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Original Article

Acute and Subchronic Toxicity Study of Sampilnorov Wurile

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ABSTRACT

Introduction: Sampilnorov Wurile (SW) is a traditional medicinal preparation containing 29 plant-, mineral-, and animal-derived components. It is used for cerebrovascular ischemia, retinal disorders, cardiovascular diseases, and neurological conditions. This study aimed to evaluate the acute and subchronic toxicities of SW. **Methods:** Acute toxicity was assessed in C57BL/6 mice based on OECD-423 guidelines, with oral doses ranging from 500 to 6,000 mg/kg body weight. Subchronic toxicity was evaluated in Wistar rats following OECD-407 guidelines, with daily oral doses of 300 and 600 mg/kg of SW daily for 4 weeks. Clinical signs, mortality, body weight, and physical condition were monitored. Hematological, biochemical, and histopathological analyses were conducted on day 29. **Results:** No mortality or significant clinical signs of toxicity were observed at doses up to 6,000 mg/kg, suggesting an oral median lethal dose exceeding this level. Subchronic toxicity assessment revealed no clinical signs of toxicity or mortality. SW at 300 and 600 mg/kg had no significant effects on serum biochemical parameters or vital organ histology compared to controls. **Conclusion:** SW exhibited no acute toxicity at doses up to 6,000 mg/kg and showed no adverse effects in a 28-d subchronic toxicity study. These findings support its safety at tested doses.
Keywords: Acute and subchronic toxicity; biochemistry; blood test; histopathological analysis; Sampilnorov Wurile.

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INTRODUCTION

Mongolian traditional medicine has evolved through generations, shaped by the nomadic lifestyle and reliance on natural resources, such as plants, animals, and minerals. It remains an integral part of Mongolia's cultural identity and is widely practiced today.[1] The global market for herbal medicines continues to expand, as many individuals use these products for therapeutic and preventive purposes. Most natural medicines contain plant- and animal-based components, making it essential to evaluate their pharmacological effects, pharmacokinetics, adverse effects, and quality.

Sampilnorov Wurile (SW) is a traditional Mongolian formulation composed of 29 ingredients, including *Pterocarpus santalinus, Pteria martensii Dunker, Myristica fragrans Houtt, Bos Taurus domesticus, Rhinoceros unicornis L, Gardenia jasminoides Ellis* seeds, *Glycyrrhiza uralensis, Polygonum aviculare L, Cassia obtusifolia L, Liquidambar formosana H, Abutilon theophrasti Medic, Carthamus tinctorius L, Eugenia caryophyllata Thunb, Elettaria cardamomum L, Nigella glandulifera L, Lactuca sativa L, Piper longum L, Serje vemiculitum, Eriocheir sinensis H.M, Terminalia chebula Retz, Terminalia bellirica Roxb, Moschus moschiferus, Santalum album Z, Aquilaria agallocha Roxb, Cinnamomum cassia P, Saussurea lappa, Inula helenium L, Calcio sinter*, and *Amomum tsao-ko Grevost*.[2] SW is among the 10 most prescribed traditional medicines for treating brain ischemia, cerebrovascular stenosis-related headaches, dizziness, insomnia, stroke recovery, epilepsy, angina pectoris, facial paralysis, vasculitis, sciatica, rheumatism, rheumatoid arthritis, and retinal disorders.[3-7] A toxicity study is essential to ensure the safety of natural medicines, minimize adverse effects, determine optimal dosages, improve efficacy, mitigate risks associated with long-term use, enhance proper medicinal application, and meet international medication standards. This study aimed to evaluate the acute and subchronic toxicity of SW.

MATERIALS AND METHODS

SW was obtained from the Inner Mongolia International Mongolian Hospital (serial number M14010081) and prepared by grinding and mixing with distilled water.

Experimental animals

C57BL/6 mice (body weight: 22–25 g, age: 6–8 weeks) and Wistar rats (200–210 g) were procured from the Core Laboratory at the Mongolian National University of Medical Science and the Institute of Pharmacology at Monos Pharm Trade LLC. Animals were housed under a 12:12 h light–dark cycle at 22–24 °C with 50% humidity and had ad libitum access to food and water.

Ethical statement

The study protocol was approved by the Ethical Review Committee of the Mongolian National University of Medical Science (approval no. 3/3/2015/-03).

