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D-AMPHETAMINE ADMINISTRATION REDUCES SUBSTANCE P CONCENTRATION IN THE RAT STRIATUM

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Substance P, a peptide containing eleven amino acids,[1] may function as a neurotransmitter in the peripheral and central nervous systems. High concentrations of immunoreactive substance P have been reported within the dorsal horn of the spinal cord[2] and in certain tracts in the brain, including a striato-nigral pathway[3,4] and a habenulo-interpeduncular tract.[5,6]

Systemic administration of the psychotropic drug, d-amphetamine, causes numerous behavioral and physiological changes, including increased motor activity,[7] anorexia,[8] stereotypy[9] and impaired thermoregulation.[10] The mode of action of d-amphetamine has been shown to involve increases in catecholaminergic transmission, caused by accelerated release of dopamine and norepinephrine, and decreased inactivation of these compounds.[11-13] Some effects of d-amphetamine have been specifically associated with the activation of dopaminergic synapses in the basal ganglia[14]--an area containing perikarya of substance P neurons.[4] Like l-dopa and 5-hydroxytryptophan, d-amphetamine in large doses can cause a transient disaggregation of brain polysomes and a partial suppression of protein synthesis.[15-17] The mechanism responsible for these effects probably involves stimulation of dopamine receptors, since dopamine receptor antagonists reverse the amphetamine-induced polysome disaggregation.[15] We now report that administration of high doses of d-amphetamine reduces the level of immunoreactive substance P in the rat striatum.

Male Sprague-Dawley rats (150-170 g; Charles River Laboratories, Wilmington, MA) were housed under a 12:12 hr light-dark cycle and given free access to food and water for several days prior to experimentation. Animals received d-amphetamine (10 mg/kg) or its diluent (0.9% saline) intraperito-

neally. In some studies, rats were pretreated with haloperidol, a dopamine receptor antagonist[18] (10 mg/kg, in 50 mM citric acid) 30 min before the d-amphetamine injection. All experiments were carried out under conditions favorable for the disaggregation of brain polysomes,[15,19] i.e. the animals were given high doses of d-amphetamine and maintained in a warm environment (26-28°) in order to produce hyperthermia (typically, a 2° increment in rectal temperature 2 hr post-injection).

Animals were killed by decaptitation at various times after d-amphetamine administration. Brain regions were rapidly dissected, homogenized in 2 N acetic acid and centrifuged, and the supernatant fluid was lyophilized. Samples were resuspended and assayed for substance P by radioimmunoassay.[20]

Two hr after administration of d-amphetamine, the concentration of immunoreactive substance P in the striatum was reduced by 20-35% (Table 1, Fig. 1); this effect was blocked by prior administration of haloperidol (Table 1). The decline in striatal substance P levels first became evident 1 hr post-injection, and persisted through hr 4 (Fig. 1). In the substantia nigra, substance P levels were slightly reduced 2-8 hr after d-amphetamine, but never significantly.

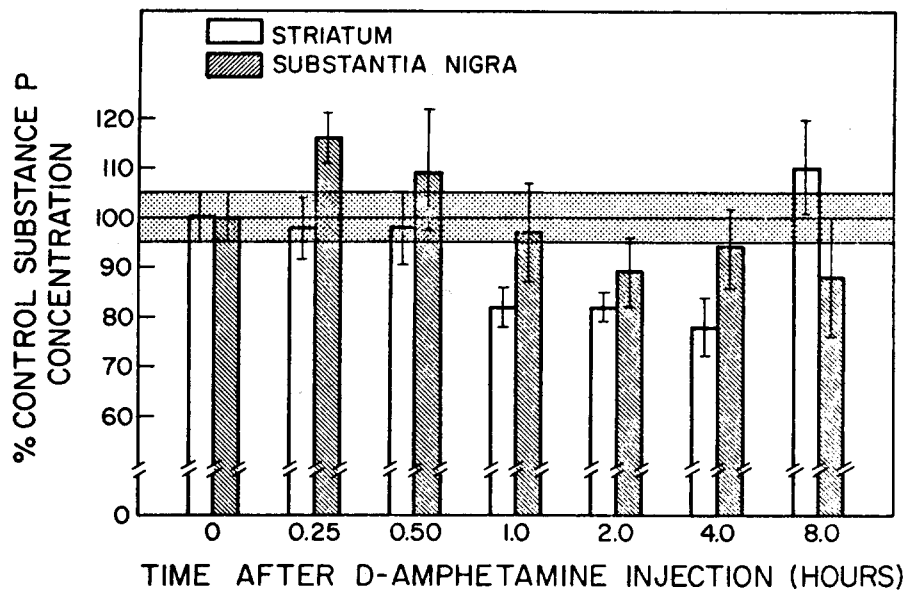

Fig. 1. Time-related changes in substance P concentration of the rat striatum and substantia nigra after d-amphetamine administration. Rats were injected with d-amphetamine (10 mg/kg, i.p.) or its diluent (0.9% saline); animals were maintained in a warm environment (26°) and killed at various times post-injection. Data are given as per cent control levels ± S. E. M. Control concentrations of substance P were: substantia nigra, 5.77 ± 0.26 pmoles/mg of protein; striatum, 1.01 ± 0.05 pmoles/10 mg of tissue. Recovery of substance P added to homogenates of both tissues is quantitative. Each group contained six to twelve rats. For the striatum, the 4-hr post-injection group differs from control (0 hr) and 8-hr groups ($P < 0.05$) (Newman-Keuls comparison test after one-way analysis of variance). No differences were observed between groups in the substantia nigra (one-way analysis of variance).

Table 1. Blockade of d-amphetamine-induced reduction of substance P concentration in the rat striatum.*

Treatment	Substance P (pmoles/10 mg tissue)
Control	1.62 ± 0.16
Haloperidol	1.40 ± 0.09
D-amphetamine	1.09 ± 0.03†
Haloperidol + d-amphetamine	1.48 ± 0.11

*Sprague-Dawley rats weighing 150-160 g received haloperidol (10 mg/kg, i.p.) or its vehicle (50 mM citric acid) and, after 30 min, d-amphetamine (10 mg/kg, i.p.) or its vehicle (0.9% saline); animals were maintained in an environment of 28°, and killed 2 hr after the second injection. Data are given as group means ± S. E. M. Each group contained four to six rats.

†Differs from control group ($P < 0.05$) (Newman-Keuls comparison test after one-way analysis of variance).

The reduction in striatal substance P levels presumably reflects processes within perikarya of substance P neurons.[4] These could involve reduced synthesis of substance P or a precursor peptide (i.e. a possible consequence of polysome disaggregation[15]), accelerated degradation of the peptide, or enhanced axoplasmic flow. That diminished substance P synthesis is involved in the effect of d-amphetamine on immunoreactive substance P concentrations in the striatum is suggested by: (1) the locus of the reduction, i.e. initially within cell bodies but not nerve terminals, and (2) the similar time courses of d-amphetamine-induced polysome disaggregation[15] and the reduction in striatal substance P concentrations. D-amphetamine could affect the synthesis of substance P or a precursor compound via a pre-synaptic action (increasing dopamine release) or by directly affecting the protein-synthetic apparatus.[15,16,21]

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