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PHYSIOLOGIC EVIDENCE FOR DESCENDING TRYPTAMINERGIC PATHWAYS IN THE SPINAL CORD

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SUMMARY

LSD was more effective in facilitating the flexor reflex in the chronic than in the acute spinal dog. On the other hand, l-tryptophan produced a significant facilitation of the flexor reflex in the acute spinal dog but not in the chronic spinal dog. These data are consistent with the hypothesis that there are long descending tryptaminergic axons which have the capacity of converting tryptophan to tryptamine and releasing it in the vicinity of postsynaptic facilitatory receptors which degenerate in the chronic spinal dog.

Previous studies have demonstrated that the spinal cord of the dog contains tryptamine (1,2) and that the brain releases tryptamine (3). Martin *et al.* (2) found that brain stem spinal cord levels of tryptamine were increased above the level of transection when compared to the intact dog and unchanged below the level of transection. They proposed on the basis of these findings that there were long ascending and descending tryptaminergic axons in the spinal cord. Tryptophan did not facilitate the flexor reflex of the chronic spinal dog (4). However, it did facilitate the C-fiber reflex in the acute spinal cat, and this facilitation was antagonized by alpha methyldopa but not by para chlorophenylalanine, indicating that tryptophan was acting through a tryptaminergic and not a serotonergic process (5). The failure of tryptophan to facilitate the flexor reflex in the chronic spinal dog could be explained by assuming that tryptophan was converted to tryptamine in the severed axons below the level of transection in the acute spinal animal, and that these axons degenerated in the chronic spinal animal with the consequence that tryptophan was not converted to tryptamine in the vicinity of the facilitatory tryptaminergic receptors. To test this hypothesis, saline, l-tryptophan and LSD were administered to the acute and chronic spinal dog and their effects on the flexor reflex were studied. LSD facilitates the flexor reflex of the chronic spinal dog and the C-fiber of the acute spinal cat by acting as a tryptaminergic agonist presumably by mimicking the actions of tryptamine on the tryptaminergic receptor (4,6).

METHODS

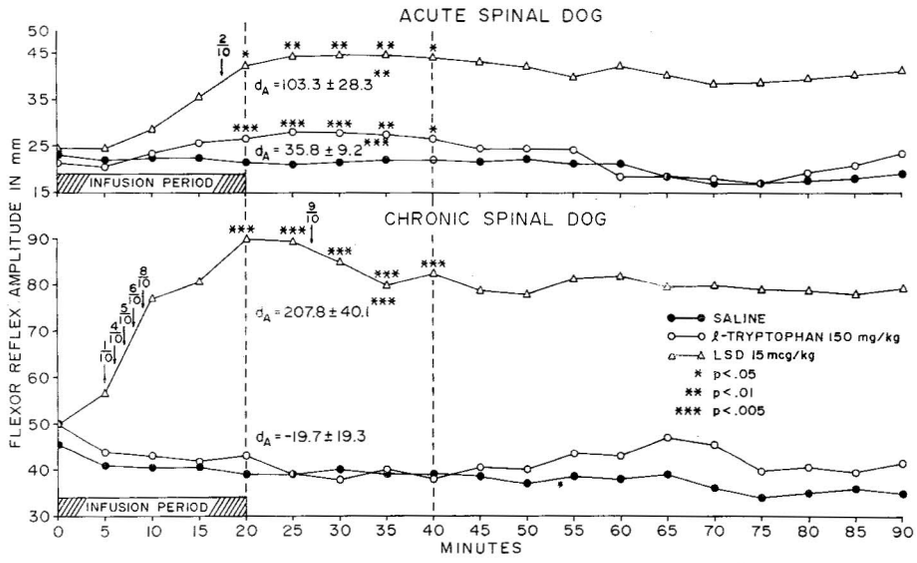
The methods used in these experiments employing the chronic spinal dog have been previously described in detail (7). Beagle type dogs whose spinal cords had been transected at the T-10 level were employed in these studies. On the 2nd through the 5th postoperative day, they received either a 20 minute infusion of saline, l-tryptophan (150 mg/kg) or LSD (15 μg/kg). The flexor reflex was evoked with a pneumatic toe squeezer. A pressure of 60 lb/in^2 was used in 1 dog, 90 lb/in^2 in 5 dogs, and 120 lb/in^2 in 4 dogs during the early part of the experiment. The drugs were administered daily using a Latin square design for the first 9 dogs. These three drug conditions were again repeated in the same dogs three weeks later; however, in the latter study the flexor reflex was evoked with a 60 lb/in^2 stimulus in 9 dogs and 90 lb/in^2 stimulus in 1 dog. In addition, other routine observations including the determination of pulse rate, respiratory rate, pupillary diameter and temperature were made. These latter observations will not be reported in this communication. It will suffice to say that the autonomic signs produced by LSD in both the acute and chronic spinal dogs were the same as have been previously described (4).