Acute toxicity study

The acute toxicity study was conducted in accordance with OECD guideline No. 423.[8] Healthy male mice were randomized into control and experimental groups. The control group received distilled water (10 mL/kg), while the experimental groups received

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SW at 500, 1,000, 2,000, 3,000, 4,000, 5,000, and 6,000 mg/kg. Physical condition and clinical signs of toxicity of the mice were monitored for 2 h, with mortality recorded at 24, 48, and 72 h.

Body weight was measured before treatment and on days 7 and 14. On day 14, all mice were euthanized, and a necropsy was performed. The liver, heart, spleen, lungs, and kidneys were weighed, and the relative organ weight was calculated using the following formula:

Relative organ weight = Absolute organ weight (g)/mouse body weight (g)×100%.

Subchronic toxicity study

The subchronic toxicity study was performed according to OECD guidelines test No. 407.[9] Wistar rats were randomly divided into three groups: the untreated healthy control group (n=10), which received distilled water daily; Wistar rats (n=10), which received SW at 300 mg/kg; and Wistar rats (n=10), which received SW at 600 mg/kg for 28 consecutive days. Clinical signs, mortality, body weight, and physical condition were monitored. Body weight was measured before treatment and on days 7, 14, 21, and 28 using a balance (OHAUS, USA). On day 29, rats were euthanized with an overdose of ketamine hydrochloride, and blood samples were collected via cardiac puncture for hematological and biochemical analyses. The tissues were excised and processed for histological examination using hematoxylin and eosin (H&E) staining.

Hematological analysis

Blood samples (3 mL) were collected in EDTA-anticoagulated tubes for hematological analysis. Hematological parameters were analyzed using an automated analyzer (Sysmex Poch 100 I, Japan).

Biochemical analysis

Blood samples (10 mL) were collected in serum separator tubes, centrifuged at 3,000 rpm for 10 minutes, and analyzed for alanine transaminase (ALT), aspartate transaminase (AST), alkaline phosphatase (ALP), urea, and creatinine using an automated biochemical analyzer (ICUBIO-iChem-520, Shenzhen, China).

Histopathology examinations

Liver, kidneys, heart, and stomach tissues were fixed in 10% buffered formaldehyde, embedded in paraffin, sectioned at 5 µm using a microtome, stained with H&E, and examined under a light microscope (Olympus BX41, Japan).[10,11]

Statistical analysis

Data were presented as mean ± SEM with a 95% confidence interval. Statistical analysis was performed using STATA-14. Group comparisons were conducted using ANOVA followed by Tukey's multiple comparison tests. Statistical significance was set at $p < 0.05$.

RESULTS

Acute toxicity study

All mice were observed for 24 h, 72 h, and 14 d post-treatment. The control group (Group 1) received 10 mL/kg distilled water orally, while the treatment group (Group 2) was administered a single oral dose of SW at 500, 1,000, 2,000, 3,000, 4,000, 5,000, and 6,000 mg/kg via gavage. Physical condition and clinical signs of toxicity were monitored for 2 h, and the results are presented in Table 1.

No signs of toxicity, such as diarrhea, excessive urination, salivation, locomotor defects, respiratory distress, or drowsiness, were observed after single doses of 500–6,000 mg/kg SW. No mortality was recorded (Table 2), indicating that the oral median lethal dose (LD_{50}) of SW exceeded 6,000 mg/kg.

According to Hodge and Sterner[12] and the OECD classification, SW is categorized as non-toxic.

Table 1. Effect of various Sampilnorov wurile dosages on C56BL/6 mice's behavioral, neurological, respiratory, physical, and clinical signs.

Clinical signs of toxicity	Group							
	Control	500 mg/kg	1,000 mg/kg	2,000 mg/kg	3,000 mg/kg	4,000 mg/kg	5,000 mg/kg	6,000 mg/kg
Sense	+	+	+	+	+	+	+	+
Breath	+	+	+	+	+	+	+	+
Movement	+	+	+	+	+	+	+	+
Salivation	+	+	+	+	+	+	+	+
Urination	+	+	+	+	+	+	+	+
Defecation	+	+	+	+	+	+	+	+
Convulsions	-	-	-	-	-	-	-	-
Tremors	-	-	-	-	-	-	-	-
Diarrhea	-	-	-	-	-	-	-	-
Edema	-	-	-	-	-	-	-	-
Drowsiness	-	-	-	-	-	-	-	-
Mortality	-	-	-	-	-	-	-	-

(+ normal; -no signs)

Table 2. Seventy-two-hour mortality assessment of Sampilnorov wurile's (SW) effects in mice.

