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Association of *AKT1* With Verbal Learning, Verbal Memory, and Regional Cortical Gray Matter Density in Twins

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Abstract

AKT1, encoding the protein kinase B, has been associated with the genetic etiology of schizophrenia and bipolar disorder. However, minuscule data exist on the role of different alleles of in measurable quantitative endophenotypes, such as cognitive abilities and neuroanatomical features, showing deviations in schizophrenia and bipolar disorder. We evaluated the contribution of *AKT1* to quantitative cognitive traits and 3D high-resolution neuroanatomical images in a Finnish twin sample consisting of 298 twins: 61 pairs with schizophrenia (8 concordant), 31 pairs with bipolar disorder (5 concordant) and 65 control pairs matched for age, sex and demographics. An *AKT1* allele defined by the SNP rs1130214 located in the UTR of the gene revealed association with cognitive traits related to verbal learning and memory (P=0.0005 for a composite index). This association was further fortified by a higher degree of resemblance of verbal memory capacity in pairs sharing the rs1130214 genotype compared to pairs not sharing the genotype. Furthermore, the same allele was also associated with decreased gray matter density in medial and dorsolateral prefrontal cortex ($P < 0.05$). Our findings support the role of *AKT1* in the genetic background of cognitive and anatomical features, known to be affected by psychotic disorders. The established association of the same allelic variant of *AKT1* with both cognitive and neuroanatomical aberrations could suggest that AKT1 exerts its effect on verbal learning and memory via neural networks involving prefrontal cortex.

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Keywords

AKT1; quantitative trait loci; magnetic resonance imaging; association

INTRODUCTION

Studies seeking to discover the molecular background of schizophrenia (SCZ) and bipolar disorder (BPD) have yielded encouraging findings of linkage and association for several loci and genes but only few findings have been consistently replicated across studies. Further, the existing findings have typically been associated with only nominal odds ratios [Craddock et al., 2005]. This has triggered a hypothesis that multiple genetic variants, each possessing only modest increase in risk, underlie the genetic etiology of SCZ and BPD. The traditional diagnostic classification of psychiatric diseases does not necessarily reflect the biological background of the diseases and may thus be insensitive to identify such variants. On the contrary, quantitative endophenotypes potentially have more direct association with the underlying biology [Gottesman and Gould, 2003]. Impairments in cognition are found more frequently in cases and their unaffected siblings compared to control subjects suggesting a manifestation of increased genetic risk. Thus, these functions are broadly accepted to serve as endophenotypes for psychotic disorders [Bearden and Freimer, 2006; Tan et al., 2008a]. Additionally, anomalous neural development has been suggested to play a major role in the vulnerability to SCZ [Pantelis et al., 2005; Rapoport et al., 2005; Schmidt-Kastner et al., 2006]. Subtle abnormalities in the brain predisposing to SCZ may manifest themselves in cognitive impairments detected already before the disease onset [Pantelis et al., 2005]. Consequently, by utilizing cognitive test measures and structural 3D MRI brain images as phenotypes in genetic analyses, we can potentially reveal causal relationships between biological mechanisms and neural phenotypes associated with both SCZ and BPD [Cannon, 2005].

V-akt murine thymoma viral oncogene homolog 1 gene (*AKT1*) encodes a serine/threonine kinase and is highly expressed in brain where the protein is involved in variety of cellular processes such as neuronal proliferation and maintenance [Wang et al., 2003]. Previously *AKT1* has been reported to be associated with predisposition to SCZ, and impaired function of the AKT1-GSK3β pathway has been related to the pathogenesis of the disorder [Emamian et al., 2004]. Several replications have confirmed *AKT1* as a plausible candidate gene for SCZ [Ikeda et al., 2004; Schwab et al., 2005; Bajestan et al., 2006; Norton et al., 2007]. Some genetic data also supports the role of *AKT1* in the etiology of BPD [Toyota et al., 2003]. Intriguingly, akt1 has been also related to spatial memory functions in rodents [Mizuno et al., 2003].

This study was designed to investigate whether specific alleles of *AKT1* influence quantitative cognitive and neuroanatomical measures related to the genetic risk for SCZ and BPD. The study sample consisted of healthy twins as well as concordant and discordant BPD and SCZ twin pairs, representing a wide cognitive spectrum ranging from normal performance to severe impairments [Seidman et al., 2002; Pirkola et al., 2005]. The twin sample, in which each pair shares fetal and early life events, enabled us to minimize the “noise” caused by environmental factors, representing the best possible harmonized human study design tuned to address the multifactorial nature of the traits.

