Increased Responsiveness of Ventral Tegmental Area Dopamine Neurons to Glutamate after Repeated Administration of Cocaine or Amphetamine Is Transient and Selectively Involves AMPA Receptors

XU-FENG ZHANG, XIU-TI HU, FRANCIS J. WHITE and MARINA E. WOLF

Department of Neuroscience, Finch University of Health Sciences/The Chicago Medical School, North Chicago, Illinois

Accepted for publication January 30, 1997

ABSTRACT

It is well established that behavioral sensitization to psychomotor stimulants is associated with adaptations in the mesoaccumbens dopamine (DA) system. We showed previously that the responsiveness of ventral tegmental area (VTA) DA neurons to glutamate was significantly enhanced in amphetamine- and cocaine-pretreated rats tested after 3 days of withdrawal, which suggests that adaptations in excitatory amino acid transmission also contribute to sensitization. The purpose of the present study was to determine the subtype of excitatory amino acid receptor responsible for this effect and to examine its persistence during withdrawal. Extracellular single cell recording and microiontophoresis were used to investigate possible alterations in the ability of glutamate agonists [(S)-α-amino-3-hydroxy-5-methyl-4-isoxazole propionate (AMPA), N-methyl-D-aspartate (NMDA), and (1S,3R)-1-aminocyclopentane-1,3-dicarboxylic acid (1S,3R-t-ACPD)] to stimulate the firing of VTA DA neurons after 3 days of withdrawal from repeated administration of saline, cocaine or amphetamine. Current-response curves showed that responses to iontophoretic AMPA, but not NMDA or 1S,3R-t-ACPD were significantly enhanced in cocaine- or amphetamine-pretreated rats in that neurons entered into a state of apparent depolarization block at significantly lower iontophoretic currents. When rats were tested for responsiveness to iontophoretic glutamate after 14 days of withdrawal, there was no significant difference between cocaine- or amphetamine- and saline-pretreated rats with respect to glutamate current-response curves. These results suggest that increased responsiveness of AMPA receptors on VTA DA neurons may contribute to sensitization at early withdrawal times, but that this alteration, like others described within the VTA, is transient.

Repeated administration of psychomotor stimulants leads to sensitization (augmentation) of their locomotor stimulatory effects (Robinson and Becker, 1986; Kalivas and Stewart, 1991). Although the ventral striatal region known as the nucleus accumbens is clearly the major site involved in stimulant-induced locomotion (Jackson et al., 1975; Pijnenburg et al., 1976) and in the expression of sensitization (Paulson and Robinson, 1991; Cador et al., 1995), the processes responsible for the initiation of the sensitization process appear to occur within the midbrain VTA (Kalivas and Weber, 1988; Vezina and Stewart, 1990; Cador et al., 1995; Perugini and Vezina, 1994), the site of DA perikarya which give rise to the mesoaccumbens DA system. The precise neuroadaptations that induce behavioral sensitization remain to be elucidated. Most evidence implicates alterations in the functioning of DA neurons, including subsensitivity of DA D2-like receptor auto-regulation of impulse activity (Kamata and Rebec, 1984; White and Wang, 1984; Henry et al., 1989; Ackerman and White, 1990), enhancement of the basal firing activity of the DA neurons (White and Wang, 1984; Henry et al., 1989) and increased release of dendritic DA in VTA (Kalivas and Duffy, 1993b), effects that are probably inextricably related to one another (Wolf et al., 1993, 1994). However, recent discoveries have placed a new emphasis on the potential role of EAAs in the initiation of behavioral sensitization. Many studies have shown that NMDA receptor antagonists prevent the development of sensitization when coadministered repeatedly with amphetamine or cocaine (Karler et al., 1989, 1990, 1991, 1994; Wolf and Khansa, 1991; Kalivas and Alesdatter, 1993; Stewart and Druhan, 1993; Wolf and Jeziorski, 1993; Haracz et al., 1995; Ida et al., 1995; Shoaib et al., 1995; Wolf et al., 1995; Kim et al., 1996),

ABBREVIATIONS: 1S,3R-t-ACPD, (1S,3R)-1-aminocyclopentane-1,3-dicarboxylic acid; AMPA, (S)-α-amino-3-hydroxy-5-methyl-4-isoxazole propionate; DA, dopamine; NMDA, N-methyl-D-aspartate; VTA, ventral tegmental area; ANOVA, analysis of variance; EAA, excitatory amino acid.
as well as with methamphetamine (Ohmori et al., 1994) or morphine (Wolf and Jezioriski, 1993; Jezioriski et al., 1994). MK-801 also blocks cellular changes in the mesoaccumbens DA system that normally accompany the development of behavioral sensitization (Wolf et al., 1994). AMPA receptors may also be involved in the initiation of behavioral sensitization, although there may be differences between stimulants and between mice and rats (Karler et al., 1991, 1994; Pierce et al., 1996; Li et al., 1996).

