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The locomotor effects of MK801 in the nucleus accumbens of developing and adult rats

Kyle Frantz *, Carol Van Hartesveldt

Department of Psychology, University of Florida, P.O. Box 112250, Gainesville, FL, 32611-2250, USA

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Abstract

This developmental study was an investigation of locomotion induced by the NMDA receptor antagonist, (+)MK-801 hydrogen maleate [(5*R*,10*S*)-(+)-5-methyl-10,11-dihydro-5*H*-dibenzo[*a*,*d*]cyclohepten-5,10-imine], at doses of 0, 3 or 10 μg injected bilaterally into the nucleus accumbens of rats at 11, 21, 31, or 61–66 days of age. During a 2-h test session, only a few 11-day-old pups responded to either dose of MK801; they displayed short bouts of obstinate progression. In contrast, 21- and 31-day-olds were not affected by 3 μg MK801 but exhibited robust activation after 10 μg MK801. The activation was greatest in 21-day-olds and also occurred after mid-striatal injections in 21- but not 31-day-old rats. Adult rats injected with MK801 were not robustly activated, but they maintained their initial level of activity throughout the test session, instead of habituating to the test monitor, as controls did. Ontological changes in MK801-induced activity are likely to reflect maturation of glutamate transmission in the nucleus accumbens.

Keywords: Dizocilpine; Ontogeny; Novel environment; Dopamine; Glutamate

1. Introduction

The nucleus accumbens is a major site of limbic–motor interface in the brain due to its integral role in locomotion, reward-related behaviors and responses to stress (Mogenson et al., 1993). Major inputs to the nucleus accumbens include glutamate and dopamine (Heimer et al., 1993, for review). Whereas dopamine has long been recognized as a major modulator of locomotion, reward-related behaviors and responses to stress (LeMoal and Simon, 1991; Salamone, 1994, 1996; for reviews), an important role for glutamate in such behaviors has only recently been confirmed (Kelley and Throne, 1992; Willner et al., 1992; Moghaddam, 1993; Burns et al., 1994; Horger and Roth, 1995; Carlezon and Wise, 1996). The behavioral functions of glutamate in the nucleus accumbens deserve further investigation.

Nucleus accumbens glutamate modulates locomotion. Locomotor behavior can result from direct intra-accumbens injections of either agonists or antagonists at the *N*-methyl-D-aspartate (NMDA) receptor subtype (e.g., Donzanti and Uretsky, 1983; Hamilton et al., 1986; Boldry and Uretsky, 1988; Svensson and Carlsson, 1992; Wu et al., 1993b; Burns et al., 1994; Svensson et al., 1994; Al-Khatib et al., 1995) or other glutamate receptor subtypes (e.g., Hamilton et al., 1986; Boldry et al., 1991; Wu et al., 1993b; Burns et al., 1994). Systemically-administered NMDA receptor antagonists, such as MK801 (dizocilpine), produce an inverse U-shaped dose–response curve with respect to locomotion in adult rats (Carlsson and Carlsson, 1989; Ford et al., 1989; Hargreaves and Cain, 1992; Willins et al., 1993) as well as developing rats (Rajachandran et al., 1991; Scalzo and Burge, 1994). In adult rats, the stimulatory aspects of the response can be eliminated by pretreatment of the nucleus accumbens with antagonists at other glutamate receptor subtypes (Willins et al., 1993). Furthermore, classic motor stimulants, such as peripherally-injected cocaine (Pulvirenti et al., 1991) or intra-accumbens-injected amphetamine (Kelley and Throne, 1992; Burns et al., 1994) or dopamine (Hamilton et al., 1986) produce locomotion that can also be blocked by pretreatment of the nucleus accumbens with an NMDA receptor antagonist. The locomotor effects of glutamate in the nucleus accumbens are obviously complex, given that both agonists and antagonists at glutamate receptors can

* Corresponding author. Tel.: +1-619-784-7233; Fax: +1-619-784-7405; E-mail: kfrantz@sage.scripps.edu

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increase locomotion and that the activity they elicit can be blocked by other glutamatergic drugs (Hamilton et al., 1986).

The complexity of glutamate's role in locomotion may be due in part to interactions between glutamate and dopamine. The stimulatory effects of peripherally-injected MK801 are attenuated when dopamine transmission in the nucleus accumbens is compromised by either dopamine receptor antagonist injection (Willins et al., 1993; Ouagazzal et al., 1994) or dopamine depletion (Carlsson and Carlsson, 1989; Carlsson and Svensson, 1990; Svensson and Carlsson, 1992; Svensson et al., 1992a,b; Criswell et al., 1993; Starr and Starr, 1993; Ferré et al., 1994; Ouagazzal et al., 1994; Starr and Starr, 1994; Svensson et al., 1994). Studies in which dopaminergic and glutamatergic drugs are either injected directly into the nucleus accumbens (Boldry and Uretsky, 1988; Wu et al., 1993b) or peripherally (e.g., Hoffman, 1992; Ögren and Goldstein, 1994; Dall-'Olio et al., 1996) also confirm that these neurotransmitters interact to alter locomotion.

