Brain, Behavior, and Immunity xxx (2011) xxx–xxx

Contents lists available at ScienceDirect

Brain, Behavior, and Immunity

journal homepage: www.elsevier.com/locate/ybrbi

Named Series: Epigenetics, Brain, Behavior, and Immunity

Meta-analysis of MTHFR gene variants in schizophrenia, bipolar disorder and unipolar depressive disorder: Evidence for a common genetic vulnerability?

Odette L.J. Peerbooms [a], Jim van Os [a,b], Marjan Drukker [a], Gunter Kenis [a], Loes Hoogveld [a], MTHFR in Psychiatry Group[1] Marc de Hert [c], Philippe Delespaul [a,d], Ruud van Winkel [a,c], Bart P.F. Rutten [a,*]

[a] Department of Psychiatry and Neuropsychology, School for Mental Health and Neuroscience, European Graduate School of Neuroscience (EURON), South Limburg Mental Health Research and Teaching Network (SEARCH), Maastricht University, Maastricht, The Netherlands
[b] King's College London, King's Health Partners, Department of Psychosis Studies, Institute of Psychiatry, London, United Kingdom
[c] University Psychiatric Centre, Catholic University Leuven, Campus Kortenberg, Belgium
[d] Mondriaan Zorggroep, Section Social Cognition, Heerlen, The Netherlands

ARTICLE INFO

Article history:
Received 1 September 2010
Received in revised form 23 November 2010
Accepted 10 December 2010
Available online xxxx

Keywords:
Genetics
Methylenetetrahydrofolate reductase
Schizophrenia
Bipolar disorder
Depressive disorder
Meta-analysis
Cross-disorder
DNA methylation
Epigenesis
Systematic review

ABSTRACT

Past analyses examining the relationship between genetic variation in the 5, 10-methylenetetrahydrofolate reductase (*MTHFR*) gene and psychiatric disorders have provided mixed and largely inconclusive findings. MTHFR is involved in the one-carbon metabolic pathway which is essential for DNA biosynthesis and the epigenetic process of DNA methylation. We conducted a meta-analysis of all published case-control studies investigating associations between two common *MTHFR* single nucleotide polymorphisms (SNPs), *MTHFR* C677T (sample size 29,502) and A1298C (sample size 7934), and the major psychiatric disorders (i) schizophrenia (SZ), (ii) bipolar disorder (BPD), and (iii) unipolar depressive disorder (UDD). In order to examine possible shared genetic vulnerability, we also tested for associations between *MTHFR* and all of these major psychiatric disorders (SZ, BPD and UDD) combined. *MTHFR* C677T was significantly associated with all of the combined psychiatric disorders (SZ, BPD and UDD); random effects odds ratio (OR) = 1.26 for TT versus CC genotype carriers; confidence interval (CI) 1.09–1.46); meta-regression did not suggest moderating effects of psychiatric diagnosis, sex, ethnic group or year of publication. Although *MTHFR* A1298C was not significantly associated with the combination of major psychiatric disorders, nor with SZ, there was evidence for diagnostic moderation indicating a significant association with BPD (random effects OR = 2.03 for AA versus CC genotype carriers, CI: 1.07–3.86). Meta-analysis on UDD was not possible due to the small number of studies available. This study provides evidence for shared genetic vulnerability for SZ, BPD and UDD mediated by *MTHFR* 677TT genotype, which is in line with epigenetic involvement in the pathophysiology of these psychiatric disorders.

1. Introduction

Numerous studies have tested for associations between polymorphisms in the gene encoding for methylenetetrahydrofolate reductase (*MTHFR*) and major psychiatric disorders including schizophrenia (SZ), bipolar disorder (BPD) and unipolar depressive disorder (UDD). These studies have yielded largely inconclusive and often mixed results (Betcheva et al., 2009; Feng et al., 2009; Gaysina et al., 2008; Gilbody et al., 2007; Pan et al., 2009; Yu et al., 2004; Yuan et al., 2008). MTHFR is a crucial enzyme involved in one-carbon metabolism (OCM), a folate-mediated pathway essential for purine and thymidylate biosynthesis, the methylation of DNA and amino acids, and is necessary for reactions forming neurotransmitters (Sugden, 2006). MTHFR and OCM play key roles in physiologic processes by regulating the channeling of one-carbon units between the DNA cycle (nucleotide synthesis) and the methylation cycle (Frankenburg, 2007; Krebs et al., 2009; Laanpere et al., 2010). Dysfunction of the OCM cycle has been linked to neural tube defects (van der Put et al., 2001; Zhang et al., 2008) and autism (Pasca et al., 2009), and may contribute to the pathogenesis of other disorders, including leukemia (de Jonge et al., 2009; Wiemels et al., 2001), dementia (Kim et al., 2008; Kronenberg et al., 2009), colorectal cancer (Kim, 1999; Levine et al., 2010), cardiovascular disease (Smulders and Stehouwer, 2005) and congenital abnormalities (Carmichael et al., 2009; Wani et al., 2008).

Given MTHFR's essential role in brain function and neurodevelopment (del Rio Garcia et al., 2009; Ueland et al., 2001), and that

* Corresponding author. Address: Department of Psychiatry and Psychology, School for Mental Health and Neuroscience, Maastricht University Medical Centre, P.O. Box 616 (VIJV), 6200 MD Maastricht, The Netherlands. Fax: +31 43 3688689.
E-mail address: b.rutten@np.unimaas.nl (B.P.F. Rutten).

0889-1591/$ - see front matter © 2010 Elsevier Inc. All rights reserved.
doi:10.1016/j.bbi.2010.12.006

family and twin studies have demonstrated considerable shared genetic variance between psychiatric disorders (Cardno et al., 2002; Lichtenstein et al., 2009; McGuffin et al., 2003; Van Snellenberg and de Candia, 2009), it is reasonable to hypothesize that genetic variation in *MTHFR* may contribute to the shared genetic vulnerability of common psychiatric disorders. In addition to this genetic contribution, environmental factors also contribute to the development of psychiatric disorders (Lahiri et al., 2009). It has been proposed that epigenetic mechanisms such as DNA methylation play important roles in biological mediation of environmental influences and underlie gene–environment interplay in the aetiology of mental disorders (Bredy et al., 2010; Iwamoto and Kato, 2009; Narayan and Dragunow, 2010). Thus variation in OCM may potentially moderate environmental influences affecting gene expression.

MTHFR is located on chromosome 1p36.3 (Frosst et al., 1995; van der Put et al., 1998). Despite this gene's potential importance, research focusing on the role of *MTHFR* in psychiatric disorders has focused almost exclusively on two common polymorphisms: (1) a CT transition at nucleotide 677 and (2) an AC transition at nucleotide 1298 (Frosst et al., 1995; Laanpere et al., 2010; van der Put et al., 1998). C677T is located in exon 4 and results in a transition from an alanine into a valine amino acid (Ala222Val) in the catalytic domain, with each copy of the 677T allele causing a 35% reduction of enzyme activity (Frosst et al., 1995). A1298C is located in exon 7 and changes glutamate into an alanine amino acid (Glu429Ala). This results in enhanced binding of inhibiting enzymes, with each copy of the 1298C allele decreasing enzyme activity (van der Put et al., 1998). Some earlier studies have indicated that demographic differences influence the effect of genetic variations of *MTHFR* on major psychiatric disorders. For example, Sazci et al. (2005) and Zintzaras (2006) reported that ethnicity and sex may influence the association between *MTHFR* variants and psychiatric disorders.

We designed this study to clarify if *MTHFR* C677T and/or *MTHFR* A1298C show significant associations with SZ, BPD and UDD, both as group of major psychiatric disorders or in isolation. In these analyses, we accounted for the potential modifying effects of ethnicity, year of publication and sex.

2. Material and methods

2.1. Identification and selection of studies

All non-familial, cross-sectional, and case-control studies examining the association between genetic variations of *MTHFR* and psychiatric disorders published before March 01, 2010 were included. The articles were identified by computer-based searches of the following databases: MEDLINE, EMBASE, PsycINFO, Pubmed, Web of Science, Science Direct, and the website of the schizophrenia research forum (SzGene; http://www.schizophreniaforum.org/res/sczgene/default.asp, accessed on January 31st 2010). Separate searches for each diagnostic group were conducted combining the following terms: "Methylenetetrahydrofolate reductase", "*MTHFR*", "polymorphisms", "C677T" and "A1298C", in the following combinations "Methylenetetrahydrofolate reductase" OR "*MTHFR*" AND "polymorphisms" OR "C677T" OR "A1298C". Separate searches used the following disorder-specific key words: "schizophrenia", "psychotic disorder", "psychosis", and "schizoaffective disorder" were used for SZ; "bipolar", "bipolar disorder ", "bipolar illness" and "manic depressive disorder" for BPD, and "depression", "depressive disorder", "major depressive disorder", "depressive episode", "major depressive episode", "unipolar disorder" and "affective disorder" for UDD. No language restrictions were used. Figs. 1–3 summarize the information flow through the different phases of the systematic review.