Because sensitization is initiated in the VTA and can be prevented by intra-VTA administration of NMDA antagonists (Kalivas and Alesdatter, 1993), it is plausible that alterations in glutamate receptor function in the VTA play an important role in the sensitization process. In a previous study, we demonstrated that repeated administration of cocaine or amphetamine resulted in enhanced responsiveness of VTA DA neurons to iontophoretic application of glutamate, whereas nucleus accumbens neurons exhibited subsensitivity to glutamate’s excitatory effects (White et al., 1995b). Repeated cocaine administration has been found to increase levels of the AMPA receptor subunit GluR1 and the NMDA receptor subunit NR1 in the VTA (Fitzgerald et al., 1996), effects which may be related to the increased electrophysiological responsiveness to glutamate in the VTA. The present study sought to identify the EAA receptor subtype(s) responsible for increased responsiveness to glutamate, using iontophoretic application of NMDA, AMPA and the metabotropic glutamate receptor agonist 1S,3R-t-ACPD. In addition, because sensitization is known to involve different cellular alterations at short and long withdrawals (e.g., Wolf et al., 1993), we determined whether enhanced responsiveness to glutamate persisted 14 days after the termination of repeated cocaine or amphetamine administration.

Methods

Animals and drug treatment. All procedures were performed in strict accordance with the Guide for the Care and Use of Laboratory Animals as adopted and promulgated by the National Institutes of Health and were approved by the Institutional Animal Care and Use Committee of the Chicago Medical School. Male Sprague-Dawley rats (Harlan, Indianapolis, IN), weighing 225 to 249 g at the start of experiments, were used in all studies. Rats were housed two per cage with free access to food and water in a colony room maintained under constant temperature (21–23°C) and humidity (40–50%) on a 12-hr light/dark schedule (7:00 A.M., on; 7:00 P.M., off). There were at least constant temperature (21–23°C) and humidity (40–50%) on a 12-hr with free access to food and water in a colony room maintained under experiments, were used in all studies. Rats were housed two per cage with free access to food and water in a colony room maintained under constant temperature (21–23°C) and humidity (40–50%) on a 12-hr light/dark schedule (7:00 A.M., on; 7:00 P.M., off). There were at least 3 days of habituation to the colony before any treatment began. Each rat received i.p. injections of either cocaine HCl (15.0 mg/kg), d-amphetamine sulfate (5.0 mg/kg) or saline (1.0 ml/kg) once daily for 5 consecutive days, with all injections administered in home cages. These treatment regimens have been demonstrated previously to produce robust behavioral sensitization (Kalivas and Duffy, 1993a; Wolf and Jezioriski, 1993).

Surgery. Each rat, on either the third or fourteenth day after the last injection of the treatment regimen, was anesthetized with chloral hydrate (400 mg/kg i.p.) and mounted in a stereotaxic frame. Body temperature was maintained at 36–37°C with a thermostatically controlled heating pad. A tail vein was catheterized for administration of additional anesthetic as needed. A burr hole was drilled in the skull and the dura was retracted from the area overlying the VTA (A 3.0–3.5, L 0.5–1.0, V 6.5–8) (Paxinos and Watson, 1986).

Single-unit recording and microiontophoresis. Procedures for extracellular recording from VTA DA neurons have been detailed previously (Henry et al., 1989). Five barrel glass micropipettes were pulled and broken back under a microscope to achieve a tip diameter of approximately 4 to 7 μm. The center recording barrel of each micropipette was filled with 2 M NaCl saturated with 1% Fast Green dye (2–5 meq/mho impedance). One side barrel of the micropipette was filled with 2 M NaCl for automatic current balancing, whereas the remaining side barrel contained combinations of the following drugs: AMPA (0.01 M, pH 8), NMDA (0.1 M, pH 8), 1S,3R-t-ACPD (0.01 M, pH 8) or l-glutamate acid monosodium salt (GLU; 0.1 M, pH 8). Retaining currents of +8 to +10 nA were applied to drug barrels (20–70 meq/mho impedance) between ejection periods. Electrical signals were passed through a high-impedance amplifier and displayed on an oscilloscope. Individual action potentials were discriminated, monitored with an audio amplifier and digitized for off-line analysis.

