Effect of Swertia chirata extract on various biochemical parameters in rats

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Abstract:

Background: The current study's objective was to compare with control and illustrate the effect of well-known plant Swertia chirata's methanol extract on kidney and liver function test. Despite being ignorant of their harmful effects, people assume that traditional plants are safe and affordable. Kidney and liver function test are a crucial for evaluating the level of toxicity on various organs. **Methodology**: Albino Wistar rats were randomly assigned into three groups of six rats each. Group 1 was control, while group 2 (LD) and group 3 (HD) received 250 mg/kgand500 mg/kg body weight of extract respectively, orally once daily. **Results**: No significant difference was observed in the the serum enzymes, alanine amino transferase (ALT), aspartate amino transferase (AST), total protein (TP), bilirubin total, urea and creatinine; however, levels of alkaline phosphatase (ALP) were significantly decreased at 500mg/kg. While all others levels were found to be non-significant as compared to the control. **Conclusion:** Oral administration of *Swertia chirata* at 500mg/kg decreased the ALP levels, while no significant difference was observed at high dose on other biochemical parameters.

Key words: ALP, ALT, AST, Bilirubin, Biochemical parameters, Creatinine, *Swertia chirata*.

1. Introduction:

Medicinal herbs have been utilized for ages in various parts of the world because they are effective in treating and preventing a wide range of diseases(1). The traditional use of medicinal herbs has been widespread in nations with limited resources due to the adverse responses and contraindications of standard medications. People prefer traditional medicines because they are readily available, safe, and affordable, partly due to cultural beliefs (2).

Worldwide, the herb Swertia chirata is commonly used to treat a variety of illnesses, especially in rural areas. This plant, often referred to as chirata, Kirataka, or Chiratika (4), is a native of the Himalayas (5) and is found in tropical regions of Asia, Europe, America, Africa, and other areas of the world (3-6). It has a bitter flavor and is a member of the Gentianaceae(3) family. Mangiferin, amarogentin, and swertimarin are the main biologically active compounds found in the crude extract of the plant's various parts. Other secondary metabolites that contribute to the plant's pharmacological action include glycosides, xanthones, secoirioid, phenolics, alkaloids, flavonoids, triterpenes, tannins, carbohydrates, and sterols (7-8).

Research on Swertia chirata in experimental animals showed that it is effective in treating diabetes (9), hypertension(10), as an analgesic, anti-inflammatory(11), other studies included anticarcinogenic(12) anticonvulsant, sedative, anxiolytic (13) antioxidant , anti-AChE properties(14), antipyretic(15), anti-bacterial(3), antiviral(16), antiulcer(17), anti-helminthic(18), anti-hyperlipidemic(19)and hepatoprotective effect(20).

Individuals believe that herbal remedies are safe and are ignorant of the harmful impact they may have on various biological systems (21). As a result, information regarding the toxicity of therapeutic herbs must be investigated. The current study was designed to evaluate the effects of swertia chirata extract on various biochemical parameters in albino wistar rats, given the plant's usefulness in the treatment of many diseases.

2. Materials and Methods:

2.1 Collection and Identification of Plant:

The dried aerial parts of Swertia chirata was bought at the Karachi, Pakistan. The sample of the plant specimen was identified by Prof. Dr Mansoor Ahmad, Department of Pharmacognosy, University of Karachi and issued a voucher specimen no 20170303-1.

2.2 Procedure of extract preparation:

Using an electronic grinder, the 500 grams of dry plant material was ground into a fine powder. Methanol was used to macerate the coarse powder for 15 days at room temperature. Following filtering, the methanol extract was evaporated in a rotary evaporator and refrigerated until needed. To improve the extract's solubility in water, 10% dimethyl sulfoxide (DMSO) was used to reconstitute the thick gummy extract (13).

2.3 Animals handling and dietary protocol:

Adult male wistar albino rats (initially weighing between 180-210 gram) were selected for the present study. All animals were acclimatized one week before commencement of experiment and kept under controlled room temperature at 23 ± 2°C in 12 h light/dark cycles with free access to food and water. The experiment was performed in accordance with National Institute of Health (NIH) guidelines for the care and use of Laboratory Animals (National Research Council, 1996), after the approval from Ethical Review Committee (ERC) of Ziauddin University Karachi, Pakistan.

