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Genetic Influences on Hippocampal Structure and Function in Recombinant Inbred Mice

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Abstract

Previously, we identified separate genetic influences on ventral versus dorsal hippocampal volume in BXD recombinant inbred mice [27]. Based on genotype at genetic markers associated with ventral hippocampal volume, we evaluated BXD mouse strains with relatively small versus large ventral hippocampal volumes using numerous behavioral paradigms known to rely upon hippocampal function and several other tasks that tap into behaviors analogous to those often impaired in schizophrenia. We observed a relationship between genotype at markers known to influence ventral hippocampal volume and working memory at an intermediate memory load. There was no association between genotype at markers known to influence ventral hippocampal volume and spatial reference memory, prepulse inhibition, or elevated plus maze performance. The relevance of these findings for understanding the pathophysiology of schizophrenia are discussed, including the possibility that genetic predisposition toward anterior hippocampal volume reductions and working memory deficits in schizophrenia may be related through a shared genetic locus.

Keywords

hippocampus; QTL; structure/function relationships; recombinant inbred mice

Introduction

A substantial proportion of normative variation in neuroanatomical structure is due to genetic factors [53]. A growing number of single nucleotide polymorphisms have been identified that influence hippocampal volume in the mammalian brain [8,10,34,40,42]. Using recombinant inbred (RI) strains of mice, we identified quantitative trait loci (QTLs) that differentially influenced ventral (anterior) and dorsal (posterior) hippocampal volume [27]. Using strains of BXD and AXB/BXA RI mice, QTLs were found on chromosomes 2, 8 and 13 that influenced ventral hippocampal volume. In contrast, QTLs were found on chromosomes 5, 13 and 15 that influenced dorsal hippocampal volume. The identification of discrete QTLs that uniquely influence ventral and dorsal hippocampal volume is consistent with observations that these two anatomical subregions serve distinct functions [3].

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There is increasing interest in the relationship between neuroanatomical structure and function [2] [51] [1] [32]. While there have been systematic investigations evaluating the role of genetics on hippocampal volume, few studies have addressed the functional consequences of such genetic influences. Much of the literature in the area of structure-function relationships has focused on the consequences of experimentally-induced physical changes in neuronal activity using animal models. These alterations have been induced largely with chemical or electrolytic lesions, rather than the effects of structural differences due to normative genetic variation [3] [21] [45] [4] [33]. Similarly, many of the relevant human studies have targeted relationships between hippocampal structure and function that emerge in the context of neural insults or neurodegenerative illnesses, such as Alzheimer's disease [for review, see [47]].

In the present study, we were interested in examining the effects of differences in ventral hippocampal volume due to spontaneous genetic variation on targeted behaviors. We tested the general hypothesis that polymorphisms at QTLs shown to influence variation in hippocampal volume in RI strains of mice would also influence performance in behavioral tests generally agreed to be hippocampal-dependent [3]. Specifically, we selected strains predicted to have large or small ventral hippocampal volumes [27], and characterized these strains using a battery of behavioral tasks thought to be dependent on the ventral, dorsal and entire structure of the hippocampus. We were particularly interested in studying the functional consequences of genetic influences on ventral hippocampal volume because of the putative dysfunction of the ventral or anterior hippocampus in schizophrenia [for review see [13] [43] [19]]. For the same reason, we also assessed performance on behavioral tasks tapping into constructs believed to be analogous to those cognitive impairments often observed in patients with schizophrenia (e.g., prepulse inhibition of the acoustic startle response) [6]. We hypothesized that smaller volumes would be associated with deficits in working memory, decreased prepulse inhibition and increased anxiety levels.

Materials and Methods

Experimentally-naïve adult mice from ten BXD strains (N = 92) served as subjects for these experiments. Based upon results of our previous study of QTLs that influenced hippocampal volume [27], we selected five strains predicted to have large ventral hippocampal volumes and five strains predicted to have small ventral hippocampal volumes based jointly on genotype at two ventral hippocampal volume QTLs, marker *D8Mit31* on chromosome 8 and SNP 13.067.300 on chromosome 13 (see Table 1). Six to twelve animals (all males) were used per strain. Animals arrived from Jackson Laboratories at two months of age and were housed in groups of 3–4 until they were tested at four to five months of age. Subjects were maintained on a 12:12 light/dark cycle with food and water available *ad libitum*. Considering the well established effects of estrous cycle on behavior, we strategically used only males in this study to decrease overall error variance and enhance test sensitivity. All experimental procedures involving animals were performed in accordance with guidelines established by the Animal Studies Committee at Washington University.