Groups	Dose	Dead/Treated mice		
Control	DW (10 mL/kg)	24 h	48 h	72 h
	500 mg/kg	0/6	0/6	0/6
	1,000 mg/kg	0/6	0/6	0/6
SW	2,000 mg/kg	0/6	0/6	0/6
	3,000 mg/kg	0/6	0/6	0/6
	4,000 mg/kg	0/6	0/6	0/6
	5,000 mg/kg	0/6	0/6	0/6
	6,000 mg/kg	0/6	0/6	0/6

Body weights of mice that received SW at doses ranging from 500 to 6,000 mg/kg were measured on days 1, 7, and 14 and compared to baseline values (Figure 1).

Statistical analysis showed no significant differences in body weight between the experimental and control groups, suggesting that SW did not affect appetite or cause weight loss. No morphological abnormalities were observed in the liver, spleen, heart, lungs, or kidneys. Absolute and relative organ weights were recorded, and results are presented in Figures 2 and 3.

Administration of SW at 500–6,000 mg/kg did not significantly alter absolute or relative organ weights compared to the control group, suggesting no substantial effects on the examined organs.

Subchronic toxicity study

The body weights of the rats were measured on day 1, 7, 14, 21, and 28 following SW administration and were compared to baseline values and control group weights (Table 3).

SW administration for 28 d did not result in significant differences in body weight between the experimental and control groups. However, a statistically significant increase in body weight was observed within the control group between days 1 and 28.

Macrostructural examination of the liver, spleen, heart, lungs, stomach, and kidneys revealed no significant differences between the control and experimental groups receiving 300 and 600 mg/kg of SW.

Absolute and relative organ weights of the liver, spleen, heart, lungs, stomach, and kidneys were assessed and compared, with results presented in Figure 4.

SW administration for 28 d did not cause statistically significant differences in the absolute or relative weights of the liver, spleen, heart, lungs, or kidneys between the experimental and control groups. These findings suggest that long-term SW use did not substantially affect organ weights.

Table 3. Body weight of rats treated orally with SW for 28 d

Days	Body weight (g), (mean±SD)		
	Control group Distilled water 10 mL/kg	Sampilnorov Wurile 300 mg/kg	Sampilnorov Wurile 600 mg/kg
0 days	206.7±10.78	205.17±8.59	207.64±10.56
7 days	219.7±11.25*	213.2±9.13*	218.08±11.31*
14 days	230.24±8.93*	222.54±8.65*	227.23±9.27*
21 days	240.82±7.40*	232.54±8.59*	235.47±10.6*
28 days	248.22±7.68*	241.56±12.5*	245±9.86*

*p<0.05, compared to the first day.

Table 4. Sampilnorov wurile (SW) effects on hematological parameters of rats in a subchronic toxicity study.

Parameters	Groups		
	Control	SW 300 mg/kg	SW 600 mg/kg
WBC, 10^3/μL	5.54±0.77	5.75±1.08	5.64±0.71
RBC, 10^6/μL	7.74±0.71	7.79±0.54	7.61±0.30
HGB, g/dL	14.8±1.17	14.8±0.86	14.51±0.76
HCT, %	42.33±3.28	42.24±3.01	40.91±2.18
MCV, fL	54.76±2.66	54.2±1.96	53.75±2.69
MCH,pg	19.2±0.73	19.01±0.76	19.05±1.14
MCHC, g/dL	35.06±0.79	35.08±0.72	35.49±0.70
PLT, 10^3/μL	740.7±124.81	702.9±132.57	669.8±201.04
LYP,10^3/μL	3.52±1.45	4.35±1.49	3.91±1.02
MID, 10^3/μL	0.82±0.18	0.86±0.35	0.87±0.33
GRA,%	21.13±10.27	14.15±3.59	17.15±5.80
RDW,%	10.75±0.77	10.6±0.74	10.71±0.70
RDW-SD,%	23.9±1.57	23.8±1.51	23.4±2.49
MPV,fL	8.9±0.41	8.43±0.34	8.8±0.68
PDW, %	28.6±5.53	26.5±4.23	27.2±5.1
PLCR,%	10.73±2.31	8.33±1.75	9.9±3.87
PCT, %	0.65±0.08	0.60±0.10	0.61±0.14

