Many indoles have been investigated in recent years for their effects on central nervous system. Some of them have been demonstrated to modify the analgesic effects of morphine. SIGG et al. (16) have shown that morphine analgesia is potentiated by 5-hydroxytryptamine (5-HT), 5-hydroxytryptophan (5-HTP) and tryptamine. SCHNEIDER and Mc ARTHUR (15) reported potentiating action of a phrenotropic indole alkaloid Ibogaine on morphine analgesia. Reserpine has been variously reported to antagonise (14, 16) and potentiate (22) morphine analgesia. The present study was undertaken to investigate the effects of LSD-25, an important phrenotropic indole on morphine analgesia. Amidone was included to see if a similar interaction occurred with other morphine like analgesics as well.

Methods

The analgesic effect was studied in albino rats (35-55 gm body weight) by the hot plate method of WOOLFE and MCDONALD (23) as modified by EDDY et al. (4). The temperature of hot plate was kept constant at 56.5°C and the first licking of the hind paws was taken as the end point. The data were analysed by the method of EDDY et al. (5). The reaction time was determined immediately before and 20, 40, 60 and 90 minutes after drug administration. Initially reaction time of 150 normal rats was determined at usual post injection time intervals and the results plotted on millimeter graph paper. The standard deviation from average
reaction time in these rats was found to be 123 sq. mm. Subsequently the post injection reaction time area had to differ from its own normal reaction time area by at least twice the standard deviation (246 sq.mm.) before the animal was considered to have developed analgesia.

The animals were divided in batches of ten and each dose was administered to at least one batch. The drugs, morphine tartrate, lysergic acid diethylamide (LSD-25) (*) and amidone (**) were administered subcutaneously, dissolved in distilled water. Whenever a combination of drugs was used the two drugs were administered simultaneously. Various doses of each drug or each combination were tested and at least four dose levels were established between the limits of hundred percent analgesia and no analgesia. The AD50 (dose producing analgesia in 50 per cent animals), AD25 and AD75 along with their fiducial limits were calculated by the method of Bliss (1, 2) and Foster (6).

For studying the effect of LSD-25 on development and duration of morphine analgesia two batches of twenty animals each were employed. One batch was given 15.0 mg/kg morphine alone and another 250.0 µg/kg LSD-25 along with morphine. Both batches were then tested at intervals of fifteen minutes for two hours and percentage of animals showing analgesia at each interval was calculated. They were then tested half hourly till 95 per cent animals showed no analgesia.

RESULTS

LSD-25 alone failed to show any analgesic activity at dose levels of 50, 250, 500, 1000 and 1250 µg/kg respectively. The animals became extremely excited with the bigger doses (1000 and 1250 µg/kg) and it was difficult to judge the end point sharply. Hence these doses were not used for combining with morphine or amidone.

The final regression lines for morphine alone and when combined with 50, 250 and 500 µg/kg of LSD-25 are shown in Fig. 1. These lines were tested for goodness of fit by the X^2 test of Finney (5). The values of X^2 were not found significant at 5 per cent level for any of the lines. The values of AD50, 25 and 75 and their fiducial limits (P = 0.05) are given in table I along with their relative potencies as compared to morphine. It will be noticed that the degree of antagonism becomes more marked at higher dose levels of either morphine or LSD-25.

(*) Courtesy Dr. E. J. Vaz, Sandoz Private Ltd., Bombay.
(**) Polamidon-C (Hoechst) brand. Each one cc. ampoule contains amidone 5.0 mg and diphenyl-piperidino ethylacetamide hydrochloride 0.25 mg.
Showing the final regression lines for morphine alone and when combined with 50, 250 and 500 μg/kg respectively of LSD-25. The points actually obtained during the experiments are also shown. The ED50 values have been marked. Note the marked divergence of regression lines when heavier doses of LSD-25 are employed.

**Table 1**

Table 1 showing AD25, AD50 and AD75 of morphine alone and in combination with 50, 250 and 500 μg/kg LSD-25 respectively along with their range and relative potency.

<table>
<thead>
<tr>
<th>Drug</th>
<th>mg/kg</th>
<th>Range (P 0.05)</th>
<th>Relative Potency</th>
</tr>
</thead>
<tbody>
<tr>
<td>Morphine</td>
<td>AD25</td>
<td>7.63</td>
<td>5.43 ± 0.83</td>
</tr>
<tr>
<td></td>
<td>AD50</td>
<td>10.30</td>
<td>8.58 ± 1.02</td>
</tr>
<tr>
<td></td>
<td>AD75</td>
<td>14.20</td>
<td>10.14 ± 1.06</td>
</tr>
<tr>
<td>LSD 50 μg/kg</td>
<td>AD25</td>
<td>9.40</td>
<td>7.69 ± 1.09</td>
</tr>
<tr>
<td>Morphine</td>
<td>AD50</td>
<td>12.34</td>
<td>10.23 ± 1.45</td>
</tr>
<tr>
<td></td>
<td>AD75</td>
<td>16.03</td>
<td>11.44 ± 1.66</td>
</tr>
<tr>
<td>LSD 250 μg/kg</td>
<td>AD25</td>
<td>8.53</td>
<td>5.23 ± 1.83</td>
</tr>
<tr>
<td>Morphine</td>
<td>AD50</td>
<td>14.36</td>
<td>12.31 ± 1.41</td>
</tr>
<tr>
<td></td>
<td>AD75</td>
<td>22.57</td>
<td>14.91 ± 1.63</td>
</tr>
<tr>
<td>LSD 500 μg/kg</td>
<td>AD25</td>
<td>9.04</td>
<td>5.07 ± 1.24</td>
</tr>
<tr>
<td>Morphine</td>
<td>AD50</td>
<td>15.25</td>
<td>12.45 ± 1.80</td>
</tr>
<tr>
<td></td>
<td>AD75</td>
<td>25.63</td>
<td>16.87 ± 1.99</td>
</tr>
</tbody>
</table>

Morphine = 1
LSD-25 ANTAGONISM OF MORPHINE ANALGESIA

Fig. 2

Showing the induction and duration of analgesia with morphine 15.0 mg/kg alone and in combination with 250 μg/kg LSD-25 respectively. Note the higher percentage of analgesia and longer duration of peak effect in the group receiving morphine alone.

