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Assessment of the acute toxicity for methanolic Leaf extract of *Solanum melongena* L. in Swiss albino mice

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Abstract

Eggplant (Solanum melongena L.) is a therapeutically significant vegetable known for its diverse therapeutic properties. The current research aimed to conduct the acute toxicity study of methanolic peel extract of S. melongena in Swiss albino mice. Mice received an oral dose of methanolic peel extract (2000 mg/kg body weight) and were observed over a period of eight consecutive days. Mice were dissected after the end of acute period and their blood was sampled for different biochemical (ALT, AST, ALP, Uric acid, Urea, Creatinine, LDL, HDL) and hematological indices (RBCs, MCV, WBCs, Hematocrit, Platelets, Hemoglobin) and some major organs (liver, kidneys, spleen, lungs, heart) were surgically removed for histopathological analysis. The treated group of mice showed no significant alterations in biochemical and hematological parameters when compared with the control group. The histopathological assessment of the major organs did not reveal any toxic effects of the extract. The results demonstrating that the methanolic peel extract of S. melongena is non-toxic and safe at the selected dose, indicating a possible applicability of the peel extract in the therapeutic practice. Nonetheless, additional sub-acute and chronic toxicity research is necessary to ascertain the safety in the long-term.

1. Introduction

Natural compounds are being extensively studied due to increasing prevalence of chronic diseases worldwide (Shakya *et al.*, 2016; Halder *et al.*, 2021). Conventional medicine has been employing herbs and vegetables due to their extensive array of phytocompounds and pharmacological effects (Samtiya *et al.*, 2021). Solanaceae plants, such as *Solanum melongena*, are well-known in terms of their nutritional and health benefits (Yarmohammadi *et al.*, 2021; Solanke *et al.*, 2019). *Solanum melongena* L. is a valuable dietary addition to the vegetarian diet providing dietary fibers and a variety of minerals, including iron, calcium, zinc, potassium, phosphorus, magnesium, and vitamins C, B1, B3, B6, A, E, D, and K (Yarmohammadi *et al.*, 2021). *S. melongena* peel and pulp have the antioxidative, anti-inflammatory, and neuroprotective effect because of the presence of chlorogenic acid, delphinidin, anthocyanins, and flavonoid compounds (Fraikue *et al.*, 2016; Fidrianny *et al.*, 2017). Eggplant extracts can be used to address hypercholesterolemia, bronchitis, asthma, and diabetes (Yarmohammadi *et al.*, 2021). Although much has been researched on the phytochemistry and bioactivity of eggplant fruit and pulp (Karimi *et al.*, 2021; Tsafack *et al.*, 2024), the peel is usually discarded as the food waste. This by-product might have concentrated secondary metabolites. The recent research suggests that the phytochemicals in various sections of the plant are accumulated in different modes (Nisha *et al.*, 2009). The study was carried out with an aim of assessment of the acute anti-inflammatory effect of aqueous leaf extract of the eggplant in albino mice. Guideline based acute toxicity was performed to determine the LD50 of test drug (Umamageswari and Maniyar, 2015). A study examines how eggplant peel chemicals affect

lipid profiles, blood glucose, liver enzymes, and renal function in diabetic mice. The effects on lipid profiles, blood glucose, and creatinine were examined for prospective benefits. These medications may reduce diabetes complications (Abdullah and Al-Lehebe, 2025). Nevertheless, a detailed study has not been conducted to determine the safety profile and toxicity of methanolic *S. melongena* peel extract in animals. The gap is addressed by this study that explores the toxicity of *S. melongena* methanolic peel extract. The objectives of this study are to (1) assess acute toxicity of the methanolic peel extract of *S. melongena* in Swiss albino mice, (2) evaluate biochemical and hematological parameters, and (3) examine histopathological changes in vital organs. The study aims to demonstrate that the potential therapeutic use of eggplant peel waste remains safe, even though it is a waste product, as it originates from typical food waste sources.

