

Hypothalamic–pituitary–adrenal axis responses to stress in subjects with 3,4-methylenedioxy-methamphetamine (‘ecstasy’) use history: correlation with dopamine receptor sensitivity

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Abstract

Fifteen 3,4-methylenedioxymethamphetamine (MDMA, ‘ecstasy’) users who did not have other drug dependencies or prolonged alcohol abuse and 15 control subjects were studied. All the subjects were exposed to the same psychosocial stressor (Stroop Color–Word Interference Task, public speaking and mental arithmetic in front of an audience) 3 weeks after MDMA discontinuation. Plasma concentrations of adrenocorticotrophic hormone (ACTH) and cortisol were measured immediately before the tests began and at their end, 30 min later. Growth hormone (GH) responses to the dopaminergic agonist bromocriptine and psychometric measures (Tridimensional Personality Questionnaire, Minnesota Multiphasic Personality Inventory, Buss–Durkee Hostility Inventory) were also obtained 4 weeks after MDMA discontinuation for the same subjects. ACTH and cortisol basal levels were significantly higher in ecstasy users than in control subjects. In contrast, ACTH and cortisol responses to stress were significantly blunted in MDMA users. The sensitivity of dopamine D2 receptors, reflected by GH responses to bromocriptine challenge, was reduced in MDMA users compared with controls. The responsiveness of the hypothalamic–pituitary–adrenal (HPA) axis (ACTH and cortisol delta peaks) correlated directly with GH areas under curves in response to bromocriptine, and inversely with psychometric measures of aggressiveness and novelty seeking. No correlation was found between hormonal measures and the extent of MDMA exposure. Reduced D2 receptor sensitivity, HPA basal hyperactivation and reduced responsiveness to stress may represent a complex neuroendocrine dysfunction associated with MDMA use. The present findings do not exclude the possibility that dopamine dysfunction partly predated MDMA exposure.

Keywords: Substance abuse; Cortisol; Growth hormone; Bromocriptine; Dopamine; Adrenocorticotrophic hormone

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1. Introduction

The relationship between the function of the hypothalamic–pituitary–adrenal (HPA) axis and substance use disorders in humans is extremely complex, particularly in the area of stimulant addictions (Goeders, 2002). It is not clear whether the changes in HPA axis function observed in stimulant users are a consequence of long-lasting exposure to drugs or reflect a preexisting condition that contributes to vulnerability to addictive behavior (Majewska, 2002).

Our previous findings, obtained in a human behavioral laboratory, showed high basal levels of cortisol and adrenocorticotrophic hormone (ACTH) in users of 3,4-methylenedioxymethamphetamine (MDMA, ‘ecstasy’), but low responses to experimental aggressiveness, in comparison with findings in normal subjects (Gerra et al., 2001). MDMA has been demonstrated to increase cortisol (Mas et al., 1999) and ACTH plasma levels in humans (Grob et al., 1996), possibly being directly responsible for the changes in HPA axis function seen in our subjects.

Otherwise, HPA axis hyperactivity and reduced reactivity to stressful stimuli in MDMA users may reflect the biological correlates of depressive symptoms (Drevets et al., 1997) that have been found to be associated with serotonin system impairment in these subjects (Parrott, 2001; Gerra et al., 1998b, 2000).

Moreover, dopamine function impairment, recently observed in MDMA users (Gerra et al., 2002), may be hypothesized to be related to HPA axis dysfunction. The dopamine agonist apomorphine was reported to significantly increase ACTH and cortisol levels (Mokrani et al., 1995), suggesting possible control by the dopamine system of the reactivity of stress hormones. Subsensitivity of the HPA axis has also been found in relationship to dopamine system derangement in children with attention deficit hyperactivity disorder (Kariyawasam et al., 2002).

For these reasons, we decided to investigate the possible relationship between HPA responses to experimental psychological stress and D2 receptor sensitivity in subjects with a history of MDMA use. The aim of the study was to better understand

the complex neuroendocrine dysfunction that may underlie impaired coping with psychological stress in subjects exposed to stimulant drugs. A reduced sensitivity of dopamine receptors was found in MDMA users included in our previous studies (Gerra et al., 2002), but a direct capacity of MDMA to impair dopaminergic system in humans was not definitely demonstrated.

The hypothesis of the present study was that reduced hormonal responses to stressful stimuli in MDMA users could be related to a disturbance in brain monoamines, including dopamine dysfunction, as a neurobiological pattern affecting the reward system threshold and coping capacity. ACTH and cortisol responses to a mixed model of psychological stress were investigated in MDMA users, 3 weeks after drug discontinuation, simultaneously evaluating dopamine receptor sensitivity with a specific D2 receptor agonist, bromocriptine (BROM). Personality and temperamental measures were also evaluated with psychometric tests in all participants.

2. Methods

2.1. Subjects

Fifteen male MDMA users, aged 20–30 years (mean $\pm$ S.D. = 23.8 ± 3.5 years), with a history of at least 25 occasions of drug use (mean $\pm$ S.D. = 58.9 ± 29.5 ; range = 25–82) before drug interruption, entered the study. All the subjects gave informed consent. The subjects had used MDMA only on the weekend, one to three pills every night. In the Italian illicit market, the variability of MDMA concentrations ranges from 25 to 125 mg (Bellomo, 1995). The duration of MDMA use was from 6 to 34 months (mean $\pm$ S.D. = 20.5 ± 10). Table 1 presents demographic data and characteristics of MDMA use.

