



## EFFECT OF RAJATA (SILVER) BHASMA ON ACQUISITION MEMORY:

### AN EXPERIMENTAL STUDY

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#### ABSTRACT

*Background: In the present era, cognitive deficits present with many neuropsychiatric conditions are faced by all age groups, due to the stressful lifestyle. In Ayurveda, Medhya refers to one which enhances memory power (acquisition memory), drugs which increases memory are known as Medhya dravyas. The Medhya property of Rajata (silver) Bhasma can be traced in classical texts like Rasa Tarangini, Rasa Ratna Samuchaya etc. In order to boost and improve cognitive functions, this current paper aims at the experimental study of Rajata Bhasma, investigated for acquisition (learning) memory in Wister albino rats. Methods: A quality Rajata (silver) Bhasma was prepared in Rasashastra and Bhaishajya Kalpana Department of SDMACH Hassan, India according to the classical guidelines. The experimental study was carried out at the SDM Research center, Udupi, India. Methodology consists of three groups of rats, test group was administered with Rajata Bhasma (11.25mg/kg per day p.o.), standard group was administered piracetam (100mg/kg per day i.p.) and control group was fed with food and water, to assess the Medhya activity in Wister albino rats followed by the Morris water maze test. The escape latencies, acquisition memory were recorded, results are analysed statistically with the help of parameters to assess the memory. Results: Indicate that Rajata (silver) Bhasma has moderate effect on learning and acquisition, statistically significant result on the paradigm of thigmotactic behaviour average duration and frequency. Conclusion: Rajata (silver) Bhasma has facilitatory effect on acquisition memory (learning and memory). Key words – Medhya, Rajata Bhasma, acquisition memory, Morris water maze test, piracetam*



## INTRODUCTION

Cognitive deficits present with many neuropsychiatric conditions are faced by all age groups, due to the stressful lifestyle. Poor memory, lower retention and slow recall are commonly faced in today's competitive world as amnesia is major health problem. Works to evaluate and bring about effective safe Medhya<sup>1</sup> (nootropic) drugs is the need of the hour. But there are very few systematic documented studies which can be used to evaluate the efficacy of Rajata Bhasma. Experimental protocols help Ayurveda to develop drug dose protocol and to evaluate the therapeutic effect of the drug. In the present era for assuring safety and to get global acceptance many experimental studies are conducted in the field of Ayurveda. Memory is the registration, retention and recollection of experiences, thoughts, feelings, sensations, ideas and knowledge.<sup>2</sup> Acquisition is the process of storage and retrieval of new information in memory. In Ayurveda, Medhya<sup>3</sup> (learning and memory) refers to one which enhances Medha<sup>3</sup> (memory power), which either grasps or

retains knowledge. Rajata Bhasma, which is the focus of the present study, was prepared by following classical guidelines<sup>4</sup> in the department of Rasashastra, SDMCAH, Hassan, India. It has been described as Medhya by various classical texts of Rasashastra<sup>5</sup> hence was investigated for acquisition (learning) memory viz. Morris water maze test.

## METHODOLOGY:

### MATERIALS AND METHODS:

Materials:- Measuring glass, Insulin syringe (2 ml), Stop watch, mortar & pestle distilled water etc. Experimental animals: Selection of animal species, healthy adult albino female rats weighing between 190-210g were procured from animal house attached to pharmacology laboratory of SDM Centre for Research in Ayurveda & Allied Sciences, Udupi, Karnataka, India. The animals were fed with Amrut brand rat pallet feed and tap water throughout the study period. The rats were grouped into 3 groups with 6 animals in each group. In each group the animals were marked with yellow colour to different body parts to permit individual identification. Drugs and treatment; Test drug- a quality Rajata Bhasma was prepared according to the classical reference, at department of Rasashastra, SDM College of Ayurveda, Hassan, India. Standard drug- Piracetam injection, brand name-



Nootropil by Dr Reddy's was obtained from pharmaceuticals, Udupi. Dose calculation: The classical dose of Rajata Bhasma is 1 Ratti (125mg). The dose for experimental study was calculated by referring Paget's and Barnes (1964) chart. It was done as follows, human dose  $\times 0.018 \times 5$ , dose for rats = human dose  $\times 0.018 \times 5$  (i.e  $125 \times 0.018 \times 5 = 11.25$  mg/kg) body weight. For Standard drug- Piracetam  $100\text{mg/kg} = 0.1\text{mg/g}$   $0.1\text{ml} = 10\text{mg}$ ,  $0.1\text{ml}$  piracetam was taken and administered through i.p as per body weight. The test drug was administered by oral route and the standard drug was administered intra peritoneal.

