Meta-analysis of 32 genome-wide linkage studies of schizophrenia

Ng, M. Y. M., Levinson, D. F., Faraone, S. V., Suarez, B. K., DeLisi, L. E., Arinami, T., ... Lewis, C. M. (2009). Meta-analysis of 32 genome-wide linkage studies of schizophrenia. Molecular Psychiatry, 14(8), 774-785. DOI: 10.1038/mp.2008.135

Published in:
Molecular Psychiatry

Document Version:
Peer reviewed version

Queen's University Belfast - Research Portal:
Link to publication record in Queen's University Belfast Research Portal

Published in final edited form as:
Mol Psychiatry. 2009 August ; 14(8): 774–785. doi:10.1038/mp.2008.135.

Meta-analysis of 32 genome-wide linkage studies of schizophrenia

MYM Ng[1], **DF Levinson**[2], **SV Faraone**[3], **BK Suarez**[4], **LE DeLisi**[5,6], **T Arinami**[7], **B Riley**[8], **T Paunio**[9,10], **AE Pulver**[11], **Irmansyah**[12], **PA Holmans**[13], **M Escamilla**[14], **DB Wildenauer**[15], **NM Williams**[13], **C Laurent**[16], **BJ Mowry**[17], **LM Brzustowicz**[18], **M Maziade**[19], **P Sklar**[20], **DL Garver**[21], **GR Abecasis**[22], **B Lerer**[23], **MD Fallin**[24], **HMD Gurling**[25], **PV Gejman**[26], **E Lindholm**[27], **HW Moises**[28], **W Byerley**[29], **EM Wijsman**[30], **P Forabosco**[1], **MT Tsuang**[31,32], **H-G Hwu**[33], **Y Okazaki**[34], **KS Kendler**[8], **B Wormley**[8], **A Fanous**[35,36], **D Walsh**[37], **FA O'Neill**[38], **L Peltonen**[39,40,41,42], **G Nestadt**[11], **VK Lasseter**[11], **KY Liang**[43], **GM Papadimitriou**[44], **DG Dikeos**[44], **SG Schwab**[45,46,47], **MJ Owen**[13], **MC O'Donovan**[13], **N Norton**[13], **E Hare**[14], **H Raventos**[48], **H Nicolini**[49], **M Albus**[50], **W Maier**[51], **VL Nimgaonkar**[52], **L Terenius**[53], **J Mallet**[54], **M Jay**[55], **S Godard**[56], **D Nertney**[57], **M Alexander**[2], **RR Crowe**[58], **JM Silverman**[59], **AS Bassett**[60], **M-A Roy**[19], **C Mérette**[19], **CN Pato**[61], **MT Pato**[61], **J Louw Roos**[62], **Y Kohn**[23], **D Amann-Zalcenstein**[63], **G Kalsi**[25], **A McQuillin**[25], **D Curtis**[64], **J Brynjolfson**[65], **T Sigmundsson**[65], **H Petursson**[65], **AR Sanders**[26], **J Duan**[26], **E Jazin**[27], **M Myles-Worsley**[66], **M Karayiorgou**[67], and **CM Lewis**[1,68]

[1]King's College London, Department of Medical and Molecular Genetics, London, UK [2]Department of Psychiatry, Stanford University, Stanford, CA, USA [3]Departments of Psychiatry and of Neuroscience and Physiology, SUNY Upstate Medical University, Syracuse, NY, USA [4]Washington University in St Louis, St Louis, MO, USA [5]Department of Psychiatry, The New York University Langone Medical Center, New York, NY, USA [6]Nathan S Kline Institute for Psychiatric Research, Orangeburg, NY, USA [7]Department of Medical Genetics, University of Tsukuba, Tsukuba, Japan [8]Department of Psychiatry, Virginia Institute for Psychiatric and Behavioral Genetics, Virginia Commonwealth University, Richmond, VA, USA [9]National Public Health Institute, Helsinki, Finland [10]Department of Psychiatry, Helsinki University Central Hospital, Helsinki, Finland [11]Department of Psychiatry & Behavioral Sciences, Johns Hopkins University School of Medicine, Baltimore, MD, USA [12]Department of Psychiatry, University of Indonesia, Jakarta, Indonesia [13]Department of Psychological Medicine, School of Medicine, Cardiff University, Cardiff, UK [14]University of Texas Health Science Center at San Antonio, San Antonio, TX, USA [15]Center for Clinical Research in Neuropsychiatry, School of Psychiatry and Clinical Neurosciences, University of Western Australia, Perth, WA, Australia [16]Department of Child Psychiatry, Université Pierre et Marie Curie and Hôpital de la Pitiè-Salpêtrière, Paris, France [17]Queensland Centre for Mental Health Research and University of Queensland, Brisbane, QLD, Australia [18]Department of Genetics, Rutgers University, Piscataway, NJ, USA [19]Department of Psychiatry, Laval University & Centre de recherche Université Laval Robert-Giffard, Québec, QC, Canada [20]Center for Human Genetic Research, Massachusetts General Hospital and Harvard Medical School, Boston, MA, USA [21]VA Medical Center, Asheville, NC, USA [22]Department of Biostatistics, University of Michigan School of Public Health, Ann Arbor, MI, USA [23]Department of Psychiatry, Hadassah-Hebrew University Medical Center, Jerusalem, Israel [24]Department of Epidemiology, Johns Hopkins Bloomberg School of Public Health, Baltimore, MD, USA [25]Department of Mental Health Sciences, University College

Correspondence: Dr CM Lewis, Department of Medical and Molecular Genetics, King's College London, 8th Floor Tower Wing, Guy's Hospital, Great Maze Pond, London SE1 9RT, UK. E-mail: E-mail: Cathryn.lewis@kcl.ac.uk.

UKPMC Funders Group Author Manuscript

London, London, UK [26]Center for Psychiatric Genetics, NorthShore University HealthSystem Research Institute and Northwestern University, Evanston, IL, USA [27]Department of Development & Genetics, Uppsala University, Uppsala, Sweden [28]University of Kiel, Kiel, Germany [29]University of California, San Francisco, CA, USA [30]Departments of Medicine and Biostatistics, University of Washington, Seattle, WA, USA [31]Center for Behavioral Genomics and Department of Psychiatry, University of California, San Diego, CA, USA [32]Harvard Institute of Psychiatric Epidemiology & Genetics, Boston, MA, USA [33]National Taiwan University Hospital and National Taiwan University College of Medicine, Taipei, Taiwan [34]Tokyo Metropolitan Matsuzawa Hospital, Tokyo, Japan [35]Washington VA Medical Center, Washington, DC, USA [36]Department of Psychiatry, Georgetown University Medical Center, Virginia Commonwealth University, Richmond, VA, USA [37]The Health Research Board, Dublin, Ireland [38]Department of Psychiatry, Queens University, Belfast, Northern Ireland [39]Department of Molecular Medicine, National Public Health Institute, Helsinki, Finland [40]Department of Medical Genetics, University of Helsinki, Helsinki, Finland [41]The Broad Institute, MIT, Boston, MA, USA [42]Wellcome Trust Sanger Institute, Wellcome Trust Genome Campus, Cambridge, UK [43]Department of Biostatistics, Johns Hopkins University Bloomberg School of Public Health, Baltimore, MD, USA [44]1st Department of Psychiatry, University of Athens Medical School, and University Mental Health Research Institute, Athens, Greece [45]Western Australian Institute for Medical Research, University of Western Australia, Perth, WA, Australia [46]School of Psychiatry and Clinical Neurosciences, University of Western Australia, Perth, WA, Australia [47]School of Medicine and Pharmacology, University of Western Australia, Perth, WA, Australia [48]School of Biology and CIBCM, University of Costa Rica, San Jose, Costa Rica [49]Carracci Medical Group and Universidad Autonoma de la Ciudad de Mexico, Mexico City, Mexico [50]State Mental Hospital, Haar, Germany [51]Department of Psychiatry, University of Bonn, Bonn, Germany [52]Departments of Psychiatry and Human Genetics, University of Pittsburgh, Pittsburgh, PA, USA [53]Department of Clinical Neuroscience, Karolinska Hospital, Stockholm, Sweden [54]Laboratoire de Génétique Moléculaire de la Neurotransmission et des Processus Neurodégénératifs, Centre National de la Recherche Scientifique, Hôpital de la Pitié Salpêtrière, Paris, France [55]Department of Child Psychiatry, Université Pierre et Marie Curie and Hôpital de la Pitiè-Salpêtrière, Paris, France [56]INSERM, Institut de Myologie, Hôpital de la Pitiè-Salpêtrière, Paris, France [57]Queensland Centre for Mental Health Research and University of Queensland, Brisbane, QLD, Australia [58]Department of Psychiatry, The University of Iowa Carver College of Medicine, Iowa City, IA, USA [59]Department of Psychiatry, Mount Sinai School of Medicine, New York, NY, USA [60]Clinical Genetics Research Program, Centre for Addiction and Mental Health and Department of Psychiatry, University of Toronto, Toronto, ON, Canada [61]Center for Genomic Psychiatry, University of Southern California, Los Angeles, CA, USA [62]Department of Psychiatry, University of Pretoria, Weskoppies Hospital, Pretoria, Republic of South Africa [63]Department of Molecular Genetics, Weizmann Institute of Science, Rehovot, Israel [64]Department of Psychological Medicine, St Bartholomew's and Royal London School of Medicine and Dentistry, London, UK [65]Department of Psychiatry, General Hospital, Reykjavik, Iceland [66]Department of Psychiatry, SUNY Upstate Medical University, Syracuse, NY, USA [67]Departments of Psychiatry and Genetics & Development, Columbia University Medical Center, New York, NY, USA [68]King's College London, MRC SGDP Centre, Institute of Psychiatry, London, UK

