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Research Article

EVALUATION OF ACUTE ORAL TOXICITY OF AQUEOUS EXTRACT *OF EUPHORBIA HELIOSCOPIA*

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ABSTRACT:

Euphorbia helioscopia is a well-known species of medicine plant in the *Euphorbiaceae* family. In traditional medicine, euphorbia plants have been used for hundreds of years to treat warts, tumors, and cancer. The study's goal was to find out how harmful the water-based extract of *Euphorbia helioscopia* was to Swiss white mice. After giving the extract by mouth in doses of 2 g/kg and 5 g/kg, each patient was watched for four hours on their own. After that, they watched them nonstop for 24 hours and then at least once a day for the next 14 days. According to the study's findings, the liquid extract of *Euphorbia helioscopia* did not cause any harm or death in any of the treatment groups that were given it. On top of that, there were no changes between the treatment and control groups in body weight, food intake, organ weights, or biochemical markers. Since this is the case, giving *Euphorbia helioscopia* extract to mice by mouth is thought to be safe and not cause any harm.

KEYWORDS: *Euphorbia helioscopia*, acute oral toxicity, Swiss albino mice, Toxicity.

INTRODUCTION:

A chemical's acute toxicity refers to the harmful effects that happen after one exposure or several exposures in a short amount of time, usually less than 24 hours (Walum, 1998). For a drug to be considered acutely toxic, its harmful effects must show up within 14 days of being given (Schlede, *et al.*, 2005). Acute toxicity is different from chronic toxicity, which is the harm done to health by being exposed to a chemical over and over again for a long time (months or years), usually at lower amounts each time (Kattamuri, *et al.*, 2012). A lot of people think it's wrong to use people as test subjects for acute (or ongoing) toxicity studies (Panditi and Vinukonda, 2011). However, looking into accidental human exposures, like accidents in factories, can give us useful information (Putikam, *et al.*, 2012). Animal tests, in vitro tests, and extrapolation from data on related substances are the main ways that acute toxicity data is gathered (Akki, *et al.*, 2022 and Saisri, *et al.*, 2021).

Herbs are used as alternative medicines to treat a wide range of illnesses because they are safe, effective, easy to get, and don't cost much (Namadeva, *et al.*, 2024). The main worries of the public, patients, and consumers are the well-being of animals and the quick availability of safe and effective drugs (RANI, *et al.*, 2023). It makes sense to think that plants used in traditional medicine will not be very harmful since people have used them for a long time

(Nataraja, *et al.,* 2023). According to new studies, some plants that are widely used in traditional medicine can have bad effects (Meenakshi, *et al.,* 2023). These secondary molecules, which come from plant biosynthesis, are very important for plant development, growth, reproduction, and defense. In addition, plants need them to stay alive. *Euphorbia helioscopia L.* is a yearly plant in the *Euphorbiacae* family (Pathan, *et al.,* 2023). It has small yellow-green flowers, a straight brown stem, and oval leaves that grow in a pattern that changes every year (Sen, *et al.,* 2023). It can grow up to 50 cm tall. This plant comes from a lot of different places, like Europe, Asia, and North Africa (Khulbe, *et al.,* 2023). People in many countries around the world have used *Euphorbia helioscopia* as a cure in their traditional medicine.

Euphorbia helioscopia is an old plant that has been used for a long time to treat many illnesses, including swelling, ascites, tuberculosis, dysentery, scabies, lung cancer, cervical carcinoma, and esophageal cancer (Kumar, *et al.,* 2022). It is also thought to have antibacterial and antifungal qualities. Scientists have found that *Euphorbia species* make a lot of different diterpenoids and triterpenoids. These chemicals have many biological effects, such as stopping tumors from growing and fighting cancer. People and animals are said to be at risk from some species of Euphorbia because they contain poisonous substances, mostly in the white, milky sap known as latex. In this study, the harmful effects of *Euphorbia helioscopia* on white mice were looked into (Surana, *et al.,* 2020).

MATERIALS AND METHODS

Collection of Plant Material:

Euphorbia specimens were collected from many places in India throughout the month of April (Surana, *et al.*, 2019).

Animal used in acute oral toxicity:

Male White to study acute poisoning, 35–45 g Wistar mice were used. The animals were kept in a controlled setting called an animal facility. The temperature was kept at 20°C, and daylight and darkness were switched on and off every 12 hours. The mice were kept in plastic cages, with three mice in each cage. They could drink tap water whenever they wanted and were given market foods to eat (Ahmad, *et al.*, 2016).

Preparation of *Euphorbia helioscopia* Aqueous Extract

The sealed parts underwent a process of drying, crushing, and cleaning using running water.

A quantity of fifty grams of powder was heated in a volume of five hundred milliliters of water for a duration of fifteen minutes. Subsequently, the mixture underwent evaporation in a rotary vacuum evaporator following filtration using Whatman filter paper.

