Hypothalamic Abnormalities in Schizophrenia: Sex Effects and Genetic Vulnerability

Jill M. Goldstein, Larry J. Seidman, Nikos Makris, Todd Ahern, Liam M. O'Brien, Verne S. Caviness, Jr., David N. Kennedy, Stephen V. Faraone, and Ming T. Tsuang

Background: This is a unique hypothalamic magnetic resonance imaging (MRI) study in schizophrenia, an important region in the limbic system. We hypothesized abnormal volumetric increases, with greater severity in multiplex families (more than one ill member) compared with simplex families (one ill). We tested the hypothesis that normal hypothalamic sexual dimorphism is disrupted in schizophrenia.

Methods: Eighty-eight DSM-III-R schizophrenia cases (40 simplex and 48 multiplex), 43 first-degree nonpsychotic relatives, and 48 normal comparisons systematically were compared. A 1.5-Tesla General Electric scanner was used to acquire structural MRI scans, and contiguous 3.1-mm slices were used to segment anterior and posterior hypothalamus. General linear model for correlated data and generalized estimating equations were used to compare cases, relatives, and controls on right and left hypothalamus, controlled for age, sex, and total cerebral volume. Spearman’s correlations of hypothalamic volumes with anxiety were calculated to begin to examine arousal correlates with structural abnormalities.

Results: Findings demonstrated significantly increased hypothalamic volume in cases and nonpsychotic relatives, particularly in regions of paraventricular and mammillary body nuclei, respectively. This increase was linear from simplex to multiplex cases, was positively correlated with anxiety, and had a greater propensity in women.

Conclusions: Findings suggest important implications for understanding genetic vulnerability of schizophrenia and the high rate of endocrine abnormalities.

Key Words: Genetic vulnerability, hypothalamus, MRI, schizophrenia, sex differences

Few current studies exist that examine hypothalamic volume in schizophrenia, even though the hypothalamus has connections to nearly every part of the central nervous system, including the cortex, the amygdala and hippocampus (Mesulam 1985; Swaab 2003, 2004a), all of which are known to be abnormal in schizophrenia. It weighs only about 4 g but is comprised of at least 14 distinct nuclei that are involved in drives, affect, autonomic and endocrine control, and immunoregulation.
Methods and Materials

Subjects

Simplex Patients. Cases, primarily outpatients, were recruited from three public Boston-area psychiatric hospitals serving primarily psychotic patients (Goldstein et al. 1998; Kremen et al. 1996; Seidman et al. 1997). The sample included subjects who were reported in previous work (Goldstein et al. 2002; Seidman et al. 2002); hypothalamic data have not been reported elsewhere. Recruitment criteria for subjects included the following: ages at MRI scanning, 23–68 years; at least an 8th-grade education; English as first language; and an estimated intelligence quotient (IQ) of \( \geq 70 \). Criteria required absence of the following: substance abuse during the past 6 months, history of head injury with documented cognitive sequelae or of loss of consciousness of longer than 5 min, neurologic disease or damage, and medical illnesses significantly impairing neurocognitive function. Cases were DSM-III-R schizophrenia probands \( n = 40 \), on the basis of interview by very experienced diagnostic interviewers and systematic review of medical records. Senior investigators (JMG, LJS) reviewed all material to determine diagnosis (see Goldstein et al. [1998, 2002] for details and excellent reliability).

Multiplex Probands. These outpatients were ascertained from the Harvard cohort of the National Institute of Mental Health (NIMH) Genetics Schizophrenia Initiative (MH6318; principal investigator, MTT). Families with at least two persons affected with DSM-III-R schizophrenia or with schizoaffective disorder, depressed type, were identified by systematic screening in psychiatric hospitals and clinics. Test–retest reliabilities were excellent (Nurnberger et al. 1994). Procedures for diagnosing multiplex probands were similar to those for the simplex probands described in the previous section. Rerecruitment of the Harvard cohort (NIMH MH56956, principal investigator, JMG) involved recruitment letters to probands, case managers, guardians, and Massachusetts area directors and involved home visits for those without phones or listed numbers. Fifty probands were scanned, for whom data from two were excluded because of motion artifacts.

Relatives. Relatives were free of lifetime psychosis. There were 28 simplex and 17 multiplex relatives from 34 unique families. Twenty-six families provided a single relative, three families had three relatives, and five had two relatives. All available relatives were interviewed (Spitzer et al. 1987). Although half of the relatives had experienced a nonpsychotic diagnosis, mainly past major depressive disorder or substance abuse, we demonstrated that these potential confounds did not significantly affect brain volumes (Seidman et al. 1999, 2002) or neuropsychological dysfunction (Faraone et al. 1995, 2000). In addition, analysis of a subset of 18 relatives from the 13 families who had a schizophrenia proband demonstrated that the results were present in subjects coming from the same families.