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MATERIALS AND METHODS

Subjects

The study protocol was reviewed and approved by the institutional review boards of the National Public Health Institute, Helsinki, Finland, and the University of California-Los Angeles, and all subjects signed institutional review board—approved informed consent forms.

The SCZ cases were identified by combining data from three national registers, including the Finnish National Hospital Discharge Register, that were screened for any psychiatric indication. Twin pairs were identified using a twin cohort of all same sex twins born in Finland between 1940 and 1957, originally identified from the national population register of Finland. Twin pairs with at least the other co-twin affected with SCZ or schizoaffective disorder were included as SCZ pairs. Pairs with no history of psychiatric indication were sampled from the remaining twin cohort members and included as control pairs [Cannon et al., 2000]. A detailed description of the sample collection procedures has been previously presented [Cannon et al., 2000]. BPD twin pairs were collected separately from SCZ and control pairs, based on the Finnish National Hospital Discharge Register and the Finnish twin cohorts [Kieseppa et al., 2005] (Fig. 1).

The study sample comprised of 157 twin pairs with altogether 298 individuals (including 61 pairs (120 individuals) with at least one member affected by SCZ or schizoaffective disorder, 31 pairs (51 individuals) with at least one member affected by BPD, and 65 pairs (127 individuals) of control twins with no psychiatric disorder) (Table I). Age and gender distributions are presented in Table II. A detailed neuropsychological test battery and clinical evaluation were performed as previously described (Table III, Supplementary Text 1) [Cannon et al., 2000]. In addition, high-resolution MR images were obtained from 48 pairs with SCZ and 47 control pairs (N=190) (Supplementary Text 1).

Genotyping

Three SNPs were selected based on the previous genetic studies (rs2494732, rs2498799, rs1130214) [Emamian et al., 2004] (Fig. 2). The genotyping was performed using Sequenom's MassARRAY platform using the homogenous massEXTEND protocol as specified by the manufacturer (Sequenom Inc., San Diego, CA). The automated allele calling from Sequneom was verified manually using the MassARRAY Typer software by two independent reviewers. For quality control, 18 duplicate samples and 10 water controls were included in each plate. Hardy–Weinberg equilibrium was calculated selecting at random one co-twin of each pair (i.e., blind with respect to affection status). All the SNPs were in Hardy–Weinberg equilibrium ($P > 0.05$) and the overall genotyping success rate was 94.7%. No discrepancies were observed among the duplicate genotype pairs.

Statistical Analyses

Basic descriptive statistics of the neuropsychological test data were computed by SPSS 11.5 software. As the subsequent statistical genetic analyses are sensitive to non-normal distribution [Allison et al., 1999], each trait, usually normally distributed in population [Spreen et al., 2006] was first transformed if necessary to regain normal or close-to normal distribution (Table III).

For genetic analyses of the quantitative neuropsychological and volumetric data of hippocampus and prefrontal cortex, linear regression was used and adjusted for age, gender, zygosity, psychosis and the co-twin being affected, in case the effect for a given covariate for the trait was found to be significant ($P < 0.05$). The analyses were performed using Stata

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8 software. The lack of statistical independence of twins was taken into account by computing robust estimates of variance for cluster-correlated data in Stata [Williams, 2000].

Bivariate correlation analysis and principal component analysis (PCA) were used to combine the four most interesting traits into on composite index to further assess a possible common underlying biological feature affected by *AKT1* using SPSS 11.5 Software. Only the four traits representing one functional domain were included to the PCA and the composite index was formed as a weighted sum of these traits.

Intra-pair resemblance was assessed by computing intra-pair differences and correlations of verbal memory composite index within monozygotic (MZ) and dizygotic (DZ) control twins and discordant SCZ pairs stratified by identity-by-state (IBS) status for the *AKT1* SNP. IBS status 0 and 1 were combined in analyses due to small numbers. The BPD pairs were excluded from these analyses due to the low number of pairs (31 pairs).

The genetic influence of *AKT1* on gray matter density at each cortical voxel was assessed using QTDT software [Abecasis et al., 2000a,b]. This analysis took advantage of the QTDT program's variance component framework, functionally equivalent to those conducted on the cognitive data using Stata 8, but computationally more feasible with the platform in the voxel-based analyses with an engineered software “bridge” between the probabilistic brain atlas algorithms and QTDT.