The VTA DA neurons were identified on line by anatomical location within the VTA and well established electrophysiological criteria (Bunney et al., 1973; Wang, 1981). Each neuron was recorded for 3 to 5 min to establish a stable baseline firing rate. Glutamate agonists were then administered iontophoretically for a 40-sec period. At the end of that 40-sec period, the current applied to the drug barrel was doubled until the cell entered a state of apparent depolarization block. This state was defined as a loss of detectable activity after increases in firing rate accompanied by burst firing, diminished spike amplitude and increased spike duration (Grace and Bunney, 1986). For purposes of quantifying the number of neurons in a state of apparent depolarization block at each iontophoretic current of glutamate agonist, we operationally defined this state as follows: drug-induced firing rate below 50% of the maximal firing rate during the 40-sec iontophoretic application of a particular drug current, with continued decline to the point of inactivity at higher currents (White et al., 1995b). Once this state of inactivity was achieved, no additional currents were tested and the cell was then assigned a firing rate of “0” for all higher currents. All cells used in this study recovered fully from apparent depolarization block when glutamate agonist iontophoresis was discontinued.

Histology. At the end of the experiment, the final recording site was marked by passing a 25 nA cathodal current through the recording barrel for 20 min to deposit a spot of Fast Green dye. The rat was then perfused with saline followed by 10% buffered formalin for 15 min. Serial coronal sections were cut at 50-μm intervals and stained with cresyl violet and neutral red. The Fast Green dye spot served as a reference point to extrapolate the location of other recorded cells. All cells included in the present analysis were confirmed to lie within the VTA.

Drugs. (+)-Amphetamine sulfate was provided by the Research Technology Branch of the National Institute on Drug Abuse. (-)-Cocaine hydrochloride was obtained from Sigma (St Louis, MO). AMPA, NMDA and 1S,3R-t-ACPD were obtained from Research Biochemicals International (Natick, MA). Doses refer to salt weights.

Statistical analysis. Data were analyzed either with Student’s t test or two-way ANOVA with repeated measures on one variable (iontophoretic current). Subsequent planned comparisons between treatment and control means were conducted with Dunnett’s test with α = 0.05. Tests for significance between two proportions were conducted with the Fisher’s exact probability test.

Results

Responses of VTA DA neurons to AMPA. In saline-pretreated (control) rats, iontophoretically applied AMPA, at currents of 1 to 4 nA, caused a current-dependent increase in the firing rate of VTA DA neurons (figs. 1A1 and 2A). As the iontophoretic current increased further, DA neurons began to fire in bursts, with decreasing spike amplitude and increasing waveform duration. Finally, they entered a state of apparent depolarization block (Grace and Bunney, 1986), as defined operationally under “Methods.” Most neurons in the control group (8 of 11) entered a state...
of apparent depolarization block at 8 nA, and the remainder did so at 16 nA (fig. 2A). In this group, the mean maximal increase in firing rate elicited by iontophoretic AMPA was 74.69 ± 20.01%. Neurons recorded from rats pretreated with either cocaine or amphetamine and tested 3 days after the last injection were less likely to be activated to high rates of firing, because they entered a state of apparent depolarization block at significantly lower iontophoretic currents (figs. 1B1, 1C1 and 2A). Thus, in both the cocaine- and amphetamine-treated groups, AMPA elicited a dose-dependent increase in firing rate of most neurons at currents of 1 to 2 nA. However, at 4 nA, a significantly higher proportion of neurons in both the cocaine group (13/21, P = .001) and the amphetamine group (6/13, P < .05) were now in a state of apparent depolarization block as compared with the saline group (0/11). All remaining neurons in the cocaine and amphetamine groups entered a state of apparent depolarization block at 8 nA (fig. 2A), a current at which 8/11 neurons from saline-pretreated rats also exhibited apparent depolarization block. The maximal increase in firing rate elicited by AMPA was 25.32 ± 7.07% for the cocaine group and 12.69 ± 2.60% for the amphetamine group. ANOVA revealed a significant difference in current-response curves between the control group and either the cocaine (F(1,30) = 12.22, P = .0018) or amphetamine (F(1,26) = 4.80, P = .037) groups.

