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Research Article

EXPERIMENTAL STUDY ON *PANCHANAN RASA* W.S.R. TO ITS ACUTE ORAL TOXICITY

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ABSTRACT

Ras sashtra is a specialized branch of Indian traditional system and medicine which deals with metals, minerals, herbal compounds, and various principles of alchemy and metallurgy etc. *Panchanan Rasa* chief reference is in *Ras Ratna Sammuchya Hridya Roga Chikitsa* for *Hridya Roga*. It is a herbo-mineral preparation made with the help of *Kharal* so called *Kharaliya* preparation and is used in powder form to be given to patient. In present era *Hridya Roga* is becoming a common problem, every one person in ten people is suffering from CAD/IHD with varied etiology and is going under long term medication, costly surgeries which every patient is not able to afford, so this article discuss about the acute oral toxicity of *Panchanan Rasa* and found it non-toxic on Albino rats.

INTRODUCTION

Ayurveda the traditional Indian system of medicine is regarded as the most methodological and efficient among all such systems practiced in different regions of the world. According to WHO, traditional medicine has established and proved itself to possess preventive, curative and rehabilitative roles. The branch which deals with *Rasa*, metals, minerals, gems and various principles of alchemy, metallurgy etc. is known as *Rasa-Shastra* or Ayurveda pharmaceutics which has become an integral part of Ayurveda. The innate qualities like quick action, lesser dose, prolonged shelf-life, better palatability of *Rasa ausadhies* have helped them to conquer the compliance of the patient as well as pharmaceutical proprietors. In the present study an attempt was made to check the acute oral toxicity of *Panchanan Rasa* on Wistar albino rats as per the OECD-423 guidelines in the animal house of VNS College of Pharmacy Neelbad, Bhopal, MP.

Need of Study

Herbal medicines are indicated in *Hridaya Roga*, but the work on *Rasa aushadhi* has not been so far much explored, *Panchanan Rasa* has been mentioned in *Rasa Ratna Samuchya* for the indication of *Hridaya Roga*, as per our knowledge is concerned there has been no experimental study on *Panchanan Rasa*, so there is need to check the acute oral toxicity so this study was taken. *Panchanan Rasa* is not commonly used formulation in present scenario so there was a need to conduct toxicity study, so that its therapeutic efficacy can be explored after knowing toxicity study aspects on animal model for safety profile, so that in future this preparation can be taken as clinical trial.

The efficacy, safety and authenticity of *Panchanan Rasa* is most important aspects and also to re-establish whether this preparation is suitable in present scenario.

Review of literature

It is one of the potent herbo-mineral drugs and the detailed description of ingredients, usage, properties, method of preparation, and therapeutic effect of *Panchanan Rasa* is mentioned in *Rasa Ratna Samuchya*, *Hridaya Roga Chikitsa* chapter 14/09.

AIMS AND OBJECTIVE

To conduct acute toxicity study on animal model and to assess the safety profile of *Panchanan Rasa*.

Methods of preparation of *Panchanan Rasa* as per chief reference

Rasa Ratna Samuchya Hridya Roga Chikitsa Chapter 14/09.

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MATERIAL AND METHODS

Materials

1) *Parad* (Mercury [Hg])
2) *Gandhak* (Sulphur [S])
3) *Aamla* (*Phyllanthus emblica*)
4) *Draksha* (*Vitis Vinifera*)
5) *Yasthimadhu* (*Glycyrrhiza glabra*)
6) *Kharjura* (*Phoenix dactylifera*),

   Was obtained from the Rasa Shashtra Pharmacy of Pt. Khusilal Sharma Govt. Ayurveda Institute, Bhopal, MP, and was purified as per the classical methods.
7) For experimental study- *Panchanan Rasa*, Wistar strain Albino rats (Total no. of rats = 12), were used.

Methods

Experimental study w.s.r to its Acute Oral Toxicity Study.

Experimental study

1) Selection of animal model as Wistar strain albino rats weighing about 200-250gram used as per the guideline of OECD-423.
2) The animals are randomly selected and marked to permit individual identification and kept in their cages for at least 5 days prior to dosing to allow for acclimatization to the laboratory conditions.

The temperature in the experimental animal room was maintained and humidity was around 30% and did not exceed 70%. The animals where maintained in standard laboratory conditions the sequence being 12 hours dark and 12 hours light cycle. After the medicine has been administered, food has been withheld for further 3-4 hours.

Inclusive Criteria

- Healthy and active albino rats selected randomly.
- Rats weighing 200-250 gram

Exclusive Criteria

- Rats below and above 200-250 grams.
- Diseased and infected rats
- Pregnant rats
- Rats which are under trial for other experiments

Dose Fixation

Dose: According to OECD 423 guideline there were 4 level of dose.

- 5mg/kg body weight
- 50mg/kg body weight
- 300mg/kg body weight
- 2000mg/kg body weight

3 rats were used for each step, i.e., total 12 rats were used.

Duration- 14 days

Schedule- Single dose, test drug and vehicle (5% CMC) were administered.