RESULTS

Association of rs1130214 With Verbal Learning and Memory

We first looked for association between *AKT1* variants and neuropsychological data in the complete sample of 298 twins (157 pairs) by linear regression analysis, adjusted for age, gender, zygosity and psychosis. In these analysis, we obtained some evidence for association between the A-allele of the 5′-end SNP (rs1130214) and lower performance on five measures related to verbal memory and derived from the CVLT: total recall, recall after 5 and 20 min delay (P=0.0028, 0.0005, and 0.0009, respectively), semantic clustering (P=0.005) and perseverative errors (P=0.024). Out of these, association of rs1130214 to recall after short delay remained significant ($P < 0.05$) even after correcting the P-values by the number of tests performed (26 traits, 3 SNPs; corrected P=0.0005 × 60=0.039). In addition, we detected nominal evidence for association for verbal working memory (P=0.039). No association was detected for the two additional SNPs (Table III).

Further Evidence From Analysis of a Composite Index of Verbal Memory

The four traits (total recall, recall after short and long time period and semantic clustering), showing the strongest evidence for association with rs1130214, were found to correlate strongly with each other (coefficients > 0.50) (data not shown). To condense the convergent information of these traits, most probably also biologically related, and to minimize the effect of sporadic variation between individual test scores, we combined these traits into one composite index of verbal memory (here after called “verbal memory”) using PCA. This composite index was formed as weighted sum with coefficients 0.278, 0.278, 0.272, and 0.260 for verbal learning, recall after short time period, recall after long time period, and semantic clustering, respectively. The composite index explained 84.3% of the total variance. As in case of the individual traits, analysis of the composite index in relation to the critical SNP (rs1130214) revealed a significant association (Coef.=−1.37, 95% CI=−2.13 to −0.60, P=0.0005).

To address the potential effect of the disease process and treatments on the association to verbal memory performance, we combined the control pairs and unaffected co-twins into a

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"healthy" subgroup and the affected co-twins into an "affected" subgroup. Interestingly, the strongest signal for the association was obtained from the healthy group (Coef.=−1.47, 95% CI=−2.47 to −0.48, *P*=0.004) while only a tendency, not reaching statistical significance, was detected in the affected co-twins, (Coef.=−1.02, 95% CI=−2.37 to 0.33, *P*=0.14).

Variation of Verbal Memory in Subgroups of the Sample and in Allelic Variety of rs1130214

We evaluated intra-pair relationships in control and discordant SCZ pairs, ascertained for their zygosity and rs1130214 genotype by calculating absolute intra-pair differences as well as intra-pair correlations. In addition to overall genetic similarity and presence of disease, we also detected a distinct contribution of *AKT1* affecting the degree of resemblance in verbal memory between co-twins. As expected, the control pairs were found to have higher overall intra-pair resemblance in verbal memory than SCZ pairs (*P* (absolute intra-pair difference)=0.0018). The intra-pair correlation was highest and the intra-pair differences were smallest among the MZ control pairs (Table IV). Interestingly, the DZ pairs sharing the same rs1130214 genotype had higher resemblance (r=0.57, SD=5.13; Mean intra-pair difference=3.41, SD=3.3) than the DZ pairs not sharing the rs1130214 genotype (r=0.19, SD=5.18; Mean intra-pair difference=5.56, SD= 3.53) (*P* (correlation)=0.056; *P* (intra-pair difference)=0.025), and only a slightly lower resemblance than the MZ pairs (r=0.62, SD=5.26; Mean intra-pair difference=3.59, SD=2.88), however not reaching statistical significance (*P* (correlation)=0.40; *P* (intra-pair difference)=0.80). In the SCZ group the DZ pairs sharing the rs1130214 genotype had point estimates of lower intra-pair difference and higher correlation indicating higher resemblance than the MZ pairs sharing the whole genomic sequence; however, this is probably due to sample variations given the low number of pairs (11 pairs) in the group (Table IV). In addition, we evaluated the difference between the healthy and affected co-twins. This differed from the absolute intra-pair difference since in some pairs the healthy co-twin performed worse than the affected co-twin. Interestingly the largest difference was observed in the discordant MZ pairs (Table IV).

