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Decreasing the SGPT level of male wistar rats induced by gentamicin with purslane ethanol extract

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Abstract:

Antibiotics from the aminoglycoside group, such as gentamicin, are frequently used for the infection therapy of gram-negative bacteria including Salmonella typhi. Some studies show that gentamicin can cause hepatotoxicity and increase the level of SGPT (serum glutamic pyruvic transaminase). The purpose of this research is to figure out the decreasing SGPT level of male Wistar rats induced by gentamicin with the purslane ethanol extract. This research employed a post-test only control group design, utilizing 25 male Wistar rats divided into 5 groups. The normal control group (NorG) was without any treatment, while the positive control (PG) was intraperitoneally injected with gentamicin 60 mg/kg of rat body weight. The treatment groups consisted of P1, P2, and P3 were intraperitoneally injected with gentamicin at the dosage of 60 mg/kg of rat body weight for 7 days and then administered with purslane ethanol extract respectively at the dosage of 200, 300, and 400 mg/kg of rat body weight per oral for 7 days. The analysis on the SGPT level was conducted with the IFCC modification method using chemistry analyzer. One way ANOVA test shows that there were significant differences in SGPT levels among groups. LSD post hoc test shows that purslane ethanol extract at the dosage of 400 mg/kg of rat body weight significantly decreased the SGPT level ($p<0.05$) when compared to the positive control group. The administration of common purslane ethanol extract at the dosage of 200, 300, and 400 mg/kg of rat body weight can decrease the SGPT level of male Wistar rats induced by gentamicin.

Keywords: SGPT, Purslane ethanol extract, Gentamicin.

INTRODUCTION

Antibiotics from the aminoglycoside group, such as gentamicin, are frequently used for the infection therapy of gram-negative bacteria including Salmonella typhi. Some studies show that gentamicin can cause hepatotoxicity and increase the level of SGPT (serum glutamic pyruvic transaminase).[1] SGPT or ALAT (alanine aminotransferase) is an enzyme specifically produced by liver cells, in which SGPT is released when the liver is in damaged condition. One medicine causing the significantly increasing SGPT level is gentamicin.[2, 3] To decrease the SGPT level, the administration of high antioxidant substances, such as purslane plant.[4, 5]

Studies on the utilization of common purslane extract have been conducted by researchers throughout the world, including in Indonesia. The research conducted by Ahangarpour *et al.* (2018) shows that the common purslane extract has a protective effect on the pancreas by decreasing hypoglycemic activity and insulin resistance.[6] The research conducted in Indonesia by Saptaningtyas et al. (2020) also shows that common purslane

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ethanol extract can improve the level of high-sensitivity CRP (Hs-CRP) with the total score of male Wistar rats' renal tubular degeneration induced by gentamicin is equal to that in normal condition.[7]

The increasing SGPT in the male Wistar rats' serum induced by gentamicin indicates an abnormality in their liver. Oxidative stress is a key mechanism responsible for liver damage resulting from the administration of gentamicin which results in the increasing level of SGPT.[8, 9] The antioxidants contained in purslane extract have an important role against oxidative stress and minimizing the negative impacts of oxidative stress. High antioxidant activity in common purslane extract has been investigated since containing gallotannin, omega-3 fatty acid, ascorbic acid, tocopherol, kaempferol, quercetin, and apigenin.[10] Gentamicin induces increasing oxidative stress and free-radical production and suppresses the antioxidant defense system in the liver. The increasing lipid peroxidation results in lipid membrane damage and causes hepatoxic necrosis triggering the increasing level of SGPT. The antioxidant substances contained in common purslane extract suppress the lipid peroxidation and increase the antioxidant defense system in the liver by increasing the glutathione content and improving the superoxidase dismutase activity.[2, 4, 11, 12]

The purpose of this research is to figure out the effect of purslane ethanol extract on SGPT level of male Wistar rats induced by gentamicin. The utilization of purslane extract in decreasing the SGPT level has never been reported before. Thus, it is expected that the results of this research may become a candidad for phytotherapy against the liver damage caused by gentamicin.