Evaluation of Acute Toxicity Study of *Euphorbia helioscopia* Aqueous Extract:

The extract's acute oral toxicity was assessed using the procedures specified in the 425 guidelines of the Organization for Economic Co-operation and Development. The animals were divided into three groups, each consisting of three individuals, with three males in each group (Jarouliya, *et al.,* 2015). The control group received a standard saline solution. The second group received a single dose of the water-based extract at a concentration of 2 g/kg, whereas the third group received a single dose at a concentration of 5 g/kg. Prior to receiving individual dosages of the extract diluted in distilled water, the animals underwent a period of fasting for four hours during which they were deprived of food and water (Gautam, *et al.*, 2015). After receiving the dose, the mice's activity was observed daily for a period of 14 days, starting two weeks later. In the first 24 hours, their behavior was also watched on and off, with extra attention paid to the first four hours. After finishing the therapy, the animals were given free access to water after not eating or drinking for the night (Gautam, *et al.*, 2015). After giving diethyl ether to put the person to sleep, blood samples were taken through a retro-orbital cut and put in a tube with heparin. The blood was obtained by centrifuging the tube at 4000 revolutions per minute for 15 minutes at a temperature of 4°C. After that, it was kept at -20°C until it was examined. The kidneys, liver, lungs, heart, and spleen were all measured.

Study of Food Consumption and Body Weight:

During the two-week study period, the body weight of each mouse was evaluated weekly. Furthermore, the quantity of food ingested by each mouse was ascertained by calculating the disparity between the amount provided and the amount remained after a 24-hour period (Tiwari *et al.,* 2021).

Biochemical Analysis Study of *Euphorbia helioscopia* Aqueous Extract:

The task was accomplished using a device known as the Beckman, which is an automated analyzer. One of the contributing factors was alanine aminotransferase (ALT), while another element was aspartate aminotransferase (AST) (Tiwari *et al*., 2023a; Tiwari *et al.,* 2023b).

Animal Organ Weights:

Following the sacrifices of all the animals, the kidneys, heart, liver, spleen and lungs were methodically extracted and individually measured for weight (Tiwari and Pathak, 2023; Tiwari *et al.*, 2022).

Statistical Analysis

The data is presented using the average value plus or minus the standard deviation. ANOVA, also known as one-way analysis of variance, was used to identify the distinctions among the groups (Tiwari *et al.*, 2021).

RESULTS

Observations revealed that the behavior and body weight of the mice were modified by specific amounts of *E. helioscopia*. The livers of mice in both the treatment and control groups were inspected using a microscope to detect the levels of aspartate aminotransferase (AST) and alanine aminotransferase (ALT). Additionally, molecular tests were conducted on the mice.

Animal Signs of Toxicity and Mortality:

Scientists observed the behavior of the mice multiple times over the 14-day acute toxicity study period. Several of the mice's measurements shown no indications of poisoning, and all of them remained in good health (Table 1).

Table 1: Mice administered a solitary oral dose of *Euphorbia helioscopia* extract exhibited death rates and displayed signs indicative of toxicity.

Prescribe Dose (mg/kg)	**Death Ratio (D/T)**	**Reported Adverse effects**
0000	0 out of 3	Normal
2000	0 out of 3	Normal
5000	0 out of 3	Normal

Body Weight Changes

The mice did not lose any weight while they were being watched while the *E. helioscopia* watery extract was being used. While the control mice gained or lost weight, the treated mice's weight stayed the same (Table 2). This is important to know because differences in

body weight can show when chemicals and drugs are making people sick. Chemicals or drugs that are poisonous make the body lose weight. Chemicals or drugs that are not poisonous, on the other hand, make the body gain weight.

Table 2: Effects of the aqueous extract derived from *Euphorbia helioscopia* on the body weight of mice.

Day	**Body Weight (g) of mice**		
	Control	**2 g/kg**	**5 g/kg**
1st Day	46.72±1.72	36.11±3.11	37.41±4.91
7th Day	40.81±1.59	45.45±2.11	49.71±1.31
14th Day	45.71±1.90	37.31±2.90	36.81±3.55

Macroscopic examination of the organs in the therapy groups revealed no anomalies. These organs were found to have the same texture as the organs in the control group.

Food Consumption

The data shown in Table 3 show that there is no statistically significant difference in the amount of food ingested by the mice in the treatment and control groups.

Table 3: Effect of *Eeuphorbia helioscopia* aqueous extract on mice's food intake

Day	**Food consumption (g)**		
	Control	**2 g/kg**	**5 g/kg**
Food	13.90±8.11	20.44±3.99	20.22±1.11

Biochemical Analysis

It was discovered that the ALT and AST levels in the treated and control groups were the same, indicating that the treatment had no effect on this parameter (Table 4).

Table 4: Effect of a water-based extract of *Euphorbia helioscopia* on mouse biochemical parameters.

	Euphorbia helioscopia on mouse biochemical parameters		
Parameters	**Control**	**2 g/kg**	**5 g/kg**
ALT (UI/L)	31.75±3.32	28.03±2.51	45.21±10.10
AST (UI/L	111.21±31.66	173.33±79.58	210.82±18.56

Organs Weight

The organ weight data is presented in Table 5, and no noticeable distinction was observed between the groups.