2.4 Grouping of animals:

The animals were divided into three groups of six rats in each group, as follows.

Group 1: Received DMSO 10% as placebo (10 ml/kg body weight)

Group 2: Received 250 mg/kg body weight of extract

Group 3: Received 500 mg/kg body weight of extract

All the animals were weighed before starting experiment. The methanol crude extract was administered according to their body weight once daily per oral by gastric intubation for 14 days.

2.5 Evaluation of Kidney and Liver function test

The blood samples (approximately 3ml) were collected to investigate the effects of herb on kidney and liver function test in a serum separating vacutainer tubes (containing serum separating gel and silica particles), then centrifuged at a rate of 4000 rpm (revolutions per minute) for 5 minutes. After that the supernatant serum was separated from the blood, collected in a labeled eppendorf and preserved in a refrigerator until 31 analytical measurements was carried out (22). The serum enzymes, alanine amino transferase (ALT), aspartate amino transferase (AST), alkaline phosphatase (ALP), total protein (TP), bilirubin total, urea and creatinine were measured using semi-automated chemistry analyzer (Microlab 200), according to the procedures described by Randox Laboratories Ltd, United Kingdom.

2.6 Statistical analysis:

Statistical analysis was carried out by using SPSS (Statistical Package for Social Sciences) version 20. Data is presented as mean and standard deviation / standard error of mean. The difference amongst three groups; control, low dose (250mg) and high dose (500mg) of *Swertia chirata* extract was measured by using one way analysis of variance (ANOVA) and where data violated the assumptions of normality, Kruskal Wallis H test (non-parametric test) was used, followed by pairwise comparison post hoc Tukey-test. A p-value of <0.05 was taken to indicate the statistical significance.

3. Results:

3.1 Effect of Swertia chirata extract at 250 mg/kg on kidney and liver function tests after treatment for 14 days

The effects of Swertia chirata at low dose (250 mg/kg) did not produce any toxicity to any biochemical parameters after administration of extract for 14 days. All biochemical parameters were not significantly different as compared to control (Table 1).

Table 1: Effect of Swertia chirata extract at 250 mg/kg on kidney and liver function tests after treatment for 14 days

Parameters Mean ± SD	Control	Low dose (SC 250 mg/kg)	P value	95% Confidence Interval	
				Lower	Upper
T. BIL (mg/dl)	0.5 ±0.2	0.5 ±0.08	0.9	-8.2	6.8
ALT (IU/L)	148 ± 28	148 ±19	0.8	-6.0	8.4
ALP (IU/L)	399 ± 111	376 ±122	0.8	-7.7	5.0
AST (IU/L)	197 ± 24	210 ±19	0.2	-2.3	9.6
T.P (g/dl)	6.7 ± 0.6	6.4 ±0.3	0.6	-10.4	5.1
Urea (mg/dl)	42.3 ± 2.4	41.6 ± 4.3	1.0	-18.3	15.7
CREAT (mg/dl)	0.7 ± 0.05	0.6 ±0.1	0.08	-10.3	0.6

*p-value<0.05 was considered significant, Data are expressed as Mean ± SD, n=6, Total bilirubin (T.BIL), Alanine aminotransferase (ALT), Alkaline phosphatase (ALP), Aspartate aminotransferase (AST), Total protein (T.P), Creatinine (CREAT), SC = *Swertia chirata*

3.2 Effect of Swertia chirata extract at 500 mg/kg on kidney and liver function tests after treatment for 14 days

The effects of Swertia chirata at high dose (500 mg/kg) did not produce any toxicity to any biochemical parameters after administration of extract for 14 days. All biochemical parameters were not significantly different as compared to control except ALP; ALP levels were significantly decreased as compared to the control (Table 2).

Table 2: Effect of Swertia chirata extract at 500 mg/kg on kidney and liver function tests after treatment for 14 days.

