Chapter - V

Results and Discussion

The obtained results have been presented and discussed in the following section. We may recall that the present investigation had been designed to study the effect of different doses of morphine on retention of a single trial passive avoidance task.

For testing the hypothesis formulated in chapter III, an experiment based on a multigroup design was conducted.

Animals were trained in a passive avoidance task. They were divided into seven groups, six of which were injected intraperitoneally with various doses of morphine and one with NaCl solution, immediately after training. Retention tests were given 1, 2, 7 and 14 days after training.

The mean latency score of each group on the various retention tests were calculated and a suitable statistical test was applied to determine whether the groups differed significantly. Since multiple comparisons had to be made, Duncan's range Test was applied. McGuigan (1978) has recommended the use for this test for experiments based on multigroup design, since it is less time
consuming and easier to compute as compared to the alternative technique - Analysis of variance (ANOVA). If ANOVA had been applied the experimenter would have to calculate a number of 't values' after computing the 'f value'.

Table 1 showing significance of difference between the means of the Morphine and saline injected subjects, on the Retention Tests by using Duncan's Range Test.

<table>
<thead>
<tr>
<th>Groups</th>
<th>2</th>
<th>4</th>
<th>3</th>
<th>1</th>
<th>5</th>
<th>6</th>
<th>7</th>
</tr>
</thead>
<tbody>
<tr>
<td>Means</td>
<td>10.975</td>
<td>13.75</td>
<td>14.3625</td>
<td>198</td>
<td>207.375</td>
<td>227.5</td>
<td>255.625</td>
</tr>
<tr>
<td>p / .05</td>
<td>0</td>
<td>2.775</td>
<td>3.3875</td>
<td>187.025*</td>
<td>196.4*</td>
<td>216.525*</td>
<td>244.65*</td>
</tr>
<tr>
<td></td>
<td>13.75</td>
<td>0</td>
<td>.6125</td>
<td>184.25*</td>
<td>193.625*</td>
<td>213.75*</td>
<td>241.875*</td>
</tr>
<tr>
<td></td>
<td>14.3625</td>
<td>0</td>
<td>183.6375*</td>
<td>193.0125*</td>
<td>213.1375*</td>
<td>241.2625*</td>
<td></td>
</tr>
<tr>
<td></td>
<td>198</td>
<td>0</td>
<td>9.375</td>
<td>29.5</td>
<td>57.625</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>207.375</td>
<td>0</td>
<td>20.125</td>
<td>48.25</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>227.5</td>
<td>0</td>
<td>28.125</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>255.625</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
FIG. 3. SHOWING THE MEAN LATENCIES OF THE MORPHINE AND NaCl INJECTED GROUPS ON RETENTION TEST I (1 DAY)
From the above table, it is evident that on the first retention test, which was given 24 hours after training session mean latency score of group II (M 3mg/kg) which was injected with a low dose of morphine, is the least while that of the saline injected group (Group VII) is the maximum. The highest morphine dose group i.e. 30 mg/kg (Group VI), also has a high latency score.

From the statistical analysis it is clear that group II, IV and III (3 mg, 20 mg, and 30 mg/kg morphine respectively) are significantly different from group I, V, VI and VII (1 mg, 25 mg, 30 mg/kg morphine and .25 ml NaCl respectively). These differences are significantly at the .01 level. However, none of these groups (II, IV and III; I, V, VI and VII) are significantly different from each other (p .05). This indicates that three of the lower doses (except 1 mg/kg) impair retention while the higher doses and the lowest dose has no effect. The same is graphically depicted in Figure 3.

Table 2 showing the significance of difference between the means of the Morphine and NaCl injected groups on the II, III and IV Retention test by using Duncan's Range Test.
FIG. 6. SHOWING THE MEAN LATENCIES OF THE MORPHINE AND NaCl INJECTED GROUPS ON RETENTION TEST IV (14 DAYS)
FIG. 5. SHOWING THE MEAN LATENCIES OF THE MORPHINE AND NaCl INJECTED GROUPS ON RETENTION TEST III (7 DAYS)
FIG. 4. SHOWING THE MEAN LATENCIES OF THE MORPHINE AND NaCl INJECTED GROUPS ON RETENTION TEST II (2 DAYS)
Table 2

