

**STUDY OF VANGA BHASMA ANTIHYPERLIPEDAEMIC ACTIVITY IN WISTAR  
ALBINO RATS- AN EXPERIMENTAL STUDY****Dr. Lalitha M. Vatar\*<sup>1</sup>, Dr. Madhuri<sup>2</sup> and Dr. Chandrashekhara Kuppi<sup>3</sup>**

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Article Received on 10/05/2021

Article Revised on 31/05/2021

Article Accepted on 20/06/2021

**ABSTRACT**

Rasashastra is a special branch of Ayurveda which deals with specific pharmaceutical procedures and techniques which convert the metals and minerals into a safe and acceptable form. Bhasma is such kind of drugs with higher efficacy in lower doses and with good palatability. Vanga is more popular, it being one among putilohas, is having gunas Tikta, kashaya, lavana rasa, Ushna veerya and is indicated in Krimi, Prameha, Kapha and Medorogas.

**KEYWORDS:** Vanga, Bhasma, Putiloha, Medo Roga.**INTRODUCTION**

Vanga is a metal which got its name due to its origin i.e. from Vanga Desha. It is placed under Puti lohas, due to its low melting point and the smell emitted during the melting. Vanga bhasma has been considered as medhoghna, medhanasha, medhohara etc.<sup>[1,2,3,4]</sup> these references directly indicate it as a lipid lowering agent.

Lipids and cholesterol are directly related to hyperlipidaemia. There is no direct reference available in classics correlating hyperlipidemia, it can be included under Santarpanajanya vyadhi as a Medoroga, It is a condition caused by derangement of agni, leads to Amarasa, there is medhodhatvagni mandya leads to improper formation of excess medodhatu. Vanga due to its Lekhana<sup>[5]</sup> property decreases Medas and its Ruksha Tikshna, guna<sup>[6,7]</sup> does Srotovishodhana.

**Acute oral toxicity**

The moral traditional test for acute oral toxicity is the oral LD<sub>50</sub> and test protocol for this is found in OECD test guideline 420.<sup>[8]</sup>

**Steps to be followed before starting the experiment<sup>[9]</sup>**

- Adequate number of animals should be available for study.
- Each animal must be individually tested.
- Injuries, abnormalities and lesions should be rejected.

- Before allocated to groups rats must undergo basic haematological and biochemistry tests. Record the weight of all animals.

Acute toxicity test of Vanga bhasma was determined as per the OECD guidelines No. 420. Mice (16 – 20 g) were used for this study. The initial dose of 2000 mg/kg of the vanga bhasma was selected and administered to a group of six animals. The treated animals were monitored for 48 hrs, for mortality and general behavior. No toxic symptoms or mortality was observed till the end of the study. The lethal dose (LD<sub>50</sub>) selected was 2 g/kg body weight. Hence, the experimental dose was selected as one-fifth as high dose (400 mg/kg), one-tenth (200 mg) medium dose, one-twenty (100 mg) low dose of the LD<sub>50</sub> dose.

**Principle**

- Groups of animals of a single sex are dosed in a stepwise procedure using the fixed doses of 5, 50, 300 and 2000 mg/kg (exceptionally an additional fixed dose of 5000 mg/kg maybe considered). The initial dose level is selected on the basis of a sighting study as the dose expected to produce some signs of toxicity without causing severe toxic effects or mortality.

Table No 1: showing study design for sighting study for acute toxicity.

Sl no	Dose Level (mg/Kg)	No. of mice	Weight of the mice (gms)	Calculation of dose (mg)	Volume administered Dose in ml
1	5	H	32	0.16	0.47
		B	33	0.165	0.48
		T	30	0.15	0.44
		HB	31	0.15	0.44
		BT	32	0.16	0.47
		TH	30	0.15	0.44
2	50	H	21	1.05	0.30
		B	32	1.6	0.47
		T	26	1.3	0.38
		HB	30	1.5	0.44
		BT	33	1.65	0.48
		TH	24	1.2	0.35
3	300	H	22	6.6	0.41
		B	26	7.8	0.48
		T	23	6.9	0.43
		HB	21	6.3	0.39
		BT	22	6.6	0.41
		TH	26	7.8	0.48
4	2000	H	31	62	0.44
		B	32	64	0.45
		T	33	66	0.47
		HB	33	66	0.47
		BT	31	62	0.44
		TH	32	64	0.45

Table No.2: Shows Behavioral profile of experimental animals of sighting study.

Behavior	5mg	50mg	300mg	2000mg
Alert	++	++	++	+
Stereotype	+	+	-	-
Grooming	-	-	+	+
Immediate touch response	+	+	+	+
Vocalization	+	+	-	-
Itching	+	+	-	+

Table No.3: shows Neurological Profile of experimental animals of sighting study.

