

Sex Differences in Striatal Dopamine Release in Healthy Adults

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Background: Sex differences in addictive disorders have been described. Preclinical studies have implicated the striatal dopamine system in these differences, but human studies have yet to substantiate these findings.

Methods: Using positron emission tomography (PET) scans with high-specific-activity [^{11}C] raclopride and a reference tissue approach, we compared baseline striatal dopamine binding potential (BP) and dopamine release in men and women following amphetamine and placebo challenges. Subjective drug effects and plasma cortisol and growth hormone responses were also examined.

Results: Although there was no sex difference in baseline BP, men had markedly greater dopamine release than women in the ventral striatum. Secondary analyses indicated that men also had greater dopamine release in three of four additional striatal regions. Paralleling the PET findings, men's ratings of the positive effects of amphetamine were greater than women's. We found no sex difference in neuroendocrine hormone responses.

Conclusions: We report for the first time a sex difference in dopamine release in humans. The robust dopamine release in men could account for increased vulnerability to stimulant use disorders and methamphetamine toxicity. Our findings indicate that future studies should control for sex and may have implications for the interpretation of sex differences in other illnesses involving the striatum.

Key Words: Addiction, amphetamine, binding potential, dopamine release, gender, sex differences, striatum

Men and women differ in their vulnerability to addictive disorders (Brady and Randall 1999; Brecht et al 2004). Sex differences in the prevalence of psychostimulant drug dependence in general, and in methamphetamine use in particular, have been identified (Brady & Randall, 1999; Brecht et al 2004; Substance Abuse and Mental Health Services Administration 2005). Moreover, men compared with women are more susceptible to methamphetamine toxicity (Dluzen et al 2003; Miller et al 1998). Except for N-methyl-D-aspartate antagonists, the amphetamines are the only class of addictive drugs known to be associated with depletion of striatal dopamine (McCann and Ricaurte 2004).

Because the ventral striatum is well recognized as an important site for reward in addictive behaviors, attempts to elucidate the neurobiology of sex differences underlying addiction have focused on gender differences in this region. These investigations have revealed the nucleus accumbens, an area within the ventral striatum, as principally important in the rewarding effects of drugs of addiction (for a review, see Di Chiara et al 2004). In preclinical studies, sex differences in the striatal dopamine system have been observed (Dluzen 2004; Pohjalainen et al 1998). Rodent studies have documented sex differences in the depletion, turnover, and extracellular accumulation of dopamine following methamphetamine administration (Dluzen and

Ramirez 1985; Hruska and Silbergeld 1980; Shimizu and Bray 1993; Xiao and Becker 1994; Yu and Wagner 1994).

In addition to addictive disorders, sex differences in the clinical presentation and age of onset of or vulnerability to other neuropsychiatric illnesses that involve the striatum, such as Parkinson's disease (Scott et al 2000), schizophrenia (Aleman et al 2003), Huntington's disease (Tamir et al 1969), obsessive-compulsive disorder (Bogetto et al 1999), and Tourette's syndrome (Kidd et al 1980), have been described. Whether these differences might also be related to striatal dopamine is not known, however.

The purpose of this study was to test the hypothesis that the magnitude of dopamine, subjective, and neuroendocrine responses to amphetamine is greater in men than in women. The hypothesis was studied by measuring striatal binding potential using the D_2/D_3 dopamine (DA) receptor radioligand [^{11}C]raclopride with positron emission tomography (PET). The ventral striatum was the primary volume of interest.

Methods and Materials

Forty-three healthy individuals (28 men, 15 women), aged 18 to 29 years, were recruited for participation by newspaper advertisements and fliers posted in the Baltimore metropolitan area. Under the auspices of the Johns Hopkins School of Medicine Institutional Review Board, all participants provided written informed consent after receiving oral and written descriptions of study procedures and aims. Subject assessment included a medical history and physical examination performed by a physician, blood chemistry profile, complete blood count, liver and renal function tests, electrocardiogram, urinalysis, alcohol breathalyzer test, and urine toxicology screen. The Semi-Structured Assessment for the Genetics of Alcoholism (SSAGA; Bucholz et al 1994) was administered by a master's-level interviewer to identify *Diagnosis and Statistical Manual* (4th edition; DSM-IV) Axis I psychiatric diagnoses. Exclusionary criteria included 1) presence of DSM-IV Axis I disorder; 2) treatment in the last 6 months with antidepressants, neuroleptics, sedative hypnotics, glucocorticoids, appetite suppressants, sex hormones, or opiate or dopamine medications; 3) use of any prescription medications

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Received November 30, 2005; revised January 10, 2006; accepted January 13, 2006.

within the past 30 days; 4) women currently using a hormonal method of birth control, hormone replacement therapy, currently pregnant or lactating women, women with oligo- or amenorrhea; 5) medical conditions, including history of seizure disorder or closed head trauma; 6) unable to provide clean urine drug screens at intake or during study participation; 7) report of drinking more than 30 alcoholic drinks per month or illicit drug use within the 30 days before participation; and 8) current smoking. Following screening procedures, eligible subjects were scheduled for admission to the Johns Hopkins General Clinical Research Center (GCRC) to complete the study.

