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Research Article

The Role of Coffee Cherry Ethanol Extract in Repairing Liver Damage in Type 2 Diabetic Rats

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Abstract:

Background. Type 2 diabetes mellitus (T2DM) is a metabolic disorder characterized by chronic hyperglycemia due to insulin resistance and pancreatic β-cell dysfunction. The process of chronic hyperglycemia triggers oxidative stress and increases inflammation, thereby affecting liver damage. The potential of cherry coffee ethanol extract as an antioxidant and anti-inflammatory agent can be used as a therapeutic option in repairing liver damage. **Objective.** This study aimed to evaluate the effect of cherry coffee ethanol extract on liver histopathology in a T2DM mouse model. **Methods**. This experimental study used 28 male Wistar rats, divided into 7 groups with 4 rats in each group, administered coffee cherry ethanol extract at doses of 100mg/kgBW and 200mg/kgBW/day/oral. Results. The results showed that the administration of coffee cherry ethanol extract improved hepatocyte structure, reduced inflammatory infiltration, and decreased fat accumulation in liver tissue compared to the untreated T2DM group ($p<0.05$). **Conclusion**. Coffee cherry ethanol extract has the potential as an adjunct therapy in slowing the progression of T2DM complications in the liver. Further studies are needed to confirm its efficacy, mechanism of action, and long-term effects in humans.

Keywords: ethanol extract, liver histopathology, oxidative stress, type 2 diabetes mellitus.

INTRODUCTION

Type 2 diabetes mellitus (T2DM) is a metabolic disorder characterized by the body's inability to use insulin effectively, leading to chronic elevated blood glucose levels and potentially serious complications.(1) The number of people with diabetes is around 537 million, with predictions that this number will increase to 643 million by 2030 and 783 million adults aged 20-79 by 2045.(2) This increase is predicted to be 42% in developed countries and 170% in developing countries, so

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globally, T2DM remains a burden for every country due to morbidity and mortality rates that continue to increase every year.(2,3) The main risk factors contributing to the incidence of T2DM include genetic factors, family history, obesity, lack of physical activity, smoking, alcohol consumption, and lifestyle changes.(4) Genetically, individuals with a family history of T2DM, especially if their parents or siblings have the disease, have a 2–3 times higher risk of developing T2DM compared to the general population.(5) In addition, obesity is a major risk factor because it plays a role in increasing insulin resistance, which is the main mechanism in the pathogenesis of T2DM.[4-5](6)

Insufficient insulin production and/or ineffective cellular response to insulin, leading to elevated blood sugar levels (hyperglycemia).(7) This condition is characterized by insulin resistance, a state in which cells such as muscle, liver, and fat tissue do not respond optimally to insulin, causing glucose to accumulate in the bloodstream.(8) Insulin is produced by the β cells of the pancreatic islets of Langerhans to regulate glucose levels in the bloodstream and induce glucose storage in the liver, muscles, and adipose tissue.(9) In T2DM, there is dysfunction and reduction in pancreatic β cell mass, which is largely associated with increased cytokines, free fatty acids (FFA), and persistent hyperglycemia.As a result, chronic exposure to these mediators produces free radicals, triggering oxidative stress that induces the production of reactive oxygen species (ROS) and worsens insulin resistance.(10) Oxidative stress and inflammatory processes contribute to the progression of liver damage in people with T2DM.(11) This damage causes hepatocyte apoptosis, lipid peroxidation, and increased free fatty acid circulation, which then leads to fat accumulation in the liver, known as steatosis.(12) In addition to steatosis, there are signs of inflammation such as infiltration of inflammatory cells (lymphocytes and macrophages) in the hepatic sinusoids. If this persists for a long time, it will leave scar tissue/fibrosis in the liver.(11,12)The combination of all these processes can not only damage the liver but also worsen other complications caused by T2DM due to the inflammatory process.(12)

