AIM: Assessing Anti-Anxiety Effects through the Elevated Plus Maze in Mice

REFERENCE:

1) M.N. Gosh Common Laboratory Animals, Fundamentals of Experimental Pharmacology, Fifth Edition, 2011
2) Kulkarni S.K., Handbook of experimental pharmacology, New Delhi: Vallabh Prakashan, 2014.

INTRODUCTION:

Anxiolytic drugs are used to reduce anxiety without causing significant sedation or motor impairment. The study of anxiolytic activity in rats or mice involves the use of experimental models that mimic anxiety-related behaviors. Commonly used models include the Elevated Plus Maze (EPM), Open Field Test, and Light/Dark Box test. These models evaluate exploratory behavior, avoidance of open spaces, or preference for darker areas—behaviors that are altered under anxiety. Anxiolytic drugs like benzodiazepines typically increase the time spent in open arms (EPM) or illuminated areas (Light/Dark Box), indicating reduced anxiety levels. These animal models are useful for screening potential antianxiety agents.

REQUIREMENTS:

Animal :   Mice (20-25g)

Drug: Diazepam (2mg/kg)

Instrument: Elevated Plus Maze

PRINCIPLE:

The principle of studying anxiolytic activity in rats or mice is based on evaluating behavioral responses that reflect anxiety-like states. Mice naturally avoid open or brightly lit areas due to fear and stress. Anxiolytic drugs reduce this avoidance behavior, allowing increased exploration of such areas. Experimental models like the Elevated Plus Maze, Open Field Test, and Light/Dark Box rely on this innate behavior. A drug with anxiolytic properties will increase the time spent in open arms or lit zones and enhance exploratory activity, indicating a reduction in anxiety levels. These behavioral changes form the basis for assessing the efficacy of anxiolytic agents.

PROCEDURE:

1) Weigh and number the animals. Divide them into 2 groups each consisting of 4–5 mice. One group is used as control and other for drug (diazepam) treatment.
2) Place the animals individually in center of the maze, head facing towards open arm & start the stop watch and note following parameters for five minutes:

a)First preference of mouse to open or enclosed arm.

b)Number of entries in open and enclosed arm.

c) Average time each animal spends in each arm.

3) Inject diazepam to the test group. After 30 min place the animal individually in center of maze and note all parameters as described under step 2.

4) Compare the preference of animal to open or enclosed arm, average time spent in open arm and number of entries in open arm in each group.

OBSERVATION TABLE:

GROUPS	DIRECTION	READING	ANIMAL (MOUSE) No.					AVERAGE
			1	2	3	4	5	
Vehicle Treated	Open Arm	No. of entries						
		Total time spent						
	Close Arm	No. of entries						
		Total time spent						
Diazepam Treated (2mg/kg) i.p	Open Arm	No. of entries						
		Total time spent						
	Close Arm	No. of entries						
		Total time spent						

INFERENCE:

GROUPS	DIRECTION	READING	ANIMAL (MOUSE) No.					AVERAGE
			1	**2**	**3**	**4**	**5**	
Vehicle Treated	Open Arm	No. of entries	5	8	6	10	8	7.4
		Total time spent	15 sec	22 sec	28 sec	12 sec	14 sec	18.2 sec
	Close Arm	No. of entries	10	12	13	17	16	13.6
		Total time spent	210 sec	28 sec	115 sec	196 sec	175 sec	144.8 sec
Diazepam Treated (2mg/kg) i.p	Open Arm	No. of entries	15	16	19	22	18	18
		Total time spent	45 sec	56 sec	68 sec	75 sec	54 sec	59.6 sec
	Close Arm	No. of entries	12	10	12	15	13	12.4
		Total time spent	175 sec	185 sec	165 sec	155 sec	170 sec	170 sec

*Observation table after completion of the experiment can be downloaded by clicking tab (RJPT SimLab)
DISCLAIMER: "The results provided here are only for reference or comparison purposes. Students are expected to perform the experiment and record their actual observations."

CONCLUSION:

The present study demonstrated the anxiolytic effect of diazepam using the Elevated Plus Maze model in mice. Diazepam-treated animals showed a significant increase in the number of entries and time spent in the open arms compared to the control group, indicating reduced anxiety levels. This confirms that diazepam possesses anxiolytic properties by enhancing exploratory behavior and reducing fear-driven avoidance of open spaces. Thus, the model effectively evaluates the anxiolytic potential of pharmacological agents.

RESULT: Diazepam is anti-anxiety drug, as its effect is observed in drug treated group.