2.2. Data extraction

After removing duplicate studies, two independent researchers (O.P. and L.H.) evaluated the relevance of the identified studies (determination of the distribution of the C677T and A1298C genotypes in controls and patients) by reading the abstract or, when necessary, the full article. All references cited in the studies were screened. Abstracts for conferences, case reports, editorials and review articles were furthermore screened to identify other published and unpublished work. Figs. 1–3 summarize the identification, screening, eligibility, and inclusion of the studies that were identified using the searches on SZ (Fig. 1), BPD (Fig. 2), and UDD (Fig. 3).

Those studies considered ineligible for inclusion included those that: (i) examined associations between *MTHFR* and disorders other than the three aforementioned psychiatric disorders; (ii) reviewed past associations studies between *MTHFR* and psychiatric disorders and did not present new case-control comparison data; (iii) used linkage and family designs; and (iv) were meta-analyses of the association between *MTHFR* and psychiatric disorders.

[1] "MTHFR in Psychiatry Group" consists of *O.L.J. Peerbooms*, Department of Psychiatry and Psychology, School for Mental Health and Neuroscience, Maastricht University Medical Centre, The Netherlands; *B.P.F. Rutten*, Department of Psychiatry and Psychology, School for Mental Health and Neuroscience, Maastricht University Medical Centre, The Netherlands; *J. van Os*, Department of Psychiatry and Psychology, School for Mental Health and Neuroscience, Maastricht University Medical Centre, The Netherlands; *O.P. Almeida*, FRANZCP, FFPOA, Western Australian Centre for Health & Ageing (WACHA), Centre for Medical Research, Department of Psychiatry, Royal Perth Hospital, Winthrop Professor & Chair of Geriatric Psychiatry, School of Psychiatry and Clinical Neurosciences, Faculty of Medicine, Dentistry and Health Sciences, University of Western Australia; *S. Lewis*, School of Community and Social Medicine, University of Bristol, UK; *R. van Winkel*, University Psychiatric Centre Catholic University Leuven, Campus Kortenberg, Belgium; *M. de Hert*, University Psychiatric Centre Catholic University Leuven, Campus Kortenberg, Belgium; *A. Reif*, Department of Psychiatry, Psychosomatics and Psychotherapy, Psychiatric Neurobiology and Bipolar Disorder Program, University of Würzburg, Germany; *J.L. Roffman*, Massachusetts General Hospital, Harvard Medical School, Charlestown, USA; *E. Vilella*, Hospital Psiquiàtric Universitari Institut Pere Mata, IISPV, Universitat Rovira i Virgili Sant Llorenç, Spain; *D.I. Toncheva*, Department of Medical Genetics, Medical Faculty, Medical University Sofia, Bulgaria; *G. Arankowsky Sandoval*, Laboratorio de Neurobiología, Centro de Investigaciones Regionales, "Dr. Hideyo Noguchi", Universidad Autónoma de Yucatán, Mexico; *E.G. Jönsson*, Department of Clinical Neuroscience, Psychiatry Section, Karolinska Institutet, Karolinska University Hospital Solna, Stockholm, Sweden; *E. Choo Tan*, KK Research Centre, KK Women's and Children's Hospital, Singapore; *D. Hyun Han*, Department of Psychiatry, Chungang University Hospital School of Medicine, South Korea; *D. Gaysina*, MRC Unit for Lifelong Health and Ageing, University College London, London, UK; *H. Kunugi*, Department of Mental Disorder Research, National Institute of Neuroscience, National Center of Neurology and Psychiatry, Tokyo, Japan; *J. Yoon*, Department of Psychiatry, Chonnam National University Medical School, Republic of Korea; *A. Sazci*, University of Kocaeli, Faculty of Medicine, Department of Medical Biology and Genetics, Turkey; *Y. Nakamura*, Human Genome Center, Institute of Medical Science, The University of Tokyo, Tokyo, Japan; *A. Evans*, Centre for Public Health, The Queen's University of Belfast, Ireland; *C. Chen*, Department of Psychiatry, Kaohsiung Medical University Hospital, Kaohsiung, Taiwan; *A.S. Whitehead*, Centers for Cancer Pharmacology, Pharmacogenetics, and Excellence in Environmental Toxicology, Department of Pharmacology, University of Pennsylvania School of Medicine, Philadelphia, Pennsylvania; *R. Joober*, Prevention and Early Intervention Program for Psychoses, Douglas Mental Health University Institute and Department of Psychiatry, McGill University, Canada; *P.P. Jagodziński*, Department of Biochemistry and Molecular Biology, University of Medical Sciences, Poland; *E.T. Betcheva*, Laboratory for International Alliance, RIKEN Center for Genomic Medicine, Yokohama City, Japan and Department of Medical Genetics, Medical Faculty, Medical University Sofia, Bulgaria; *I. Bjelland*, Department of Child and Adolescent Psychiatry, Haukeland University Hospital and Department of Clinical Medicine, University of Bergen, Norway; *M. Mavros*, Department of Psychiatry, "Gr. T. Popa" University of Medicine and Pharmacy, Iasi, Romania; *J. Muntjewerff*, Department of Psychiatry, Radboud University Nijmegen Medical Centre, The Netherlands; *A. Yosivofa*, Laboratory for International Alliance, RIKEN Center for Genomic Medicine, Japan; Department of Medical Genetics, Medical University, Bulgaria; *R. Philibert*, Psychiatry Department, University of Iowa Carver College of Medicine, USA; *E. Susser*, Department of Epidemiology, Mailman School of Public Health, Columbia University, USA and New York State Psychiatric Institute; *D.C. Steffens*, Department of Psychiatry and Behavioral Sciences, Duke University Medical Center, Durham, NC, USA; *W.D. Taylor*, Department of Psychiatry and Behavioral Sciences, Duke University Medical Center, Durham, USA.

Please cite this article in press as: Peerbooms, O.L.J., et al. Meta-analysis of MTHFR gene variants in schizophrenia, bipolar disorder and unipolar depressive disorder: Evidence for a common genetic vulnerability?. Brain Behav. Immun. (2011), doi:10.1016/j.bbi.2010.12.006

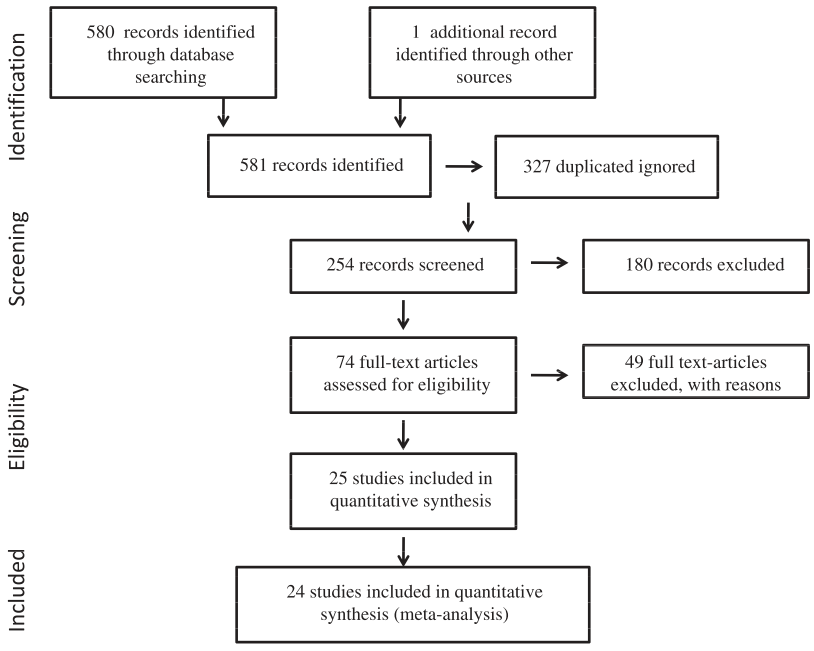

Fig. 1. Flowchart illustrating the identification, screening, eligibility and inclusion of studies on schizophrenia.

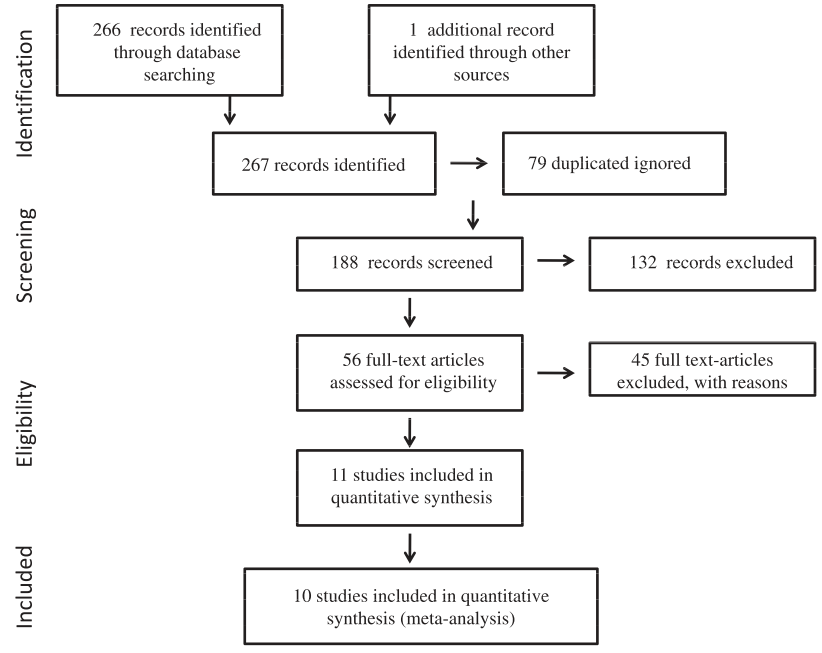

Fig. 2. Flowchart illustrating the identification, screening, eligibility and inclusion of studies on bipolar disorder.