2. Materials and Methods

2.1. Collection of plant material

Fresh vegetables (*Solanum melongena*) were purchased from a local market and their peels were removed using a knife. The peels were washed with tap water and then dried in the shade at room temperature.

2.2. Preparation of methanolic extract of *Solanum melongena* peel

The dried peels were pulverized into fine powder using an electric grinder. A 50 g sample of the peel powder was dissolved in 75% methanol at a ratio of 1 g to 10 mL. The solution was then shifted to a conical flask and agitated on an orbital shaker at 24 °C for 72 hours. The mixture was filtered by Whatman filter paper no. 1. The solvent was evaporated from the extract at 40 °C under reduced pressure using a rotary evaporator. After drying, it was kept at 4 °C for subsequent use (Nauroze *et al.*, 2023).

2.3. Toxicity profiling in Swiss albino mice

2.3.1. Maintenance of animals

Swiss albino mice (male and female) of 5-6 weeks of age and weighed 20 to 30 g, were acquired. The mice were maintained in controlled conditions (23 to 25 °C, 12 light/12 dark cycle) with unrestricted access to normal pellet food and water. The animals were given one week of adaptation to the laboratory conditions prior to commencement of the experiment.

2.3.2. Ethical approval

To initiate this study, ethical approval was obtained under letter No. UO/ERC/2023/Z14, dated: 23-12-2023, from the Ethical Research Committee, University of Okara, Punjab, Pakistan.

2.3.3. Measurement of body weight

The initial body weights of all mice were recorded using an electric weighing balance. The body weights of the control and treatment group mice were also recorded on a daily basis throughout the experimental period. The dose was prepared on the basis of average body weight of the mice.

2.4. Acute toxicity study

Swiss albino mice were split into two groups, with five mice in each group. The treatment group received a single oral dose of 2000 mg/kg body weight of the methanolic peel extract of *Solanum melongena*. The control group had a regular feeding on food and water. The mice were observed closely in the case of abnormal behavior and general toxicity appearance within the first 24 hours of drug administration. The observations were made for the 8 consecutive days. By the conclusion of the experiment, the animals were not fed overnight and then anesthetized using chloroform and cardiac puncture was used to take blood samples. Two separate tubes were taken, one for serum analysis and the other for hematological analysis (Kanwal *et al.*, 2022).

2.4.1. Biochemical analysis

To separate serum and plasma, blood samples were incubated in room temperatures (5-10 minutes) followed by centrifugation using biochemical tubes at 3000 rpm (10 minutes). The biochemical parameters including AST, ALT, ALP, Urea, Uric acid, Creatinine, LDL, and HDL were analyzed on serum samples using commercially available diagnostic kits (Nauroze *et al.*, 2023).

2.4.2. Hematological analysis

To conduct hematological analysis, blood samples were collected in EDTA tubes. A hematology analyzer (Coulter counter S-plus VI) was used to carry out complete blood count (CBC) to identify red blood cells (RBCs), white blood cells (WBCs), platelets (PLT), mean corpuscular volume (MCV), hematocrit, and hemoglobin (Hb) (Nauroze *et al.*, 2023).

2.4.3. Histopathological examination

Surgical removal of the liver, kidneys, spleen, lungs and heart were performed followed by rinsing with phosphate buffer saline to eliminate debris. The individual weighing of each organ in grams was done using an analytical balance. The organs were then stabilized on 10% formalin solution (pH 7.4) after 24 hrs. It was preceded by the dehydration of tissues in progressive degrees of alcohol (70%, 90% and 100%), clearing in xylene and embedding in paraffin wax at 58 to 60 °C. Tissue blocks were made, and sections of 5 µm thickness were cut with a microtome. All the tissue sections were stained using hematoxylin and eosin (H&E). A photomicroscope was used to examine the stained areas at various magnifications (Kanwal *et al.*, 2022).

