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Original Article

An Evaluation of the Oral Toxicity of Shwaskas Chintamani Rasa in Wistar Rats for 90-Days Repeated Administration

Mahalaxmi Mohan[1,*], Shubhangi Hemraj Pawar[1], Sanskruti Sanjay Kashmire[1], Shishir Purushottam Pande[2], Rajshree Abhay Kulkarni[2], Abhay Narayan Kulkarni[2]

[1]Department of Pharmacology, Mahatma Gandhi Vidyamandir's Pharmacy College, Panchavati, Nashik, Maharashtra, INDIA.
[2]Department of Rasashastra, A.S.S. Ayurved Mahavidyalaya, Nashik, Maharashtra, INDIA.

ABSTRACT

Objectives: Toxicological research employing Herbo-Metallic (HM) medicine is essential to confirm that such medication has no harmful effects on humans. Despite the fact that the mercury level in Shwaskas Chintamani rasa which is used for respiratory infections has generated safety concerns, there are currently no scientifically validated data on toxicological features and the safety of the drug. As a result, the current study on Shwaskas Chintamani rasa used a sub-chronic study with albino Wistar rats to characterize its toxicological effects. The purpose of this study was to evaluate the toxicity of Shwaskas Chintamani Rasa in Wistar rats following repeated oral administration for 90 days. **Materials and Methods:** Wistar rats (160-180 g) were divided into four groups (*n=6*), each containing an equal number of male and female rats. Group II, III and IV were administered with low (51.36 mg/kg, p.o.), moderate (205.44 mg/kg, p.o.), or high (513.6 mg/kg, p.o.) doses of the test drug, respectively, for 13 weeks. The control group received the vehicle (2:3 of honey: water). Physical examination for body weight, food and water intake was carried out. Urine analysis was performed one week before sacrifice. On the 91st day, the animals were sacrificed and hematological, biochemical parameters, organ histology was examined. **Results:** Study revealed that there were no apparent harmful effects or deaths from Shwaskas Chintamani rasa in 3 dose levels. **Conclusion:** Thus, this research provides scientific evidence about safety of Shwaskas Chintamani rasa as it does not produce any toxicity in Wistar rats and may be clinically used to treat pregnancy-related illnesses.

Keywords: Blood Analysis, Chronic, Gross Pathology, Shwaskas Chintamani, Toxicity.

Correspondence:
Dr. Mahalaxmi Mohan
Department of Pharmacology, Mahatma Gandhi Vidyamandir's Pharmacy College, Panchavati, Nashik-422003, Maharashtra, INDIA.
Email: mm_nasik@yahoo.co.in
ORCID: 0000-0003-0326-0222

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INTRODUCTION

Ayurveda is a traditional science of healing system that has been practiced for centuries and is now accepted on a global scale. The current top draws in the health care and personal care sector include ayurvedic medications, nutraceuticals and dietary supplements. Due to consumer awareness of the negative side effects of synthetic drugs, there has been a movement in customer preference toward Ayurveda products.[1] Several ayurveda medications are designed as Herbo-Metallic (HM) formulations due to their numerous advantages, including increased stability, simplicity in storage, reduced dosage and ongoing availability.[2] Herbo- Metallic preparations are developed by integrating herbal components with heavily processed, biocompatible, non-metallic forms of heavy metals and minerals.[3] In order to rule out any adverse consequences, their thorough scientific investigation has been drawn to the rising usage and presence of heavy metals in ayurveda remedies.[4] An ayurvedic HM drug Shwaskas Chintamani Rasa is a mercury-based drug, hence named Kalpa. Shwaskas Chintamani Rasa also contains purified mercury, iron, mica, cow urine and a number of medicinal compounds. According to the Indian ayurveda regimen, sulfur is added to mercury and triturated without the addition of any liquid to create kajjali, a fine black powder. It has been discovered that mercury's inherent poisonous properties account for its toxicity rather than any impurities that may be present in it. Due to its anti-microbial characteristics, mercury was originally widely employed in western medicine; however, due to its toxicity and health hazards, its usage in health goods has since been questioned. Also, it was noted that quality control was a very difficult task in India because of the absence of post-market surveillance and the paucity of research studies on ayurveda medications.[5] Ayurveda experts support the idea that some hazardous medications and metals that are commonly utilized in Rasashastra undergo shodhana for purifying or detoxification in order to improve their medicinal properties. Toxicological research using HM medication is required to confirm the fact that such medication does not cause any hazardous effects on human beings. As a

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result, several toxicity studies on the safety evaluation of different HM medications need to be performed.[6] There are currently no scientifically validated data on toxicological characteristics and the safety of the medicine, despite the fact that Shwaskas Chintamani rasa's mercury level has raised safety concerns. As a result, the current investigation on Shwaskas Chintamani rasa was conducted to characterize its toxicological effects utilizing a sub-chronic study that involved albino Wistar rats receiving three doses of the drug orally every day for 90 days before undergoing toxicity testing for various organs, particular enzymes. A series of biochemical measurements, including those for the enzymes alanine amino transferase and aspartate amino transferase were made in serum. In conclusion, we conducted toxicity testing and produced 90 days toxicity data on Shwaskas Chintamani rasa during sub-chronic exposure *in vivo* to gain a thorough understanding of Shwaskas Chintamani rasa safety and support ayurvedic medication management.