Morris water maze test<sup>6</sup>: The technique of this test is that rats are placed into large circular pool of water, can swim and escape on to hidden platform (refer figure no.1). Only one platform is hidden by its placement just below 2cm of the water

surface and is kept constant. Thus the platform offers no local cues to guide escape behaviour. The rat can escape from swimming by climbing on to platform and overtime the rat apparently learns the spatial location of the platform from any starting position at the circumference of the pool. Before the training started, rats were allowed to swim freely into the pool for 60 sec without platform. They were given 4 trials (once from each starting position) per session for 6 days; each trial had a ceiling time of 60 sec and a trial interval of approximately 30 sec. After climbing on to the platform, the animal was allowed to remain there for 30 sec before the commencement of the next trial. Latency to reach the platform was recorded. If the rat failed to reach the escape platform within the maximum allowed time of 60 sec, it was gently placed on the platform and allowed to remain there for the same interval of time. On 7<sup>th</sup> day of drug treatment - by repeating the procedure, acquisition memory at all the regions (north, west, south, east), thigmotatic duration and thigmotatic frequency were recorded.<sup>10</sup>



Figure no.1: Morris water maze



Figure no.2: Administration of Rajata bhasma

Figure no.3: : Administration of Piracetam

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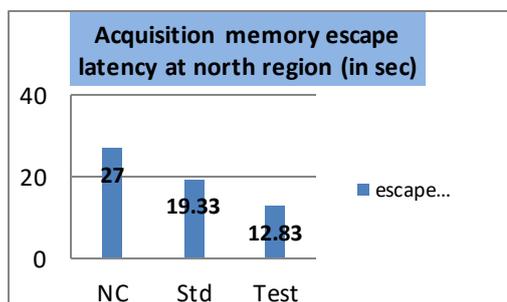


Statistical data analysis:

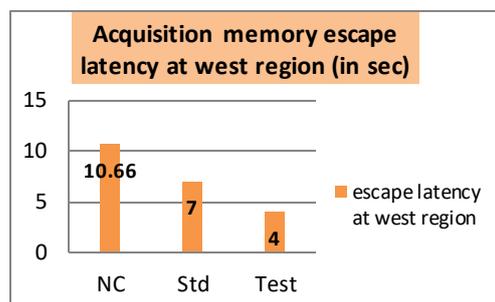
All the values were expressed as mean SEM (Standard Error of Mean). The data was analysed by one way ANOVA.

Acquisition memory; recorded at all the 4 quadrants.

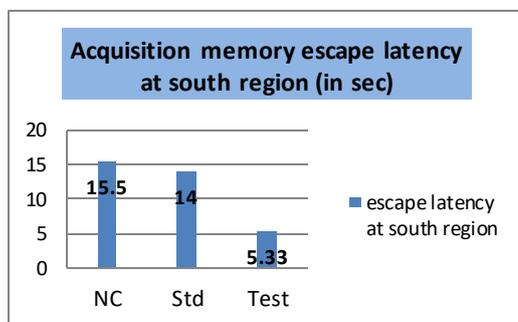
*Graph no: 1 (a): Effect of Rajata Bhasma on Acquisition memory at north region*



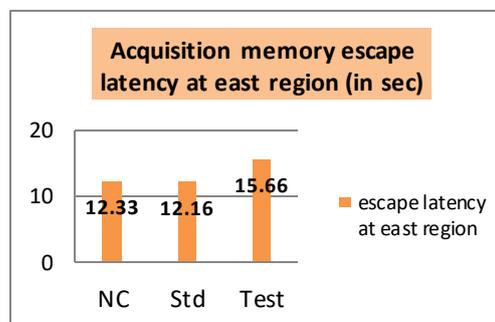
*Graph no: 1 (b): Effect of Rajata Bhasma on Acquisition memory at West region*



*Graph no: 1 (c): Effect of Rajata Bhasma on Acquisition memory at south region*



*Graph no: 1 (d): Effect of Rajata Bhasma on Acquisition memory at east region*



NC- Normal control group  
Std- Standard group  
Test – Rajata Bhasma group

*Graph no: 1 (a): Effect of Rajata Bhasma on Acquisition memory at north region*

The data in the graph no.1(a); shows there was decrease in escape latency at north region in Rajata bhasma group when compared to the normal control group, the observed decrease was found to be statistically non significant.

