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BRIEF COMMUNICATION

Decreases in Behavioral and Striatal Neuronal Responses to Dexamphetamine With Aging

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WARENYCIA, M. W. AND G. M. McKENZIE. *Decreases in behavioral and striatal neuronal responses to dexamphetamine with aging.* PHARMACOL BIOCHEM BEHAV **33**(2) 489–491, 1989.—Striatal neurons of mature rats responded to 2.5 mg/kg dexamphetamine with increased multiple unit activity that followed the time course of drug-increased behavior. In contrast, in older (middle-aged) rats striatal neurons failed to respond to dexamphetamine with excitation. Behavioral responses were reduced by half as compared to mature rats. Retesting of these middle-aged rats with dexamphetamine did not result in either improved behavioral or neuronal responses and decreases in spontaneous MUA suggested dexamphetamine neurotoxicity in older animals. Since striatal neuronal responses and behavioral responses to dexamphetamine are greatly reduced, age-related impairment of dopaminergic neurotransmission may lead to in reductions in striatal neuronal excitation as well as feedback from dexamphetamine-induced behavior.

Striatum Dexamphetamine Age-dependent reductions Behavioral response Neuronal response Neurotoxicity

SEVERAL studies have now established that in freely-moving animals most striatal neurons respond to dexamphetamine with dose-dependent increases in discharge rate as well as behavioral and locomotor activation (5,18). Furthermore, it has been shown that this excitatory response is critically dependent on the simultaneous presence of the three major afferent systems innervating the striatum, viz. the thalamostriatal, the nigrostriatal and the corticostriatal (20–22).

Little attention has been directed toward examining the actual effects of aging on responsiveness of striatal neurons. The present study focused on examining the effects of aging on behavior and the striatal neuronal response to dexamphetamine. Several reports have indicated that the aging process may represent the development of an as yet an undefined neurochemical lesion within the striatum or its afferent systems (1,7). Furthermore, it has been reported that a single dexamphetamine treatment can restore motor performance following extensive cortical injury (3) and enhance rotational behavior as well as striatal dopamine release in normal animals (15). Therefore, the effects of a single dexamphetamine treatment on subsequent striatal neuronal responses and behavior challenge in aged animals were also examined to see if age deficits could be at least partially reversed by the use of this indirect dopamine agonist.

METHOD

Male Long-Evans rats (2–3.5 months old) weighing 250–370 g (called mature adult controls in this study) and 500–750 g (middle-aged: 597 ± 50 g, mean ± SE; corresponding to a mean age of 10 months for this rat strain as defined by the breeder, Charles River Laboratories) were used exclusively in this study. Animals were individually housed, given unrestricted access to food and water, and kept on a 12-hr dark/light photoperiod. Bipolar recording electrodes were implanted in the striatum under halothane anaesthesia as described previously (5,19) using stereotaxic coordinates (12) of AP +2.4; ML ±2.5 and DV −5.0. Animals received intramuscular Penicillin G 30,000 I.U. for prophylaxis against infection and were allowed 5–7 days to recover prior to the acute recording experiment.

Neuronal action potentials from striatal neurons were initially amplified by a preamplifier (WPI Instruments) and then differentially amplified, filtered by a band pass filter (0.3–10 kHz) and fed to a Mentor window discriminator. The output from the discriminator was then displayed on an oscilloscope (Tektronix); only preparations with signal-to-noise ratios between 2.5:1 and 10:1 were accepted. Waveforms were extracellular action potentials with durations of less than 0.5 msec, and were asymmetric and biphasic about the zero potential line with amplitudes (peak-to-peak) between 100 and 150 μV. Standard pulses generated by the window discriminator, each representing an action potential, were recorded by an Ortec counter, and 4-min bin totals printed. Animals were observed directly for signs of stereotyped behavior and locomotor activity monitored in each experiment with a