The locomotor effects of both glutamate and dopamine in the nucleus accumbens vary with the novelty of the testing environment. Generally, novelty in the external environment induces locomotor exploration followed by habituation in adult rats (O'Keefe and Nadel, 1978). Intra-accumbens injections of a glutamate receptor antagonist decrease locomotion that is induced by the presence of novel objects (Mogenson and Nielsen, 1984; Bradberry et al., 1991; Hooks et al., 1992), but increase locomotor activity in animals habituated to their environment (e.g., Svensson et al., 1994; Al-Khatib et al., 1995) or exposed to an environment that does not contain novel objects (e.g., Boldry and Uretsky, 1988; Maldonado-Irizarry and Kelley, 1994). Likewise, intra-accumbens injections of a dopamine agonist decrease novelty-induced locomotion (Mogenson and Wu, 1991a,b), but increase locomotion in animals habituated to their environments (Wu et al., 1993a). Novelty in the environment can even reverse the direction of locomotor responding to a dopamine agonist injected peripherally (Van Hartesveldt, 1997). The neural mechanisms underlying these effects are not clear, but studying the ontological development of locomotor behavior may help to elucidate them.

Locomotor responses to novelty and dopaminergic drugs vary in an age-dependent manner. Locomotor exploration by an isolated rat pup in a novel environment is barely exhibited by 10-day-old rats, rises rapidly to a peak in 15-day-olds, falls by 25 days of age, but can peak again during the periadolescent period before reaching adult levels (Campbell et al., 1969; Randall and Campbell, 1976; Barrett et al., 1982; Spear and Brake, 1983). While intra-accumbens or peripheral injections of dopamine agonists can suppress locomotion induced by novelty in post-weanling rat pups and adult rats (Mogenson and Wu, 1991a,b; Van Hartesveldt et al., 1992, 1994), intra-accumbens injections of quinpirole do not suppress the low level of novelty-induced locomotion in 11-day-old pups (Frantz and Van Hartesveldt, 1995). In general, novelty-dependent, dopamine agonist-induced locomotor suppression occurs only in rat pups older than approximately 3 weeks of age and reaches adult levels only after postnatal day 30 (Shalaby and Spear, 1980; Spear and Brake, 1983; Arnt, 1983; Frantz et al., 1996). The late ontological onset of the interaction between novelty and dopaminergic compounds indicates that it requires a neural mechanism that does not mature until approximately the third postnatal week.

The goal of the present study was to explore the ontological development of the role of the nucleus accumbens in behavior by examining the locomotor effects of glutamate activity at NMDA receptors in the nucleus accumbens of developing rats. The non-competitive NMDA receptor antagonist, MK801, was employed because MK801 is a use-dependent, open-channel blocker of the NMDA receptor-associated ion channel and its effects on locomotion therefore indicate that endogenous glutamate had already activated the NMDA receptor and opened the associated ionophore. MK801 was injected directly into the nucleus accumbens of both male and female, developing and adult rats, and subsequent locomotor activity was analyzed.

2. Materials and methods

2.1. Subjects

Subjects were derived from Sprague–Dawley dams and sires from Zivic–Miller Laboratories (Portersville, PA). Female rats were placed in breeding cages with males and given daily vaginal lavage to check for sperm. Pregnant females were checked twice daily for litters, so that the time of birth was recorded within 12 h. The day of birth was recorded as day 0. On day 1, litters were culled to 10 pups with approximately equal numbers of males and females. On day 25, the rat pups were weaned, separated by sex and housed with at least one other littermate. Colony rooms were maintained at 21°C on a 14:10 light:dark cycle with lights on at 0800 h. Testing took place between 0800 and 1800 h in a room maintained at 26°C. All procedures were approved by the Institutional Animal Care and Use Committee (IACUC) at the University of Florida and were in accordance with the principles of laboratory animal care established by the National Institutes of Health (Bethesda, MD, USA).

Rat pups were implanted with injection cannulae at 10, 20 and 30 days of age and tested the next day at 11, 21, and 31 days of age. Adult rats were between 60 and 65 days of age at the time of cannula implantation. Each dose group consisted of 6 to 15 rats with approximately equal numbers of males and females in each group. Rats from 2 to 5 different litters were tested at each drug dose. Each rat was tested only once.

2.2. Surgery

Stereotaxic surgery was carried out while 10-day-old rat pups were under hypothermic anesthesia (Danneman and Mandrell, 1997). Older pups and adults were administered a ketamine/xylazine anesthesia cocktail in doses dependent on the age and sex of the animal (Wixson et al., 1987). Rat pups were placed on foam platforms during surgery in order to raise their body levels close to that of the ear bars. For 10-day-old pups in which the ears were not yet open, long incisions in the skin over the skull were made so that the ear bars could be secured directly against the exposed ear cavity. Guide cannulae were constructed from 23 gauge stainless steel tubing (7 mm in length). They were implanted bilaterally into the cortex above the nucleus accumbens (2.5 mm below the skull surface) and secured to the skull with dental acrylic. Coordinates from bregma on the antero-posterior and medio-lateral planes were determined empirically for each age group, as were the lengths of injection needles (30 gauge) that would enable drug placement in the nucleus accumbens (3 to 4.5 mm ventral from the tip of the guide cannulae; Table 1).

In order to analyze the specificity of the intra-accumbens drug placement, the injection site was varied along the dorsal–ventral plane by using shorter injection needles to inject 10 μg MK801 into the striatum of 21- and 31-day-old rat pups. Eleven-day-old pups were not used in the placement analysis because pilot experiments revealed no profound locomotor responding to the drug placed at several cerebral locations. Nor were adult rats used in the placement analysis, because adult rats did not exhibit robust activation in response to intra-accumbens MK801 and intra-striatal MK801 was reported to produce even less locomotion (Al-Khatib et al., 1995).