MATERIAL AND METHOD

Time and Place of Research

The research was conducted after obtaining ethical approval from the Ethics Committee of FK UNISSULA (Decree no. 285/ VII/2018/Bioethics Committee).This research was conducted from April to September 2020. The place of this research was carried out at clinical pathology laboratory, Medical Laboratory Technology, Universitas Muhammadiyah Semarang.

Research Design

This research used a completely randomized with post-test only control group design. 25 samples of male Wistar rats aged 8-12 weeks ah the body weight of 150-200 grams. The samples were divided into 5 treatment groups respectively consisting of 5 male Wistar rats. The control group was the normal group (NorG) which was only treated with distilled water, while the positive control group (PG) was a group intraperitoneally injected only with gentamicin at the dosage of 60 mg/kg of rat body weight for 7 days. The treatment groups respectively consisted of P1, P2, and P3 intraperitoneally injected with gentamicin at the dosage of 60 mg/kg of rat body weight for 7 days and then administered with purslane ethanol extract at the dosage of respectively 200, 300, and 400 mg/kg of rat body weight per oral for 7 days.

Materials and Tools

Materials and tools needed in this research are purslane, ethanol (96%, MERCK), male Wistar rats' serum, gentamicin, SGPT reagent (Diasys), rotary evaporator, erlenmeyer, tube test, and chemistry analyzer (Mindray BA-88A).

Research Procedures

Purslane Extraction

Purslane plants were washed thoroughly and dried in an oven at 550 C, then grind them well. One liter of 96% ethanol was added to 100 grams of grinded purslane, mixed, and soaked overnight. The supernatant of the 96% ethanol purslane was taken to rotary evaporator to be evaporate until we get the purslane extract.

Research termination

The research termination was performed on day 8 for NorG and day 15 for PG, P1, P2, and P3 by taking their blood through the orbital sinusitis of Wistar rats' eyes. The obtained blood was collected in the red vacutainer, waited until frozen, and then centrifuged to take the serum.

SGPT and Statisitical analysis

The serum was then analyzed based on their SGPT level with the modification method of IFCC (International Federation of Clinical Chemistry and Laboratory Medicine) using chemistry analyzer in Unit/liter (U/L). The data resulted from the examination of SGPT level were the analyzed using SPSS with One way ANOVA and LSD post hoc test ($p<0.05$.

RESULTS AND DISCUSSION

The data obtained from the mean results of SGPT level examination on the influence of common purslane ethanol extract on male Wistar rats induced by gentamicin were presented in Figure 1.

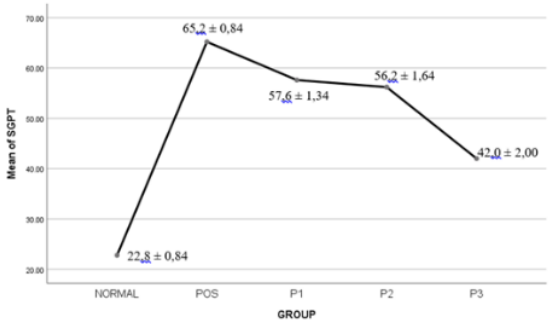

Figure 1. Chart of SGPT Level (Mean ± SD)

Based on Figure 1, it can be seen that the highest mean SGPT level was in positive group (PG), while the lowest was in NorG. The increasing SGPT level significantly happened in PG. The highest decreasing mean SGPT level was in P3 respectively followed by P2 and P1. The decrease was not yet equal with the average SGPT level in NorG, however, has reached the reference value of SGPT level, that is, 18-45 U/L.[13] The results of ANOVA test show that the value of $p < 0.05$ so that it was considered having a significant difference. Meanwhile, post hoc test was then performed to know the differences among groups.