2.5. Statistical analysis

The results were given in the form of means and standard error of mean (SEM). GraphPad Prism (version 5) was used to analyze the data with unpaired t-test. A P-value < 0.05 was considered statistically significant.

3. Results

All mice survived for a period of eight days after receiving a single oral dose (2000 mg/kg body weight) of the methanolic peel extract of *Solanum melongena*. The mice in both the control and treated groups were carefully observed. No significant alterations were observed in various behavioral patterns. The body weight of animals in both the control and treated groups was recorded every day. As presented in Table 1, the control and treatment groups showed no significant variation in body weight. Organ weights did not differ significantly between the control and treatment groups, as shown in Table 2. The biochemical profiles of the mice's blood revealed no significant variations in AST, ALT, ALP, creatinine, uric acid, urea, HDL, and LDL levels (Figure 1). Furthermore, the methanolic peel extract of *S. melongena* had no significant impact on the levels of RBCs, WBCs, PLT, MCV, Hb, and hematocrit in the blood of the mice (Figure 2). Histopathological examinations of the liver, heart, spleen, kidneys, and lungs of the dissected mice were conducted to assess the toxicity of the plant extract. The histopathological analysis of these vital organs revealed no signs of damage, indicating that the methanolic *S. melongena* peel extract did not adversely affect these organs (Figure 3).

Table 1. Body weight of control and treated group of mice.

Days	Body weight (g) of Swiss albino mice	
	Control group	Treated group
Before Dose	22.4 ± 1.2	23.4 ± 1.3
After Dose		
Day 1	22.4 ± 1.5	22.8 ± 1.5
Day 2	22.6 ± 1.2	22.7 ± 1.3
Day 3	22.1 ± 1.9	22.8 ± 1.2
Day 4	22.4 ± 1.8	22.9 ± 1.2
Day 5	23.8 ± 1.0	23.4 ± 1.3
Day 6	23.2 ± 1.2	23.8 ± 1.5
Day 7	24.8 ± 0.9	24.9 ± 1.0

Values are articulated as Mean ± SEM. Values are the mean of five replicates.

Table 2. Organ weight of control and treated group of mice.

Sr. No.	Organs	Control group mice	Treated group mice
1.	Kidney	0.3 ± 0.0	0.3 ± 0.0
2.	Liver	1.5 ± 0.2	1.5 ± 0.3
3.	Spleen	0.2 ± 0.1	0.2 ± 0.0
4.	Heart	0.1 ± 0.0	0.1 ± 0.0
5.	Lungs	0.3 ± 0.1	0.4 ± 0.1

Values are articulated as Mean ± SEM. Values are the mean of five replicates.