MATERIALS AND METHODS

Test Formulation

The Ayurveda Seva Sangha provided the certified raw materials (Table 1) for the test formulation Shwaskas Chintamani Rasa.[7] After extensive drying in the shade, the individual ingredients were transformed into coarse powder. To create a uniform blend, all the powders were completely combined in the designated ratios. The combination was then transformed into granules with the use of granulation equipment and 500 mg tablets were created in a tablet manufacturing machine and used in the experimental study.

Chemicals

Anesthetics like Ketamine (60 mg/kg, i.p.) and Xylazine (16 mg/kg, i.p.)[8] were purchased from Unitech Life Science (Kandivali west, Mumbai, Maharashtra, India). All other chemicals were of reagent grades and obtained from Modern Science Apparatus Pvt. Ltd., (MIDC Satpur, Nashik, Maharashtra, India).

Animals Selection

Albino Wistar rats of 7-8 weeks old of either sex weighing 160-180 g were procured from the National Institute of Biosciences (Bhor, Pune, Maharashtra, India).[9] They were fed with Nutrimix std.1020 rat pellet feed supplied by Nutrivet Life Sciences while confined to huge, airy polypropylene cages and purified water was given *ad libitum*. Before the experiment began, the animals were accustomed to the environment for at least a week in conventional laboratory circumstances, which included 12 hr of dark and light cycles, a temperature of 21°C±3°C and 50%-60% humidity. According to the recommendations of the Committee for Control and Supervision of Experiments on Animals, the institutional animal ethics committee had approved the experimental protocol (Approval number: MGV/PC/CPCSEA/XXXIX/01/2022-23/03, Date: 19/09/22).

Dose Fixation

The traditional text mentions the following medicinal dosage of Shwaskas Chintamani Rasa: (Approx.500 mg). Using the Paget and Barnes standard table, the same dose was converted to an animal dose based on the body surface area ratio. Based on this, it was determined that the rat dose was 51.37 mg/kg body weight.[10] This was considered as low-dose. As per OECD guideline 408, the moderate dose and high dose were categorized as 205.44 mg/kg and 513.7 mg/kg.[11] The test formulation was made into a uniform suspension with the appropriate concentration depending on the body weight of the animals and supplied orally with the aid of oral gavage in a mixture of honey and distilled water in a ratio of (2:3).[12]

Experimental Protocol

The four groups, each with six animals of either sex, were formed by randomly selecting the animals. Group I, the control group, was given honey and water (2:3) as vehicle. Group II (Low): Shwaskas Chintamani Rasa (51.36 mg/kg, p.o.) for 13 weeks. Group III (Moderate): Shwaskas Chintamani Rasa (205.44 mg/kg, p.o.) for 13 weeks. Group IV (High): Shwaskas Chintamani Rasa (513.7 mg/kg, p.o.) for 13 weeks. These doses are one, four and ten times the converted suggested therapeutic amount for humans.[13]

The test drug was administered to respective groups for 90 consecutive days. Water and food intake was evaluated daily and body weight was measured weekly. The following profiles were evaluated.[11]

- Behavioural Profile: Awareness, Mood, Motor Activity.
- Neurological Profile: Central excitation, Motor incoordination, Muscle tone, Reflexes.
- Autonomic Profile: Optical signs, Secretory signs, General signs.

On the 87th day, urine was collected for urine analysis using metabolic cages. On the 91st day, under the anesthesia of ketamine and xylazine (60 mg/kg i.p) and (16 mg/kg i.p) blood was drawn from fasted rats by cardiac puncture.[14] The collected blood sample was centrifuged at 3000 rpm for 10 min and used for serum biochemistry.[15] Blood collected in anti- coagulant, ethylenediamine tetra-acetic acid tubes was fed into an auto cell analyzer (Erba Mannheim (H 360), which was automatically drawn into the device to estimate various parameters, including hemoglobin content, total Red Blood Cell (RBC) count, total White Blood Cell (WBC) differential WBC count and platelet count.[16]

The following serum chemistry parameters were evaluated: Total bilirubin, Aspartate aminotransferase, Alanine transaminase, Glucose, Total Cholesterol, Serum triglycerides, High-density lipoprotein, Low-density lipoprotein, Very low-density lipoprotein, Total protein, Albumin and Globulin, A/G ratio, Blood Urea, Serum creatinine, BUN.[17] On the 91st day, the animals were weighed and sacrificed. The liver, brain, heart, lungs, pancreas and kidney, among other significant organs, were removed, cleaned of extraneous tissue and weighed. All organs underwent normal processing with 10% buffered formalin fixation and paraffin embedding. According to protocol, 4 µm paraffin slices were cut on glass slides and stained with hematoxylin and eosin. The slides were examined under a light microscope at 400 magnifications to document changes in the microscopic features of the tissues under study.[18]

Statistical Analysis

Results are expressed as mean±SEM and analyzed by Graphpad Prism software version 9.3.2. Biostatistical analysis was done with a one-way ANOVA test followed by Dunnett's multiple tests for comparison.