*Graph no: 1 (b): Effect of Rajata Bhasma on Acquisition memory at West region*

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The data in graph no.1(b); shows there was decrease in escape latency at west region in Rajata Bhasma group when compared to the normal control group, the observed decrease was found to be statistically non significant.

*Graph no: 1(c): Effect of Rajata Bhasma on Acquisition memory at south region*

The data in graph no. 1(c); shows there was decrease in escape latency at south region in Rajata Bhasma group when compared to the normal control group, the observed decrease was found to be statistically non significant.

*Graph no: 1(d): Effect of Rajata Bhasma on Acquisition memory at east region*

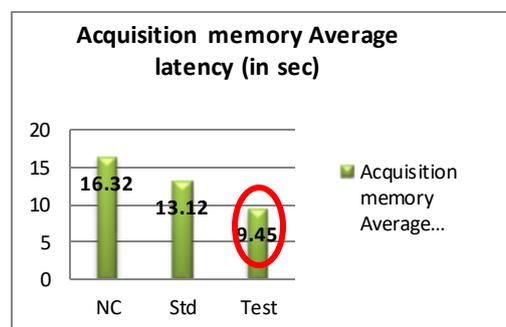
The data in graph no. 1(d); shows there was increase in escape latency at east region in Rajata Bhasma group when compared to the normal control group, the observed increase was found to be statistically non significant.

Average Latency;

*Table no. 1: Effect of Rajata Bhasma on acquisition memory at average latency:*

Group	Average latency (sec) MEAN±SEM	% changes
Normal Control	16.32±2.59	--
Standard	13.12±3.58	19.60↓
Rajata Bhasma	9.45±1.26	42.09↓

*Graph no: 2 Effect of Rajata Bhasma on Acquisition memory (Average Latency)*



Data: MEAN±SEM

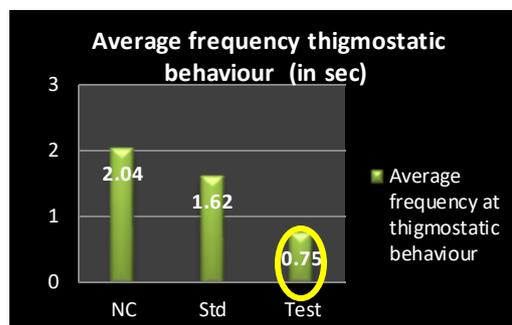
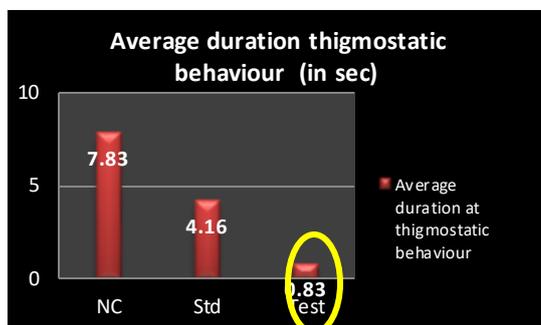
The data related to the effect of Rajata Bhasma on escape latency or average latency can be found in table no. 1; and graph no.2; shows there was decrease in escape latency in standard group and Rajata Bhasma group when compared to the normal control group, the observed decrease was found to be statistically non significant.

Thigmostatic Behaviour;

*Graph no: 3 Effect of Rajata Bhasma on Average duration Thigmostatic behaviour*

*Graph no: 4 Effect of Rajata Bhasma on Average frequency- Thigmostatic Behaviour*

*Graph no: 4 Effect of Rajata Bhasma on Average duration- Thigmostatic Behaviour*



The data in graph no.3; shows there was decrease in average duration of thigmostatic behaviour in Rajata Bhasma group when compared to the normal control group, the observed decrease was found to be statistically significant.

Graph no: 4 Effect of Rajata Bhasma on Average frequency- Thigmostatic Behaviour

The data in graph no.4; shows there was decrease in average frequency of thigmostatic behaviour in Rajata bhasma group when compared to the normal control group, the observed decrease was found to be statistically significant.

## RESULTS AND OBSERVATIONS

The data for different quadrants were acquired and quadrant based analysis and pooled data analysis was observed. In the test group, though shorter duration of latency to find the hidden platform was observed the prolongation did not reach statistically significant level for all the parameters but was statistically significant for thigmostatic average duration and thigmostatic average frequency.