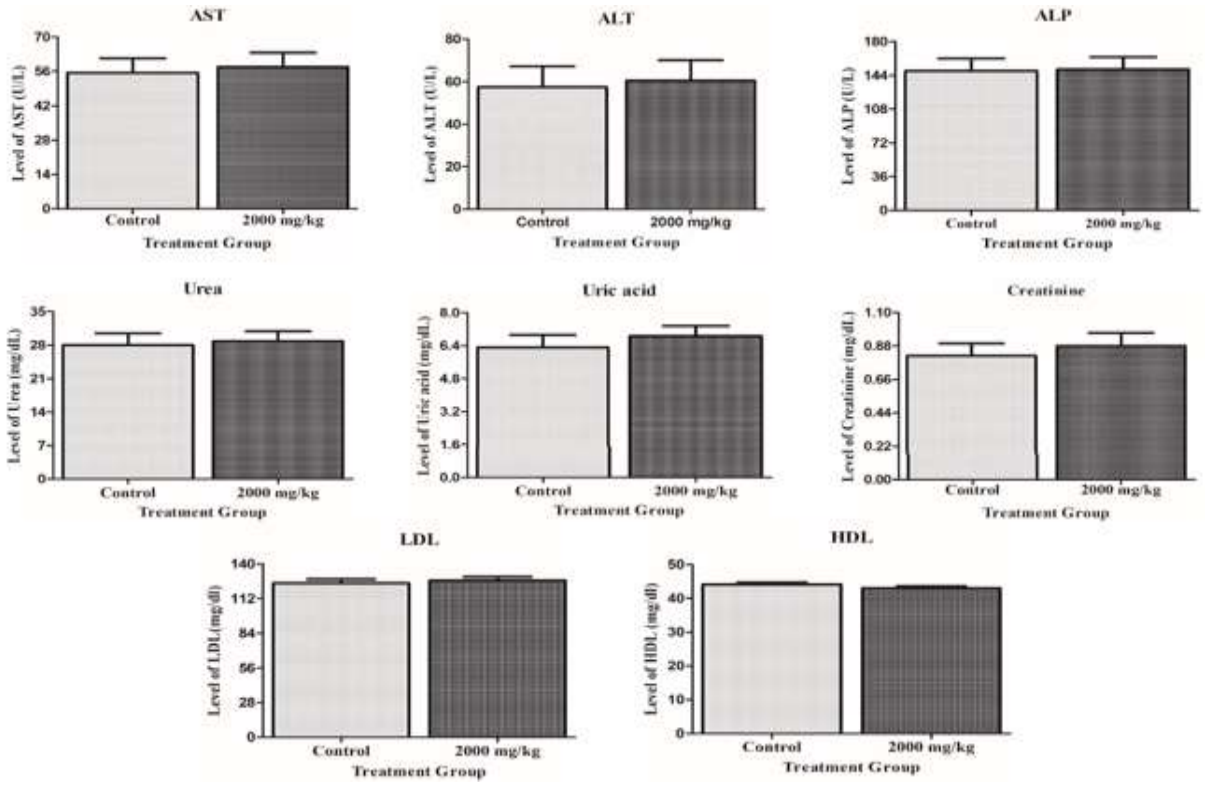

Figure 1. Effect of methanolic peel extract of *Solanum melongena* on the level of AST, ALT, ALP, Urea, Uric acid, Creatinine, LDL, and HDL of Swiss albino mice in acute toxicity study.