All rats were allowed approximately 24 h to recover from surgery. Rat pups were returned to their pre-surgery housing conditions for the interval between surgery and testing. Adult rats, which had previously been group-housed, were isolated for the interval between surgery and testing in order to protect the implanted cannulae.

2.3. Drug procedure

(+)MK-801 hydrogen maleate [(5*R*,10*S*)-(+)-5-methyl-10,11-dihydro-5*H*-dibenzo[*a*,*d*]cyclohepten-5,10-imine, dizocilpine maleate, Research Biochemicals International, MA, USA], was dissolved over heat in glacial acetic acid, then pH-balanced with 1 N NaOH to a pH of approximately 4.85. The solution was either used fresh or frozen in aliquots for use on the day of testing. MK801 was injected bilaterally into the nucleus accumbens at doses of 0 [distilled water (dH_2O) control injection], 3 or 10 μg in volumes of 0.25 μl. The solutions were equivalent to 0, 0.039 or 0.12 M concentrations. A separate control group of seven rats (54–56 days of age) was injected with 0.5 μl of a glacial acetic acid/NaOH solution with pH = 4.85 and showed that an intra-accumbens injection of acidic solution does not affect the locomotion (data not shown).

Table 1
Stereotaxic coordinates (mm measurements) for injection placement in the nucleus accumbens of rats at various ages

Age	Nose bar	A–P	M–L	D–V
10 days	−2.0	+1.4	+2.0	−10.5
20 days	−2.0	+1.8	+2.0	−10.5
30 days	−3.0	+1.9	+1.8	−11.0
Adult	−3.3	+2.2	+1.6	−11.5

Dorso-ventral measurements indicate the length of the injection needle.

2.4. Behavioral procedure: intra-accumbens MK801 injections

On the day of testing, rats were transported in their home cages from the colony rooms to the working laboratory. Rats, 11 and 21 days old, were maintained with their dams until just before drug injection. Drug injection took place in the testing room. The 30 gauge injection needles were inserted through the guide cannulae into the target region and the vehicle or drug was infused at a rate of 0.50 μl/min. The injection needles were left in place for an additional 30 s before removal. Immediately thereafter, each rat was placed in a randomly assigned activity monitor which recorded locomotion for 2 h.

Each Omnitech Digiscan Animal Activity Monitor consisted of a $41.91 \times 41.91 \times 30.48$ cm^3 Plexiglas cage with a wire mesh floor. Photocell beams projected across the arena. They were spaced 2.54 cm apart such that 16 beams crossed side to side and 16 beams front to back, all 3 cm above the mesh floor. Solid flooring was added for the 10-day-old rats, so that beams crossed 1.5 cm above this floor board. The interruption of photocell beams was translated into various measures of locomotor activity by the Digiscan analyzer. Total distance travelled in cm was analyzed in 5 min intervals over a period of 2 h. Experimenter observation of locomotor behavior was carried out simultaneously through one-way observation glass, in order to gauge qualitatively the various motor behaviors.

2.5. Histology

After behavioral testing of intracerebral drug injections, rats were administered an overdose of sodium pentobarbital (i.p.) and perfused intracardially with 0.9% NaCl followed by 10% formalin. The brains were removed and placed in a 10% sucrose–10% formalin solution. At least 24 h later, the brains were frozen, sectioned, mounted on slides and stained with thionine. The locations of the injection needle tips were identified, and only animals with bilateral injections within the nucleus accumbens (or dorsal to the nucleus accumbens for the placement analysis) were used in the data report.

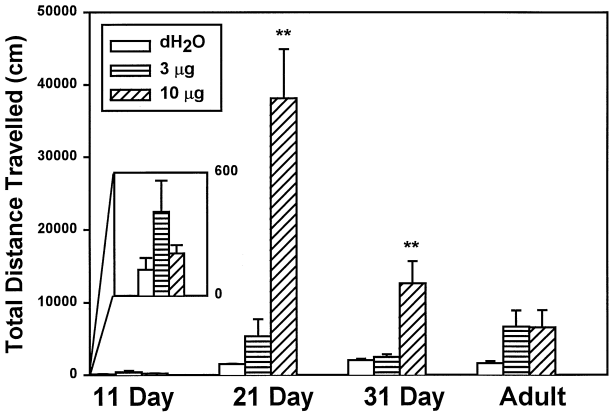

Fig. 1. Total distance (cm) travelled in a 2-h test session by rats of several different ages, following intracerebral injection with various doses of MK801. Error bars represent the standard error of the mean. Significant differences from dH_2O-injected controls are indicated ($^{**}P < 0.01$, $^{*}P < 0.05$).

2.6. Statistics

The total distance travelled in centimeters was analyzed in all the conditions tested. Separate three-way analyses of variance (ANOVAs) with repeated measures (time factor) were carried out for each age group with gender, drug dose, and time interval (5 min) as the main factors. Follow-up comparisons were made using two-way and one-way ANOVAs followed by Duncan's New Multiple Range Test to determine significance ($P < 0.05$).