WBC: white blood count, RBC: red blood cell, HGB: hemoglobin, HCT: hematocrit, MCV: mean corpuscular volume, MCH: mean corpuscular hemoglobin, MCHC: Mean corpuscular hemoglobin concentration, PLT: platelet,

LYP: lymphocyte count, MID: middle cells (monocytes, eosinophils and basophils), GRA: granulocye, RDW: red cell distribution width RDW-SD: red cell distribution width-standard deviation, MPV: mean platelet volume, PDW: platelet distribution width

PLCR: platelet-larger cell ratio, PCT: plateletcrit

Table 5. Sampilnorov wurile (SW) effects on biochemical parameters of rats in the subchronic toxicity study.

Parameters Groups	Alanine transaminase, U/L	Aspartate transaminase, U/L	Alkaline phosphatase, U/L	Urea mmol/L	Creatinine μmol/L
Control	52.27±6.44	109.85±11.64	135.7±22.12	3.5±0.75	38.3±3.34
SW 300 mg/kg	56.19±5.81	113.3±11.93	131.4±18.4	3.36±1.12	38.36±2.73
SW 600 mg/kg	59.01±6.84	121.4±12.41	132.9±23.18	3.61±1.45	39.12±5.33

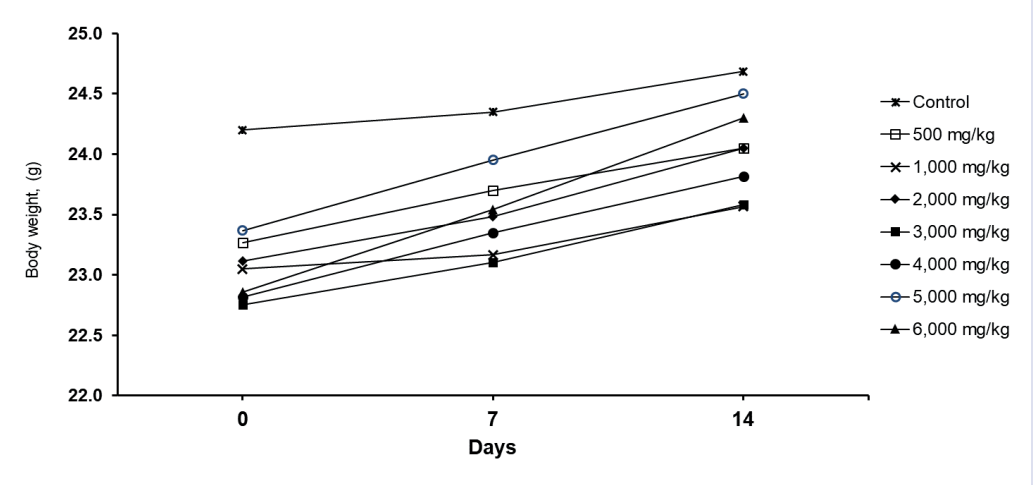

Figure 1. Body weights measurements in mice administered Sampilnorov wurile.

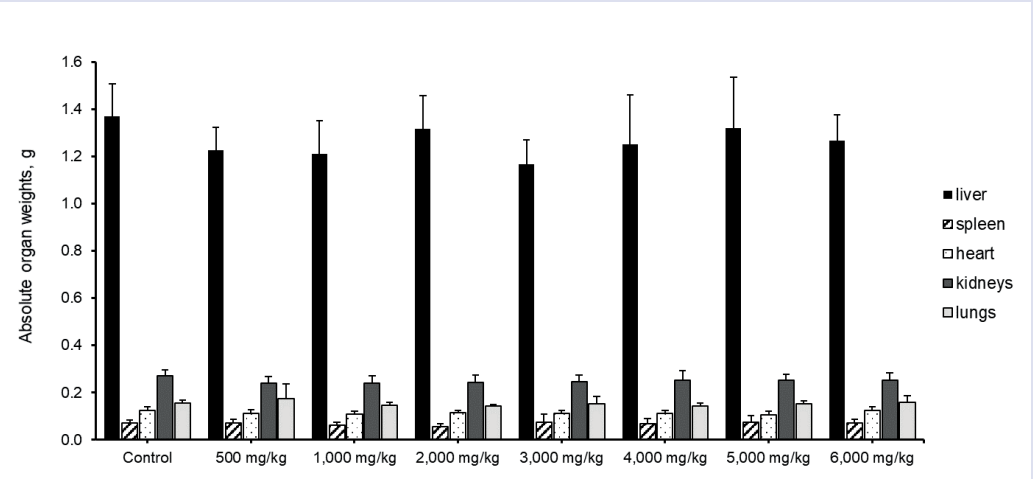

Figure 2. Absolute organ weights of mice treated with SW for 14 d.