Normal Comparison Subjects. These individuals were recruited through advertisements in the catchment areas and through posted notices in services from which the patients were ascertained (Goldstein et al. 2002; Seidman et al. 2002). They were selected to be comparable to patients and relatives on age, sex, ethnicity, parental socioeconomic status (SES), and handedness and were screened for current psychopathology (Vincent et al. 1984) and for family history of psychoses or psychiatric hospitalizations. Potential controls were excluded if they had current psychopathology or lifetime history of any psychosis, family history of psychosis, or psychiatric hospitalization or if the score for any Minnesota Multiphasic Personality Inventory clinical or validity scale, except Masculinity-Femininity, was above 70.

Blindness of assessments was maintained among MRI data and psychiatric status. Written informed consent was obtained from all subjects after providing a complete description of the study, and they were compensated for their time and participation. This study was approved by Harvard Medical School and hospital (Massachusetts Mental Health Center and Massachusetts General Hospital) human-studies committees. Permission was obtained from patients to contact their relatives. Ascertainment of relatives used the same inclusion/exclusion criteria as patients and controls and blindness of assessments was maintained.

Table 1 shows that multiplex and simplex cases and normal comparison individuals were similar on age, Caucasian ethnicity, middle- to lower-middle parental SES, and right-handedness. Although not significantly different, age at MRI scanning was slightly older in multiplex probands compared with controls, thus controlled in analyses. Parental SES was lower among multiplex probands than among controls. The probands’ education typically was high-school completion and some college, although multiplex probands had less education and lower reading ability and IQ estimates (Kremen et al. 1996) than did simplex probands and normal controls.

Probands primarily had undifferentiated or paranoid subtypes (see Table 1) and were clinically stable, living in the community with mild to moderate negative and positive symptomatology. Multiplex subjects had significantly higher global ratings on affective flattening, avolition, formal thought disorder, and attention, on the basis of assessments that were conducted using our clinical diagnostic instruments, mentioned above (see Table 1). Patients were a chronically disabled group, with the average chlorpromazine-equivalent neuroleptic daily dose not signifi-
IQ, intelligence quotient; MRI, magnetic resonance imaging; WRAT-R, Wide Range Achievement Test–Revised (Jastak and Jastak 1985).

All data are mean (SD) unless otherwise indicated.

IQ, intelligence quotient; MRI, magnetic resonance imaging; WRAT-R, Wide Range Achievement Test–Revised (Jastak and Jastak 1985).

*Data are F statistic unless otherwise indicated. Multiplex and simplex probands were not significantly different on sociodemographic, handedness, diagnostic type, age at first hospitalization, or number of hospitalizations. Multiplex probands were older than normal controls, and thus, age was controlled in analyses. All subjects were clinically stable. Chlorpromazine equivalents were calculated using a standard formula for converting the specific typical and atypical antipsychotic medications.

**IQ estimate derived from vocabulary and block-design age-scale scores (Brooker and Cyr 1986).

Substance abuse ratings: 0 = never/occasional use; 1 = recreational (episodic) use; 2 = regular use; 3 = abuse (for a period of 6 months to 5 years); 4 = sustained abuse (for more than 5 years).

...cently different between family types (see Table 1). Approximately 43% of patients were on typical antipsychotic medications, and 57% were on atypical (primarily clozapine, and to a lesser extent, olanzapine and risperidone) antipsychotic medications.

Groups did not differ significantly on age, parental education, ethnicity, handedness, or past alcohol use (see Table 1). There were significant differences by education, IQ, and past drug use, and these differences were significant across all categories. There were no significant differences between male and female relatives compared with controls; thus, sex was controlled in analyses with relatives.

### MRI Parameters and Segmentation Procedures

MRI scans were acquired at the Athinoula Martinos Biomedical Imaging Center at Massachusetts General Hospital with a 1.5-Tesla General Electric Signa scanner (Waukesha, Wisconsin). Contiguous 3.1-mm coronal spoiled gradient echo images of the entire brain were obtained by using the following parameters: TR = 40 msec, TE = 8 msec, flip angle = 50°, field of view = 30 cm, matrix = 256 × 256, and averages = 1. MR images were analyzed at the Massachusetts General Hospital Center for Morphometric Analysis. Images were positionally normalized to a three-dimensional coordinate system on MR scans (Caviness et al. 1996; Filipek et al. 1994) and were resliced into normalized 3.1-mm coronal scans.