Responses of VTA DA neurons to NMDA. Iontophoretic administration of NMDA, at currents of 1 to 4 nA, caused a current-dependent increase in the firing rate of VTA DA neurons in all three groups. At currents of 8 nA or more, NMDA drove the neurons into a state of apparent depolarization block (figs. 1A2, 1B2, 1C2 and 2B). The number of neurons entering depolarization block at each current was similar in all groups (fig. 2B). ANOVA revealed no significant differences in the current-response curves between the control group and either the cocaine (F(1,25) = 0.024, P = .85) or amphetamine (F(1,26) = 0.13, P = .72) groups.

Responses of VTA DA neurons to glutamate after 14 days of withdrawal from repeated cocaine or amphetamine. In all experiments described above, as well as our previous report that responsiveness to iontophoretic glutamate was enhanced in VTA DA neurons recorded from amphetamine- or cocaine-pretreated rats (White et al., 1995b), electrophysiological recordings were performed 3 days after...
the last injection of psychomotor stimulant. To examine the persistence of altered responsiveness, responses of VTA DA neurons to glutamate were examined after 14 days of withdrawal from treatment with saline, cocaine or amphetamine. In all three groups, currents of 1 to 4 nA caused a current-dependent increase in firing rate, whereas higher currents (8 nA or more) drove neurons into a state of apparent depolarization block (figs. 3 and 4). ANOVA revealed no significant differences in current-response curves between the control and cocaine ($F_{1,27} = 0.068, P = .89$) or amphetamine ($F_{1,26} = 0.41, P = .54$) groups. These findings suggest that psychomotor stimulant-induced alterations in the responsiveness of VTA DA neurons to glutamate are relatively transient in nature. Additional studies demonstrated that VTA DA neurons recorded after 14 days of withdrawal from cocaine or amphetamine also failed to exhibit alterations in responsiveness to iontophoretic application of AMPA or NMDA (data not shown).

**Discussion**

We showed previously that repeated administration of psychomotor stimulants increased the responsiveness of VTA DA neurons to glutamate in that neurons entered a state of apparent depolarization inactivation (or block) at lower ion...
trophoretic ejection currents of glutamate (White et al., 1995b). In the present study, we determined the subtype(s) of glutamate receptors involved in the alteration. After a 3-day withdrawal from repeated administration of cocaine or amphetamine, VTA DA neurons were significantly more likely to enter a state of apparent depolarization block during administration of AMPA. No differences in responsiveness to NMDA or the metabotropic glutamate agonist 1S,3R-t-ACPD were found between saline- and stimulant-pretreated rats. These findings suggest that the lower threshold for induction of depolarization block in stimulant-pretreated rats is caused by alterations in AMPA receptor responsiveness. In additional experiments, we found that responsiveness of VTA DA neurons to iontophoretic glutamate had returned to normal after 14 days of withdrawal from amphetamine or cocaine.

Repeated cocaine or amphetamine selectively alters responsiveness to AMPA. In saline-pretreated rats, VTA DA neurons exhibited excitation after iontophoretic administration of AMPA, NMDA and the metabotropic glutamate receptor agonist 1S,3R-t-ACPD, consistent with previous reports that all three glutamate receptor subtypes are present on midbrain DA neurons (Seutin et al., 1990; Meretu et al., 1991; Mercuri et al., 1992, 1993; Overton and Clark, 1992; Wang and French, 1993a, b; Wu et al., 1994; Zhang et al., 1994). Our results demonstrate a selective alteration in the responsiveness to AMPA in VTA DA neurons recorded 3 days after withdrawal from repeated cocaine or amphetamine. The major effect was decreased threshold for induction of apparent depolarization inactivation in response to iontophoretic application of AMPA, as was reported previously for iontophoretic application of glutamate (White et al., 1995b). Why was increased responsiveness to AMPA manifest as a change in threshold for depolarization inactivation? As illustrated by figure 2, DA neurons are firing at almost maximal rates in the absence of iontophoretic drug application, presumably in response to endogenous glutamatergic drive. Because responses to iontophoretic AMPA are superimposed on this endogenous drive, AMPA produces only small increases in firing rate before driving cells into depolarization inactivation. Under such conditions, it is not surprising that a change in responsiveness to iontophoretic AMPA was most evident at the high end of the current-response relationship, that is, at currents capable of eliciting depolarization inactivation.

