A STUDY ON THE EFFECTS OF MO SINH CO PLUS OINTMENT ON GENERAL CONDITION AND HEMATOLOGICAL PARAMETERS IN EXPERIMENTAL ANIMALS

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Abstract

Objectives*: To evaluate the effect of Mo sinh co plus according to the route of application on the skin on the general state and hematological indicators in practice.* ***Methods:*** *White rats were continuously lubricated with Mo sinh co plus declared levels of 0.5 g/kg/day and 2.0 g/kg/day for 12 consecutive weeks. Evaluation of the general status, body weight and hematopoietic function of rats at the time points before applying the drug, after 6 weeks and 12 weeks of applying the drug.* ***Results****: The results of the study showed that Mo sinh co plus applied topically on the skin claimed to be 0.5 g/kg/day and 2.0 g/kg/day continuously for 12 weeks without affecting the general condition, Weighted numbers and unique serum student numbers in white rats.*

Keywords: *Mo sinh co plus, general state, hematological index.*

I. INTRODUCTION

Traditional Vietnamese medicine has long placed great importance on external medications. With the development of traditional medicine, externally used drugs have also made continuous progress, as evidenced by pharmacological studies and the constant accumulation of new formulas. "Mỡ sinh co", (Mo sinh co) produced by the Military Institute of Traditional Medicine for many years, has been applied in the research and treatment of various diseases such as hemorrhoids, anal fistulas, and anal fissures. Additionally, preliminary studies on its application in the treatment of skin ulcers have yielded promising results. The product features a simple formulation as an ointment for preclinical and clinical research, making it suitable for implementation in many medical facilities. Its topical application, compact packaging, and ease of use and storage are advantageous. To supplement and upgrade the product for broader therapeutic effects, we have undertaken the study entitled "Research on the formulation, toxicity assessment, and some pharmacological effects of Mo sinh co plus in experiments." The objective of this study is to evaluate the effects of Mo sinh co plus on the general condition and hematological parameters when applied topically to the skin of white rats, providing a basis for conducting subsequent research stages.

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II. METHODS

2.1. Materials

2.1.1. Composition of Mo sinh co plus: Each 20g tube of Mo sinh co plus was prepared using the following medicinal herbs:

Nghe (*Curcuma longa),* Duong quy (*Radix Angelicae sinensis),* Bach chi (*Radix Angelicae dahuricae),* Cot toai bo (*Rhizoma Drynariae),* Nguu tat (*Radix Achyranthis bidentatae),* Sinh dia (*Radix Rehmanniae glutinosae),* Khuong hoat (*Rhizoma et Radix Notopterygii),* Thuong truat (*Rhizoma Atractylodis),* Dai hoang (*Rhizoma Rhei),* Tru ma can (*Rhizoma et Radix Boehmeria nivea),* Ngu boi tu (*Galla sinensis),* Que nhuc (*Cortex Cinnamomi),* Long nao (*Cinnamomum camphora N. Et E.).* And sufficient excipients.

The ointment was produced by the Center for Research and Application of Traditional Medicine, Military Institute of Traditional Medicine. All medicinal herbs met the standards of the Vietnamese Pharmacopoeia V [1], and Mo sinh co plus was tested to meet the established standards.

2.1.2. Equipment and Chemicals:

- Analytical chemicals and other laboratory equipment met the standards for research.

- Swelab Alfa hematology analyzer (Sweden)

2.2. Subjects

30 Wistar strain white rats, both male and female, with an average weight of 180g ± 20g, were provided by the Animal Breeding Department of the Military Academy of Medicine.

The experimental rats were housed in the laboratory of the Experimental Research Department, Military Institute of Traditional Medicine for 5 days before the study with standard food and water ad libitum.

2.3. Methods

The sub-chronic toxicity of Mo sinh co plus in white rats via topical application was evaluated according to the guidelines of the WHO, regulations of the Vietnamese Ministry of Health, and related regulations [2],[3],[4]. The study was conducted at the Experimental Research Department, Military Institute of Traditional Medicine.

The rats were randomly divided into 3 groups, each with 10 rats, with a similar male/female ratio:

- Control group: no treatment (no application of the study drug).

- Treatment group 1: Mo sinh co plus at a dose of 0.5 g/kg/day (equivalent to the projected human dose).

- Treatment group 2: Mo Sinh Co plus at a dose of 2.0 g/kg/day (4 times the dose of group 1).

Mo sinh co plus was applied directly to the shaved skin on both sides of the rat's back (approximately 10% of the body surface area) and covered with sterile gauze. The drug was changed once a day at 8 am for 12 consecutive weeks.

***Monitoring indicators:**

- General condition and body weight of rats.

- Assessment of hematopoietic function through the number of red blood cells, hemoglobin, hematocrit, mean corpuscular volume, white blood cell count, white blood cell differential, and platelet count.