RESULTS

As can be seen from Figure 1, despite the fact that nearly twice the stimulus strength was used in the acute as in the chronic spinal dog (a mean

FIG. 1

The effects of l-tryptophan and LSD in the acute and chronic spinal dog



Each point represents the mean of observations made in 10 dogs. The asterisks above each point indicate the level of significance. The area and its standard deviation between the experimental and saline curve for the 20th through the 40th minute were calculated as well as its mean and standard error (d_A). The fractions and arrows indicate the time of onset and the proportion of dog exhibiting fragmentary stepping movements.

of 99 *vs* 55 lb/in) the amplitude of the flexor reflex was greater in the chronic than in the acute spinal dog, indicating the presence of spinal shock in the acute spinal dog.

There was no change in the amplitude of the reflex or its response to LSD or tryptophan across days during the first experimental period. As can be seen, the rate of accommodation of the flexor reflex across time in the saline-treated dogs is somewhat greater in the chronic than the acute spinal dog. Further, tryptophan produced a highly significant increase in the amplitude of the flexor reflex in the acute spinal dog but had no significant effect in the chronic spinal dog. The facilitatory effects of tryptophan became apparent near the end of the infusion and were clearly evident for approximately 40 minutes following the infusion. LSD also produced a significant facilitation of the flexor reflex in both the acute and chronic spinal dog. The facilitation produced by LSD in the chronic spinal dog was over twice that produced in the acute spinal dog even though the stimulus strength in the chronic spinal dog was approximately half that in the acute spinal dog. Despite the increased responsivity of the flexor reflex to both the stimulus and to LSD, l-tryptophan did not facilitate the flexor reflex in the chronic spinal dog. As a matter of fact, during the 20th to the 40th minute following the onset of the infusion there was a modest decrement in amplitude of the flexor reflex. The variance and the amplitude of the flexor reflex in the chronic spinal dog were greater than that observed in the acute spinal dog.

In previous studies it has been shown that LSD also evokes the stepping reflex. In the acute spinal dog, only 2 dogs demonstrated fragmentary stepping which appeared at around the 17th minute of infusion and persisted for about 30 minutes after the end of the infusion. In the chronic spinal dog, LSD evoked more vigorous fragmentary stepping movements in 9 of 10 dogs. In general, as can be seen from Figure 1, fragmentary stepping had an earlier onset in the chronic than in the acute spinal dog. Fragmentary stepping also persisted for a longer time.

DISCUSSION

The major purpose of this study was to reconcile observations made in the acute spinal cat and the chronic spinal dog on the effect of l-tryptophan on nociceptive spinal cord reflexes. Tryptophan facilitates the flexor reflex in the acute spinal dog and the C-fiber reflex in the acute spinal cat. Tryptophan increases the synthesis of both serotonin and tryptamine, both of which are known to have facilitatory effects on spinal cord reflex activity. The mechanism whereby tryptophan facilitates the flexor reflex in the acute spinal dog has not been explored; however, it is probably tryptaminergic because the facilitatory effect of tryptophan on the C-fiber reflex in the acute spinal cat is due to tryptamine and not serotonin since this action is antagonized by alpha methyldopa but not by pCPA. It is relevant that tryptophan does not facilitate the C-fiber reflex in the chronic spinal cat (J. A. Bell, personal communication), a finding analogous to observations made on the flexor reflex in the chronic spinal dog.

The fact that the facilitatory response to the tryptaminergic agonist LSD was enhanced in the chronic as compared to the acute spinal dog would argue that failure of tryptophan to facilitate the flexor reflex in the chronic spinal dog cannot be due to decreased tryptamine receptors or decreased responsivity of the system. An alternative explanation is that nerve fibers and terminals which convert tryptophan to tryptamine and liberate it in the vicinity of the tryptaminergic neurones have degenerated. These observations and interpretations are consistent with the hypothesis that there are long descending tryptaminergic facilitatory pathways in the dog spinal cord (2) and with the observations that

intraventricularly administered 5,6-dihydroxytryptamine decreases spinal cord tryptamine and serotonin levels in the rat (8).

The fact that the responsivity of the flexor reflex and the facilitatory effects of LSD were enhanced in the chronic spinal dog would suggest that denervation supersensitivity to tryptaminergic agonists occurred. Cannon *et al.* (9) first described enhanced afterdischarge of the ipsilateral flexor reflex in chronic hemisected cats. Not only is the flexor reflex more responsive in its own right and to the facilitatory actions of LSD in the chronic than in the acute preparation but the incidence of fragmentary stepping was also greater. LSD (15 µg/kg) usually evokes continuous stepping which persists for several hours in dogs that have been spinal for several months (4).

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