Using both extracellular and intracellular recordings from in vitro slice preparations of the VTA, Wang and French (1993a, b) showed that low concentrations of glutamate excite VTA DA neurons via a preferential effect on the NMDA receptor, with AMPA receptors coming into play only at higher glutamate concentrations. These investigators also demonstrated concentration-dependent biphasic effects of AMPA and NMDA, with low concentrations depolarizing and activating VTA DA neurons and high concentrations inducing depolarization block (Wang and French, 1993a, b; Wang et al., 1994). Such findings are consistent with our results regarding alterations in the effects of EAs after chronic treatment with psychomotor stimulants. Thus, if only AMPA receptors are altered by repeated administration of amphetamine or cocaine, one would expect no alteration in the rate-enhancing effects of low glutamate currents (mediated by NMDA receptors), but significant alteration in the response to higher iontophoretic glutamate currents (mediated by AMPA receptors), as was observed with iontophoretic glutamate administration in our previous study (White et al., 1995b). Interestingly, when AMPA was used as the iontophoretically applied agonist, VTA DA neurons in stimulant-pretreated rats showed a trend toward increased responsiveness to the rate-enhancing effects of low iontophoretic currents, as well as lower threshold for induction of depolarization block (fig. 2). This is presumably because AMPA receptors are mediating both components of the response in stimulant-pretreated rats because of increased AMPA receptor sensitivity.

Increased responsiveness of VTA DA neurons to AMPA is transient. Previous work has established that the induction and maintenance of behavioral sensitization is a complex process, with different cellular alterations contributing at different times. At short withdrawal times, cellular alterations in the VTA predominate, consistent with studies establishing VTA as the site of initiation of sensitization (Kalivas and Weber, 1988; Vezina and Stewart, 1990; Cador et al., 1995; Perugini and Vezina, 1994). Such alterations are generally transient. For example, psychomotor stimulant-induced subsensitivity of impulse-modulating somatodendritic DA autoreceptors is obvious at short withdrawal times (1–3 days), is less evident at intermediate withdrawal times (5–8 days) and is absent at longer withdrawal times (10–14 days) (White and Wang, 1984; Ackerman and White, 1990; Wolf et al., 1993). Decreased VTA levels of the inhibitory G-protein subunits Gi and Go have been observed in chronic cocaine-treated rats 1 or 6 hr, but not 24 hr, after discontinuing daily injections (Nestler et al., 1990; Striplin and Kalivas, 1992). Basal extracellular DA levels in the VTA are elevated in cocaine-compared with saline-pretreated rats when measured 1 day after discontinuing daily treatments, but not after 14 days (Kalivas and Duffy, 1993b). These various measures are probably related to each other, and to the increase in basal DA cell firing rate observed after short withdrawals from repeated administration of amphetamine.

Fig. 4. Current-response curves illustrating the effects of iontophoretic glutamate on VTA DA neurons in rats after 14 days of withdrawal from repeated administration of saline, cocaine or amphetamine. The increased sensitivity to depolarization inactivation demonstrated after 3 days of withdrawal from cocaine or amphetamine is no longer present. In saline-pretreated rats, VTA DA neurons showed a trend toward increased responsiveness to AMPA after 14 days of withdrawal from amphetamine or cocaine. The numbers in the different brackets indicate the number of cells in each group that had entered a state of apparent depolarization block, as defined operationally under "Methods." Data points represent the mean ± S.E.M.
or cocaine (White and Wang, 1984; Henry et al., 1989). Our
failure to observe increased basal firing rates in the present
study likely relates to the larger sample size and shorter
withdrawal periods used in these previous studies (White