Abstract

A genome scan meta-analysis (GSMA) was carried out on 32 independent genome-wide linkage scan analyses that included 3255 pedigrees with 7413 genotyped cases affected with schizophrenia (SCZ) or related disorders. The primary GSMA divided the autosomes into 120 bins, rank-ordered the bins within each study according to the most positive linkage result in each bin, summed these ranks (weighted for study size) for each bin across studies and determined the empirical probability of a given summed rank (P_{SR}) by simulation. Suggestive evidence for linkage was observed in two single bins, on chromosomes 5q (142-168 Mb) and 2q (103-134 Mb). Genome-wide evidence for

linkage was detected on chromosome 2q (119-152 Mb) when bin boundaries were shifted to the middle of the previous bins. The primary analysis met empirical criteria for 'aggregate' genome-wide significance, indicating that some or all of 10 bins are likely to contain loci linked to SCZ, including regions of chromosomes 1, 2q, 3q, 4q, 5q, 8p and 10q. In a secondary analysis of 22 studies of European-ancestry samples, suggestive evidence for linkage was observed on chromosome 8p (16-33 Mb). Although the newer genome-wide association methodology has greater power to detect weak associations to single common DNA sequence variants, linkage analysis can detect diverse genetic effects that segregate in families, including multiple rare variants within one locus or several weakly associated loci in the same region. Therefore, the regions supported by this meta-analysis deserve close attention in future studies.

Keywords

genome; human; humans; schizophrenia/genetics; genetic predisposition to disease; linkage (genetics); meta-analysis

Introduction

We report here on a new genome scan meta-analysis (GSMA)[1-3] of genome-wide linkage scans (GWLS) of schizophrenia (SCZ). We previously published a GSMA of 20 scans that included 1208 pedigrees with 2945 genotyped affected individuals.[4] Here we analyze 32 scans that included 3255 pedigrees with 7413 affected individuals.

Genome-wide association (GWA) study methods have proven more successful than GWLS for common diseases: they detect smaller effects of common single nucleotide polymorphisms (SNPs)[5] as well as some copy number variants (CNVs),[6] and have identified many common disease associations.[7] But an adequately powered GWLS can detect a signal from many kinds of susceptibility variants (common SNPs, multiple rare SNPs, tandem repeats and heritable structural variations) in one or more[8] loci in a region. Meta-analysis can achieve much larger sample sizes than single studies. Consensus linkage regions may deserve further study using methods such as high-throughput resequencing.

Family and twin data[9] suggest that most of the genetic risk to SCZ is conferred by multiple interacting loci, each causing a small increase in risk. As SCZ has similar symptoms and roughly similar prevalence throughout the world,[10] a reasonable hypothesis is that at least some loci have effects in many populations. SCZ linkage findings do not replicate consistently, and although there may be some true positives in small samples from unique populations, in general, power is probably limited by inadequate sample size and differences in ascertainment, marker sets, ancestry and statistical methods. There must also be some measurement error due to limitations in diagnostic methods. Current diagnostic criteria produce the largest known estimates of heritability (twins) and of increased risk to first-degree relatives,[11,12] and there is good diagnostic reliability within and across research groups.[13] But diagnostic judgments are based on data from interview and medical records that vary in completeness and quality. Fortunately diagnostic differences are usually among psychotic disorders that coaggregate in families with similar risks.[14] It is not known whether a further subdivision of cases would increase power.

We present a GSMA of all 32 scan analyses, and a secondary analysis of 22 European-ancestry analyses. The new samples (some of them large) are from European, non-European and isolate populations, but are too diverse to consider a subset other than Europeans. We comment briefly below on an analysis of heterogeneity between Asian and European samples. Statistical significance has been evaluated with permutation tests and simulations. In the primary analysis,

2 bins met empirical genome-wide criteria for suggestive linkage; and a set of 10 bins (in eight chromosomal regions) met an empirical aggregate criterion for genome-wide significant linkage, indicating that loci linked to SCZ are present in some or all of these bins. Five of these bins were consistent with our previous report.[4] When bin boundaries were shifted to the middle of the previous bins, one genome-wide significant result was observed on chromosome 2q, consistent with our previous report.[4]

Materials and methods

Study selection

We identified GWLS of SCZ (in addition to those studied previously[4]) through PubMed literature searches, conference abstracts and personal contacts with investigators, selecting the largest analysis for each study. Study characteristics are summarized in Table 1. Investigators of each of these studies agreed to contribute data to this analysis. All scans ascertained probands with SCZ by one of three closely related sets of diagnostic criteria. Most scans included relatives with SCZ and those with one or all subtypes of schizoaffective disorder (SA). A few scans included only SCZ cases, or included broader diagnoses in alternative models. Scans were included if informative families ascertained through a SCZ case had at least 30 genotyped SCZ and SA cases. We included only primary genome-wide analyses with few gaps in marker coverage, and did not consider subsequent 'fine mapping' with additional markers or pedigrees in selected regions. We excluded a small Utah study[48] whose RFLP markers were difficult to place on the current map; a study of a large Costa Rican pedigree[49] with too few affected cases; a small study from an Italian isolate population[50] that combined SCZ and bipolar cases; and a study of isolated villages in Daghestan[51] that used a very broad phenotype definition. We lacked detailed results for the Icelandic study of Stefansson *et al*.[52] We included two earlier studies of Icelandic families for which we had complete results,[15,20] and discuss below a secondary analysis substituting results from Stefansson *et al*. inferred from published graphs.

A companion paper[38] reports on a combined analysis of studies 24-31 (Table 1). Older data for all or parts of six of those data sets were included as separate scans in our previous GSMA, so all eight are treated separately here using the newer data.[38] To eliminate sample overlap, investigators provided us with data for NUH (study 28) that excluded families that were also in the US/International study (study 9); and data for sample 15 (US/Sweden) without families in sample 26 (Cardiff). The investigators determined from genotypes that there were no overlapping families in samples 10 and 22.

There were 14 studies of European-ancestry pedigrees (including partial isolates such as Finland and Ashkenazim); 8 of both European and non-European families; and 10 non-European samples, including 3 Asian (Japanese, Han Chinese and Indonesian), 2 Latino, 1 Arab, 2 Pacific Island isolates (Palau and Kosrae) and 2 entirely (study 20) or predominantly (study 6) African-American samples. Study 6 (Texas) included a few European-ancestry families for which we lacked separate data. The ALL analysis included all 32 samples; the EUR (European-ancestry) analysis included 22 samples or subsamples (Table 1). Data for chromosome X markers were available for 23 studies, so this chromosome was considered in a secondary analysis.

Linkage scores were obtained from investigators or internet postings. Data for each study were rank ordered based on the designated 'primary' analysis, and if this included more than one test or model, we took the most positive linkage score across tests for each chromosomal bin. From our previous GSMA,[4] data were carried over unchanged for studies 1-10 in Table 1; the Utah study[48] was omitted as discussed above; and nine studies were updated with new genotyping results, some with larger samples. Studies 13 and 24-31 used SNP markers from

the Illumina version 4 panel; all other studies used microsatellite markers at approximately 8-10 cM density.

Marker mapping

Marker locations were determined from the Rutgers combined linkage map (Build 36),[53] or interpolated based on physical position, other maps or flanking markers. Chromosomes were divided into bins of approximately equal genetic widths—120 30-cM bins (our primary analysis), or (for secondary analyses) 179 20-cM bins and 88 40-cM bins. Six additional 30-cM bins were defined on the X chromosome (female map length). If a study had no marker in a bin (for example, the narrower 20 cM bins), we used the mean linkage score for the two flanking markers, or the single closest marker for an empty telomeric bin. Bin boundaries in cM and Mb are listed in online Supplementary Table 1.

Statistical analysis

GSMA is a nonparametric method to combine data generated with different maps and statistical tests.[1-3] A GSMA divides the genome into approximately equal-width bins (N_{bins}), labeled *ch.K*, where *ch* = chromosome and *K* = bin number (that is, 1.4 is the fourth bin of chromosome 1). For each study, bins are ranked by their highest LOD, NPL or Z score or minimum *P*-value. Ranks for each sample was weighted by the square root of the number of genotyped affected cases, and then, for comparison, analyzed without weights (results of unweighted analyses are provided in the online Supplementary Materials). Note that '120' was the best rank, and higher summed ranks indicate stronger evidence for linkage (whereas in our previous report,[4] '1' was the best rank in a study and we reported averaged rather than summed ranks).

We used GSMA software[2] to evaluate nominal (binwise) significance by randomly permuting the ranks for bins within each study and then resumming the ranks across studies to determine the empirical summed rank probability (P_{SR})—the nominal probability that a bin would achieve or exceed the observed summed rank under the null hypothesis of no linkage in the bin. We also determined the empirical ordered rank *P*-value (P_{OR}—the probability that the *k*th highest summed rank achieves or exceeds an observed summed rank under the null). Thresholds for genome-wide significant and suggestive evidence for linkage[54] were determined empirically (see below). For the primary analysis, these thresholds were $P_{SR} < 0.00037$ and $P_{SR} < 0.0077$ respectively.