The following information was extracted for each eligible study: authors, journal, year of publication, country of origin and ethnic group of study population, selection and characteristics of cases and controls, diagnosis and diagnostic instruments used, demographics of cases and controls, matching of controls, genotyping method, and the number of cases and controls for which C677T and/or A1298C genotyping was conducted. Genotype distributions, allele frequencies as well as genotype distributions per sex were extracted for cases and controls. When not reported in the original article, allele frequencies were calculated from the genotype distributions.

In case of overlapping samples in more than one article, we examined the most recently described sample and/or the largest sample. When different articles utilized the same control group (Arinami et al., 1997; Jonsson et al., 2008; Kempisty et al., 2007, 2006; Kim et al., 2009b; Kunugi et al., 1998; Reif et al., 2005; Tan et al., 2004), odds ratios (ORs) for these samples were calculated by comparing the genotypic distribution of the group with the relevant psychiatric disorder(s) with the control group.

The presence of a psychiatric disorder was defined as a diagnosis of this disorder using validated diagnostic instruments, standardized diagnostic interviews, or questionnaires (in some publications on UDD). To assess the impact of studies making use of questionnaires, the robustness of the analyses was tested with sensitivity analyses (please see detailed description in the section below).

Please cite this article in press as: Peerbooms, O.L.J., et al. Meta-analysis of MTHFR gene variants in schizophrenia, bipolar disorder and unipolar depressive disorder: Evidence for a common genetic vulnerability?. Brain Behav. Immun. (2011), doi:10.1016/j.bbi.2010.12.006

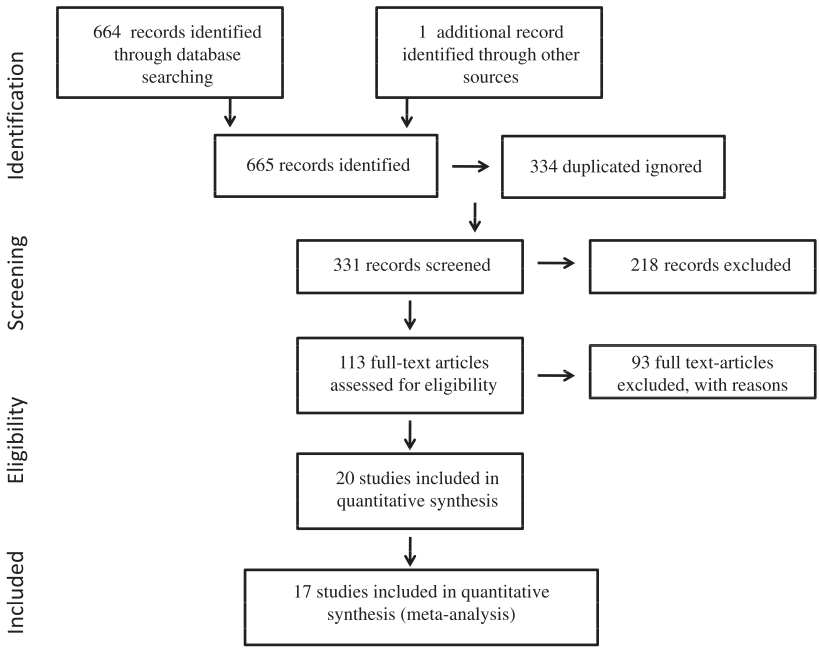

Fig. 3. Flowchart illustrating the identification, screening, eligibility and inclusion of studies on unipolar depressive disorder.

2.3. Statistical methods

Pooled ORs and 95% confidence intervals (CI) were calculated using five genetic models of analysis (Table 1): model 1 is the recessive genotype model, model 2 is the dominant genotype model, model 3 and 4 are co-dominant genotype models, and model 5 is the allele frequencies model. Heterogeneity between studies was tested using the chi-squared test (cut-off: $p < 0.05$) and quantified using the I^2 metric (which is independent of the number of studies in the meta-analysis). The pooled OR was estimated using either the fixed effects (FE) model or the random effects (RE) model depending on the level of heterogeneity.

Meta-regression was used to investigate effects of the possible modifiers of diagnosis, sex, ethnic group and year of publication. Because missing data of some of the factors would reduce the power of the analysis, the possible modifiers were tested in isolation as well as together in each of the five genetic models. The following additional tests were used for evaluation of robustness of the results. The distributions of the genotypes in the control groups were tested for Hardy–Weinberg equilibrium using the exact test. Sensitivity analyses were conducted for samples not in Hardy–Weinberg equilibrium ($p < 0.05$ in the exact test) to evaluate the impact of these studies. In a second sensitivity analysis, we excluded samples with p-values <0.1 in the exact test; this was done because the exact test for Hardy–Weinberg equilibrium may be influenced negatively by small effects and small sample sizes of the included studies (Salanti et al., 2005).

Table 1
Overview of the genetic models used in the meta-analysis for *MTHFR* C677T and A1298C.

Model	MTHFR C677T	MTHFR A1298C
Model 1 (recessive)	CC/CT versus TT genotypes	AA/AC versus CC genotypes
Model 2 (dominant)	CC versus CT/TT genotypes	AA versus AC/CC genotypes
Model 3 (co-dominant)	CC versus TT genotypes	AA versus CC genotypes
Model 4 (co-dominant)	CT versus TT genotypes	AC versus CC genotypes
Model 5 (allele-frequency)	C-allele versus T-allele	A-allele versus C-allele

Cumulative meta-analyses, i.e. repeated performances of meta-analyses whenever a new study was available, were conducted to evaluate the impact of the year of publication. Publication bias was investigated with funnel plot analyses, the Egger regression test for asymmetry, and Harbord's modification of the Egger's test (Sterne, 2009). When present, the impact of publication bias was determined with the trim and fill method.

All analyses were conducted using STATA version, 11.0, software (Stat Corp., College Station, Texas), using the commands metan, metacum. metabias, metafunnel, metatrim, and metareg.

3. Results

In total, 56 articles (25 studies on SZ, eleven on BPD, and 20 on UDD) were eligible for inclusion. Because three articles reported ORs for all three disorders (Arinami et al., 1997; Kunugi et al., 1998; Tan et al., 2004) and four articles reported ORs for two disorders (Jonsson et al., 2008; Kempisty et al., 2006; Kim et al., 2009b; Reif et al., 2005), a total of 46 articles were included. The corresponding author, first author and (if necessary) other co-authors were contacted for retrieval of information not reported in the original article. The authors of four articles/abstracts could not be reached, thus their data were not used or only in part (Hickie et al., 2005; Kim et al., 2009b; Lok et al., 2008; Perez et al., 2009). Therefore, this meta-analysis included data reported in 42 publications describing 51 samples that consisted of 29,502 subjects (9648 patients and 19,854 controls) with genotyping of *MTHFR* C677T, and 7934 subjects (3507 patients and 4427 controls) with genotyping of *MTHFR* A1298C. Table 2 summarizes, for the different studies, the name of the first author, year of publication, country, ethnic group, case/control characteristics and *MTHFR* SNP genotyped.

3.1. MTHFR C677T

The meta-analysis of the association between *MTHFR* C677T and major psychiatric disorders (SZ, BPD and UDD combined) was conducted on a total of 36 articles describing 48 samples comprising 22 SZ samples, nine BPD samples and 17 UDD samples. Table 3 provides an overview of the included samples with *MTHFR* C677T genotype distribution.

Please cite this article in press as: Peerbooms, O.L.J., et al. Meta-analysis of MTHFR gene variants in schizophrenia, bipolar disorder and unipolar depressive disorder: Evidence for a common genetic vulnerability?. Brain Behav. Immun. (2011), doi:10.1016/j.bbi.2010.12.006

Table 2
Overview of studies included in the current meta-analysis, with information about country, ethnic group, number of cases and controls and the *MTHFR* SNP examined.