Route of Drug Administration

Prepared drug was given orally with help of suitable gastric catheter.

Criteria for Assessment

- **Observation-** All animals were observed at ½, 1, 2, 3, 4, 5, 6 hours after dosing.
- **Body Weight-** The body weight of each rats were recorded just prior to dosing 1, 7, 14 days.
- **Terminal Study-** At last of 14 days rats were kept for overnight fasting and next day were sacrificed by over dose of ether anesthesia, and autopsy were carried out for any histo-pathological changes in organs like liver, kidney and heart.

Investigations

- **Haematological Parameters**- Hb, WBC count, RBC count, lymphocytes, platelets counts.
- **Biochemical Parameters**- LFT-Serum glutamic oxalo acetic Transaminase (SGOT), Serum Glutamic Pyruvic Transaminase (SGPT), ALP (alkaline phosphatase test), Total bilirubin, RFT- Blood Urea nitrogen, creatinine, uric acid and total protein were estimated.
- **Histo-pathological Study**- Liver, kidney and heart were analysed after animal sacrifices.

RESULTS

Table 1: Complete Blood Count

S. No	Group	Hb% (g/dl)		RBC count		WBC count		Platelet counts	
	Group – 4	**Before**	**After**	**Before**	**After**	**Before**	**After**	**Before**	**After**
1	R1	14.9	13.5	6.7	7.83	4800	3170	10.55	7.86
2	R2	14.75	13.1	6.82	8.55	4200	8440	8.46	7.21
3	R3	14.6	13.2	6.94	8.32	3600	10580	6.37	8.44
	P value		**0.0031**		**0.0148**		**0.3352**		**0.7015**

Note: All the data were expressed as MEAN ± SEM (n= 03). Analyzed by Two tailed Paired 't' test , p<0.05 will be considered as non-significant

Table 2: Liver Function Test

S.No	Group	Total Bilirubin		SGOT		SGPT		ALP	
	Group – 4	**Before**	**After**	**Before**	**After**	**Before**	**After**	**Before**	**After**
1	R1	0.9	0.37	47.9	143.7	42.6	60.20	160.2	374.20
2	R2	0.65	0.32	54.95	129.4	48.35	52.10	159.40	245.20
3	R3	0.4	0.41	62	192.4	54.1	49.20	158.6	196.40
	P value		**0.2141**		**0.0255**		**0.4908**		**0.3701**

Note: All the data will be expressed as MEAN ± SEM (n=03). Analyzed by Two-tailed Paired 't' test followed by p<0.05 will be considered as non-significant

Table 3: Kidney Function Test

Sr.No	Group	Blood Urea		S. creatinine	
	Group- 4	**Before**	**After**	**Before**	**After**
1	R1	46.9	42.10	0.9	0.26
2	R2	46.10	44.10	0.85	0.38
3	R3	45.3	44.10	0.8	0.25
	P value		**0.1345**		**0.0078**

Note: All the data will be expressed as MEAN ± SEM (n = 03). Analyzed by Two-tailed paired 't' test followed by p<0.05 will be considered as non-significant.

Table: 4 Body Weight

Sr.No	Group	1st day	7th day	14th day
	Group – 4			
1	R1	170gm	214gm	244gm
2	R2	182gm	224gm	249gm
3	R3	170gm	210gm	238gm
	P value			**0.0001**

Note: All the data will be expressed as MEAN ± SEM. Analyzed by ANOVA followed by Bonferroni multiple comparison test, p<0.05 will be considered as non-significant.

DISCUSSION

On the basis of above test parameters and their statistical data computed for each test with before and after values and applying the test applicable as per data two tailed paired 't' test and One away ANOVA test was implied to group-4 and the 'p' value for Hb% is 0.0031, RBC is 0.0148. WBC is 0.3352, platelet is 0.7015, total bilirubin is 0.2141, SGOT is 0.0255, SGPT is 0.4908, ALP is 0.3701, blood urea is 0.1345, Sr. Creatinine is 0.0078, and for weights the 'p' value is 0.0001. On the basis of above results and the slides of different organs observed we can say that the acute oral toxicity of *Panchanan Ras* done as per the OECD – 423 guidelines, was found non-significant and so the drug is not toxic.

LD50 Value: As per observations and calculations from acute oral toxicity (OECD Guidelines 423), the LD50 value of *Panchanan rasa* was found to be more than 2000mg/kg body weight.

CONCLUSION

Panchanan rasa does not exhibit toxic effects when given orally at concentration of 2000mg/kg body weight. However the normalcy and insignificant changes in wellness parameters and body weights reveals the safety of *Panchanan rasa* at a dose of 2000mg/kg body weight.

