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# Toxicity Effect of *Nectaroscordum tripedale* Extract on Hematological and Biochemical Parameters in Mice

**Ali Asghar Kiani[1], Behrouz Ezatpour[1], Massumeh Niazi[2], Sareh Jahanbakhsh[3, 4 *]**

*[1]Razi Herbal Medicines Research Center, Lorestan University of Medical Science, Khorramabad, Iran*
*[2]Student Research Committee, Lorestan University of Medical Science, Khorramabad, Iran*
*[3]Bam University of Medical Sciences, Bam, Iran*
*[4]Research Center for Tropical and Infectious Diseases, Kerman University of Medical Sciences, Kerman, Iran*

**ABSTRACT**

*Currently, a broad spectrum of medicinal herbs is existing; while this seems to be necessary to present scientific validations for finding their side effects for therapeutic goals. In this study, the toxicity effects of Nectaroscordum tripedale extract on hematological and biochemical parameters in mice were assessed. Toxicity effects of N. tripedale extract was assessed by measuring the biochemical and hematological parameters of the treated mice after oral administration of N. tripedale extract at the doses of 0.5, 1, and 2 g/kg, respectively, for two weeks. The obtained results of the present investigation revealed that N. tripedale extract at the tested doses had no significant toxicity on the biochemical parameters of ALT, AST, ALP, and bilirubin for the liver function, and also Cr and BUN for renal function as well as hematological parameters such as HGB, Hct, WBC, RBC, and PLT counts. However additional studies are mandatory to assess other toxicity aspects such as genotoxicity, chronic toxicity.*

**Keywords:** Toxicity; mice; liver; kidney; hematology

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**Corresponding author**: Ms. Sareh Jahanbakhsh

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## INTRODUCTION

From ancient times to now, the use of medicinal plants for the treatment and prevention of diseases has always been interested by human societies in all countries of the world [1,2]. Currently, a broad spectrum of medicinal herbs is existing; while this seems to be necessary to present scientific validations for finding their side effects for therapeutic goals [3-5].

One of the most widely used herbs in Iranian folk medicine is *Nectaroscordum tripedale*, from the Allium genus; which is widely grows in various regions of the world particularly in Iran [6]. From many years ago, this *N. tripedale* has demonstrated a lot of health benefits in treating many diseases such as rheumatic and joint pains, bladder and kidney stones as well as infectious diseases [6, 7]. On the other hand, studies carried out in recent years have shown the high antimicrobial potency of this plant; so that its antibacterial effects against some pathogenic bacterial strains as well as antileishmanial and protoscolicidal activity against some parasites have been proven [8-10].

Recent studies on laboratory animals have shown that liver and kidney are considered as the key target tissues of drug toxicity; therefore, evaluating the performance of this organ is one of the important ways to measure drug toxicity [11]. To date, liver damage is being investigated by measuring serum levels of enzymes such as alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP) and bilirubin. Considering kidney damage in measuring drug toxicity, the creatinine blood test (Cr) and blood urea nitrogen (BUN) are considered the main serum enzymes to assess the kidney function [12].

Given that there is currently no documented study on the toxicity of this plant, this study was designed to assess the toxicity effect of *N.*

*tripedale* extract on hematological and biochemical parameters in mice.

## MATERIALS AND METHODS

### Collection of plant materials

The aerial parts of *N. tripedale* were collected in April 2017 from mountainous areas of Lorestan Province, Iran. Then, the plant was identified by a botanist in Herbarium of Agriculture and Natural Resource Research Center, Khorramabad, Iran.

### Preparation of extract

Extraction of air dried plant materials (200 g) was done by percolation method using methanol (80%) for three days in 21°C. To remove the artifacts, the extract was passed through filter paper (Whatman No.3, Sigma, Germany). Next, the extract was concentrated in vacuum at 50°C by means of a rotary evaporator (Heidolph, Germany) and kept at -20°C, until use [13, 14].

### Animals

Totally 24 male NMRI mice (6–8 weeks old) were purchased from the Animal Breeding Stock Facility of Razi Herbal Medicines Research Center, (Khorramabad, Iran). Animals were housed in a colony room with a 12:12-h light/dark cycle at 21 ± 2˚C and handled according to the standard protocols for the use of laboratory animals.

### Study design

Mice were randomly divided into four groups (6 mice per each group) as following:

First group (Control group): received normal saline intraperitoneally for two consecutive weeks

Second group: mice which received the *N. tripedale* extract at the dose of 0.5 g/kg intraperitoneally for two consecutive weeks

Third group: mice which received the *N. tripedale* extract at the dose of 1 g/kg intraperitoneally for two consecutive weeks

Forth group: mice which received the *N. tripedale* extract at the dose of 2 g/kg intraperitoneally for two consecutive weeks.

### Blood collection

After 14 days of administration, the animals were anesthetized by Ketamine-Xylizine, and then the blood samples from each mouse after opening the heart were collected.

### Determination of hematological parameters

For measuring the hematological parameters, the obtained total blood was put into tubes with ethylenediaminetetraacetic acid (EDTA) anticoagulant. Then a number of hematological parameters including hemoglobin, hematocrit, white blood cell counts, red blood cell, and platelet counts were assessed [15,16].

### Determination of biochemical parameters

For measuring the biochemical parameters in serum, some collected blood was put into tubes without anticoagulant, to process the clot. After separating the serum by centrifugation at 5000g for 10 min, various clinical chemistry parameters such as aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), creatinine (Cr), blood urea nitrogen (BUN), and bilirubin (direct and total), were determined by commercial diagnostics kits [17, 18].

### Statistical analysis

The analysis of the obtained results was done using SPSS 17.0 statistical package (SPSS Inc., Chicago, IL, USA). Differences between test and control groups were analyzed by t-test and $P<0.05$ was considered statistically significant.