Behavioral Tests

Strains were characterized using tasks known to rely upon ventral hippocampal function (spatial working memory and elevated plus maze activity), a dorsal hippocampal-dependent task (spatial reference memory). In addition, prepulse inhibition of the acoustic startle response performance was evaluated, as this task taps into constructs of cognition that have been repeatedly shown to be disturbed in schizophrenia [3] [6] [17] [30] [24]. Locomotor activity was also evaluated since hyperactivity is frequently cited as a component of the behavioral phenotype analyzed in mouse models of schizophrenia [16,31]. Testing order was consistent across animals.

8-Arm Radial Water Maze

Work in rats [29], monkeys [11] and humans [35] suggests that the dorsal (posterior) hippocampus processes spatial reference information. In contrast, spatial working memory appears to be more dependent on ventral (anterior) hippocampal function. In one study of healthy human subjects, it was reported that activation of left anterior hippocampus was associated with maintenance of information during a working memory task [33]. Similarly, Zarahn et al (2005) [54] observed that the hippocampus is preferentially activated when demands on working memory are moderate, but not when these demands are minimal or excessive. Interestingly, both working memory impairments and anterior hippocampal volume deficits have been reported in patients with schizophrenia [13] [30] [24] and their non-psychotic siblings [7,43]. In the present study, spatial reference and working memory were simultaneously assessed using an 8-arm radial water maze, as described previously [22] [18] [5].

The maze was located in a room with distinct extra-maze visual cues. The maze was filled with water at least 12-hours prior to testing and was made opaque with non-toxic white Tempera paint. Escape platforms were submerged just below the surface of the water in the distal ends of four arms. Platforms were not located in three adjacent arms or in the start arm and platform location was consistent across animals and across the daily sessions. For each trial, a mouse was released from the start arm and permitted 2 minutes to find a platform. Once a platform was located, the animal remained on it for 15 seconds. If the subject failed to locate a platform before the trial expired, it was guided gently to the nearest platform. The mouse was then returned to its heated home cage for 10 seconds during which time the platform recently found was removed from the maze for that session until all of the platforms were found. This strategy of incrementally increasing the demands on working memory across successive trials within a day, while keeping demands on reference memory constant permitted the simultaneous assessment of working and reference memory [23]. Animals were tested for 12 consecutive days with days 1 and 2 considered training days. Our analysis focused on data from the terminal ten days of testing. Data were blocked by averaging the number of errors on the first five and last five days to permit trend analysis without the undue influence of individual variability.

Acoustic Startle Response and Prepulse Inhibition of the Acoustic Startle Response

The acoustic startle reflex (ASR) is a contraction of the skeletal and facial musculature in response to a sudden, brief, intense auditory stimulus that is elicited by a loud tone in both humans and rodents. Prepulse inhibition of startle (PPI) is a reduction in the ASR amplitude that occurs when the startling stimulus is preceded 30 – 500 ms by the presentation of a weaker auditory stimulus. Although the startle responses measured are different across species, (eye blink in humans and whole body flinch in rodents), virtually identical stimulus parameters are used, and considerable evidence supports a high degree of homology in startle/PPI measures between rodents and humans [39] [6]. Of relevance to the present work, patients with schizophrenia exhibit little change in the ASR to repeated presentations of a startling stimulus, whereas healthy controls quickly habituate this response. In addition, impairments in PPI have been reported a host of disorders such as schizophrenia, Huntington's disease, Tourette syndrome, and several other neuropsychiatric illnesses [39]. PPI is an often-used measure in rodent models of schizophrenia, and recent work suggests that it may be dependent on both dorsal and ventral hippocampal functions [for review, see [4]].

The ASR/PPI apparatus consisted of a single holding unit housed inside a sound-attenuating, lighted chamber (Hamilton-Kinder, LLC, Poway, CA). Units were constructed to contain, but not overly stress, rodents during the procedure.. Testing was conducted using our previously published methods for assessing the ASR and PPI in mice [20]. Mice were acclimated to 70 decibel (dB) background white noise for five minutes and then exposed them to five

consecutive startle trials in which the ASR was measured in response to a noise pulse (40 ms broadband 120 dB burst). Subjects were then exposed to ten blocks of five different types of trials, including: a startle trial (120 dB pulse alone); no stimulus; or three different types of PPI trials during which a 20-ms long prepulse stimulus (4, 12 or 20 dB above background noise) was followed by the startle stimulus (120 dB pulse). The session ended with five consecutive startle trials. Maximum force generated by each subject at the onset of the 120 dB startle stimulus was recorded. The ASR was evaluated by comparing the means from the four blocks (5 trials per block) of startle trials, while habituation of the ASR was assessed by comparing means from the first and last startle blocks. The degree of PPI induced was evaluated by analyzing percent PPI scores for each mouse and each PPI trial type. Percent PPI scores were derived using a mathematical transformation comparing the amplitude of the response resulting from a given PPI (prepulse + pulse) trial with the mean amplitude of the middle 10 startle (pulse alone) trials, and deriving a percentage of reduction (or increase) in response [see [20] for greater details].