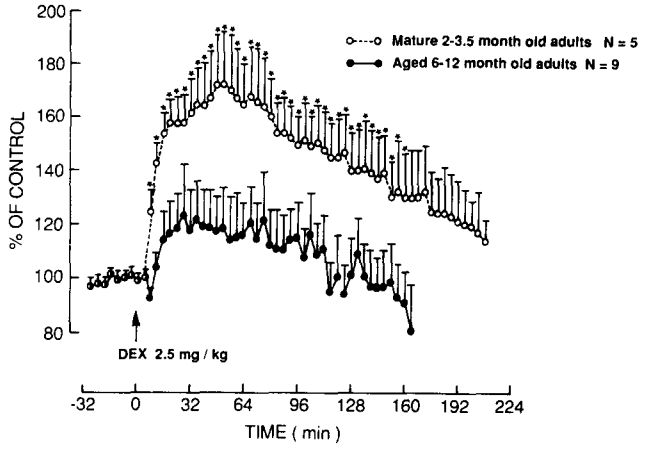

FIG. 1. Effects of 2.5 mg/kg dexamphetamine on striatal neurons of freely-moving mature (controls; ○, 5 recording sites in 5 animals) and middle-aged Long-Evans rats (●, 9 recording sites, 7 animals). Spontaneous MUA for mature and middle-aged groups was 5537 ± 970 and 5426 ± 1105 spikes/4 min repectively, and was not statistically different. Control interval data corresponding to predrug intervals was pooled for the two groups. Each point represents the mean ± SE, expressed as % of control. *$p<0.05$ compared to predrug control intervals. Behavioral activation followed the time course of striatal neuronal excitation in control animals; in middle-aged animals increases in behavior or locomotor activation lasted 60 min or less.

Stoelting activity monitor, the analog output of which was recorded on a Grass polygraph. An adjacent channel receiving the output of the discriminator was used for the ratemeter record.

Animals were allowed to move freely for 40–80 min before IP dexamphetamine administration. Bin totals from 8 consecutive 4-min recording intervals preceding dexamphetamine (baseline multiple unit activity) were averaged to obtain spontaneous MUA and the resulting mean MUA set as the predrug control level (i.e., 100%). All data points were then expressed as % of control and MUA changes after dexamphetamine were evaluated by the Student-Neuman-Keuls multiple comparison test with $p<0.05$ considered significant. Spontaneous MUA between the groups was compared by Student's t-test. Comparison of mean spontaneous MUA between first and second trials for the same animals was carried out with the paired t-test.

Following each set of experiments, a lesioning current was passed through each striatal electrode and animals perfused with saline followed by 10% formalin. Frozen 100-μM sections were then examined with a projector microscope (Reichert) to verify the striatal location of each electrode.

RESULTS

Comparison of Dexamphetamine Effects on Striatal Neurons in Control and Aged Rats

In mature 3–6-month-old rats (controls) the striatal neuronal response to 2.5 mg/kg dexamphetamine consisted of an increase in MUA (Fig. 1) that reached a maximum of $172 \pm 20\%$ of baseline levels between 30 and 60 min after dexamphetamine administration. This sustained elevation in MUA remained statistically significant compared to predrug baseline levels for over 100 min before declining to predrug firing levels by 160 min. Behavioral activation followed a similar time course to that of increased neuronal discharge rate. In contrast, in middle-aged animals, no significant increase in striatal MUA could be demonstrated over the same time course (Fig. 1).

This lack of responsiveness to dexamphetamine could not be ascribed to differences in spontaneous (baseline) MUA between control and aged animals as spontaneous MUA was nearly identical in the two groups (controls $= 5537 \pm 90$ spikes/4 min vs. 5426 ± 1105 spikes/4 min, aged). In addition, in middle-aged rats, the duration of the behavioral and locomotor activation normally elicited by this dose of dexamphetamine (as measured by the Stoelting activity monitor and direct observation) was reduced to just less than half of that seen in control animals. Behavioral observation of the aged animals prior to electrode implantation showed that these rats were not as active as the controls, spending considerably more time sleeping.