## RESULTS

### Hematological parameters

In this investigation, the doses of 0.5, 1, and 2 g/kg of *N. tripedale* extract were used to determine its toxicity effects of hematological parameters in mice. Table 1 shows the results of the effects of various doses of *N. tripedale* extract of hematological parameters in mice. At first, the obtained findings revealed no death for the mice after administration of *N. tripedale* extract after 2 weeks. No significant difference ($p> 0.05$) between oral administrations of *N. tripedale* extract was observed at the doses of 0.5, 1, 2, and 4 g/kg and control group.

**Table 1.** Hematology parameters in whole blood of tested mice (Mean ± SD).

| Parameters | *N. tripedale* extract (g/kg) | | | Control |
|---|---|---|---|---|
| | 0.5 | 1 | 2 | |
| RBC (×l0$^6$/μL) | 3.8 ± 1.15 | 4.1 ± 0.33 | 4.6 ± 0.6 | 4.2 ± 0.3 |
| HGB (g/dL) | 11.1 ± 1.23 | 10.8 ± 1.07 | 10.6 ±1.15 | 10.9 ± 0.66 |
| Hct (%) | 34.3 ± 6.5 | 36.2 ± 3.1 | 40.1± 6.6 | 38.6 ± 3.51 |
| WBC (×l0$^3$/μL) | 6.8 ± 0.33 | 7.1 ± 0.43 | 8.1 ± 0.85 | 7.5 ± 0.6 |
| PLT (×l0$^3$/μL) | 356.6 ± 26.6 | 381.6 ± 21.3 | 401.6 ± 33.6 | 379.3 ± 30.3 |

RBC, red blood cell; HGB, hemoglobin; Hct, hematocrit; WBC, white blood cell; PLT, platele lkanes

**Biochemical parameters**

Table 2 shows the results of toxicity effects of *N. tripedale* extract on serum biochemical parameters in mice receiving *N. tripedale* extract at the doses of 0.5, 1, and 2 g/kg for two weeks. Table 1 shows the results of the effects of various doses of *N. tripedale* extract on hematological parameters in mice. The obtained results demonstrated that although these parameters increased with increasing dosage of extract, there is no significant difference (p> 0.05) between oral administrations of *N. tripedale* extract at the doses of 0.5, 1, and 2 g/kg and control group.

**Table 2.** Clinical chemistry parameters in mice sera (Mean ± SD).

| Parameters | *N. tripedale* extract (g/kg) | | | Control |
|---|---|---|---|---|
| | 0.5 | 1 | 2 | |
| AST (U/L) | 86.6 ± 6.5 | 98.3 ± 13.3 | 107± 18.3 | 81.3 ± 11.5 |
| ALT (U/L) | 61.2 ± 6.6 | 72.3 ± 5.6 | 80.6± 11.3 | 64.6 ± 6.3 |
| ALP (U/L) | 173.3 ± 18.6 | 191.3 ± 20.1 | 198± 16.3 | 168.6 ± 16.5 |
| Cr (mg/dL) | 0.96 ± 0.07 | 0.78 ± 0.08 | 0.91 ± 0.21 | 0.8 ± 0.05 |
| BUN (mg/dL) | 41.3 ± 6.1 | 47.1 ± 6.6 | 46.6 ± 6.3 | 39.6 ± 6.15 |
| TB (mg/dL) | 0.7 ± 0.23 | 0.84 ± 0.15 | 0.88± 0.15 | 0.83 ± 0.1 |
| DB (mg/dL) | 0.11 ± 0.05 | 0.15 ± 0.02 | 0.2± 0.01 | 0.2 ± 0.015 |

AST, aspartate aminotransferase; ALT, alanine aminotransferase; ALP, alkaline phosphatase; Cr, creatinine; BUN, Blood urea nitrogen; TB, Total bilirubin; DB, Direct bilirubin

## DISCUSSION

The statistics has shown that a significant number of world populations are using medicinal herbs for treatment and curative goals. About one fifth of all the plants are used for medical purposes; while approximately 10% of them are used for commercial aims [1, 2]. In this study, it was aimed to assess toxicity of *N. tripedale* extract on hematological and biochemical parameters in mice. Nowadays, one of the most important tests to evaluate the liver function as well as inflammations and injuries such as hepatitis and cirrhosis is evaluation of serum liver enzymes. Measuring the serum BUN and creatinine is mostly used to evaluate the kidney function in a large number of conditions, to make detecting kidney diseases possible and screen individuals with acute or chronic renal dysfunction or failure (12).

Obtained finding in the present study revealed that although biochemical parameters of ALT, AST, ALP, and bilirubin for evaluation of the liver function and also Cr and BUN for renal function have increased with increasing dosage of the extract; there is no significant difference (p> 0.05) between oral administrations of *N. tripedale* extract at the doses of 0.5, 1, and 2 g/kg and control group. Moreover, we found that *N. tripedale* extract had no significant modification in hematological parameters such as HGB, Hct, WBC, RBC, and PLT counts.

Regarding the toxicity effects of plants in the genus of *Allium*, [19] have shown that after oral administration of garlic (*Allium sativum*) extract for 5 weeks, no significant changes were observed in serum AST, ALP, total bilirubins, Na, K, Cr, RBC, HCT, Hb, mean corpuscular hemoglobin concentration, granulocyte and organs-body weight ratio in the tested rats.

## CONCLUSION

The obtained findings of the current investigation demonstrated that *N. tripedale* extract at the tested doses had no significant toxicity on the liver and kidney organs and also against hematological parameters in NMRI mice for two weeks; however additional studies are mandatory to assess other toxicity aspects such as genotoxicity, chronic toxicity.

**Conflict of interest**

None.

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