3. Results

Generally, the MK801-induced activity consisted of hyperlocomotion, stereotyped sniffing, 'frantic exploration' of the test monitor, hyper-reactivity and hyper-excitability. The dH_2O-injected animals explored the novel environment initially but habituated within approximately 20 min.

3.1. Eleven-day-old rat pups

Intra-accumbens MK801 (3 or 10 μg) elicited infrequent bouts of a motor response that resembled obstinate progression (head pressed into a corner with limbs continuing to step in a coordinated movement pattern) and was sometimes followed by wall-climbing. As determined by experimenter observation, only 7 out of 19 pups given

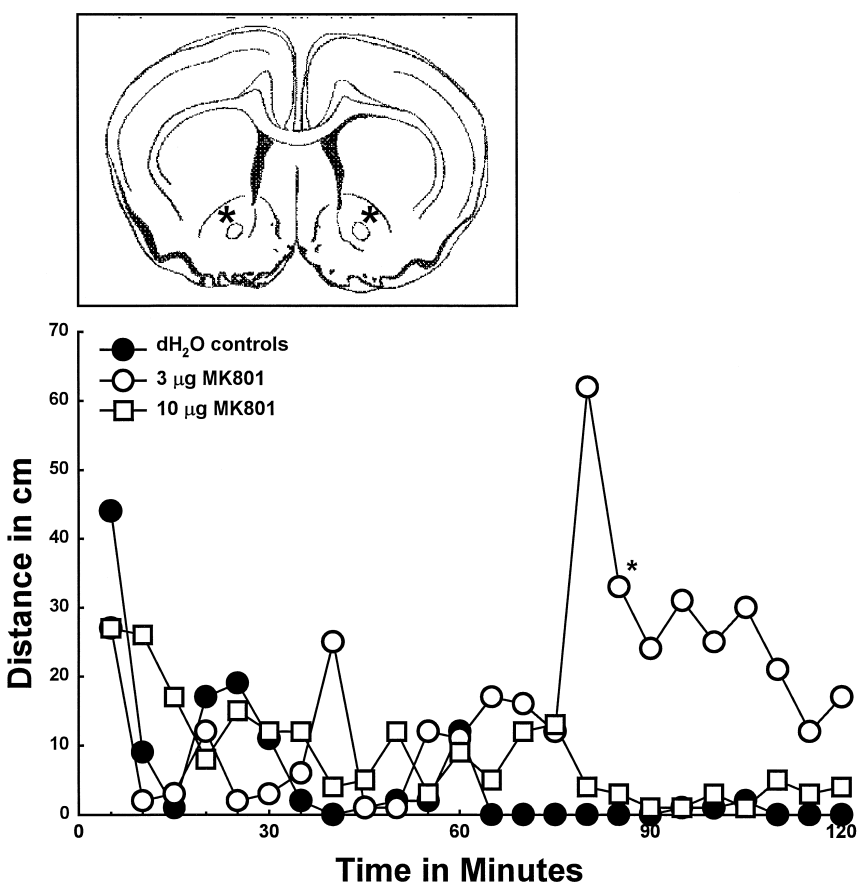

Fig. 2. Distance travelled (cm) by 11-day-old rat pups following intra-accumbens injection of various doses of MK801. A significant difference from the dH_2O-injected control group is shown ($* P < 0.05$). Error bars have been omitted for clarity. Note that the range on the ordinate is lower for 11-day-old rats than for older rats. Inset shows approximate location of MK801 injection into the nucleus accumbens.

MK801 responded in this manner. Other pups did not exhibit any other noticeable behaviors different from the dH_2O-injected control group. Thus, there was no significant main effect of dose [$F(2,22) = 2.10$] (Fig. 1). There was a significant gender × dose × time interaction, however [$F(46,437) = 1.53$, $P < 0.018$] (Fig. 2). Two of the female rat pups were active near the end of the test session following injection of 3 μg MK801 and their locomotor scores were the only source of significant difference between males and females. Therefore, Fig. 2 depicts the male and female groups combined and shows the significant dose × time interaction [$F(46,506) = 1.55$, $P < 0.014$]. The 3 μg dose of MK801 increased activity above control levels at only the 85 min time interval.

3.2. Twenty-one-day-old rat pups

In 21-day-old rat pups, only the 10 μg dose increased the distance travelled in the 2 h test session, according to a significant effect of dose [$F(2,20) = 19.99$, $P < 0.0001$] (Fig. 1). A significant dose × time interaction [$F(46,460) = 1.77$, $P < 0.002$] revealed that the 10 μg dose increased the distance travelled from 15 min post-injection through the end of the test session, compared with rats injected with 3 μg MK801 or dH_2O (Fig. 3). No significant gender effects were recorded.

3.3. Thirty-one-day-old rat pups

In 31-day-old rat pups, 10 μg MK801 increased the distance travelled during the 2 h test session, as confirmed by the significant effect of dose [$F(2,25) = 14.74$, $P < 0.0001$] (Fig. 1). There were also significant time effects [$F(23,575) = 16.29$, $P < 0.0001$] and a significant dose × time interaction [$F(46,575) = 5.74$, $P < 0.0001$] (Fig. 4). Although there was a significant gender × dose × time interaction [$F(46, 575) = 2.30$, $P < 0.0001$], at no particular time interval did the activity levels induced by either dose of MK801 in males vs. females differ significantly. Furthermore, the time intervals at which the activity of MK801-injected rats differed significantly from activity levels in same-sexed control groups were highly similar for males and females. Therefore, Fig. 4 depicts the male and female groups combined. In that case, 10 μg MK801 increased the distance travelled from 20–100 min post-injection, compared with vehicle-injected rats.