In middle-aged animals a subsequent dexamphetamine challenge 48 hr later did not result in either behavioral enhancement or restoration of the striatal neuronal response to dexamphetamine. Striatal MUA showed no evidence of any increased response to 2.5 mg/kg dexamphetamine as compared to the first trial (data not shown). Furthermore, the overall spontaneous MUA of striatal neurons (the MUA of the entire predrug period corresponding to baseline activity) was significantly reduced by approximately 36%, whereas a similar reduction was not seen in control animals. Responses to dexamphetamine to control animals in the second trial consisted of excitation and were indistinguishable from that seen in the first. Differences in responsiveness between the two groups could not be attributed to electrode location as all recording sites were localized to the anterior striatum.

DISCUSSION

Reduction in behavioral and striatal neuronal responsiveness to dexamphetamine observed in middle-aged animals may in part be due to impaired dopaminergic neurotransmission. Striatal dopamine content declines with increasing age (8) as does the rate of dopamine synthesis (24) and uptake by striatal synaptosomes in older animals (4). Decreases in both D-1 and D-2 striatal dopamine receptor binding sites also occur with increasing age (6). As the dopamine content of the striatum appears to determine the striatal neuronal response to dexamphetamine (21), the attenuated responses observed in this study may indicate an overall decline in dopaminergic neurotransmission. Since corticostriatal afferent activity may also be modulated by dopamine (11), reductions in dopaminergic neurotransmission could lead to further decreases in the response of striatal neurons to dexamphetamine as well as decreases in dexamphetamine-induced behavior.

The reduction in dexamphetamine-induced behavior activation animals is consistent with the decrease seen with aging in other behavioral indices by other investigators (9). Decreases in striatal neuronal responsiveness to dexamphetamine may thus also reflect a decreased capability of striatal afferent systems to convey feedback from drug-induced behavior to striatal neurons as well as reduced feedback from attenuated behavioral responses. Lastly, increased neurotoxicity of dexamphetamine may result in decreased striatal neuronal responsiveness and behavior, thus agreeing with an earlier study that demonstrated that reductions in behavioral activity accompanied decreased striatal excitation [19].

Age-related changes in other striatal afferent systems could also contribute to the observed reduction in the responsiveness of striatal neurons to dexamphetamine. Decreased release of striatal acetylcholine (4) may also suggest a decrease in the efficiency of neurotransmission by thalamostriatal afferents. Similarly, the 20% decrease in striatal dopamine D-2 receptor-binding sites (6) seen with increasing age may indicate loss of corticostriatal afferents, since it has been shown that approximately 30% of dopamine receptor sites are located on these fibres (16) and play a significant role in the response of striatal neurons to dexamphetamine (22).

A subsequent challenge with dexamphetamine 48 hr later did

not result in either increased behavioral responses or restoration of the excitatory response of striatal neurons to dexamphetamine. Other studies have shown a single dexamphetamine treatment resulted in either deficit reversal (3) or behavioral or neurochemical augmentation (15). Thus, the absence of an improved striatal neuronal response contrasts with results where dexamphetamine appeared to enhance striatal neuronal function after a cortical lesion (23). The lack of improvement in middle-aged animals strongly suggests that the potential for recovery of neuronal or behavioral responses is also impaired due to aging.

Another explanation for the lack of improvement may be due to the neurotoxicity of dexamphetamine previously observed only in much younger animals (10). Increases in amphetamine neurotoxicity in older animals could negate any restorative effects of the drug. Indeed, such a view is consistent with observations that dopaminergic neurons in aged rats show an enhanced susceptibility to several neurotoxins (7,14). Alternatively, middle-aged rats may require a longer time to recover from the desensitization that accompanies amphetamine action.

Changes in pharmacokinetic parameters such as the volume of distribution, tissue accumulation, or rate of metabolism of dexamphetamine could also partially explain the present findings as there appears to be some evidence for increased brain amphetamine levels in older animals (17). However, in a study of dexamphetamine uptake in various brain regions, there was no difference in uptake between 440 g rats and 350 g animals (2), suggesting that pharmacokinetic considerations may be a minor factor with respect to the effects of dexamphetamine in older animals. Further work is required to more accurately define the exact mechanism(s) underlying the decreased efficacy of dexamphetamine in exciting striatal neurons of older animals.

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