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Short communication

A novel neurotensin analog blocks cocaine- and D-amphetamine-induced hyperactivity

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Abstract

Neurotensin is a tridecapeptide that exhibits selective anatomic and neurochemical interactions with dopaminergic systems. Since dopaminergic neurotransmission underlies many of the behavioral properties of psychostimulants, and since neurotensin has been implicated in modulating dopaminergic neurotransmitter systems, we tested the effect of our novel neurotensin analog, NT69L (*N*-methyl-Arg^{8},L-Lys^{9},L-*neo*-Trp^{11},tert-Leu^{12}]neurotensin-(8-13)), on hyperactivity caused by cocaine and D-amphetamine. Previously, we showed that NT69L reduces body temperature, blocks apomorphine-induced climbing, and haloperidol-induced catalepsy. In this study, NT69L blocked the hyperactivity induced by both cocaine and D-amphetamine administered at three different doses each, when this peptide was injected intraperitoneally. These results provide further evidence for the involvement of the neurotensin system in some of the behavioral properties of psychostimulants and suggest that NT69L may find clinical application in patients who abuse this class of compounds.

Keywords: Neurotensin; Cocaine; D-Amphetamine; Hyperactivity

1. Introduction

The involvement of the dopaminergic system in the behavioral effects of the psychostimulants, cocaine and D-amphetamine, has been intensively investigated (Kleven et al., 1990; Ng et al., 1991; Peris et al., 1990). However, more attention has been recently directed to the involvement of neuropeptide systems as well. Neurotensin is such a neuropeptide that has intrigued many researchers in this field because it exhibits a number of well-documented interactions with dopaminergic systems (Nemeroff, 1986; Nemeroff et al., 1982), and is implicated in mediating some of the behavioral effects of cocaine (Betancur et al., 1998). Intraventricular administration of neurotensin in rats attenuates the locomotor hyperactivity induced by a single injection of cocaine or D-amphetamine (Ervin et al., 1981; Nemeroff, 1986; Nemeroff et al., 1983). Also, chronic cocaine treatment modifies neurotensin binding in rat brain in regions associated with dopaminergic pathways (Pilotte et al., 1991) and increases concentrations of neurotensin-like material in the striatum and substania nigra (Hanson et al., 1989). However, after a single injection of cocaine, neurotensin-like material increased only in the substania nigra (Hanson et al., 1989).

In view of the evidence implicating the neurotensin system in the neurochemical response to psychostimulants, we evaluated the effect of our neurotensin analog, NT69L (*N*-methyl-Arg8,L-Lys9,L-*neo*-Trp11,tert-Leu12]neurotensin-(8-13)), administered peripherally, due to its ability to cross the blood–brain barrier in blocking the hyperactivity induced by injection of cocaine or D-amphetamine in the rat.

2. Materials and methods

2.1. Animals

Male Sprague–Dawley rats weighing 150–250 g were used for all experiments. Rats were housed in a temperature-controlled room with free access to food and water.

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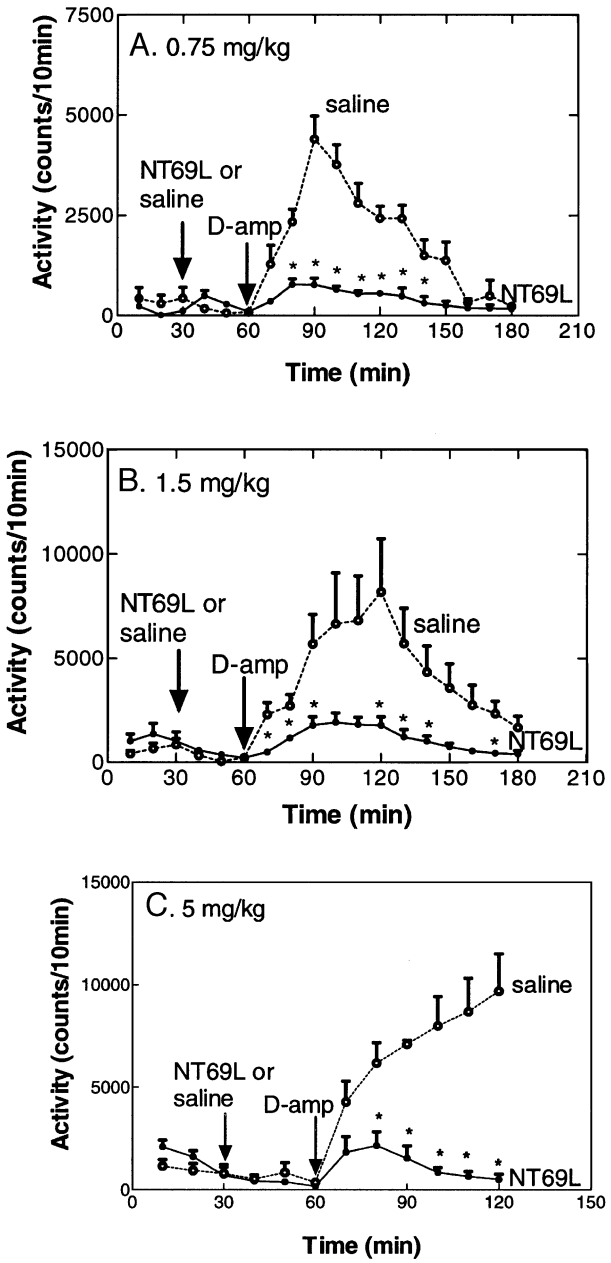

