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EVALUATION OF ACUTE TOXICITY PROPERTIES OF YARROW AND COMMON OAK PLANT EXTRACTS

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Abstract: Nowadays, preparations made from medicinal plant raw materials are being studied with great interest by the global community compared to synthetic biologically active substances. Due to the side effects of synthetic drugs considered suitable for treating chronic diseases, people around the world prefer plant-based preparations. Traditional medicinal plants can provide new compounds that counter the high cost and toxicity of available drugs, especially for rural populations in developing countries [1].
This article presents data on the acute toxicity properties of an extract sample obtained from Achillea millefolium (yarrow) and Quercus robur (common oak), studied at the Pharmacology and Screening Laboratory of the Institute of Bioorganic Chemistry of the Academy of Sciences of Uzbekistan.

Keywords: Acute toxicity, plant extract, white laboratory mice, LD_{50}, toxicity class, evaluation indicators.

Introduction: In today's era of medical advancements, the demand for medicines derived from natural plant raw materials is increasing. This is because synthetic drugs clearly show adverse effects. Under the initiative of our President, great importance is being given to expanding the production of local pharmaceuticals using regional plants. According to item 31 of Appendix 1 of the Presidential Decree No. PQ-4670, dated April 10, 2020, "On measures for the conservation, cultivation, processing and rational use of wild-growing medicinal plants", 50 plants are listed, including "yarrow".
Our research focuses on analyzing the alcoholic extract obtained from a mixture of yarrow and oak plants and collecting data on its toxicity for potential use in medicine [2].
Description of Plants and Composition: Common Yarrow (Achillea millefolium) – A perennial herbaceous plant of the Asteraceae family, growing up to 20–80 cm tall. It has a branched rhizome and erect stems ending in corymb-like inflorescences. The leaves are sessile, double-pinnately divided. The flowers are in heads that collectively form a corymb. It blooms from June to late summer and fruits from August.
Chemical composition: Contains carotene, vitamins K and C, alkaloids like achilleine and betonicine, up to 0.8% essential oil, matricarin isomers, millefin lactone, 0.31% choline, asparagine, resins, tannins, and bitter substances like prochamazulene (achillin). According to the XI Pharmacopoeia, the essential oil content must be at least 0.1%. The oil contains up to 4% chamazulene, thujone, camphor, borneol, caryophyllene, up to 10% cineole, and various acids.

Uses: Used for gastrointestinal diseases, to stimulate appetite, and as a hemostatic for internal bleeding and external wounds (nose, gums, injuries).
Common Oak (Quercus robur) – A tree species of the Fagaceae family, found widely in Ukraine, Belarus, Moldova, the Baltics, and Russia, and cultivated in Uzbekistan. Grows up to 40–50 meters tall.
Chemical composition: Contains 7–20% tannins (including up to 4% in old bark), mainly from the pyrogallol group, 1.6% gallic and ellagic acids, flavonoids (quercetin), flobaphene, pentoses, and pectic acids. According to the XI Pharmacopoeia, tannin content must be at least 8%.
Uses: Preparations from oak bark are used as astringent and antiseptic agents for oral cavity diseases (gingivitis, stomatitis), sore throat, and for treating burns with 20% decoctions.

Objective of the Study: To evaluate the acute toxicity effect of an extract obtained from yarrow and oak plants.

Materials and Methods:
The acute oral toxicity was assessed using the fixed dose method recommended by the OECD (2001), Test No. 420 (Acute Oral Toxicity - Fixed Dose Procedure, OECD Guidelines for the Testing of Chemicals, Section 4, OECD Publishing, Paris, https://doi.org/10.1787/9789264070943-enra).
The plant extract was administered orally to animals at a fixed dose of 5000 mg/kg. Experiments were conducted on male, non-breed white laboratory mice with an average body weight of 22±2.0 g. Each group included 5 mice, with a total of 10 animals. Animals were healthy, sexually mature, and quarantined for 10–14 days prior to testing.

Experimental Procedure: The experiment was conducted in two stages. In the first stage, two mice were administered the plant extract at a dose of 5000 mg/kg in 0.5 ml volume via gastric intubation. No mortality was observed during the 2–3 days of observation. In the second stage, the remaining 3 mice of the group received the same dose. A control group received the same volume of distilled water.
During the first day of both stages, hourly monitoring was carried out for general condition, tremors, and signs of death. For up to two weeks, daily monitoring included assessment of overall health, activity, skin and fur condition, respiration rate and depth, urination, body weight changes, and other parameters. The animals were kept under standard conditions with free access to food and water. At the end of the study, the extract's median lethal dose (LD_{50}) and toxicity class were determined [4,5].
The data were statistically processed using arithmetic mean (M), standard error (m), and results were considered statistically significant at $p<0.05$.

Results: After oral administration of the extract at 5000 mg/kg, mice exhibited increased respiratory rate, huddling behavior, and eye constriction within 10 minutes. These effects lasted for 15–25 minutes, after which the animals began returning to normal condition.
No mortality was observed in the group administered the plant extract at 5000 mg/kg (0/5).
Compared to the control group, there were no significant body weight reductions on days 7 and 14 ($p>0.05$). The extract's LD_{50} was determined to be >5000 mg/kg.
The results are summarized in Table 1.

Table 1.
Assessment indicators of acute toxicity of plant extracts in male mice

(M±m, n=5)

Groups	Dose mg/ml, ml	Number of animals/tested	Average body weight, g	LD_{50}, mg/kg
			Day 1	Day 7
Control	0.5 ml	5/0	22.0 ± 0.3	23.0 ± 0.3
Plant extract	5000 mg/ml	5/0	21.0 ± 0.2	22.0 ± 0.3

Note: *P<0.05 compared to control group.

Table 2.
General appearance and behavioral observation of control and test groups during acute toxicity study [1]

Observation	Control Group	5000 mg/kg Dose – After 10 minutes	5000 mg/kg Dose – After 15–25 minutes
Digestion	No change	No change	No change
Temperature	Normal	Increased	Normal
Urination	Normal	Altered	Normal
Respiratory rate	Normal	Increased	Normal
Skin condition	No effect	No effect	No effect
Insomnia	Absent	Absent	Absent
Sedation	No effect	Observed	Observed
Eye appearance	No effect	Eye narrowing observed	Eye narrowing observed
Diarrhea	Absent	Absent	Absent
General physical activity	Normal	Decreased	Decreased
Coma	Absent	Absent	Absent
Mortality	Alive	Alive	Alive
Food consumption	Normal	Normal	Normal
Body weight	Normal	Normal	Normal

Conclusion: When the plant extract was administered orally to mice at a single dose of 5000 mg/kg (relative to the mass of the alcoholic extract), and the results were classified according to the OECD guidelines, it was determined that the sample belongs to the Class VI – substances with low hazard, with an LD_{50} > 5000 mg/kg.

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