Observations	5mg	50mg	300mg	2000mg
Convulsions	-	-	-	-
Righting reflex	+	+	+	+
Piloextension	-	-	-	+
Straub's tail movement	-	-	-	-
Tremor	-	-	-	-
Jerky movements	-	-	-	+
Dilated pupil	-	-	-	-
Opisthones	-	-	-	-

**Mortality of the rats:** In groups of 5mg, 50mg, 300mg and 2000mg no mortality was observed.

#### Experimental Study

**Hyperlipidemia,<sup>[10]</sup> hyperlipoproteinemia, or hyperlipidaemia** (British English) is a heterogeneous group of disorders characterized by an excess of lipids in

the bloodstream. [It is the most common form of dyslipidemia (which includes any abnormal lipid levels)]. Albino Wistar rats (200 – 250 g) were obtained from the Central Animal House, V.L. College of Pharmacy Raichur, and housed in a group of six animals, for one week, in a 12:12 hour light and dark cycle, in a temperature and humidity controlled room. The animals

were given free access to food and water. After the one-week adaptation period, the healthy animals were used for the study.

## MATERIALS AND METHODS

### Materials

1. Vanga bhasma (Test drug)
2. Sibutramine (Standard drug)
3. Cafeteria diet (Control)
4. Albino rats (Wistar Strain)

### Sample size

36 albino rats were taken for the experimental study, distributed 6 in each group.

### Determination of Antihyperlipidaemic activity

Cafeteria diet induced Hyperlipidaemia in rats- cafeteria diet includes 3 diets.

- 1) 48 g condensed milk, 48 g of bread.
- 2) 18 g of chocolate, 36 g of dried coconut.
- 3) 48 g of cheese, 60 g of potatoes.

- These diets consist of a simple exchange of carbohydrate-derived calories with fat-derived calories when compared to low fat or chow control diets.
- Three diets will be given to 6 groups of rats each containing 6 rats on days 1, 2, 3, and then repeated for 40 days in same succession, in addition to normal pellet chow diet.

### Experimental Protocol

- Group 1-Normal control which receives normal pellet chow diet for 40 days
- Group 2-Cafeteria diet control + normal pellet chow diet 40 days
- Group 3-Cafeteria diet + Sibutramina (5mg/kg) for 40 days
- Group 4-Cafeteria diet + Lower dose of Vanga bhasma for 40 days
- Group 5-Cafeteria diet + Medium dose of Vanga bhasma for 40 days
- Group 6-Cafeteria diet + High dose of Vanga bhasma for 40 days.

**Fixation and preparation of rat dose:** The classically advised, normal human adult dose of Vanga Bhasma is 1-2Ratti, which is equal to 125-250mg.

## METHOD

36 healthy albino rats (18 male and 18 female) were taken for the experiment.

1. The rats were divided into 6 groups of six animals in each group.
2. Male and female rats were kept in different cages.
3. On 0 day all the animals were starved for 14 hrs and water ad libitum.
4. On 1<sup>st</sup> day, blood was drawn from all orbital

plexuses of albino rats and serum was separated.

5. Then medicines (Test drug, Standard drug and Control drug) were administered daily once for 40 days.
6. On 40<sup>th</sup> day, morning in all the groups drugs were administered to all the rats then from evening all the animals were starved for 14 hrs and water ad libitum.
7. On 41<sup>th</sup> day, blood was drawn from all albino rats and serum was separated for the analysis.

Wistar albino rats (36) of either sex weighing 200 g to 250 g of 36 rats were divided into six different groups, six in each group.

**Control group** rats were administered with Gum Acasia at a dose of 0.5 ml /kg with Normal control which receives normal pellet chow diet and water for 40 days.

**The second group** rats were administered with Gum Acasia<sup>[11]</sup> at a dose of 0.5 ml /kg CD +normal pellet chow diet for 40 days.

**The third Group** was administered with - CD + Sibutramina (5mg/kg) for 40 days.

Animals	Body weight	Dose in mg	Dose in ml
H	225	1.125	0.5
B	225	1.125	0.5
T	225	1.125	0.5
HB	225	1.125	0.5
BT	225	1.125	0.5
TH	225	1.125	0.5

**The fourth group** were administered with- CD + Low dose of VB for 40 days (1/20<sup>th</sup> of LD<sub>50</sub> i e 100 mg).

Animals	Body weight	Dose in mg	Dose in ml
H	250	25	0.5
B	220	22	0.44
T	180	18	0.36
HB	190	19	0.38
BT	190	19	0.38
TH	215	21.5	0.43

**The fifth group** were administered with CD + Medium dose of VB for 40 days (1/10<sup>th</sup> of LD<sub>50</sub> i e 200 mg).

