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Effects of Haloperidol, Methyltyrosine and Morphine on Recovery from Lesions of Lateral Hypothalamus[1]

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HYNES, M. D., C. D. ANDERSON, G. GIANUTSOS AND H. LAL. *Effects of haloperidol, methyltyrosine and morphine on recovery from lesions of the lateral hypothalamus.* PHARMAC. BIOCHEM. BEHAV. 3(5) 755–759, 1975. – In rats loss of body weight and lethality were measured after bilateral electrolytic lesions of the lateral hypothalamus. The rats with incomplete lesions initially lost body weight but recovered spontaneously. Treatment with haloperidol (0.8 mg/kg/day) or dl-alpha-methyl-p-tyrosine (100 mg/kg/day) for three days prior to surgery facilitated the recovery in those rats. The rats with more complete lesions lost body weight which resulted in eventual death. Treatment with haloperidol (4 mg/kg/day) or morphine (60 mg/kg/day) given daily for six days before surgery promoted recovery and reduced the incidence of death. The drugs used in this study inhibit activity of brain dopamine receptors to result in the supersensitivity and/or promotion of regenerative processes. These effects are considered to be related to the facilitation of recovery from nerve injury.

Lateral hypothalamus Nigrostriatal fibers Brain lesions Haloperidol Alphy-methyl-tyrosine Morphine Weight Loss Lethality

BILATERAL electolytic lesions of the far lateral areas of the lateral hypothalamus produce a severe aphagia and adipsia [24, 34]. The lesioned animals starve to death in spite of free access to ample food and water. However, when animals which have received lateral hypothalamic lesions are force-fed they gradually recover from the deficit of their food ingestive behavior [33].

The basis of recovery after lateral hypothalamic lesions is not known despite much investigation. Several workers proposed supersensitivity of noradrenergic reward systems to be the basis of recovery [3, 4, 13]. Their experiments utilized intraventricular injections of norepinephrine [3] and nerve growth factor [4] or systemic administration of alpha-methyl-p-tyrosine [13], an inhibitor of catecholamine synthesis [30]. More recent studies based upon the damage to histochemically localized dopaminergic fibers which pass through the area of lesion and the resulting depletion of striatal dopamine in preference to other amines [24, 34, 35] suggest that destruction of the nigrostriatal dopamine system is the basis of aphagia and adipsia.

Most previous studies lack specificity in that they often employed chemical agents which inhibit synthesis of all catecholamines. Use of the synthesis inhibitor not only produced inhibition of synthesis of both norepinephrine and dopamine, it did not permit distinction between deficit in the transmitter release-process [5] and the blockade of postsynaptic receptors due to transmitter depletion.

In the present study we lesioned the lateral hypothalamus while employing drugs known to decrease dopamine receptor activity through direct and indirect mechanisms. Through this approach we observed that lesioning of the nigrostriatal pathway is critical to the production of aphagia and adipsia and that dopamine receptor activity is critical for recovery.

METHOD

Animals

We employed the Long-Evans strain of male hooded rats which were housed individually and maintained on ad lib food and water in a room with a 12 hr light-dark cycle. Drugs were administered intraperitoneally twice a day at 8 a.m. and 8 p.m. in equal doses for either a 3 or 6 day period with the last injection being given 24 hr prior to surgery.

Procedure

Under ether anesthesia, electrolytic lesions were made with anodal current from a monopolar stainless steel electrode with a non-insulated tip. With the dorsal cranium horizontal, the coordinates were 1.5 mm posterior to bregma, 2.0 mm lateral, and 8.5 mm subcranial. Following

the lesioning the rats were housed as before and given free access to dry food and water. Force feeding was not attempted. The rats were weighed daily.

A number of animals from the various drug treated and saline control groups were used to evaluate the site of lesioning. After each brain was embedded in paraffin, sections were taken from the area containing the lesion and were processed according to the method of Luna [20] for eosin staining.