All the subjects were studied 3 weeks after their last dose of MDMA. In the month before MDMA discontinuation, urinary screens for amphetamines, methamphetamines, morphine, methadone, cannabis, cocaine, barbiturates and alcohol were performed three times a week. Screens were positive for MDMA, before discontinuation, in all MDMA users and were occasionally positive for cannabis

Table 1
Demographics and characteristics of MDMA use (means $\pm$ S.E.)

	Age (years)	Height (cm)	Weight (kg)
MDMA subjects	23.8 $\pm$ 3.5	171.9 $\pm$ 12.1	69.9 $\pm$ 11.4
Control subjects	22.9 $\pm$ 3.8	174.9 $\pm$ 14.3	76.8 $\pm$ 13.1
Number of exposures	58.9 $\pm$ 29.5		range 25–82
Duration of use	20.5 $\pm$ 10 months		range 6–34
Frequency of use	8.6 $\pm$ 4.6 per month		range 3.0–14.0

in four subjects. The use of other illicit drugs, apart from MDMA, was occasional: cannabis was found only in two urine screens in four subjects.

Previous prolonged consumption or dependence on other drugs of abuse and psychotropic agents, or continuous excessive alcohol intake was excluded on the basis of a structured interview. Many subjects were excluded after the first contact because of their previous long-lasting use of other drugs. Most of the subjects included in the study reported occasional use of cannabis and cocaine or abuse of alcohol (two or three times) in the last 6 months.

Information about MDMA and/or other drug use was obtained in the following ways: self-report during the first clinical evaluation; an MDMA questionnaire that asked about duration, frequency, doses and time of last dose; a questionnaire concerning other drugs and alcohol; a double interview with the family to correct for possible denial; and direct medical evaluation. Previous episodes of cannabis and cocaine use or alcohol abuse were truly 'occasional' in the subjects included in the study, with 4- to 6-week intervals between each episode.

The subjects had contacted one of the investigators at the Drug Addiction Service in Parma (Az. USL) seeking initial information about MDMA ($n=5$) or treatment ($n=6$); the remaining subjects ($n=4$) had contacted the center through a teacher or parent. Six subjects were students, five subjects were workers, and four were unemployed. Four of the six students showed academic underachievement. Eight of the 15 subjects were still living in their parents' home, and their socioeconomic status was medium or high. All MDMA users included in the study spent their weekend

nights in disco-clubs and manifested a preference for techno-music or progressive music.

The subjects were admitted to a long-term psychosocial rehabilitation program. If twice-weekly analyses for urine metabolites of the main substances of abuse excluded their consumption in the first 3 weeks after admission, they were included in the study.

Criteria for exclusion also included severe chronic liver (transaminases > 50 U/l and gamma-globulins $> 20\%$ g/dl) or renal (creatinine clearance: $100\text{--}120\text{ mg}^{-1}\text{ l}^{-1}\text{ min}^{-1}$) diseases or other chronic physical disorders, significant weight loss ($> 10\%$) or obesity, endocrinopathies and immunodeficiencies (the subjects were HIV-negative).

Fifteen healthy male volunteers, matched for age (19–30 years; mean $\pm$ S.D. = 22.9 ± 3.8 years) and demographic/cultural data, were recruited from the hospital staff and the high school of Parma as controls. The controls never used psychoactive drugs or abused alcohol; urine screens for 4 weeks and a double interview with the parents confirmed the self-report data. Exclusion criteria were the same as for the MDMA users.

2.2. Personality assessment

DSM-IV clinical evaluation and psychometric measures were performed 3 weeks after MDMA discontinuation and repeated after 12 months of abstinence from MDMA at the end of the study. Following the same schedule, control subjects were retested after 12 months. Axis I and II disorders were evaluated by a trained psychiatrist utilizing the Structured Clinical Interview (SCID) for Axis I disorders (Spitzer et al., 1990) (Italian Version: Intervista Clinica Strutturata per il DSM-III-R,

Organizzazioni Speciali, Florence) and the Structured Interview for DSM-IV Personality Disorders (SIDP) for Axis II disorders (Black et al., 1993) (Italian Version: Maggini and Piccini, draft edition, 1994).

The SCID was used to exclude Axis I disorders among MDMA users and controls. The evaluation with the SIDP for Axis II disorders identified five subjects with overt personality disorders out of 15; three subjects had borderline personality disorder, and two had avoidant personality disorder. Another two subjects showed symptoms partially corresponding to avoidant personality disorder criteria, and one subject to antisocial personality disorder, but the subjects did not show the complete clinical picture of this Axis II disorder. No Axis II disorders were found among the control subjects. Although major depression was not diagnosed among MDMA users, eight subjects showed dysphoria and mood changes in the weeks following MDMA discontinuation; seven subjects reported tiredness and fatigue; and four subjects had subtle cognitive impairment and episodes of confusion. Nine subjects among the MDMA users showed novelty-seeking behavior as a characteristic of their life style.