Animals	Body weight	Dose in mg	Dose in ml
H	200	20	0.2
B	210	21	0.21
T	225	22.5	0.225
HB	250	50	0.5
BT	180	18	0.18
TH	190	19	0.19

The Sixth group were administered with CD + High dose of VB for 40 days (1/5<sup>th</sup> of LD<sub>50</sub> i.e 400 mg).

Animals	Body weight	Dose in mg	Dose in ml
H	180	72	0.36
B	250	100	0.5
T	190	76	0.38
HB	200	80	0.4
BT	225	90	0.45
TH	200	80	0.4

As the body weight increase dose also increases. Test

## EXPERIMENTAL RESULTS

**Table No 5: Change in Body weight (Gm) (mean±SEM) of different Groups of rats selected for the experimental study on different time intervals.**

Sl. No	Groups	0 day	1 <sup>st</sup> week	2 <sup>nd</sup> week	3 <sup>rd</sup> week	4 <sup>th</sup> week	5 <sup>th</sup> week	6 <sup>th</sup> week
1	Control group	190±4.472	194.166±4.556	199±4.487	203.5±3.998	204.833±3.410	209.166±4.415	212.833±4.556
2	Cafeteria diet+normal pelleted chow	197.5±2.814	203±3.138	213.373±2.348	227.166±2.548	237.33±2.404	248.66±3.756	259.5±3.594
3	CD+Standard (sibutramine)	200±0.00	205±1.033	211.166±2.348	217±1.789	225.5±1.086	231.166±1.887	237.666±1.476
4	CD+Lowerdose of VB	207.5±10.626	214±10.507	221.66±10.213	230.166±10.035	237.66±9.584	246.166±10.166	253.33±1.476
5	CD+Medium dose VB	209±10.367	213.166±10.550	217.833±10.297	224.166±9.910	229.166±9.946	234.66±9.965	241.66±9.878
6	CD+Highdose VB	207.5±10.468	212±10.142	216.166±10.142	220±10.045	223.06±9.744	227±9.496	232.16±9.63

**Table No 6: The % change in biochemical parameters due to the administration of standard drug & Vanga Bhasma (low, medium, high doses) in Cafeteria diet induced hyperlipidemic rats.**

Parameters	Normal Control	Cafeteria +normal pellet diet	CD+Sibutramine	CD+Low dose VB	CD+Medium dose VB	CD+High dose VB
RBS	6.831±1.308	46.346±2.734	15.584±6.051	39.914±3.700	17.354±1.549	13.058±3.243
UREA	29.623±10.539	96.137±14.541	31.2636±12.425	87.962±10.302	57.897±4.005	50.172±12.068
Creatinine	15.277±8.170	45.574±7.343	13.491±7.936	63.686±19.00	45.972±7.215	53.519±8.779
SGOT	12.799±3.264	33.0666±7.683	18.274±5.485	28.488±6.055	20.171±7.495	16.032±2.818
SGPT	10.585±2.822	45.545±8.819	14.627±2.959	21.152±3.234	22.523±2.958	12.576±2.112
ALP	8.726±1.902	29.84833±3.436	13.4918±3.120	35.180±3.148	22.601±1.430	18.4023±1.094
T PROTEIN	5.383±0.9018	18.553±3.360	7.399±2.430	16.617±2.430	12.426±1.114	6.642±1.895
ALBUMIN	6.0253±0.8750	35.6126±5.134	11.024±2.696	31.945±4.956	21.867±3.093	18.0466±3.548
GLOBULIN	4.948±0.7057	23.734±1.723	8.545±2.406	11.809±1.290	13.080±0.890	6.260±1.486
T CHOLESTEROL	5.0720±0.3869	32.226±1.783	8.714±0.8847	24.6495±0.9824	22.467±0.8179	8.225±0.5815
TRIGLYCERIDES	7.8761±0.6980	35.948±3.474	11.745±1.232	27.258±1.257	18.618±1.857	14.280±2.420
HDL	16.9145±2.678	3.06366±0.4518	20.99016±2.2064	2.81605±0.4562	4.23666±0.8157	9.5203±1.557
LDL	8.9025±0.7354	28.141±2.407	11.660±1.393	24.555±2.334	18.839±1.527	12.866±1.416
VLDL	8.173±1.404	33.703±10.870	15.499±2.448	33.491±7.473	30.689±4.740	19.712±2.987

Table No.7: Caffeterian Diet +Normal pallet diet (GroupII Vs all Grops).

