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Hepatoprotective Potential Of Temu Kunci Ethanol Extract In Increasing Glutathione In Paracetamol-Induced Rats

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Abstract–Liver damage induced by hepatotoxic compounds such as N-acetyl-p-aminophenol (N-APAP) remains a significant global health concern. Hepatotoxicity caused by N-APAP, particularly in cases of overdose or prolonged use, can lead to a reduction in glutathione levels in the liver, resulting in oxidative stress and cellular damage. This condition contributes to an increased incidence of liver diseases, including toxic hepatitis and acute liver failure, which can lead to serious complications and even death. Although various pharmacological therapies, such as N-acetylcysteine (NAC), are available to address N-APAP-induced liver damage, limitations such as side effects, high costs, and variable efficacy have driven the search for safer and more affordable therapeutic alternatives. Therefore, research on natural hepatoprotective agents, such as extracts from the rhizomes of fingerroot (Boesenbergia rotunda), has become increasingly relevant in addressing this issue. The aim of this study was to evaluate the hepatoprotective potential of ethanol extract from fingerroot rhizomes (Boesenbergia rotunda) in protecting the liver from N-acetyl-p-aminophenol (N-APAP)-induced damage. Specifically, this research focuses on measuring glutathione levels as a key indicator of the liver's protective mechanism against oxidative stress. In this study, 30 male White rats were randomly divided into six groups, each consisting of five rats. All rats were orally administered the extract at predetermined doses for 10 consecutive days, except for the negative and positive control groups. On the 10th day, all groups (except group 1) were treated with paracetamol (800 mg/kg body weight) to induce liver damage. Twenty-four hours after induction, the rats were sacrificed and liver samples were collected. Glutathione levels were measured using the ELISA immunosorbent assay. Statistical analysis showed that administering the ethanol extract of Fingerroot rhizomes (EERTK) for 10 days prior to N-APAP induction significantly increased the average antioxidant enzyme GSH-Px levels in the liver homogenate of rats in the EERTK 250 mg/kg BW (43.36 ± 1.34 U/mg protein), EERTK 500 mg/kg BW (55.70 ± 2.15 U/mg protein), and 750 mg/kg BW (53.14 ± 2.57 U/mg protein) group compared to the negative control group (N-APAP 800 mg/kg BW), which had the lowest average antioxidant enzyme GSH-Px level (33.86 ± 3.52 U/mg protein).

Keywords: N-APAP; Oxidative Stress; Gluthatione; Boesenbergia rotunda

1. INTRODUCTION

The liver is a vital organ that plays an important role in the metabolism, detoxification, and regulation of various physiological functions of the body. As a center for toxin processing, the liver is often exposed to various toxic compounds, both exogenous and endogenous, produced by metabolism.(Dey et al., 2013; Mahmood & Hackl, 2024). One of the compounds known to cause hepatotoxicity is N-acetyl-p-aminophenol (N-APAP), also known as paracetamol. Although Paracetamol is a commonly used analgesic and antipyretic drug, overdose or long-term use can cause liver damage through oxidative stress mechanisms and decrease glutathione levels, the main antioxidant compound in liver cells. This condition triggers the accumulation of free radicals that damage cell membranes, disrupt liver function, and are potentially life threatening(Babyvanitha & Jaykar, 2023; Stavrakeva et al., 2024).

To protect the liver from damage, increasing attention is being paid to the use of natural substances as hepatoprotective agents.(Shikh et al., 2024). One of the plants with great potential

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in this regard is Temu Kunci (Boesenbergia rotunda), a traditional plant widely used in herbal medicine. Temu Kunci rhizome contains various bioactive compounds, such as flavonoids, pinostrobin, and boesenbergin, which are known to have antioxidant and anti-inflammatory activities. This makes it a promising candidate for the development of herbal-based hepatoprotective therapy, especially in the context of liver damage induced by hepatotoxic compounds such as N-APAP(Gonfa et al., 2024; Harahap et al., 2023; Sugiharto et al., 2024).