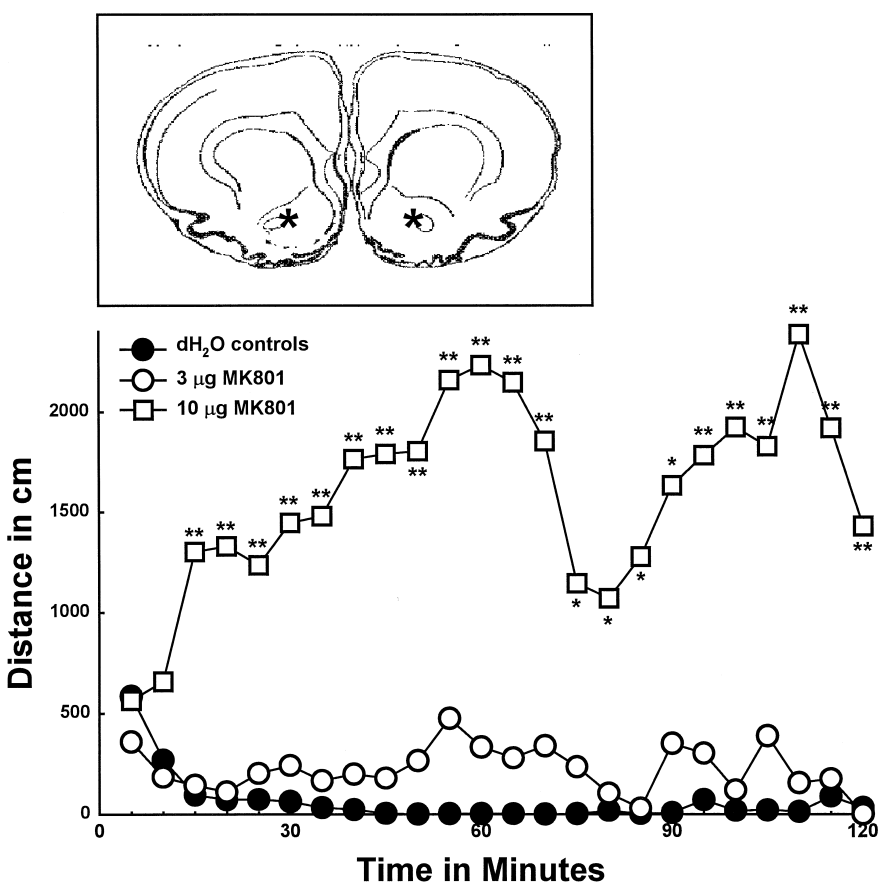

Fig. 3. Distance travelled (cm) by 21-day-old rat pups following intra-accumbens injection of various doses of MK801. Significant differences from the dH_2O-injected control group are shown ($^{**}P < 0.01$, $^{*}P < 0.05$). Error bars have been omitted for clarity. Inset shows approximate location of MK801 injection into the nucleus accumbens.

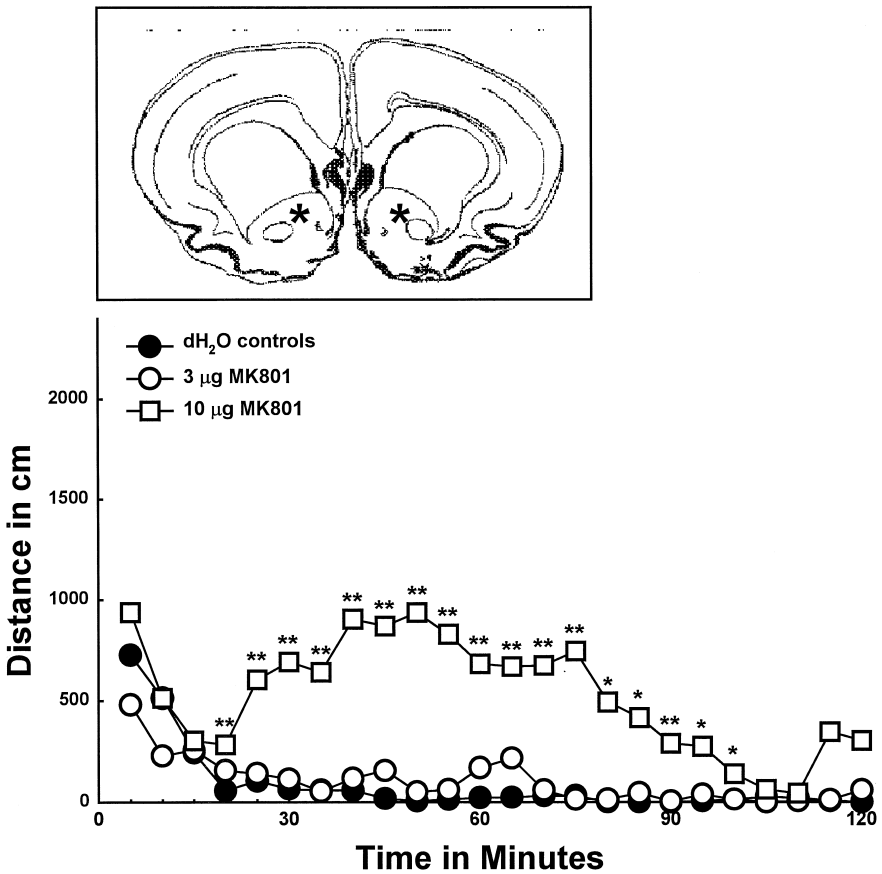

Fig. 4. Distance travelled (cm) by 31-day-old rat pups following intracerebral injection of various doses of MK801. Significant differences from the dH_2O-injected control group are shown ($^{**}P < 0.01$, $^{*}P < 0.05$). Inset shows approximate location of MK801 injections into the nucleus accumbens.