Biochemical parameters	Controlgroup	Sibutramine + CD	Low doseVB+CD	Medium dose VB+CD	High dose VB+CD
RBS	6.831±1.308 (**P<0.01)	15.584±6.051 (**P<0.01)	39.914±3.700 ns(P>0.05)	17.354±1.549 (**P<0.01)	13.058±3.243 (**P<0.01)
Urea	29.623±10.53 (*P<0.05)	31.2636±12.425 (*P<0.05)	87.962±10.302 ns(P>0.05)	57.897±4.005 ns(P>0.05)	50.172±12.068 (*P<0.05)
Creatinine	15.277±8.170 (*P<0.05)	13.491±7.936 (*P<0.05)	63.686±19.00 ns(P>0.05)	45.972±7.215 ns(P>0.05)	53.519±8.779 (*P<0.05)
SGOT	12.799±3.264 (*P<0.05)	18.274±5.485 (**P<0.01)	28.488±6.055 ns(P>0.05)	20.171±7.495 ns(P>0.05)	16.032±2.818 (**P<0.01)
SGPT	10.585±2.822 (**P<0.01)	14.627±2.959 (**P<0.01)	21.152±3.234 ns(P>0.05)	22.523±2.958 (*P<0.05)	12.576±2.112 (**P<0.01)
ALP	8.726±1.902 (**P<0.01)	13.4918±3.120 (**P<0.01)	35.180±3.148 ns(P>0.05)	22.601±1.430 (*P<0.05)	18.4023±1.094 (**P<0.01)
Total protein	5.383±0.9018 (**P<0.01)	7.399±2.430 (**P<0.01)	16.617±2.430 ns(P>0.05)	12.426±1.114 ns(P>0.05)	6.642±1.895 (**P<0.01)
Albumin	6.0253±0.8750 (**P<0.01)	11.024±2.696 (**P<0.01)	31.945±4.956 ns(P>0.05)	21.867±3.093 ns(P>0.05)	18.0466±3.548 (**P<0.01)
Globulin	4.948±0.7057 (**P<0.01)	8.545±2.406 (**P<0.01)	11.809±1.290 (*P<0.05)	13.080±0.890 (**P<0.01)	6.260±1.486 (**P<0.01)
Total Cholestrol	5.0720±0.3869 (**P<0.01)	8.714±0.8847 (**P<0.01)	24.6495±0.9824 ns(P>0.05)	22.467±0.8179 (**P<0.01)	8.225±0.5815 (**P<0.01)
Triglycerides	7.8761±0.6980 (**P<0.01)	11.745±1.232 (**P<0.01)	27.258±1.257 (*P<0.05)	18.618±1.857 (**P<0.01)	14.280±2.420 (**P<0.01)
HDL	16.9145±2.678 (**P<0.01)	20.99016±2.2064 (**P<0.01)	2.81605±0.4562 ns(P>0.05)	4.23666±0.8157 (*P<0.05)	9.5203±1.557 (**P<0.01)
LDL	8.9025±0.7354 (**P<0.01)	11.660±1.393 (**P<0.01)	24.555±2.334 ns(P>0.05)	18.839±1.527 (**P<0.01)	12.866±1.416 (**P<0.01)
VLDL	8.173±1.404 (**P<0.01)	15.499±2.448 (**P<0.01)	33.491±7.473ns (P>0.05)	30.689±4.740 (*P<0.05)	19.712±2.987 (**P<0.01)

ns - p > 0.05 , \* - p < 0.05 , \*\* - p < 0.01, \*\*\* - p < 0.001

In all the above tables the study was significant at the level of \*\* **p<0.01** by adopting Dunnett's multiple comparison test.<sup>[12]</sup> From all the above calculations it is evident that Vanga Bhasma showed a considerable Antihyperlipidaemic activity.

## CONCLUSION

- Vanga Bhasma has Shown no mortality or behavioural abnormality recorded in mice at the highest dose level of 2000 mg/kg tested for LD<sub>50</sub> studies.
- The Standard drug (Group III) at **\*\*P<0.01**, High dose of VB (Group VI) at **\*\*P<0.01** and medium dose VB (Group V) at **\*P<0.05** shown significantly reduced the serum biochemical parameters like serum Glucose, Total Cholestrol, Triglycerides, LDL and VLDL cholesterol levels, Urea, Creatinine, Alkaline phosphate, Protein, SGOT, SGPT, Albumin, Globulin, with an increase in HDL levels in CD induced hyperlipidemic rats.
- Cafeteria diet (Group II) **ns P>0.05**, Low dose of VB (Group IV) **ns P>0.05** shown insignificantly decrease in the serum biochemical parameters like serum Glucose, Total Cholestrol, Triglycerides, LDL and VLDL cholesterol levels, Urea, Creatinine,

Alkaline phosphate, Protein, SGOT, SGPT, Albumin, Globulin with an increase in HDL levels in CD induced hyperlipidemic rats.

- VB at High dose and Medium dose owing to its Rasadi gunas and Lekhana, Medhophakarma helps in Samprapthi vighatana (breaking the Pathogenesis) of Medoroga which is due to impaired kapha and vata dosha and helps in reducing the biochemical parameters and regulation of metabolism, As no physiological or adverse behavioral changes were noticed in the experimental animals, so it is considered as a safer drug for Hyperlipidaemic condition.

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