



Research Journal of Pharmaceutical, Biological and Chemical Sciences

Evaluation of Memory Enhancement Activity and Shock Motivated Brightness Discrimination Response by using Y-Maze.

D Eswar Tony*, T Vijaya, D Sathish Kumar, M Sathish Kumar, A Narendra Babu, and
N Rama Rao

Department of Pharmacology, Chalapathi Institute of Pharmaceutical Sciences, Guntur, Andhrapradesh.

ABSTRACT

Spatial memory, according to neuroscience, is the part of memory responsible for recording one's environment and its spatial orientation. For example, spatial memory is needed to learn the location of food at the end of the maze. It is often argued that in both humans and animals. Spatial memory has representations within working, short term and long term memory. Thus for measuring the spatial memory many mazes were came into existence such as Y-maze, rectangular maze and labyrinth maze. Here is an introduction to Y maze and their purpose, procedure and evaluation of memory enhancement activity.

Keywords: Spatial memory, Y-maze, Memory enhancement, spontaneous alterations.

**Corresponding author*



INTRODUCTION

Spontaneous alteration in rats and mice refers to the natural tendency to spontaneously choose alternative arms in mazes [1]. This spontaneous alteration behavior has been ascribed to the operation of a variety of mechanisms. It is evident that the animal must remember to which arm it had entered on a previous occasion to enable it to alternate its choice on a following trial. Therefore, spontaneous alteration has been embraced by behavioral pharmacologists as a quick and relatively simple test of spatial working memory, devoid of fear, reward of reinforces. Free running or continuous spontaneous alteration in symmetrical Y-mazes or other mazes was first applied to the study of drug effects. The basic procedure allowed to individual rats and mice free access to the arms of the mazes for several minutes during which the time sequences of entries of the arms were recorded. From this sequence, proportions of arms entered that were different from those previously visited were calculated to produce an alternation score.

Y Maze spontaneous alteration is a behavioural test for measuring the willingness of rodents to explore new environments. Rodents typically prefer to investigate a new arm of the maze rather than returning to one that was previously visited. Many parts of the brain including the hippocampus, septum, basal forebrain, and prefrontal cortex are involved in this task [2].

EXPERIMENTAL ANIMALS

Wistar rats of either sex (200-300g) were maintained for 7 days in the animal house of Chalapathi Institute of Pharmaceutical Sciences, Guntur under standard conditions temperature (24 ± 10 C), relative humidity (45-55%) and 12:12 light: dark cycle. The animals were fed with standard rat pellet and water ad libitum. The animals were allowed to acclimatize to laboratory conditions 48 h before the start of the experiment. 5 rats/group were used in all sets of experiments. All the experiments were conducted after obtaining permission from the Institutional Animal Ethics Committee (IAEC) Chalapathi Institute of Pharmaceutical Sciences, Guntur.

Purpose and Activity

The mazes are used mainly to measure the intensity of the spatial memory by comparing with standard drugs of memory suppressors and also memory enhancers. Many new drugs like synthesized products and other herbal extracts are used for the testing of memory enhancement activity.

Equipment: Y-maze

Y-maze

The Y-maze is designed for studying shock motivated brightness discrimination response, i.e., simultaneous brightness discrimination in rats [4]. The Y-maze has been designed to make the animal to learn to discriminate between two arms- one illuminated

without shock and other non-illuminated with shock and learn to reach the correct 'arm', the illuminated one.

The spatial working memory was measured through the spontaneous alteration of behaviour in Y-maze (INCO). The Y-maze consists of three identical removal sun mica lined chambers arranged in Y-shape connected to the central chamber. Each arm has a working dimension of approx. 30 ×15× 15 cm with rat presence indicator and hinged top. Each mouse is placed in the central chamber and allowed to move freely through the maze during an 8-minute session. The mouse trend to explore the maze systematically, entering each arm in turn. When mouse enters one arm the rat presence indicator glows. The series of arm entries was recorded. Alteration is defined as the number of successive entries in to the three arms on overlapping triplet sets. The percentage alteration was calculated as the ratio of actual to possible alterations [3].



FIGURE 1

These kinds of experiments are known as simultaneous discrimination learning where both CS+ and CS- stimulus are present together simultaneously.

- Y-maze is divided into 4 parts. It consists of
 - Three identical removable sun mica lined chambers arranged in Y shape connected to central chamber.
 - The central chamber is about 20 cm height with three openings which allow the other three arms to be connected.
 - Each arm has its own power plugs and is connected to the semiautomatic power controller tool.
 - Each arm has a working dimension of approximately 30X15X15 cms with electric grill and has chamber light or cue light with indicator, grill charge indicator, rat presence indicator and hinged top. The central compartment also contains a wire grill.
 - Each chamber along with central compartment contains grills which gives electric shock.



FIGURE 2

- After assembling the unit testing for correct assembly should be done to ensure the correct working of the apparatus. Testing of the Y-maze is performed by the following steps
 - Connect the mains lead to power outlet and switch the unit ON.
 - Set the 'program selector' rotary switch to any one of the three program positions (A/B/C) and compare the illuminated lights on replica of maze on control unit to that on the corresponding arm.
 - If the program selector is set to position A, then the cue light appears in arm A and the other two arms i.e., B and C have shock without cue light.
 - Similarly if the program selector is set to position B, then the cue light appears in arm B and other two arms A and C has shock without cue light, similarly this transversion applies to arm C.
- This completes the installation and testing of Y-maze.