A similar decline in the relationship between verbal memory and psychosis load was evident by comparing the mean values of verbal memory in various subgroups of the sample (Supplementary Fig. 1).

MRI Data

We had also access to the MRI data of the SCZ and control twins and used this information of gray matter density to assess the impact of *AKT1* to another quantitative parameter, relevant for psychosis Genetic association analysis using a variance components framework and adjusting for dependence of observations among twin pairs [Abecasis et al., 2000a,b] was used to assess association of gray matter density at each cortical voxel with the three SNPs. In parallel with the findings related to cognitive deficits, the A-allele of the 5′-end SNP (rs1130214) of *AKT1* was associated with reduced gray matter density in the medial and bilateral dorsal prefrontal cortex, as well as in inferior prefrontal cortex in the left hemisphere ($P < 0.05$) (Fig. 3). To correct for multiple comparisons, we computed 50,000 permutations of the statistical brain maps in which rs1130214 AKT1 allelic value was randomly assigned to subjects' MRI scan data. Thresholding the observed minor AKT1 allelic effect on reduced gray matter density at $P<0.0001$, the permutation analysis confirmed that the observed number of voxels showing association with reduced gray matter density was not observed by chance at an empirical $P<0.03$ in right prefrontal cortex overall and at an empirical $P<0.025$ in right dorsal prefrontal cortex. However, unlike for the neuropsychological measures, the association of rs1130214 with prefrontal gray matter reduction was stronger in the SCZ patients than in the control subjects, with the patients showing a much larger surface area in the frontal lobe region associating with the A-allele (Fig. 3).

DISCUSSION

We have studied the role of *AKT1*, a highly relevant candidate gene for SCZ and BPD, in relation to quantitative cognitive and neuroanatomical phenotypes in Finnish twin pairs. We obtained evidence for association between *AKT1* and four quantitative verbal memory-related functions as well as prefrontal cortical gray matter density. The association between *AKT1* short-term verbal memory reached statistical significance (original P=0.0005, corrected P=0.03) even after applying conservative measures to correct for multiple testing. Importantly, the same AKT1 allele was associated with worse cognitive functioning and with decreased gray matter density in the medial, dorsolateral, and inferior pre-frontal cortex, suggesting a consistent biological effect. This study design, profiting from information content of discordant twin pairs in analysis of intermediate phenotypes, is obviously not suitable for addressing plausible association with the disease diagnosis; it rather facilitates monitoring for the impact of the disease status on the phenotypes followed. Not surprisingly, we did not see evidence of association between *AKT1* alleles and psychosis or the DSM-IV based end state diagnoses of SCZ spectrum psychosis and affective psychosis (data not shown).

The strongest contribution of *AKT1* to verbal memory was observed among the control pairs and unaffected co-twins of SCZ and BPD pairs, but similar trend was also seen in affected co-twins, whereas the association to gray matter density was strongest in the patients, having the largest measurable changes. There are numerous potential explanations for this, including statistical power due to differences in sample sizes and in magnitude of the measurable changes for applied methods. Furthermore, these parameters (verbal memory and gray matter density), potentially affected by *AKT1*, could also be independent from each other and be altered at a different and individual pace during the disease process.

We obtained support for the association by taking advantage of the twin design of the study that allowed us to compare intra-pair resemblance between co-twins relative to their *AKT1* genotypes. Pairs sharing the rs1130214 genotype had a significantly lower intra-pair variation in verbal memory compared to co-twins not sharing the genotype. Interestingly, genotype-related quantitative differences can also be observed in the gray matter in the prefrontal cortex, the allele associating with lower performance in verbal memory also associating with the reduced gray matter density.

Neuropsychological functions are known to be impaired both in patients suffering from SCZ, and to a less extent, in their first degree relatives [Cannon et al., 1994]. Some of these deficits seem to be heritable and indicate increased disease risk, whereas others probably are more directly (even diagnostically) a result of the disease itself [Tuulio-Henriksson et al., 2002]. SCZ patients typically have a more generalized and severe impairment whereas BPD patients are found to differ from control population especially in verbal memory functions [van Gorp et al., 1999; Seidman et al., 2002]. Verbal memory functions are shown to have a fundamental genetic component explaining 56% of the total variation in a study of elderly twins [Swan et al., 1999]. The performance of the healthy siblings or co-twins of BPD patients is reported to be comparable to healthy individuals or show only mild impairment [Kieseppa et al., 2005; Antila et al., 2007] whereas the siblings of SCZ patients perform worse in CVLT than healthy controls [Egan et al., 2001]. Previous studies in twins discordant for SCZ [Cannon et al., 2000], as well as heritability estimates in a family based study [Tuulio-Henriksson et al., 2002], have indicated that verbal memory deficits are strongly influenced by the SCZ disease process. Furthermore, in line with these observations, in a recent study of five domains of cognition (processing speed, reasoning, verbal memory, working memory, and vigilance) and *AKT1* conducted in patients with SCZ no evidence for association was detected [Pinheiro et al., 2007]. The current study supports