Liver damage induced by hepatotoxic compounds such as N-acetyl-p-aminophenol (N-APAP) continues to be a significant global health problem. Hepatotoxicity caused by N-APAP, especially in cases of overdose or long-term use, can trigger a decrease in glutathione levels in the liver, resulting in oxidative stress and cellular damage.(Hionides-Gutierrez et al., 2024; Tsai et al., 2024). This condition contributes to an increased incidence of liver diseases, including toxic hepatitis and acute liver failure, which can lead to serious complications or even death. Although various pharmacological therapies, such as N-acetylcysteine (NAC), have been used to treat N-APAP-induced liver damage, limitations, such as side effects, high costs, and varying effectiveness, have prompted the search for safer and more affordable therapeutic alternatives. Therefore, research on natural hepatoprotective agents, such as Temu Kunci rhizome extract (Boesenbergia rotunda), is becoming increasingly relevant to address this problem.(Jaeschke & Ramachandran, 2024; Mahajan et al., 2024).

This study aimed to evaluate the hepatoprotective potential of the ethanolic extract of Temu Kunci rhizome (Boesenbergia rotunda) in protecting the liver from damage induced by N-acetyl-p-aminophenol (NN-APAP). This study specifically focused on measuring glutathione levels as the main indicator of liver protection against oxidative stress.(Sarkar et al., 2024). By identifying and testing the biological activity of the bioactive compounds in Temu kunci, this study is expected to provide scientific evidence regarding its effectiveness as a hepatoprotective agent. The results of this study are expected to not only broaden the insight into the therapeutic benefits of Temu Kunci, but also support its development as a safe and efficient herbal therapy candidate for the prevention and treatment of liver damage caused by toxic compounds.(Ahmed & Khamees, 2024; Babu et al., 2024; Prottasha et al., 2024).

Although a number of studies have explored the antioxidant activity and hepatoprotective potential of herbal plants, studies specifically evaluating the effects of the ethanol extract of Temu Kunci rhizome (Boesenbergia rotunda) on liver damage induced by N-acetyl-p-aminophenol (NN-APAP) are still very limited. Most previous studies have only focused on in vitro activity or used different biological models; therefore, empirical evidence regarding the protective mechanism associated with increased glutathione levels in the liver in animal models

has not been sufficiently revealed. In addition, most studies are still general in nature and have not explored the relationship between the specific bioactive compounds of Temu Kunci and its hepatoprotective ability. Therefore, there is an urgent need to fill this gap in the literature with more focused and comprehensive studies, which can strengthen the scientific basis for the use of Temu kunci as a natural hepatoprotective agent. This study offers a new contribution by exploring the potential of the ethanol extract of Temu Kunci rhizome (Boesenbergia rotunda) as a hepatoprotective agent by increasing glutathione levels in the liver. This approach provides a new perspective in the study of hepatoprotective herbs, considering that most previous studies have not specifically linked biochemical activities to liver protection against damage induced by N-acetyl-p-aminophenol (NN-APAP). The novelty of this study lies in the combination of biochemical evaluation and pharmacological observations in one series of studies, which has the potential to open up new opportunities in the development of natural ingredient-based therapies. In addition to providing a strong scientific justification for the therapeutic benefits of Temu Kunci, the results of this study are expected to provide safer, more economical, and more effective solutions for the prevention and treatment of liver damage, thus contributing significantly to the development of hepatology and herbal-based therapies.

2. RESEARCH METHODS

Research Design

This study used a completely randomized experimental design (Randomized Complete design) with male white rats (Rattus norvegicus), which was divided into six treatment groups. Each group was administered a different treatment to evaluate the hepatoprotective effect of the ethanol extract of Temu Kunci rhizome (Boesenbergia rotunda) against liver damage induced by N-acetyl-p-aminophenol (paracetamol). The control and treatment groups were tested by administering Temu Kunci extract at various doses, followed by induction with paracetamol on the 10th day to assess the biochemical response, especially the increase in glutathione levels in the liver.