The development and duration of analgesia in the two batches is shown in Fig. 2. There was no difference in the time of onset and duration of analgesia in the two cases. However, the degree of analgesia was much more in the batch where no LSD-25 had been used. The

Fig. 3

Showing final regression lines for amidone and amidone + 250 μg/kg LSD-25. The ED₅₀ values have been marked. The points refer to experimental data obtained.
duration of peak effect also was more marked in the same group. In both groups ninety five per cent animals recovered within three and a half hours.

LSD-25 was combined with amidone to find out if this property of LSD-25 was specific for morphine. The results (Fig. 3 and Table II) show that LSD-25 antagonised the effect of amidone also and that the pattern of antagonism was the same in both cases.

**Table II**

<table>
<thead>
<tr>
<th>Drug</th>
<th>mg/kg</th>
<th>Range (P - 0.05)</th>
<th>Relative Potency</th>
</tr>
</thead>
<tbody>
<tr>
<td>Amidone</td>
<td>AD25</td>
<td>2.36</td>
<td>0.70-4.36</td>
</tr>
<tr>
<td></td>
<td>AD50</td>
<td>4.61</td>
<td>3.01-5.21</td>
</tr>
<tr>
<td></td>
<td>AD75</td>
<td>8.31</td>
<td>5.11-11.31</td>
</tr>
<tr>
<td>LSD 250 µg/kg</td>
<td>AD25</td>
<td>5.10</td>
<td>2.71-7.49</td>
</tr>
<tr>
<td>Amidone</td>
<td>AD50</td>
<td>10.21</td>
<td>6.37-14.05</td>
</tr>
<tr>
<td></td>
<td>AD75</td>
<td>20.42</td>
<td>10.21-24.63</td>
</tr>
</tbody>
</table>

The effect of LSD-25 on the Straub tail reaction (19) was also observed. LSD-25 itself did not produce any tail reaction in a dose of 500 µg/kg. The tail reaction produced by 50 mg/kg of morphine (21) subcutaneously was, however, increased by LSD-25 (250-500 µg/kg).

**Discussion**

Few reports of morphine-LSD-25 interactions are available in literature. Cook and Weidley (3) have investigated the behavioural effects of LSD-25 using the “Conditioned avoidance escape response” (CR) in trained rats. They found that CR-blocking effect of morphine was antagonised by LSD-25 in doses of 100-500 µg/kg. The results of the present study show a marked antagonism of morphine analgesia by LSD-25. The antagonism also extends to other narcotic agents like amidone. The dose response curves obtained (Fig. 1 and 3) suggest that the antagonism is probably not competitive in nature. LSD-25 does not affect the rate of induction of morphine analgesia. The duration
LSD-25 ANTAGONISM OF MORPHINE ANALGESIA

of analgesia also is not much different in the LSD-25 treated group (Fig. 2). These findings suggest that LSD-25 probably does not influence absorption or fate of morphine in the body. It is interesting to note that the Straub's tail effect of morphine — a stimulant effect — is on the contrary increased by LSD-25.

The available data give little clue as to the mechanism of the LSD-25-morphine antagonism. Some workers have linked cholinesterase inhibition by morphine to its analgesic activity (17, 24) and postulate that morphine potentiation by certain drugs is also brought about by this mechanism (15, 18). LSD-25 has been reported to evoke central parasympathetic activity (11, 12). Poloni and Maffezzoni (10) have observed an increase of acetylcholine in the brain of guinea pigs after administration of LSD-25. Thompson et al. (20) and Fried and Antopol (7) have observed an inhibition of cholinesterase activity of plasma and brain by LSD-25. If cholinesterase inhibition by morphine is linked to its analgesic effect, LSD-25 should have potentiated morphine analgesia. On the contrary LSD-25 antagonises the analgesic action of morphine. These findings are more in consonance with those of Young et al. (25) who reported that analgesia of morphine is probably not dependent on anticholinesterase activity.

The antagonism could be ascribed to the central stimulant effects of LSD-25 but many other central stimulants like ibogaine (15), mescaline, methamphetamine (13) and d-amphetamine (8, 16) have been shown to potentiate morphine analgesia.

The LSD-25 antagonism might be related to alteration of 5-HT level of brain. Morphine has been shown to cause some increase in brain 5-HT level (9). Morphine analgesia is potentiated by 5-HT and 5-HTP (16). It is possible that LSD-25 antagonism of morphine analgesia may be linked with its anti 5-HT effect.

SUMMARY

LSD-25 has been found to antagonise the analgesic effect of morphine and amidone in rats. The antagonism is more marked when higher doses of either LSD-25 or analgesics are employed.

The induction or duration of morphine analgesia is not affected by LSD-25.

The Straub's tail reaction to morphine, a stimulant effect, is increased by LSD-25.
References

2. — Bliss, C. I. J. Econ. Entomol., 1935, 28, 646.
15. — Schneider, J. A. and Mc Arthur, M. Experientia, 1956, 12, 323.