Strong linkage signals can cover a wide genetic region in complex disorders, and often produce high summed ranks in adjacent bins,[1] but GSMA can miss a weak linkage signal near the boundary of two bins. Therefore, regions showing suggestive evidence for linkage were reanalyzed using 20 and 40 cM bin widths, and using 30 cM bins starting at the midpoint of the bins used in the primary analysis (Supplementary Figure 1).

We tested for heterogeneity in study ranks between three outbred Asian studies (Taiwan, Japan, Indonesia) and the 22 EUR studies, using the nonparametric Wilcoxon rank sum test. We did not test for heterogeneity arising from an arbitrary set of studies;[55] these tests detect bins where study ranks have a wider spread than expected by chance, but they have low power and do not identify the study subgroups contributing to the heterogeneity.[56] We also carried out sensitivity analyses, dropping one study at a time and analyzing the remaining studies with GSMA (weighted and unweighted analyses, 30 cM bins).

Simulation study of power and significance thresholds

We performed a simulation study to determine: (1) power; (2) permutation-based estimates of type I error and (3) aggregate criteria for genome-wide significance. Details are provided in Supplementary Online Materials and in a previous report.[1] Briefly, genome-wide data were

simulated for affected sibling pairs under the assumption of no linkage or assuming a range of genetic models (1-10 linked bins with locus-specific sibling relative risks (λ_S) of 1.15 or 1.3, at the edge or near the middle of chromosomes), and 1000 replicates of the 32-sample data set (or of alternative subsets of the data) were created by replacing each sample with a number of ASPs with roughly comparable linkage information.

For estimates of type 1 error, we determined empirical bin-wise thresholds for genome-wide suggestive linkage (the nominal *P*-value observed on average once per GSMA replicate) or genome-wide significant linkage (the nominal *P*-value observed on average in 5% of GSMA replicates).

We also determined criteria for aggregate genome-wide significant linkage, that is, that the total number of bins achieving a specified nominal *P*-value threshold was greater than that observed in 5% of genome-wide replicates with no linkage present. This criterion was determined for ALL and EUR studies by establishing a *P*-value threshold, P_0, for which exactly 5% of unlinked replicates had *N* or more bins with $P_{SR} < P_0$. P_0 was initially set at 0.05, and the number of unlinked replicates with $\geq N$ bins achieving $P_{SR} < P_0$ was tabulated (for $N = 1, 2, 3,...$). P_0 was then reduced incrementally until exactly 5% of simulations had at least *N* bins with $P_{SR} < P_0$, for some value *N*. For example, for the primary weighted analysis of 32 studies, the 5% genome-wide threshold by this procedure was 10 bins with $P < 0.046$; for EUR studies, it was 10 bins with $P < 0.048$.

For power calculations, we determined how often, on average, the linked bins exceeded the P_{SR} threshold for ALL and for EUR studies, and, to investigate heterogeneity, for ALL studies assuming that linkage arose only in EUR studies.

Results

Evidence for linkage (ALL and EUR studies)

Figure 1 illustrates *P*-values for all bins in the ALL and EUR analyses, and the figure legend states the empirical *P*-value thresholds for each study. For ALL studies (3255 pedigrees, 7413 genotyped cases), Table 2 lists the 10 bins (in eight nonadjacent chromosomal regions) that achieved $P_{SR} < 0.05$ in the primary analysis (weighted, 30 cM bins). These 10 bins met the empirical aggregate criterion for genome-wide significant linkage (10 bins with nominal $P_{SR} < 0.046$). Suggestive evidence for linkage was observed for single bins on chromosome 5q (bin 5.6, $P_{SR} = 0.0046$) and chromosome 2q (bin 2.5, $P_{SR} = 0.0075$). Figure 1 illustrates the results for all bins (ALL and EUR analyses), and Figure 2 illustrates the relative ranks of bins in each study. (Results for all analyses are available in Online Supplementary Tables; ranks for each study are available online at www.kcl.ac.uk/mmg/ngdata.html.)

Table 3 summarizes results for 22 EUR samples or subsamples (1813 pedigrees, 4094 genotyped cases). Suggestive evidence for linkage was observed on chromosome 8p ($P_{SR} = 0.00057$, close to the genome-wide threshold for 22 EUR studies of 0.00044), with a further five bins in four different regions achieving nominal significance.

On chromosome X, there were no nominally significant bins for ALL studies; there was one such bin (X.5, 130-162 cM, 119-141 Mb, $P_{SR} = 0.0247$) for EUR studies.

When estimated ranks (interpolated from published graphs) for the Icelandic study of Stefansson *et al*.[52] were substituted for the results of Gurling *et al*.[20] and Moises *et al*.,[15] results were similar to those shown in Table 1 except that bin 3.4 failed to achieve nominal significance. For EUR studies, results again were similar except that bins 8.2 and 2.8 both achieved suggestive significance.

Effect of bin width

Online Supplementary Figures 2-4 illustrate results of analyses using four alternative bin widths (20, 30 and 40 cM, and 30 cM bins shifted by 50% for chromosomes with suggestive evidence for linkage (chromosomes 2 and 5 for ALL, chromosome 8 for EUR). On chromosome 2, genome-wide significance was obtained for the shifted 30 cM bin (P_{SR} = 0.00035, 132.2-161.6 cM, 118.7-152 Mb), and suggestive linkage evidence was seen in the adjacent bin (102.8-132.2 cM, 84.9-118.7 Mb). For the region of chromosome 5q that produced suggestive evidence for linkage in the primary analysis, the signal was similar regardless of bin width. On chromosome 8, using 20 cM bins extended the signal by approximately 5 cM in both directions; if linkage is present, this pattern could indicate a broader signal (multiple loci) or the effects of a single strong signal on adjacent bins.

Heterogeneity and sensitivity analyses

In tests of differences between 22 EUR vs 3 Asian studies, none of the 120 bins exceeded nominal $P < 0.01$, and only 4 bins achieved nominal $P < 0.05$, thus, no significant difference was observed. The most significant evidence for heterogeneity between the Asian and European studies was in bin 1.5, but this did not reach the suggestive threshold (online Supplementary Figure 5). Results of sensitivity analyses, omitting each study in turn, are shown in online Supplementary Figures 6 and 7. The largest changes: bin 8.2 (8p) achieves the suggestive linkage threshold without the Taiwan data set; bin 2.8 (2q) becomes substantially more significant (but not achieving genome-wide significance) without the Japan data set; bin 5.6 (5q) becomes substantially more significant (but not genome-wide significant) without the Suarez *et al*. data set (study 21); and bin 6.3 (6pq) would join the list of nominally significant bins without either the Taiwan or Japan data set.

Type I error

Type I error was calculated for the unlinked GSMA simulations (120 000 30-cM bins) by determining the number of observed bins in which the GSMA program computed P_{SR} values (based on permutation alone) that were less than the theoretical threshold for nominal significance (0.05); for genome-wide suggestive linkage, or a value expected once per GSMA (1/120 = 0.00833) or genome-wide significant linkage (0.05/120 = 0.00042). The GSMA program's permutation procedure produced type I errors that were slightly liberal for ALL (0.053, 0.0096 and 0.00051) and less liberal for EUR studies (0.051, 0.0091 and 0.00042). The source of the discrepancy is not clear, but we have used the permutation-based suggestive and significant values here for single bins (0.0077 and 0.00037 for ALL, and 0.0078 and 0.00044 for EUR studies) and an empirical threshold for aggregate genome-wide significance as described above.

Power

Table 4 summarizes the results of power calculations from simulation studies. Power is lower for edge bins for multipoint analyses, so we computed a weighted average of power assuming 20% edge and 80% mid-chromosome bins (see Table 4 legend). Power was excellent to detect significant linkage at multiple loci with observed population-wide $\lambda_S = 1.3$ (30% increased risk in sibs) across ALL studies or in EUR studies alone. For $\lambda_S = 1.15$, there was excellent power to detect significant linkage in multiple bins (for ALL studies), or to detect suggestive evidence for linkage in multiple bins (if limited to EUR studies); there was reduced power in the presence of substantial heterogeneity. Genetic effects limited to a defined subset (EUR) were more readily detected in a separate analysis.

Discussion

Meta-analysis of 32 GWLS, encompassing 7413 individuals affected with SCZ or related disorders in 3255 pedigrees, produced evidence for significant linkage in the genome based on empirical aggregate criteria: 10 30-cM bins that achieved bin-wise $P_{SR} < 0.046$. Genome-wide evidence for linkage was detected on chromosome 2q (118.7-152 Mb) in a secondary analysis that shifted bin location by 50%, consistent with our previous finding of significant linkage in the same region in our previous GSMA of 20 studies.[4] Suggestive evidence for linkage was detected in ALL studies in two single bins, on chromosomes 5q and 2q. Separate analysis of European-ancestry samples produced linkage evidence falling just short of the genome-wide significance in bin 8.2 (15.7-32.7 Mb).

It is widely assumed that many loci contribute to SCZ susceptibility. Table 4 shows that for the GSMA method, power to detect at least one true linkage goes up with the number of linked loci, but the power to detect any specific locus decreases, because in a rank-ordered test there is a finite number of high ranks in each study. But if there were many loci with locus-specific λ_S values of 1.3 or even 1.15 (averaged across all study populations and accounting for the effects of various sources of measurement error), we would expect to detect genome-wide significant linkage in several individual bins, and suggestive evidence for linkage in many more than two bins. It would be reasonable to conclude that there are probably no loci with such strong effects worldwide or across all European populations, although they might exist within particular populations for which samples of this size are not available or within subsets of families, which we cannot yet identify in advance.