Behavioral Measures

Measures of psychiatric symptoms and perceived stress were administered during the initial assessment interview. These assessments included the following: State-Trait Anxiety Inventory (STAI; Spielberger 1983), Beck Depression Inventory (2nd edition; BDI-II; Beck et al 1996), Brief Symptom Inventory (BSI; Derogotis and Melisaratos 1993), Perceived Stress Scale (PSS; Cohen et al 1983), Life Experiences Survey (LES; Sarason et al 1978), and the Combined Hassles and Uplifts Scale (Lazarus and Folkman 1989).

Analog Rating Scales (Bigelow and Walsh, 1998)

At 5 min before each scan and 3, 6, 10, 15, 25, 55, and 85 min during scans, subjects were asked to rate verbally, on a 5-point scale (0 = least, 4 = most), the degree to which they were experiencing each of 10 possible drug effects. Positive effects included “high,” “rush,” “good effects,” “liking,” and “desire for drug.” Negative effects included “fidgety,” “anxious,” “dizziness,” “dry mouth,” and “distrust.”

Magnetic Resonance Imaging Assessment and Mask Fitting

Use of magnetic resonance imaging (MRI) allowed coregistration of the emission images so that anatomically accurate volumes of interest (VOIs) could be drawn (see VOI Definition). To minimize head motion during MRI acquisition, each subject was fitted for a thermoplastic mask modeled to his or her face before admission to the General Clinical Research Center (GCRC). The MRIs were acquired with an SPGR (spoiled gradient) sequence (TE = 5, TR = 25, flip angle = 40°, slice thickness = 1.5 mm, image matrix = 256 × 192, field of view = 24 cm) for anatomic identification of brain structures, and a double echo (proton density and T2-weighted, 5-mm-thick slices) sequence, used as a diagnostic scan and to segment extracerebral cerebrospinal fluid.

PET Procedures and Data Acquisition

Subjects were admitted to the GCRC in-patient unit the day before the PET procedures. They were instructed not to ingest any alcohol, drugs, or over-the-counter medications for 48 hour before admission. Laboratory studies at admission included a urine toxicology screen, alcohol breathalyzer test, hematocrit, electrolyte panel, and urine pregnancy screen for women. A calorie-controlled, caffeine-free breakfast was provided to subjects before the PET procedures. Beginning at 8:30 AM, subjects underwent two consecutive 90-min PET scans with [¹¹C] raclopride. This radioligand is a benzamide antagonist at D2 and D3 receptors, previously shown to be sensitive to stimulant-induced changes in brain dopamine concentration (Endres et al 1997; Volkow et al 1994). At the beginning of each scan, a high-specific-activity intravenous bolus injection of approximately 18 mCi [¹¹C] raclopride was administered. The first scan was preceded at –5 min by an intravenous injection of saline; the second

scan was preceded at –5 min by .3 mg/kg amphetamine, each delivered over 3 min. The amphetamine free base used in this study was 73.4% of the amphetamine sulfate. The .3 mg/kg of amphetamine sulfate given to each subject is .22 mg/kg amphetamine free base as a bolus over 3 min, starting 5 min before radiotracer injection of bolus [¹¹C] raclopride. The scanning image protocol consisted of up to 30 scan acquisitions in three-dimensional (3D) mode, starting from a 15-sec duration and increasing to 6 min in length over a 90-min period. All images were acquired on the 3D GE Advance whole body PET scanner and were preceded by a 10-min attenuation scan employing a rotating germanium-68 source. Subjects were under continuous cardiovascular monitoring during the scans. They were permitted to arise briefly after the first scan and were repositioned on the scanner table for the second. Subjects were escorted back to the GCRC following the scans. Before discharge, they were evaluated by a physician.

Volumes of Interest Definition

Volumes of interest (VOIs) were defined using interactive segmentation software on spoiled gradient (SPGR) MRI volumes for the caudate nucleus and the putamen bilaterally to obtain regional BP values. The software program allowed for the selection of upper and lower MRI intensity thresholds to delineate striatal structures from surrounding structures and required minimal hand drawing. The ventral striatum (VS) was automatically defined on the SPGR MRI volume, reoriented so the plane containing the midline separating the left and right halves of the brain is orthogonal to the horizontal plane containing the points representing the anterior commissure and the posterior commissure (anterior commissure–posterior commissure plane). On each coronal slice, the portion of the striatal volumes of interest ventral to the line crossing the ventral corner of the lateral ventricle and perpendicular to the bisector of the internal capsule defined the VS (Baumann et al 1999). The MRI volumes were spatially aligned to the PET volumes (averaged volumes across frames taken between 30 and 90 min after tracer-injection) using information theory (Collignon et al 1995) and implemented in SPM2b software (Friston 2002; see <http://www.fil.ion.ucl.ac.uk/spm/>). The same transformation parameters were applied to transfer VOIs from MRI space to PET space. The cut-off level of VOIs in PET spaces was set at .5; the value of VOI voxels in the MRI spaces was set to 1, and that of remaining voxels was set to 0.