The dopamine concentration of the corpus striatum was determined spectroflourometrically. The animals were food deprived for 18 to 24 hr and were then sacrificed by decapitation. The corpus striatum was rapidly dissected and homogenized in 0.4 N perchloric acid. The homogenized samples were centrifuged at 10,000 RPM for 10 min. The pH of the supernatant was then adjusted to 8.5. All manipulations were carried out in ice chilled containers. The dopamine concentration was determined spectroflouro-metrically following cation exchange chromotography and oxidation [8].

RESULTS

The typical localization of the 1 mA, 15 sec lesion employed in these experiments is shown in Fig. 1. Increasing the current and duration to 2 mA for 30 sec produced a greater destruction of brain tissue as shown in Fig. 2. Examination of these sections showed that the lesion was localized in the area of the lateral hypothalamus, with destruction of the nigro-striatal pathway and portions of the medial fore brain bundle.

Twenty-four hours following the 1 mA, 15 sec lesion there was an increase in the dopamine level in the corpus striatum. At both 4 and 7 days following this lesion, corpus striatal dopamine levels were significantly decreased. The decrease in dopamine levels was greatest on the seventh post lesion day.

Striatal dopamine levels were seen to significantly increase ($p<0.05$), when more extensive lesions, 2 mA for 30 sec were made 24 hr prior to measurement. The concentration of dopamine in the corpus striatum 4 days after lesioning was dramatically and significantly decreased ($p<0.05$). These results were summarized in Fig. 3.

Three groups of animals in which a current of 1 mA for 15 sec was used to make partial lesions lost body weight as expected (Fig. 4). On the fifth postoperative day these lesioned animals began to gain weight. These results are in agreement with those of previous investigators [24, 34, 35]. Pretreatment for 3 days with either dl-alpha-methyl-para-tyrosine (100 mg/kg/day) or haloperidol (0.8 mg/kg/day) promoted the recovery from the effects of the lesions. In these drug-treated animals the extent of total weight loss was diminished and the process of recovery began earlier than in the control group.

An analysis of variance for repeated measures [39] on Days 2 through 7 comparing the haloperidol with the control condition indicated a significant ($p<0.05$) difference.

Where a current of 2 mA for 30 sec was used to make more complete lesions, the resulting aphagia and adipsia was more severe and was irreversible for the control group. All of the rats in the control group died within 2 weeks of the lesion, nearly half of them in less than 8 days. In the group receiving drug pretreatments a significant percentage of the subjects were still alive at fifteen days when they were sacrificed for histological examination (Fig. 5).

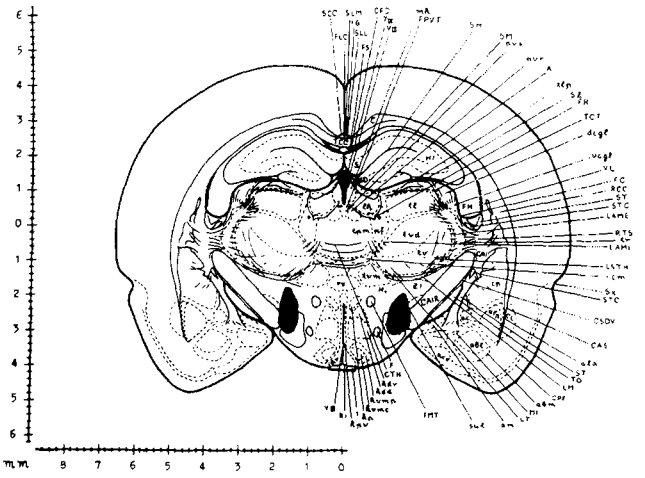

FIG.1. Diagramatic representation of a small (1 mA 15 sec) nigro-striatal lesion. Figure was taken from the König and Klippel atlas [19].