The power of linkage analysis for a locus is predicted by its contribution to the relative risk (RR) to siblings of probands (λ_S),[57] whereas the power of association analysis is predicted by allelic or genotypic RRs, that is, the increased risk to individual carriers.[5] GWA studies have produced robust association findings for many complex genetic disorders[58-60] typically with RRs of 1.1-1.5. A variant found on 10% of chromosomes conferring a ‘large’ RR of 1.5 per allele in a multiplicative model (2.25 for homozygous carriers) would produce a population-wide sibling RR of only 1.02 (undetectable by linkage). But GWA chips are unlikely to tag all pathogenic variants. For example, in NOD2 there are three rare variants, each with a frequency of less than 5%, that confer moderate risks of Crohn’s disease. Their individual effects generate only weak signals when assayed by the Affymetrix 500K chip because they are poorly tagged; but combined, they confer a sibling RR of 1.16, which is detectable in a very large linkage sample.[58,61] This illustrates a strength of linkage analysis: allelic heterogeneity is difficult to detect in association studies, as the genetic effect is split across variants, but linkage can detect the pooled effect of all variants (not only SNPs) within one or more susceptibility genes or elements in a region.

Readers may be interested to know the location of SCZ candidate genes or regions in relation to bins that produced evidence for linkage here. On chromosome 1q, NOS1AP (160 Mb; previously known as CAPON),[62,63] RGS4 (161.3 Mb)[64,65] and UHMK1 (160.7 Mb)[66] are near the telomeric edge of bin 1.6 (114.6-162.1 Mb), and the rare (usually *de novo*) SCZ-associated CNV is located in a different part of the same bin (145-146 Mb).[67,68] On chromosome 8p, PPP3CC (22.35 Mb)[69] and NRG1 (31.6-32.7 Mb)[52,70] are both within bin 8.2 (15.7-32.7 Mb). On chromosome 2q, ZNF804A, which contains a SNP that produced genome-wide significant evidence for association in a combined SCZ-bipolar disorder sample, [71] is in bin 2.7 at 185 Mb. This bin was not nominally significant in the primary analysis, but 185 Mb is within nominally significant bins in two secondary analyses (one that shifted bin boundaries by 50% and one that considered 40 cM bin widths). DTNBP1, DAOA, TRAR4/STX7, CHRNA7, COMT/ARVCF, DISC1, AKT1, HTR2A and DRD2 are not in bins listed in Tables 2 or 3. Many of the research groups whose samples are included here are actively

investigating association of SCZ to sequence variants in genes identified in linkage regions or by the more recent genome-wide association methodology.

In conclusion, this updated GSMA of 32 SCZ studies showed significant evidence for linkage on chromosome 2q in a secondary analysis, suggestive evidence for linkage on chromosomes 5q and 2q in the primary analysis, and suggestive evidence for linkage on 8p in European samples. Genome-wide significant aggregate evidence for linkage (that is, more modestly significant results than expected by chance) was observed based on results for 10 bins in 8 nonadjacent chromosomal regions. As results from genome-wide association studies emerge, it may be important to keep in mind that there may be SCZ susceptibility loci in some of these consensus linkage regions that cannot be detected by common tag SNPs. It is likely that diverse methods will be required to identify specific DNA sequence variants and to confirm and define their role in the etiology of SCZ.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

The work reported here was supported by: Medical Research Council (UK) Grants G0400960 (CML) and G9309834 (MO); National Institute of Mental Health Grants 7R01MH062276 (to DFL [Aust/US], CL [France/La Réunion], MO [Cardiff] and DW [Bonn]), 5R01MH068922 (to PG [ENH]), 5R01MH068921 (to AEP [Johns Hopkins]), 5R01MH068881 (to BR [VCU/Ireland]), MH-41953 (to KSK [VCU/Ireland]); MH63356 and MH80299 (to WB [Palau]); MH58586 (to JMS [Aust/US]); MH 56242 (to VLM [US/Sweden]), MH61399 (EMW, MK [Kosrae, South Africa]), NIMH Grant MH062440, Canadian Institutes of Health Research Grants MOP-53216 and MOP-12155, a National Alliance for Research on Schizophrenia and Depression Distinguished Investigator Award and Canada Research Chair in Schizophrenia Genetics (LMB, ASB [Canada]); Australian National Health and Medical Research Council Grants 910234, 941087, and 971095 (to BJM [Aust/US]), MRC project Grant G880473N, The European Science Foundation, SANE, the Iceland Department of Health, the General Hospital Reykjavik, the Joseph Levy Charitable Foundation, the Wellcome Trust Grant 055379, The Priory Hospital, the Neuroscience Research Charitable Trust, the University of Iceland and the Icelandic Science Council (HMDG [UCL]); Warner-Lambert, Parke-Davis Pharmaceuticals Company and NIMH Grant R01-MH44245 (LEL [US/International]); the Deutsche Forschungsgemeinschaft (HWM [Kiel]; MA WM, SGS, DBW [Indonesia]); the German Israeli Foundation for Scientific Research (BL; DBW); Mammalian Genotyping Service HV48141 (DBW [Indonesia]; CREST of JST (Japan Science and Technology Agency) TA [Japan]; Pfizer, Inc. and the SANE Foundation (LEL [Costa Rica]); the Israel Science Foundation, US. Israel Binational Science Foundation, the National Alliance for Research on Schizophrenia and Depression, and the Harry Stern Family Foundation (BL and YK [Israel]); the VA Merit Review Program (AF); recruitment of the NIMH Genetics Initiative sample was supported by NIMH Grants 5 UO1MH46318, UO1MH46289 and UO1MH46276; the Taiwan Schizophrenia Linkage Study was supported by NIMH Grant 1R01 MH59624-01 and Grant NHRI-90-8825PP, NHRI -EX91,92-9113PP from the National Health Research Institute, Taiwan, and support from the Genomic Medicine Research Program of Psychiatric Disorders, National Taiwan University Hospital; the VA Linkage Study was supported by funds from the Department of Veterans Affairs Cooperative Studies Program; the US/Mexico/Central America study was supported by a collaborative NIMH grant (‘Genetics of Schizophrenia in Latino Populations’) (MH60881 and MH60875) to ME [University of Texas Health Science Center at San Antonio], R Mendoza [University of California at Los Angeles-Harbor], HR [University of Costa Rica, San Jose, Costa Rica], A Ontiveros [Instituto de Informacion de Investigacion en Salud Mental, Monterrey, Mexico], HN [Medical and Family Research Group, Carracci SC, Mexico City, Mexico], and R Munoz [Family Health Centers of San Diego, CA].

References

1. Levinson DF, Levinson MD, Segurado R, Lewis CM. Genome scan meta-analysis of schizophrenia and bipolar disorder, part I: Methods and power analysis. Am J Hum Genet 2003;73:17–33. [PubMed: 12802787]
2. Pardi F, Levinson DF, Lewis CM. GSMA: software implementation of the genome search meta-analysis method. Bioinformatics 2005;21:4430–4431. [PubMed: 16249265]
3. Wise LH, Lanchbury JS, Lewis CM. Meta-analysis of genome searches. Ann Hum Genet 1999;63(Part 3):263–272. [PubMed: 10738538]