First author and year (Ref. No.)	Country	Ethnic group	Cases	Controls	*MTHFR* SNP	
Schizophrenia						
Arinami, 1997	Japan	Asian	297	419	C677T	
Kunugi, 1998	Japan	Asian	343	258	C677T	
Virgos, 1999	Spain	White	210	452	C677T	
Joober, 2000	Canada	White	105	90	C677T	
Sazci, 2003	Turkey	White	130	226	C677T	A1298C
Tan, 2004	Singapore	Asian	236	120	C677T	
Yu, 2004	Scotland	White	426	628	C677T	A1298C
	China	Asian	230	251	C677T	A1298C
Muntjewerff, 2005	Netherlands	White	254	414	C677T	
Sazci, 2005	Turkey	White	297	341	C677T	A1298C
Vilella, 2005	Spain	White	234	392	C677T	A1298C
Kempisty, 2006	Poland	White	200	300	C677T	
Lee, 2006	Korea	Asian	235	235	C677T	A1298C
Philibert, 2006	USA	White	200	359	C677T	
Crisan, 2006	Romania	White	93	85	C677T	A1298C
Kempisty, 2007	Poland	White	200	300		A1298C
Jonsson, 2008	Norway	White	163	177	C677T	A1298C
	Denmark	White	419	1006	C677T	A1298C
	Sweden	White	258	293	C677T	A1298C
Roffman, 2008	USA	White	79	75	C677T	
Mavros, 2008	Romania	White	44	35	C677T	A1298C
Feng, 2009	China	Asian	123	123	C677T	
Betcheva, 2009	Bulgaria	White	185	184	C677T	A1298C
Garcia-Miss, 2010	Mexico	White	105	108	C677T	
Bipolar disorder						
Arinami, 1997	Japan	Asian	40	419	C677T	
Kunugi, 1998	Japan	Asian	143	258	C677T	
Tan, 2004	Singapore	Asian	167	120	C677T	
Reif, 2005	Germany	White	92	284	C677T	A1298C
Kempisty, 2006	Poland	White	200	300	C677T	
Kempisty, 2007	Poland	White	200	300		A1298C
Jonsson, 2008	Norway	White	117	177	C677T	A1298C
Ozbek, 2008	Turkey	White	197	238	C677T	A1298C
Chen, 2009	China	Asian	501	461	C677T	
Yosifova, 2009	Bulgarian	White	94	184		A1298C
Depressive disorder						
Arinami, 1997	Japan	Asian	32	419	C677T	
Kunugi, 1998	Japan	Asian	71	258	C677T	
Hickie, 2001	Australia	Not reported	47	21	C677T	
Bjelland, 2003	Norway	Not reported	243	8944	C677T	
Tan, 2004	Singapore	Asian	88	120	C677T	
Kelly, 2004	UK	Not reported	100	100	C677T	
Reif, 2005	Germany	White	45	284	C677T	A1298C
Almeida, 2005	Australia	White	42	198	C677T	
Chen, 2005	Taiwan	Asian	39	20	C677T	
Lewis, 2006	UK	White	545	2942	C677T	
Almeida, 2008	Australia	White	513	3239	C677T	
Gaysina, 2008	UK	White	1222	835	C677T	
Slopien, 2008	Poland	Not reported	83	89	C677T	
Yuan, 2008	China	Asian	116	80	C677T	
Kim, 2009	South Korea	Asian	101	631	C677T	
Hong, 2009	USA	White	178	85	C677T	
Hernadez-Sanchez, 2009	Spain	White	21	21	C677T	

ORs for more than one diagnosis were reported in six articles (Arinami et al., 1997; Jonsson et al., 2008; Kempisty et al., 2006; Kunugi et al., 1998; Reif et al., 2005; Tan et al., 2004). The presence of both a control group and a group with psychiatric disorder(s) was reported in 39 samples, with 34 samples reporting a case-control design and five samples using a cross-sectional design in the general population. UDD was diagnosed with questionnaires rather than with clinical interviews in these five cross-sectional general population samples (Almeida et al., 2005, 2008; Bjelland et al., 2003; Kim et al., 2009a; Lewis et al., 2006) as well as in one case-control sample (Slopien et al., 2008). Screening for psychiatric symptoms or disorders in control subjects was reported in 29 samples, but not in eight other samples (Arinami et al., 1997; Chen et al., 2009; Kempisty et al., 2006; Kunugi et al., 1998; Mavros et al., 2008; Philibert et al., 2006; Reif et al., 2005; Sazci et al., 2003); information on psychiatric screening of control subjects was not available for two samples (Yu et al., 2004). Matching of control subjects on ethnic group, geographical area, sex and/or age was performed in twelve samples (Almeida et al., 2005; Chen et al., 2009; Crisan, 2006; Feng et al., 2009; Gaysina et al., 2008; Hernandez Sanchez et al., 2009; Jonsson et al., 2008; Kelly et al., 2004; Roffman et al., 2008; Sazci et al., 2003, 2005; Tan et al., 2004). Genotyping was performed using polymerase chain reaction (PCR) based methods in all studies.

Meta-analysis. Significant between-study heterogeneity was found for all genetic models (Table 4) and for this reason random effects were used to calculate the ORs. All statistical models showed significant statistical associations between *MTHFR* C677T and major psychiatric disorder (SZ, BPD and UDD combined). The highest OR was found for TT genotype carriers

Please cite this article in press as: Peerbooms, O.L.J., et al. Meta-analysis of MTHFR gene variants in schizophrenia, bipolar disorder and unipolar depressive disorder: Evidence for a common genetic vulnerability?. Brain Behav. Immun. (2011), doi:10.1016/j.bbi.2010.12.006

Table 3
Overview of *MTHFR* C677T genotype distribution of cases and controls per diagnostic group and sample, and results of Hardy–Weinberg equilibrium testing.

First author, year	CC		CT		TT		T-allele		Hardy–Weinberg *p*-value
	Cases	Controls	Cases	Controls	Cases	Controls	Cases	Controls	
Schizophrenia									
Arinami, 1997	96	154	138	214	63	51	264	316	0.10*
Jonsson, 2008	200	490	177	413	42	103	261	619	0.27
	75	80	70	75	18	22	106	119	0.50
	137	156	104	113	17	24	138	161	0.56
Joober, 2000	30	41	52	36	23	13	98	62	0.35
Kempisty, 2006	113	210	68	79	19	11	106	101	0.30
Kunugi, 1998	121	95	168	129	54	34	276	197	0.43
Lee, 2006	74	99	128	115	33	21	194	157	0.14
Muntjewerff, 2005	112	212	111	166	31	36	173	238	0.72
Philibert, 2006	107	176	83	137	16	46	115	229	0.02*
Roffman, 2008	41	35	27	32	11	8	49	48	1.00
Sazci, 2003	59	106	49	103	22	17	93	137	0.27
Sazci, 2005	144	161	115	156	38	24	191	204	0.12
Tan, 2004	136	80	84	33	16	7	116	36	0.16
Virgos, 1999	75	88	90	115	29	39	148	193	0.89
Yu, 2004	91	85	96	126	43	40	182	206	0.60
	199	306	186	260	41	62	268	384	0.57
Feng, 2009	17	40	67	65	39	18	145	101	0.36
Betcheva, 2009	76	84	85	76	24	22	133	120	0.50
Mavros, 2008	14	23	21	7	7	5	35	17	0.01*
Crisan, 2006	52	64	29	19	12	2	53	23	0.64
Garcia-Miss, 2010	29	22	45	54	31	31	107	116	1.00
Bipolar disorder									
Chen, 2009	178	153	231	235	92	73	415	381	0.29
Arinami, 1997	15	154	20	214	5	51	30	316	0.10*
Jonsson, 2008	58	80	49	75	10	22	69	119	0.50
Kempisty, 2006	108	210	73	79	19	11	111	101	0.30
Kunugi, 1998	41	95	74	129	28	34	130	197	0.43
Tan, 2004	99	80	60	33	8	7	76	47	0.16
Reif, 2005	47	75	35	80	10	21	55	122	1.00
Yosifova, 2009	43	84	45	76	6	22	57	120	0.50
Ozbek, 2008	104	116	76	97	17	25	110	147	0.54
Depression									
Arinami, 1997	9	154	14	214	9	51	32	316	0.10*
Kunugi, 1998	30	95	31	129	10	34	51	197	0.43
Tan, 2004	49	80	34	33	5	7	44	47	0.16
Reif, 2005	24	75	15	80	8	21	31	122	1.00
Almeida, 2008	235	1423	218	1457	60	359	338	2175	0.66
Almeida, 2005	13	85	26	87	3	26	32	139	0.64
Bjelland, 2003	127	2375	85	1996	30	381	145	2758	0.18
Chen, 2005	22	11	15	9	2	0	19	9	0.53
Gaysina, 2008	545	351	513	379	64	106	641	591	0.82
Hickie, 2001	33	12	33	9	9	1	51	11	1.00
Kelly, 2004	30	40	56	37	14	12	84	61	0.48
Kim, 2009	16	84	28	248	19	126	66	500	0.06*
Slopien, 2008	26	46	38	36	19	7	76	50	1.00
Yuan, 2008	27	46	38	48	15	22	68	92	0.17
Hong, 2009	75	32	84	44	19	9	122	62	0.35
Hernadez-Sanchez, 2009	9	11	8	8	4	2	16	12	1.00
Lewis, 2006	221	1344	251	1269	73	329	397	1927	0.26

* Indicates *p*-values < 0.10 in the exact test for Hardy-Weinberg equilibrium.

compared with CC genotype carriers (genetic model 3, random effects OR_{TTvCC} = 1.26, 95 % confidence interval (CI): 1.09–1.46; I^2 52.4%). Fig. 4 shows the results from the random effects meta-analysis of the co-dominant genotype model 3 of the association between *MTHFR* C677T and major psychiatric disorder. Significant ORs were also found in the other genetic models and varied from 1.11 in genetic model 2 (random effects $OR_{CCvCT/TT}$ = 1.11, 95 % confidence interval (CI): 1.01–1.23; I^2 58.2%) to 1.24 in genetic model 1 (random effects $OR_{CC/CTvTT}$ = 1.24, 95 % confidence interval (CI): 1.10–1.39; I^2 39.5%; Table 4). Meta-regression analyses showed no significant moderation of the meta-analytic effect size by diagnosis, sex, ethnic group and year of publication (Table 5).

