Psychopharmacology (2000) 148:322–324 © Springer-Verlag 2000

RAPID COMMUNICATION

Liesbeth Reneman · Jan Booij · Ben Schmand
Wim van den Brink · Boudewijn Gunning

Memory disturbances in "Ecstasy" users are correlated with an altered brain serotonin neurotransmission

Received: 20 August 1999 / Final version: 25 November 1999

Abstract *Rationale:* Methylenedioxymethamphetamine (MDMA) is known to damage brain pre-synaptic serotonin (5-HT) neurons. Since loss of 5-HT neurons has been implicated in memory loss, it is important to establish whether MDMA use may produce changes in postsynaptic 5-HT receptors and memory function in humans. *Objectives:* To investigate whether MDMA use leads to compensative alterations in post-synaptic 5-HT_{2A} receptors and whether there is a relation with memory disturbances. *Methods:* Brain cortical 5-HT_{2A} receptor densities were studied with [^{123}I]-5-I-R91150 SPECT in five abstinent MDMA users and nine healthy controls. Memory performance was assessed using RAVLT. *Results:* [^{123}I]-5-I-R91150 binding ratios were significantly higher in the occipital cortex of MDMA users than in controls, indicating up-regulation. Mean cortical 5-HT_{2A} receptor binding correlated positively with RAVLT-recall in MDMA users. *Conclusion:* Our preliminary results may indicate altered 5-HT neuronal function with correlated memory impairment in abstinent MDMA users.

Key words MDMA · Ecstasy · SPECT · 5-HT_2 receptor · Memory

L. Reneman · J. Booij
Graduate School of Neurosciences,
Department of Nuclear Medicine, Academic Medical Center,
1105 AZ Amsterdam, The Netherlands

B. Schmand
Department of Neurology, Academic Medical Center,
1105 AZ Amsterdam, The Netherlands

W. van den Brink
Amsterdam Institute for Addiction Research
and Department of Psychiatry, Academic Medical Center,
1105 BC Amsterdam, The Netherlands

B. Gunning
Department of Child-Adolescent Psychiatry,
Academic Medical Center, 1105 AZ Amsterdam, The Netherlands

L. Reneman (✉)
Department of Nuclear Medicine, F2N, Academic Medical Center,
Meibergdreef 9, 1105 AZ Amsterdam, The Netherlands
e-mail: l.reneman@amc.uva.nl, Fax: +31-20-6976508

Use of the popular recreational drug (±)3,4-methylenedioxymethamphetamine (MDMA, "Ecstasy") leads to toxic effects on brain serotonin (5-HT) presynaptic neurons in humans, as recently reported (McCann et al. 1998; Semple et al. 1999). While neurotoxic effects of MDMA on 5-HT neurons lead to 5-HT depletion, little is known about the effects of this depletion on postsynaptic 5-HT_2 receptors. Furthermore, since MDMA-induced 5-HT depletion may lead to impairment of functions in which 5-HT and 5-HT_2 receptors are involved (such as learning and mnemonic function), it is important to study the effects of MDMA on 5-HT_2 receptors and memory (Buhot et al. 1997). This is of particular interest, since several studies have found that recreational MDMA users display significant memory impairments, whereas their performance on other cognitive tests is generally normal (Krystal et al. 1992; Parrott et al. 1998).

Recent development of [^{123}I]-5-I-R91150, a radioligand with high affinity and selectivity for the 5-HT_{2A} receptor subtype, has made it possible to assess the density of postsynaptic 5-HT_{2A} receptors in the living human brain, using single photon emission computed tomography (SPECT). Cortical binding of [^{123}I]-5-I-R91150 for 5-HT_{2A} receptors is specific and reversible, as shown by inhibition of binding by ritanserin and displacement by ketanserin. Furthermore, the cortico-cerebellar ratios at pseudo-equilibrium reflect a distribution similar to that expected from post-mortem studies (Busatto et al. 1997).

The present pilot study was designed to investigate whether MDMA use leads to quantitative alterations in [^{123}I]-5-I-R91150 labelled post-synaptic 5-HT_{2A} receptors and related memory functions.