4. Lewis CM, Levinson DF, Wise LH, DeLisi LE, Straub RE, Hovatta I, et al. Genome scan meta-analysis of schizophrenia and bipolar disorder, part II: Schizophrenia. Am J Hum Genet 2003;73:34–48. [PubMed: 12802786]
5. Risch N, Merikangas K. The future of genetic studies of complex human diseases. Science 1996;273:1516–1517. [PubMed: 8801636]
6. McCarroll SA, Altshuler DM. Copy-number variation and association studies of human disease. Nat Genet 2007;39(7 Suppl):S37–S42. [PubMed: 17597780]
7. Manolio TA, Brooks LD, Collins FS. A HapMap harvest of insights into the genetics of common disease. J Clin Invest 2008;118:1590–1605. [PubMed: 18451988]
8. McMillan I, Roberson A. Power of detection of major genes affecting quantitative characters. Heredity 1974;32:349–356. [PubMed: 4528799]
9. Risch N. Genetic linkage and complex diseases, with special reference to psychiatric disorders. Genet Epidemiol 1990;7:3–16. [PubMed: 2184091]discussion 17-45
10. Jablensky A, Sartorius N, Ernberg G, Anker M, Korten A, Cooper JE, et al. Schizophrenia: manifestations, incidence and course in different cultures. A World Health Organization ten-country study. Psychol Med Monogr Suppl 1992;20:1–97. [PubMed: 1565705]
11. Cardno AG, Marshall EJ, Coid B, Macdonald AM, Ribchester TR, Davies NJ, et al. Heritability estimates for psychotic disorders: the Maudsley twin psychosis series. Arch Gen Psychiatry 1999;56:162–168. [PubMed: 10025441]
12. Farmer AE, McGuffin P, Gottesman II. Twin concordance for DSM-III schizophrenia. Scrutinizing the validity of the definition. Arch Gen Psychiatry 1987;44:634–641. [PubMed: 3606329]
13. Faraone SV, Blehar M, Pepple J, Moldin SO, Norton J, Nurnberger JI, et al. Diagnostic accuracy and confusability analyses: an application to the Diagnostic Interview for Genetic Studies. Psychol Med 1996;26:401–410. [PubMed: 8685296]
14. Kendler KS, Neale MC, Walsh D. Evaluating the spectrum concept of schizophrenia in the Roscommon Family Study. Am J Psychiatry 1995;152:749–754. [PubMed: 7726315]
15. Moises HW, Yang L, Kristbjarnarson H, Wiese C, Byerley W, Macciardi F, et al. An international two-stage genome-wide search for schizophrenia susceptibility genes. Nat Genet 1995;11:321–324. [PubMed: 7581457]
16. Devlin B, Bacanu SA, Roeder K, Reimherr F, Wender P, Galke B, et al. Genome-wide multipoint linkage analyses of multiplex schizophrenia pedigrees from the oceanic nation of Palau. Mol Psychiatry 2002;7:689–694. [PubMed: 12192612]
17. Brzustowicz LM, Hodgkinson KA, Chow EW, Honer WG, Bassett AS. Location of a major susceptibility locus for familial schizophrenia on chromosome 1q21-q22. Science 2000;288:678–682. [PubMed: 10784452]
18. Paunio T, Ekelund J, Varilo T, Parker A, Hovatta I, Turunen JA, et al. Genome-wide scan in a nationwide study sample of schizophrenia families in Finland reveals susceptibility loci on chromosomes 2q and 5q. Hum Mol Genet 2001;10:3037–30348. [PubMed: 11751686]
19. Garver DL, Holcomb J, Mapua FM, Wilson R, Barnes B. Schizophrenia spectrum disorders: an autosomal-wide scan in multiplex pedigrees. Schizophr Res 2001;52:145–160. [PubMed: 11705708]
20. Gurling HM, Kalsi G, Brynjolfson J, Sigmundsson T, Sherrington R, Mankoo BS, et al. Genomewide genetic linkage analysis confirms the presence of susceptibility loci for schizophrenia, on chromosomes 1q32. 2, 5q33. 2, and 8p21-22 and provides support for linkage to schizophrenia, on chromosomes 11q23. 3-24 and 20q12. 1-11. 23. Am J Hum Genet 2001;68:661–673. [PubMed: 11179014]
21. Lindholm E, Ekholm B, Shaw S, Jalonen P, Johansson G, Pettersson U, et al. A schizophrenia-susceptibility locus at 6q25, in one of the world's largest reported pedigrees. Am J Hum Genet 2001;69:96–105. [PubMed: 11389481]
22. DeLisi LE, Shaw SH, Crow TJ, Shields G, Smith AB, Larach VW, et al. A genome-wide scan for linkage to chromosomal regions in 382 sibling pairs with schizophrenia or schizoaffective disorder. Am J Psychiatry 2002;159:803–812. [PubMed: 11986135]
23. DeLisi LE, Mesen A, Rodriguez C, Bertheau A, LaPrade B, Llach M, et al. Genome-wide scan for linkage to schizophrenia in a Spanish-origin cohort from Costa Rica. Am J Med Genet 2002;114:497–508. [PubMed: 12116183]

24. Wijsman EM, Rosenthal EA, Hall D, Blundell ML, Sobin C, Heath SC, et al. Genome-wide scan in a large complex pedigree with predominantly male schizophrenics from the island of Kosrae: evidence for linkage to chromosome 2q. Mol Psychiatry 2003;8:695–705. 643. [PubMed: 12874606]
25. Lerer B, Segman RH, Hamdan A, Kanyas K, Karni O, Kohn Y, et al. Genome scan of Arab Israeli families maps a schizophrenia susceptibility gene to chromosome 6q23 and supports a locus at chromosome 10q24. Mol Psychiatry 2003;8:488–498. [PubMed: 12808429]
26. Arinami T, Ohtsuki T, Ishiguro H, Ujike H, Tanaka Y, Morita Y, et al. Genomewide high-density SNP linkage analysis of 236 Japanese families supports the existence of schizophrenia susceptibility loci on chromosomes 1p, 14q, and 20p. Am J Hum Genet 2005;77:937–944. [PubMed: 16380906]
27. Fallin MD, Lasseter VK, Wolyniec PS, McGrath JA, Nestadt G, Valle D, et al. Genomewide linkage scan for schizophrenia susceptibility loci among Ashkenazi Jewish families shows evidence of linkage on chromosome 10q22. Am J Hum Genet 2003;73:601–611. [PubMed: 12929083]
28. Williams NM, Norton N, Williams H, Ekholm B, Hamshere ML, Lindblom Y, et al. A systematic genomewide linkage study in 353 sib pairs with schizophrenia. Am J Hum Genet 2003;73:1355–1367. [PubMed: 14628288]
29. Sklar P, Pato MT, Kirby A, Petryshen TL, Medeiros H, Carvalho C, et al. Genome-wide scan in Portuguese Island families identifies 5q31-5q35 as a susceptibility locus for schizophrenia and psychosis. Mol Psychiatry 2004;9:213–218. [PubMed: 14699422]
30. Abecasis GR, Burt RA, Hall D, Bochum S, Doheny KF, Lundy SL, et al. Genomewide scan in families with schizophrenia from the founder population of Afrikaners reveals evidence for linkage and uniparental disomy on chromosome 1. Am J Hum Genet 2004;74:403–417. [PubMed: 14750073]
31. Maziade M, Roy MA, Chagnon YC, Cliche D, Fournier JP, Montgrain N, et al. Shared and specific susceptibility loci for schizophrenia and bipolar disorder: a dense genome scan in Eastern Quebec families. Mol Psychiatry 2005;10:486–499. [PubMed: 15534619]
32. Faraone SV, Skol AD, Tsuang DW, Young KA, Haverstock SL, Prabhudesai S, et al. Genome scan of schizophrenia families in a large Veterans Affairs Cooperative Study sample: evidence for linkage to 18p11. 32 and for racial heterogeneity on chromosomes 6 and 14. Am J Med Genet B Neuropsychiatr Genet 2005;139:91–100. [PubMed: 16152571]
33. Suarez BK, Duan J, Sanders AR, Hinrichs AL, Jin CH, Hou C, et al. Genomewide linkage scan of 409 European-ancestry and African American families with schizophrenia: suggestive evidence of linkage at 8p23. 3-p21. 2 and 11p13. 1-q14. 1 in the combined sample. Am J Hum Genet 2006;78:315–333. [PubMed: 16400611]
34. Escamilla MA, Ontiveros A, Nicolini H, Raventos H, Mendoza R, Medina R, et al. A genome-wide scan for schizophrenia and psychosis susceptibility loci in families of Mexican and Central American ancestry. Am J Med Genet B Neuropsychiatr Genet 2007;144:193–199. [PubMed: 17044102]
35. Faraone SV, Hwu HG, Liu CM, Chen WJ, Tsuang MM, Liu SK, et al. Genome scan of Han Chinese schizophrenia families from Taiwan: confirmation of linkage to 10q22. 3. Am J Psychiatry 2006;163:1760–1766. [PubMed: 17012687]
36. Levinson DF, Mahtani MM, Nancarrow DJ, Brown DM, Kruglyak L, Kirby A, et al. Genome scan of schizophrenia. Am J Psychiatry 1998;155:741–750. [PubMed: 9619145]
37. Mowry BJ, Ewen KR, Nancarrow DJ, Lennon DP, Nertney DA, Jones HL, et al. Second stage of a genome scan of schizophrenia: study of five positive regions in an expanded sample. Am J Med Genet 2000;96:864–869. [PubMed: 11121199]
38. Holmans P, Riley B, Pulver AE, Owen MJ, Wildenauer DB, Gejman PV. Genomewide linkage scan of schizophrenia in a large multicenter pedigree sample using single nucleotide polymorphisms. Mol Psychiatry. in press
39. Schwab SG, Hallmayer J, Albus M, Lerer B, Eckstein GN, Borrmann M, et al. A genome-wide autosomal screen for schizophrenia susceptibility loci in 71 families with affected siblings: support for loci on chromosome 10p and 6. Mol Psychiatry 2000;5:638–649. [PubMed: 11126394]
40. Bonnet-Brilhault F, Laurent C, Campion D, Thibaut F, Lafargue C, Charbonnier F, et al. No evidence for involvement of KCNN3 (hSKCa3) potassium channel gene in familial and isolated cases of schizophrenia. Eur J Hum Genet 1999;7:247–250. [PubMed: 10196711]
41. Campion D, d'Amato T, Bastard C, Laurent C, Guedj F, Jay M, et al. Genetic study of dopamine D1, D2, and D4 receptors in schizophrenia. Psychiatry Res 1994;51:215–230. [PubMed: 7911585]