Table 5: Effect of a water-based extract of *Euphorbia helioscopia* on the weight of mice's organs

	Organs Weight		
Organ (mg)	**Control**	**2 g/kg**	**5 g/kg**
Liver	1.81±0.21	1.70±0.07	1.51±0.18
Lungs	0.31±0.11	0.23±0.11	0.20±0.19
Kidneys	0.33±0.22	0.29±0.04	0.22±0.07
Spleen	0.19±0.06	0.22±0.08	0.21±0.03
Heart	0.14±0.04	0.21±0.04	0.22±0.04

DISCUSSION

There are many ways that medicinal plants can be used to make new drugs, whether they are an extract, a pure component, or a product (Vyas, *et al.,* 2010). Most of the natural substances used in traditional medicine have strong science evidence to back up their biological properties. However, there isn't a lot of information or proof about the bad affects that therapeutic herbs might have on people who use them. To make people more confident in the safety of medicinal plants and the products made from them, especially those used in pharmaceuticals, study on acute and sub-chronic toxicity is needed (Vyas, *et al.,* 2010). Because of this, one important part of figuring out if a medicinal plant extract could be dangerous is finding out what toxicological effects it has, whether it is meant for people or animals.

There were no instant changes in the test animals' overall behavior. The animals that were treated behaved normally, just like the animals in the control group (Jain, *et al.,* 2010). Individual animals in the group lived for up to 14 days, and no deaths were seen. The results of our study supported what other researchers had found about the latex and methanol extract (L.MT) from *Euphorbia helioscopia* leaves (Sharma, *et al.,* 2010). The results of the study on how E. Helioscopia L. aqueous extract affected the body weight of mice showed that the plant was not poisonous, since the aqueous extracts did not change the mice's weight gain or food intake. Because nutrients are important for the body's processes, it is important to keep track of how much water and food the animal eats while testing the safety of a chemical

meant to be used for medical reasons. Regardless of the treatment group, the mice in this study ate and drank normally. The same thing was found by in his study in 2015. The fact that this was found says that the animal's body is metabolizing fat, carbs, and proteins normally (Vyas, *et al.*, 2010).

To figure out how dangerous drugs are, you need to know a lot about clinical biochemistry. This is because transaminases, especially AST and ALT, are useful measures for both predicting drug toxicity and checking liver function (Surana, *et al.*, 2024). High amounts of these enzymes mean that they are getting into the bloodstream because the parenchymal cells in the liver are damaged. It is possible for hepatic enzymes to leave cells that have been damaged by liver damage because the membranes are more permeable. ALT and aspartate aminotransferase values that are too high are signs of liver damage (Khairnar, *et al.*, 2024).

The current investigation shows that the biochemical parameters (ALT and AST) in the group that was treated with *E. helioscopia* L. extract were not significantly different from those in the normal control group. The groups that got latex and LMT treatment in the Butin study had lower amounts of bilirubin, ALT, and ALP than the control group (Parkhe, *et al.*, 2024). These results show that *E. helioscopia* protects the liver, which may be because the plant has antioxidant properties. The results show that the methanol extract of *E. helioscopia* leaves (L.MT) is a strong antioxidant in living things and may help treat diseases caused by oxidative stress (Lokhande, *et al.*, 2024).

During this experiment, there were no changes in organ weights that were statistically significant. As a result, it is possible to say that the test substances are not toxic. In 2014, the same result was found. It is possible for any poison that targets the heart, liver, kidneys, lungs, or spleen to cause metabolic effects (Aher, *et al.*, 2024). The heart, liver, kidneys, lungs, and spleen of the control group did not show any problems when looked at under a microscope.

It was found that the plant's LD50 was more than 5000 mg/kg. Based on the globally standardized classification system, the plant can be put into category 5 according to his study from 2015. In line with the OECD, this ranking is based on the fact that the LD50 values for latex and LMT were both more than 2000 mg/kg (Shelke, *et al.*, 2024).

CONCLUSION

Based on the results, we may infer that the aqueous extract of *Euphorbia helioscopia* L. is non-toxic and does not cause any adverse reactions, even when consumed at various amounts. The extract of *E. helioscopia* did not modify the composition of the blood's

constituents or induce any cellular apoptosis. The initial results indicate multiple areas that warrant additional exploration on the therapeutic and rejuvenating capabilities of *E. helioscopia* L. extract, while posing minimal risk of adverse effects. Additional research is required to examine the long-term and ongoing harmful effects of this extract in order to ensure its safety. Moreover, prior to the widespread endorsement of a novel phytotherapeutic medicine generated from this plant, it is imperative to conduct clinical trials to ensure its safe application.

DECLARATIONS:

Ethics approval and consent to participate:

Not applicable.

Consent for publication:

All the authors approved the manuscript for publication.

Availability of data and material:

All required data is available.

Competing interests:

All authors declare no competing interests.

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