Scans were segmented by using a semiautomated intensity contour mapping algorithm and signal-intensity histogram distributions. This technique (described in the following articles: Caviness et al. 1996], Goldstein et al. [1999], Rademacher et al. [1992], and Seidman et al. [1999]) yields separate compartments of neocortex, subcortical gray nuclei, white matter, and ventricular system subdivisions that generally correspond to natural tissue boundaries, distinguished by signal intensities in the T1-weighted images. The neocortex was subdivided into bilateral parcellation units on the basis of the system in Caviness et al. (1996) that was applied in Goldstein et al. (1999) to schizophrenia. This is a comprehensive system for neocortical subdivision that is designed to approximate architectonic and functional subdivisions and is based on specific topographical anatomic landmarks that are present in all brains (Caviness et al. 1996; Rademacher et al. 1992). Volumes, measured in cubic centimeters, were calculated for brain regions by multiplying the slice thickness by area measurements on each slice and summing slices on which the region appeared. Very good inter- and intrarater reliability of regions has been established in previous studies (Caviness et al. 1996; Goldstein et al. 1999; Seidman et al. 1999). Further, the concurrent, discriminant, and predictive validity of these techniques have been demonstrated in numerous studies of normals and patient populations (Caplan et al. 1995; Filipek et al. 1994; Goldstein et al. 1999; Rauch et al. 2000; Seidman et al. 1999; Vaina et al. 1998). Detailed analyses of cases versus normal comparisons are reported (Goldstein et al. 1999, 2002).

### Hypothalamic Segmentation

The hypothalamus occupies the ventral and rostral half of the diencephalon and is situated on either side of the third ventricle (Figure 1; Figure 2A–C). It is composed of approximately 20 nuclei and is traversed by white-matter fiber pathways that interconnect components of the limbic system and neocortex. When using morphometric MRI, the precise delineation of the hypothalamic region borders is challenging because of the...
A mixture of gray- and white-matter tissue that at times is not easily distinguishable from adjacent classes of tissue. Thus, the detailed knowledge of basal forebrain and diencephalic topography is necessary for an accurate segmentation of the hypothalamus using MRI (examples given in Nieuwenhuys et al. [1988], Nolte [2002], Parent [1996], Saper [2004], and Swanson [1987]).

In addition, to reliably segment the hypothalamus, it is necessary to provide conventions regarding morphologic anatomic landmarks that consistently will be observable with MRI. Thus, the detailed knowledge of basal forebrain and diencephalic topography is necessary for an accurate segmentation of the hypothalamus using MRI (examples given in Nieuwenhuys et al. [1988], Nolte [2002], Parent [1996], Saper [2004], and Swanson [1987]).

In addition, to reliably segment the hypothalamus, it is necessary to provide conventions regarding morphologic anatomic landmarks that consistently will be observable with MRI. Thus, the detailed knowledge of basal forebrain and diencephalic topography is necessary for an accurate segmentation of the hypothalamus using MRI (examples given in Nieuwenhuys et al. [1988], Nolte [2002], Parent [1996], Saper [2004], and Swanson [1987]).

In addition, to reliably segment the hypothalamus, it is necessary to provide conventions regarding morphologic anatomic landmarks that consistently will be observable with MRI. Thus, the detailed knowledge of basal forebrain and diencephalic topography is necessary for an accurate segmentation of the hypothalamus using MRI (examples given in Nieuwenhuys et al. [1988], Nolte [2002], Parent [1996], Saper [2004], and Swanson [1987]).

In addition, to reliably segment the hypothalamus, it is necessary to provide conventions regarding morphologic anatomic landmarks that consistently will be observable with MRI. Thus, the detailed knowledge of basal forebrain and diencephalic topography is necessary for an accurate segmentation of the hypothalamus using MRI (examples given in Nieuwenhuys et al. [1988], Nolte [2002], Parent [1996], Saper [2004], and Swanson [1987]).

In addition, to reliably segment the hypothalamus, it is necessary to provide conventions regarding morphologic anatomic landmarks that consistently will be observable with MRI. Thus, the detailed knowledge of basal forebrain and diencephalic topography is necessary for an accurate segmentation of the hypothalamus using MRI (examples given in Nieuwenhuys et al. [1988], Nolte [2002], Parent [1996], Saper [2004], and Swanson [1987]).

In addition, to reliably segment the hypothalamus, it is necessary to provide conventions regarding morphologic anatomic landmarks that consistently will be observable with MRI. Thus, the detailed knowledge of basal forebrain and diencephalic topography is necessary for an accurate segmentation of the hypothalamus using MRI (examples given in Nieuwenhuys et al. [1988], Nolte [2002], Parent [1996], Saper [2004], and Swanson [1987]).