UKPMC Funders Group Author Manuscript

42. Cao Q, Martinez M, Zhang J, Sanders AR, Badner JA, Cravchik A, et al. Suggestive evidence for a schizophrenia susceptibility locus on chromosome 6q and a confirmation in an independent series of pedigrees. Genomics 1997;43:1–8. [PubMed: 9226366]
43. Blouin JL, Dombroski BA, Nath SK, Lasseter VK, Wolyniec PS, Nestadt G, et al. Schizophrenia susceptibility loci on chromosomes 13q32 and 8p21. Nat Genet 1998;20:70–73. [PubMed: 9731535]
44. Faraone SV, Matise T, Svrakic D, Pepple J, Malaspina D, Suarez B, et al. Genome scan of European-American schizophrenia pedigrees: results of the NIMH Genetics Initiative and Millennium Consortium. Am J Med Genet 1998;81:290–295. [PubMed: 9674973]
45. Kaufmann CA, Suarez B, Malaspina D, Pepple J, Svrakic D, Markel PD, et al. NIMH Genetics Initiative Millenium Schizophrenia Consortium: linkage analysis of African-American pedigrees. Am J Med Genet 1998;81:282–289. [PubMed: 9674972]
46. Straub RE, MacLean CJ, Ma Y, Webb BT, Myakishev MV, Harris-Kerr C, et al. Genome-wide scans of three independent sets of 90 Irish multiplex schizophrenia families and follow-up of selected regions in all families provides evidence for multiple susceptibility genes. Mol Psychiatry 2002;7:542–559. [PubMed: 12140777]
47. Irmansyah, Schwab SG, Heriani, Handoko HY, Kusumawardhani I, Widyawat I, et al. Genome-wide scan in 124 Indonesian sib-pair families with schizophrenia reveals genome-wide significant linkage to a locus on chromosome 3p26-21. Am J Med Genet B Neuropsychiatr Genet 2008;147B:1245–1252. [PubMed: 18449910]
48. Coon H, Jensen S, Holik J, Hoff M, Myles-Worsley M, Reimherr F, et al. Genomic scan for genes predisposing to schizophrenia. Am J Med Genet 1994;54:59–71. [PubMed: 7909992]
49. Cooper-Casey K, Mesen-Fainardi A, Galke-Rollins B, Llach M, Laprade B, Rodriguez C, et al. Suggestive linkage of schizophrenia to 5p13 in Costa Rica. Mol Psychiatry 2005;10:651–656. [PubMed: 15700049]
50. Vazza G, Bertolin C, Scudellaro E, Vettori A, Boaretto F, Rampinelli S, et al. Genome-wide scan supports the existence of a susceptibility locus for schizophrenia and bipolar disorder on chromosome 15q26. Mol Psychiatry 2007;12:87–93. [PubMed: 16969366]
51. Bulayeva KB, Glatt SJ, Bulayev OA, Pavlova TA, Tsuang MT. Genome-wide linkage scan of schizophrenia: a cross-isolate study. Genomics 2007;89:167–177. [PubMed: 17140763]
52. Stefansson H, Sigurdsson E, Steinthorsdottir V, Bjornsdottir S, Sigmundsson T, Ghosh S, et al. Neuregulin 1 and susceptibility to schizophrenia. Am J Hum Genet 2002;71:877–892. [PubMed: 12145742]
53. Matise TC, Chen F, Chen W, De La Vega FM, Hansen M, He C, et al. A second-generation combined linkage physical map of the human genome. Genome Res 2007;17:1783–1786. [PubMed: 17989245]
54. Lander E, Kruglyak L. Genetic dissection of complex traits: guidelines for interpreting and reporting linkage results. Nat Genet 1995;11:241–247. [PubMed: 7581446]
55. Zintzaras E, Ioannidis JP. Heterogeneity testing in meta-analysis of genome searches. Genet Epidemiol 2005;28:123–137. [PubMed: 15593093]
56. Lewis CM, Levinson DF. Testing for genetic heterogeneity in the genome search meta-analysis method. Genet Epidemiol 2006;30:348–355. [PubMed: 16586403]
57. Risch N. Linkage strategies for genetically complex traits. I. Multilocus models. Am J Hum Genet 1990;46:222–228. [PubMed: 2301392]
58. Wellcome Trust Case Control Consortium. Genome-wide association study of 14,000 cases of seven common diseases and 3,000 shared controls. Nature 2007;447:661–678. [PubMed: 17554300]
59. Frayling TM. Genome-wide association studies provide new insights into type 2 diabetes aetiology. Nat Rev Genet 2007;8:657–662. [PubMed: 17703236]
60. Mathew CG. New links to the pathogenesis of Crohn disease provided by genome-wide association scans. Nat Rev Genet 2008;9:9–14. [PubMed: 17968351]
61. Lewis CM, Whitwell SC, Forbes A, Sanderson J, Mathew CG, Marteau TM. Estimating risks of common complex diseases across genetic and environmental factors: the example of Crohn disease. J Med Genet 2007;44:689–694. [PubMed: 17660460]
62. Brzustowicz LM, Simone J, Mohseni P, Hayter JE, Hodgkinson KA, Chow EW, et al. Linkage disequilibrium mapping of schizophrenia susceptibility to the CAPON region of chromosome 1q22. Am J Hum Genet 2004;74:1057–1063. [PubMed: 15065015]

63. Xu B, Wratten N, Charych EI, Buyske S, Firestein BL, Brzustowicz LM. Increased expression in dorsolateral prefrontal cortex of CAPON in schizophrenia and bipolar disorder. PLoS Med 2005;2:e263. [PubMed: 16146415]

64. Guo S, Tang W, Shi Y, Huang K, Xi Z, Xu Y, et al. RGS4 polymorphisms and risk of schizophrenia: an association study in Han Chinese plus meta-analysis. Neurosci Lett 2006;406:122–127. [PubMed: 16904822]

65. Levitt P, Ebert P, Mirnics K, Nimgaonkar VL, Lewis DA. Making the case for a candidate vulnerability gene in schizophrenia: convergent evidence for regulator of G-protein signaling 4 (RGS4). Biol Psychiatry 2006;60:534–537. [PubMed: 16860780]

66. Puri V. Fine mapping by genetic association implicates the chromosome 1q23.3 gene UHMK1, encoding a serine/threonine protein kinase, as a novel schizophrenia susceptibility gene. Biol Psychiatry 2007;61:873–879. [PubMed: 16978587]

67. Stefansson H, Rujescu D, Cichon S, Pietilainen OP, Ingason A, Steinberg S, et al. Large recurrent microdeletions associated with schizophrenia. Nature 2008;455:232–236. [PubMed: 18668039]

68. Stone JL, O'Donovan MC, Gurling H, Kirov GK, Blackwood DH, Corvin A, et al. Rare chromosomal deletions and duplications increase risk of schizophrenia. Nature 2008;455:237–241. [PubMed: 18668038]

69. Gerber DJ, Hall D, Miyakawa T, Demars S, Gogos JA, Karayiorgou M, et al. Evidence for association of schizophrenia with genetic variation in the 8p21. 3 gene, PPP3CC, encoding the calcineurin gamma subunit. Proc Natl Acad Sci USA 2003;100:8993–8998. [PubMed: 12851458]

70. Stefansson H, Sarginson J, Kong A, Yates P, Steinthorsdottir V, Gudfinnsson E, et al. Association of neuregulin 1 with schizophrenia confirmed in a Scottish population. Am J Hum Genet 2003;72:83–87. [PubMed: 12478479]

71. O'Donovan MC, Craddock N, Norton N, Williams H, Peirce T, Moskvina V, et al. Identification of loci associated with schizophrenia by genome-wide association and follow-up. Nat Genet 2008;40:1053–1055. [PubMed: 18677311]