Research Sample

The sample used in this study was the rhizome of Temu Kunci (Boesenbergia rotunda) obtained from an area known for its quality. After identification and verification by botanists, the rhizome of Temu Kunci was selected as the raw material for extraction because of its bioactive compound content, which is known to have hepatoprotective potential.

Ethanol Extract Preparation Procedure

The ethanol extract of Temu Kunci rhizome was prepared using the maceration method. The washed and dried Temu Kunci rhizomes were then cut into small pieces and weighed up to 500 g. The rhizome pieces were then soaked in 2 liters of 96% ethanol and left for 72 h with shaking every 12 h. After the maceration time was completed, the filtrate obtained was separated and evaporated using a rotary evaporator to remove the ethanol solvent until a thick extract was obtained. The extract was stored at 4°C until further use(Debnath et al., 2024).

Making Ethanol Extract Suspension of Temu Kunci

The following steps were taken to prepare the EERTK suspension, which was filled with 250 mg of ERRTK, and 2 drops of Tween 20 were added. Furthermore, while being crushed until homogeneous, a 0.5% CMC-Na suspension was gradually added. After that, it was placed into a 10 mL measuring flask. CMC-Na (0.5%) was used to add the volume to the marked line. The same process was used to prepare EERTK suspensions at doses of 500 and 750 mg/kg.

Test Animals and Treatment Groups

In this study, 30 male White rats were randomly divided into six groups, each consisting of five rats. The treatment groups were as follows:

1. **Group 1 (Positive Control)**: No N-APAPU treatment was given.
2. **Group 2 (Negative Control)**: Paracetamol induction at 800 mg/kg BW on the 10th day as a negative control.
3. **Group 3 (Acetylcysteine)**: Acetylcysteine 200 mg/kg BW was induced with paracetamol 800 mg/kg BW on the 10th day.
4. **Group 4 (Kemu Kunci Extract 250 mg/kg BW)**: given 250 mg/kg BB of Temu Kunci extract, then induced with 800 mg/kg BB of paracetamol on the 10th day.
5. **Group 5 (Kemukin Extract 500 mg/kg BW)**: Temu Kunci extract 500 mg/kg BB was induced with paracetamol 800 mg/kg BB of paracetamol on the 10th day.
6. **Group 6 (Ketum Kunci Extract 750 mg/kg BW)**: 750 mg/kg BB of Temu Kunci extract was induced with 800 mg/kg BB of paracetamol on the 10th day.

All rats were orally administered the extract at a predetermined dose for 10 consecutive days, except for the negative and positive control groups. On the 10th day, all groups (except group 1) were treated with paracetamol (800 mg/kg BW) to induce liver damage. After 24 hours post-induction, rats were sacrificed for liver sampling.

Testing of Extracts on Test Animals

After paracetamol treatment and induction, liver tissue from each group of test animals was analyzed to measure glutathione peroxidase (GSH-Px) levels, which were used as

parameters to assess the ability of Temu Kunci extract to protect the liver. Glutathione peroxidase (GSH-Px) was measured using ELISA.

Data Analysis Techniques

The data obtained from this study will be analyzed using one-way analysis of variance (ANOVA) to test for significant differences between treatment groups. If the ANOVA results show significant differences, further testing using Tukey's test will be carried out to determine which groups have significant differences. All statistical analyses were performed at a significance level of $P < 0.05$.

This study aimed to evaluate the effectiveness of Temu Kunci extract in increasing glutathione peroxidase (GSH-Px) levels and protecting the liver from damage caused by oxidative stress due to paracetamol, and to provide scientific evidence regarding the hepatoprotective potential of this plant.