In the treatment of T2DM, some patients choose to consume traditional medicines as a complement to therapy. Indonesia has a diversity of biological resources, including plants that have many pharmacological benefits, one of which is cherry coffee (cherry coffee/berry coffee).(13) Cherry coffee is one of the most widely consumed beverages in the world and has a significant impact on glucose metabolism.(14) Various studies have reported the beneficial effects of whole coffee cherry extract (WCCE) in reducing the risk of T2DM and related complications.(15) In addition to being rich in caffeine, coffee cherry ethanol extract also contains chlorogenic acid, a polyphenol compound with high antioxidant activity and anti-inflammatory effects.(16) In cases of T2DM and its complications,

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chlorogenic acid has therapeutic potential in improving insulin resistance and coffee cherry ethanol extract also shows positive effects in reducing oxidative stress in the liver, inhibiting ROS formation, and improving liver function in animal models.(17) Based on the above background, the researchers were motivated to conduct research on the role of coffee cherry ethanol extract on the histopathological picture of the liver of rats induced with diabetes mellitus.

METHOD

This experimental study used male white rats (Wistar strain) weighing 100-120 g. The rats were adapted in closed cages and in a standard room with adequate lighting (12hour light and 12-hour dark cycles) and were fed and watered ad libitum.

Creation of a diabetic mouse model(18)

Mice were injected with 30 mg/kg body weight streptozotocin intraperitoneally after fasting for 24 hours.[16]

Production of High-Fat Feed(18)

High-fat feed was slightly adjusted and modified, prepared by mixing 200 grams of standard feed, 100 grams of flour, 8 grams of duck egg yolk, 20 grams of oil, and 71.2 mL of water. The maximum feeding portion for each rat was 15 grams per day.

The number of samples per group is 4, divided as follows: Group 1 normal mice

Group 2 normal mice + streptozotocin

Group 3 normal mice + high-fat feed

Group 4: Normal mice + streptozotocin + high-fat diet

Group 5: Normal mice + streptozotocin + high-fat diet + metformin 200 mg/kgBW/day/oral

Group 6: Normal mice + streptozotocin + high-fat diet + cherry coffee ethanol extract 100 mg/day/oral

Group 7 normal rats + streptozotocin + high-fat diet + cherry coffee ethanol extract 200 mg/day/oral

Maceration process of cherry coffee ethanol extract; Cherry coffee powder was placed in a maceration container, then 500 mL of 70% ethanol was added as a solvent. Stir the mixture for 5 minutes using a glass stirrer or magnetic stirrer. Cover the container and store it in a light-protected condition (e.g., in a dark cabinet) at room temperature for 48 hours. Shake or stir the mixture every 12 hours to increase extraction efficiency. Filtration and Concentration of Extract. After 48 hours, filter the mixture using Whatman No. 1 filter paper. Collect the filtrate and repeat the maceration process with the residue using a new solvent (250 mL of 70% ethanol) for 24 hours. Combine all filtrates and concentrate using a rotary evaporator at 40-50°C until a thick extract is obtained. If solvent remains, continue evaporation using a water bath at 40°C. Store the extract in dark glass bottles.

Blood glucose testing using Autocheck strips; Measurements were taken on day 0

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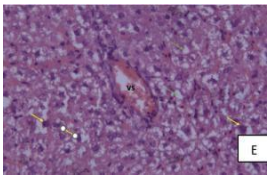
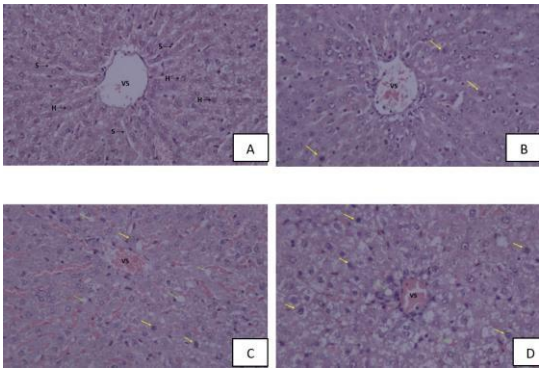
(after acclimatization), day 13 (after STZ induction and high-fat feeding), and day 28 (after administration of metformin and coffee fruit extract).