RESULTS

Body Weight

During the course of 90 days, the Low, Moderate and High groups all exhibited a typical, progressive increase in body weight identical to the Control group (Table 2).

Water and Food Intake

As compared to the control group, daily food and water intake was not significantly changed in Shwaskas Chintamani rasa-treated groups (Figures 1 and 2).

Hematological and Biochemical Analysis

Total WBC count was non-significantly increased in the Low, Moderate and High doses of Shawaskas Chintamani Rasa (Table 3).

When compared to the control group, the low group's chloride percentage non-significantly decreased as a result of the administration of Shwaskas Chintamani Rasa. In the low group of Shwaskas Chintamani rasa compared to a control group, SGPT and SGOT levels were non- significantly elevated (Table 4).

Urinalysis

No anomalies were discovered in any of the urine parameters examined on day 83 in the treatment groups when compared to the control group, including appearance, volume, pH, protein, glucose and blood.

Organ weight

Administration of Shwaskas Chintamani Rasa leads to a non significant increase in the relative weight of the Brain, Heart, Lungs, Liver, Pancreas and Kidney of Wistar rats when compared to the control group (Table 5).

Histopathological evaluation

There were no observable major pathological abnormalities related to the therapy during the necropsy. At all dose levels, the

Table 1: Composition of Shwaskas Chintamani Rasa.

Sl. No.	Name of Content	English/Official Name	Part Used	Proportion
1	Shuddha Parada	Purified mercury	NA	10.87 mg
2	Suvarnamakshik Bhasma	Copper iron sulphide	NA	10.87 mg
3	Suvarna Bhasma	Gold particles	NA	10.87 mg
4	Mouktik Bhasma	Pearl Particles	NA	5.43 mg
5	Shuddha Gandhak	Purified Sulphur	NA	21.74 mg
6	Abhrak Bhasma	Biotite Mica	NA	21.74 mg
7	Loha Bhasma Bhavanadravya Kantakarirasa	Iron Particles		43.48 mg
8	-	*Solanum xanthocarpum*	Whole Plant	q.s.
9	Bakrichadudha	-	NA	q.s.
10	Decoction of Jeshthmadh	*Glycerrhiza glabra*	Roots	q.s.
11	Nagawell rasa	Piperbetle	Leaf	q.s.

NA-Not Applicable.

Table 2: Effect of Shwaskas Chintamani Rasa on Body Weight of Wistar Rats.

Weeks	Group I (Control)	Group II (Low)	Group III (Moderate)	Group IV (High)
Week 1	152±8.53	146.7±5.15	144.1±3.66	143.3±4.57
Week 2	163.7±8.64	152.5±6.23	149.7±3.28	148.5±5.75
Week 3	164.6±9.32	156.5±6.43	155.4±3.96	154.2±3.96
Week 4	186.2±10.78	163.2±6.65	161.8±3.70	161.8±8.93
Week 5	184.7±10.89	172.3±6.09	176.5±5.25	174.8±9.28
Week 6	177.2±10.74	174.5±8.54	185.1±6.41	179.4±8.59
Week 7	179.2±11.39	183.4±11.00	193.1±8.76	180.3±7.27
Week 8	217.3±13.49	199±12.00	192.8±8.40	187.5±5.83
Week 9	230.2±14.49	204.7±12.65	200±8.01	191.3±7.84
Week 10	233.2±17.72	211.3±14.19	215±8.78	201.5±8.66
Week 11	244.2±18.25	212.2±15.07	218.5±10.11	214.3±10.23
Week 12	244.8±18.38	215.6±16.52	223.2±11.12	230.3±12.67
Week 13	222.8±17.37	224.3±16.45	219.2±9.70	219.3±11.81

Note. Values are mean±SEM of six animals per group.