3. RESULTS AND DISCUSSION

Measurement of GSH-Px enzyme in rat liver homogenate aims to analyze the effect of varying the dose of ethanol extract of T. kunci rhizome in rats induced by N-APAP at a dose of 800 mg/kg BW on the value of GSH-Px enzyme in rat liver homogenate, measurements were carried out using an ELISA kit by chopping the rat liver into small pieces and then rinsing with cold PBS pH 7.4 to completely remove excess blood. The rat liver pieces were then weighed and homogenized with PBS at a ratio of 1:9, using a homogenizer glass on ice to further break down the cells. Furthermore, sonication was performed using a freeze-thaw cycle. The homogenate was centrifuged for 5 min at 5000 rpm to obtain the supernatant, which was then measured using an ELISA kit. Enzyme-linked immunosorbent assay (ELISA) is a quantitative method used to measure peptide, protein, antibody, and hormone levels based on the principle of antigen-antibody binding. The average levels of GSH-Px enzymes in liver homogenates are shown in Table 1.

Table 1. Average results of GSH-Px enzyme levels in rat liver tissue.

Group	GSH-Px (U/mg protein) ± SD
Normal Control (P1)	58.01 ± 1.24ad
N-APAP 800 mg/kg BW (P2)	33.86 ± 3.52c
Acetylcysteine 200 mg+N-APAP (P3)	54.60 ± 2.19ad
EERTK 250 mg/kg BW+N-APAP (P4)	43.36 ± 1.34b
EERTK 500 mg/kg BW+N-APAP (P5)	55.70± 2.15ad
EERTK 750 mg/kg BW+N-APAP (P6)	53.14 ± 2.57ad

Description: a = significantly different from the negative control group ($p < 0.05$), b = significantly different from the normal control group, positive control, and significantly different from the negative control ($p < 0.05$); c = significantly different from the normal control group and positive control group ($p < 0.05$), d = not significantly different from the positive control group ($p > 0.05$).

The results of the one-way ANOVA statistical analysis showed a significance value of 0.001 ($p < 0.05$), indicating a difference in the average levels of GSH-Px enzyme in rat liver homogenates between the treatment groups. The average level of the antioxidant enzyme GSH-Px in the normal control group was58.01 ± 1.24 U/mg protein. The average value is the standard value of GSH-Px enzyme in normal (healthy) rats. Statistical analysis revealed that the average value of the GSH-Px enzyme in the normal control group was higher than that in the negative control group. The positive control group (acetylcysteine 200 mg) had an average GSH-Px enzyme level of 54.60 ± 2.19 U/mg protein and the negative control group (N-APAP 800 mg/kg BW) had the lowest average GSH-Px enzyme level of 33.86 ± 3.52 U/mg protein. These results suggest that administration of N-APAP 800 mg/kg BW can cause oxidative stress, which is characterized by low levels of the antioxidant enzyme GSH-Px in the negative control group(Coelho et al., 2023).

The results of the one-way ANOVA analysis showed that administration of N-APAP induced oxidative stress and resulted in low levels of the antioxidant enzyme GSH-Px. Low levels of the antioxidant enzyme GSH-Px in liver homogenates indicated hepatotoxicity due to N-APAP overdose.(Tanino et al., 2024).

The results of the advanced statistical analysis of Turkey Post-Hoc showed more specific differences between the treatment groups: the normal control group with negative control was significantly different, with a significance value of 0.001 ($p < 0.05$), and the negative control group when compared with the acetylcysteine group and the EERTK treatment group (250 mg/kg BW, 500 mg/kg BW, and 750 mg/kg BW) showed a significant difference in the average level of the antioxidant enzyme GSH-Px when compared with the negative control, with a significance value of 0.001 ($p < 0.05$).

The normal control group, when compared with the positive control group (acetylcysteine 200 mg), obtained a significance value of 0.334 ($p > 0.05$), and when the normal group was compared with the EERTK 500 mg/kg BB treatment group, the significance value was 0.716 ($p > 0.05$). In the 750 mg/kg BB EERTK group, the significance value = 0.073 ($p > 0.05$) showed no significant difference in the average level of the antioxidant enzyme GSH-Px when compared with the normal control. In EERTK 250 mg/kg BB, a