Parameters Mean ± SD	Control	High dose (SC 500mg/kg)	P value	95% Confidence Interval	
				Lower	Upper
T. BIL (mg/dl)	0.5 ±0.2	0.5 ±0.1	0.6	-10.1	4.9
ALT (IU/L)	148 ± 28	128 ±20	0.3	-11.1	3.3
ALP (IU/L)	399 ± 111	269 ±20	0.02*	-13.5	-0.7
AST (IU/L)	197 ± 24	177 ±14	0.1	-10.6	1.3
T.P (g/dl)	6.7 ± 0.6	6.5 ±0.5	0.9	-8.8	6.6
Urea (mg/dl)	42 ± 2.4	43 ± 1.5	0.9	-3.9	4.9
CREAT (mg/dl)	0.7 ± 0.05	0.7 ± 0.08	0.5	-3.1	5.6

*p-value<0.05 was considered significant, Data are expressed as Mean ± SD, n=6, Total bilirubin (T.BIL), Alanine aminotransferase (ALT), Alkaline phosphatase (ALP), Aspartate aminotransferase (AST), Total protein (T.P), Creatinine (CREAT), SC = *Swertia chirata*

4. Discussion:

Numerous Swertia species are used medicinally around the world, however because of its pharmacological qualities, Swertia chirata is regarded as the most more beneficial herb (3). The effects of Swertia chirata were observed at two doses, i-e 250mg/kg and 500mg/kg. The results of biochemical parameters showed that the bilirubin total, ALT, and AST did not alter significantly in both doses as compared to the control (Table 1 and 2). However, ALP levels were significantly decreased at 500mg/kg dose of Swertia chirata as compared to the control.

It could be due to the fact that Swertia chirata might causes the depletion of the magnesium or zinc levels in rats (23).

Aminotransferases, aspartate aminotransferase (AST) and alanine aminotransferase (ALT) are remarkable indicators of hepatocelluar injury. ALT, is a specific and sensitive marker for the liver, as mainly found in the liver and elevated levels indicate parenchymal injury of the liver. AST is present in skeletal and cardiac muscles, liver, kidney, lungs, leucocytes and red blood cells. Whereas, alkaline phosphatase (ALP) originates mainly form liver, bone and also found in kidney, intestine, leukocytes and placenta; the elevated levels of ALP, indicates metabolic stimulation of all these tissues. While, bilirubin is the end product of RBCs destruction; it helps to diagnose the problem related to liver disease, hemolytic anemia and blockage of the bile ducts (24). Our results indicate that methanolic extract of swertia chirata low as well as high dose, did not exhibit any remarkable hepatotoxic effects. The assessment of serum total proteins in Swertia chirata at low and high dose groups did not show any significant alteration as compared to control (Table 1, 2).Serum total proteins are measured to access overall health and normal functioning of liver, kidneys and muscles of body; any variation in their levels are closely correlated to alteration in catabolism, nutritional status, gastrointestinal and urinary loss (24). Similar to serum total proteins, the results of biochemical parameters, urea and creatinine did not show any significant variation in both doses as compared to control (Table 1, 2). Urea is the major metabolite derived from tissue protein turn over and dietary protein; While, creatinine is the end product of muscles metabolism. Being the main excretory organ of the body, kidney filters out most of the creatinine. Whereas, increased urea and creatinine levels in the serum may indicate impaired function or disease states of the kidneys due to the accumulation of these end products in blood (25). On the basis of above mentioned, our results indicate that the methanolic extract of Swertia chirata at low as well as high dose, did not exhibit any remarkable heptatotoxic and nephrotoxic effects.

Conclusion:

No significant difference was observed at high and low dose of *Swertia chirata* extract on different biochemical parameters of rats. Whereas, 500mg/kg dose of extract significantly decreased the value of ALP. Based on these facts, we can conclude that the *swertia chirata* may not have toxic effects on various biochemical parameters. However, further studies

should be carried out to evaluate the effect of *Swertia chirata* on various other biochemical parameters.

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