Elevated Plus Maze

Performance on the elevated plus maze has been associated with ventral hippocampal volume and has been interpreted as a measure of anxiety-related behaviors. The elevated plus maze was constructed from black Plexiglas and consisted of two opposing open arms and two opposing closed arms ($35 \times 15 \times 6.1$ cm), equidistantly spaced, that extended from a central platform (5.5×5.5 cm). A total of 32 photoelectric beams were configured along the X- and Y-axis and beam breaks were recorded by a computer and interface assembly (Hamilton-Kinder, LCC, Poway, CA). Anxiety-like behaviors were evaluated using our previously published protocol [36], which involved placing a mouse in the central area of the plus maze and recording its behavior through the analysis of beams broken during a 5-minute test session. Mice were tested over three consecutive days to assess the extent to which the mice exhibited avoidance of the open arm across the test sessions. Dependent variables that were analyzed included percentage of total entries made into the open arms, percentage of time spent in the open arms, and percentage of distance traveled in the open arms.

Locomotor Activity

Locomotor hyperactivity is often considered a component of the behavioral phenotype of schizophrenia [28,38]. This stems from findings relating the effects of hallucinogenic drugs on monoaminergic systems and locomotion in rodents, and the blockade of such effects by antipsychotic drugs. Interestingly, rats with neonatal excitotoxic ventral hippocampal lesions show locomotor hyperactivity in response to novelty by around two months of age [25,49]. For these reasons and to assess the possibility that performance differences on the radial water maze or the elevated plus maze might be due to non-specific differences in locomotion, subjects were evaluated on a one-hour activity test between the hours of 0800–1200. Mice were placed individually in transparent polystyrene cages ($47.6 \times 25.4 \times 20.6$ cm) where each cage was surrounded by a 4×8 matrix of photocell pairs which were used to quantify activity (MotorMonitor, Hamilton-Kinder, LLC, Poway, CA), as previously described [52]. The system software also defined a 33×11 cm central zone and a peripheral zone that was 5.5 cm along the sides of the cage, and behaviors were analyzed in these zones as well. We assessed the total number of ambulations, rearings, distance traveled in the peripheral zone as well as entries made, time spent and distance traveled in the center of the open field.

Structural confirmation of genotype effects

To confirm the expected effects of genotype on ventral hippocampal volume we measured ventral hippocampal volume in all animals. Following anesthesia, animals were perfused with 0.1 M heparanized phosphate buffered saline for two minutes followed by approximately 75

ml 2.5% glutaraldehyde and 2% paraformaldehyde in 0.1 M PBS for ten minutes. Brains were removed from the skull and weighed after removing the olfactory bulb. The tissue was then post-fixed for 48 hours in a solution of 2.5% glutaraldehyde, 2% paraformaldehyde, and 30% sucrose in 0.1M PBS, embedded in Tissue-Tek (Electron Microscopy Sciences, PA) and sectioned in the horizontal plane on a cryostat at -20 degrees C. Every fifth section was mounted on 2% porcine skin coated slides and Nissl-stained. Slides were coverslipped, sections were imaged and files were formatted for stereological measurement of volume in Analyze 7.0, as described previously [27].

Data Analysis

Data were analyzed by repeated measures ANOVA with strain nested within genotype and with day and/or trial as repeated measures variables, or by ANOVA with strain nested within genotype using SAS v8. Data were analyzed by ANOVA with strain nested within genotype to avoid bias related to strain-dependent variation in hippocampal volume. To simplify the reference and working memory data, we grouped the data into two blocks by averaging the first five and last five days. Three animals were not included in the structural analyses due to histological complications.

Results

Genotype Effects on Volume

Age ($F(1,88) = 0.16$, $p = 0.69$) did not predict ventral hippocampal volume. Therefore, ventral hippocampal volume was not corrected for age-related variation. As expected, there was a significant effect of genotype on ventral hippocampal volume ($F(1,88) = 4.91$, $p = 0.029$).