Five individuals with a history of MDMA use ["MDMA group"; mean age: 23.6 years (SD 5.3), men/women: 4/1, time since last dose: 4.6 months (range 2–11), lifetime number of tablets: 218 (50–500), mean education: 13 years (6)], and nine age-, and education-matched control subjects [mean age: 22.8 years (SD 2.9), men/women: 4/5, mean education: 15 years (5)] participated in the study. All MDMA subjects had used at least 50 tablets. The controls were healthy subjects with no

self-reported use of psychoactive drugs, including MDMA. Recruitment was through advertisements (local newspapers). Participants agreed to abstain from use of psychoactive drugs for at least 2 months before the study, and were asked to undergo urine drug screening (with an enzyme-multiplied immunoassay for amphetamines, barbiturates, benzodiazepine metabolites, cocaine and metabolite, opiates, and marijuana) before enrolment. After testing urine samples, exclusion criteria were: a positive drug screen; pregnancy; a severe medical or neuropsychiatric illness that precluded informed consent; claustrophobia; and neuropsychiatric disease in which 5-HT has been implicated. Written informed consent was obtained from all participants. The procedures used were approved by the local ethics board and have therefore been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki.

For SPECT scanning, the Strichmann Medical Equipment 810X tomographic system was used. The transaxial resolution of this camera is 7.6 mm full-width at half-maximum of a line source in air, while the axial resolution is 13.5 mm. Each acquisition consisted of at least 15 slices (acquired in a 128×128 matrix), 3 min per slice, and with a slice distance of 5 mm. The energy window was set at 135–190 keV. Subjects lay in the supine position with the head aligned in a parallel to the orbitomeatal line, and were positioned such that the scanning volume initially included the cerebellum. Acquisition was commenced 2 h after IV injection of approximately 140 MBq [^{123}I]-5-I-R91150 (radiolabelling as described by Busatto et al. 1997), a time when specific binding is maximal and stable for up to 8 h following injection. Analysis of scans was performed blind to subject status. For analysis of [^{123}I]-5-I-R91150 binding, a standard template with regions of interest (ROIs) was constructed manually from co-registered MR images. For positioning, we used these MR images as a guide. The template, including ROIs for the frontal, parietal, and occipital cortex, was placed on three consecutive SPECT slices. Additional templates were constructed with ROIs for the cerebellum and temporal cortex. Mean cortical signal densities were calculated (mean counts/pixel of frontal, parietal, temporal, occipital cortex). An investigator unaware of the participant's history performed ROI analysis. The uptake in the cerebellum, presumed free from 5-HT_{2A} receptors, was used as a reference for background radioactivity (non-specific binding+free ligand) (Busatto et al. 1997). Relative indices of "specific" binding are calculated as: "mean" ROI binding/cerebellar binding=5-HT_{2A} binding ratio.

Memory was assessed the day prior to SPECT imaging using the Rey Auditory Verbal Learning Test (RAVLT). The RAVLT is a verbal memory test. The immediate verbal memory comprised RAVLT logical memory, the delayed verbal memory incorporated RAVLT-recall, and RAVLT-recognition.

Overall, [^{123}I]-5-I-R91150 binding ratios were higher in the MDMA group than in controls, and reached statistical significance in the occipital cortex [mean 2.04 (SD 0.20) versus 1.74 (0.19), P<0.05 Mann-Whitney U-test Fig. 1], indicating an up-regulation of postsynaptic 5-HT_{2A} receptors.

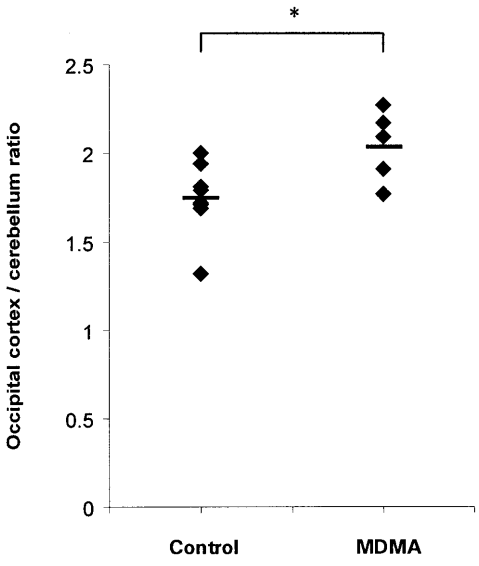

Fig. 1 The mean and individual [^{123}I]-5-I-R91150 binding ratio in the occipital cortex: controls versus MDMA users. Mean occipital binding ratios were calculated as occipital binding/binding in the cerebellum. *Statistical significant difference in binding ratio between controls and MDMA users