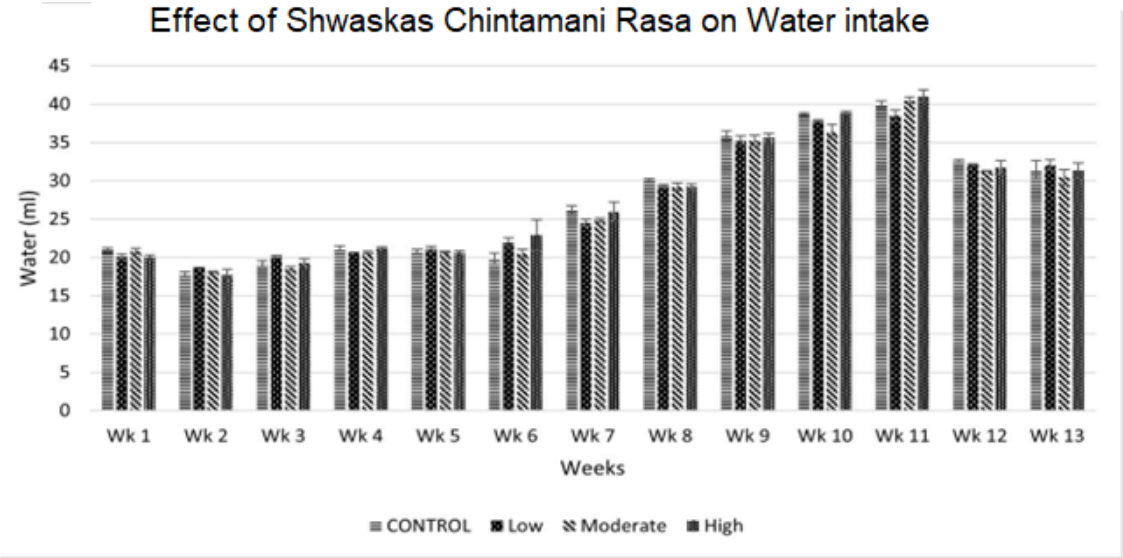

Figure 1: Water Intake following 13 weeks of administration of Low, Moderate and High Doses of Shwaskas Chintamani Rasa.

test medication did not result in any appreciable alterations in the cytoarchitecture of any organ examined (Figure 3).

DISCUSSION

In light of the fact that Complementary and Alternative Medicines (CAMs) like herbal remedies are becoming more widely used worldwide, therefore a comprehensive and thorough study of their effectiveness and safety is required.[18] There are many reasons for the rising use of CAM, including the shortcomings of current therapies and the adverse effects of prescription medications. The most significant aspect of CAM is traditional herbal medicine, which has been used for thousands of years. Consumers continue to have concerns about the efficacy and safety of traditional herbal remedies, despite the fact that there are many of them available and some of them have been supported by clinical investigations.[19]

Ayurvedic medicine Shwaskas Chintamani Rasa, which comes in tablet or pill form, is used to treat respiratory illnesses. Because this medication includes heavy metals, it is best effective when used under strict medical care. The most frequent usage of this medication is in north Indian ayurvedic treatment. It is used in the treatment of breathing disorders such as bronchitis, bronchial asthma, bloodless cough and others. It is also used as an anti-inflammatory, analgesic, anti-hypoxic and anti-diarrheal.[20]

The study was constructed in accordance with OECD Guideline 408. For each dose level, albino Wistar rats were split into 4

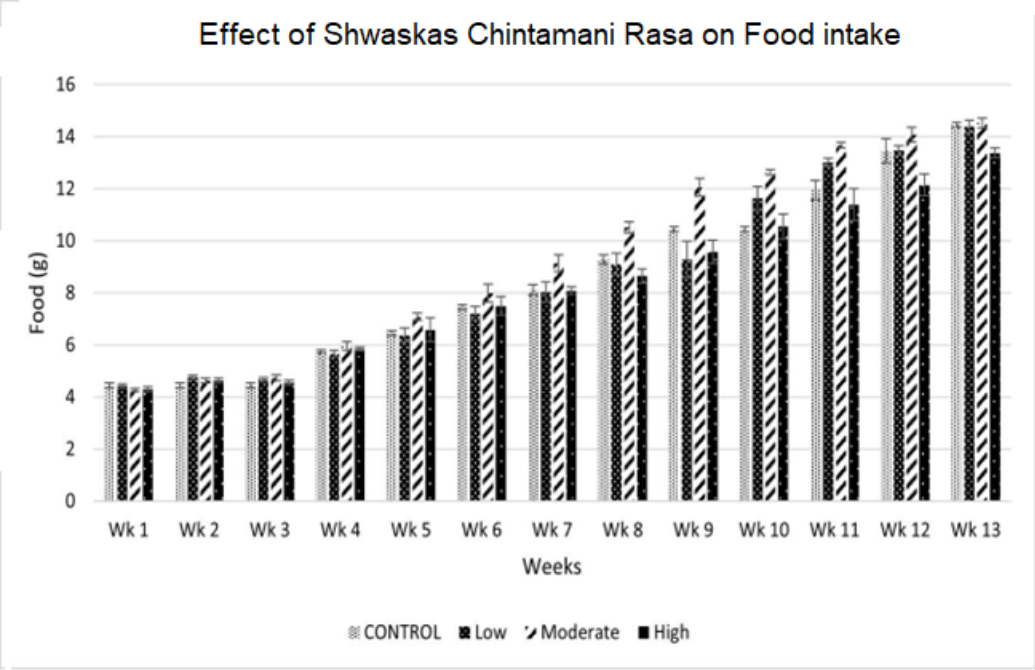

Figure 2: Food Intake following 13 weeks of administration of Low, Moderate and High doses of Shwaskas Chintamani Rasa.

Table 3: Effect of Shwaskas Chintamani Rasa on Hematological Parameters.