We have now demonstrated another transient alteration in
the VTA of psychomotor stimulant-pretreated rats, that is,
enhanced responsiveness of VTA DA neurons to the excita-
tory effects of AMPA. Is this effect related to the others
discussed above, and if so, which is primary? On the one
hand, increased excitatory tone at AMPA receptors could
account for enhanced basal activity of DA neurons and in-
creased somatodendritic DA release, as well as for subsensi-
tivity to the inhibitory effects of DA autoreceptor activation.
If increased excitatory tone was completely responsible for
DA autoreceptor subsensitivity, then generalized subsensi-
tivity to all inhibitory transmitters would be expected. Yet,
VTA DA neurons in amphetamine- or cocaine-pretreated rats
do not exhibit subsensitivity to GABA (White and Wang,
1984; Henry et al., 1989). On the other hand, decreased
inhibitory tone caused by DA autoreceptor subsensitivity
might lead to an apparent increase in responsiveness to the
excitatory transmitter glutamate. This is unlikely to account
completely for the change in glutamate responsiveness, be-
cause Fitzgerald et al. (1996) have found increased levels of
the NMDA subunit NR1 and the AMPA receptor subunit
GluR1 in the VTA after 1 day of withdrawal from repeated
cocaine administration. The elevation in GluR1 levels could
contribute to the observed increase in electrophysiological
responsiveness to AMPA reported herein. Our failure to ob-
serve a change in responsiveness to NMDA after a 3-day
withdrawal may suggest that the increase in NR1 levels is
less persistent than that of GluR1, that the increase in NR1
levels occurs in non-DA neurons within the VTA or that the
increase in NR1 levels is not sufficient to increase respon-
siveness of the population of oligomeric NMDA receptors on
VTA DA neurons. Taken together, the available evidence
suggests that both DA and EAA receptor mechanisms in VTA
are altered by repeated stimulant administration. Such
alterations are likely to be interrelated, given that EAA recep-
tor blockers prevent the development of DA autoreceptor
sub sensitivity (Wolf et al., 1994) and that the level of EAA
receptor stimulation can regulate DA D2 receptor expression
(Healy and Meador-Woodruff, 1996; Nair et al., 1996).

The mechanism by which repeated psychomotor stimulant
administration might alter AMPA receptor expression or re-
captor sensitivity is unknown. However, cocaine and amphet-
amine have been reported to increase extracellular glutama-
te levels in the VTA (Kalivas and Duffy, 1985; Xue et al.,
1996). It is possible that repeated elevation of glutamate
levels in the VTA, upon repeated stimulant injection, results in
compensatory alterations in EAA receptor function.

Role of AMPA receptors in behavioral sensitization.

Much evidence is consistent with the idea that excitatory
tone to VTA DA neurons is increased shortly after discontin-
uation of repeated psychomotor stimulant administration. As
discussed above, increased excitatory tone could account for
increased basal activity of DA cells and increased somatoden-
dritic DA release. Does increased excitatory tone play a role in
the development of sensitization? Perhaps it does, given that:
1) disinhibition of VTA DA cells by uncoupling inhibi-
tory DA and GABA<sub>H</sub> receptors from associated G-proteins
with pertussis toxin results in an augmented locomotor re-
sponse to cocaine (Steketee and Kalivas, 1991); and 2) re-
peated electrical stimulation of the VTA elicits locomotor
sensitization to amphetamine (Ben-Shahar and Ettenberg,
1994).

The present results, combined with those of Fitzgerald
et al. (1996), argue that up-regulation of EAA receptors in VTA
may be a mechanism underlying increased tonic excitation of
VTA DA neurons. Indeed, midbrain DA neurons in amphet-
amine-pretreated rats show enhanced reactivity to electrical
stimulation of the prefrontal cortex, which sends EAA-con-
taining projections to the VTA (Tong et al., 1995). Interest-
ingly, the latter study reported different changes at 2 and 10
days of withdrawal, although both were consistent with in-
creased excitability of DA neurons. It is also possible that
alterations in the activity of prefrontal cortical neurons that
project to VTA contribute to the postulated increase in exci-
tatory tone (White et al., 1995a). However, even if this is not
the case and alterations occur exclusively at the level of EAA
receptor expression by VTA DA neurons, excitatory inputs
originating in the prefrontal cortex are likely to provide the
stimulation needed for the expression of such alterations.
Thus, locomotor sensitization is prevented by interruption of
the prefrontal cortex-VTA pathway, whether by intra-VTA
administration of NMDA antagonists (Kalivas and Alesdat-
ter, 1993) or by ibotenic acid lesions of the prefrontal cortex
(Wolf et al., 1995; Li and Wolf, 1997). Conversely, electrical
kindling of the prefrontal cortex results in behavioral sensi-
tization to cocaine (Schenk and Snow, 1994). These reports
are consistent with the important role of prefrontal cortical
afferents in regulating the firing pattern and activity of VTA
DA neurons (Gariano and Groves, 1988; Murase et al., 1993;
Svensson and Tung, 1989; Tong et al., 1996).