In addition, to reliably segment the hypothalamus, it is necessary to provide conventions regarding morphologic anatomic landmarks that consistently will be observable with MRI. Thus, the detailed knowledge of basal forebrain and diencephalic topography is necessary for an accurate segmentation of the hypothalamus using MRI (examples given in Nieuwenhuys et al. [1988], Nolte [2002], Parent [1996], Saper [2004], and Swanson [1987]).

In addition, to reliably segment the hypothalamus, it is necessary to provide conventions regarding morphologic anatomic landmarks that consistently will be observable with MRI. Thus, the detailed knowledge of basal forebrain and diencephalic topography is necessary for an accurate segmentation of the hypothalamus using MRI (examples given in Nieuwenhuys et al. [1988], Nolte [2002], Parent [1996], Saper [2004], and Swanson [1987]).

In addition, to reliably segment the hypothalamus, it is necessary to provide conventions regarding morphologic anatomic landmarks that consistently will be observable with MRI. Thus, the detailed knowledge of basal forebrain and diencephalic topography is necessary for an accurate segmentation of the hypothalamus using MRI (examples given in Nieuwenhuys et al. [1988], Nolte [2002], Parent [1996], Saper [2004], and Swanson [1987]).

In addition, to reliably segment the hypothalamus, it is necessary to provide conventions regarding morphologic anatomic landmarks that consistently will be observable with MRI. Thus, the detailed knowledge of basal forebrain and diencephalic topography is necessary for an accurate segmentation of the hypothalamus using MRI (examples given in Nieuwenhuys et al. [1988], Nolte [2002], Parent [1996], Saper [2004], and Swanson [1987]).

In addition, to reliably segment the hypothalamus, it is necessary to provide conventions regarding morphologic anatomic landmarks that consistently will be observable with MRI. Thus, the detailed knowledge of basal forebrain and diencephalic topography is necessary for an accurate segmentation of the hypothalamus using MRI (examples given in Nieuwenhuys et al. [1988], Nolte [2002], Parent [1996], Saper [2004], and Swanson [1987]).

In addition, to reliably segment the hypothalamus, it is necessary to provide conventions regarding morphologic anatomic landmarks that consistently will be observable with MRI. Thus, the detailed knowledge of basal forebrain and diencephalic topography is necessary for an accurate segmentation of the hypothalamus using MRI (examples given in Nieuwenhuys et al. [1988], Nolte [2002], Parent [1996], Saper [2004], and Swanson [1987]).

In addition, to reliably segment the hypothalamus, it is necessary to provide conventions regarding morphologic anatomic landmarks that consistently will be observable with MRI. Thus, the detailed knowledge of basal forebrain and diencephalic topography is necessary for an accurate segmentation of the hypothalamus using MRI (examples given in Nieuwenhuys et al. [1988], Nolte [2002], Parent [1996], Saper [2004], and Swanson [1987]).

In addition, to reliably segment the hypothalamus, it is necessary to provide conventions regarding morphologic anatomic landmarks that consistently will be observable with MRI. Thus, the detailed knowledge of basal forebrain and diencephalic topography is necessary for an accurate segmentation of the hypothalamus using MRI (examples given in Nieuwenhuys et al. [1988], Nolte [2002], Parent [1996], Saper [2004], and Swanson [1987]).
MBs, was segmented. Usually two consecutive coronal sections captured the entire MB in each side. The lateral border of the hypothalamus was with the substantia nigra and cerebral peduncle, whereas its medial border was at the midline of the hemisphere opposing the contralateral posterior hypothalamic nucleus and MB. Its superior border was with the third ventricle and the diencephalic fissure. This border was prompted by the white-matter fibers above the MB and lateral to the third ventricle, which belong principally to the mammillothalamic tract. The inferior border of the posterior hypothalamus was the hemispheric margin (Figure 1G and G′).

Interrater reliability, assessed by intraclass correlation coefficient, for 10 brains by two raters was very good, at .81 for right and left. To localize specific hypothalamic nuclei from which the differences between groups may have arisen, thirteen 1.5-mm slices were made on the scanned images, and voxels of overlap and nonoverlap mapped to anatomic nuclei were presented in Paxinos’ atlas templates, representing 1.3- to 1.4-mm slices (Paxinos and Huang 1995). Although this is not direct evidence of localization of abnormal nuclei, this technique can be thought of as providing the flashlight for where to look in subsequent postmortem studies on hypothalamic abnormalities.