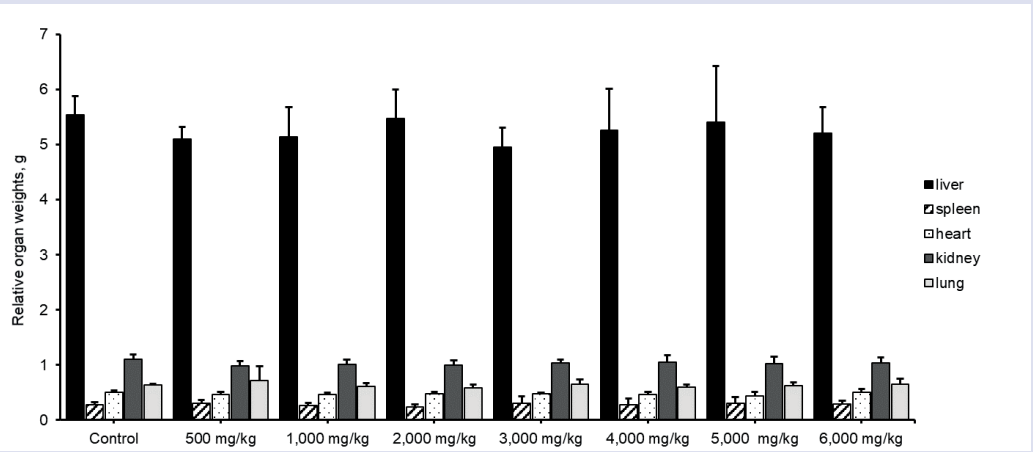

Figure 3. Relative organ weights of mice treated with SW for 14 d.

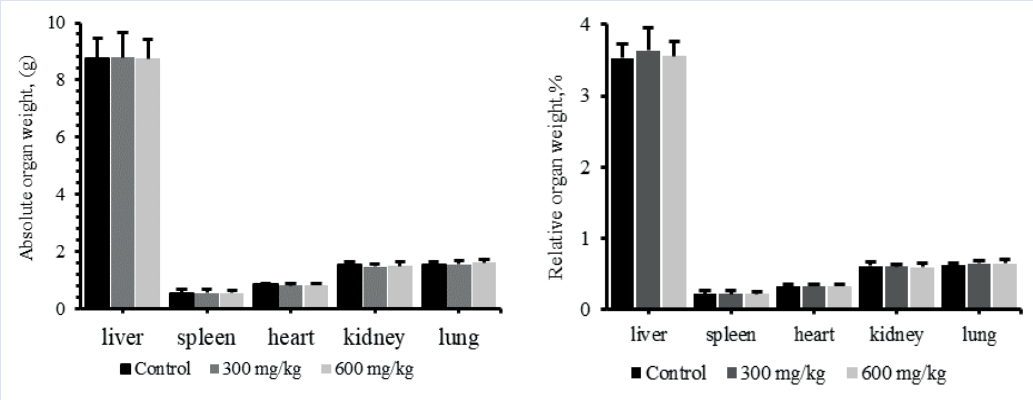

Figure 4. Absolute (a) and relative (b) organ weights of rats treated with Sampilnorov wurile for 28 d.