Fig. 1. Effect of a single injection of NT69L on blocking D-amphetamine-induced hyperactivity at 0.75 (A), 1.5 (B), and 5 mg/kg (C). Each Sprague–Dawley rat was placed in an activity chamber for 1 h for acclimation, removed for injection with NT69L (1 mg/kg) or saline i.p., and then placed back in the activity chamber. After obtaining a 30-min baseline, each rat was removed from the chamber, injected with D-amphetamine i.p., and returned to the chamber. Activity was recorded for the times indicated. (*) Significantly different ($P < 0.05$) from saline/D-amphetamine.

The animals were kept on a 12-h light/dark cycle. All tests were performed during the light cycle. All procedures were approved by the Mayo Foundation Institutional Animal Use and Care Committee. The treated groups were injected intraperitoneally (i.p.) with the indicated compounds, while the control groups were injected with an equal volume of saline (0.9% NaCl).

2.2. D-amphetamine-induced hyperactivity

Following an acclimation period of 1 h in a plexiglass Opto-Varimex Minor motility chamber (Columbus Instru-

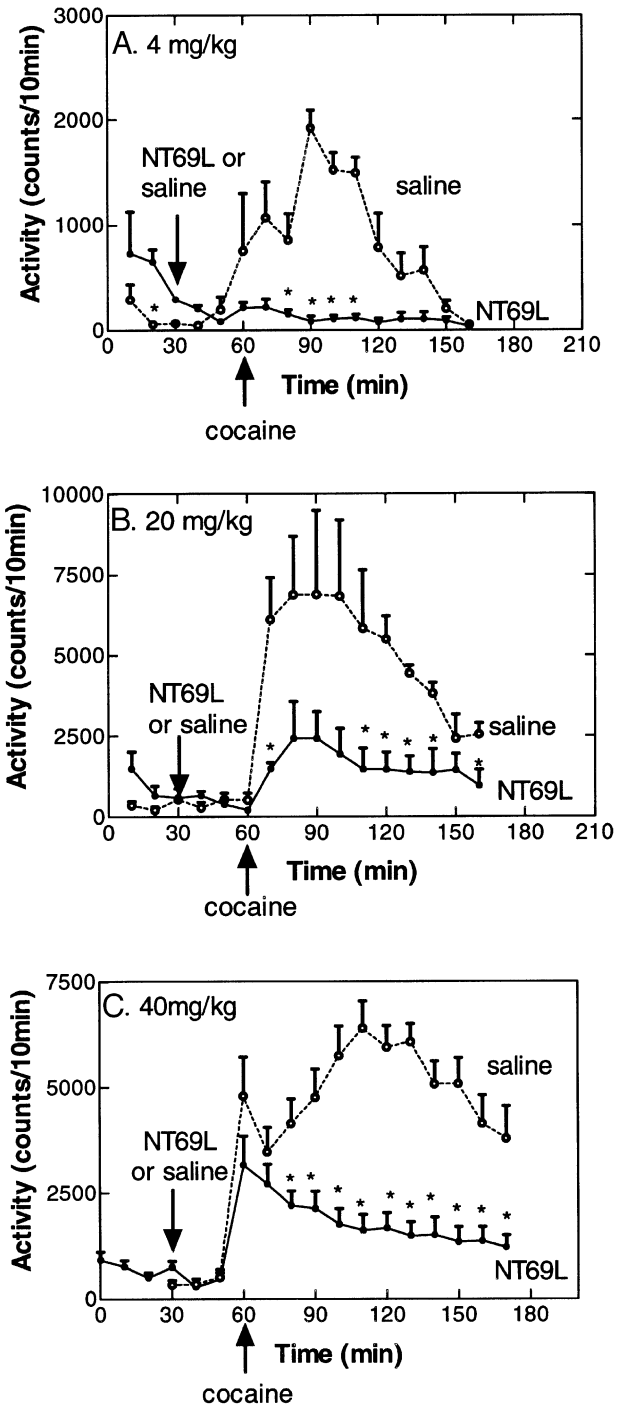

Fig. 2. Effect of single injection of NT69L on blocking cocaine-induced hyperactivity at 4 (A), 20 (B), and 40 mg/kg (C). Each Sprague–Dawley rat was placed in an activity chamber for 1 h for acclimation, removed for injection with NT69L (1 mg/kg) or saline i.p., and then placed back in the activity chamber. After obtaining a 30-min baseline, each rat was removed from the chamber, injected with cocaine i.p., and returned to the chamber. Activity was recorded for 2 h. (*) Significantly different ($P < 0.05$) from saline/cocaine.

ments, Columbus, OH), the rats were injected with NT69L (1 mg/kg) or saline and placed back in the activity chamber for 30 min to obtain a baseline, after which they were injected with D-amphetamine at 0.75, 1.5, or 5 mg/kg i.p. Activity was recorded for 1 h.