**Statistical Analyses**

The general linear model for correlated data (GLM-CD [Cnaan et al. 1997]) was used to compare cases, controls, and relatives on the right and left hypothalamus, controlled for age, sex, and total cerebral volume. We tested for interactions of sex and group to investigate whether different differences existed in hypothalamic size in men and women, comparing the groups. GLM-CD is a multivariate model that controls for intraperson correlation between brain regions, thus providing a protected test of group effects. For multiplex cases and controls, parental SES and age were controlled. Parental SES and age were not significantly different between the two proband types. Effect sizes in hypothalamic volume were calculated comparing male and female patients (or relatives) with male and female controls, divided by the pooled SD, representing group differences in SDs. Group comparisons within a sex used unadjusted volumes, whereas analyses between the sexes used adjusted volumes, given that men have larger cerebrums than women. Asymmetry was measured as in Geschwind and Galaburda (1985), using two times the difference between left minus right hemisphere volume divided by left plus right volumes. Handedness was controlled in GLMs of the hypothalamic asymmetry. An average score of anxiety symptoms was calculated on the basis of the five-point severity scale for anxiety from the clinical diagnostic interview (Schedule for Affective Disorders and Schizophrenia; Spitzer and Endicott, 1978) and a dichotomous anxiety score on the basis of the Diagnostic Interview for Genetic Studies (Nurnberger et al. 1994). This score was correlated with hypothalamic volume by using Spearman’s correlation to begin to assess potential functional correlates of hypothalamic abnormalities and stress-response deficits, given the role of the hypothalamus in anxiety.

For GLMs involving relatives, we used generalized estimating equations (GEE) under a working independence assumption for
model estimation to account for intrafamilial correlation (Liang and Zeger 1986). That is, siblings may show familial aggregation on structural brain volumes; therefore, we have response variables that may be correlated within sibships. Estimates of the SEs of the regression coefficients then were obtained by using the empirical variance estimator (Huber 1967). Both the GLM-CD and GEE approaches are valid for analyzing correlated data. However, the GEE provides consistent estimates of the regression coefficients when the model for the correlation is misspecified and allows for variable family sizes.

Results

Table 2 presents the means, SDs, and results from the GLM-CD for patients and controls. The GLM-CD showed a significant enlargement of the right and left hypothalamus in cases versus controls [brain region by group effect: \( F(2, 105) = 3.53, p = .03 \)]. There was a linear positive increase comparing (sequentially) controls and simplex and multiplex subjects, with multiplex subjects exhibiting the largest size (Table 2). Right and left hypothalami were larger in multiplex cases (particularly right) and simplex cases.

Results from the univariate GLMs showed a greater propensity for women to exhibit abnormal volumetric enlargement than men, as reflected in the large differences in effect sizes (ESs) in female simplex cases compared with female normal controls in contrast to comparisons among male simplex and controls. That is, the ES among females was \(-.64\) on the left and \(-.71\) on the right, reflecting an enlargement of more than two thirds of an SD in both hemispheres. In contrast, among male simplex cases compared with male controls, the ES was \(-.03\) on the left and \(.09\) on the right, indicating little difference in hypothalamic volume among simplex males. However, both men and women in multiplex families showed significant hypothalamic enlargement compared with their normal counterparts \([t(76) = 2.56, p = .01]\). Correlational analyses testing whether hypothalamic size correlated with antipsychotic medication showed a Spearman’s \(r\) of \(.09\) (ns) between chlorpromazine equivalent and volume within patients.

Formal tests of abnormal asymmetries were nonsignificant. That is, in normal subjects, the right was larger regardless of one’s sex, as was true in patients with schizophrenia, although in the patients, it was exaggerated.

Figure 2 provides a visual representation of the hypothalamus in three dimensions and a flattened representation of the amount of overlap in voxels between patients and controls. Figure 3A and B illustrates the nuclei that were particularly large in patients compared with controls; for example, in Figure 3B, the PVN in patients versus controls. This was true for females and males and for simplex and multiplex cases when examined separately.

As an initial test of functional significance of the enlarged hypothalamic size and discriminant validity of this finding, we compared correlations of anxiety with hypothalamic size among simplex females and males, given that simplex females had significantly enlarged volume, whereas simplex males did not. We predicted the presence of a correlation among the simplex females but not the males. Results showed a Spearman’s corre-

---

**Figure 3.** (A) Localization of the hypothalamic increase in volume in patients with schizophrenia (SCZ) versus controls. (B) Patients with schizophrenia versus normal controls: a closer look at the anterior hypothalamus. In panels, slices 1–4 illustrate the most anterior 4 of 13 1-mm-thick coronal slices through hypothalamus beginning at the anterior commissure (AC). Images in each leftmost column refer to Figure 4, and the black line depicts the coronal level illustrated in the center and rightmost columns. The center column in either panel represents the amount of overlap at the indicated level, superimposed on a representative magnetic resonance (MR) image. Purple, area of common overlap; blue, areas of normal hypothalamus alone; red, areas of increased SCZ hypothalamus. Rightmost column in either panel contains anatomical sketches, at the same approximate level, adapted from an atlas (Mai et al. 1993 The MR image stands as the right hemisphere, and the sketch stands as the left.).
Table 2. Average Hypothalamic Volumes (in cc3) by Group and Sex in Subjects with Schizophrenia and Normal Controls