These indicators were measured before drug administration, after 6 weeks, and after 12 weeks of drug administration. The tests were conducted at the Department of Labor Physiology, Military Academy of Medicine.

2.4. Data Analysis

The research data were analyzed using biomedical statistical methods with SPSS 20.0 software. Data are presented as ($\overline{X}$ ± SD**).** The difference was considered statistically significant when $p<0.05$.

III. RESULTS

3.1. General condition and weight changes in rats

- General condition: Throughout the experiment, rats in all three groups ate, drank, and were active as normal. They had bright eyes, smooth fur, and good appetites, producing dry feces. No significant abnormalities were observed in any of the three rat groups throughout the study period.

- Weight changes:

Table 1. Effect of Mo sinh co plus on rat weight (g)

Time point	Control group (n=10) (1)	Treatment group 1 (n=10) (2)	Treatment group 2 (n=10) (3)	p
Before applied	171.30 ± 2.91	173.20 ± 4.22	172.05 ± 4.49	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
After 6 weeks applied	194.05 ± 3.78	195.10 ± 4.26	190.80 ± 6.37	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
p (before- after)	< 0.01	< 0.01	< 0.01	
After 12 weeks applied	213.30 ± 5.01	216.30 ± 4.05	211.50 ± 6.65	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
p (before- after)	< 0.01	< 0.01	< 0.01	

Comment: Table 1 shows that after 6 and 12 weeks of applying Mo sinh co plus, the weight of rats in all three groups (control and two experimental groups) increased compared to before treatment ($p<0.01$). There was no significant difference in weight gain between the control group and the treatment groups at the same time points ($p>0.05$).

3.2. Effect of Mo sinh co plus on hematopoiesis

Table 2. Effect of Mo sinh co plus on red blood cell count in white rats

Time point	Red blood cell count (T/l)			p
	Control group (n=10) (1)	Treatment group 1 (n=10) (2)	Treatment group 2 (n=10) (3)	
Before applied	7.51 ± 0.98	7.46 ± 1.38	7.37 ± 0.99	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
After 6 weeks applied	7.99 ± 0.48	8.08 ± 0.44	8.19 ± 1.02	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
p (before-after)	> 0.05	> 0.05	> 0.05	
After 12 weeks applied	7.53 ± 1.04	7.73 ± 0.59	7.72 ± 0.39	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
p (before-after)	> 0.05	> 0.05	> 0.05	

Table 3. Effect of o Mo sinh co plus on hemoglobin levels in white rats

Time point	Hemoglobin levels (g/dl)			p
	Control group (n=10) (1)	Treatment group 1 (n=10) (2)	Treatment group 2 (n=10) (3)	
Before applied	13.75 ± 1.88	13.31 ± 2.38	13.17 ± 1.45	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
After 6 weeks applied	13.80 ± 0.44	14.06 ± 0.87	14.19 ± 1.44	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
p (before-after)	> 0.05	> 0.05	> 0.05	
After 12 weeks applied	13.32 ± 1.10	13.35 ± 0.80	13.48 ± 0.68	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
p (before-after)	> 0.05	> 0.05	> 0.05	

Comment: The results from Tables 2 and 3 indicate that after 12 weeks of applying Mo sinh co plus, there were no significant differences in red blood cell count and hemoglobin levels between the experimental groups and the control group, as well as between the pre- and post-treatment time points ($p > 0.05$).

Table 4. Effect of Mo sinh co plus on hematocrit in white rats

Time point	Hematocrit (%)			p
	Control group (n=10) (1)	Treatment group 1 (n=10) (2)	Treatment group 2 (n=10) (3)	
Before applied	40.27 ± 5.38	39.23 ± 7.10	38.84 ± 4.23	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
After 6 weeks applied	38.89 ± 2.64	40.29 ± 2.54	41.51 ± 5.02	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
p (before-after)	> 0.05	> 0.05	> 0.05	
After 12 weeks applied	38.15 ± 3.78	38.17 ± 2.52	39.15 ± 1.55	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
p (before-after)	> 0.05	> 0.05	> 0.05	

Table 5. Effect of Mo sinh co plus on mean corpuscular volume (MCV) in white rats

Time point	MCV (fl)			p
	Control group (n=10) (1)	Treatment group 1 (n=10) (2)	Treatment group 2 (n=10) (3)	
Before applied	50.61 ± 4.12	51.69 ± 3.71	51.84 ± 3.25	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
After 6 weeks applied	48.50 ± 3.78	49.86 ± 1.38	50.71 ± 1.94	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
p (before-after)	> 0.05	> 0.05	> 0.05	
After 12 weeks applied	50.85 ± 2.10	50.57 ± 1.32	50.76 ± 2.12	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
p (before-after)	> 0.05	> 0.05	> 0.05	

Comment: The results from Tables 4 and 5 show that after 12 weeks of applying Mo sinh co plus, hematocrit and mean corpuscular volume in both experimental groups 1 and 2 showed no significant differences compared to the control group and between the pre- and post-treatment time points ($p > 0.05$).