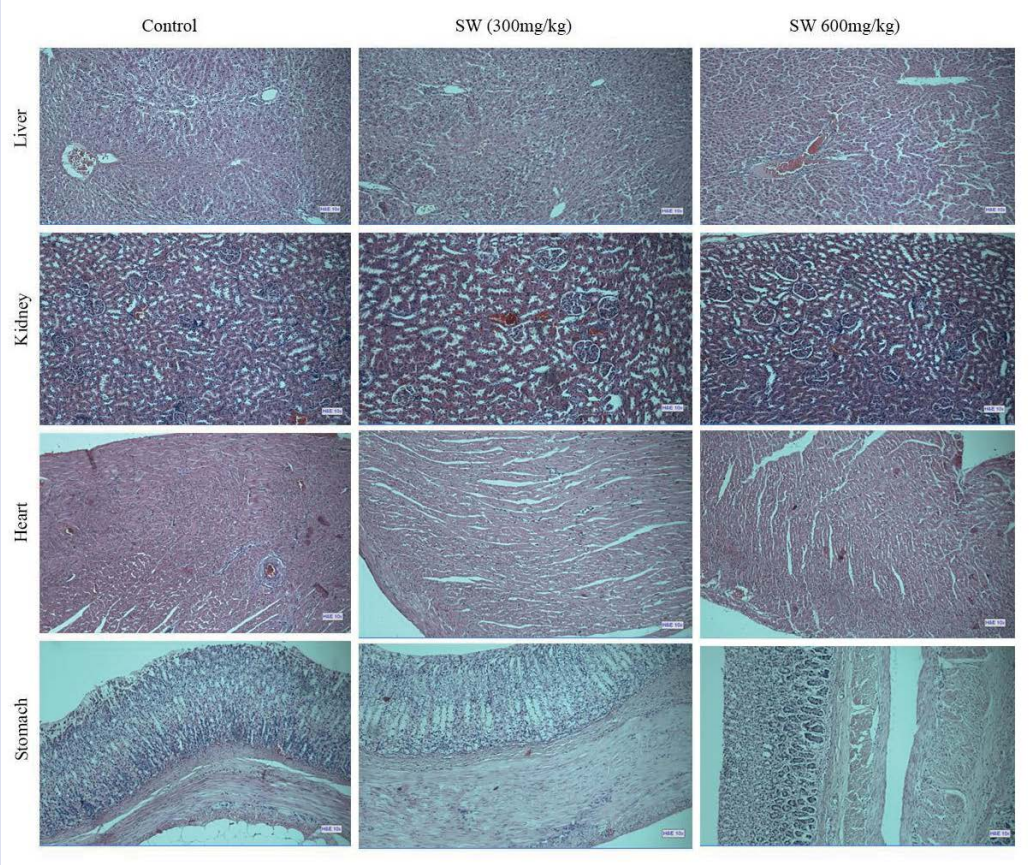

Figure 5. SW effects on the liver, kidney, heart and stomach in histomorphology of rats

SW: Sampilnorov wurile
hematoxylin-eosin, ×10.

Hematological analysis of the rats treated with 300 and 600 mg/kg of SW showed no statistically significant differences in key blood parameters, including red and white blood cell counts, platelets, hemoglobin, lymphocytes, and hematocrit, compared to controls (Table 4).

Similarly, biochemical analysis revealed no significant differences in liver function markers (AST, ALT, ALP) or kidney function markers (creatinine and urea) between the experimental and control groups after 28 d (Table 5).

These findings suggested that prolonged SW exposure did not induce hepatotoxicity or nephrotoxicity.

Histopathological study

Microscopic examination of liver sections showed no necrosis, fatty degeneration, inflammation around portal areas, or fibrosis in untreated control rats or those administered 300 mg/kg of SW. In the 600 mg/kg group, mild inflammation was observed around the portal veins, although necrosis, fatty degeneration, and fibrosis were absent. Kidney tissue analysis revealed a normal glomerular structure with no focal or diffuse necrosis in proximal or distal tubules. Histological assessment of the cardiac muscle showed normal cardiomyocyte morphology, with no pathological changes in the control or treatment

groups. Stomach histology showed no pathological alterations in the control or experimental groups treated with 300 and 600 mg/kg of SW (Figure 5).

DISCUSSION

Since ancient times, preparations derived from plants, animals, and minerals have been used to prevent and treat diseases. Traditionally, these natural products were considered safe but were often used without scientific validation or documented evidence of potential toxicity.[13] Paracelsus, the father of toxicology, stated, "All substances are poisons; there is none which is not a poison. The right dose differentiates a poison from a remedy."[14] Toxicological research is fundamental to pharmacology, providing essential information on the safety and efficacy of substances.[15] Regulatory authorities worldwide mandate toxicological evaluations of medicinal products before clinical application.[16]

Toxicity studies assess mortality and toxic doses while identifying direct and indirect effects on the body, making it essential for evaluating safety and potential risks. This study investigated the acute and subchronic toxicity of SW, a formulation containing 29 raw materials from plants, minerals, and animals.