Personality was also investigated with the Minnesota Multiphasic Personality Inventory (MMPI 2; Hathaway and McKinley, 1989). This test was compared with the DSM structured interviews to investigate possible differences in personality analysis with the self-administered methods. The character and the degree of aggressiveness (defined as direct, indirect or verbal; irritability; negativism; resentment; suspiciousness; guilt; and total score) were assessed with the Buss–Durkee Hostility Inventory (BDHI; Buss and Durkee, 1957) (Italian version: ‘Questionario per la Tipizzazione della Aggressività’, QTA; Castrogiovanni et al., 1993). QTA raw scores, in accordance with Castrogiovanni, were used for the total score and for the individual subscale scores. Depression was monitored by the 21-item Hamilton Rating Scale for Depression (HRS-D; Hamilton, 1960). All the subjects were given the Tridimensional Personality Questionnaire (TPQ; Cloninger, 1987) to investigate temperamental aspects, particularly

‘harm-avoidant,’ ‘novelty-seeking,’ and ‘reward-dependent’ characteristics.

2.3. *Experimental stress procedure*

MDMA users underwent an experimental stress procedure 3 weeks after drug discontinuation. Control subjects were tested 4 weeks after they were contacted.

Physical training and other stressful events (e.g. school examinations) were carefully avoided in the 7 days preceding the tests. Food and drinks, except for water, were avoided from midday to 16.00 h of the day of the tests, which started at 14.00 h and lasted until 16.00 h. The subjects had a light lunch at 11.00 h, including a sandwich with ham and orange juice.

After arrival at the laboratory, each subject was taken into room A, and a catheter was inserted into an antecubital vein kept patent by saline infusion. The subject rested 30 min, after which blood was drawn for hormone assays (time 0). Previous evaluations of two basal blood samples, 30 min from one another, evidenced that the second baseline hormonal value was not influenced by i.v. insertion (Kirschbaum et al., 1993; Gerra et al., 1998a). Then he was transferred into room B and acquainted with the tasks to be performed. The Stroop Color-Word Interference Task (McCann et al., 1993) was presented for 10 min, during which the subject was asked to identify rapidly the colors in which words were printed (e.g. the word ‘red’ printed in the color green, the correct response being ‘green’) (time 10). After that, a mental arithmetic test (Kirschbaum et al., 1992) was given for another 10 min, during which the subject was asked to subtract numbers of three digits from numbers of four digits (such as 412 from 3215), as fast and as accurately as possible (time 20). On every mistake, the subject had to start over again from the first subtraction. At this point, he was taken into room C, where three persons were already sitting at a table with a video camera and a tape recorder. The subject was asked to stand at a microphone in front of the three persons and to speak for 10 min, introducing himself and describing his personality, his interpersonal relationships, behavior, life style, personal

projects and beliefs (Kirschbaum et al., 1993). Immediately after the third task (time 30), a second blood sample was drawn for hormonal assays.

Heart rate (HR) and systolic and diastolic blood pressure (SBP, DBP) were measured before the three tests began and after their completion.

Plasma concentrations of cortisol were measured with a competitive enzymatic immunoassay by commercial kits (AIA-PACK., Eurogenetics Italy, Torino, Italy). ACTH was measured with commercial kits (Medical System DPC—Immuline, Los Angeles, CA, USA). The respective intra-assay and inter-assay coefficients of variation were 3.7 and 7.5% for cortisol, and 6 and 10% for ACTH. Assay sensitivities were 0.3 nmol/l for cortisol and 15 pg/ml for ACTH.

2.4. Neuroendocrine challenge

Challenges with bromocriptine (a dopamine D2 receptor agonist) were performed 4 weeks after MDMA discontinuation. Controls were also tested with bromocriptine 5 weeks after they were contacted.

The bromocriptine challenge was done at 09.00 h with a catheter inserted into an antecubital vein, kept patent by saline infusion. At 10.00 h, 5 mg of bromocriptine (Parlodel, Sandoz, Italy) were administered orally. Blood specimens for growth hormone (GH) assays were drawn into EDTA-containing tubes immediately before the administration of bromocriptine, and 15, 30, 60, 90 and 120 min afterwards. Blood was immediately centrifuged, and plasma stored at -20°C until assayed. GH concentrations were measured by chemiluminescence with the commercial kits of Medical System (Italy). The sensitivity of the method for determining GH was 0.5 ng/ml; the inter-assay variability for GH was 7.3% and the intra-assay variability 4.9%.

2.5. Statistical analysis

A chi-square test was performed to assess the normality of distribution of the data: no ‘outliers’ were included in the results. The hormonal responses to stress and dopaminergic stimulus in

the groups of MDMA users and controls were analyzed statistically by one-way analysis of variance (ANOVA) and by two-way ANOVA for repeated measures to evaluate the effects of time, group and time $\times$ group. Analysis of covariance and Pearson correlations were also used, with Bonferroni correction of P values.