Modeling of PET Outcome Measures

The binding potential (BP) = B_{max}/K_d was used to measure [¹¹C]raclopride D2-like receptor-specific binding (Wong 2002). The BP used in this work is based on a simplified reference tissue model (SRTM), which is based on the BP defined as k_3/k_4 or $(DV_{total} - DV_f + n) / (BP = f_2 B'_{max} / K_d)$, where f_2 is the free fraction of tracer in brain tissue, B'_{max} is the available receptor density for tracer binding in nM, and K_d is the equilibrium dissociation constant in nM; see Gunn et al 2001). The cerebellum was the reference tissue used to estimate BP (Lammertsma and Hume 1996). Because the cerebellum is nearly devoid of D2 and D3 receptors, specific binding of [¹¹C]raclopride is thought to be negligible in the cerebellum. A linear regression with spatial constraint algorithm was used to fit SRTM model to measured voxel kinetics, and parametric BP images were generated (Zhou et al 2003). The VOIs defined on MRI images were transferred to BP images to obtain VOI BP values. The percent change in BP from baseline was used to estimate dopamine release as $((BP_{placebo} - BP_{amphetamine}) / BP_{placebo}) \times 100$, with lower BP values

Table 1. Subject Demographics

| | Men | Women | <i>p</i> Value |
|-----------------------------|------------|------------|----------------|
| Sample size (<i>n</i>) | 28 | 15 | |
| Age | 22.0 (3.0) | 21.7 (3.1) | .731 |
| Race, <i>n</i> (%) | | | .297 |
| Caucasian | 19 (67.9) | 8 (53.3) | |
| African American | 4 (14.3) | 5 (33.3) | |
| Asian | 4 (14.3) | 1 (6.7) | |
| Hispanic | 0 (0.0) | 1 (6.7) | |
| Other | 1 (3.6) | 0 (0.0) | |
| BMI (kg/m ²) | 24.9 (3.1) | 23.2 (2.4) | .074 |
| Education, years | 14.7 (1.7) | 14.5 (2.1) | .762 |
| Drinking episodes per week | 0.7 (0.7) | 0.6 (0.4) | .418 |
| Drinks per drinking episode | 3.3 (2.7) | 2.7 (1.7) | .544 |

Except for sample size and race, all variables are presented as mean (SD).

during the amphetamine scan indicating greater levels of endogenous dopamine.

Hormone Assays

Cortisol, estradiol, progesterone, total testosterone, and free testosterone were measured by radioimmunoassay (Diagnostic Products Corporation, Los Angeles, California). Plasma concentrations of growth hormone (GH) were assayed by a two-site IRMA (Nichols immunoradiometric assay). Blood for estradiol, progesterone, and testosterone measurement were collected on the day of the scan. Women with progesterone levels ≥ 2 ng/mL were identified as being in the luteal phase of the menstrual cycle. Blood was collected for amphetamine measurement at 10, 20, 45, 55, and 85 min following injection of amphetamine. Plasma amphetamine levels were assessed by gas chromatography mass spectroscopy (Quest Diagnostics). Inter- and intraassay coefficient of variation was less than 10% for all assays.

Statistical Analysis

All statistical analyses were conducted using SPSS 12.0 for Windows. Demographic characteristics of men and women were compared using *t* tests or Chi-Square tests, as appropriate. Men's and women's scores on psychological symptom measures administered at baseline were compared with a series of *t* tests, and differences between men and women on these measures were entered as covariates in subsequent analyses. In the ventral striatum, BP and dopamine release were examined separately using *t* tests, with sex as the independent variable and then with analyses of covariance (ANCOVA) with baseline differences between men and women entered as covariates. In the anterior and posterior regions of the putamen and caudate nuclei, BP and dopamine release were examined using multivariate analyses of variance (MANOVA), with sex as the independent variable and BP or dopamine release in the four volumes of interest as the dependent variables. BP and dopamine release were also examined with multivariate analyses of covariance (MANCOVA) to control for baseline differences between men and women. Subjective analog scales of drug effect were examined by identifying each subject's highest (peak) rating for each scale first under the placebo condition and then under the amphetamine condition. To adjust for nonnormal distribution, all peak values were square-root transformed. Each square-root-transformed peak value under the placebo condition was subtracted from the square-root transformed peak value under the amphetamine condition to obtain a "response." The five positive scales were highly correlated. Therefore, a single "positive" score was de-

rived by computing the mean of the five square-root-transformed positive "response" scores. This measure was used in the analysis comparing men and women. The five negative scales were also highly correlated; a "negative" scale was thus derived in the same manner as the "positive" scale for use in analyses. A MANOVA was used to compare men's and women's responses on the "positive" and "negative" scales. An ANOVA was then used to explore any sex differences for each of the five scales comprising the "positive" and "negative" response scores. Cortisol and GH for men and women were compared by subtracting the hormone level under the placebo condition at each time point from the hormone level in the amphetamine condition at each time point. The resulting "response" values were then compared in a MANOVA for repeated measures. For exploratory analyses investigating the association between women's menstrual phase (follicular vs luteal) and dopamine release, BP, subjective responses, and cortisol and GH, univariate analyses of variance were performed.