Parameters	Group I (Control)	Group II (Low)	Group III (Moderate)	Group IV (High)
WBC (10^3/µL)	5.897±1.59	6.797±1.24	10.41±2.41	10.15±1.76
Lymphocyte (%)	84.77±4.99	86.35±1.72	86.97±1.33	76.34±10.22
Granulocyte (%)	4.383±1.48	3.533±0.55	4.467±1.03	9.92±5.65
RBC (M/µL)	7.823±0.43	7.383±0.40	7.432±0.32	7.948±0.42
HGB (g/dL)	14.62±0.82	13.7±0.63	14.07±0.79	15.16±0.64
MCV (fL)	51.65±0.94	50.43±0.91	51.18±0.86	52.08±1.12
MCH(pg)	18.68±0.30	18.62±0.34	18.88±0.40	19.14±0.40
MCHC (g/dL)	36.2±0.16	36.87±0.36	36.9±0.32	36.74±0.44
PLT (10^3/µL)	684±105.5	536.8±109.1	658.3±73.00	730±87.04

Note: Values are mean±SEM of six animals per group.

groups. The oral administration was carried out seven days a week for a period of 13 weeks. Every day, water usage was recorded. It was noted that food and water consumption were normal during the 90-day sub-chronic toxicity study and there was no noticeably different body weight. Clinical symptoms and behavior were unaltered in all treated animals when compared to the normal control.

There were no appreciable changes in body weight. When compared to the normal control, none of the treated animals' clinical symptoms or behavior changed.

Hematological and serum biochemical data are crucial for determining the toxicological consequences of therapies. Hematological markers are typically utilized to diagnose inflammation and infections, whereas serum biochemical parameters are used to detect organ-related dysfunction. Therapeutic drugs can have negative effects on the immune system by blocking the function of essential hormones and enzymes, as well as interfering with normal hematopoiesis.[21] Bioactive chemicals may have an inverse effect on hematopoiesis, resulting in various types of anemia. As part of the natural body's defense mechanism, the body increases the production of several types of WBCs in reaction to xenobiotics. In order to establish the potential harmful effects of herbal products on hematological function, hematological parameters were tested in the current study.[22]

AST is largely found in cardiac muscle, liver and skeletal muscle cytoplasm and mitochondria.[23] ALT, on the other hand, is predominantly present in the cytoplasm of hepatic cells. Because

Table 4: Effect of Shwaskas Chintamani Rasa on Biochemical Parameters.

Parameters	Group I (Control)	Group II (Low)	Group III (Moderate)	Group IV (High)
Glucose (mg/dL)	71.33±5.60	82.83±6.81	54.17±12.81	76.17±13.53
Total Bilirubin (mg%)	0.75±0.06	1.033±0.16	1.35±0.26	0.9167±0.12
SGOT (IU/L)	36±7.16	74.33±14.68	63.17±10.9	48.17±7.88
SGPT (IU/L)	31.17±4.26	69.83±16.14	55.83±12.25	35±5.25
Total Protein (g%)	5.65±0.39	5.283±0.29	5.85±0.52	6.7±0.35
Albumin (g%)	3.283±0.16	2.95±0.48	3.183±0.15	3.133±0.20
Globulin (g%)	2.367±0.30	2.2±0.38	2.667±0.43	3.567±0.31
Triglycerides (mg/dL)	73.17±7.49	69.33±5.03	82.33±25.95	75.67±11.62
Cholesterol (mg%)	107.8±14.92	108.2±8.87	76.33±7.71	114.2±17.87
HDL (mg%)	41.83±3.48	37.67±4.17	44.67±7.04	47.83±5.45
LDL (mg%)	44.67±15.42	63.5±6.37	30.5±5.38	51.5±12.24
VLDL (mg/dL)	14.17±1.49	13.5±0.96	15.83±5.31	14.83±2.39
Sodium (m Eq/L)	141.1±0.50	143.7±2.27	138.7±1.33	139.5±1.11
Potassium (m Eq/L)	7.29±1.37	8.465±1.16	6.432±0.25	6.782±0.45
Chloride (mEq/L)	101.3±0.44	97.97±30.15	99.44±0.78	100.5±0.55
T3 (µg/dL)	147.3±6.92	123.7±9.03	145.7±7.18	130.8±7.98
T4 (µg/dL)	9.733±0.66	8.553±1.35	9.28±0.66	8.4±0.54
TSH (µIU/mL)	1.412±0.38	27.8±25.91	1.466±0.56	1.26±0.27
Urea (mg/dL)	18.5±1.43	19.5±3.69	55.17±7.30	23.5±2.60
Creatinine (mg/dL)	0.9333±0.23	0.8833±0.11	1.083±0.09	0.9833±0.07
BUN (mg/dL)	8.667±0.61	9.167±1.70	25.33±3.35	10.83±1.14

Note. Values are mean±SEM of six animals per group.

Table 5: Effect of Shwaskas Chintamani Rasa on Relative Organ Weight.