All the correlation analyses were performed in the groups of MDMA users and controls separately, and not combined, because the two groups did not have a univariate normal distribution. The correlations of ACTH and cortisol responses with the measures of dopamine D2 receptor sensitivity (GH areas under curves—AUCs) were investigated using the Pearson test.

The correlations of psychometric measures with endocrine responses (ACTH delta peaks, cortisol delta-peaks, and GH AUCs) were also evaluated by Pearson analysis. Among the psychometric variables direct aggressiveness on the BDHI, depression on the D subscale of the MMPI and on the HRS-D, and novelty-seeking on the TPQ were specifically correlated with hormonal findings.

The AUCs were calculated with the Sibson Rule method: AUCs had positive values when over the basal levels ($T=0$) and negative values when under the basal levels ($T=0$).

The responses to the stress and neuroendocrine challenges (delta peaks and AUCs) were correlated with time and number of exposures to MDMA (use extent).

3. Results

Table 2 presents psychological data (mean score $\pm$ S.E.) obtained from the MMPI II, BDHI, HRS-D, and TPQ at 3 weeks of abstinence. MDMA users showed significantly higher scores than control subjects on the MMPI subscale D ($t=3.61$; d.f. = 1, 28; $P<0.005$). No other significant difference between MDMA users and controls was found for the MMPI.

In comparison with control subjects, MDMA users showed significantly higher scores on the BDHI direct aggressiveness subscale ($t=5.18$; d.f. = 1, 28; $P<0.001$), on the BDHI guilt subscale ($t=5.19$; d.f. = 1, 28; $P<0.001$), on the HRS-D ($t=4.22$; d.f. = 1, 28; $P<0.001$) and on the nov-

Table 2

Psychometric measures in MDMA users and control subjects including the depression scale D of the MMPI, the direct and guilt subscales of the Buss–Durkee Hostility Inventory (BDHI), the Hamilton Rating Scale for Depression (HRS-D), and the novelty-seeking (NS) subscale of the Three-Dimensional Personality Questionnaire (TPQ)

	MMPI D	BDHI direct	BDHI guilt	HRS-D	TPQ NS
MDMA users	60.0 ± 3.0	58.1 ± 2.6	59.33 ± 1.9	12.6 ± 2.3	20.4 ± 1.8
Control subjects	46.1 ± 2.3	41.2 ± 1.8	43.7 ± 2.2	4.1 ± 1.5	11.0 ± 1.2
d.f. = 1, 28	$t = 3.61$	$t = 5.18$	$t = 5.19$	$t = 4.22$	$t = 4.351$
	$P < 0.005$	$P < 0.001$	$P < 0.001$	$P < 0.001$	$P < 0.001$

elty-seeking TPQ subscale ($t = 4.35$; d.f. = 1, 28; $P < 0.001$).

3.1. Stress response: hormonal findings

Mean basal levels of ACTH and cortisol were significantly higher in MDMA users than in controls (ACTH: 27.33 ± 1.38 pg/ml in MDMA users vs. 19.00 ± 1.7 pg/ml in controls; cortisol: 337.49 ± 18.28 pg/ml in MDMA users vs. 276.75 ± 12.60 pg/ml in controls) ($t = 3.6$; d.f. = 1, 28; $P < 0.001$; $F = 2.82$; d.f. = 1, 28; $P < 0.005$).

As shown in Fig. 1, ACTH concentrations increased significantly more after the stimulus in control subjects than in MDMA users. ANOVA for repeated measures revealed significant effects of group ($F = 4.6$; d.f. = 1, 28; $P < 0.05$), time ($F = 37.9$; d.f. = 1, 28; $P < 0.001$), and time by group

($F = 19.22$; d.f. = 1, 28; $P < 0.001$). ACTH delta peak values were significantly higher in controls than in MDMA users (MDMA users = 2.53 ± 2.127 vs. controls = 23.05 ± 3.71 ; $t = 3.82$; d.f. = 1, 28; $P < 0.001$).

Cortisol concentrations increased significantly after the stimulus (Fig. 2) in control subjects, an effect that was more pronounced than in MDMA users. ANOVA for repeated measures revealed significant effects of group ($F = 5.6$; d.f. = 1, 28; $P < 0.05$), time ($F = 16.59$; d.f. = 1, 28; $P < 0.001$), and time by group ($F = 13.37$; d.f. = 1, 28; $P < 0.001$). Cortisol delta peak values were significantly higher in control subjects than in MDMA users (MDMA users = 11.38 ± 31.68 vs. controls = 225.76 ± 42.28 ; $t = 5.83$; d.f. = 1, 28; $P < 0.001$).

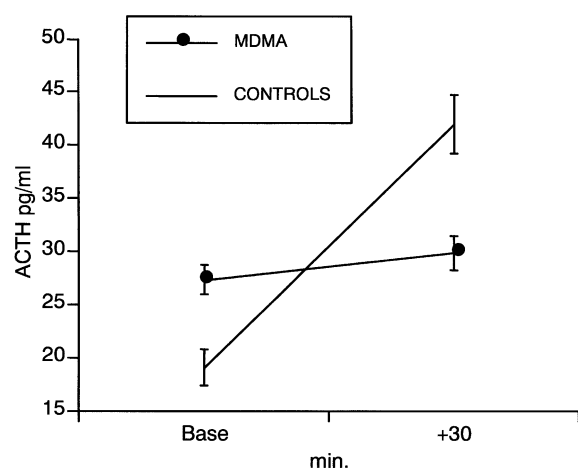


Fig. 1. ACTH responses during experimentally induced psychological stress session (mean ± S.E.) in MDMA users (●—●) and control subjects (○---○).