FIGURE 3 & FIGURE 4

Procedure

- A group of rats are trained to provide colonies for pharmacological studies.
- Every rat, in turn is placed in one of the chambers for a period of 30 seconds without any stimulus to allow an accommodation to the stimulation.
- The naive animal is allowed to explore Y-maze apparatus for 5 min at the start of their training.

- The rat is then put into central compartment and set the program selector accordingly.
- After 5 sec (approximately) shock is applied by switching the unit ON and by using the program selector set the position to either A/B/C.



FIGURE 5

- Observe the indicators till the animal reaches the goal in the illuminated arm.
- Shift the positions of the program selector by rotating and note the observations as follows
 - First preference of the rat to illuminated or non-illuminated arms
 - Number of entries in illuminated arm and non-illuminated arm. (An arm entry is defined as the entry of four paws into the arm)
 - Average time each animal spends in each arm (Average time = Total duration in arm/Number of entries)
- Inject standard or test drug to the animal group, after 30 mins, place the animal individually in the center of the maze and note all parameters.
- Compare the preference of animal to illuminated or non-illuminated arms, average time spent in illuminated or non-illuminated arms and number of entries in both the arms.

Selection of Dose and Treatment period

The test animals were randomly chosen and divided into four groups having five rats in each as follows:

- Group-1- Control group (0.9% Normal saline 5ml/kg i.p),
- Group-2 – Standard (Diazepam at a dose of 100mg/ kg i.p),
- Group-3 – Piracetam (300mg kg i.p),
- Group-4 – Diazepam+ Piracetam, Piracetam pretreated with Diazepam.

Here we can also estimate the number of choices of the arms chosen by the animals and the memory enhancement activity can be evaluated with the animals which opted the correct choice in the respective program either A/B/C.

TABLE (1) Choices taken by the animal at different treatment groups

S.No	Group	Trails	Treatment	Program			Correct Choice	No. of Correct Choices
				A	B	C		
1	I	1	Control	L	S	S	N	2
		2		S	S	L	Y	
		3		S	L	S	Y	
		4		S	S	L	N	
2	II	1	Diazepam (Standard)	L	S	S	N	1
		2		S	S	L	N	
		3		S	L	S	Y	
		4		S	S	L	N	
3	III	1	Piracetam	L	S	S	Y	3
		2		S	S	L	N	
		3		S	L	S	Y	
		4		S	S	L	Y	
4	IV	1	Diazepam + Piracetam	L	S	S	N	2
		2		S	S	L	Y	
		3		S	L	S	N	
		4		S	S	L	Y	

L – Light, S – Shock Y – Yes, N – No

STATISTICAL ANALYSIS

The values are expressed as mean± SEM. The results were analyzed for statistical significance using student’s t-test.

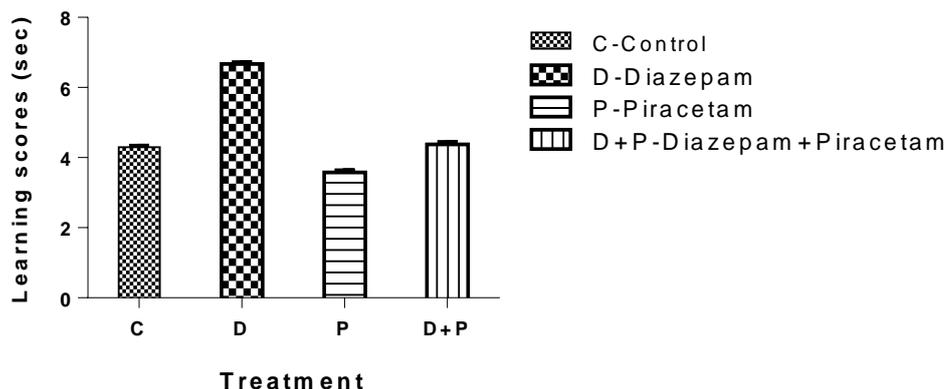
Evaluation

The average time taken to reach the arm is estimated by calculating the total time taken to reach the three arms divided by number of arms.

RESULTS AND DISCUSSION

Preclinical evaluation is necessary in all instances. As the neuroscience plays an important role in our daily life, it is necessary to improve the techniques in case of memory enhancement which leads to the advancement of treating diseases like Alzheimer’s. Behavioural studies also play a vital role and by placing a step forward in this results in treating some of the psychotic and neuroleptic disorders. In this regard Y-maze is one of the very useful models for evaluation of neurodegenerative disorders related to aging, Alzheimer’s, drug screening and phenotyping. In the present review control and standard showed significant learning remembering (memory) and reasoning enhancing activity.

Evaluation of memory enhancement activity by using Y-maze



CONCLUSION

The number of correct choices increases for group of animals which are treated with the drug piracetam because of having memory enhancement activity and the standard drug Diazepam increases the choosing time due to its depressant action. The animals which are pretreated with the piracetam before administering Diazepam has also better activity when compared with the Diazepam alone. The parameters like the time taken to reach the correct arm and total time spent in the arm are to be calculated for the difference in the activity of the drugs.

REFERENCES

- [1] Detrait, E. Proceedings of Measuring Behavior 2010:141-144.
- [2] http://sbfnl.stanford.edu/cs/bm/lm/bml_ymaze.html
- [3] Abhisek. Pharmacologyonline 2010;3:102-111 .
- [4] Hughes RN. Neurosci Biobehavioral Rev 2004; 28 : 497–505