<table>
<thead>
<tr>
<th></th>
<th>Total n</th>
<th>Mean (SD)</th>
<th>Left Mean (SD)</th>
<th>Right Mean (SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Controls</td>
<td>27</td>
<td>.92 (.11)</td>
<td>.44 (06)</td>
<td>.48 (.06)</td>
</tr>
<tr>
<td>Male</td>
<td>21</td>
<td>.78 (.16)</td>
<td>.37 (08)</td>
<td>.40 (09)</td>
</tr>
<tr>
<td>Female</td>
<td>27</td>
<td>.92 (.14)</td>
<td>.44 (.07)</td>
<td>.48 (.08)</td>
</tr>
<tr>
<td>Simplex Patients</td>
<td>13</td>
<td>.88 (.08)</td>
<td>.42 (.04)</td>
<td>.46 (.05)</td>
</tr>
<tr>
<td>Male</td>
<td>32</td>
<td>1.00 (.12)</td>
<td>.47 (06)</td>
<td>.53 (07)</td>
</tr>
<tr>
<td>Female</td>
<td>16</td>
<td>.83 (.17)</td>
<td>.39 (08)</td>
<td>.44 (10)</td>
</tr>
</tbody>
</table>

Using multivariate GLM–CD, brain region by group effect was as follows: F(2,105) = 3.53, p = .03. Univariate GLM demonstrated significant increases between simplex females versus control females (at p < .03), multiplex males versus control males (at p < .006), and multiplex versus simplex males (at p = .01). Effect sizes were as follows. Compared with controls: simplex female patients, −.71; multiplex female patients, −.49; simplex male patients, −.035; multiplex male patients, −1.1.

GLM, general linear model; CD = correlated data.

Discussion

This is the first MRI study of hypothalamic volume in schizophrenia that demonstrates that patients have significantly larger volumes than normal controls. These abnormalities were more prevalent in women than men, more severe in multiplex cases, and relatively more pronounced on the right side. Consistent with this, we demonstrated a significantly larger hypothalamic volume in nonpsychotic first-degree relatives of schizophrenia cases. The effect was stronger in multiplex versus simplex relatives, as in patients. Further, there were no significant differences in hypothalamic volume in patients compared with in relatives. The validity of our findings is underscored given the comparability of the groups within sex, and the analyses held regardless of sex or family type, controlled for potential confounders.

These findings are consistent with a recent postmortem study of MB abnormalities in patients with schizophrenia (Briess et al. 1998), demonstrating significantly larger MB volume compared with normal controls. Both hemispheres were affected, although the difference was statistically significant on the left. Further, as with the Briess et al. patients, our findings suggest that MB nuclei were particularly affected in relatives. Finally, enlarged hypothalamic volumes in schizophrenia, particularly in women, may be related to increased pituitary volume abnormalities that are found in female first-episode patients (Pariente et al. 2004).

Why an Increase? Potential Explanatory Mechanisms

In general, enlarged hypothalamic volume may reflect increased number of neurons, size of neurons, or increased neuropil. This may result from a disruption in normal brain development. In animals, normal hypothalamic sexual dimorphism is associated with high density of gonadal hormones during hypothalamic development and adulthood (Gorski 2000; Park et al. 1996; Pilgrim and Hutchison 1994; Tobet et al. 1995). Hypothalamic nuclei and volume have been found primarily to be larger in males than females (Allen et al. 1989; Goldstein et al. 2001; Swaab and Flinters 1985; Zhou et al. 1995). One mechanism for hypothalamic sexual differentiation is the conversion of testosterone to estrogen by the enzyme aromatase (Kawata 1995; Park et al. 1996).

Table 3. Average Hypothalamic Volumes (in cc3) by Group and Sex in First-degree Relatives of Subjects with Schizophrenia

<table>
<thead>
<tr>
<th></th>
<th>Total n</th>
<th>Mean (SD)</th>
<th>Left Mean (SD)</th>
<th>Right Mean (SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Controls</td>
<td>27</td>
<td>.92 (.11)</td>
<td>.44 (06)</td>
<td>.48 (.06)</td>
</tr>
<tr>
<td>Male</td>
<td>21</td>
<td>.78 (.16)</td>
<td>.37 (08)</td>
<td>.40 (09)</td>
</tr>
<tr>
<td>Female</td>
<td>27</td>
<td>.92 (.14)</td>
<td>.44 (.07)</td>
<td>.48 (.08)</td>
</tr>
<tr>
<td>Simplex Patients</td>
<td>13</td>
<td>.88 (.08)</td>
<td>.42 (.04)</td>
<td>.46 (.05)</td>
</tr>
<tr>
<td>Male</td>
<td>32</td>
<td>1.00 (.12)</td>
<td>.47 (06)</td>
<td>.53 (07)</td>
</tr>
<tr>
<td>Female</td>
<td>16</td>
<td>.83 (.17)</td>
<td>.39 (08)</td>
<td>.44 (10)</td>
</tr>
</tbody>
</table>