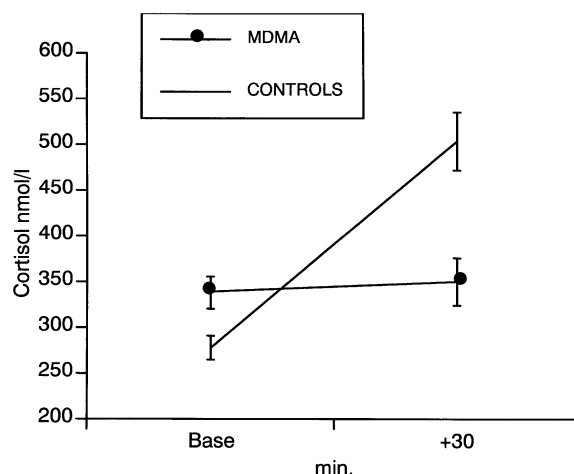


Fig. 2. Cortisol responses during experimentally induced psychological stress session (mean ± S.E.) in MDMA users (●—●) and control subjects (○---○).

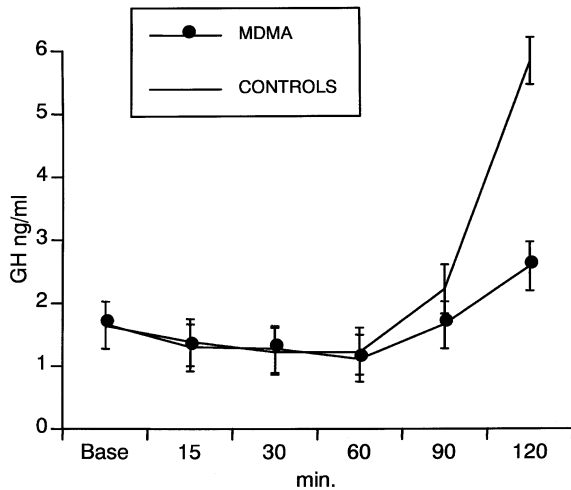


Fig. 3. Growth hormone (GH) responses during experimentally induced psychological stress session (mean $\pm$ S.E.) in MDMA users (●—●) and control subjects (—○—).

ACTH and cortisol values of the MDMA users who were previously exposed to occasional use of cannabis or cocaine and alcohol abuse were scattered throughout the 15 sets of values, and not concentrated at the high or the low end of the range (chi-square), suggesting that the effect of other pharmacological substances was not significant.

3.2. Bromocriptine challenge: hormonal findings

No significant differences were found between basal values of GH in MDMA users and controls (MDMA users: 1.66 ± 0.4 ng/ml; controls: 1.64 ± 0.3) ($t=0.09$; d.f. = 1, 28; $P=0.46$). In the comparison between the GH responses to bromocriptine of MDMA users and control subjects, ANOVA for repeated measures showed significant effects of time ($F=4.39$; d.f. = 5, 28; $P<0.05$), of group ($F=35.99$, d.f. = 1, 28; $P<0.001$) and group by time ($F=11.58$, d.f. = 5, 28; $P<0.001$; Fig. 3).

3.3. Correlations

Pearson's analysis revealed that GH values after bromocriptine were directly correlated with ACTH

delta peaks ($r=0.54$, $P<0.01$) and cortisol delta peaks ($r=0.63$, $P<0.001$) in response to stressful stimuli in MDMA users. ACTH delta peaks inversely correlated with direct aggressiveness scores on the BDHI ($r=-0.49$, $P<0.05$) and with novelty-seeking scores on the TPQ ($r=-0.51$, $P<0.01$) in MDMA abusers. The same was true for cortisol delta peaks (BDHI: $r=-0.56$, $P<0.005$; novelty seeking: $r=-0.64$, $P<0.001$).

Pearson's analysis revealed that GH values after bromocriptine were inversely correlated with novelty-seeking scores on the TPQ both in control subjects ($r=-0.45$, $P<0.05$) and MDMA users ($r=-0.53$, $P<0.005$). No correlation was found between GH AUCs and HRS-D, MMPI and BDHI subscale scores. GH AUCs and ACTH-cortisol delta peaks did not correlate with the measures of extent of MDMA exposure in the present sample of MDMA users.

4. Discussion

Higher basal levels of ACTH and cortisol were observed in MDMA users than in controls, as well as a blunted HPA axis response to psychological stress, in agreement with our previous data obtained during experimental aggressiveness sessions (Gerra et al., 2001). Accordingly, diminished activation of the HPA axis was found in recently detoxified cocaine addicts (Wilkins et al., 1997; Majewska and Wilkins, 2000), particularly among those who relapsed almost immediately.