Reference Memory

We blocked data into two five-day blocks. To examine whether the animals showed improvement over the ten-day testing period, we examined the effect of block of the total number of reference memory errors. The total number of reference memory errors was analyzed by ANOVA with strain nested within hippocampal volume QTL genotype. There was a significant effect of block ($F(2,183) = 13.93$, $p = 0.0002$) and a significant effect of strain ($F(8,183) = 2.31$, $p = 0.0185$) on the total numbers of reference memory errors committed. The significant effect of block indicates a general decrease in reference memory errors over time, showing improved performance on the part of the mice, and suggesting learning had occurred. Hippocampal volume QTL genotype was not significantly related to reference memory performance.

Working Memory

The nature of the task is such that the difficulty is incrementally increased with each successive trial. We examined each effect of block, genotype and strain on performance independently by ANOVA with strain nested within genotype. The ANOVA revealed a significant genotype X trial interaction effect on working memory performance ($F(1,183) = 3.41$, $p = 0.03$), and thus we proceeded to analyze performance across trials. This analysis showed a significant effect of block on working memory performance on trials 2 and 3 (suggesting learning had occurred), but not trial 4 (Trial 2: $F(1,183) = 4.55$, $p = 0.03$; Trial 3, $F(1,183) = 6.35$, $p = 0.01$; Trial 4: $F(1,183) = 0.03$, $p = 0.86$). There was a highly significant effect of hippocampal volume QTL genotype ($F(1,183) = 13.38$, $p = 0.0003$) and strain ($F(1,183) = 6.30$, $p < 0.0001$) on performance during trial 3. Likewise, there was a significant effect of strain on performance on trial 4 ($F(1,183) = 16.4$, $p < 0.0001$). No other statistically significant effects of strain or genotype were detected for working memory performance (see Figure 1).

Correlation between Water-Maze Performance and Ventral Hippocampal Volume

The Pearson correlation between ventral hippocampal volume and block 1 and block 2 total reference memory errors was 0.06 ($p = 0.57$) and 0.27 ($p = 0.01$), respectively. The Pearson correlation between ventral hippocampal volume and total number of errors during working memory was not significant for any trial (trial 2, block 1 – $r = 0.04$ ($p = 0.74$); trial 2, block 2 – $r = 0.06$ ($p = 0.06$); trial 3, block 1 – $r = 0.04$ ($p = 0.68$); trial 3, block 2 – $r = 0.11$ ($p = 0.30$); trial 4, block 1 – $r = 0.13$ ($p = 0.24$); trial 4, block 2 – $r = 0.07$ ($p = 0.50$)).

Acoustic Startle Response and Pre-Pulse Inhibition

There was a significant effect of strain ($F(8,83) = 4.19$, $p < 0.001$), but not genotype, on the magnitude of the ASR, and a significant effect of strain ($F(1,83) = 3.27$, $p = 0.0028$), but not genotype, on habituation of the ASR as well. In contrast, there were no statistically significant effects of strain or genotype on PPI. There was not a significant correlation between ventral hippocampal volume on any of the variables.

Elevated Plus Maze

There was a significant effect of trial on the percentage of arm entries made into the open arms ($F(2,86) = 22.30$, $p < 0.001$), the percentage of total arm time spent in the open arms ($F(2,86) = 17.25$, $p < 0.001$) and the percentage of total distance traveled in the open arms ($F(2,86) = 12.88$, $p < 0.001$). In addition, there was a significant effect of strain on these three variables [$F(8,83) = 7.36$, $p < 0.0001$]; ($F(8,83) = 4.81$, $p < 0.001$); ($F(8,83) = 7.13$, $p < 0.0001$), respectively], although there was no effect of hippocampal volume QTL genotype on elevated plus maze performance. There was a significant negative correlation between ventral hippocampal volume and percentage of distance traveled in the open arms ($r = -0.25$) and time ($r = -0.22$, $p = 0.03$). No other elevated plus variables were significantly correlated with ventral hippocampal volume.