Organs	Group I (Control)	Group II (Low)	Group III (Moderate)	Group IV (High)
Brain	0.8471±0.06	0.6883±0.05	0.8784±0.06	0.792±0.05
Heart	0.3697±0.01	0.3529±0.01	0.3427±0.02	0.3541±0.02
Lungs	0.8569±0.05	0.9087±0.09	1.07±0.09	0.8009±0.04
Liver	3.218±0.16	3.45±0.27	3.792±0.25	3.525±0.30
Kidney	0.3895±0.02	0.4028±0.04	0.3088±0.07	0.4301±0.02
Pancreas	0.4293±0.06	0.3792±0.05	0.5249±0.11	0.516±0.07

Note. Values are mean±SEM of six animals per group.

liver enzymes leak into the bloodstream after necrosis, liver injury, or changes in hepatocellular permeability, serum levels of these enzymes serve as effective indicators for assessing hepatotoxicity. AST and ALT, specific liver enzymes, are used to measure hepatic cell integrity because they indicate the degree of hepatocyte deterioration.[24] Furthermore, the liver is important in protein and lipid biosynthesis in the body. Approximately 80% of the cholesterol required in mammals is produced endogenously. The liver is the source of the majority of serum endogenous cholesterol and its levels can reveal information about the liver's ability to synthesize cholesterol following medication delivery, among other things.

Hematology testing at any dosage level showed no appreciable abnormalities. However, alanine transaminase and serum glutamic-oxaloacetic transaminase levels non-significantly increased at moderate dose levels. While comparing the treatment groups to the control group on day 83, no anomalies were seen in any of the urine parameters tested. Animals administered with the low, moderate and high doses of Shwaskas Chintamani Rasa had no significant change in organ weight as compared to the control

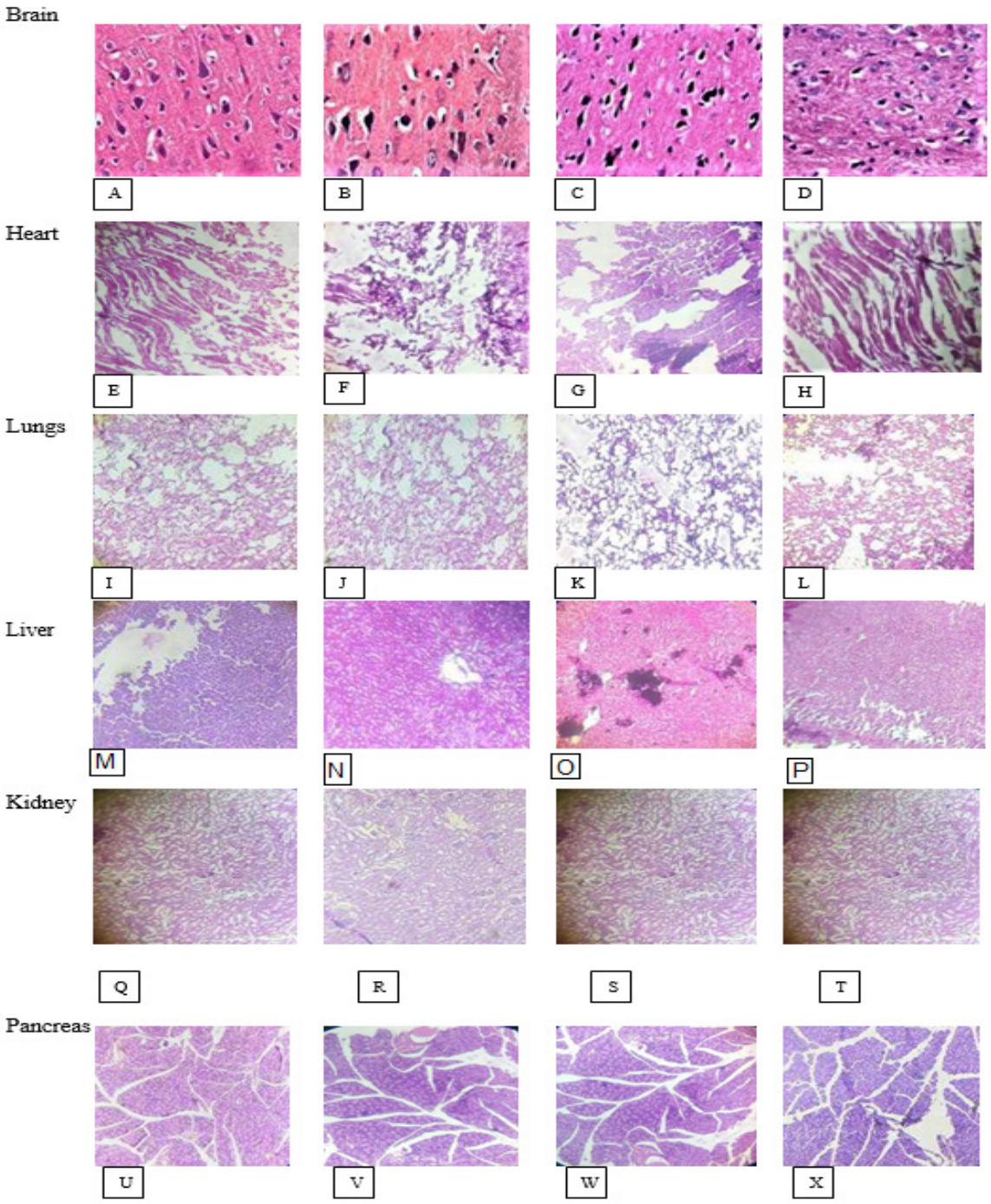

Figure 3: Photomicrographs of sections of the Brain, Heart, Lungs, Liver, Kidney and Pancreas of Wistar Rats (×400 magnification). Figure 3A: Histopathology of normal Brain. Figure 3B-D: Histopathology of three dose-treated Brain. Figure 3E: Histopathology of normal Heart. Figure 3F-H: Histopathology of three dose-treated Heart. Figure 3I: Histopathology of normal Lungs. Figure 3J-L: Histopathology of three dose-treated Lungs. Figure 3M: Histopathology of normal Liver. Figure 3N-P: Histopathology of three dose-treated Liver. Figure 3Q: Histopathology of normal Kidney. Figure 3R-T: Histopathology of three dose-treated Kidney. Figure 3U: Histopathology of normal Pancreas. Figure 3V-X: Histopathology of three dose-treated Pancreas.