High basal levels of ACTH and cortisol in MDMA users may reflect a variety of factors. Possibly increased worry and the perception of the challenge as more stressful by MDMA users in comparison with controls may explain the higher basal levels of stress hormones as an anticipatory reaction (Gerra et al., 1998a). Higher cortisol basal levels with lower responsiveness to stress may also represent the neuroendocrine pattern reported in depressed adolescents at risk for substance abuse: the HPA axis was found to be active when the system is normally quiescent and unable to express any response during coping processes with stressful conditions (Rao et al., 1999).

Alternatively, increased basal levels of cortisol and ACTH could be attributable to a direct pharmacological action of MDMA (Grob et al., 1996; Mas et al., 1999) that may exhaust HPA axis responsiveness, but the lack of a significant correlation between hormonal data and the extent of MDMA exposure seems incompatible with this explanation. Furthermore, the inverse correlations between ACTH and cortisol responses and the measures of aggressiveness and novelty-seeking temperament, evidenced in our experiment, support the hypothesis that HPA dysfunction may be influenced more by personality traits than by MDMA exposure.

Reduced D2 receptor sensitivity to the dopamine agonist bromocriptine was not unexpected in MDMA users, because of similar results obtained in a different sample of subjects with a history of MDMA use included in our previous studies (Gerra et al., 2002). The lack of GH response to bromocriptine, evidenced in MDMA users, once again suggests a possible involvement of the dopamine system in MDMA action. The capacity of MDMA to partially affect the dopamine system was also suggested by electroencephalographic (EEG) findings. Comparison of the MDMA-specific EEG pattern with that of various serotonin, dopamine and noradrenaline agonists indicates that serotonin, noradrenaline and, to a lesser degree, dopamine contribute to the effects of MDMA on the EEG, and possibly also on mood and behavior (Frei et al., 2001). Accordingly, pretreatment with the dopamine D2 antagonist haloperidol has been reported to reduce MDMA-induced positive mood (Liechti and Vollenweider, 2000; Liechti et al., 2001), indicating a possible action of MDMA on the function of the dopaminergic system in humans. More recently, Ricaurte et al. (2002) demonstrated that non-human primates exposed to several sequential doses of MDMA, a regimen modeled after one used by humans, developed severe brain dopaminergic neurotoxicity, in addition to less pronounced serotonergic neurotoxicity.

The correlation between reduced D2 receptor sensitivity and impaired HPA axis responses to stress could simply reflect the effects of MDMA in influencing both brain monoamine function and ACTH-cortisol reactivity (Mayerhofer et al.,

2001). On the other side, the subjects with high basal levels of ACTH and cortisol in association with low reactivity to stress were characterized by monotony avoidance, type-A behavior (competitive, rushed, physically aggressive) and mastery (Grossi et al., 1998), all personality traits reflecting a dopamine dysfunction as described in the reward deficiency syndrome (Blum et al., 2000). Accordingly, no change in GH after bromocriptine administration was found to correspond to psychosocial stress by alexithymic and overadaptive behaviors (Okuse and Anzai, 1992) that could have affected MDMA users before MDMA exposure.

The impairment of D2 receptor sensitivity, together with HPA basal hyperactivation and reduced responsiveness to stress, may represent a complex neuroendocrine dysfunction associated with stimulant use. Impaired dopamine function and impaired reward circuitry, possibly connected to changes in dopamine transporter density (Volkow et al., 1999), may underlie the perception of fewer rewards from usually interesting activities and a generalized underarousal in response to stress, as an individual attitude, also including HPA axis dysfunction (Kariyawasam et al., 2002).

In the same way, high dopamine turnover with high levels of homovanillic acid appears to be linked to substance abuse and antisocial behavior in males with a positive history for family substance abuse (Gabel et al., 1995), maladaptation to stress, reward delay and risk-taking behavior preexisting substance use.

Finally, dopamine system changes and HPA impairment in MDMA users might be due to the serotonergic dysfunction induced by prolonged exposure to MDMA. Brain serotonin interacts with dopamine function in a complex fashion. Increased 5-HT activity in the nucleus accumbens was found to inhibit dopamine-dependent behavior, indicating that the activation of serotonin receptors is particularly important in dopamine regulation (Fletcher et al., 2002). The striatal serotonin depletion in MDMA users (Kish et al., 2000) could also be responsible for the defective serotonergic control of the pituitary–adrenal axis that has been reported in other pathological conditions in humans (Volpi et al., 1997).

Although our data are preliminary and need to be interpreted with caution because of the small number of subjects, the evidence of HPA axis and dopamine system dysfunction in human subjects with a history of MDMA use, as well as the association between behavioral and biological changes, indicate that MDMA may be implicated in a complex psychobiological disturbance. Our preliminary findings suggest the need for further studies designed to investigate the effects of MDMA on human brain in relationship to a possible neuroendocrine substrate partly preexisting stimulant exposure.