Overall test of relatives versus controls, using GEE adjustment of standard errors for intrafamilial correlation: b = .07 (.03), z = 2.36, p = .01. Effect was significant for males and females. Effect sizes are as follows. Compared with controls: simplex female relatives, −.33; multiplex female relatives, −.52; simplex male relatives, −.65; multiplex male relatives, −.80. GEE = generalized estimated equations.
Figure 4. (A) Localization of hypothalamic increase in volume in first-degree relatives (1REL) versus normal controls. (B) First-degree relatives versus normal controls: a closer look at the posterior hypothalamus. In panels, slices 10–13 illustrate the most posterior 4 of 13 1-mm-thick coronal slices through hypothalamus, beginning at the anterior commissure (AC). The vertical black line in each leftmost column depicts the coronal level illustrated in the center and rightmost columns; the middle column in each panel represents amount of overlap at the indicated level, superimposed on a representative MR image. Light blue, area of normal hypothalamus alone; green, areas of increased first-degree relative hypothalamus. The rightmost column in each panel contains sketches, at the same approximate level, adapted from an atlas (Mai et al. 1993). The MR image stands as the right hemisphere, and the sketch stands as the left.
MacLusky *et al.* 1987). In our previous studies of the disruption in normal sexual dimorphisms in schizophrenia (Goldstein *et al.* 2002), we hypothesized that the disruption of midgestational aromatization in HPG and HPA systems may contribute to producing sex differences in brain abnormalities in schizophrenia. We suggest that a disruption in aromatization in males may lead to an increase in hypothalamic size in schizophrenia through its effect on developmental apoptosis or normal cell death, which are known to be regulated partly by testosterone (Dodson and Gorski 1993; Hammond *et al.* 2001) and estrogen (Kawata 1995). Recent studies demonstrated that testosterone has a protective effect on apoptosis (Dodson and Gorski 1993; Hammond *et al.* 2001). If developmental aromatization is disrupted in males, this may lead to less hypothalamic apoptosis and to an increase in hypothalamic neurons compared with normal men. Further, previous studies also have demonstrated a disruption in the HPG system in females, resulting in low estrogen and high androgens relative to normal population values (Canuso *et al.* 2002; Riecher-Rossler *et al.* 1994). Again, high testosterone in females with schizophrenia also may lead to disrupted normal apoptosis, resulting in an increase in hypothalamic size compared with normal women.

We also suggested that the PVN may be particularly enlarged in patients compared with controls. PVN has a high density of CRH, which is involved in the release of norepinephrine, both of which are implicated in the stress response. CRH neurons in the PVN colocalize with sex-hormone receptors, such as estrogen receptors (ERx and ERβ), suggesting a role for ERs in the PVN release of CRH (Bao *et al.* 2005). Further, in major depression, CRH neurons in PVN that colocalized with ERs were significantly greater in number than in normal controls, and in part explained greater PVN neuronal density and size (Bao *et al.* 2005), findings that may have direct implications for our schizophrenia study. In addition, abnormalities in numbers of vasopressin and oxytocin neurons and levels were found in schizophrenia, regardless of medication (Beckmann *et al.* 1985; Legros *et al.* 1992; Mai *et al.* 1993).

Previous animal studies demonstrated that inhibiting aromatase resulted in a rise in norepinephrine in the cortex (Stewart and Rajabi 1994). In fact, early postmortem schizophrenia studies found significantly higher norepinephrine levels in the limbic forebrain than normal controls (Farley *et al.* 1978). Several studies also have reported higher rates of baseline cortisol activity in schizophrenia and hyperresponsivity to stress (Fredrikson *et al.* 1995; Walker and Diforio 1997; Williams *et al.* 2004). Our finding of enlarged hypothalamic volume (particularly PVN) is consistent with this, also given that higher levels of anxiety were associated with larger hypothalamic volumes.