significance value of 0.001 ($p < 0.05$) was obtained, which indicated a significant difference in the average level of the antioxidant enzyme GSH-Px when compared with the normal control. However, if the EERTK 250 mg/kg BB group was compared with the negative control, a significance of 0.001 ($p < 0.05$) was obtained, which indicates a significant difference, namely, the average level of the antioxidant enzyme GSH-Px was higher than that of the negative control. It can be concluded that the EERTK dose of 250 mg/kg BB can cause a high average level of the antioxidant enzyme GSH-Px in rat liver homogenates, but with an average level that is still significantly different when compared with the average level of the antioxidant enzyme GSH-Px in normal controls. EERTK 500 and 750 mg/kg BB had a significant effect ($p > 0.05$) when compared with normal controls. This explains why there is no significant difference; therefore, from the results of the analysis, it is known that the doses of acetylcysteine 200 mg and EERTK 500 and 750 mg/kg BW have the ability to cause high average levels of the antioxidant enzyme GSH-Px in rat liver homogenates, which is almost the same as the average level of the antioxidant enzyme GSH-Px in normal rat liver homogenates.

The positive control group (acetylcysteine 200 mg), when compared with the EERTK treatment group of 500 and 750 mg/kg BW, showed significant differences of 0.983 and 0.944, respectively, and no significant differences were observed in the average levels of the antioxidant enzyme GSH-Px.

Based on statistical analysis, administering EERTK for 10 days before induction with N-APAP had a significant effect on increasing the average levels of the antioxidant enzyme GSH-Px in liver homogenates of rats in the EERTK 250 mg/kg BW group (43.36 ± 1.34 U/mg protein), EERTK 500 mg/kg BB (55.70± 2.15 U/mg protein), and 750 mg/kg BW (53.14 ± 2.57 U/mg protein) compared to the negative control group (N-APAP 800 mg/kg BW) which had the lowest average value of the antioxidant enzyme GSH-Px (33.86 ± 3.52 U/mg protein). This indicates that the administration of an ethanol extract of T. kunci rhizome effectively protected the liver by increasing the enzymatic defense system against N-APAP-induced injury, thereby increasing the capacity of the antioxidant enzyme GSH-Px in the body, which can ultimately prevent oxidative stress, although it did not reach normal values. A graph of the effect of the treatment group on GSH-Px enzyme levels is shown in Figure 1.

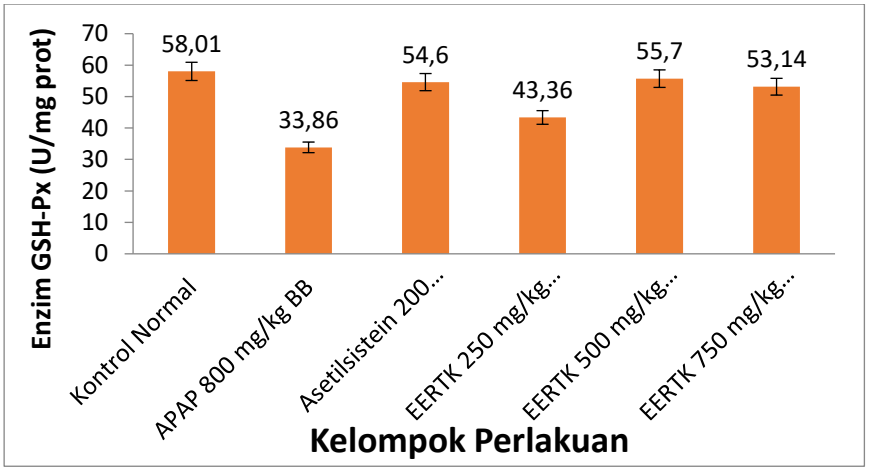

Figure 1. Graph of average GSH-Px enzyme levels in rat liver homogenates

Based on the graphic image, the statistical results also indicated that the EERTK doses of 250 and 750 mg/kg BW provided a high average value of the antioxidant enzyme GSH-Px when compared to the average value of the antioxidant enzyme GSH-Px in the negative control. However, the best EERTK dose is at a dose of 500 mg/kg BW. The use of acetylcysteine 200 mg as a positive control has been shown to cause a high average value of the antioxidant enzyme GSH-Px, which is almost the same as the EERTK dose of 500 mg/kg BW.