References

- Bellomo, R.D., 1995. Aspetti di polizia scientifica. Atti del Seminario Studi sui Derivati Amfetaminici. Italian Ministry of Interior, Rome, pp. 43–48.
- Black, D.W., Noyes Jr, R., Pfohl, B., Goldstein, R.B., Blum, N., 1993. Personality disorder in obsessive-compulsive volunteers, well comparison subjects, and their first-degree relatives. *American Journal of Psychiatry* 150, 1226–1232.
- Blum, K., Braverman, E.R., Holder, J.M., Lubar, J.F., Monastera, V.J., Miller, D., Lubar, J.O., Chen, T.J., Comings, D.E., 2000. Reward deficiency syndrome: a biogenetic model for the diagnosis and treatment of impulsive, addictive, and compulsive behaviors. *Journal of Psychoactive Drugs* 32 (Suppl. i–iv), 1–112.
- Buss, A.H., Durkee, A., 1957. An inventory for assessing different kinds of hostility. *Journal of Consulting Psychology* 21, 343–349.
- Castrogiovanni, P., Maremmi, I., Di Muro, A., 1993. Aggressive behaviour and hostility in depression: clinical aspects. In: Placidi, G.E., Dell’Osso, L., Nisticò, G., Akiskal, H.S. (Eds.), *Recurrent Mood Disorders*. Springer-Verlag, Berlin Heidelberg, pp. 51–65.
- Cloninger, C.R., 1987. A systematic method for clinical description and classification of personality variants. *Archives of General Psychiatry* 44, 573–588.
- Drevets, W.C., Price, J.L., Simpson Jr, J.R., Todd, R.D., Reich, T., Vannier, M., Raichle, M.E., 1997. Subgenual prefrontal cortex abnormalities in mood disorders. *Nature* 386 (6627), 824–827.
- Fletcher, P.J., Azampanah, A., Korth, K.M., 2002. Activation of 5-HT(1B) receptors in the nucleus accumbens reduces self-administration of amphetamine on a progressive ratio schedule. *Pharmacology, Biochemistry and Behaviour* 71 (4), 717–725.
- Frei, E., Gamma, A., Pascual-Marqui, R., Lehmann, D., Hell, D., Vollenweider, F.X., 2001. Localization of MDMA-induced brain activity in healthy volunteers using low resolution brain electromagnetic tomography (LORETA). *Human Brain Mapping* 14 (3), 152–165.
- Gabel, S., Stadler, J., Bjorn, J., Shindledacker, R., 1995. Homovanillic acid and dopamine-beta-hydroxylase in male youth: relationships with paternal substance abuse and antisocial behavior. *American Journal of Drug and Alcohol Abuse* 21 (3), 363–378.
- Gerra, G., Zaimovic, A., Chittolini, B., Giucastro, G., Folli, F., Maestri, D., Tesson, A., Avanzini, P., Caccavari, R., Bernasconi, S., Brambilla, F., 1998a. Neurotransmitter-hormonal responses to psychological stress in peripubertal subjects: relationship to aggressive behavior. *Life Sciences* 62 (7), 617–625.
- Gerra, G., Zaimovic, A., Giucastro, G., Maestri, M., Monica, C., Sartori, R., Caccavari, R., Delsignore, R., 1998b. Serotonergic function after ($\pm$)3, 4-methylene-dioxy-methamphetamine (MDMA, ‘ecstasy’) in humans. *International Clinical Psychopharmacology* 13, 1–9.
- Gerra, G., Zaimovic, A., Zambelli, U., Timpano, M., Neri, E., Marzocchi, G.F., Delsignore, R., Brambilla, F., 2000. Long-lasting effects of $\pm$ (3,4-methylenedioxy-methamphetamine (ecstasy) on serotonin system function in humans. *Biological Psychiatry* 47, 127–136.
- Gerra, G., Zaimovic, A., Giusti, F., Delsignore, R., Raggi, M.A., Laviola, G., Macchia, T., Brambilla, F., 2001. Experimentally-induced aggressive behaviour in subjects with 3,4-methylenedioxy-methamphetamine (MDMA; ‘ecstasy’) use history; psychobiological correlates. *Journal of Substance Abuse* 13, 471–491.
- Gerra, G., Zaimovic, A., Moi, G., Giusti, F., Gardini, S., Delsignore, R., Laviola, G., Macchia, T., Brambilla, F., 2002. Effects of ($\pm$)3,4-methylene-dioxy-methamphetamine (ecstasy) on dopamine system function in humans. *Behavioural Brain Research* 134, 403–410.
- Goeders, N.E., 2002. The HPA axis and cocaine reinforcement. *Psychoneuroendocrinology* 27 (1–2), 13–33.
- Grob, C.S., Poland, R.E., Chang, L., Ernst, T., 1996. Psychobiologic effects of 3,4-methylenedioxy-methamphetamine in humans: methodological considerations and preliminary observations. *Behavioral Brain Research* 73 (1–2), 103–107.
- Grossi, G., Ahs, A., Lundberg, U., 1998. Psychological correlates of salivary cortisol secretion among unemployed men and women. *Integrative Physiological Behavioral Science* 33 (3), 249–263.
- Hamilton, M., 1960. A rating scale for depression. *Journal of Neurology, Neurosurgery and Psychiatry* 23, 56–62.
- Hathaway, S.R., McKinley, J.C., 1989. *Minnesota Multiphasic Personality Inventory-2. Manual*, University of Minnesota, Minneapolis.
- Kariyawasam, S.H., Zaw, F., Handley, S.L., 2002. Reduced salivary cortisol in children with comorbid attention deficit hyperactivity disorder and oppositional defiant disorder. *Neuroendocrinology Letters* 23 (1), 45–48.
- Kirschbaum, C., Diedrich, O., Gehrke, J., Wüst, S., Hellhammer, D.H., 1992. Cortisol and behavior. ‘The Trier challenge test (TMCT)’: first evaluation of a new psychological stress test. In: Ehlers, A., Fiegenbaum, W., Florin, I., Margraf, J. (Eds.), *Perspectives and Promises of Clinical Psychology*. Plenum, New York, pp. 67–75.