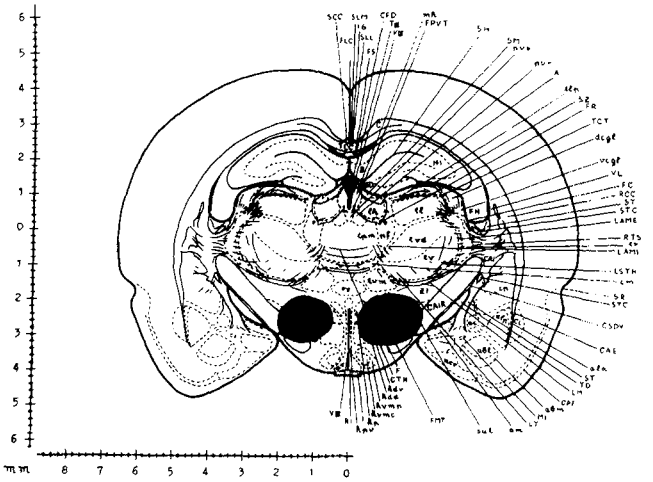

FIG. 2. Diagramatic representation of a large (2 mA 30 sec) nigro-striatal lesion superimposed on a figure from the König and Klippel atlas [19].

Although all groups began to lose weight at the same time, the weight loss in the control group was irreversible (Fig. 6). The weight loss in drug treated groups was similar to that of the control group for the first 6 days. But following this initial weight loss, the groups treated with haloperidol (4 mg/kg/day) or morphine sulfate (60 mg/kg/day) for 6 days prior to surgery, began to recover until sufficient weight was gained to insure survival. Beginning on the ninth postoperative day the weight of the drug treated animals was significantly higher than the weight of the controls ($p<0.05$). The haloperidol treated rats gained more weight than morphine treated rats, but this difference did not reach statistical significance.

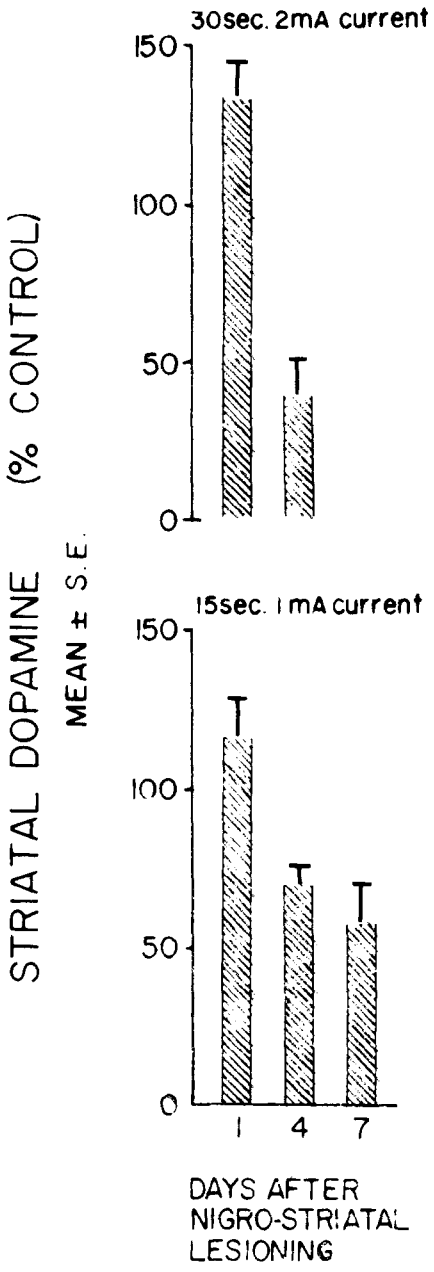

FIG. 3. Striatal dopamine levels at various time intervals following bilateral lesions of the nigro-striatal bundle. Dopamine concentrations are expressed as percent of non-lesioned controls (7.83 ± 0.44 μg/g, N = 17). Dopamine levels were measured at 1 (N = 9), 4 (N = 11) and 7 (N = 8) days following 15 sec 1 mA current lesions and at 1 (N = 8) and 4 (N = 8) days after 30 sec 2 mA current lesions. The animals with the larger lesions did not survive long enough to permit 7 day determination.