Results

Demographics

Table 1 summarizes the demographic characteristics of the sample. Men and women did not differ in age, race, body mass, education, or the frequency or amount of alcohol consumed weekly.

Psychological Measures

Scores on mood assessments and measures of distress are shown in Table 2. Women had higher trait anxiety (STAI), endorsed a greater severity of subjective distress (BSD), and perceived more events as negative (LES) than did men.

Dopamine Binding and Release

Figure 1 illustrates D2 receptor availability during the placebo and amphetamine challenge and the volumes under investigation. There was no sex difference in baseline BP in the ventral striatum (Table 3). In contrast, dopamine release in the ventral striatum was higher in men than in women ($p = .010$; Figure 2). Secondary analyses revealed that baseline BP in the other striatal regions did not differ between men and women but that men had greater dopamine release in three of four striatal regions examined [$F(4,38) = 2.628, p = .049$]. Differences were revealed in the anterior putamen ($p = .017$), as well as the anterior and posterior

Table 2. Psychological Symptom Measures

| | Men | Women | <i>p</i> Value |
|--|------------|------------|----------------|
| Sample Size | 28 | 15 | |
| Trait Anxiety (STAI) | 27.8 (6.6) | 34.1 (9.4) | .017 |
| State Anxiety (STAI) | 26.8 (7.3) | 30.1 (6.8) | .156 |
| Depression (BDI) | 2.2 (3.2) | 3.6 (4.0) | .257 |
| Global Severity Index (BSI) | 0.2 (0.2) | 0.3 (0.3) | .020 |
| Perceived Stress Scale (PSS) | 11.0 (6.5) | 13.6 (7.0) | .295 |
| Hassles Frequency/Severity (H-U) | | | |
| No. items | 16.7 (8.9) | 17.8 (4.8) | .683 |
| Mean item score | 1.2 (0.3) | 1.4 (0.3) | .062 |
| Life Experiences Survey (Negative Items) | | | |
| No. items | 2.0 (2.2) | 4.1 (2.3) | .015 |
| Mean item score | 1.5 (0.7) | 1.4 (0.3) | .638 |

Except for sample size, all variables are presented as mean (SD). BDI, Beck Depression Inventory; BSI, Brief Symptom Inventory; H-U, Combined Hassles and Uplift Scale; PSS, Perceived Stress Scale; STAI, State-Trait Anxiety Inventory.

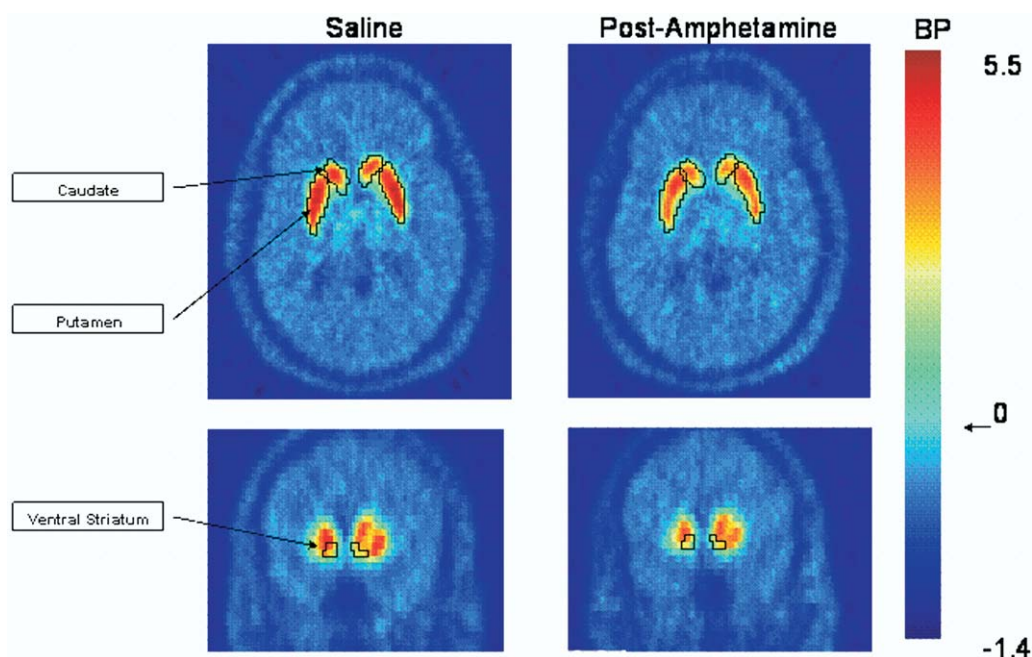


Figure 1. Representative transaxial (top row) and coronal images (bottom row) of parametric binding potential (BP) volumes, baseline saline (left panel), and post-amphetamine (right panel) scans taken from one subject (20-year-old man). Outlines of volumes of interest for the caudate nucleus, putamen, and ventral striatum are shown. Color scale bar indicates voxel BP values that can assume negative in cerebrospinal fluid space and outside the brain.