Preparation of liver histopathology specimens; After the liver was removed from the mice, it was fixed overnight in 10% buffered formalin. The tissue was processed by washing, dehydration, clearing, paraffin embedding, casting, and sectioning into 5 µm slices for hematoxylin and eosin staining.



RESULT

The ethics for this study were obtained from the Health Research Ethics Committee of the Faculty of Medicine, University of Muhammadiyah Sumatera Utara, with Letter Number: 322/KEPK/FKUMSU/2024. In this study, a kappa test was performed to determine the level of agreement between two observers in assessing histopathological features in experimental animals. The kappa value obtained was 0.667, indicating substantial agreement. This value shows that both observers had a relatively high level of consistency in their assessments. The p-value for both was 0.000, indicating that the agreement between observers was statistically significant ($p < 0.05$), so it was unlikely that this agreement occurred by chance. The Hematoxylin-Eosin (HE) staining method was used in this study to observe histopathological changes in rat liver tissue after various treatments. HE staining allows visualization of detailed cellular structures, including hepatocyte damage, inflammation, and tissue morphological changes (Figure 4.1) due to diabetes mellitus and the effects of the therapy administered.



Gambar 4.1 Hepatopathology of the liver shows hepatocytes (H), sinusoids (S), central veins (CV), hepatocyte necrosis (yellow arrows) in the form of pyknotic, karyorrhexis, lipid degeneration (green arrows). (A) Score 0 is seen in the normal group, (B) Score 1 is seen in the treatment (K5 and K7) indicating liver damage of 0.1-5%, (C) Score 2 is seen in K6 indicating liver damage of 6-25%, (D) Score 3 is seen in K2 and K3 indicating liver damage of 50%, (E) Score 4 is seen in K4, indicating liver damage >50%, (H) hepatocytes, (S) sinusoids, (VS) central veins, hepatocyte cell necrosis (yellow arrows) in the form of pyknotic, karyorhexis, lipid degeneration (green arrows), HE staining. Magnification 100X.

In the histopathological assessment of liver damage, the results showed that the histopathological findings in the treatment group improved compared to the positive group. Minimal liver damage may still be present but is lower than in the positive group. The comparison between the treatment groups showed significant results at the highest extract dose. The treatment group receiving

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metformin also showed improvement, but not as much as the 200 mg extract group. Therefore, if metformin and the 200 mg extract have the same effect in improving liver function, herbal medicine is a better choice because synthetic drugs have lower toxicity.

DISCUSSION

Coffee cherry ethanol extract is rich in polyphenols, such as chlorogenic acid, which have antioxidant and anti-inflammatory properties.(19) As antioxidants, these polyphenols play a role in neutralizing free radicals, preventing oxidative damage to liver cells (hepatocytes), and supporting the regeneration of damaged liver tissue. (17)In addition, the anti-inflammatory properties of polyphenols can inhibit the production of pro-inflammatory cytokines, such as interleukin-6 (IL-6) and tumor necrosis factor-alpha (TNF-α), thereby reducing inflammation and further damage to the liver. (19) Although specific research on the effects of cherry coffee ethanol extract on IL-6 and liver damage in T2DM model rats is still limited, other studies have shown that consumption of polyphenol-rich foods can reduce the production of TNF-α and IL-6, as well as increase levels of interleukin 10 (IL-10), an anti-inflammatory cytokine.(20) Therefore, coffee cherry ethanol extract has potential as a therapeutic agent in reducing inflammation and repairing liver damage through antioxidant and anti-inflammatory mechanisms. The antioxidant and anti-inflammatory mechanisms in repairing liver damage and reducing pro-inflammatory cytokine levels involve various important interrelated processes.(21,22) As antioxidants, compounds such as flavonoids and phenolic acids play a role in neutralizing free radicals that can cause oxidative stress in hepatocytes.27 With reduced oxidative stress, the structure and function of cell membranes and their organelles can be protected from further damage, thereby accelerating the regeneration process of damaged liver tissue. In addition, the anti-inflammatory effects of these bioactive compounds work by inhibiting the production of proinflammatory cytokines such as IL-6, IL-1β, and TNF-α, which play a role in the inflammatory process and liver tissue damage.(23,24)