Sensitivity analysis. Two out of a total of 39 control samples were in Hardy–Weinberg disequilibrium (*p*-value < 0.05) (Mavros et al., 2008; Philibert et al., 2006) and another two in possible disequilibrium ($p < 0.10$) (Arinami et al., 1997; Kim et al., 2009b) (Table 3). Exclusion of these four samples from the meta-analysis did not alter the results (Table 4). Cumulative meta-analysis by year of publication showed that the association between major psychiatric disorders and *MTHFR* C677T was reduced in strength and magnitude over the years, but nevertheless remained statistically significant over the entire period (Fig. 5).

Analysis of possible publication bias, using funnel plot analyses, Egger's test and Harbord's modification of the Egger's test, indicated publication bias when using genetic models 2 and 5 (Table 4). Subsequent data correction using the trim and fill method led to a (borderline) non-significant *p*-value in genetic model 2 ($p = 0.051$; OR: 1.10; 95% CI 1.00–1.22), while the *p*-value remained significant in genetic model 5 ($p = 0.002$; OR: 1.12; 95% CI 1.04–1.21). No publication bias or small study effects were found in the other genetic models.

Please cite this article in press as: Peerbooms, O.L.J., et al. Meta-analysis of MTHFR gene variants in schizophrenia, bipolar disorder and unipolar depressive disorder: Evidence for a common genetic vulnerability?. Brain Behav. Immun. (2011), doi:10.1016/j.bbi.2010.12.006

Table 4
Odd ratio's (ORs) and confidence intervals (CI) of the five genetic models tested in the meta-analysis of MTHFR C677T and the risk of major psychiatric disorders,[a] including both fixed and random effects models, testing for heterogeneity with, publication bias with and sensitivity analyses excluding studies in Hardy–Weinberg disequilibrium.

	OR fixed	95% CI	Heterogeneity *p*-value	I^2 (%)	OR random	95% CI	Egger's test	Harbord's modification	Sensitivity analysis	
									OR random	95% CI
Model 1	1.22	1.12–1.33	$p = 0.007$	39.5	1.24	1.10–1.39	$p = 0.33$	$p = 0.31$	1.26	1.12–1.42
Model 2	1.07	1.02–1.14	$p < 0.001$	58.2	1.11	1.01–1.23	$p = 0.06$	$p = 0.07$	1.11	1.01–1.22
Model 3	1.23	1.12–1.34	$p < 0.001$	52.4	1.26	1.09–1.46	$p = 0.13$	$p = 0.14$	1.29	1.11–1.49
Model 4	1.21	1.11–1.32	$p < 0.001$	28.2	1.21	1.08–1.36	$p = 0.91$	$p = 0.80$	1.23[b]	1.13–1.35
Model 5	1.09	1.05–1.14	$p = 0.054$	60.1	1.12	1.05–1.21	$p = 0.05$	$p = 0.06$	1.13	1.05–1.21

[a] Schizophrenia, bipolar disorder and depression.
[b] Fixed effects model OR.

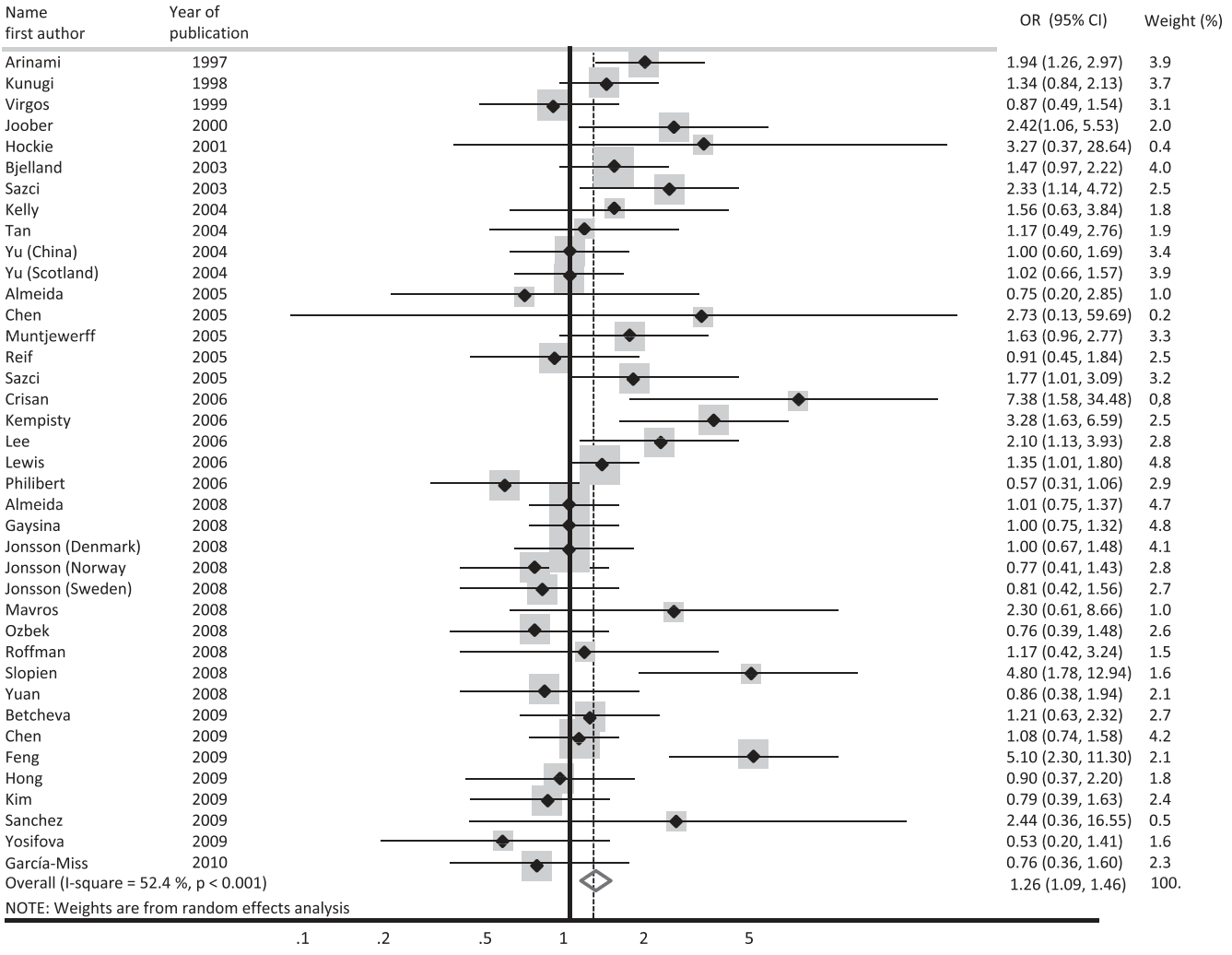

Fig. 4. Results of the random effect meta-analysis of the association between *MTHFR* C677T and the combined diagnostic group of major psychiatric disorders schizophrenia, bipolar disorder and unipolar depressive disorder, as tested in genetic model 3.

Table 5
Results of the meta-regression analyses of the MTHFR C677T meta-analysis for the five genetic models tested, including diagnosis, year of publication, ethnic group and sex as possible modifiers in separate (one modifier) and combined analyses (multiple modifiers).