these findings. Intra-pair comparisons of verbal memory revealed a decrease in intra-pair resemblance among discordant SCZ pairs compared to controls. However, the current study does not enable us to make a distinction between influences secondary to the illness expression and treatment, nor can we differentiate these from etiologically relevant non-genetic factors underlying verbal memory.

The finding that *AKT1* associates specifically with gray matter reduction in the prefrontal cortex is in line with prior work showing that gray matter density in this region correlates with the degree of genetic proximity to an individual affected with SCZ [Rapoport et al., 2005] or BPD [Shastry, 2005] and with evidence of a role of prefrontal cortex in verbal memory encoding and retrieval. In addition, SCZ patients are reported to have lower AKT1 protein levels and decreased phosphorylation of GSK-3β, a substrate of AKT1, in lymphocytes and prefrontal cortex [Emamian et al., 2004]. Furthermore, several three-SNP haplotypes with a common two-SNP core in *AKT1* were recently associated with SCZ and decreased AKT1 levels in lymphocyte-derived cell lines [Emamian et al., 2004]. Several genetic studies have since then replicated the association of *AKT1* in the etiology of SCZ [Ikeda et al., 2004; Schwab et al., 2005; Bajestan et al., 2006; Norton et al., 2007] but also negative reports exist [Ohtsuki et al., 2004; Ide et al., 2006; Liu et al., 2006; Turunen et al., 2007]. Our associations between verbal memory, prefrontal cortical gray matter, and *AKT1* in Finns are all with the same "risk" allele A of rs1130214 which is also part of the originally reported "risk" core haplotype [Emamian et al., 2004]. Previously the 3′ end of *AKT1* specifically the SNP rs2498799/rs1130233 was associated with a factor phenotype containing measures of IQ, executive function and processing speed, and with inefficient prefrontal activation and gray matter density [Tan et al., 2008b]. It is highly intriguing that two independent studies show evidence for a role of AKT1 in cognitive functions and brain morphology. However, we were not able to directly replicate the association with the same SNP as Tan et al. This divergence between the studies is unlikely explained by genetic proximity of the two SNPs. In our sample the LD between rs1130214, we found to be associated, and rs2498799 is low ($D' = 0.157$ and $r^2 = 0.016$) which is in agreement with the Caucasian haplotype structure of the International HapMap Project (http://www.hapmap.org). In contrast the differences might rather be suggestive of multiple different causative variants in the gene affecting to a relatively broad range of nervous functions.

More direct functional data also support the role of AKT1 in complex brain functions, disturbed in psychosis. Phosphorylation levels of AKT1 are increased in hippocampus of rats, trained in a maze for several days compared to untrained controls [Mizuno et al., 2003]. Akt1 deficient mice exhibit changes in the expression of genes related to synaptic function, neuronal development, myelination, and actin polymerization in prefrontal cortex. Furthermore, there were morphological changes in the dendritic architecture and complexity of V-pyramidal neurons [Lai et al., 2006]. Further, alterations in blood glucose concentration have been reported to influence higher brain functions such as memory, while oscillations in glucose concentration affect the phosphorylation states of AKT1 and GSK3β in mouse brain, suggesting that this pathway might play an important role in memory deficits occurring in some patients with diabetes [Clodfelder-Miller et al., 2005]. Similarly, insulin dependent AKT1-GSK3β signaling cascade has been found to be significantly reduced in dorsolateral prefrontal cortex of patients suffering from SCZ [Zhao et al., 2006]. Lithium, commonly used to treat mood disorders, has been related to decreased performance in verbal memory tasks, although the effect appears to be somewhat modest and a few negative findings have also been reported [Macqueen and Young, 2003; Pachet and Wisniewski, 2003]. AKT1-GSK3β signaling cascade is a target of lithium [Coyle and Duman, 2003; Beaulieu et al., 2008] raising the possibility that the proposed lithium related defects in verbal memory are mediated through the same pathway that we now report to be