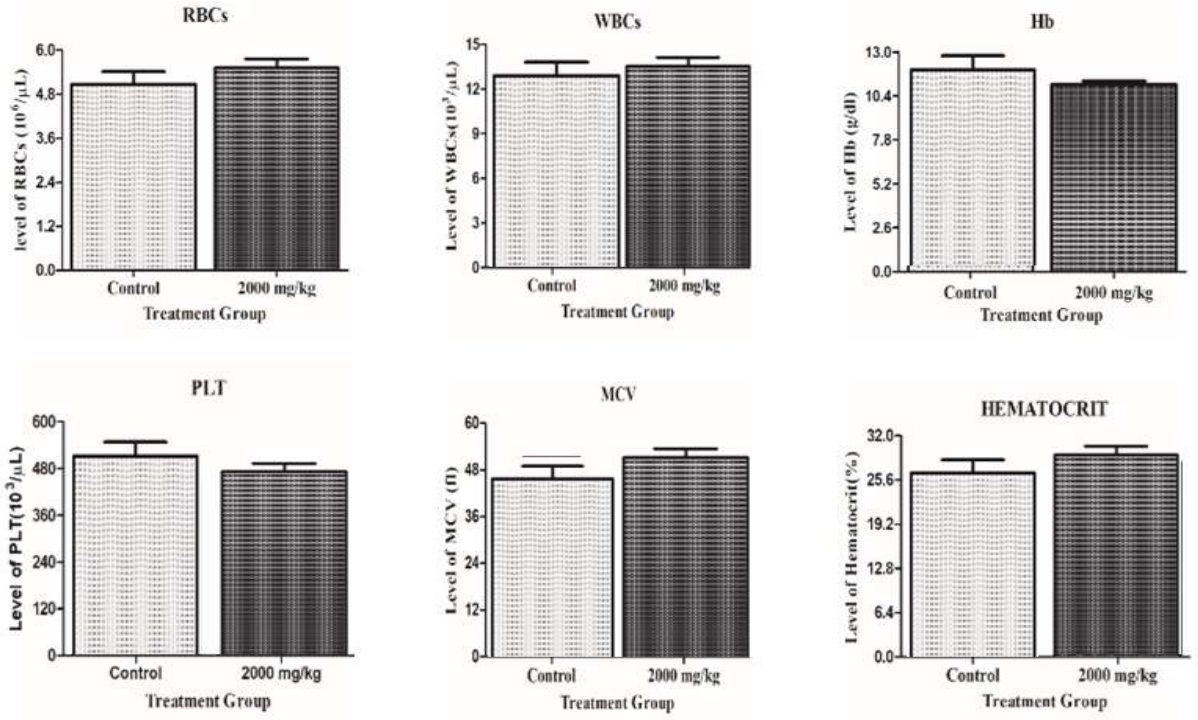

Figure 2. Effect of methanolic peel extract of *Solanum melongena* on the level of RBCs, WBCs, PLT, MCV, Hb, and Hematocrit of Swiss albino mice in acute toxicity study.

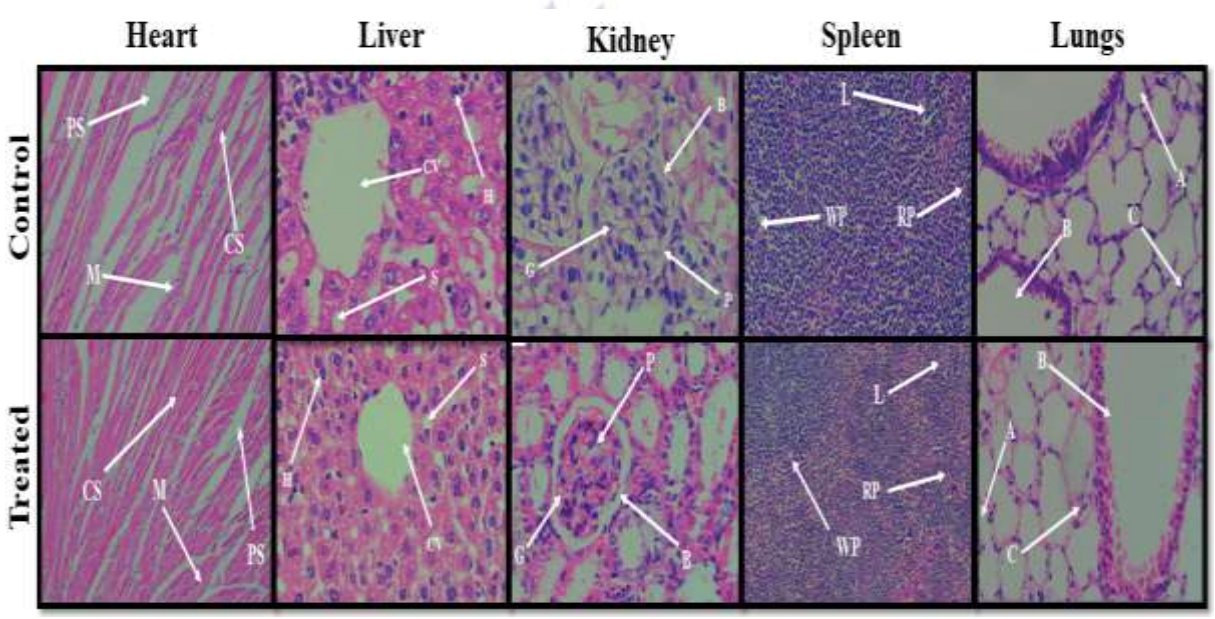

Figure 3: Histopathological comparison of vital organs in control versus *Solanum melongena* peel-extract treated mice. Hematoxylin Eosin-stained sections were examined at 10 x and 40 x. Abbreviations: 'C' for Capillary, 'N' for Nuclei, 'ID' for Intercalated disc 'H' for Hepatocytes,'S' for Sinusoidal cells, 'CV' for Central vein 'G' for Glomerulus, 'B' for Bowman's space, 'P' for Podocytes 'WP' for White pulp, 'RP' for Red pulp, 'L' for Lymphocytes 'E' for Epithelium, 'A' for Alveoli.