2.3. Cocaine-induced hyperactivity

The rats were acclimated for 1 h in the activity chamber, injected with NT69L (1 mg/kg) or saline, followed by placement in the activity chamber for 30 min, after which time each animal was injected with cocaine at 4, 20, or 40 mg/kg i.p. Over the next 2 h, activity was measured.

2.4. Statistical analysis

Statistical analysis was carried out by one-way analysis of variance (ANOVA) followed by Tukey's test for multiple comparisons, using Sigma Stat software, with $P < 0.05$ being considered significant.

3. Results

3.1. D-amphetamine-induced activity

Administration of NT69L (1 mg/kg) blocked the hyperactivity caused by amphetamine at doses 0.75 mg/kg (Fig. 1A), 1.5 mg/kg (Fig. 1B), and 5 mg/kg (Fig. 1C).

3.2. Cocaine-induced activity

Acute administration of NT69L (1 mg/kg) resulted in blocking cocaine- (4, 20, and 40 mg/kg) induced hyperactivity, (Fig. 2A, B, and C, respectively).

4. Discussion

The behavioral effects of cocaine have long been associated with alteration in the dopamine system (Johanson and Fischman, 1989). The interaction of dopamine with the neuropeptide neurotensin prompted researchers to test the hypothesis that neurotensin plays an important role in the behavioral properties of psychostimulants. Centrally administered neurotensin attenuates the locomotor hyperactivity induced by D-amphetamine and cocaine in rats (Nemeroff et al., 1983). In addition, central nervous system stimulants such as cocaine and D-amphetamine substantially impact neurotensin in different brain regions (Cain et al., 1993). In this study, we tested a novel neurotensin analog (called NT69L or [*N*-methyl-Arg^{8},L-Lys^{9},L-*neo*-Trp^{11},tert-Leu^{12}]neurotensin-(8-13)) that our laboratory has developed. Unlike neurotensin, NT69L crosses the blood–brain barrier, and retains all the classic properties of neurotensin, such as hypothermia and antinociception (Tyler et al., 1999). It also blocks haloperidol-induced catalepsy and apomorphine-induced climbing (Cusack et al., 2000), and reduces food intake in Sprague–Dawley and obese Zucker rats (Boules et al., 2000). In addition, NT69L increases dopamine and serotonin levels in brain homogenates (Boules et al., 2000) and increases the dopamine turnover rate in freely moving animals as measured by microdialysis (Warrington et al., manuscript submitted for publication). In the present work, we showed that NT69L attenuated the hyperactivity caused by injection of cocaine or D-amphetamine at three different doses (0.75, 1.5, and 5 mg/kg for D-amphetamine, and 4, 20, and 40 mg/kg for cocaine. The results are in agreement with those reported for neurotensin directly injected into the brain (Nemeroff et al., 1983). The exact mechanism by which neurotensin exerts its effects on the behavioral properties of psychostimulants is not totally elucidated. Neurotensin could be acting presynaptically to counteract increased dopamine in the synaptic cleft (Nemeroff et al., 1983) or it could have postsynaptic inhibitory influences on dopaminergic systems (Nemeroff, 1986; Quirion et al., 1982), since it decreases behaviors induced by pharmacological stimulation of dopamine receptors (Ervin et al., 1981; Jolicoeur et al., 1983). On the other hand, neurotensin can attenuate behavioral manifestations induced by hypo-dopaminergic conditions (Jolicoeur et al., 1991) and can markedly inhibit behaviors produced by small doses of a dopamine agonist that are thought to activate the inhibitory dopamine autoreceptors (Jolicoeur et al., 1985). With NT69L being systemically administered and potentially reaching all brain areas, it can affect the dopaminergic system by interacting with other neurotransmitter systems (Chapman and See, 1996; Ferraro et al., 1998; Kasckow and Nemeroff, 1991; Kinkead et al., 1999). It can also be involved in the formation of "hybrid" receptors by association with dopamine receptors, a mechanism that will increase the diversity of cellular responses to extracellular signals, as is the case with insulin and growth factor receptors (Jacobs and Moxham, 1991). Nonetheless, our results provide further evidence for the involvement of neurotensin in the behavioral and/or addictive properties of psychostimulants. They indicate that neurotensin plays an important role in psychostimulant abuse-related phenomena and suggest that NT69L may be useful in the treatment of cocaine abuse and other psychostimulant drug dependencies.

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