genetically associated with verbal memory. In addition, dopamine, a neurotransmitter tightly linked to multiple neurological and psychiatric disorders, is suggested to exert at least some of its effect on behavior via AKT1/GSK3β pathway. Mice knocked out for gene encoding for a dopamine transporter (DAT) have reduced levels of active phosphorylated AKT1 (pAKT1) while the amount of pAKT1 increases in depletion of dopamine. Importantly, the dopamine associated hyperactivity in the $DAT^{-/-}$ mice is antagonized by lithium treatment [Beaulieu et al., 2004]. AKT1 has been implicated in a variety of functions in the central nervous system such as neuronal migration [Beffert et al., 2002], growth factor dependent survival of neurons, as well as axon growth and branching [Markus et al., 2002], while alterations in these functions may be the cause of changes in cognitive functions.

In conclusion, using a unique study design of same sex twin pairs, we obtained evidence that *AKT1* is connected to several quantitative measures of verbal learning and memory as well as to gray matter density in the prefrontal cortex. The association was fortified by a decrease in variance in verbal memory among pairs sharing the same critical *AKT1* allele. Our study further highlights the effectiveness of the use of measurable quantitative cognitive and neuroanatomical traits as endophenotypes for SCZ and BPD. Recognition of the genetic variation affecting cognitive functioning and brain structure in the population level may provide tools to identify pathways related to disease pathogenesis.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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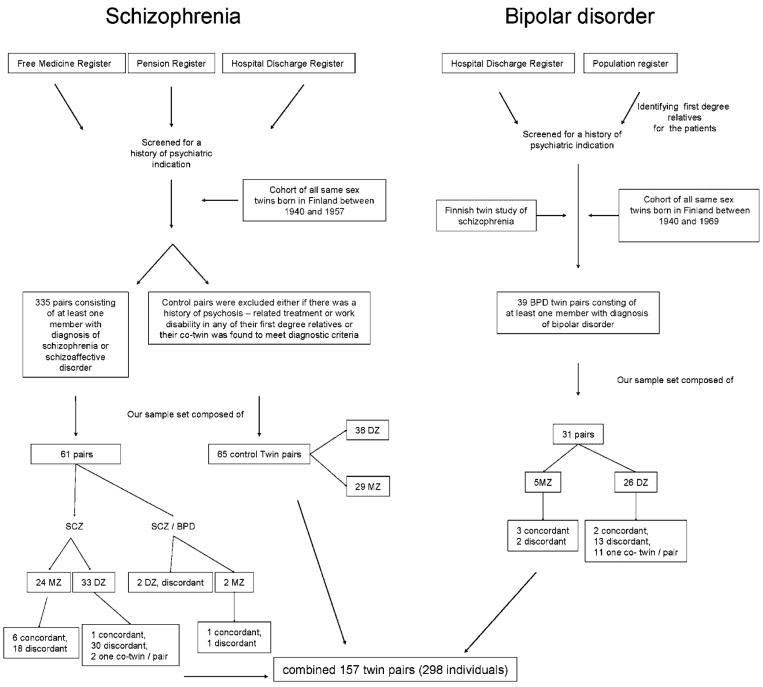

FIG. 1.
The collection of the twin sample.

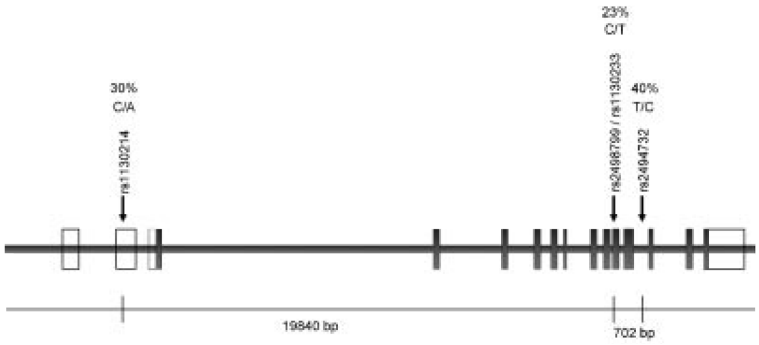

FIG. 2.
Genomic structure of AKT1 and the positions of the genotyped SNPs. Minor allele frequencies and alleles are presented above each SNP.