	Diagnosis (coef; *p*-value)		Year of publication		Ethnic group		Sex	
	One modifier	Multiple modifiers	One modifier	Multiple modifiers	One modifier	Multiple modifiers	One modifier	Multiple modifiers
Model 1	0.020; 0.70	0.02; 0.77	− 0.029; 0.053	− 0.03; 0.29	0.04; 0.71	− 0.10; 0.35	0.10; 0.34	0.09; 0.48
Model 2	− 0.008; 0.85	0.02; 0.70	− 0.013; 0.41	− 0.03; 0.16	− 0.06; 0.56	0.03; 0.70	0.10; 0.22	0.10; 0.25
Model 3	0.00009; 1.00	0.03; 0.70	− 0.034; 0.11	−0.04; 0.15	− 0.024; 0.87	−0.09; 0.48	0.13; 0.33	0.11; 0.39
Model 4	0.032; 0.50	0.02; 0.81	− 0.026; 0.051	−0.02; 0.53	0.071; 0.47	− 0.11; 0.33	0.060; 0.56	0.04; 0.74
Model 5	− 0.001; 0.97	0.02; 0.58	− 0.013; 0.27	− 0–2; 0.13	− 0.051; 0.43	−0.01; 0.90	0.077; 0.23	0.07; 0.27

Please cite this article in press as: Peerbooms, O.L.J., et al. Meta-analysis of MTHFR gene variants in schizophrenia, bipolar disorder and unipolar depressive disorder: Evidence for a common genetic vulnerability?. Brain Behav. Immun. (2011), doi:10.1016/j.bbi.2010.12.006

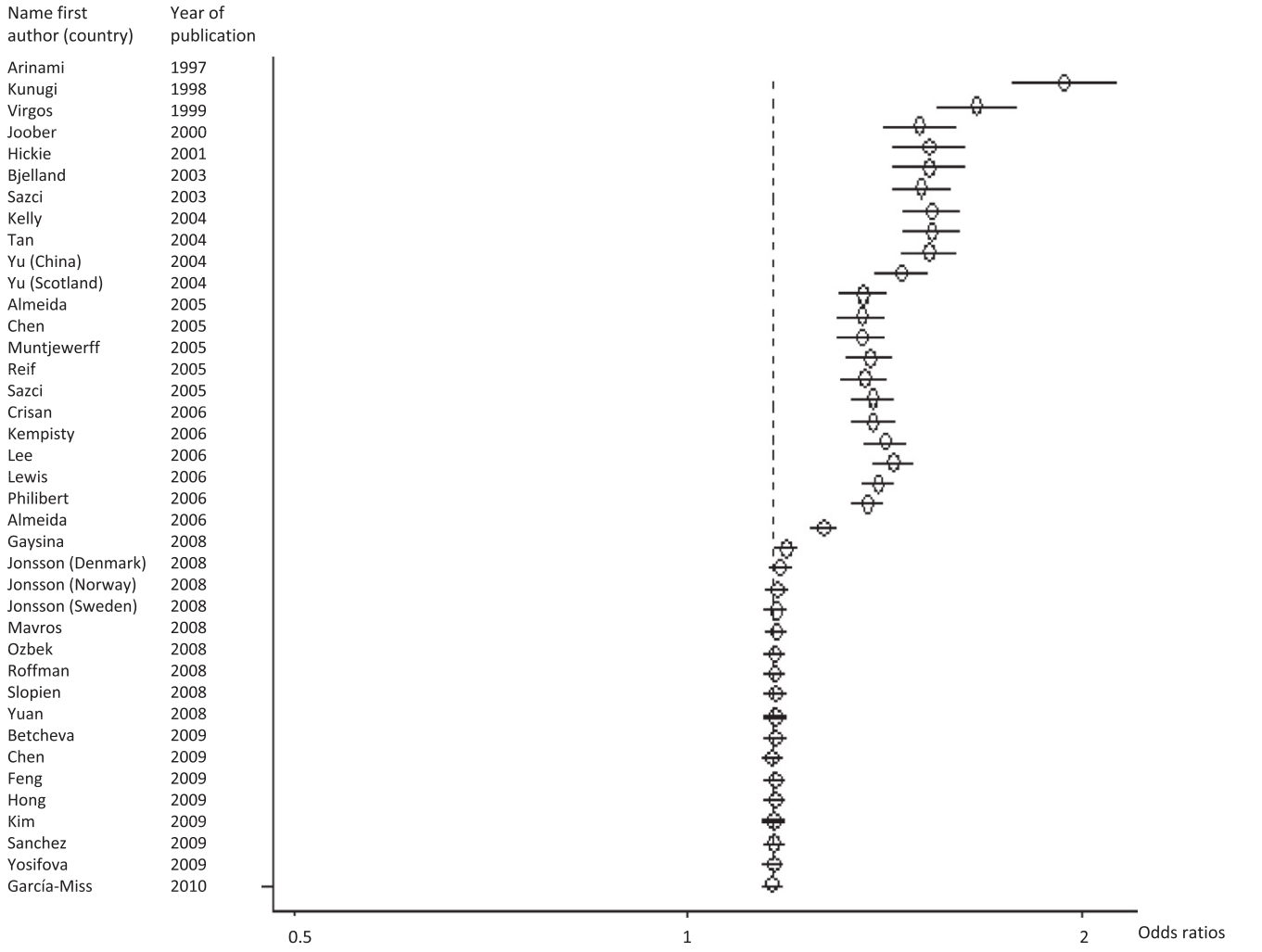

Fig. 5. Cumulative results of the meta-analysis of the association between *MTHFR* C677T and the combined diagnostic group of schizophrenia, bipolar disorder and unipolar depressive disorder, according to year of publication.

3.2. MTHFR A1298C

The meta-analysis of the associations between *MTHFR* A1298C and major psychiatric disorder (SZ, BPD and UDD combined) included 13 articles describing a total of 18 samples comprising 13 SZ samples, four BPD samples and one UDD sample (Table 6). Of these 13 articles, three articles reported results for two psychiatric disorders (Jonsson et al., 2008; Kempisty et al., 2007; Reif et al., 2005) and two articles included samples from more than one country (Jonsson et al., 2008; Yu et al., 2004). All studies used a case-control design and clinical diagnoses of psychiatric disorders were made by interviews or examination of patients' records by psychiatrists. Seven articles described screening of the control group for psychiatric disorders (Betcheva et al., 2009; Crisan, 2006; Jonsson et al., 2008; Lee et al., 2006; Ozbek et al., 2008; Sazci et al., 2005; Vilella et al., 2005) and five articles reported matching of the control group (Crisan, 2006; Jonsson et al., 2008; Lee et al., 2006; Sazci et al., 2003, 2005). All samples used a PCR-based method for genotyping the A1298C polymorphism.

Meta-analysis. Analyzing the data from the18 samples for association between *MTHFR* A1298C and major psychiatric disorders revealed significant ORs for each of the five genetic models. However, meta-regression analyses showed that diagnosis was a significant moderator and therefore further analyses were conducted stratified for diagnosis (Table 8).

3.2.1. Schizophrenia

Ten studies reported data on the association between *MTHFR* A1298C and SZ (Table 6). Because the level of heterogeneity between studies varied, both fixed effects models and random effects models were used. Diagnosis of SZ was not significantly associated with *MTHFR* A1298C (Table 7 and Fig. 6). In addition, meta-regression analyses revealed no significant influence of sex or ethnic group. Year of publication (Table 8) modified the association significantly in genetic models 2, 3 and 5; later year of publication was associated with a decreased OR.

Sensitivity analyses. Hardy–Weinberg disequilibrium was observed in one study (p-value <0.01;(Lee et al., 2006)), and seemed likely for another one (p-value = 0.07; (Jonsson et al., 2008)) (Table 6). Exclusion of the aforementioned study, as well as exclusion of both studies from the analyses did not change the results of the meta-analysis (Table 7). Funnel plot analyses, Egger's test and Harbord's modification of Egger's test did not support presence of publication bias (Table 7).

3.2.2. Bipolar disorder

Four articles reported data on the association between BPD and *MTHFR* A1298C (Jonsson et al., 2008; Kempisty et al., 2007; Ozbek et al., 2008; Reif et al., 2005) (Table 6). Because of variable study heterogeneity among the five genetic models, the random effects model was used in genetic model 3 (random effects OR_{AAvCC} = 2.03,

Please cite this article in press as: Peerbooms, O.L.J., et al. Meta-analysis of MTHFR gene variants in schizophrenia, bipolar disorder and unipolar depressive disorder: Evidence for a common genetic vulnerability?. Brain Behav. Immun. (2011), doi:10.1016/j.bbi.2010.12.006

Table 6
Overview of MTHFR A1298C genotype distribution of cases and controls per diagnostic group and sample, and results of Hardy–Weinberg equilibrium testing.

First author and year	AA		AC		CC		C allele		Hardy–Weinberg *p*-value
	Cases	Controls	Cases	Controls	Cases	Controls	Cases	Controls	
Schizophrenia									
Jonssori, 2008	89	82	60	79	14	16	88	111	0.73
	184	462	186	419	48	123	282	665	0.07
	110	122	113	129	35	42	183	213	0.45
Kempisty, 2007	109	185	74	105	17	10	108	125	0.38
Lee, 2006	157	145	7	14	71	77	149	168	0.00*
Sazci, 2003	57	114	59	93	14	19	73	131	1.00
Sazci, 2005	130	159	129	155	38	27	205	209	0.25
Vilella, 2005	98	128	67	100	18	13	103	126	0.32
Yu, 2004	199	292	186	272	41	64	268	400	1.00
	130	154	78	81	22	16	122	113	0.28
Betcheva, 2009	91	80	72	79	18	24	108	127	0.52
Mavros, 2008	26	17	13	20	2	3	17	26	0.49
Crisan, 2006	54	73	39	12	0	0	39	12	1.00
Bipolar disorder									
Jonsson, 2008	47	82	56	79	12	16	80	111	0.73
Kempisty, 2007	99	185	78	105	23	10	124	125	0.38
Reif, 2005	18	75	22	96	8	13	38	122	0.02*
Ozbek, 2008	91	113	84	101	22	24	128	149	0.88
Depression									
Reif, 2005	18	75	22	96	8	13	38	122	0.02*

* Indicates *p*-values < 0.10 in the exact test for Hardy-Weinberg equilibrium.