group. In all groups treated with ayurvedic formulations at low (51.36 mg/kg), moderate (205.44 mg/kg) and high doses (513.6 mg/kg), no mortality nor moribound stage were seen. Shwaskas Chintamani Rasa is a traditional Ayurvedic herbo-mineral formula. Human health is seriously threatened by the use of metals and minerals in ayurvedic treatments. The results show that Wistar rats received oral treatment of any of the three formulations with little to no negative effects. This formulation's safety at all doses (Low, Moderate and High) was established in Wistar rats.

CONCLUSION

It is concluded that Shwaskas Chintamani Rasa is safe for usage in therapeutic settings and has no harmful effects on Wistar rats as tested in the 90-day chronic toxicity study.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

AUTHORS CONTRIBUTION

Dr. Shubhangi Pawar monitored and reviewed the study and Ms. Sanskruti Kashmire carried out the study and contributed in article writing. Dr. Mahalaxmi Mohan conceptualized, revised and approved the article. Dr. Shishir Pandey, Dr. Rajshree Kulkarni and Dr. Abhay Kulkarni reviewed the article.

SUMMARY

The study investigated the safety and efficacy of Shwaskas Chintamani Rasa, a traditional Ayurvedic herbo-metallic formulation used to treat respiratory illnesses. Given its inclusion of heavy metals, the medication was administered under strict medical supervision.

Study Design and Methodology: Following OECD Guideline 408, albino Wistar rats were divided into four groups, receiving oral doses daily for 13 weeks. Food and water intake were monitored, revealing no significant changes in consumption or body weight across treatment groups.

Toxicological Assessments: Clinical symptoms and behaviors remained consistent with the control group, indicating no adverse effects. Hematological and biochemical parameters were analyzed to assess potential toxicity:

Hematological Findings: No significant abnormalities were observed in hematological markers, which are crucial for detecting inflammation and infections.

Biochemical Analysis: Serum levels of Alanine aminotransferase (ALT) and Aspartate aminotransferase (AST) showed non-significant increases at moderate dose levels, indicating potential hepatic effects but not reaching toxicity levels.

Additionally, urine parameters tested on day 83 showed no anomalies and organ weights did not differ significantly from the control group across all dosage levels (low: 51.36 mg/kg, moderate: 205.44 mg/kg, high: 513.6 mg/kg). Importantly, there were no observed mortalities or severe health declines in any of the groups.

Conclusion: The results demonstrate that Shwaskas Chintamani Rasa is safe for use at all tested doses in Wistar rats, showing minimal to no negative effects.