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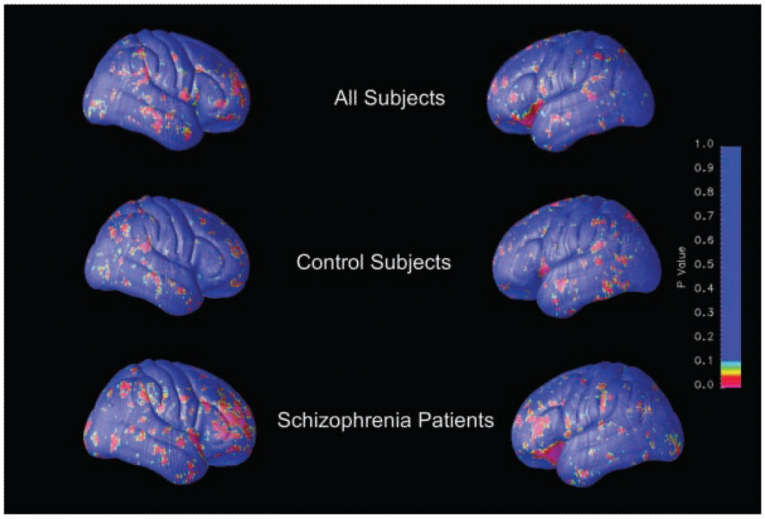

FIG. 3.

Statistical anatomic maps of areas of the cortical surface showing significant reductions in gray matter density associated with rs1130214 in AKT1. Maps are thresholded to display regions showing association with gray matter density reductions at $P<0.05$. Significance scale is presented on the right side of the picture. The images on the left represent the right hemisphere and the images on the right represent the left hemisphere.

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TABLE I

Study Sample as Number of Twin Pairs and Individuals

Group	Individuals	Pairs	DZ pairs with both co-twins available	DZ pairs with only one co-twin available[a]	MZ pairs with both co-twins available	Concordant DZ pairs	Concordant MZ pairs	Discordant DZ pairs	Discordant MZ pairs
Control	127	65	33	3	29	33	29	0	0
Schizophrenia	112	57	31	2	24	1	6	30	18
Schizoaffective	8	4	2	0	2	0	1	2	1
Bipolar	51	31	15	11	5	2	3	13	2
Combined	298	157	81	16	60	36	39	45	21

MZ, monozygotic; DZ, dizygotic.

[a]The twins lacking a co-twin were not included in pairwise analysis taking advantage of the twin design of the study. Out of the 16 single individuals 2 were diagnosed with schizophrenia and 8 with bipolar disorder.

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TABLE II

Age and Gender Distributions of Twins

				Age	
Group	**Individuals**	**Males**	**Females**	**Mean**	**SD**
All	298	146 (49%)	152 (51%)	47.5	5.6
Control	127	65 (51%)	62 (49%)	48.5	4.0
SCZ[a]	120	59 (49%)	61 (51%)	47.7	5.5
BPD	51	22 (43%)	29 (57%)	44.7	7.9

SCZ, schizophrenia; BPD, bipolar disease; SD, Standard deviation.

[a]Including both schizophrenia and schizoaffective twins.

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TABLE III

Neuropsychological Test Battery Administered for the Study Subjects Clustered Along Cognitive Domains and the Two Brain Volume Measures