The acute toxicity of several SW components has been previously reported. For example, oral administration of *Pterocarpus santalinus* water extract at 1 or 2 g/kg and intraperitoneal injection at 0.5 or 1 g/kg in Swiss albino mice did not cause mortality within 72 h or induce central nervous system effects within the first 3 h.[17]

Pearls have historically been considered safe in ancient Chinese medical texts. Kai Bao Ben Cao (974 AD) described them as "non-toxic," while Shao Xing Ben Cao (1159 AD) classified them as "slightly cold, non-toxic." In toxicity studies, mice administered high doses of water-soluble pearl powder, pearl powder, and dyed black pearls (10, 15, and 10 g/kg, respectively) exhibited no signs of poisoning, and no mortality occurred within 14 d.[18]

Similarly, *Myristica fragrans Houtt* seed extracts in methanol or n-hexane did not induce mortality or behavioral toxicity within 24 h at doses up to 5,000 mg/kg body weight.[19] In acute toxicity studies, the oral LD_{50} of *Bos Taurus domesticus* in mice exceeded 15 g/kg.[20] *Gardenia jasminoides* fruit extract administered to rats at 15 g/kg over 14 d showed no abnormalities, with an LD_{50} exceeding 15 g/kg.[21,22]

A previous study reported similar findings. Rats administered *Glycyrrhiza uralensis* root extract orally at 2,000 mg/kg showed no toxicity or mortality, classifying it as non-toxic.[23] Rats administered *Polygonum aviculare* flavonoids at 300, 2,000, or 5,000 mg/kg showed no mortality or behavioral changes.[24] Similarly, oral administration of *Liquidambar formosana H* extract at 0.25–2 g/kg to Balb/c mice showed no toxicity or mortality.[25] Acute toxicity experiments in mice determined that the LD_{50} of *Eugenia caryophyllata* essential oil or eugenol exceeded 4,500 mg/kg,[26] while a 2 g/kg dose of *Nigella glanduliferia L* water extract did not affect behavior or growth.[27] The LD_{50} of *Piper longum L* ethanol extract exceeded 2,000 mg/kg.[28] Additionally, rats orally administered 5,000 mg/kg of *Terminalia chebula* water extract exhibited no behavioral abnormalities or mortality, with histopathological analysis confirming normal organ structures.[29,30]

Similarly, *Terminalia bellirica Roxb* ethanol extract at 5,000 mg/kg was classified as non-toxic.[31] Following injection of *Moschus moschiferus* water extract and extracted ketone into the mouse tail vein, the LD_{50} values were found to be 848 mg/kg and 152 mg/kg, respectively.[32] The LD_{50} of *Saussurea Lappa* roots ethanol extract exceeded 5,000 mg/kg.[33]

Our study determined that the oral LD_{50} of SW exceeded 6,000 mg/kg, classifying it as non-toxic according to Hodge and Sterner and the OECD classification.

These findings align with those of previous studies on the toxicity of SW components. Subchronic toxicity studies are essential for identifying potential long-term adverse effects, particularly for traditional herbal medicines used over extended periods.[9] In this study, rats received SW at 300 and 600 mg/kg for 28 consecutive days, during which the rats' physical condition, body weight, biochemistry, hematology, and histopathology were evaluated. No mortality or signs of toxicity were observed throughout the study.

Body weight is a critical parameter in toxicity studies, as weight loss may indicate reduced appetite or metabolic dysfunction.[34] In this study, body weight gain was normal across all SW-treated and control groups. Organ weight, whether absolute or relative, is an important indicator of systemic toxicity, particularly in metabolically active organs, such as the liver, kidneys, spleen, heart, and lungs.[35] No significant differences were found in the absolute or relative organ weights between the control and treatment groups.

Hematological parameters are essential for assessing systemic toxicity, as blood plays a critical role in transporting microelements and substrates to organs.[36] As hematological changes are strong predictors of human toxicity, animal studies often evaluate blood components to assess chemicals effects. In this study, key hematological parameters, including hemoglobin, white blood cell counts, and platelets, did not differ significantly between SW-treated and control rats, suggesting that SW did not induce anemia or other blood disorders.

Biochemical markers, such as urea and creatinine, reflect renal filtration efficiency,[37] while liver enzymes (ALT, AST, ALP) indicate hepatocellular function and bile secretion.[38] No significant changes in these parameters were observed between SW-treated and control rats, suggesting that SW did not induce hepatotoxicity or nephrotoxicity and maintained normal liver and kidney function at the tested doses. Toxic substances can disrupt cellular metabolism, impair enzyme activity, and alter protein, lipid, and carbohydrate structures, ultimately leading to cell dysfunction.[39] However, our findings suggest that SW did not exert such harmful effects.