UKPMC Funders Group Author Manuscript

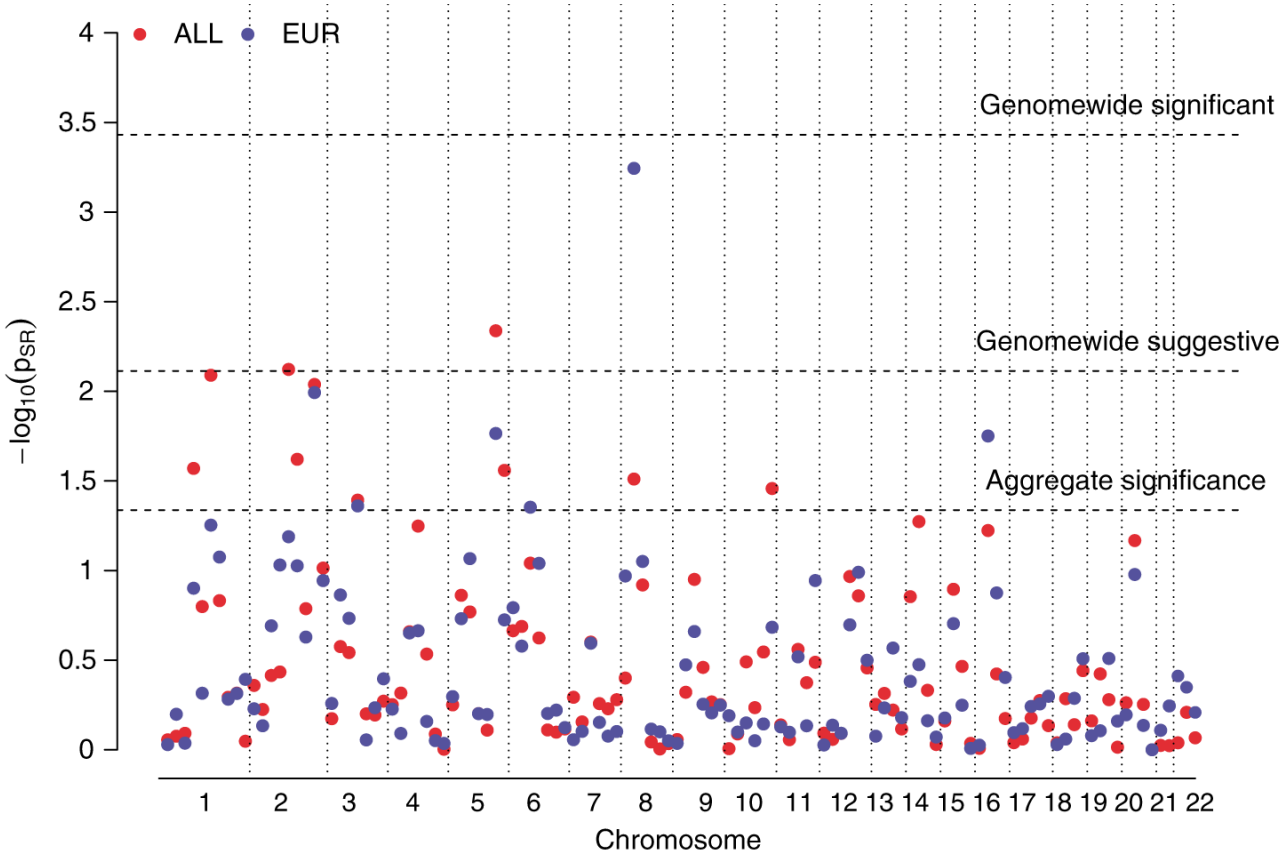

Figure 1.

Genomewide results for primary analysis (weighted, 30 cM bins) for ALL studies (red) and EUR studies (blue). The two upper dashed lines represent empirical thresholds for genomewide significant (theoretical P_{SR} = 0.00037) and suggestive (theoretical $P_{SR=}$ 0.0077) evidence for linkage for a single bin for ALL studies; and the lower dashed line represents the threshold for aggregate significance for ALL studies (theoretical P_{SR} = 0.046 in 10 or more bins). The thresholds for EUR studies were slightly less stringent (P_{SR} values of 0.00044, 0.0078 and 0.0475 (in 10 or more bins) respectively), but the Figure shows all EUR bins within the region for their correct threshold of significance.

UKPMC Funders Group Author Manuscript

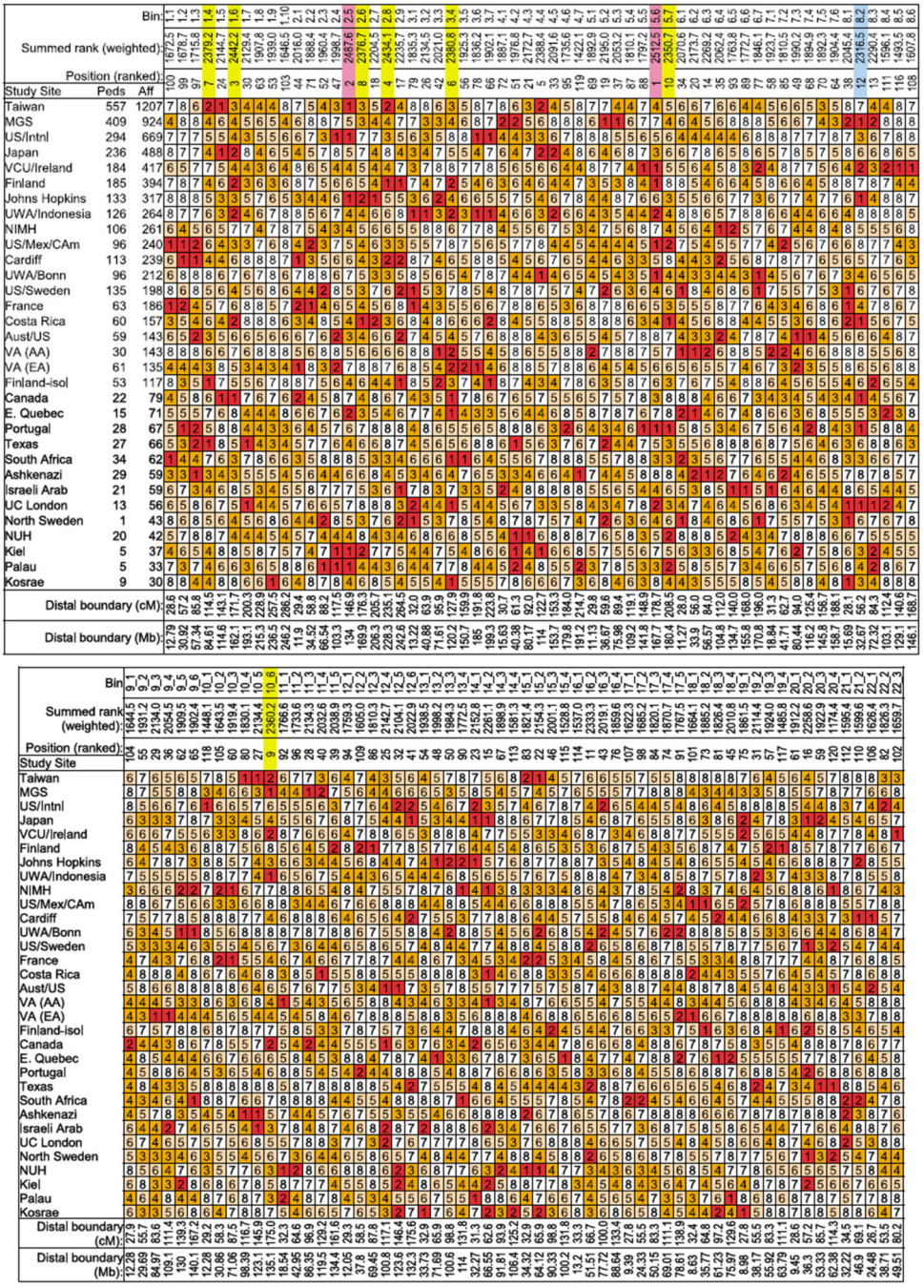

Figure 2.

Ranks by study and summed ranks. Shown for each study is the within-study rank of each bin. '1' against a red background indicates bins with ranks of 116-120 (the bin containing the most significant linkage score in a study is ranked 120); 2 indicates ranks 111-115; 3—ranks 101-110; 4—ranks 81-100; 5—ranks 61-80; 6—ranks 41-60; 7—ranks 21-40; and 8—ranks 1-20 (cells with 7 and 8 have a white background). Shown at the top of each part of the figure are the bin number, the weighted summed rank for each bin across studies (i.e., weighted for each sample by the square root of the N of genotyped affected individuals), and the rank-ordered position of that bin in the ALL analysis (i.e., 1 is the best position, see details in Table 2). Purple background indicates bins with empirical suggestive evidence for linkage in the ALL

analysis; 10 bins shaded in purple, yellow or blue met the threshold for aggregate genome-wide evidence for linkage (10 bins with nominal $P < 0.046$); and bin 8.2 (shaded blue) also met empirical suggestive evidence for linkage in the EUR analysis. Shown at the bottom is the distal boundary of each bin in Rutgers cM and in Mb (build 36). Note that tied ranks can result in uneven numbers of bins in a grouping, particularly for lower ranks when there are many zero or negative scores. See Table 1 for the references associated with each study.

UKPMC Funders Group Author Manuscript

UKPMC Funders Group Author Manuscript

Table 1

Genome-wide linkage scan characteristics (ALL and European-ancestry analyses)

	Study site or project[a] *(previous(p)/ modified(m)/ new(n))*	*Diagnostic criteria*[b]	*ALL analysis*		*EUR analysis*		*Markers*	*Program*	*Statistics*	*Ethnicity*[c]
			Ped	*Aff*	*Ped*	*Aff*				
1	Kiel[15] (p)	SCZ, SA (RDC or D3R)	5	37	5	37	413	WRPC	*P*-values	Icelandic
2	Palau[16] (p)	SCZ, SAS (RDC)	5	33			496	LINKAGE	LOD	Palau
3	Canada[17] (p)	SCZ, SA (D3R)	22	79	22	79	381	FASTLINK	HLOD	Celtic Canada
4	Finland—isolate[18] (p)	SCZ; +SA (D4)	53	117	53	117	345	MLINK	LOD	Finnish
5	Finland—national[18] (p)	SCZ; +SA (D4)	185	394	185	394	345	MLINK	LOD	Finnish
6	Texas[19] (p)	SCZ (D4)	27	66			406	Genehunter Plus	NPL	AA, few EA
7	UC London[20] (p)	SCZ,SA,UFP (RDC)	13	56	13	56	365	MFLINK	*P*-values	Icelandic
8	North Sweden[21] (p)	SCZ;+SAD;+PNOS;+SAB (D4)	1	43	1	43	371	LINKAGE	LOD	Swedish
9	US/International[22] (p)	SCZ, SA (D3R)	294	669	294	669	396	Mapmaker/Sibs	MLS	EA (inc Chile)
10	Costa Rica[23] (p)	SCZ, SA (D4)	60	157			404	Genehunter Plus	KCLOD	Costa Rican
11	Kosrae[24] (n)	SCZ, SA (D4)	9	30			398	FASTLINK	LOD	Kosrae
12	Israeli Arab[25] (n)	SCZ, SAD (D4)	21	59			350	Allegro	NPL	Arab
13	Japan[26] (n)	SCZ, SA (D4)	236	488			5861	Merlin	KCLOD	Japanese
14	Ashkenazi[27] (n)	SCZ, SA (D4)	29	59	29	59	382	Genehunter	LOD	Ashkenazi
15	US/Sweden[28] (m)	SCZ, SA (D4)	135	198	135	198	392	Mapmaker/Sibs	MLS	Swedish, US
16	Portugal[29] (n)	SCZ, SAD (D4)	28	67	28	67	366	Genehunter	NPL	Portuguese
17	South Africa[30] (n)	SCZ, SAD (D4)	34	62	34	62	388	Merlin	KCLOD	Afrikaner
18	Eastern Quebec[31] (n)	SCZ (D3R)	15	71	15	71	350	FASTLINK	LOD	Quebecois
19	VA (EA)[32] (n)	SCZ, SAD (D3R)	61	135	61	135	414	Merlin	KCLOD	EA
20	VA (AA)[32] (n)	SCZ, SAD (D3R)	60	143			414	Merlin	KCLOD	AA
21	MGS[33] (n)	SCZ, SA (D4)	409	924	263	571	400	Genehunter Plus	Zlr	EA, AA
22	US/Mexico/Central America[34] (n)	SCZ, SA (D4)	96	240			404	Merlin	KCLOD	Mexican, Cent Am
23	Taiwan[35] (n)	SCZ (D4)	557	1207			286	Merlin	Zlr	Han Chinese
24	Aust/US[36-38] (m)	SCZ, SA (D3R)	59	143	49	114	5868	Allegro	Zlr	EA, AA, Other