caudate nuclei ($p = .010$ and $.012$, respectively) but not in the posterior putamen ($p = .128$; see Table 3). After controlling for differences between men and women on a measure of trait anxiety (STAI), severity of distress related to psychiatric symptoms (BSD), and number of life events judged as having a negative impact (LES), results for all analyses were unchanged. Plasma amphetamine concentrations obtained at 10, 20, 45, 55, and 85 minutes following the injection of amphetamine did not differ by sex.

Estradiol and progesterone levels are provided in Table 4. Women in the luteal phase of the menstrual cycle ($n = 6$) had lower baseline BP in both the anterior putamen ($p = .045$) and posterior putamen ($p = .034$) but not in the caudate ($p = .123$ anterior, $.351$ posterior) or ventral striatum ($p = .199$), when compared with women in the follicular phase ($n = 9$). Dopamine release, however, did not differ by phase of the menstrual cycle in any brain region explored (all p values $> .206$). Neither estradiol nor progesterone level was correlated with baseline BP or dopamine release.

In men, neither total nor free testosterone levels correlated with baseline BP or dopamine release in the ventral striatum.

Subjective Drug Effects

Subjective responses to amphetamine were greater in men than in women [$F(2,40) = 3.902$, $p = .028$] above the effects of the placebo. In particular, men had greater positive ($p = .008$) but not negative ($p = .262$) responses than women to amphetamine. Examination of each scale comprising the “positive” and “negative” subjective responses indicated that men’s ratings on four of the five positive scales were higher than women’s (all significant p values $< .026$), whereas none of the negative scales differed by sex. Results for each subscale comprising the “positive” and “negative” scales are shown in Figure 3. As previously shown (Oswald et al 2005), dopamine release in all regions examined correlated with positive subjective responses to am-

phetamine in the whole sample (R values ranged from $.309$ to $.365$, p values ranged from $.019$ to $.049$).

Women in the follicular and luteal phases of the menstrual cycle did not differ in either positive or negative subjective responses (data not shown). In men, neither total nor free

Table 3. Raclopride Binding Potentials During Placebo and Amphetamine PET Scans^a

| Region | Placebo | Amphetamine | Dopamine Release ^b |
|----------------|-------------|-------------|-------------------------------|
| aPU | | | |
| Men | 3.06 ± 0.33 | 2.67 ± 0.31 | 12.59 ± 6.30 |
| Women | 3.09 ± 0.25 | 2.84 ± 0.27 | 8.19 ± 3.56 |
| <i>p</i> value | .777 | .091 | .017 |
| pPU | | | |
| Men | 3.04 ± 0.40 | 2.43 ± 0.33 | 19.94 ± 6.59 |
| Women | 3.19 ± 0.27 | 2.65 ± 0.20 | 16.97 ± 4.56 |
| <i>p</i> value | .185 | .021 | .128 |
| aCN | | | |
| Men | 2.63 ± 0.30 | 2.45 ± 0.28 | 6.58 ± 5.62 |
| Women | 2.70 ± 0.25 | 2.64 ± 0.26 | 2.20 ± 3.72 |
| <i>p</i> value | .409 | .032 | .010 |
| pCN | | | |
| Men | 1.86 ± 0.40 | 1.68 ± 0.34 | 9.59 ± 7.09 |
| Women | 1.95 ± 0.30 | 1.86 ± 0.29 | 4.16 ± 5.00 |
| <i>p</i> value | .463 | .079 | .012 |
| VS | | | |
| Men | 2.12 ± 0.32 | 1.88 ± 0.31 | 11.64 ± 5.52 |
| Women | 2.08 ± 0.21 | 1.94 ± 0.22 | 7.13 ± 4.54 |
| <i>p</i> Value | .686 | .512 | .010 |

aCN = anterior caudate nucleus; aPU = anterior putamen; pCN = posterior caudate nucleus; PET = positron emission tomography; pPU = posterior putamen; VS = ventral striatum.

^aValues represent mean ± SD.

^bDopamine release = ((placebo BP – amph BP)/placebo BP) * 100.