95 % CI: 1.07–3.86; I^2 64.0%), while fixed effects models were used in genetic models 1, 2, 4 and 5 (Table 7 and Fig. 7). Diagnosis of BPD was significantly associated with *MTHFR* A1298C in all five genetic models (Table 7). Meta-regression analyses revealed no significant moderation by year of publication or sex. Ethnic group could not be analyzed as a possible modifier in these analyses because all studies were conducted in White samples (Table 8).

Sensitivity analyses. Hardy–Weinberg disequilibrium was observed in one study ($p = 0.02$; (Reif et al., 2005)), (Table 6) and sensitivity analyses indicated that exclusion of this study (Reif et al., 2005) led to a loss of statistical association with mental disorders in genetic models 1, 3 and 4 but not in genetic models 2 and 5 (Table 7). Analysis of funnel plots, Egger's test and Harbord's modification to Egger's test indicated no evidence for publication bias.

3.2.3. Depression

The study conducted by Reif et al. (2005) was the only study that examined the association between *MTHFR* A1298C and UDD (Reif et al., 2005) (Table 6). Their data indicated that the diagnosis of UDD was significantly associated with *MTHFR* A1298C in genetic model 1 (fixed-effect $OR_{AA/ACvCC} = 2.63$, 95 % CI: 1.02–6.77; I^2 0%) but not in the other genetic models (Table 7).

4. Discussion

4.1. Main findings

The present meta-analysis examining *MTHFR* C677T in 9648 patient and 19,854 control subjects indicated that carriers of the

Table 7
Odd ratio's (ORs) and confidence intervals (CI) of the 5 genetic models tested in the meta-analysis of MTHFR A1298C and the risk of major psychiatric disorders,[a] including both fixed and random effects models, testing for heterogeneity with the I^2 test, publication bias with Egger's test and Harbord's modification of the Egger test and sensitivity analyses excluding studies in Hardy–Weinberg disequilibrium. Analyses were conducted separately as diagnosis was found to be a modifier.

	OR		Heterogeneity		OR		Egger's test	Harbord's modification	Sensitivity analysis	
	Fixed	95% CI	*p*-Value	I^2 (%)	Random	95% CI			OR	95% CI
Schizophrenia										
Model 1	1.08	0.93–1.26	$p = 0.140$	31.4	1.12	0.92–1.37	$p = 0.21$	$p = 0.22$	1.12	0.95–1.33
Model 2	1.08	0.98–1.19	$p = 0.002$	61.4	1.07	0.90–1.26	$p = 0.90$	$p = 0.84$	1.10[a]	0.92–1.30
Model 3	1.10	0.93–1.29	$p = 0.090$	37.6	1.13	0.91–1.41	$p = 0.43$	$p = 0.43$	1.19	0.93–1.51
Model 4	1.12	0.94–1.33	$p = 0.225$	22.2	1.16	0.94–1.43	$p = 0.06$	$p = 0.07$	1.10	0.92–1.31
Model5	1.06	0.99–1.15	$p = 0.002$	61.1	1.07	0.94–1.21	$p = 0.70$	$p = 0.72$	1.09[a]	0.96–1.25
Bipolar disorder										
Model 1	1.78	1.25–2.54	$p = 0.051$	61.4	1.85	1.02–3.34	$p = 0.41$	$p = 0.57$	1.68[a]	0.78–3.59
Model 2	1.33	1.08–1.64	$p = 0.409$	0.0	1.33	1.08–1.64	$p = 0.94$	$p = 0.92$	1.31	1.04–1.64
Model 3	1.95	1.34–2.82	$p = 0.040$	64.0	2.03	1.07–3.86	$p = 0.44$	$p = 0.61$	1.83[a]	0.80–4.18
Model 4	1.64	1.13–2.38	$p = 0.126$	47.6	1.67	0.98–2.84	$p = 0.47$	$p = 0.65$	1.49	0.98–2.27
Model 5	1.32	1.13–1.55	$p = 0.107$	50.8	1.32	1.05–1.66	$p = 0.99$	$p = 0.97$	1.29	1.08–1.54
Depression										
Model 1	2.63	1.02–6.77								
Model 2	1.15	0.60–2.21								
Model 3	2.56	0.93–7.11								
Model 4	2.69	0.99–7.26								
Model 5	1.32	0.83–2.10								

[a] OR random effects model.

Please cite this article in press as: Peerbooms, O.L.J., et al. Meta-analysis of MTHFR gene variants in schizophrenia, bipolar disorder and unipolar depressive disorder: Evidence for a common genetic vulnerability?. Brain Behav. Immun. (2011), doi:10.1016/j.bbi.2010.12.006

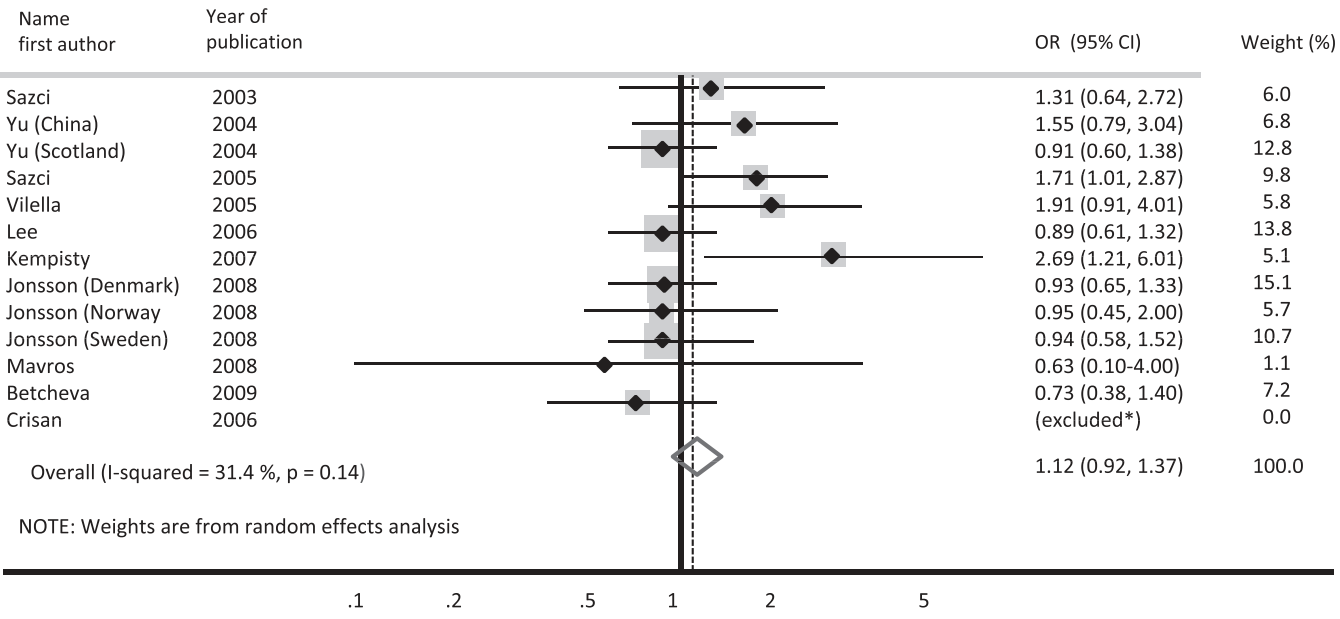

Fig. 6. Results of the random effect meta-analysis of MTHFR A1298C and schizophrenia as tested in genetic model 1.

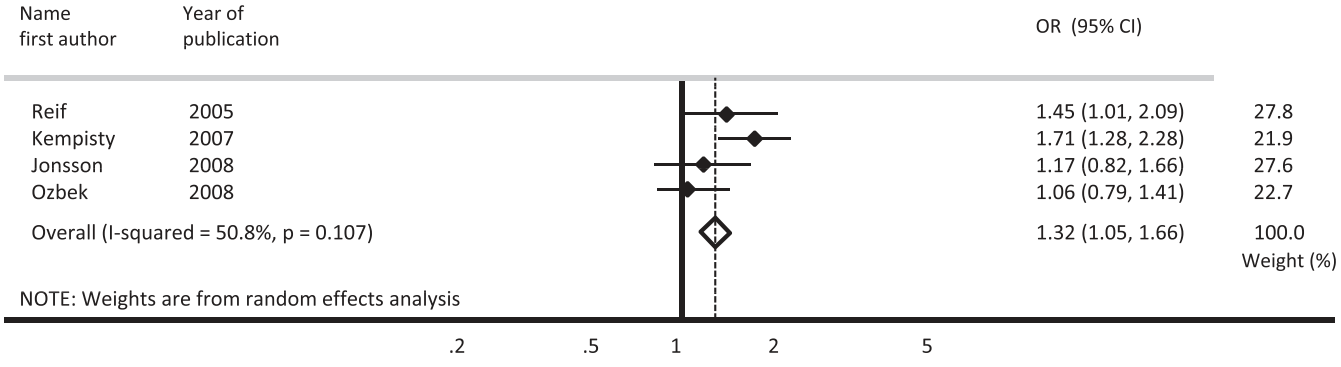

Fig. 7. Results of the random effect meta-analysis of MTHFR A1298C and bipolar disorder as tested in genetic model 5.