UKPMC Funders Group Author Manuscript

	Study site or project[a] (previous(p)/ modified(m)/ new(n))	Diagnostic criteria[b]	ALL analysis		EUR analysis		Markers	Program	Statistics	Ethnicity[c]
			Ped	Aff	Ped	Aff				
25	UWA/Bonn[38,39] (m)	SCZ, SA (D3R)	96	212	96	212	5868	Allegro	Zlr	German,Israeli
26	Cardiff[28,38] (m)	SCZ, SA (D3R)	113	239	113	239	5868	Allegro	Zlr	UK
27	France/ LaRéunion[38,40,41] (n)	SCZ, SA (D3R)	63	186	29	75	5868	Allegro	Zlr	EA, AA, Other
28	NUH[38,42] (n)	SCZ, SA (D3R)	20	42	19	40	5868	Allegro	Zlr	EA, AA
29	Johns Hopkins[38,43] (m)	SCZ, SA (D3R)	133	317	124	295	5868	Allegro	Zlr	EA, AA, Other
30	NIMH[38,44,45] (m)	SCZ, SA (D3R)	106	261	61	142	5868	Allegro	Zlr	EA, AA, Other
31	VCU/Ireland[38,46] (m)	SCZ, SAPO (D3R)	184	417	184	417	5868	Allegro	Zlr	Irish
32	UWA/Indonesia[38,47] (n)	SCZ (D3R)	124	267			400	Genehunter	MLS	Indonesian
		Total	3283	7476	1835	4141				

Other abbreviations: Ped = informative pedigrees; Aff = genotyped affected cases. VA = US Veterans' Administration; MGS = Molecular Genetics of Schizophrenia collaboration; Aust/US = Australia/US collaboration; NUH = NorthShore University HealthSystem Research Institute; NIMH = National Institute of Mental Health Schizophrenia Genetics Initiative; VCU = Virginia Commonwealth University; UWA = University of Western Australia. LOD = log of the odds ratio under one or more parametric models; HLOD = heterogeneity LOD scores (generally, maximized over at least two models); NPL = nonparametric linkage *Z*-score; MLS = maximum LOD score (affected sib-pair analysis, constrained to possible triangle); Zlr = Z-likelihood ratio statistic; KCLOD = Kong-Cox equivalent LOD score computed from Zlr.

[a] Sorted by year of publication of the data used in this analysis. The letter in parentheses indicates whether the dataset was included in the previous analysis (p), has been modified with new genotyping and/or additional pedigrees (m), or is new (n).

[b] D3R = DSM-IIIR criteria; D4 = DSM-IV criteria; RDC = Research Diagnostic Criteria; SA = schizoaffective disorder (SAS = RDC mainly schizophrenic type; SAD = depressed type; SAB = bipolar type; SAPO = poor outcome; PNOS = psychosis not otherwise specified; UFP = unspecified functional psychosis (RDC). Diagnoses separated by commas constitute one model; sets separated by semicolons are alternative models over which scores were maximized, with '+' indicating that diagnoses were added to the ones in the next narrower model.

[c] EA = mixed European ancestry (usually from the US, but also mixed US, European and Australian samples); AA = African American.

UKPMC Funders Group Author Manuscript

Table 2

Bins achieving nominally significant evidence for linkage (ALL studies)

Pos	*Bin*	*cM*	*Mb*	P_{SR}	P_{OR}
1	5.6*	148.9-178.7	141.8-167.7	**0.00459**	0.43156
2	2.5*	117.5-146.9	103.3-134.0	**0.00755**	0.22798
3	1.6*	143.1-171.7	114.6-162.1	0.00814	0.06639
4	2.8	205.7-235.1	206.3-228.3	0.00916	0.01970
5	2.6*	146.9-176.3	134.0-169.9	0.02395	0.14269
6	1.4	85.8-114.5	57.3-84.6	0.02692	0.08449
7	5.7	178.7-208.5	167.7-180.4	0.02760	0.03430
8	8.2*	28.1-56.2	15.7-32.7	0.03086	0.02070
9	10.6	145.9-175.0	123.1-135.1	0.03487	0.01350
10	3.4	95.9-127.9	71.6-120.2	0.04047	0.01140

Shown are the bins with nominally significant evidence for linkage (P_{SR} <0.05), for ALL studies (30cM bins, weighted analysis), ordered by their position (1 = best) according to weighted analysis P_{SR} (summed rank *P*-value). P_{OR} : ordered rank *P*-value. Bold P_{SR} values achieved an empirical threshold for suggestive evidence for linkage (P_{SR} < 0.0077). No single bin achieved an empirical genome-wide significant threshold (0.00037).

Empirically (see text), 10 or more bins with nominal P_{SR} < 0.046 was observed in 5% of simulated replicates of this dataset in the absence of linkage. Therefore, it is likely that at least some of the bins listed here contain loci linked to SCZ. Asterisks indicate which regions were consistent with our previous GSMA[4] of 20 schizophrenia scans (i.e., bins that overlap with those in the previous analysis that met aggregate significance criteria—that analysis used the Marshfield rather than the Rutgers map, so bin boundaries were not identical). See Figure 2 for further details.

UKPMC Funders Group Author Manuscript

Table 3

Bins achieving nominally significant evidence for linkage (European studies)

Pos	*Bin*	*cM*	*Mb*	P_{SR}	P_{OR}
1	8.2*	28.1-56.2	15.7-32.7	**0.00057**	0.06659
2	2.8	205.7-235.1	206.3-228.3	0.01016	0.35077
3	5.6*	148.9-178.7	141.8-167.7	0.01718	0.33717
4	16.2*	33.3-66.7	13.2-51.5	0.01775	0.15808
5	3.4	95.9-127.9	71.6-120.2	0.04359	0.62724
6	6.3*	56.0-84.0	33.9-56.6	0.04433	0.44626

Shown are bins with nominally significant evidence for linkage ($P_{SR} < 0.05$), in 22 samples or subsamples of European-ancestry families (see Table 1); the bolded p-value met empirical criteria for suggestive evidence for linkage ($P_{SR} < 0.0078$). The results here did not meet aggregate criteria for significant genome-wide evidence for linkage in this subset of the data (10 bins with nominal $P_{SR} < 0.0475$). See legend for Table 2 for additional details.

UKPMC Funders Group Author Manuscript

Table 4

Power to detect linkage in simulation studies (weighted GSMA)

	Level of significance								
	Nominal			*Suggestive*			*Genome-wide*		
Number of linked bins:	***1***	***5***	***10***	***1***	***5***	***10***	***1***	***5***	***10***
$\lambda_S = 1.3$									
ALL studies	1.000	1.000	1.000	1.000	1.000	0.998	1.000	0.996	0.975
European samples	1.000	0.998	0.993	1.000	0.992	0.953	0.950	0.912	0.745
ALL, Heterogeneity[a]	0.988	0.952	0.918	0.898	0.834	0.705	0.600	0.474	0.325
$\lambda_S = 1.15$									
ALL studies	1.000	1.000	0.989	0.996	0.980	0.932	0.904	0.850	0.705
European samples	0.970	0.948	0.908	0.874	0.822	0.697	0.598	0.468	0.323
ALL, Heterogeneity[a]	0.864	0.774	0.740	0.592	0.546	0.403	0.186	0.180	0.116

Shown is the power of weighted GSMA to detect linkage in the simulation study of samples of affected sibling pairs with information approximately comparable to the 32 studies in the schizophrenia GSMA (120 30-cM bins). λ_S = relative risk to siblings of case probands vs population risk. For 1 linked bin per simulated genome, power is a weighted average assuming that 20% of bins lie at a chromosome end ('edge,' see text), and 80% of are mid-chromosomal ('mid'). Simulations with 5 linked loci included 1 edge and 4 mid bins; those with 10 linked included 2 edge and 8 mid bins.

[a]Heterogeneity analyses modelled cases where all samples were included, but European samples were linked as shown while non-European studies were unlinked.