Locomotor Activity

We observed a significant effect of strain on total number of ambulations ($F(8,83) = 8.13$, $p < 0.0001$), but no significant effect of genotype, while the effects of strain and genotype were both without effect on the frequency of rearing. There was also a significant effect of strain on the number of entries made into the center of the open field ($F(8,83) = 7.76$, $p < 0.0001$), time spent in the center ($F(8,83) = 2.95$, $p = 0.0061$), and distance traveled in the center ($F(8,83) = 7.91$, $p < 0.0001$). In addition, a significant effect of genotype was found for time spent in the center ($F(1,87) = 4.72$, $p = 0.033$), and for distance traveled in the center ($F(1,87) = 4.21$, $p = 0.043$), with smaller hippocampal volume being associated with decreased activity in the center of the open field (suggesting increased anxiety). However, there were no effects of genotype on number of entries made into the center of the open field ($F(1,87) = 2.399$, $p = 0.124$). The direct correlation between ventral hippocampal volume and the total number of ambulations was not significant ($r = 0.18$, $p = 0.11$).

Discussion

The focus of this study was to test the hypothesis that QTLs that influence ventral hippocampal volume would also influence behaviors usually associated with ventral hippocampal function. As hypothesized, animals with genotypes associated with larger ventral hippocampal volumes showed significantly better working memory performance, but only during moderate memory. As hypothesized, there was no overall effect of ventral hippocampal volume QTL genotype on spatial reference memory on any trial. However, contrary to our hypotheses, genotype at ventral hippocampal volume QTLs did not predict degree of PPI, magnitude or habituation of the ASR, level of anxiety-like behaviors in the elevated plus maze, or alterations in general

locomotor activity. There were no significant correlations between ventral hippocampal volume and working memory performance on any trial, although there was an unexpected significant positive correlation between ventral hippocampal volume and reference memory errors, with larger volume being associated with more errors. The finding of no significant correlation between ventral hippocampal volume and working memory performance is problematic since it suggests that either the influence of genotype on working memory is not directly mediated through ventral hippocampal volume, or that a truncation in the range of hippocampal volumes may have led to spuriously low correlations.

The results of *in vivo* functional neuroimaging studies of human subjects performing working memory tasks suggest that ventral (anterior) hippocampal projections to the prefrontal cortex are required for normal working memory function, as both of these regions show activation during working memory tasks [9]. Moreover, Fos imaging in mice has demonstrated activation of both the dorsal and ventral hippocampal during spatial memory tasks. [48] and work in rodents suggests the ventral hippocampal-PFC pathway is important for long-term working memory in the radial arm [15] and T-maze tasks [50]. Another recent study suggested the hippocampus is strategically recruited during non-spatial tasks that involve working memory, especially at intermediate memory loads. This is in contrast to the engagement of the dorsolateral prefrontal cortex, which appears to be active at all working memory loads [54]. Our finding of no effect of ventral hippocampal volume on spatial reference memory was not surprising, as studies in humans suggests a relationship between posterior, but not anterior, hippocampal volume and spatial memory performance [26].

Gene variants shown to influence hippocampal volume also have been shown to influence working memory performance in separate studies. The catechol-O-methyltransferase (COMT) val^{108/158}met polymorphism was shown to influence verbal working memory in healthy controls, with subjects homozygous for the val allele performing most poorly on the n-back [14]. The same polymorphism also was shown to influence hippocampal and prefrontal gray matter volume in a separate study, which reported a positive correlation between the number of met genes and hippocampal gray matter (Cerasa 2008). DISC1 variation also was shown to influence gray matter volume of the hippocampus and hippocampal bold fMRI during working memory tasks. Our finding of a shared genetic influence on both ventral hippocampal volume and working memory performance is consistent with these findings.

Recent work has implicated the hippocampus as an important modulator of startle and PPI [4], although the relationship between hippocampal volume and the capacity for sensorimotor gating has not been studied. Our finding that genotype was not related to PPI suggests that normative variation in hippocampal volume in mice does not influence this behavior, or that if volume/function relationships exist they are not influenced by the genotypes that were the focus of this study. In the current study, anxiety-like behavior in the elevated plus maze was assessed by measuring the percent of total arm entries made into the open arms, the percent of total arm time spent in the open arms, and the percent of total arm distance traveled in the open arms. There was a statistically significant effect of strain on each of these three variables indicating genetic variation between strains. However, we found no relationship between ventral hippocampal volume QTL genotype and anxiety-like behaviors as measured by elevated plus maze. When we assessed anxiety-related behaviors in the open field, we observed a significant effect of genotype on two out of the three open field variables. Specifically, we found that animals with smaller hippocampal volumes spent less time in and traveled a shorter distance in the center of the field (i.e., were more anxious) than animals with larger hippocampal volumes. However, support for a significant effect of genotype on anxiety-like behavior was not wholly consistent, since there was no effect of genotype on the number of entries into the center of the field. As with the elevated plus maze, we also observed a significant strain effect on anxiety-related behaviors in the open field. Taken together, the results of this

study suggest the relationship between hippocampal volume and anxiety measures may be both strain- and task-dependent. Alternatively, it is possible that this relationship is also affected by environmental influences (e.g., stressors).