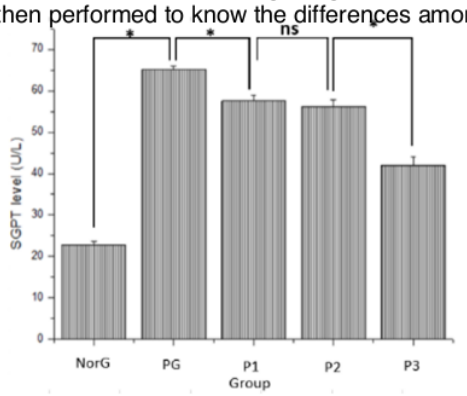

Figure 2. POSTHOC Test on SGPT Level

Figure 2 show that purslane ethanol extract was able to decrease the SGPT level of male Wistar rats induced by gentamicin. SGPT is enzyme catalyzing the amino group changes to form the oxaloacetic metabolism in liver. SGPT can be found in the cytosol of hepatocytes. When liver injury occurs, SGPT is released from injured liver cells and causes a significant increase in serum SGPT activity. The increasing SGPT level may occur due to the use of certain medicine, such as gentamicin.[14, 15] Gentamicin at the dosage of 60 mg/kg of rat body weight induced intraperitoneally causes the increasing SGPT level when compared with that in normal control group. The increasing SGPT level of male Wistar rats induced by gentamicin was in accordance with the result of research previously conducted by Aboubakr in 2016 and Mishra in 2018.[9, 11] Gentamicin is antibiotics from aminoglycoside group which may result in hepatotoxicity. Hepatotoxicity or liver damage has been confirmed with the increasing SGPT level and eventually causing the mitochondrial disfunction. Gentamicin enter the cell by endocytosis or kation channel. Endocytosis transfer gentamicin into endoplasmic reticulum and lysosome, causing cathepsin release, and trigger cell death.[16, 17] Gentamicin increases the production of superoxide anion, hydrogen peroxide, and hydroxyl radical produced by mitochondria. The formed free radicals cause the peroxidase of phospholipid membrane, DNA damage, and protein denaturation.[9, 11, 18]

Gentamicin is covalently related to intracellular protein which possibly decreases the ATP and actin disorder may occur. The destruction of actin fibril on the surface of hepatocyte causes the inflaming cells and breaking cell membranes. Toxin in hepatocyte induces the oxidative stress of cell organelles, such as endoplasmic reticulum and mitochondria possibly causing necrosis or apoptosis.[19] Hepatotoxic metabolite oxidates the thiol protein group and results in reactive oxygen species (ROS). The increasing production of ROS influences the permeabilities of mitochondrial membranes which also then influences the ATP synthesis and expulsion of protein between membranes then triggering necrosis or apoptosis.[20, 21, 22] The research results show that purslane ethanol extract at the dosage of respectively 200, 300, and 400 mg/kg of rat body weight influenced the SGPT level of male Wistar rats induced by gentamicin. Purslane ethanol extract at the dosage of 400 mg/kg of rat body weight could significantly decrease the SGPT level. Some studies have shown the utilization of purslane extract as antioxidant, anti-inflammation, antitumor and anti-bacteria.[23, 24, 25, 26, 27] The chemical content of common purslane ethanol extract includes alkaloid, flavonoid, terpenoid, vitamin, and mineral. The antioxidant content in purslane ethanol extract against free radical formed by reacting with the free radicals stabilizes free radicals and changes free radicals into non-reactive compounds.[28, 29] The antioxidant binds electrons from free radicals and inhibits the chain reaction from the formation of free radicals. Flavonoid and alkaloid in antioxidant react as antioxidant donating hydrogen atoms to free radicals. Vitamin and mineral have the function as cell maintainer by increasing phagocytosis and suppress the occurring inflammation.[9, 20, 30].

CONCLUSION

The administration of purslane ethanol extract at the dosage of 200, 300, and 400 mg/kg of rat body weight can decrease the SGPT level of male Wistar rats induced by gentamicin.

AUTHORS' CONTRIBUTIONS

All authors contributed equally to this work.

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DATA AVAILABILITY STATEMENT

The utilized data to contribute to this investigation are available from the corresponding author on reasonable request.

DISCLOSURE STATEMENT

The views and opinions expressed in this article are those of the authors and do not necessarily reflect the official policy or position of any affiliated agency of the authors. The data is the result of the author's research and has never been published in other journals.

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