Chronic Toxicity of Leaf Extract from Sphagneticola trilobata (L.) Pruski

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ABSTRACT
Context: Sphagneticola trilobata (L.) Pruski. is a member of the family Asteraceae and has been traditionally used in the prevention and treatment of various diseases. Aim: The research was aimed to determine chronic toxicity of 80% ethanolic leaf extract from S. trilobata (STLE). Materials and Methods: STLE at the doses of 200 or 400 mg/kg b. w. was oral given to the healthy Wistar rats daily for 90 days. Statistical analysis used: Statistical analysis was carried out using F-test (One-Way ANOVA) followed by Duncan’s New Multiple Range Test. Results: STLE did not produce any signs or symptoms of chronic toxicity. And also, the mortal rat was not observed during a period of an observation. Furthermore, STLE did not alter the body weight, relative organ (liver, pancreas, kidney and heart) weight, hemoglobin (Hb), hematocrit (Hct), red blood cell (RBC), white blood cell (WBC), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), neutrophil, lymphocyte, monocyte, platelet, alkaline phosphatase (ALP), aspartate aminotransferase (AST), alanine aminotransferase (ALT), blood urea nitrogen (BUN), creatinine, blood cell characteristics, ultrastructure of RBC, and histological features of hepatic, pancreatic and renal tissues in the STLE treated rats comparing to control rats. Conclusions: These findings indicate that the leaf extract from S. trilobata exerts non chronic toxicity in rats and can be used safely as a traditional medicine or diet complement without any effect on hepatic and renal functions.

Key words: S. trilobata, Chronic toxicity, Hematological values, Blood biochemistry, Histological feature.

INTRODUCTION
Medicinal plants are the major resource of traditional medicines. Traditional and alternative medicines are extensively practiced in the prevention and treatment of various ailments.1

Sphagneticola trilobata (L.) Pruski or Wedelia trilobata (L.) A.S. Hitch., is a member of the family Asteraceae. Its leaves or aerial parts are used for backache, muscle cramp, rheumatism, sores, swelling and arthritic painful joints.2 The leaves and stems are used in childbirth and in the treatment of bites and stings, fever and infection,1 wound healing.2 S. trilobata has antioxidant,4 antibacterial,5 analgesic,6 and antimicrobial activities,7 and is a potential candidate in the management of diabetes.8 These pharmacological activities may depend on its phytoconstituents such as tannin, saponins, flavonoids, phenol and terpenoids.8

Despite non acute toxicity of the leaf extract from S. trilobata has been reported in our previous study.9 To see whether it is safe for long term administration, the present study was therefore, designed to determine the chronic toxicity of 80% ethanolic leaf extract from S. trilobata to Wistar rats for 90 days.

MATERIALS AND METHODS
Preparation of STLE
Fresh and mature leaves of S. trilobata were collected from the local gardens in Maha Sarakham and Roi-Ét Provinces, Thailand and identified by the Plant Varieties Protection Division, Department of Agriculture, Ministry of Agriculture and Cooperatives, Thailand. A voucher specimen (Code: MSU-Sci/PA001) is deposited in the Department of Biology, Faculty of Science, Mahasarakham University, Thailand. The plant leaves were washed with tap water, sliced into small pieces, air dried at ambient temperature and ground into powder. The plant powder was extracted by macerating in 80% ethanol for 7 days and the mixture was then filtered. The filtrate was evaporated in a rotary evaporator followed by freeze drying. The obtained powder extract (STLE) was stored at 4°C until be used.

Animals
The animals used in the study were albino Wistar rats weighing 180-200 g purchasing from the National Laboratory Animal Centre, Mahidol University, Thailand. The rats were housed in the clean cages under the conditions at 25 ± 2°C, 50 ± 5% RH with a 12 h D/L cycle in the animal laboratory at the Department of Biology, Faculty of Science, Mahasarakham University. All animals were given a standard laboratory diet and water ad libitum. The experimental protocol and performance of the rats were approved by the Institutional Ethical Committee for the Purpose of Use and Control, and Supervi-
RESULTS AND DISCUSSION

Body weight and Relative organ weight

STLE at the doses of 200 and 400 mg/kg b.w. did not produce any signs of toxicity (diarrhoea, sleep, lethargy, salivation, eyes, skin and fur) and mortality of the rats observed twice daily during a period lasting 90 days. Body weight of each rat was recorded weekly. The results revealed that the initial (day 0) body weight of controls and STLE treated rats was not different. The same result also occurred at the final (day 90) body weight (Table 1). Relative organ weight, the relationship between each organ weight and body weight, between controls and STLE treated rats was not different (Table 1).

Biochemical and hematological values

Glucone, BUN, creatinine, Uric, TP, Alb, Glob, TB, AST, ALT, and ALP from rats treated with different doses of STLE were not different from the normal controls (Table 2). The enzyme activities of AST, ALT, and ALP from controls and the rats received 200 or 400 mg/kg b. w. STLE were not different (Figure 1).

Histological examination

Pancreatic tissue: Light microscopic observation revealed that the islet of Langerhans, which is the round blind beginning of the nephron. It is invaginated by a reticulin network. Hepatocytes in the controls and the rats received 200 or 400 mg/kg b. w. STLE were not different (Figure 1).

Blood biochemistry and hematological values

Hb, Hct, RBC, MCV, MCH, MCHC, WBC, neutrophils, lymphocytes, monocytes and platelet from the rats treated with different doses of STLE and from controls were not different (Table 2).

DISCUSSION

Phyto-therapeutic using medicinal plants or plant products have become universally popular in primary healthcare, particularly in developing countries. The medicinal plants and plant products are presumed to be safe. Nevertheless, there is a lack of proven scientific studies on the toxicity and adverse and/or undesirable effects of these remedies. Therefore, oral chronic toxicity study of the leaf extract from S. trilobata was inves- tigated.

STLE at the doses of 200 and 400 mg/kg b.w. exerted non chronic toxicity on albino Wistar rats as they did not exhibit any signs of toxicity and mortality of the rats. Furthermore, the body weight, relative organ weight, blood biochemistry, hematological values, histological feature of white blood cells, and structural feature of red blood cells in all experimental rats and those from controls were not different.

The values represent the meanSEM within the same row followed by the different superscript letters are significantly different at the p<0.05. 
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CONCLUSION

CONFLICT OF INTEREST

None

ABBREVIATIONS USED


REFERENCES


TEM micrographs revealed that the ultrastructure of red blood cells STLE was not different from that in the controls. This study showed the same result as the extracts from Nelumbo nucifera flowers.

Figure 1: Light micrographs showed hepatic tissue (A), pancreatic tissue (B), renal tissue (C), lymphocytes (D), monocytes (E), neutrophils (F) and eosinophils (G).

Figure 2: Transmission Electron Micrographs showed lead citrate and uranyl acetate staining of the red blood cell ultrastructure in control (A), rats received 200 mg/kg b.w. STLE (B) and rats received 400 mg/kg b.w. STLE (C).

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