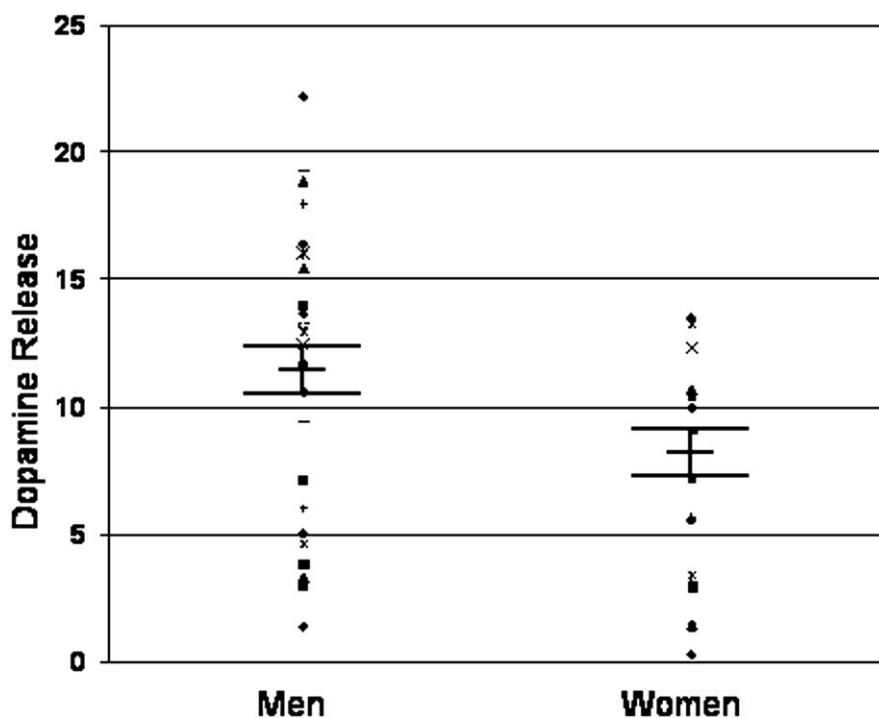


Figure 2. Dopamine release in the ventral striatum by sex. Bars represent mean and standard error.

testosterone levels correlated with subjective responses to amphetamine.

Cortisol and Growth Hormone

Measurements of plasma cortisol and GH were obtained at baseline (−25 and −5 min) and at scheduled intervals (15, 35, 55, and 75 min) during the scans. Although both cortisol and GH increased following administration of amphetamine, these increases did not differ between men and women. Comparison of women in the follicular phase to those in the luteal phase also revealed no differences (data not shown).

Discussion

The aim of this study was to determine whether striatal dopamine response following administration of amphetamine was similar in men and women. Our primary finding was a robust sex difference; men exhibited greater dopamine release than women in the ventral striatum, anterior putamen, and anterior and posterior caudate nuclei. These findings were maintained whether or not the analyses was adjusted for sex differences on psychological symptom measures, for phase of the menstrual cycle in women, or for testosterone levels in men. Supporting this neurochemical observation was the finding that men also rated the positive effects of amphetamine higher than did women. Plasma amphetamine levels did not differ by sex and therefore cannot explain differences in dopamine release or subjective responses to the drug.

To our knowledge, this is the first report in humans of a sex difference in dopamine release. Prior studies have compared men and women on other aspects of the striatal dopaminergic system. Consistent with our finding of equivalent baseline binding potential in men and women, two previous studies found no sex difference in dopamine receptor density (Farde et al 1995; Pohjalainen et al 1998). Dopamine receptor affinity, in contrast, was found to be lower in women than men in one study (Pohjalainen et al 1998). This is not, however, a consistent

finding (see Farde et al 1995). In an investigation of dopamine synthesis capacity, women had higher [^{18}F] fluorodopa uptake than men in striatum (Laakso et al 2002), suggesting that female sex hormones enhance presynaptic dopamine turnover. Results of preclinical studies support this claim (Dluzen and Ramirez 1985; Shimizu and Bray 1993; Xiao and Becker 1994).

In women, surges of estrogen are associated with increased dopamine activity (DiPaolo et al 1988; Levesque et al 1989). For example, striatal dopamine turnover is high (Shimizu and Bray 1993) and extracellular dopamine concentrations in the striatum and nucleus accumbens are elevated in rats during high estrogen states associated with estrus (Dluzen and Ramirez 1985; Xiao and Becker 1994). Furthermore, estradiol administration has been shown to increase receptor density in the striatum (Hruska and Silbergeld 1980) as well as increase dopamine turnover in the nucleus accumbens (Shimizu and Bray 1993). In contrast, progesterone has an overall blunting effect on the striatal dopamine system, opposing the actions of estradiol (Fernandez-Ruiz et al 1990; Shimizu and Bray 1993; White et al 2002). In fact, progesterone administration to men dampens subjective and physiological responses to cocaine (Sofluoglu et al 2004). It has been posited that the ratio of estrogen to progesterone, which changes throughout the menstrual cycle, helps determine responsiveness to amphetamine (White et al 2002). We observed lower baseline BP measurements in the putamen during the luteal phase compared with the follicular phase of the menstrual cycle. Dopamine release did not differ as a function of menstrual phase in any striatal region, however. Sample size and the between-subject design may have precluded capturing intercycle varia-

Table 4. Hormone Levels by Menstrual Phase

| | Follicular | Luteal |
|--------------------------|---------------|----------------|
| Sample Size | 9 | 6 |
| Estradiol, pg/mL (SD) | 59.63 (45.03) | 118.41 (69.62) |
| Progesterone, ng/mL (SD) | 0.83 (0.48) | 10.18 (7.12) |