When administered in therapeutic doses, N-acetyl-p-aminophenol (N-APAP) is a safe and effective analgesic. However, in overdose conditions, N-APAP is converted to NAPQI mainly by CYP2E1, the cytochrome-P450 isozyme responsible for N-APAP bioactivation. N-acetyl-p-benzoquinone imine (NAPQI) is a reactive metabolite that depletes glutathione (GSH) and induces oxidative stress. NAPQI covalently binds to cellular proteins, resulting in mitochondrial damage, ATP depletion, lipid peroxidation, DNA damage, parenchymal cell necrosis, and liver necrosis. Extensive investigation into the source of free radicals after N-APAP overdose has established that mitochondrial damage and the production of free radicals, such as superoxide, are critical for N-APAP-induced hepatocyte necrosis. The reactive N-APAP metabolite, N-acetyl-p-benzoquinone imine (NAPQI), forms a bond with mitochondrial proteins, particularly components of the electron transport chain, such as ATP synthase, resulting in mitochondrial dysfunction. Furthermore, the activity of this complex, which has been identified as a source of free radicals in the mitochondria, is higher after N-APAP overdose, and this is associated with the severity of liver damage.(Jaeschke & Ramachandran, 2024; Jollow, 2024).

Aerobic reactions lead to the accumulation of free radicals, which can be toxic to the cells. Biotic and abiotic stresses can trigger a significant increase in the generation of free

radicals such as superoxide radicals, hydroxyl radicals, and hydrogen peroxide in the intracellular environment. Two forms of antioxidants–enzymatic and non-enzymatic–have been found to influence the free radical response. The enzymatic system includes a set of gene products such as superoxide dismutase, catalase, ascorbate peroxidase, and glutathione peroxidase (GSH-Px). The body defends itself against ROS by reducing the levels of lipid hyperperoxide and H2O2 through antioxidant enzymes, avoiding lipid peroxidation, and maintaining the structure and function of cell membranes. Glutathione peroxidase (GSH-Px) is the general name for a family of multiple isozymes that catalyze the reduction of H2O2 or organic hydroperoxides to water or alcohols, using reduced glutathione (GSH) as an electron donor. Glutathione peroxidase (GSH-Px) is found in almost all mammalian tissues and is especially abundant in the liver. Therefore, the activity of the antioxidant enzyme GSH-Px can be used as an indicator of liver injury.(Tanino et al., 2024).

To understand the protective mechanism of the ethanol extract of T. kunci rhizome (EERTK) on hepatotoxicity induced by N-APAP, GSH-Px was used as an index to evaluate the level of oxidative stress in the liver. N-APAP overdose induces an oxidative stress response in the liver. As shown in this study. To understand the antioxidant activity of EERTK, the level of the antioxidant enzyme GSH-Px in the liver was determined. GSH-Px catalyzes the reduction of H2O2 or organic hydroperoxide into water or alcohol using reduced glutathione (GSH) as its substrate. As expected, the administration of EERTK significantly increased liver GSH-Px levels induced by N-APAP and inhibited the occurrence of oxidative stress induced by free radicals. This study showed that EERTK effectively protected the liver by enhancing the enzymatic antioxidant defense system against N-APAP-induced liver injury.(Ahmed & Khamees, 2024; Prottasha et al., 2024).

The results of phytochemical screening of the ethanol extract of T. kunci rhizome revealed flavonoids that are likely responsible for the enzymatic antioxidant activity of GSH-Px, as shown by the increase in the levels of the antioxidant enzyme GSH-Px. Polyphenol compounds, such as flavonoids, act as indirect antioxidants by increasing the expression of enzymatic antioxidant genes through various pathways. One way to increase the expression of antioxidant genes is to activate nuclear factor erythroid 2 related factor 2 (Nrf2), which results in an increase in the expression of genes involved in the production of endogenous antioxidant enzymes, such as superoxide dismutase (SOD) and glutathione peroxidase (GSH-Px), to prevent oxidative stress, which can then prevent damage to liver cells.(Sarkar et al., 2024; Sugiharto et al., 2024).