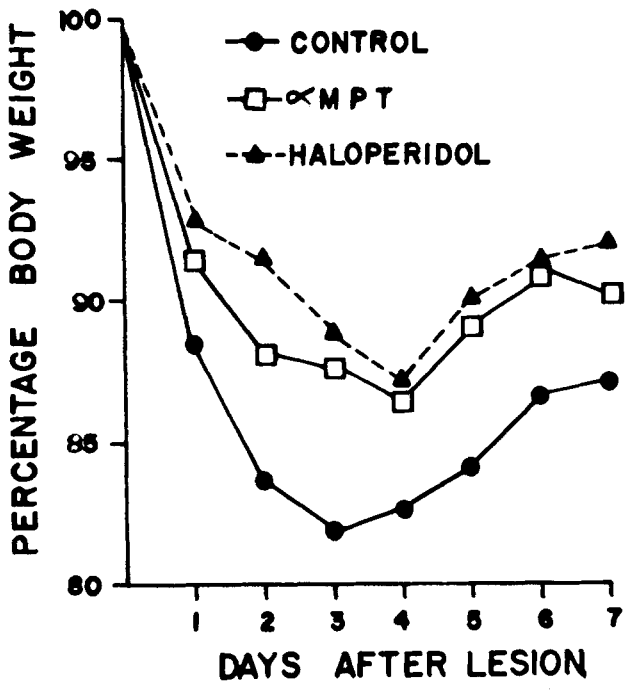

FIG. 4. Loss of body weight expressed as percent of presurgery weight (275 – 325 g) after small (1 mA 15 sec) lesions of the nigro-striatal bundle. Prior to surgery the rats were treated with either saline (N = 8), haloperidol (N = 8) or alpha-methyl-para-tyrosine (N = 5).

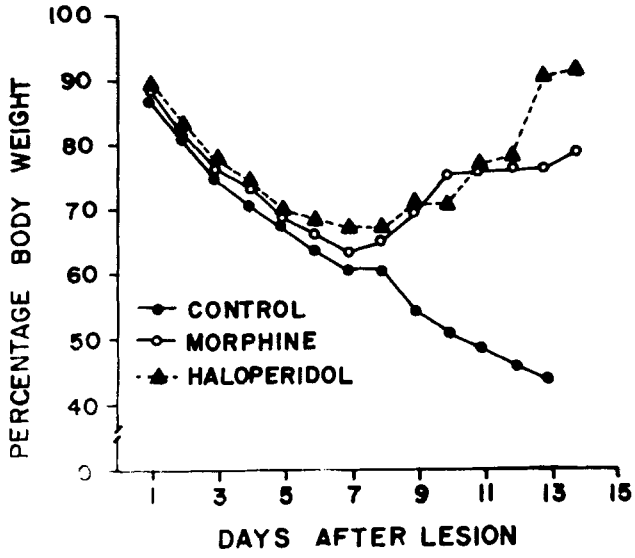

FIG. 5. Loss of body weight expressed as percentage of the presurgery weight (275 – 325 g) after large (2 mA 30 sec) lesions of the nigro-striatal bundle. Prior to surgery the rats were treated with either saline (N = 11),haloperidol (N = 28) or morphine sulfate (N = 14).

DISCUSSION

The histological examination of the lesion sites and the decrease in corpus striatal dopamine produced by the lesion indicates that the extent of weight loss was dependent upon the amount of nigro-striatal damage. Several prev ious studies have suggested that the critical areas for producing weight loss may be outside or include only a far lateral segment of the lateral hypothalamus [14, 15, 22, 38]. These critical areas include portions of the globus pallidus, the medial portion of the internal capsule, and a portion of the lateral hypothalamus adjacent to the internal capsule. Fibers of the nigrostriatal dopamine neurons system