Psychomotor stimulants are known to inhibit the firing of
VTA DA neurons by increasing somatodendritic DA levels
and thereby activating impulse-modulating DA autorecep-
tors (see Wolf and Roth, 1987; White, 1996 for reviews). Is
this inconsistent with the hypothesis that sensitization re-
quires increased excitatory tone to VTA DA neurons? Not
necessarily. Although the acute effect of elevation of VTA DA
levels by psychomotor stimulants may be inhibition of DA
cell firing, their acute administration also leads to rapid
desensitization of DA autoreceptors (Seutin et al., 1991).
With chronic administration of psychomotor stimulants, the
duration of DA autoreceptor desensitization is prolonged (see
above). This, along with other compensatory changes, includ-
ing increased responsiveness to AMPA, leads to increases in
excitatory drive during the time between repeated injections
of psychomotor stimulants, as evidenced by increased basal
firing rates of VTA DA neurons after short withdrawals from
repeated stimulant administration (White and Wang, 1984;
Henry et al., 1989). Many lines of evidence argue that tran-
sient changes in VTA observed at short withdrawals, all of
which are consistent with increased excitatory drive to VTA
DA cells (see above), are necessary prerequisites for the in-
duction of longer lasting changes in the nucleus accumbens
that may underlie the persistence of behavioral sensitization.
For example, several previous studies have suggested that
autoreceptor subsensitivity must occur, albeit transiently,
for D1 receptor supersensitivity in nucleus accumbens to
show persistence (e.g., see Ackerman and White, 1990; Wolf
et al., 1994). Those who argue against a role for DA autore-

Downloaded from jpet.aspetjournals.org on May 25, 2017
ceptor subsensitivity in the initiation of sensitization, based on the inability of D2 receptor antagonists to prevent its development (Vezina and Stewart, 1989; Bijou et al., 1996), have overlooked the fact that D2 receptor antagonists also inhibit the activity of VTA DA neurons (see White, 1996, for review).

The major difference observed in VTA DA neurons recorded from cocaine- or amphetamine-pretreated rats in the present study was increased sensitivity to depolarization block. How might this be related to the function of VTA DA neurons and to psychomotor stimulant withdrawal? Midbrain DA neurons respond to salient (motivationally arousing) environmental stimuli with transient bursts of impulses that are thought to contribute to the setting of a motivational state by alerting other neuronal systems, in particular the nucleus accumbens, that subserve reward-seeking and goal-directed activity (Schultz and Romo, 1990). Increased sensitivity to depolarization block, perhaps in response to alterations in cortical drive, would render DA neurons less capable of communicating information about salient stimuli to their targets. This, in turn, might render rats less responsive to their environment. This could be related to the reduced motivation, anergia and anhedonia observed in humans during withdrawal from psychomotor stimulant addiction (Gawin and Ellinwood, 1988; Gawin, 1991).

Because increased sensitivity to glutamate-mediated depolarization inactivation is a transient effect, it is likely involved only in behavioral alterations accompanying short withdrawals (see above), or perhaps also in the “transfer” of sensitization to the nucleus accumbens, where it is expressed (Paulson and Robinson, 1991; Cador et al., 1995). However, it is possible that alterations in EAA transmission elsewhere in brain contribute, albeit via different mechanisms and circuits, to behavioral effects at longer withdrawals. Thus, recent reports suggest that repeated administration of psychomotor stimulants can lead to long-lasting changes in EAA systems. For example, repeated administration of low-dose methamphetamine results in enhancement of K⁺-stimulated glutamate efflux in prefrontal cortex measured after 7 days of withdrawal (Stephans and Yamamoto, 1995). Alterations in the effect of cocaine on glutamate efflux in nucleus accumbens core, and in behavioral effects of intra-nucleus accumbens injections of AMPA and the AMPA receptor antagonist 6-cyano-7-nitroquinoxaline-2,3-dione (CNQX), have been demonstrated after 21 days of withdrawal from repeated cocaine (Pierce et al., 1996). We have found decreased levels of mRNA for the AMPA receptor subunits GluR1 and GluR2 in the nucleus accumbens after 14 days, but not 3 days, of withdrawal from repeated amphetamine (Lu et al., in press). Thus, stimulant-induced alterations in EAA transmission may contribute to both the development and the persistence of behavioral sensitization.

Acknowledgments

The authors gratefully acknowledge Nha Lien and Pamela Alvarcz for technical assistance.

References


ZHANG et al.