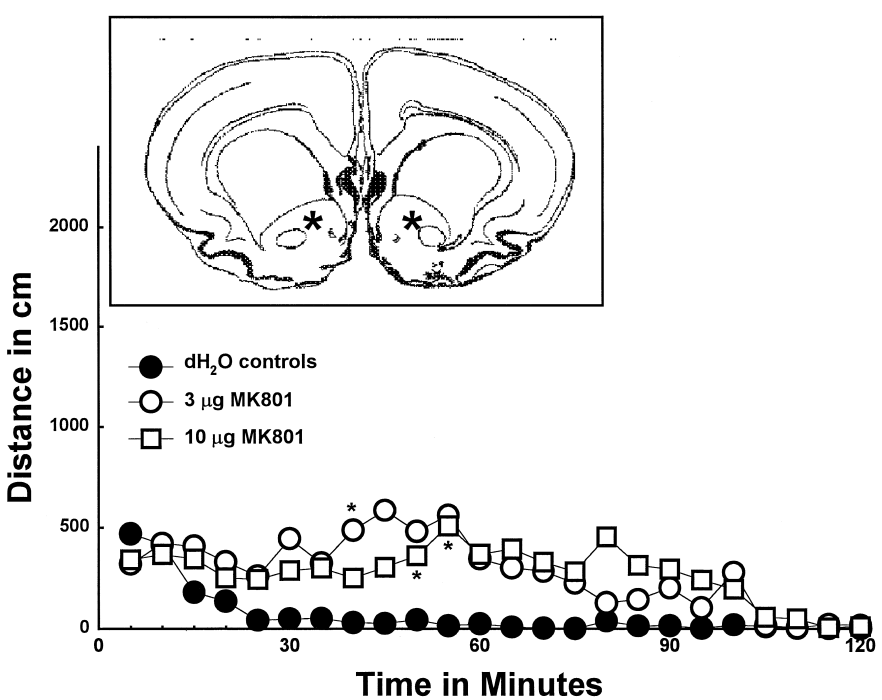

Fig. 5. Distance travelled (cm) by adult rats following intracerebral injection of various doses of MK801. Significant differences from the dH_2O-injected control group are shown ($^{**}P < 0.01$, $^{*}P < 0.05$). Inset shows approximate location of MK801 injections into the nucleus accumbens.

3.4. Adult rats

In adult rats, although MK801 (3 or 10 μg) did not alter the total distance travelled in cm over the entire 2 h test session (Fig. 1), there was a time effect [$F(23,598) = 5.73$, $P < 0.0001$] and a dose × time interaction [$F(46,598) = 2.03$, $P < 0.0001$] (Fig. 5). The major effect of MK801 in adult rats was to delay locomotor habituation. The dH_2O-injected control rats displayed little activity after the initial 10 min of exploration, as revealed by significant within-