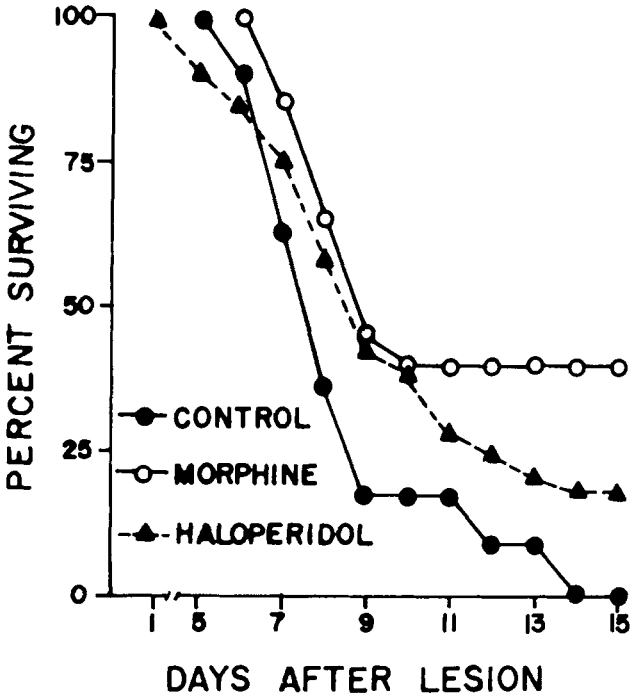

FIG. 6. Surviving rats at different days after large (2 mA 30 sec) lesions of the nigro-striatal bundle. For treatments see legion of Fig. 5.

are known to pass through areas described as critical for producing lesion induce weight loss. Our results thus support previous suggestions [24, 34, 35] that the destruction of the nigro-striatal pathway is critical for the production of the lateral hypothalamic syndrome.

In rats with partial lesions, prior treatment with dl-alpha-methyl-para-tyrosine or haloperidol, markedly facilitated recovery. More complete destruction of these nigrofugal fibers produced continuous loss of weight resulting in death. However, where lesioning was performed after pretreatment with haloperidol or morphine sulfate, a significant recovery took place after an initial period of weight loss.

While we cannot as yet propose a definite mechanism underlying recovery from the near lethal consequences of the destruction of the nigrofugal nerve tracts, several suggesions are worth considering, first an increase in sensitivity of postsynaptic receptors [7, 13, 31], secondly the regenerative axon sprouting of transected axons, [18, 23], third and lastly is an increase in catecholamine turnover in surviving neurons [1, 37]. Any or all of these processes are likely to promote recovery. All the drugs found active in promoting recovery have one property in common, that is, they produce dopamine deficiency at receptor sites. Alpha-methyl-p-tyrosine is a well known inhibitor of catecholamine synthesis [30], haloperidol blocks dopamine receptors directly [2, 17, 40] and morphine inhibits the activity of dopamine receptors possibly indirectly through an effect on nondopaminergic mechanism involved in regulating the activity of dopaminergic systems [9, 25, 27, 28]. The other pharmacological effects of the drugs employed in this study are not common to all three drugs.

A drug induced deficiency of receptor activity can cause both an increase in receptor sensitivity as a consequence of pharmacological denervation [29] and stimulate neuronal feedback mechanisms [2, 25]. Chronic treatment with haloperidol [10,12], methyl-p-tyrosine [21,32] and morphine sulfate [11,26] is known to produce supersensitivity of dopamine receptors in the central nervous system. Both haloperidol and morphine sulfate increase dopamine synthesis in the nigro-striatal system [16, 17, 25, 27, 28].

The pretreatment with these drugs, that increase neurotransmitter synthesis, could promote the development of a compensatory change in the level of activity of the surviving dopamine containing neurons [1, 6, 37]. The stimulation of neurotransmitter synthesis could possibly accompany stimulation of several other processes which play a part in the regenerative process. Which of the suggested effects of these drugs under consideration actually account for the observed promotion of recovery after nerve injury needs further investigation.

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