It is unlikely that antipsychotic medication explained the enlarged hypothalamic volume. Medication levels did not significantly relate to hypothalamic size, and most important, enlarged hypothalamic volumes were present in nonpsychotic relatives who never had taken antipsychotic medications. Preclinical studies in monkeys that were chronically exposed to typical and atypical antipsychotic medications have reported an effect of decreased, not increased, volumes of various brain regions (Dorph-Petersen *et al.* 2005). In contrast, another monkey study reported increased cortical glial density with long duration of neuroleptics (Selemmon and Goldman-Rakic 1999). Increased glial density could result in enlarged regional volumes, given the substantial number of white-matter fibers entering and exiting hypothalamic nuclei as well as their interconnectivity. However, no one has investigated these issues with regard to hypothalamic nuclei. Further, our findings of enlarged hypothalami in unmedicated relatives would not be explained by medication effects on glial density.

Enlarged hypothalamic volumes also most likely were not a result of any potential alcohol confounds, given that recent studies reported decreased hypothalamic volume of the mamillary bodies in patients with various alcohol disorders compared with normal controls, in contrast to the enlarged hypothalamic volume found in our study (Shear *et al.* 1996; Sullivan *et al.* 1999). In fact, enlarged hypothalamic volumes were replicated in four independent samples in our study: simplex and multiplex patients and simplex and multiplex relatives.

Genetic Vulnerability Implications

Our findings in first-degree relatives suggest that they may represent part of the vulnerability to schizophrenia rather than be a reflection of the psychosis per se. The relatives’ results are underscored by the linear effect found in multiplex versus simplex patients, with a greater volumetric size in multiplex than simplex families in whom genetic factors are more likely presumed. We and other investigators have demonstrated abnormalities in nonpsychotic relatives in other limbic and paralimbic regions that have been implicated in schizophrenia (e.g., Faraone *et al.* 2000; Keshavan *et al.* 1997; Seidman *et al.* 2002, 2003), which have strong connections with the hypothalamus. Findings presented here extend the description of the genetic vulnerability to include the hypothalamus, a critical brain region in controlling appetitive drives, affect, the reward system, and endocrine control.

Our findings also may provide leads to hypotheses about what is transmitted in schizophrenia. The MBs, in which relatives showed the greatest volumetric abnormality, excite the adrenergic system and regulate prolactin and luteinizing hormone via inhibition (Beltramino and Taleisnik 1984; Caceres and Taleisnik 1982; Ivanisevic-Milovanovic and Musicki 1992). Lesioning of MBs in animals produced an increase in, and stimulation produced suppression of, plasma corticosterone levels (Feldman *et al.* 1976; Suarez and Perassi 1988), which was not surprising given the high density of CRF receptors in MBs. MBs also receive projections from, among others, the anterior hypothalamus (including PVN), which also is dense in CRF receptors and was enlarged significantly in patients. Thus, MBs play a role in arousal and may be a site for antianxiety action of benzodiazepines (Kataoka *et al.* 1982). Elevated levels of cortisol have been found in schizotypal personality-disorder patients, underscoring the premise that adrenal dysfunction may be part of schizophrenia vulnerability rather than a result of psychosis or medication effects (Hansen *et al.* 1985). Overall, our results suggest that etiologic factors that explain abnormalities in the HPA system in schizophrenia are established during development and genetically transmitted, rather than a consequence of the psychosis per se.

In summary, this is a unique MRI study demonstrating significant abnormal hypothalamic enlargement in schizophrenia and nonpsychotic first-degree relatives. The patients’ abnormalities were greater in females than males, greater in multiplex than simplex cases, and present in nonpsychotic first-degree relatives. Normal hypothalamic sexual dimorphism was disrupted. Hormonal mechanisms involved in the regulation of hypothalamic development may have implications for the consequences from midgestational insults implicated in schizophrenia that occur simultaneously with the sexual dimorphism of the hypothalamus, a hypothesis that is supported by data in Goldstein *et al.*
This work was supported by grants from the National Institute of Mental Health, ROI MH50695 (to JMG, which was, in part, supported by the National Institutes of Health Office of Research on Women’s Health), and by Merit Award MI 43518 and MI 46318 (MTT) and Stanley Medical Research Institute (to LJS). We thank Martha Shenton, Ph.D., for help with ascertainment scans for our final three multiplex subjects and thank Valerie Thompson for manuscript preparation. TA currently is affiliated with the Department of Neuroscience, Emory University, Atlanta, Georgia.


Beltramino C, Taleisnik S (1984): Inhibitory influence of the nuclei of the amygdala and the PVN, associated with anxiety, also suggest that these abnormalities may help to explain high rates of endocrine disorders in schizophrenia (Canuso et al. 2002; Oades and Schepker 1994; Riecher-Rossler et al. 1994; Wechsler et al. 1981).


Faraone SV, Seidman LJ, Kremen WS, Toomey R, Pepple JR, Tsuang MT (2001, 2002). These abnormalities are established during development, because our findings suggest that they are genetically transmitted and present in nonpsychotic first-degree relatives.