REFERENCES

1. Kumari M, Bajad SM, Kshirsagar SR, Chinde S, Balaji AS, Jerald Mahesh Kumar MJ, *et al.* Sub-chronic oral toxicity evaluation of herbo-metallic formulation Arshakuthar rasa in rats. J Ethnopharmacol. 2022;298:115306. doi: 10.1016/j.jep.2022.115306, PMID 35443217.
2. Nagarajan S, Sivaji K, Krishnaswamy S, Pemiah B, Rajan KS, Krishnan UM, *et al.* Safety and toxicity issues associated with lead-based traditional herbo-metallic preparations. J Ethnopharmacol. 2014;151(1):1. doi: 10.1016/j.jep.2013.10.037, PMID 24216165.
3. Krishnamachary B, Pemiah BR, Krishnaswamy S, Krishnan UM, Sethuraman S, Sekar R. Elucidation of a core-shell model for lauha bhasma through physicochemical characterization. Int J Pharm Pharm Sci. 2012;4(2):644-9.
4. Phua DH, Zosel A, Heard K. Dietary supplements and herbal medicine toxicities-when to anticipate them and how to manage them. Int J Emerg Med. 2009;2(2):69-76;15(39):4886. doi: 10.1007/s12245-009-0105-z, PMID 20157447.
5. Dwivedi V, Anandan EM, Mony RS, Muraleedharan TS, Valiathan MS, Mutsuddi M, et al. *In vivo* effects of traditional Ayurvedic formulations in *Drosophila melanogaster* model relate with therapeutic applications. PLOS One. 2012;7(5):e37113. doi: 10.1371/journal.pone.0037113, PMID 22606337.
6. Maurya SK, Seth A, Laloo D, Singh NK, Gautam DN, Singh AK. Śodhana: an Ayurvedic process for detoxification and modification of therapeutic activities of Poisonous medicinal plants. Anc Sci Life. 2015;34(4):188-97. doi: 10.4103/0257-7941.160862, PMID 26283803.
7. Govindadas, Ratnavali B, edited by Rajeshwar Dattashastri, chapter 16, verse. 8th ed. Vols. 57-60. p. 334.
8. Parasuraman S, Raveendran R, Kesavan R. Blood sample collection in small laboratory animals. J Pharmacol Pharmacother. 2010;1(2):87-93. doi: 10.4103/0976-500X.72350, PMID 21350616.
9. Kumar S, Kumar R, Srivastava R, Rai RK, Sasty JL, Gupta A. Subchronic oral toxicity study of a herbo-metallic ayurvedic formulation Swarna Guggulu in Wistar rats. Indian J Nat Prod Resour (IJNPR)[Formerly Natural Product Radiance (NPR)]. 2018;9(3):244-52.
10. Reagan-Shaw S, Nihal M, Ahmad N. Dosetranslation from animalto human studies revisited. FASEB J. 2008;22(3):659-61. doi: 10.1096/fj.07- 9574LSF. OECD/OCDE408. oecdguidelineforthetestingofchemicals, Repeated dose 90-day oral toxicity study in rodents [Internet]. 2018. PMID 17942826.
11. Jamadagni PS, Jamadagni SB, Singh A, Singh RK, Upadhyay SN, Gaidhani SN, *et al.* Toxicity study of Swarna bhasma, an ayurvedic medicine containing gold, in Wistar rats. Toxicol Int. 2015;22(3):11. doi: 10.22506/ti/2015/v22/i3/137618.
12. Patel C, Dadhaniya P, Hingorani L, Soni MG. Safety assessment of pomegranate fruit extract: acute and subchronic toxicity studies. Food Chem Toxicol. 2008;46(8):2728-35. doi: 10.1016/j.fct.2008.04.035, PMID 18571823.
13. Shokryazdan P, Faseleh Jahromi M, Liang JB, Kalavathy R, Sieo CC, Ho YW. Safety assessment of two new Lactobacillus strains as probiotic for human using a rat model. PLOS One. 2016;11(7):e0159851. doi: 10.1371/journal.pone.0159851, PMID 27467068.
14. Vigneshwar R, Arivuchelvan A, Mekala P, Imayarasi K. Sex-specific reference intervals for Wistar albino rats: hematology and clinical biochemistry. Indian J Anim Health. 2021;60(1):58-65. doi: 10.36062/ijah.60.1.2021.58-65.
15. Lavekar G.S, Ravishankar B, Venugopal Rao S, Gaidhani S. N, Ashok B. K, Shukla V.J. safety / Toxicity studies of ayurvedic formulation-Navratna rasa. Toxicol Int. 2009;16(1):37-42.
16. Mensah FA, Kaukanen CT, Kauhanen J, Henneh IT, Agyei PE, Akakpo Kand Ekor M. Sub-chronic toxicity evaluation of top three commercial herbal antimalarial preparations in the Kumasi Metropolis, Ghana. Biosci Rep. 2020;40.

17. Firenzuoli F, Gori L. Herbal medicine today: clinical and research issues. Evid Based Complement Alternat Med. 2007;4 Suppl 1:37-40. doi: 10.1093/ecam/nem096, PMID 18227931.
18. Cheng CW, Bian ZX, Wu TX. Systematic review of Chinese herbal medicine for functional constipation. World J Gastroenterol. 2009 Oct;15(39):4886-95. doi: 10.37 48/wjg.15.4886, PMID 19842218.
19. Amin MN. *In vivo* evaluation of anti-nociceptive, anti-inflammatory, Antipyretic, Hypoxia and Gastro-intestinal potentials of Swas Kas Chintamani Ras. Discovery. Phytomedicine Journal of Natural Products Researchand Ethnopharmacology. 2019;6(1): 1.
20. Lubran MM. Hematologic side effects of drugs. Ann Clin Lab Sci. 1989;19(2):114-21. PMID 2665627.
21. Wang X, Zhang W, Wang Y, Peng D, Ihsan A, Huang X, *et al.* Acute and sub-chronic oral toxicological evaluation sofquinocetone in Wistar rats. Regul Toxicol Pharmacol. 2010;58(3):421-7. doi: 10.1016/j.yrtph.2010.08.008, PMID 20713111.
22. Upur H, Amat N, Blazeković B, Talip A. Protective effect of *Cichorium glandulosum* root extract on carbon tetrachloride-induced and galactosamine-induced hepatotoxicity in mice. Food Chem Toxicol. 2009;47(8):2022-30. doi: 10.1016/j.fct.2009.05.022, PMID 19477217.
23. Wong LL, Lacar L, Roytman M, Orloff SL. Urgent liver transplantation for dietary supplements: an under-recognized problem. Transplant Proc. 2017;49(2):322-5. doi: 10.1016/j.transproceed.2016.11.041, PMID 28219592.
24. Karri SK, Saper RB, Kales SN. Lead encephalopathy due to traditional medicines. Curr Drug Saf. 2008;3(1):54-9. doi: 10.2174/157488608783333907, PMID 18690981.

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