To account for the confounding effects of differences in locomotor activity on elevated plus maze activity or water maze performance, we assessed ambulatory behavior over a one-hour period. In the current study, there was a significant effect of strain, but not genotype, on locomotor activity. These results suggest that differences in working memory performance in animals predicted to have large versus small ventral hippocampal volumes are not attributable to differences in locomotor activity.

Limitations of the current study include the small sample size (only 5 strains per genotype) and the fact that only male animals were used. It is possible that volume QTLs could influence behavior differently in males than in females. One short-coming of this work was the variation in number of animals representing each strain. It is possible that the variation in the number of animals used per strain may have biased results. Further, we only examined the effects of two ventral hippocampal volume QTLs on behavior. Other genetic influences on volume are likely to exist and influence behavior in yet uncharacterized ways. An additional limitation of the current study was that we did not examine environmental influences on hippocampal volume. This initial study was not suitable for such analyses, as animals were not reared under systematically variable housing conditions nor did we systematically vary maternal influences (i.e., animals within strains were reared by dams from the same strain). In future work, the effects of differences in hippocampal volume on behavior could be studied in isogenic animals reared in systematically different environments.

These results may have important implications for our understanding of schizophrenia. The results of numerous studies suggest that there is an association between hippocampal neuropathy and schizophrenia (for review, see [19]). Hippocampal volume reductions are present in both discordant monozygotic twins [37], and non-affected siblings of patients with schizophrenia [43], suggesting a genetic basis for its development. Recent studies suggest that this volume reduction is selectively pronounced in the anterior hippocampus of subjects with schizophrenia [13] and their relatives [43]. In turn, the heritability of working memory has been estimated at 36% [46], and the non-psychotic relatives of schizophrenics also display working memory deficits, relative to normal controls [44]. Thus, the results of the present study support the hypothesis that the differences in hippocampal volumes and working memory deficits observed in schizophrenia may have a shared genetic basis, at least in part. Among genes that could influence both hippocampal volume and spatial working memory performance, it is interesting to consider *Drebrin*, which is also located within the ventral hippocampal volume QTL on chromosome 13. *Drebrin* is signal transduction molecule that appears to play a role in hippocampal development and dendritic spine morphogenesis [41]. Moreover, *Drebrin* expression has also suggested to play a role in the neural mechanisms of cognition and reductions in *Drebrin* expression has been reported to be associated with cognitive decline in elderly subjects [12].

We observed several strain effects that were independent of the genotypes used for strain selection. Measures of anxiety in both the elevated plus maze and open field were strain dependent in BXD mice. Reference memory and working memory performance were also influenced by strain. These results suggest the behavioral tests used in the current study would be useful in QTL mapping of anxiety, reference memory and working memory using an expanded number of BXD strains.

Further research is needed to determine whether a single gene or multiple genes within the QTLs selected are responsible for the effects observed in this study. Future fine-mapping of

such loci may lead to the identification of novel susceptibility genes for schizophrenia and may aid in our understanding of the basis of neuroanatomical abnormalities and cognitive dysfunction in schizophrenia.

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Figure 1.

Genotype A was associated with smaller ventral hippocampal volume and the DBA and C57 genotypes at marker D8Mit30 and SNP13.067.300, respectively. Genotype B was associated with larger hippocampal volume and the C57 and DBA genotype at marker D8Mit30 and SNP 13.067.300, respectively. No significant effects of genotype were observed on trial 2 or 4, but there was a significant effect of genotype on the number of working memory errors committed on trial 3.

Table 1
Subjects. Total Number of Animals Per Strain and Genotype Used in Behavioral Tests

	Genotype at Marker D8Mit30	Genotype at SNP 13.067.300
<u>Small Volumes Strains</u>		
BXD1 (n = 10)	DBA	C57
BXD5 (n = 6)	DBA	C57
BXD19 (n = 9)	DBA	C57
BXD22 (n = 6)	DBA	C57
BXD40 (n = 10)	DBA	C57
<u>Large Volume Strains</u>		
BXD6 (n = 10)	C57	DBA
BXD12 (n = 10)	C57	DBA
BXD14 (n = 10)	C57	DBA
BXD33 (n = 12)	C57	DBA
BXD28 (n = 9)	C57	DBA