In the MDMA group, a significant lower number of recalled words in the RAVLT-recall was observed than in controls [8.14 (3.4) versus 12.3 (1.8), P<0.001; Fig. 2]. In the MDMA group, but not in controls, mean cortical 5-HT_{2A} receptor binding highly correlated (Spearman's rho) with recall [r=–0.98 (P=0.005), r=–0.29 (P=0.46), respectively; Fig. 2]. Age, sex, extent of previous MDMA use, and education had no significant effect on this correlation (P=0.28, 0.17, 0.25, and 0.16, respectively).

The high 5-HT_{2A} receptor binding in the occipital cortex in the MDMA group may be caused by 5-HT depletion. It is known that severe 5-HT depletion causes up-regulation of 5-HT_2 receptors (Heal et al. 1985). Moreover, MDMA-treated monkeys showed most severe 5-HT depletion in the occipital cortex. In these monkeys, 14 months after MDMA administration 5-HT levels were still reduced in the occipital cortex by 97% (Scheffel et al. 1998). In a recent SPECT study, MDMA users showed a significant reduction only in occipital 5-HT neurons (Semple et al. 1999). Thus, the presently observed up-regulation of 5-HT_{2A} receptors in the occipital cortex may reflect MDMA-induced brain 5-HT neurotoxicity.

In the present study, MDMA users showed significant deficits in delayed memory tasks, consistent with reports of memory problems in previous studies (Bolla et al. 1998; Parrott et al. 1998). Numerous laboratory studies with rats and monkeys have shown that MDMA produces serotonergic neurodegeneration. This has been demonstrated in various brain areas including the hippo-

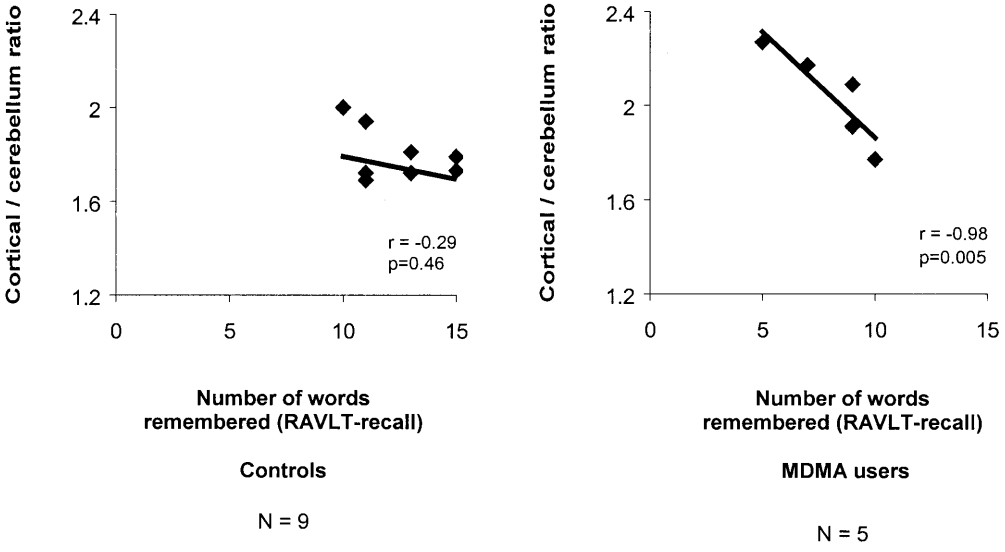

Fig. 2 Correlation between mean cortical [^{123}I]-5-I-R91150 binding ratios and memory function (number of words remembered on RAVLT recall trial) in control and MDMA group. SPECT data are expressed as cortical binding over cerebellar binding

campus, which is important for memory functioning (Hatzidimitriou et al. 1999). There is also clinical evidence for 5-HT brain damage in humans (Squier et al. 1995; McCann et al. 1998; Semple et al. 1999). There is therefore consistent evidence that memory deficits found in the present study may at least be attributed to MDMA-induced 5-HT deficits, particularly since high densities of 5-HT$_{2A}$ receptors (an indirect measure of 5-HT depletion) were associated with lower performance on the delayed memory tests. This provisional finding is in agreement with a recent study which showed that the extent of memory impairment correlated with the reduction of brain 5-HT, as indexed by CSF 5-HIAA (Bolla et al. 1998). We observed in this study that only individuals with apparent higher densities of occipital 5-HT$_{2A}$ receptors (presumably reflecting a greater extent of 5-HT injury) demonstrated detectable difficulties with memory function.

