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SHORT COMMUNICATION article

A toxicological study of ecballium elaterium plant in mice

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Abstract: Ecbalium elaterium has a very violent effect on the body and has little use in modern herbalism. Little is known about the acute and chronic toxicities of ecbalium elaterium in human. This study aims to determine the acute toxicity (LD_{50}) of fruit extract and another aerial part extract of the ecballium elaterium in experimental animals. Thus, male albino mice were divided into different groups each group consists of six mice receiving 40, 46, 52, 61, and 69 mg/kg of fruit extract of ecballium elaterium, respectively. Other groups were given 1000, 1412, 1995, 2818, and 3981 mg/kg of the areal part of ecballium elaterium extract, respectively. The LD_{50} in both treatments was determined by using the Spearman-Karber method. The LD_{50} of the fruit and aerial parts of ecballium were 55 mg/kg and 2112.5 mg/kg, respectively. The present findings showed significant weight loss after one month of treatment with 1400 mg/kg and 40 mg/Kg of fruit and aerial part extracts, respectively. The results indicated that the fruit extract is highly toxic as compared to the extract of the aerial parts.

Introduction

Ecballium elaterium (EE) is a flowering plant of one of the cucurbitaceous family. It is known as a squirting cucumber. It is a Mediterranean plant used in folk medicine. Especially fruits and fruit juice are administered for several therapeutic uses, although they can be toxic at high doses [1, 2]. Fruit has traditionally been used orally and topically for the treatment of various diseases such as sinusitis, fever, rheumatic diseases, hepatic diseases, hypertension constipation, edema, jaundice and fungous infectious [3-5]. Pharmacological studies have demonstrated that the plant's fruit has anticancer effect [6, 7], and antibacterial, antifungal, analgesic, antipyretic, and anti-inflammatory properties [4, 8, 9]. Hassan and Paulis [10] have observed that the administration of EE fruit juice to rats ameliorates the hepatotoxicity induced by the cyclophosphamide compound through its antioxidant and anti-inflammatory activities. Thus, this study aims to determine the acute toxicity of the fruit and the aerial part extract of the EE in mice.

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Materials and methods

Experimental animals: Male albino mice weighing 25 - 30 gm from the local animal house of the Faculty of Medicine, University of Benghazi, Libya were used throughout the current study. Ethical approval was obtained from the ethics committee of the University of Benghazi, Libya (ref. No. 02-2021).

Experimental protocol: Extraction procedure: An acceptable quantity of EE plant was collected in October 2018 from the area around Benghazi City, Benghazi, Libya. The plant was identified by two members of the Department of Botany, Faculty of Science, University of Benghazi. The plant was cleaned by washing with cold water and dried in the shade at room temperature, then crushed and ground by an electrical blender to obtain the powder. 100 gm of the dried fruit or areal part of plant powder was placed in the thimble siphons of the Soxhlet apparatus. The flask of the Soxhlet was filled with a half-liter of the solvent, and the heating of the solvent started at a temperature between 60 and 80^0C. The solvent extraction was performed in order of increasing polarity by using petroleum ether, chloroform, ethyl acetate, ethanol, and water. The process continued till the extraction was achieved. Extracts were completely evaporated by using a rotary evaporator and kept until used. The ethanolic fraction was used during all the experiments.

Acute toxicity: The LD_{50} was calculated according to the Spearman-Karber method as described by Gené [11]. Mice were divided into five groups, each of six mice receiving 40, 46, 52, 61, and 69 mg/kg of fruit extract of EE, respectively. Other groups were given 1000, 1412, 1995, 2818, and 3981 mg/kg of the areal part of EE extract, respectively. The symptoms of the toxicity were observed in these different groups. The number of deaths in each group of mice was also recorded. The median lethal dose (LD_{50}) in treatment after 48 hours was determined by using the Spearman-Karber method.

