extract 100 mg/Kgwb	56,437± 3,982
Extract 200 mg/Kgwb	51,839±18,992
Extract 400 mg/Kgwb	36,897±10,345
KontrolPembanding	67,931±19,199

Table 1. MDA level of Extract ethanol permot (passiflorafoetida L.) with treatment 7 days

KelompokPerlakuan	Average Induced byCCl ₄	Average after treatment 7 days
Normal Group	64,000 ± 8,638	61,993 ± 8,951
Control Group	$184,233 \pm 3,855$	78,633 ± 7,023
Extract 100 mg/Kgwb	$124,267 \pm 18,020$	67,400 ± 3,279
Extract 200 mg/Kgwb	$101,067 \pm 8,832$	57,167 ± 6,668
Extract 400 mg/Kgwb	$116,200 \pm 10,987$	45,600 ± 5,820
KontrolPembanding	107,433 ±8,016	53,500 ± 17,135

Table 2. SGPT levels

n = 3

Blood rats from the retro orbital sinus taken carefully using a syringe and use anticoagulants (EDTA) to prevent clotting. Blood was placed in tubes centrifuges were clean and dry, and then centrifuged for 10 minutes at 10,000 rpm. After separating the upper layer (plasma) that is colored translucent yellowish taken for measurement of MDA (Malondialdehyde) and SGPT

Determination of MDA levels being done with TBA test . Supernatant as much as 100 mL eppendorf put in , plus 550 mL of distilled water , 100 mL of TCA 10 %, 250 mL of 1 N HCl and 100 mL of Na - thio . After that, homogenized and centrifuged at 500 rpm for 10 minutes . The supernatant was taken and incubated in a 100 $^{\circ}$ C water bath for 30 minutes. The supernatant was left at room temperature and then measured the absorbance at the maximum wavelength of 532 nm⁴. Absorbance was then plotted on a linear regression equation obtained in order to obtain the levels of MDA .SGPT was measured by human analyzer. The analysis data between the all groups using One Way Anova, and continue by using Duncan comparison test after vitamin C and permot extract administration among the treatment groups was MDA level and SGPT

Result and Discussion

Disscusion

Antioxidants can be derived from plant material is one permot(passiflorafoetida L.). Where the plant permot (Passiflorafoetida L), namely fruits, seeds, and leaves contain a chemical component that is unstable and hydrocyanic acid lactone. In addition, permot also contains alkaloids, steroids, saponins and flavonoids. The main component of this plant contain alkaloids , phenols, flavonoids and components cyanogenic glycosides ² and passifloricin, polyketida and alpha - pyrones (Echeverri et al., 2001) which contained bioactive compounds as antioxidants which can inhibit the oxidation reaction. Therefore, the research conducted testing of ethanol leaf extract antioxidant permot(passiflorafoetida L.) with MDA parameters (Malondialdehyde)

This study used inducer of carbon tetrachloride (CCl4), because carbon tetrachloride (CCl4) is a xenobiotic that is commonly used to induce lipid peroxidation and poisoning . Constituent fatty acids of cell membranes , especially long- chain fatty acids polyunsaturated (PUFAs) could be vulnerable to free radicals . According to Jeon et all . Number of CCl4 induced proportional to the reduced amount of PUFAs in the endoplasmic reticulum membrane phospholipids. In the endoplasmic reticulum of the liver CCl4 is metabolized by cytochrome P450 2E1 (CYP2E1) into free radicals triklorometil (CCl3*). Triklorometil with oxygen to form radicals which can attack triklorometilperoxi endoplasmic reticulum membrane lipids at speeds in excess of free radicals triklorometil . Furthermore triklorometilperoxi cause lipid peroxidation that disrupts Ca2 + homeostasis , and ultimately cause cell death. Administration of CCl4 in high doses can damage the endoplasmic reticulum , disrupt the process of oxidation, causing swelling of the liver so that the liver weight be increased, and long-term administration can lead to necrosis sentrilobular and fatty degeneration in the liver.

Enzyme ALT is the best indicator of liver damage in the notice. In mild liver cell disruption it will seep into the cytoplasmic enzyme, especially serum alanine aminotransferase enzyme. Therefore, the levels of ALT enzyme is unique and specific to liver cell damage so it is suitable as a test to determine the presence of liver dysfunction can be caused by free radicals, although in a minor degree. In mice, the normal value of SGPT levels ranged from 19.3 to $68.9 \text{ U}/\text{L}^5$.

Absorbance of MDA (Malondialdehyde) measured by UV - Vis spektofotomeri method. This method is the most widely used method to measure the presence of free radicals and lipid peroxides, have a fairly high sensitivity, easy to apply to a wide range of samples. This method is based on a complex reaction between MDA (Malondialdehyde) and TBA (tiobarbiturat acid) pink then then measured in a spectrophotometer at 532 nm wavelength.

Based on statistical calculations showed that the results of measurements for the normal group showed significantly different with induction group. This shows that the oxidation reaction occurs with an increase in absorbance of MDA in group induction. For the normal group showed a nonsignificant results with comparison group and the test group extract dosage of 100 mg / kgbw, 200 mg / kgbw, and 400 mg / kgbw. This shows that the comparator and the test preparation extracts have antioxidant effects. For the comparison group and the test extract 100 mg / kg did not show good antioxidant activity. For a group of test extract 200 mg / kg showed significant results against the induction, this shows the test extract 200 mg / kg gave good antioxidant effects. For a group of test extract 400 mg / kgbw body weight showed highly significant results of the induction group, this suggests that the test extract 400 mg / kgbw body weight provide excellent antioxidant effect

Conclusion

The results of research are permotekstracts has antioksidant activity in hepatic rat and effective dose is 400 mg/kgbw.

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