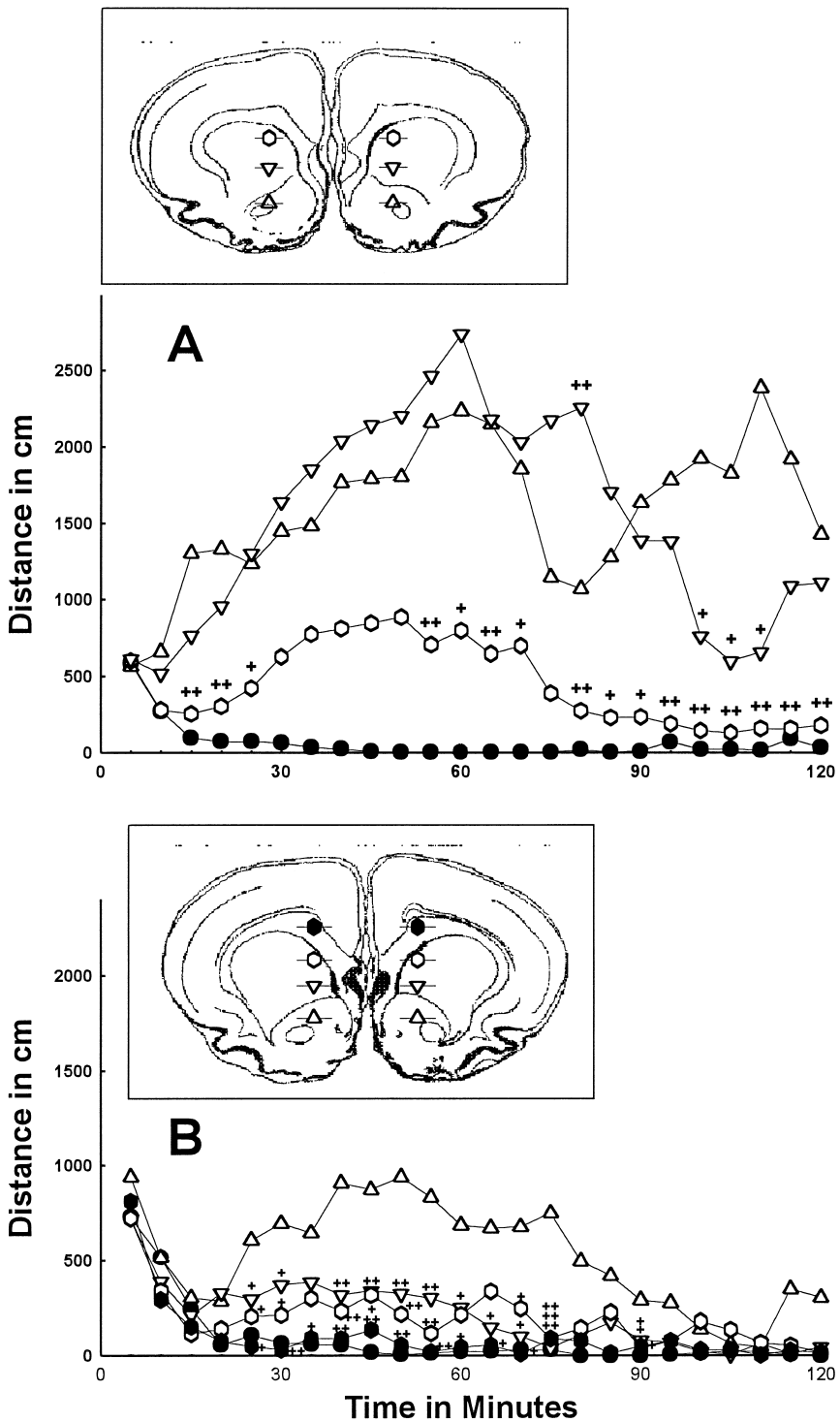

Fig. 6. (A) Distance travelled (cm) by 21-day-old rat pups following injection of 10 μg MK801 to the nucleus accumbens, mid-striatum or dorsal striatum; (B) the same measure for 31-day-old pups, including a fourth intra-cerebral injection placement near the corpus callosum. Insets: Approximate location of MK801 injections to the nucleus accumbens and striatum. Significant differences between the intra-accumbens injection site and the other sites are shown ($^{++}P < 0.01$, $^{+}P < 0.05$). Closed circles represent activity of control groups injected with dH_2O into the nucleus accumbens.

group differences in activity levels during the first two time intervals compared with all subsequent intervals ($P < 0.01$). In contrast, the activity levels of MK801-injected rats did not differ within group over time. Analyses across dose at each time interval showed few points of significant difference (Fig. 5). No significant gender effects were found.

3.5. Histology

Histological analyses showed that the injections were placed in the nucleus accumbens for rats of all ages. Slight differences in injection site were noted across ages such that the injection was predominantly just lateral to the anterior commissure for 11-day-old pups (Fig. 2 inset), but more medial for 21- and 31-day-old pups and adults (Figs. 3–5 insets).

3.6. Examination of placement specificity

In 21-day-old rat pups, there were significant effects of placement [$F(2,26) = 11.386$, $P < 0.0003$] and time [$F(23,598) = 5.229$, $P < 0.0001$] and a significant placement × time interaction [$F(46.598) = 2.01$, $P < 0.0001$] (Fig. 6A). The mid-striatal injection of MK801 induced activity that differed from the intra-accumbens injection group at only four time intervals in the test session, whereas the dorsal striatal injection induced significantly less activity than the intra-accumbens injection from 15–120 min post-injection. In 31-day-old rat pups, the effects of placement [$F(3,32) = 7.023$, $P < 0.0009$], time [$F(23,736) = 13.894$, $P < 0.0001$], and a placement × time interaction [$F(69,736) = 2.16$, $P < 0.0001$] were also significant (Fig. 6B). In all three of the striatal placements, MK801 elicited activity levels that were significantly lower than the intra-accumbens injection group from approximately 25–90 min post-injection. Striatal placements of MK801 did not induce other stereotypies in pups of either age that might have interfered with the expression of locomotion, as determined by experimenter observation.

4. Discussion

The present experiment shows that MK801 injected into the nucleus accumbens of developing rats increases locomotion in a novel environment in a dose- and age-dependent manner. Ontological changes in MK801-induced activity are likely to reflect maturational changes in glutamate activity in the limbic-motor circuitry involved in the locomotor effects of intra-accumbens MK801. For example, given that the prefrontal cortex and hippocampus mature relatively late in ontogeny (Angevine, 1975; Alexander and Goldman, 1978; O'Keefe and Nadel, 1978; Benes, 1989), it is conceivable that their glutamatergic projections into the nucleus accumbens are not yet fully functional in 11-day-old rats, few of which respond to intra-accumbens MK801. Anatomical analyses of the development of glutamate inputs to the nucleus accumbens that could confirm this idea have not been carried out yet. Nevertheless, the similarity of obstinate progression exhibited by the present 11-day-old rats given MK801 and decorticate cats (Villablanca et al., 1976) supports the contention that a lack of cortical efferents could be involved in the motor behavior of 11-day-old rats. In addition, cognitive tasks thought to involve the medial prefrontal cortex and hippocampus are not performed accurately by young rat pups (Green and Stanton, 1989) and motor activity is not affected by neonatal lesions of the hippocampus until after puberty (Flores et al., 1996; Wan et al., 1996), suggesting further that these brain regions mature late in ontogeny. Alternatively, mechanisms postsynaptic to glutamate inputs in the nucleus accumbens may not yet be mature in the 11-day-old rat. NMDA receptors may not exist in sufficient density, may not be associated with the appropriate intracellular mechanisms, or may not be present on the appropriate output neurons. In this case, the sporadic motor response to MK801 exhibited by some 11-day-olds could possibly have been mediated by blockade of the NMDA ionophore in other brain regions. While it could also be postulated that nuclei to which the efferent fibers of the nucleus accumbens project are not developed enough to facilitate locomotion, intra-accumbens injections of a dopamine D2 receptor subfamily agonist can increase locomotion substantially in 11-day-old pups (Frantz and Van Hartesveldt, 1995). Therefore, output from the nucleus accumbens is at least capable of producing locomotion at 11 days of age.