Table 6. Effect of Mo sinh co plus on white blood cell count in white rats

Time point	White blood cell count (G/l)			p
	Control group (n=10) (1)	Treatment group 1 (n=10) (2)	Treatment group 2 (n=10) (3)	
Before applied	9.39 ± 2.89	11.04 ± 5.11	10.73 ± 2.98	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
After 6 weeks applied	11.33 ± 2.42	12.26 ± 2.86	11.67 ± 1.79	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
p (before-after)	> 0.05	> 0.05	> 0.05	
After 12 weeks applied	9.64 ± 3.60	10.09 ± 3.06	9.97 ± 2.81	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
p (before-after)	> 0.05	> 0.05	> 0.05	

Table 7. Effect of Mo sinh co plus on white blood cell differential in white rats

Time point	White blood cell (%)	Group			p
		Control group (n=10) (1)	Treatment group 1 (n=10) (2)	Treatment group 2 (n=10) (3)	
Before applied	Lympho	70.05 ± 8.63	69.01 ± 8.91	70.38 ± 9.46	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
	Neutrophil	22.91 ± 7.71	26.53 ± 5.95	24.01 ± 8.63	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
After 6 weeks applied	Lympho	69.35 ± 7.69	65.88 ± 7.05	66.89 ± 4.07	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
	Neutrophil	25.71 ± 7.84	28.74 ± 6.79	28.80 ± 5.88	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
p (before- after)		> 0.05	> 0.05	> 0.05	> 0.05
After 12 weeks applied	Lympho	66.80 ± 6.97	63.64 ± 5.22	64.36 ± 9.16	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
	Neutrophil	26.36 ± 5.00	29.56 ± 7.62	28.12 ± 6.89	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
p (before- after)		> 0.05	> 0.05	> 0.05	> 0.05

Table 8. Effect of Mo sinh co plus on platelet count in white rats

Time point	Platelet count (G/l)			p
	Control group (n=10) (1)	Treatment group 1 (n=10) (2)	Treatment group 2 (n=10) (3)	
Before applied	419.20 ± 77.98	414.00 ± 81.91	402.30 ± 83.32	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
After 6 weeks applied	415.30 ± 88.62	400.20 ± 70.95	403.90 ± 57.75	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
p (before-after)	> 0.05	> 0.05	> 0.05	
After 12 weeks applied	396.60 ± 65.23	421.20 ± 88.83	437.30 ±85.34	$p_{1-2}; p_{1-3}; p_{2-3} > 0.05$
p (before-after)	> 0.05	> 0.05	> 0.05	

Comment: The results from Tables 6, 7, and 8 indicate that after 12 weeks of applying Mo sinh co plus, the white blood cell count, white blood cell differential, and platelet count in both experimental groups 1 and 2 showed no significant differences compared to the control group and between the pre- and post-treatment time points ($p > 0.05$).

IV. DISCUSSION

4.1. Effects of Mo sinh co plus on General Condition

The general condition and weight changes reflect the overall health status of animals and are mandatory indicators in subchronic toxicity studies. Throughout the study period, experimental animals in all three groups (negative control and two treatment groups) exhibited normal food intake, activity, bright eyes, smooth fur, and dry feces. After 6 and 12 weeks of Mo sinh co plus application, the weight of rats in all three groups increased compared to the pre-dose values ($p<0.01$); there was no significant difference in weight between the groups treated with Mo sinh co plus and the control group ($p > 0.05$).

Thus, Mo sinh co plus at doses of 0.5 g/kg and 2.0 g/kg body weight did not affect the general condition and weight of rats when applied topically for 12 consecutive weeks.

4.2. Effects of Mo sinh co plus on Hematological Parameters

Blood is a crucial component of the body. Peripheral blood cell counts are valuable in assessing hematopoietic function [4]. According to WHO guidelines, hematological parameters are mandatory tests in the evaluation of the toxicity of test substances [5]. Therefore, red blood cell count, white blood cell count, differential white blood cell count, and platelet count in rats were determined. The results showed that all hematological parameters in both the control group and the two treatment groups remained within normal limits, with no statistically significant differences compared to pre-dose values ($p > 0.05$) and compared to the control group at both 6 and 12 weeks of continuous Mo sinh co plus application. Thus, Mo sinh co plus did not exhibit toxicity to hematopoietic organs.

V. CONCLUSION

The results of this subchronic dermal toxicity study of Mo sinh co plus demonstrated that topical application of Mo sinh co plus at doses of 0.5 g/kg and 2.0 g/kg (four times the projected clinical dose) for 12 consecutive weeks did not affect the general condition, weight gain, or hematological parameters in rats.

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