Acquisition memory: Results are summarized in the graph no.1(a), (b), (c) and (d). When the rats in the reference standard groups were placed in northern, western, southern and eastern region exhibited decrease in the

escape latency indicating faster learning in comparison to normal control group. Where as in Rajata Bhasma treated group significant shortening was observed when the animals were tested in all the quadrants compared to standard and control, it showed moderate effect compared to other groups.

Average latency: Analysis of the combined data for all the quadrants showed significant reduction in escape latency in the test drug group can be seen in graph no.2. Rats in Rajata Bhasma group, performed good in comparison to the other groups, but showed moderate effect when compared to normal and standard groups (MEAN±SEM 13.12±3.58). The



effect on how fast the rats learn to search and reach the hidden platform was assessed in different groups. The Rajata bhasma group rats, (MEAN $\pm$ SEM 9.45 $\pm$ 1.26) required less time to learn, to locate, the hidden platform indicating that it has learning promoting property probably through faster establishment of working and spatial memory.

Thigmotactic behaviour average duration: Results are summarised in graph no. 3. The tendency of the rats to keep close to the side of the pool as they swim in the Morris water maze is known as thigmotaxis and was recorded during different stages of the experiment. The data generated shows that the duration of thigmotactic behaviour was significantly reduced by reference standard and Rajata bhasma treated groups. But Rajata Bhasma (p<0.05) treated group, produced statistically significant result.

Thigmotactic behaviour average frequency: Results are seen in graph no. 4. The data generated shows that the thigmotactic behaviour frequency was significantly reduced by reference standard and Rajata Bhasma treated groups compared to normal group but Rajata Bhasma treated group produced statistically significant result (p<0.05). This indicates that Rajata Bhasma has moderate effect on acquisition (learning) memory.

## DISCUSSION

Rajata Bhasma has been extensively used as medhya rasayana<sup>8</sup>. Medhya rasayana acts as a brain tonic and cognition facilitator in Ayurveda.<sup>9</sup> Rajata Bhasma has been characterised as globular particles of silver with an average size of 20-100 nm, which directly enters blood stream and stimulates the sensory register & triggers the targeted hippocampus region of brain which may result in enhancement of memory. Medhya drugs act by nutritive mechanism, providing micro nutrients to the brain cells and improve their action. Rajata Bhasma is basically a tridosahara<sup>10</sup> with qualities like Kashaya, Sheeta Veerya, Laghu, Singdha guna, Agni, Bala vardaka, Vrishya, Brumhana, Rasayana, Sarva roga hara and Smruti vardaka etc mainly indicated in all type of manovikaras.<sup>11</sup> These properties increases the kapha, which is responsible for nutrition of body cells and nourishment of brain cells. Its absorption and assimilation in body is very fast due to its Laghu guna and because as it is in bhasma form nano size particles. As it possesses all the above qualities it; maintains the balance of all Manasika and Shareerika dosha. Analysis of Rajata Bhasma was done in the MIT College of Manipal, India by instrument models like SEM; showed the size of the silver particles ranging in nanometres, making it easy for absorption into the blood. Piracetam is



the prototype of this class of psychotropic agents which can facilitate learning acquisition and retention.<sup>12,13</sup> Measuring thigmotaxis (results of graph no. 3 and 4) gives an indication of whether the rat is making use of spatial learning or non-spatial strategies. It can be considered to provide an indication of problem-solving capability of the rat. Excessive thigmotaxis suggests that the animal is not recognizing that the platform is the goal, or not focusing on the task of finding it. This may be the result of drugs interfering with sensor motor function. The results of Rajata Bhasma treated group are statistically significant ( $p < 0.05$ ) for both the thigmotactic duration and frequency. It clearly shows that the test drug ensures proper functioning of sensor motor function resulting in acquisition of spatial memory in quicker time. In the present study Rajata Bhasma can be regarded as a memory enhancer in view of its

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facilitatory effect on acquisition memory on paradigms used and results are comparable to piracetam.

## CONCLUSION

The Medhya (learning and memory) effect of Rajata bhasma in Wistar albino rats, can be concluded based on the results for acquisition memory. Rajata Bhasma has a statistically significant result on thigmotactic average duration and frequency; it may act as memory enhancer, it has over all moderate effect on acquisition (learning) memory. However this can be established after further detailed studies on various parameters like retention memory, long term memory etc using different protocols. Further detailed histopathological studies of brain, relating learning and memory effect in various CNS disorders can be elucidated.

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