This decrease in cytokine production also reduces macrophage activity that contributes to chronic inflammation, thereby accelerating the healing process of liver tissue.(19) Furthermore, the interaction between antioxidant and anti-inflammatory effects creates synergy in suppressing the inflammatory pathway triggered by oxidative stress, which ultimately reduces pro-inflammatory cytokine levels and improves liver function. (24) Thus, the use of compounds with antioxidant and anti-inflammatory activities can be an effective strategy in reducing inflammation, repairing liver damage, and restoring liver function in conditions affected by oxidative stress and excessive inflammation. (19,25) Overall, the results of this study indicate that cherry coffee

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ethanol extract, especially at a dose of 200 mg/kgBW, has the potential to be an effective alternative therapy in reducing inflammation in liver tissue due to T2DM. Its effectiveness, which is close to or even better than metformin, shows that coffee cherry ethanol extract can be a safer therapeutic option with a lower risk of toxicity compared to synthetic drugs. However, further research is still needed to understand the mechanism of action of coffee cherry ethanol extract in more depth and to evaluate its long-term effects on liver function.

Conclusion

The administration of metformin and 200 mg extract to diabetic-induced rats had the same effect on liver repair, so herbal options are preferable because synthetic drugs have lower toxicity. However, there are still many shortcomings in this study because phytopharmacological testing has not been conducted.

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REFERENCES

1. World Health Organization (WHO). Diabetes [Internet]. 2023 [cited 2024 Jul 9]. Available from: https://www.who.int/health-topics/diabetes#tab=tab_1
2. IDF International Diabetes Atlas 10th ed. Brussels, Belgium. International Diabetes Federation. 2021 [cited 2024 Jul 9]; Available from: https://www.diabetesatlas.org
3. Khan MAB, Hashim MJ, King JK, Govender RD, Mustafa H, Al Kaabi J. Epidemiology of Type 2 Diabetes – Global Burden of Disease and Forecasted Trends. J Epidemiol Glob Health. 2019;10(1):107.
4. Wu Y, Ding Y, Tanaka Y, Zhang W. Risk Factors Contributing to Type 2 Diabetes and Recent Advances in the Treatment and Prevention. Int J Med Sci. 2014;11(11):1185–200.
5. Meigs JB, Cupples LA, Wilson PW. Parental transmission of type 2 diabetes: the Framingham Offspring Study. Diabetes. 2000 Dec 1;49(12):2201–7.
6. Banday MZ, Sameer AS, Nissar S. Pathophysiology of diabetes: An overview. Avicenna J Med. 2020 Oct 4;10(04):174–88.
7. Galicia-Garcia U, Benito-Vicente A, Jebari S, Larrea-Sebal A, Siddiqi H, Uribe KB, et al. Pathophysiology of Type 2 Diabetes Mellitus. Int J Mol Sci. 2020 Aug 30;21(17):6275.
8. Kilanowska A, Ziółkowska A. Role of Phosphodiesterase in the Biology and Pathology of Diabetes. Int J Mol Sci. 2020 Nov 3;21(21):8244.
9. Lu X, Xie Q, Pan X, Zhang R, Zhang X, Peng G, et al. Type 2 diabetes mellitus in adults: pathogenesis, prevention and therapy. Signal Transduct Target Ther. 2024 Oct 2;9(1):262.
10. Bashir H, Hayat Bhat M, Majid S. Molecular Pathogenesis of Inflammatory Cytokines in Insulin Resistance Diabetes Mellitus. In: Insights on Antimicrobial