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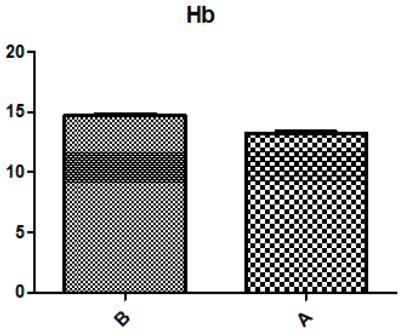
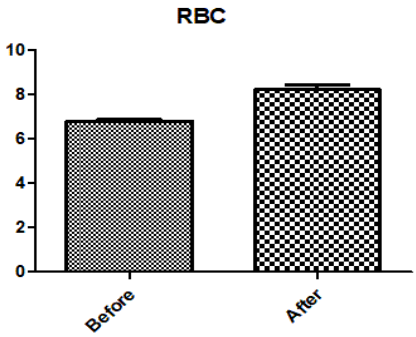
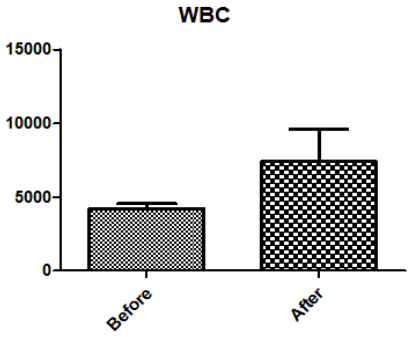
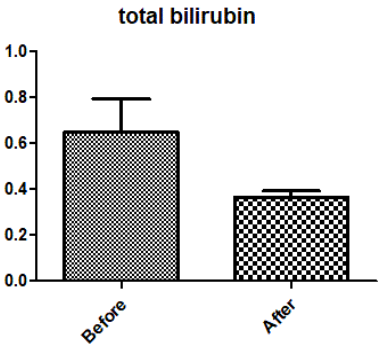
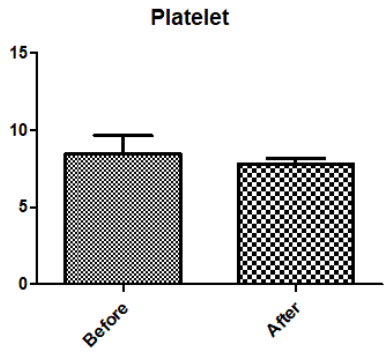
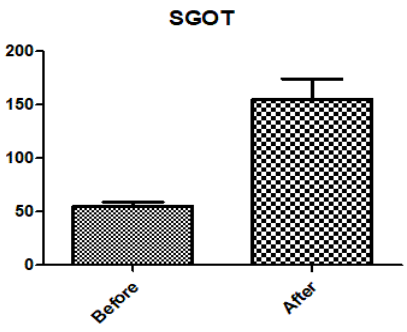

Hb
20
15
10
5
0
B
A

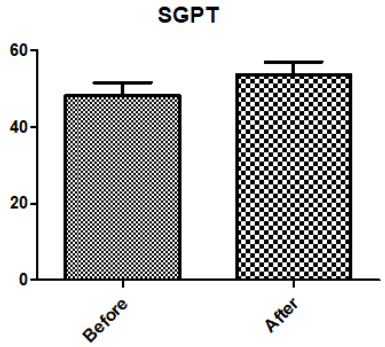

RBC
10
8
6
4
2
0
Before
After

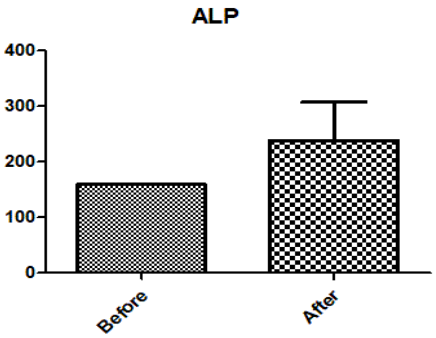

WBC
15000
10000
5000
0
Before
After

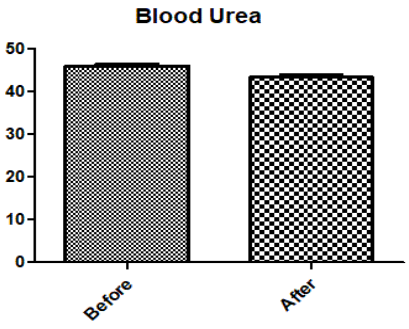

total bilirubin
1.0
0.8
0.6
0.4
0.2
0.0
Before
After


Platelet
15
10
5
0
Before
After

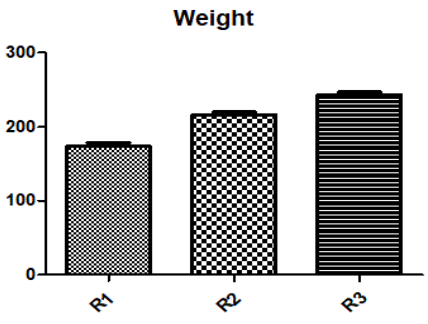

SGOT
200
150
100
50
0
Before
After


SGPT
60
40
20
0
Before
After


ALP
400
300
200
100
0
Before
After


Blood Urea
50
40
30
20
10
0
Before
After


S. creatinine
1.0
0.8
0.6
0.4
0.2
0.0
Before
After


Weight
300
200
100
0
R1
R2
R3

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