Calculation of LD_{50}: The LD_{50} is calculated from the following equation: $M = Xk + 1/2d - dr/N$

Where: M = log LD50
Xk = log dose causing 100% mortality
d = log dose interval
r = the sum of the number of animals
N is the number of animals at each dose level

Results and discussion

About eighty percent of the world's population uses traditional medicine for primary health care [12]. Even though the use of these plants has shown promising potential with high global demand, there are still concerns about not only their use but also their safety [13]. During the acute toxicity study and as indicated in **Table 1**, tachypnea, hypoactivity lethargy, diarrhea, staggering, gasping, and death were observed at higher doses of fruit extract of EE. As presented in **Table 2**, and through observation of the animals for 24 hours, the number of dead mice increased as the dose increased. Data shown in **Table 2**, indicated that 69.0 mg/kg of the fruit extract of EE causes 100% death. By using of Spearman-Karber method [11], the LD_{50} of the fruit extract of EE was nearly equal to 55.0 mg/kg. As shown in **Table 3**, the signs and symptoms which include tachypnea, lethargy, hypoactivity, diarrhea, gasping, and ended with death were observed upon administration of 1995 mg/kg, and 100% of death was observed with 3981 mg/kg of the areal parts of EE extract.

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Table 1: Signs of acute toxicity of fruit extract of ecballium elaterium in mice

Dose (mg/kg)	No of mice	Signs of acute toxicity	No dead mice
40	6	None	0
46	6	None	0
52	6	Tachypnea, lethargy, hypoactivity, diarrhea, gasping & death	2
61	6	Tachypnea, lethargy, hypoactivity, diarrhea, gasping & death	5
69	6	Tachypnea, lethargy, hypoactivity, diarrhea, gasping & death	6

Table 2: Evaluation of LD_{50} of fruit extract of ecballium elaterium according to Spearman-Karber

Dose mg/kg	No of mice	No dead mice	Dead/total No	Percentage of mortality	Log dose	Log dose interval
40	6	0	0/6	00.0%	1.60	0.06
46	6	0	0/6	00.0%	1.66	0.06
52	6	2	2/6	25.0%	1.72	0.06
61	6	5	5/6	62.5%	1.78	0.06
69	6	6	6/6	100%	1.84	0.06

Table 3: Signs of acute toxicity of the areal parts of ecballium elaterium in mice

Dose (mg/kg)	No of mice	Signs of toxicity	No dead mice
1000	6	None	0
1412	6	None	0
1995	6	Tachypnea, lethargy, hypoactivity, diarrhea, gasping & death	3
2818	6	Tachypnea, lethargy, hypoactivity, diarrhea, gasping & death	5
3981	6	Tachypnea, lethargy, hypoactivity, diarrhea, gasping & death	6

The principal aim of evaluating the safety of any medicinal plant is to identify the nature and significance of adverse effects and to establish the exposure level at which this effect is observed [14]. According to the Organization for Economic Development (OECD), substances with LD_{50} > 2000 to 5000 mg/kg are categorized as unclassified or category 5 [15]. This suggests that the oral LD_{50} of the plant being greater than 2000 mg/kg may be safe. This study evaluated the potential toxicity of ethanolic extract of the fruit and the aerial parts of EE through acute and sub-acute oral intake in mice. As shown in **Table 4**, the calculated LD_{50} of the extract of the areal part was 2113.5 mg/kg. The results of the toxicity study indicate that the LD_{50} of the fruit and the aerial parts of EE were 55.0 mg/kg and 2112 mg/kg in mouse, respectively. However, further studies are needed to evaluate the sub-chronic and chronic toxicity and to explore the mechanism of toxicity of the fruit extract in experimental animals.

Table 4: Evaluation of the LD_{50} of areal parts of ecballium elaterium according to Spearman- Karber

Dose (mg/kg)	No of mice	No dead mice	Dead/total No	% of mortality	Log dose	Log dose interval
1000	6	0	0/6	00.0%	03.0	0.15
1412	6	0	0/6	00.0%	3.15	0.15
1995	6	3	3/6	37.5%	03.3	0.15
2818	6	5	5/6	62.5%	3.45	0.15
3981	6	6	6/6	100%	03.6	0.15

Conclusion: According to the OECD, the fruit extract of EE should be considered toxic, whereas the aerial part extract is less toxic or may be safe.

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