E-ISSN :2722-0877

Peptides. IntechOpen; 2022.
11. Ghasemi-Dehnoo M, Amini-Khoei H, Lorigooini Z, Rafieian-Kopaei M. Oxidative stress and antioxidants in diabetes mellitus. Asian Pac J Trop Med. 2020;13(10):431.
12. Mohamed J, H. NNA, H. ZA, B. BS. Mechanisms of Diabetes-Induced Liver Damage: The role of oxidative stress and inflammation. Sultan Qaboos Univ Med J. 2016 May 15;16(2):e132-141.
13. Lestari W, Hasballah K, Listiawan MY, Sofia S. Antioxidant and phytometabolite profiles of ethanolic extract from the cascara pulp of Coffea arabica collected from Gayo Highland: A study for potential anti-photoaging agent. F1000Res. 2023 Sep 18;12:12.
14. Coffee-Around-the-World, National Coffee Association of U.S.A., Inc. https://www.ncausa.org/About-Coffee/Coffee-Around-the-World.
15. Van der Werf R, Walter C, Bietiger W, Seyfritz E, Mura C, Peronet C, et al. Beneficial effects of cherry consumption as a dietary intervention for metabolic, hepatic and vascular complications in type 2 diabetic rats. Cardiovasc Diabetol. 2018 Dec 20;17(1):104.
16. Bahadoran Z, Mirmiran P, Azizi F. Dietary polyphenols as potential nutraceuticals in management of diabetes: a review. J Diabetes Metab Disord. 2013 Aug 13;12(1):43.
17. Krawczyk M, Burzynska-Pedziwiatr I, Wozniak LA, Bukowiecka-Matusiak M. Impact of Polyphenols on Inflammatory and Oxidative Stress Factors in Diabetes Mellitus: Nutritional Antioxidants and Their Application in Improving Antidiabetic Therapy. Biomolecules. 2023 Sep 17;13(9):1402.
18. Speakman JR. Use of high-fat diets to study rodent obesity as a model of human obesity. Int J Obes. 2019 Aug 9;43(8):1491–2.
19. Simón J, Casado-Andrés M, Goikoetxea-Usandizaga N, Serrano-Maciá M, Martínez-Chantar ML. Nutraceutical Properties of Polyphenols against Liver Diseases. Nutrients. 2020 Nov 15;12(11):3517.
20. Liu C, Feng X, Li Q, Wang Y, Li Q, Hua M. Adiponectin, TNF-α and inflammatory cytokines and risk of type 2 diabetes: A systematic review and meta-analysis. Cytokine. 2016 Oct;86:100–9.
21. Yan Y, Zhou X, Guo K, Zhou F, Yang H. Use of Chlorogenic Acid against Diabetes Mellitus and Its Complications. J Immunol Res. 2020 May 28;2020:1–6.
22. Abhilasha, Mitra P, Suri S, Saxena I, Shukla RKG, Shukla KK, et al. Downregulation of interleukin-10 receptor (IL-10R) along with low serum IL-10 levels in newly diagnosed type 2 diabetes mellitus patients. Gene Rep. 2021 Sep;24:101251.
23. Nemzer B, Kalita D, Abshiru N. Quantification of Major Bioactive Constituents, Antioxidant Activity, and Enzyme Inhibitory Effects of Whole Coffee Cherries (Coffea arabica) and Their Extracts. Molecules. 2021 Jul 16;26(14):4306.
24. Schmidt-Arras D, Rose-John S. IL-6 pathway in the liver: From physiopathology to therapy. J Hepatol [Internet]. 2016 Jun;64(6):1403–15. Available from: https://linkinghub.elsevier.com/retrieve/pii/S0168827816000830
25. Bilal HM, Riaz F, Munir K, Saqib A, Sarwar MR. Histological changes in the

E-ISSN :2722-0877

liver of diabetic rats: A review of pathogenesis of nonalcoholic fatty liver disease in type 1 diabetes mellitus. Cogent Med. 2016 Dec 31;3(1):1275415.