Trait	Test	Transformation[a]	Kurtosis[b]	Skewness[b]	*P*-value*	*P*-value*	*P*-value*
Attention and working memory					rs2494732	rs2498799	rsll30214
Verbal attention (Digit span forward)	WMS-R	None	−0.22	−0.14	0.24	0.10	0.60
Verbal working memory (Digit span backward)	WMS-R	None	0.13	−0.04	0.29	0.65	**0.04**
Visual attention (Visual span forward)	WMS-R	None	−0.41	0.11	0.71	0.49	0.57
Visual working memory (Visual span backward)	WMS-R	None	0.03	−0.04	0.86	0.52	0.61
Attention and mental tracking (Trails-A)	TMT-A	Inversion	0.98	0.91	0.97	0.60	0.26
Verbal learning and memory							
Learning (recalling words of a word list during five trials)	CVLT	None	−0.31	−0.42	0.45	0.75	**0.003**
Recalling words after short delay	CVLT	None	−0.60	−0.32	0.10	0.16	**<0.001**
Recalling word after long delay	CVLT	None	−0.25	−0.54	0.21	0.26	**<0.001**
Clustering words along semantic categories, a learning strategy	CVLT	Square root	0.29	0.10	0.39	0.83	**0.006**
Making perseverative recall errors (repeating words)	CVLT	$\log_{10}$	−0.76	−0.31	0.49	0.27	**0.02**
Making intrusive recall errors (words not on the list)	CVLT	$\log_{10}$	0.23	1.13	0.91	0.76	0.75
Recognition memory index	CVLT	Cubed	1.27	−1.03	0.37	0.94	0.12
Immediate recall of story	WMS-R	None	−0.24	−0.26	0.19	0.96	0.37
Delayed recall of story	WMS-R	None	−0.57	−0.10	0.32	0.66	0.48
Visual memory							
Immediate recall (visual reproduction)	WMS-R	Squared	0.08	−0.84	0.54	0.33	0.97
Delayed recall (visual reproduction)	WMS-R	None	−0.15	−0.78	0.16	0.09	0.21
Executive functions							
Stroop interference score, distractibility	Stroop task	$\log_{10}$	1.12	0.65	0.22	0.17	0.15
Strategy shifting (Trails B)	TMT_B	$\log_{10}$	−0.19	0.29	0.28	0.58	0.21
Number of perseverative errors during WCST	WCST	Inversion	0.26	1.03	0.31	0.54	0.25
WCST, number of categories	WCST	None	−0.63	−0.88	0.32	0.55	0.20
Ability functions							
Concept formation and abstraction (Similarities)	WAIS-R	Square root	7.60	−0.45	0.51	0.18	0.22
Premorbid and basic intelligence (Vocabulary)	WAIS-R	Square root	4.54	0.10	0.77	0.13	0.22
Mental tracking, processing speed (Digit symbol)	WAIS-R	$\log_{10}$	2.28	−0.98	0.16	0.19	0.19

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Trait	Test	Transformation[a]	Kurtosis[b]	Skewness[b]	*P*-value[*]	*P*-value[*]	*P*-value[*]
Visuospatial reasoning (Block design)	WAIS-R	Square root	2.29	0.15	0.53	0.14	0.23
Brain volumetric data							
Total frontal cortex volume		In	−0.10	−0.17	0.64	0.17	0.70
Total hippocampal volume		$\log_{10}$	0.03	−0.29	0.10	0.10	0.88

[a] Usually normally distributed in the population (Spreen et al., 2006] Some of the traits were transformed to normalize distribution.

[b] Kurtosis and skewness are shown after transformation, if applicable.

[*] *P*-values obtained from linear regression in the complete sample. *P*-value <0.05 are shown in bold.

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TABLE IV

Intra-Pair Comparisons of the Composite Index of Verbal Memory in Subgroups of Twins

Health status	Zygosity	N (pairs)[a]	rsll30214 genotype	Mean verbal memory performance	Absolute intra-pair difference (SD)	Difference between healthy and affected co-twin (SD)	Intra-class correlation (95% CI)
Control	MZ	28	Same	22.03	2.61 (1.87)	na	0.61 (0.41–0.75)
	DZ	16	Same	21.53	2.92 (2.42)	na	0.49 (0.17–0.72)
	DZ	16	Different	19.72	4.56 (2.78)	na	0.45 (0.12–0.69)

Health status	Zygosity	N (pairs) [a]	rsll30214 genotype	Mean verbal memory performance		Absolute intra-pair difference (SD)	Difference between healthy and affected co-twin (SD)	Intra-class correlation (95% CI)
				Healthy	Affected			
SCZ	MZ	18	Same	16.56	14.53	5.12 (3.53)	2.03 (5.99)	0.21 (−0.13–0.51)
	DZ	11	Same	18.27	14.98	4.12 (4.26)	3.29 (5.00)	0.42 (0–0.72)
	DZ	10	Different	20.05	13.60	7.17 (4.12)	6.46 (5.28)	−0.48 (−0.76–(−0.05))

na, not applicable; SCZ, schizophrenia; MZ, monozygotic; DZ, dizygotic; SD, standard deviation.

[a]The number of pairs may differ from that presented in Table 1 due to missing data.