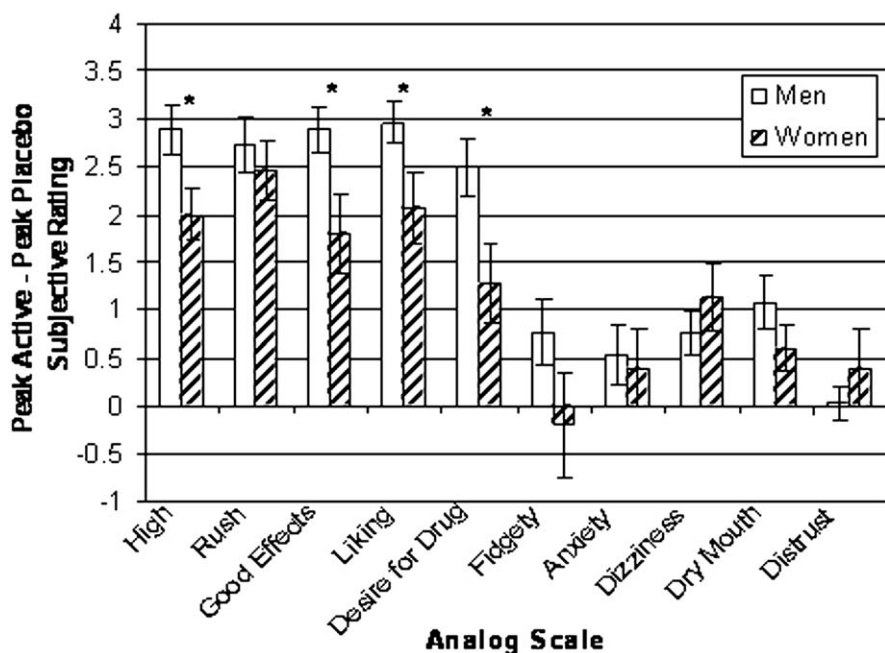


Figure 3. Subjective analog scale responses by sex.

tions in dopamine release or subjective responses. Regardless of menstrual phase differences, however, men had greater dopamine release than women.

The second finding from this investigation was that men rated the positive effects of amphetamine higher than did the women. Previous research (Laruelle et al 1995; Oswald et al 2005; Volkow et al 1999) as well as this study demonstrate a correlation between dopamine release and subjective responses to stimulant drugs; greater subjective responses to amphetamine, cocaine, and methylphenidate are associated with greater dopamine release. Our findings of greater dopamine release and subjective responses in men compared to women are thus compatible with this observation. In contrast to our finding regarding the subjective responses to amphetamine, preclinical studies have shown that female subjects exhibit greater behavioral response and sensitization to stimulants than do male subjects (Becker et al 2001). Perhaps the fact that our findings were seen in the associative, but not in the sensorimotor, areas of the striatum (Martinez et al 2005) accounts for the apparent discrepancy between behavioral observations made from preclinical studies and sex differences in subjective responses seen in our study.

Our findings are in agreement with clinical observations regarding drug dependence. The ventral striatum is well recognized as an important site for reward in response to various drugs of abuse (Bonci et al 2003; Koob 1992; Robinson et al 1988; Volkow et al 1997). Pharmacological studies have shown that men have greater subjective responses to amphetamine and cocaine compared with women, especially when women are in the luteal phase of the menstrual cycle (Sofuoglu et al 2004; White et al 2002). Sex studies have also shown a higher prevalence of stimulant and alcohol use disorders in men than women (Substance Abuse and Mental Health Services Administration 2005). Men are also more vulnerable to methamphetamine toxicity (Dluzen et al 2003; Miller et al 1998), and male stimulant abusers show greater electroencephalogram abnormalities than female stimulant users (King et al 2000). Our findings suggest that differences between men and women in dopamine release may serve as a possible mechanism underlying the observed sex differences in the clinical presentation and neuro-

logical consequences of stimulant use. That is, given that the ventral striatum is a reward center for drugs of abuse, men's higher level of dopamine release in this vulnerable substrate may predispose them to greater use and abuse of stimulant drugs.

The third finding in our study was that there was no significant sex difference in the degree to which cortisol and growth hormone responded to amphetamine. Although amphetamine is a robust activator of the hypothalamic-pituitary-adrenal axis, the gender effect on dopamine release does not appear to affect the more distal event, namely, the release of cortisol or growth hormone.