4. Discussion

A study previously done by Ur Rehman *et al.*, (2021) indicated that medicinal plants and extracts are applicable in the treatment of diseases. People normally consider medicinal plants and herbs to heal or cure diseases and restore health in the body. Our study also highlights the importance of medicinal plants,

and we also indicate that plants are of both medicinal and commercial importance.

Solanum melongena, commonly known as eggplant, is a member of the Solanaceae family. It is rich in various beneficial components, including chlorogenic acid, lanosterol, steroid alkaloids, glycoalkaloids, nasunin, oxalic acid, selenium, zinc, and vitamins A, C, and E. Notably, eggplant is abundant in vitamins, phytochemicals, and antioxidants, which contribute to the treatment and prevention of numerous ailments such as cancer, diabetes, cardiovascular disease, and inflammation (Tsafack *et al.*, 2024).

According to Karimi *et al.* (2021), the calyx and peel of the eggplant are prevalent by products in industrial food processing. The peels are particularly rich in phenolic compounds, including chlorogenic acid, anthocyanins, and flavonoids, which enhance their antioxidant capacity. Research has demonstrated that methanol-dissolved extracts of eggplant peel possess antioxidant properties, which mitigate oxidative stress and thereby help prevent the onset of chronic diseases (Nisha *et al.*, 2009). We also prepare methanolic extracts under standardized conditions and conduct toxicity profiling in Swiss albino mice.

S. melongena is commonly referred to as eggplant and has enormous applications in the field of medicine. The plant has many anti-inflammatory, antibacterial, and antioxidant bioactive phenolic compounds, flavonoids, and anthocyanins. Since previous researchers have found that eggplant can perform numerous biological functions, we also observed that in our research, eggplant has antioxidant, anti-inflammatory, antibacterial, and numerous other biological functions (Fraikue *et al.*, 2016). Consistent with the present study, the previous study by Sarhan *et al.*, (2014) found that the hematological parameters of the treatment group had no significant changes when they were exposed to dosage of 2000 mg/kg of eggplant peel extract and were not toxic to liver, kidney, spleen, heart and lungs of the mice. Acute toxicity shows that the plant extract was considered to be safe and non-toxic. This is in favor of safe usage of methanolic extract *S. melongena* peel in pharmacological practice.

The biochemical parameters and histopathology were the methods of assessing the impact of natural materials on the major organs of mice (Islam *et al.*, 2014). Histopathology of the major organs (kidney, spleen, liver, lungs and heart) in this study demonstrated that none of the vital organs showed structural alteration or damage during the toxicity study. Present research demonstrated that the methanolic peel extract of *Solanum melongena* exhibited a non-toxic profile in mice.

Conclusion

The current study showed that *Solanum melongena* methanol peel extract proved to be safe and non-toxic in Swiss albino mice. There were no significant variations in either biochemical or hematological indices investigated in acute administration of the extract in Swiss albino mice as compared with control group indicating no nephrotoxicity, hepatotoxicity, or hematopoietic disruption. The histopathological study of vital organs (liver, kidneys, spleen, heart, and lungs) did not show any structural damages or abnormalities. These findings prove the safety profile of eggplant peel extract and justify the potential in the use of the same in therapeutic application. More sub-acute and chronic toxicity studies are however suggested to completely determine the safety profile of long-term effects. Such research would potentially bring new opportunities regarding the advancement of safe plant-based therapeutic reagents based on products of agricultural wastes.

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