All participants in the MDMA group in our study reported that they had abstained from use of MDMA or other psychoactive drugs for at least 2 months before the study. Although most of the MDMA users had experimented with other recreational drugs (mainly alcohol and cannabis), none was a known 5-HT neurotoxin in human beings, and was therefore not likely to account for changes in [^{123}I]-5-I-R91150 binding to 5-HT$_{2A}$ receptors.

This pilot study was performed using small samples. Nevertheless, this study at least suggests an intriguing relationship between 5-HT$_{2A}$ receptor densities and memory performance in MDMA users. Because of the small sample size, the observed correlation cannot be said to be a definitive finding, and future studies investigating 5-HT neurotransmission and memory performance in MDMA users need to be conducted.

In conclusion, we provide additional evidence suggesting that human MDMA users are susceptible to MDMA-induced brain 5-HT neuronal injury and related functional disturbances, by showing a correlation between 5-HT$_2$ receptor densities and memory disturbances. Thus, reductions in 5-HT, as indexed by elevated cortical 5-HT$_2$ receptor densities, may be responsible for decrements in abstinent MDMA users.

References

Bolla KI, McCann UD, Ricaurte GA (1998) Memory impairment in abstinent MDMA ("Ecstasy") users. Neurology 51:1532–1537

Buhot MC (1997) Serotonin receptors in cognitive behaviors. Curr Opin Neurobiol 7:243–254

Busatto GF, Pilowsky LS (1997) Initial evaluation of ^{123}I-5-R91150, a selective 5-HT$_{2A}$ ligand for single-photon emission tomography, in healthy human subjects. Eur J Nucl Med 24: 119–124

Hatzidimitriou G, McCann UD, Ricaurte GA (1999) Altered serotonin innervation patterns in the forebrain of monkeys treated with (±)3,4-methylenedioxymethamphetamine seven years previously; factors influencing abnormal recovery. J Neurosci 19:5096–5107

Heal DJ, Philpot K, Molyneux SG, Metz (1985) Intracerebroventricular administration of 5,7-dihydroxytryptamine to mice increases both head-twitch response and the number of cortical 5-HT$_2$ receptors. Neuropharmacology 24:1201–1205

Krystal JH, Price LH, Opsahl C, Ricaurte GA, Heniger GR (1992) Chronic 3,4-methylenedioxymethamphetamine (MDMA) use: effects on mood and neuropsychological function? Am J Drug Alcohol Abuse 18:331–341

McCann UD, Szabo Z, Scheffel U, Dannals RF, Ricaurte GA (1998) Positron emission tomographic evidence of toxic effect of MDMA ("Ecstasy") on brain serotonin neurons in human beings. Lancet 352:1433–1437

Parrott AC, Lees A, Garnham NJ, Jones M, Wesnes K (1998) Cognitive performance in recreational users of MDMA or "ecstasy": evidence for memory deficits. J Psychopharmacol 12: 79–83

Scheffel U, Szabo Z, Mathews WB, Finley PA, Dannals RF, Ravert HT, Szabo K, Yuan J, Ricaurte GA (1998) In vivo detection of short- and long term MDMA neurotoxicity-A positron emission tomography study in the living primate brain. Synapse 29:183–92

Semple DM, Ebmeier KP, Glabus MF, O'Carroll RE, Johnstone EC (1999) Reduced in vivo binding to serotonin transporters in the cerebral cortex of MDMA ("ecstasy") users. Br J Psychiatry 175:63–69

Squier MV, Jalloh S, Hilton-Jones D, Series H (1995) Death after ecstasy ingestion: neuropathological findings. J Neurol Neurosurg Psychiatry 58:756