It remains unclear how male sex accounts for greater dopamine responses to amphetamine throughout the striatum, but it most likely relates to the influence of sex hormones on the dopaminergic system. Preclinical studies have shown that men have greater amphetamine-induced striatal dopamine release as well as dopamine depletion than women (Yu and Wagner 1994). Findings from our study concur with preclinical studies. Although estradiol may enhance presynaptic dopaminergic function, preclinical studies have shown opposite effects in regard to dopamine release. For example, using striatal tissue fragments from gonadectomized and intact male and female mice, the amount of dopamine evoked by methamphetamine was significantly reduced when estrogen was coinfused (Myers et al 2003). In contrast, testosterone failed to produce an overall change in methamphetamine-evoked dopamine output. It appears that estrogen but not testosterone exerts modulatory effects on methamphetamine-evoked dopamine output (Bisagno et al 2003; Gao and Dluzen 2001). In agreement with these observations, our study found no relationship between testosterone and dopamine measures in men. Combining the various findings outlined earlier, we posit that in women, estrogen dampens dopamine release but also enhances presynaptic dopamine turnover and thus limits the kind of severe dopamine depletion observed in men following chronic amphetamine exposure. This difference between estrogen and testosterone may explain why estrogen plays a neuroprotective role in modulating the effects of stimulants on the central nervous system (Bisagno et al 2003).

The relevance of our findings may extend beyond addiction

to include other disorders involving the striatum. Obsessive-compulsive disorder, for example, is associated with reduced availability of striatal dopamine transporter (Hesse et al 2005). In this disorder, men have an earlier age of onset, more tics, and worse outcomes than women (Bogetto et al 1999). Tourette's syndrome, associated with altered dopamine release in response to amphetamine (Singer et al 2002), is more prevalent in boys than in girls (Kidd et al 1980). Reduced transporter binding in the striatum has also been found in patients with Parkinson's disease (Schwarz et al 2000). Sex differences in the symptom profiles of patients with Parkinson's disease, as well as greater prevalence of this disease in men compared with women, have also been reported (Fall et al 1996; Scott et al 2000). By far, the most widely researched neuropsychiatric disorder in terms of its relation to abnormalities in the striatal dopaminergic system is schizophrenia. Sex differences, such as earlier onset and more negative symptoms in men, are consistently reported (Aleman et al 2003). Although the mechanisms for sex differences in disorders involving the striatal dopamine system are poorly understood, the consistency with which men appear to be more vulnerable than women is striking. To the extent that our findings suggest an increased reactivity of the striatal dopamine system in men compared with women, we speculate that the sex differences in these various neuropsychiatric disorders is at least partially related to this difference in striatal dopaminergic reactivity. Our findings bear relevance to these differences and indicate that future studies of dopamine release should control for sex.

This study has several strengths. First, the sample size is larger than that of most PET studies examining changes in [¹¹C]raclopride binding potential. Second, inclusion of this larger number of participants allowed us to perform a meaningful, comprehensive assessment of mood, stress, and sex hormones and then control for these important measures in assessing dopamine response to amphetamine. Third, the PET procedure employed has been well validated (Hietala et al 1999; Singer et al 2002). Fourth, adjusting the analysis for comparing dopamine release in multiple striatal regions (MANOVA) provides a conservative assessment, thus minimizing type I error.

Several weaknesses of this investigation should be noted. First, because of the potential for prolonged sensitization to amphetamine, the order of the scans was not counterbalanced. The placebo scan always preceded the amphetamine scan. Second, the rationale for this investigation was based in part on findings from preclinical data. Because the species differences in pharmacokinetics and neurotoxicity are far from understood, caution should be taken when extrapolating to humans. Third, we based this study on literature concerning methamphetamines and extrapolated it to amphetamine. Because methamphetamine is converted to amphetamine in the body and it would seem logical to assume that the findings from methamphetamine would apply to amphetamine. This assumption remains to be proven, however, because amphetamine may not have the same neurotoxicity as methamphetamine. A number of studies have reported negligible displacement of the radioactivity in the cerebellum after amphetamine challenge using bolus-plus-infusion scheme in which the radioactivity in the cerebellum remained constant (e.g., Breier et al 1997). Nevertheless, we cannot determine to what extent this approach might have affected our results. Fourth, although the term "dopamine release" has been used conventionally in the PET literature to describe amphetamine-induced changes in [¹¹C]raclopride BP, the increases in dopamine concentrations that occur following amphetamine administration probably result

from several mechanisms, including dopamine reuptake blockade, reverse transport of dopamine through the dopamine transporter (Schmitz et al 2001) as well as possible actions on endogenous opioid systems (Schad et al 2002). Other mechanisms such as internalization of dopamine receptors (Ginovart et al 2004; Laruette 2000) and change in the affinity status (Narendran et al 2004) are also under investigation to explain the sustained [¹¹C]raclopride displacement after amphetamine administration. Our use of the term "dopamine release," therefore, does not convey a full description of the mechanisms by which amphetamine alters dopamine concentration. Fifth, although the study was adequately powered for a meaningful comparison between men and women, the relatively small number of women in the luteal phase of the cycle precludes definitive statements about dopamine release as a function of menstrual cycle phase.

Conclusion

We report for the first time in humans a sex difference in dopamine release in vivo. This finding has implications for observed sex differences in a wide variety of neuropsychiatric illnesses involving the striatum and indicates that future studies of these disorders need to control for sex.

Supported by Grant Nos. AA10158 (GSW), AA12837 (MEM), AA12839 (DFW), and GCRC (NIH/NCRR M01RR00052).

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