4. CONCLUSION

Administration of the ethanol extract of T. kunci rhizome can increase the level of antioxidant enzyme GSH-Px and prevent oxidative stress in male white rats induced by N-APAP. Based on statistical analysis, administration of EERTK for 10 days before induction with N-APAP had a significant effect on increasing the average level of antioxidant enzyme GSH-Px in liver homogenate of rats in the EERTK 250 mg/kg BW group (43.36 ± 1.34 U/mg protein), EERTK 500 mg/kg BB (55.70± 2.15 U/mg protein), and 750 mg/kg BW (53.14 ± 2.57 U/mg protein) compared to the negative control group (N-APAP 800 mg/kg BW) which had the lowest average value of the antioxidant enzyme GSH-Px (33.86 ± 3.52 U/mg protein). This may indicate that administration of an ethanol extract of T. kunci rhizome effectively plays a role in protects the liver by increasing the enzymatic defense system against N-APAP-induced injury, thereby increasing the capacity of the antioxidant enzyme GSH-Px in the body, which can ultimately prevent oxidative stress, although it has not reached normal values.

REFERENCES

Ahmed, M.N., & Khamees, A.H. (2024). Study the hepatoprotective effect of the methanolic extract of Plumbago auriculata Lam. against CCl4-induced hepatocyte damage in mice. Plant Science Today. https://doi.org/10.14719/pst.3572

Babu, S., Ranjit, S. K., Pattnaik, G., Ghosh, G., Rath, G., & Kar, B. (2024). Ameliorative Effect of Herbal Medicine in the Management of Hepatotoxicity. Endocrine, Metabolic & Immune Disorders - Drug Targets, 24. https://doi.org/10.2174/0118715303319815240819053924

Babyvanitha, S., & Jaykar, B. (2023). PROTECTIVE ROLE OF HERBAL PLANTS IN HEPATOTOXICITY. In Global Journal of Health Sciences (Vol. 8, Issue 2). On line. www.iprjb.org

Coelho, A.M., Queiroz, I.F., Lima, W.G., Talvani, A., Perucci, L.O., Oliveira de Souza, M., & Costa, D.C. (2023). Temporal analysis of paracetamol-induced hepatotoxicity. Drug and Chemical Toxicology, 46(3), 472–481. https://doi.org/10.1080/01480545.2022.2052891

Debnath, S., Kumar, D., Das, M., Mondal, S., & Babu, G. (2024). EXTRACTION METHODS OF BIOACTIVE COMPOUNDS FROM THE PLANTS. In Futuristic Trends in Herbal Medicines and Food Products (pp. 103–122). Iterative International Publishers, Selfypage Developers Pvt Ltd. https://doi.org/10.58532/nbennurch246

Dey, P., Saha, M.R., & Sen, A. (2013). An overview on drug-induced hepatotoxicity. In Asian Journal of Pharmaceutical and Clinical Research (Vol. 6, Issue 4, pp. 1–4). https://doi.org/10.47583/ijpsrr.2024.v84i01.013

Gonfa, Y.H., Bachheti, A., Semwal, P., Rai, N., Singab, A.N., & Bachheti, R.K. (2024). Hepatoprotective activity of medicinal plants, their phytochemistry, and safety concerns:

a systematic review. Zeitschrift Für Naturforschung C. https://doi.org/10.1515/znc-2024-0116

Harahap, MS, Harahap, U., & Sitorus, P. (2023). Phytochemical Screening Of Ethanol Extract Of Temu Kunci (Boesenbergia Rotunda (L.) Mansf) Rhizome Extract And Testing Of Bilirubin Levels In Male White Mice. International Journal of Science, Technology & Management, 4(3), 582–588. https://doi.org/10.46729/ijstm.v4i3.819

Hionides-Gutierrez, A., Goikoetxea-Usandizaga, N., Sanz-García, C., Martínez-Chantar, M.L., & Cubero, F.J. (2024). Novel Emerging Mechanisms in Acetaminophen (<scp>APAP</scp>) Hepatotoxicity. Liver International. https://doi.org/10.1111/liv.16167