<table>
<thead>
<tr>
<th>Groups</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
</tr>
</thead>
<tbody>
<tr>
<td>Retention</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>II</td>
<td>8.126</td>
<td>9.9</td>
<td>13.75</td>
<td>163.25</td>
<td>178.625</td>
<td>199.375</td>
</tr>
<tr>
<td>III</td>
<td>3.344</td>
<td>6.31</td>
<td>11.81</td>
<td>143.87</td>
<td>165</td>
<td>167.37</td>
</tr>
</tbody>
</table>

Further study of the mean latency scores, (Table 2 and figures - 4, 5 and 6) indicate that group V and VI (M, 25 and 30 mg/kg) are significantly different from the saline injected group (Group VII p .05) on the second retention test. The difference between group I (1 mg/kg Morphine) and saline are significant (p .05) on the last retention test. Other than this the trend is the same as the retention test I.

Thus, on the basis of these results it can be inferred that moderate doses of morphine (2 mg to 20 mg)
disrupt retention. Even higher doses (25 to 30 mg/kg) has an inhibitory effect. These results are in con-tradiction to those of Jensen et al. (1978), Gallagher and Kepp (1978) Mondadori and Waser (1979) and Staubli et al (1980), who reported that Morphine in extreme doses facilitate retention while moderate doses have an inhibitory effect.

Thus our hypotheses which predicted that post-training administration would lead to retention impairment in a dose dependent manner.

However the 2nd hypotheses predicted that low and high doses of morphine enhance retention, an inverted U-shaped relationship would be obtained between varying doses of morphine and retention.

Moderate doses, i.e. from 2 to 20 mg/kg have an inhibitory effect and the effect decreases with subsequent decrease or increase in the dosage of morphine.

The reason why the present investigator failed to observe the facilitatory effects of Morphine could be that doses below 1 mg or above 30 mg/kg were not used.

The investigator had initially planned to vary the dosage of morphine from 1 to 100 mg/kg. However, during the pilot work, it was observed that the animals could not
even tolerate doses as high as 40 mg/kg, therefore the upper limit was brought down to 30 mg/kg. Even, 30 mg/kg of morphine produced behavioural abnormalities in the animals. Three animals, who were injected with this dose expired within 24 hours.

It has been reported although higher doses of morphine are lethal, tolerance can be developed by continuous administration of small amount of Morphine after frequent intervals. The present investigation was a short term project, therefore this technique was not feasible.

Since Morphine is an opiate agonist which mimics the action of the endogeneous peptides, it can be hypothesised that this drug influences memory via the same mechanism as the endogeneous peptides.

Izquierdo and Grandenzi (1980) have suggested that the inhibitory action of Morphine on retention might be due to its inhibitory actions on the noradrenergic synapses. This view is supported by the fact that release of these systems from the inhibitory influence of the endogeneous peptide system by administration of opiate antagonists reverses the induced amnesia.
This postulate can also explain the facilitation of retention observed after administration of extreme doses of morphine. It has been reported by Gold and van Buskirk (1978) that hyper or hypo secretion of NE at the adrenergic synapses results in amnesia, while moderate secretions are positively correlated with good retention.

Since the opiate peptides have an opposite action i.e. facilitation at high and low, and inhibition at moderate levels their memory modulating effects might be due to their effects on the adrenergic synapses.

Alternatively the facilitation of retention observed after administration of higher doses of morphine might be due to its direct action on reinforcement mechanisms by activation of opiate receptors (Mondadori et al., 1979).

White et al. (1978) have demonstrated that morphine facilitates retention of a single trial appetitive learning task. They suggest that this effect is due to the effect of morphine on reinforcement mechanisms.

Sunita and Mohar (1983) showed that morphine impairs retention of an inhibitory avoidance task while naloxone enhances it. They have proved that immediate post-training administration of morphine causes impairment of retention in a dose dependent manner, in a single trial avoidance task.

We may now sum up the findings of the present work. A multi group design was used to investigate the dose dependent effect of morphine on retention. It was
hypothesised that lower and higher doses of Morphine would facilitate retention while moderate doses would cause amnesia. Also an inverted U-shaped relationship would be observed between dosage of Morphine and retention performance. The first hypothesis is not confirmed by the results of the present study since the investigator failed to observe any facilitatory effect of Morphine.

The second hypothesis is verified. These findings provide strong evidence of the involvement of the endogenous opiate peptides in memory modulation.