- Kirschbaum, C., Pirke, K.M., Hellhammer, D.H., 1993. The 'Trier Social Stress Test'—a tool for investigating psychological stress responses in a laboratory setting. *Neuropsychobiology* 28, 76–88.
- Kish, S.J., Furukawa, Y., Ang, L., Vorce, S.P., Kalasinsky, K.S., 2000. Striatal serotonin is depleted in brain of a human MDMA (ecstasy) user. *Neurology* 55 (2), 294–296.
- Liechti, M.E., Geyer, M.A., Hell, D., Vollenweider, F.X., 2001. Effects of MDMA (ecstasy) on prepulse inhibition and habituation of startle in humans after pretreatment with citalopram, haloperidol or ketanserin. *Neuropsychopharmacology* 24 (3), 240–252.
- Liechti, M.E., Vollenweider, F.X., 2000. Acute psychological and physiological effects of MDMA ('ecstasy') after haloperidol pretreatment in healthy humans. *European Neuropsychopharmacology* 10 (4), 289–295.
- Majewska, M.D., Wilkins, J., 2000. Low plasma levels of DHEAS as markers for refractory cocaine dependence. *Neuropsychopharmacology* 23, S46 [Abstract].
- Majewska, M.D., 2002. HPA axis and stimulant dependence: an enigmatic relationship. *Psychoneuroendocrinology* 27 (1–2), 5–12.
- Mas, M., Farre, M., de la Torre, R., Roset, P.N., Ortuno, J., Segura, J., Cami, J., 1999. Cardiovascular and neuroendocrine effects and pharmacokinetics of 3,4-methylenedioxy-methamphetamine in humans. *Journal of Pharmacology and Experimental Therapeutics* 290 (1), 136–145.
- Mayerhofer, A., Kovar, K.A., Schmidt, W.J., 2001. Changes in serotonin, dopamine and noradrenaline levels in striatum and nucleus accumbens after repeated administration of the abused drug MDMA in rats. *Neuroscience Letters* 308 (2), 99–102.
- McCann, B.S., Carter, J., Vaughan, M., Raskind, M., Wilkinson, C.W., Veith, R.C., 1993. Cardiovascular and neuroendocrine responses to extended laboratory challenge. *Psychosomatic Medicine* 55, 497–504.
- Mokrani, M.C., Duval, F., Crocq, M.A., Bailey, P.E., Macher, J.P., 1995. Multihormonal responses to apomorphine in mental illness. *Psychoneuroendocrinology* 20 (4), 365–375.
- Okuse, S., Anzai, T., 1992. Psychosomatic aspects of stress. *Rinsho Byori* 40 (3), 234–240.
- Parrott, A.C., 2001. Human psychopharmacology of ecstasy (MDMA): a review of 15 years of empirical research. *Human Psychopharmacology* 16 (8), 557–577.
- Rao, U., Ryan, N.D., Dahl, R.E., Birmaher, B., Rao, R., Williamson, D.E., Perel, J.M., 1999. Factors associated with the development of substance use disorder in depressed adolescents. *Journal of the American Academy of Child and Adolescent Psychiatry* 38 (9), 1109–1117.
- Ricaurte, G.A., Yuan, J., Hatzidimitriou, G., Cord, B.J., McCann, U.D., 2002. Severe dopaminergic neurotoxicity in primates after a common recreational dose regimen of MDMA ('ecstasy'). *Science* 297 (5590), 2185–2187.
- Spitzer, R.L., Williams, J.B.W., Gibbon, M., First, M.B. 1990, Structured Clinical Interview for DSM-III-R First published by American Psychiatric Press, Washington D.C. and London, England.
- Volkow, N.D., Fowler, J.S., Wang, G.J., 1999. Imaging studies on the role of dopamine in cocaine reinforcement and addiction in humans. *Journal of Psychopharmacology* 13 (4), 337–345.
- Volpi, R., Caffarra, P., Scaglioni, A., Boni, S., Saginario, A., Chiodera, P., Coiro, V., 1997. Defective 5-HT₁-receptor-mediated neurotransmission in the control of growth hormone secretion in Parkinson's disease. *Neuropsychobiology* 35 (2), 79–83.
- Wilkins, J., Van Gorp, W., Hinken, C., Welch, B., Wheatley, S., Plotkin, D., Moore, L., Setoda, D., Ashofteh, A., Majewska, M.D., 1997. Relapse to cocaine use may be predicted in early abstinence by low plasma dehydroepiandrosterone. *NIDA Research Monograph* 174, 183.