Oral administration of SW at 300 mg/kg did not induce harmful or abnormal changes in the kidney, heart, stomach, or liver tissues, as confirmed by biochemical and histopathological analysis. Rats administered 600 mg/kg SW exhibited mild liver inflammation and a slight increase in ALT and AST levels; however, these changes were not statistically significant compared to controls.

The subchronic toxicity of individual SW components has been reported in previous studies. Rats administered water-soluble pearl powder for 30 d exhibited normal body weight with no changes in hematology, biochemistry, or histopathology.[18] In contrast, histopathological analysis of rats administered 1,000 mg/kg of *Myristica fragrans* methanol and n-hexane extracts for 14 d revealed hepatotoxicity attributed to myristicin in nutmeg.[19] Similarly, *Gardenia jasminoides* extracts demonstrated varying toxicity effects.[21,22] While no mortality was recorded after administering 0.5–1.5 g/kg of *Gardenia jasminoides* Ellis fruit extract to Sprague Dawley rats for 90 d, an aqueous extract at 30 g/kg for 14 d led to increased liver weight and hepatotoxicity due to geniposide accumulation.[21,22]

Toxicity evaluations of *Glycyrrhiza uralensis* root extract at 50–1,000 mg/kg over 120 d showed no evidence of toxicity, mortality, or hematological, biochemical, or histopathological changes.[23] Similarly, rats receiving 1 mL of *Polygonum aviculare L* extract orally for 10 d exhibited no pathological alterations in the liver, kidneys, intestines, hypothalamus, or adrenal glands.[24] Pregnant mice injected intraperitoneally with *Carthamus tinctorius L* methanol extract at 10–40 mg/kg for 25 d showed no hematological changes.[40] However, at 40 mg/kg, hepatic and renal interstitial inflammation was observed.

In contrast, prolonged administration of *Eugenia caryophyllata* Thunb essential oil at 400 mg/kg reduced body weight, but relative organ weights and histopathology remained normal.[26] These findings align with those of our study, which determined SW to be non-toxic.

CONCLUSIONS

In the acute toxicity study, the LD_{50} of SW exceeded 6,000 mg/kg, designating it as practically non-toxic according to OECD guidelines. In the subchronic toxicity study, no mortality, clinical signs of toxicity, or significant changes in hematological, biochemical parameters, and histopathological parameters were observed following 28 d of SW administration at 300 and 600 mg/kg. These findings suggest that SW has a low toxicity profile.

Further chronic toxicity studies on SW would be beneficial for a comprehensive evaluation of its safety.

AUTHOR CONTRIBUTIONS

Conceptualization, L.Ch. and Ye.Ch.; methodology, Ch.Ts. and E.G.; software, E.G., M.G., B.B. and L.Ch.; validation, L.Ch., E.G., and Ye.Ch.; formal analysis, L.Ch., E.G.; investigation, L.Ch. and Ye.Ch.; resources, L.Ch., D.D.; data curation, L.Ch., E.G., Ch.Ts., and Ye.Ch.; writing—original draft preparation, L.Ch., E.G., and Ye.Ch.; writing—review and editing, L.Ch., E.G., and Ye.Ch.; visualization, B.B., M.G., M.B, U.A., N.M; supervision, E.G., and Ye.Ch.; funding acquisition, L.Ch. All authors have read and agreed to the published version of the manuscript.

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INSTITUTIONAL REVIEW BOARD STATEMENT

Protocols for the animal study were approved by the Ethical Review Committee of the Mongolian National University of Medical Science (№3/3/2015/-03).

INFORMED CONSENT STATEMENT

Not applicable.

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CONFLICTS OF INTEREST

The authors declare no conflicts of interest.

ABBREVIATIONS

The following abbreviations are used in this manuscript:

SW	Sampilnorov Wurel
OECD	Organisation for Economic Co-operation and Development
ALT	Alanine Transaminase
AST	Aspartate Transaminase
ALP	Alkaline Phosphatase
EDTA	Ethylenediaminetetraacetic Acid
SEM	Standard Error of the Mean
ANOVA	Analysis of Variance
LD_{50}	Median lethal dose
H&E	Hematoxylin and Eosin

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