Jaeschke, H., & Ramachandran, A. (2024). Acetaminophen Hepatotoxicity: Paradigm for Understanding Mechanisms of Drug-Induced Liver Injury. Annual Review of Pathology: Mechanisms of Disease, 19(1), 453–478. https://doi.org/10.1146/annurev-pathmechdis-051122-094016

Jollow, D. (2024). Acetaminophen-Induced Hepatic Necrosis: A Reminiscence. Drug Metabolism and Disposition, 52(8), 707–711. https://doi.org/10.1124/dmd.123.001278

Mahajan, P., Palkar, M., & Pingili, R.B. (2024). Drug reactive metabolite-induced hepatotoxicity: a comprehensive review. Toxicology Mechanisms and Methods, 34(6), 607–627. https://doi.org/10.1080/15376516.2024.2332613

Mahmood, F., & Hackl, F. (2024). Hepatic Function and Metabolism. In A. Abd-Elsayed (Ed.), Basic Anesthesia Review (pp. 707–708). Oxford University Press New York. https://doi.org/10.1093/med/9780197584569.003.0290

Prottasha, MK, Haq, K., Nafsan, MNR, Shawon, SJ, & Rahmat, S. (2024). Evaluation of Hepatoprotective Activity of Ethanolic Extract of the Seed of Flacourtia jangomas against \(ccl_4\)-Induced Liver Damage in Rat Model. Journal of Complementary and Alternative Medical Research, 25(10), 1–9. https://doi.org/10.9734/jocamr/2024/v25i10573

Sarkar, S., Das, M., & Nanjappan, S. K. (2024). Exploration of the pharmacological mechanisms of herbal medicines against hepatic disorders: A systematic review and network pharmacological analysis. Pharmacological Research - Natural Products, 3, 100048. https://doi.org/10.1016/j.prenap.2024.100048

Shikh, EV, Khaytovich, E.D., & Tsvetkov, D.N. (2024). Herbal extracts in hepatoprotection: antioxidant and immunomodulatory effects. A review. Terapevticheskii Arkhiv, 96(8), 836–845. https://doi.org/10.26442/00403660.2024.08.202841

Stavrakeva, K., Popova, M., Esad, M., Apostolova, E., Kokova, V., Bacelova, M., Alakidi, A., & Bivolarska, A. (2024). Drug-Induced Liver Toxicity. Acta Medica Bulgarica, 51(4), 77–85. https://doi.org/10.2478/amb-2024-0083

Sugiharto, S., Faradila, D., Rizqi Aningrum, K., Nur Hakiki, FD, Zahrotus Sa'adah, A., Fatimatuz Zahroh, U., Soepriandono, H., & Sri Wulan Manuhara, Y. (2024). Potential of Red Ginger (Zingiber officinale) and Temu Kunci (Boesenbergia rotunda): As Stimulator of Antioxidant Enzyme Activity and Anticancer in HepG2 Cell Line. Research Journal

of Pharmacy and Technology, 3599–3606. https://doi.org/10.52711/0974-360X.2024.00562

Tanino, T., Ueda, Y., Nagai, N., Ishihara, Y., Saijo, M., & Funakami, Y. (2024). In vivo upstream factors of mouse hepatotoxic mechanism with sustained hepatic glutathione depletion: Acetaminophen metabolite-erythrocyte adducts and splenic macrophage-generated reactive oxygen species. Chemico-Biological Interactions, 398, 111091. https://doi.org/10.1016/j.cbi.2024.111091

Tsai, Y.-F., Chen, C.-H., Wu, Y.-M., Hung, C.-L., Fang, M.-C., Yu, I.-S., Sheu, J.-C., Hsu, Y.-C., & Lin, S.-W. (2024). Hepsin as a potential therapeutic target for alleviating acetaminophen-induced hepatotoxicity via gap-junction regulation and oxidative stress modulation. Cell Biology and Toxicology, 40(1), 80. https://doi.org/10.1007/s10565-024-09915-z