Whereas 11-day-old rat pups exhibited low, inconsistent levels of responding to MK801, 21-day-old rat pups exhibited the highest magnitude of MK801-induced locomotion, with the shortest latency to onset and the longest duration, compared with rats of other ages. They also exhibited high levels of activation in response to MK801 injected into the striatum dorsal to the nucleus accumbens, whereas 31-day-old rat pups did not. Such characteristics imply that the neural mechanisms mediating MK801-induced activation are intact, widespread, and even supersensitive at this age. Indeed, at least in the striatum, several aspects of glutamate function reach adult levels during the second and third postnatal weeks and could be supersensitive early in their development. Not only do levels of glutamate and glutamate uptake sites in the striatum double during the second postnatal week (Campochiaro and Coyle, 1978), but also a plateau of [^{3}H]MK801 binding just above adult levels begins at this age in both dorsal and ventral subregions of the striatum (Subramaniam and McGonigle, 1994). In addition, a slight overshoot of [^{3}H]glutamate binding at NMDA receptors occurs in the striatum at that stage and a robust overexpression of NMDA receptors in other limbic brain regions (Insel et al., 1990) and motor regions

(Greenamyre et al., 1987) occurs at approximately 20 days of age. Moreover, this hypothesized maturation of glutamate transmission at approximately 3 weeks of age correlates with the ontological onset of the interactions between novelty and dopaminergic compounds (Shalaby et al., 1981; Arnt, 1983; Spear and Brake, 1983; Van Hartesveldt et al., 1994; Frantz et al., 1996), perhaps indicating a role for glutamate in such interactions.

If high levels of glutamate release, a high density of NMDA receptors or overabundant postsynaptic connections mediate the supersensitivity of 21-day-old rat pups to intra-accumbens MK801, then stabilized levels of glutamate release, programmed cell death, synaptic pruning or receptor elimination could contribute to the decreased magnitude and duration of MK801 effects in 31-day-old and adult rats that were shown in the present study as well as in two previous investigations with adult rats (Raffa et al., 1989; Al-Khatib et al., 1995). Maturational changes in neural connectivity could also contribute to the decreased size of the anatomical region in which MK801 injection served to stimulate locomotion in older animals shown both in this study (31-day-olds) and another (Al-Khatib et al., 1995). An additional consideration may be, however, that in the present study, only the adult rats were isolated for 24 h prior to locomotor testing. The effects of isolation stress on locomotion deserve further investigation. Finally, it should be noted that the present results confirm a role for glutamate in novelty-induced locomotion because 31-day-olds and adults were certainly affected by intra-accumbens MK801 in that the drug at least delayed locomotor habituation in these older animals.

The present age-related changes in responding are fairly typical of those induced by other locomotor-activating drugs, such as dopamine receptor agonists (Spear and Brake, 1983; Van Hartesveldt et al., 1994; Frantz et al., 1996). Generally, 31-day-old pups exhibit less drug induced activity than 21-day-olds. By adulthood, stimulation elicits even less locomotion. However, the present results differ from those of peripheral administration of MK801 in two ways. First, 30-day-olds are less active than adults following peripheral injections (Frantz and Van Hartesveldt, in press) and second, 30-day-old and adult female rats are more sensitive than male rats to the locomotor effects of peripherally-injected MK801 (e.g., Fleischmann et al., 1991; Blanchard et al., 1992; Criswell et al., 1993; Hönack and Löscher, 1993; Haggerty and Brown, 1996; Frantz and Van Hartesveldt, in press). These developmental and gender-dependent differences are therefore likely to reflect the activity of MK801 outside the nucleus accumbens.

The clinical significance of this research relates to schizophrenia, a human disorder of cognitive, emotional and motor behaviors. The condition involves abnormal development of mesocorticolimbic feedback loops between the ventral tegmental area, nucleus accumbens, cortex and hippocampus (Doran et al., 1987; Weinberger, 1988; Weinberger et al., 1988; Walker, 1994). The neurotransmitters glutamate and dopamine are major determinants of activity in these pathways and, as such, they are targets for the pharmacological treatment of this disorders (Wachtel and Turski, 1990; Moghaddam, 1994; Halberstadt, 1995; Olney and Farber, 1995a,b; Carlsson et al., 1997). The ontological development of glutamate transmission and its role in behavior should therefore be understood.

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