Table 8
Results of the meta-regression analyses of the MTHFR A1298C meta-analysis for the 5 genetic models tested, including diagnosis, year of publication, ethnic group and sex as possible modifiers in separate (one modifier) and combined analyses (multiple modifiers).

	Year of publication (coef; *p*-valu e)		Ethnic group (coef; *p*-value)		Sex (coef; *p*-value)	
	One modifier	Multiple modifier	One modifier	Multiple modifiers	One modifier	Multiple modifiers
Schizophrenia						
Model 1	−0.08; 0.14	−0.22; 0.07	−0.05; 0.86	−0.49; 0.07	−0.10; 0.62	−0.07; 0.71
Model 2	−0.07; 0.05	−0.06; 0.24	−0.35; 0.07	−0.34; 0.14	−0.05; 0.69	0.07; 0.59
Model 3	−0.10; 0.06	−0.23; 0.013	−0.08; 0.77	−0.57; 0.04	−0.08; 0.72	−0.05; 0.82
Model 4	−0.05; 0.39	−0.20; 0.029	0.37; 0.27	0.38; 0.45	−0.11; 0.67	−0.09; 0.70
Model 5	−0.06; 0.03	−0.08; 0.052	−0.24; 0.13	−0.32; 0.07	0.01; 0.90	0.03; 0.80
Bipolar disorder						
Model 1	−0.28; 0.23	−0.27; 0.21			−0.08; 0.86	−0.13; 0.78
Model 2	−0.09; 0.40	−0.09; 0.46			0.16; 0.48	0.16; 0.51
Model 3	−0.30; 0.23	−0.30; 0.20			0.07; 0.90	0.02; 0.98
Model 4	−0.27; 0.16	−0.26; 0.20			−0.19; 0.65	−0.22; 0.62
Model 5	−0.10; 0.33	−0.09; 0.38			0.10; 0.63	0.09; 0.66

T allele and TT genotype are at a small but statistically significant increased risk of receiving the diagnosis of major psychiatric disorders (SZ, BPD and UDD). No moderating effects were observed for specific diagnosis, sex or ethnic group. The robustness of these findings is underscored by the lack of publication bias. The present meta-analysis on *MTHFR* A1298C in 3507 patient and 4427 control subjects indicated that carriers of the C allele and CC genotype are small but statistically significant increased risk of being diagnosed with BPD, but not SZ.

4.2. Strengths of the meta-analysis

The current report represents the largest published meta-analysis on genetic variations of *MTHFR* in psychiatric disorders (SZ,

Please cite this article in press as: Peerbooms, O.L.J., et al. Meta-analysis of MTHFR gene variants in schizophrenia, bipolar disorder and unipolar depressive disorder: Evidence for a common genetic vulnerability?. Brain Behav. Immun. (2011), doi:10.1016/j.bbi.2010.12.006

BPD and UDD) to date. This analysis examined two SNP's associated with three psychiatric disorders, examining both combined major psychiatric disorders as well as individual diagnoses when there was evidence for diagnostic moderation. This is the first meta-analysis examining the association between *MTHFR* and multiple psychiatric disorders using a cross-disorder design – most meta-analyses available to date have examined the association between *MTHFR* C677T and diagnoses of a single psychiatric disorder (Allen et al., 2008; Chen et al., 2009; Cohen-Woods et al., 2010; Gaysina et al., 2008; Shi et al., 2008; Yoshimi et al., 2010). Meta-analyses conducted by Lewis et al. (6 articles, 2427 subjects) (2005) and Muntjewerff (10 articles, 4986 subjects) (2006) tested for an association between *MTHFR* C677T and schizophrenia; both reported an increased risk of SZ with the TT genotype, respectively $OR_{TTvCC/CT}$ = 1.48, 95% CI: 1.18–1.86; OR_{TTvCC} = 1.36, 95% CI: 1.07–1.72 (Lewis et al., 2005; Muntjewerff et al., 2006).

Meta-analyses conducted by Gilbody et al. (2007) and Zintzaras (2006) on *MTHFR* C677T did include several psychiatric disorders. The meta-analysis by Gilbody et al. (2007) included twelve articles on SZ (6125 subjects), four on BPD (1648 subjects), and ten on UDD (11,709 subjects) (Gilbody et al., 2007). Zintzaras (2006) conducted his meta-analysis at the same time as Gilbody et al. (2007) and included the same articles but differed by including two additional articles (Kempisty et al., 2006; Reif et al., 2005). Jonsson et al. (2008) conducted a meta-analysis in 2008 examining the association between SZ and both *MTHFR* C677T and A1298C in a total sample of, respectively 9548 subjects and 6118 subjects (Jonsson et al., 2008). In addition, the association between BPD and *MTHFR* C677T and A1298C was examined in a total sample of, respectively 2211 subjects and 792 subjects. Compared with our current report, most of these meta-analyses included fewer studies, a smaller total sample, and *MTHFR* A1298C was not always consistently examined.

Thus, by examining both the two *MTHFR* SNP's C677T and A1298C in all five genetic models while including sex, year of publication and ethnic background in the analysis, this meta-analysis provides a good overview of the association between *MTHFR* gene variants and the psychiatric disorders SZ, BPD and UDD.

4.3. Limitations of the meta-analysis

First, although the current study analyzed the largest combined sample so far, the meta-analysis on *MTHFR* A1298C was performed on a 'relatively' low number of subjects, especially given the minor allele frequency (MAF) of 36% in combination with the small effect sizes reported in the literature. The limited number of studies investigating the *MTHFR* A1298C polymorphism suggests that type II error cannot be dismissed (Dudbridge and Gusnanto, 2008; O'Donovan et al., 2009). Second, the use of different designs in the studies included in the meta-analysis may influence the association found between *MTHFR* and psychiatric disorders. However, all significant gene-disorder associations remained after sensitivity analyses for differences in study design. Third, matching of control subjects was not performed in all studies used in this meta-analysis. However, sensitivity testing indicated that it is unlikely that non-matching of control subjects had substantial influences. Fourth, it is possible that non-uniformity in diagnostic measures may have resulted in heterogeneity between studies and we must acknowledge that the diagnosis of depression in some studies was not based on accepted diagnostic criteria but cut-points in scales. This can create two potential problems: uncertainty about (i) the validity of the diagnosis of depression and (ii) misclassification (for example, if a patient with bipolar disorder was experiencing a depressive episode when s/he completed the rating of the scale). In the first instance, this type of error would lead to a decrease in effect size; in the second it would lead to misclassification bias. Given that the analyses combined the diagnoses of SZ, BPD and UDD, misclassification is not a problem. We may, however, have underestimated the effect of the association. Fifth, the generalizability of the present meta-analysis is limited to the ethnic groups investigated, i.e. White and Asian.

4.4. Comparison with other meta-analyses on specific diagnoses

4.4.1. Schizophrenia

In line with the current results, the meta-analysis of Yoshimi et al. (2010) supported an association between *MTHFR* C677T with schizophrenia (Yoshimi et al., 2010), similar to earlier meta-analyses (Yoshimi: random effects OR = 1.17, 95% CI: 1.07–1.29) (Allen et al., 2008; Gilbody et al., 2007; Jonsson et al., 2008; Lewis et al., 2005; Muntjewerff et al., 2006; Shi et al., 2008; Zintzaras, 2006).

Regarding *MTHFR* A1298C, Zintzaras (2006) concluded in his meta-analysis on 2.565 subjects that this SNP was associated with the diagnosis of schizophrenia, however not in all genetic models examined (fixed effects OR_{CvA} = 1.16, 95% CI: 1.03–1.31; $OR_{AC/CCvAA}$ = 1.19, 95% CI: 1.02–1.40; OR_{CCvAA} = 1.37, 95% CI 1.03–1.82; random effects $OR_{CCvAC/AA}$ = 1.33, 95% CI: 0.94–1.88) (Zintzaras, 2006). The meta-analysis of Gilbody et al. (2007) on 994 subjects reported only an association in the co-dominant model (fixed effects OR_{CCvAA} = 1.64, 95% CI: 1.05–2.54; genetic model 3), but not in the other model tested (fixed effects OR_{ACvAA} = 1.10, 95% CI 0.84–1.43) (Gilbody et al., 2007). Jonsson et al. (2008) reported non-significant or only borderline significant results in favor of an association between *MTHFR* A1298C and SZ in a sample of 6118 subjects (fixed effects OR_{CvA} = 1.09, 95% CI: 1.01–1.18; $OR_{AAvAC/CC}$ = 1.10, 95% CI: 0.99–1.23; OR_{CCvAA} = 1.20, 95% CI 0.99–1.44; $OR_{CCvAC/AA}$ = 1.15, 95% CI: 0.96–1.37; OR_{ACvAA} = 1.08, 95% CI 0.97–1.21) (Jonsson et al., 2008). Thus, the current sample size in the present meta-analysis (7466 subjects) exceeds earlier studies and indicates that *MTHFR* A1298C is not significantly associated with SZ in any of the five genetic models.

4.4.2. Bipolar disorder

MTHFR C677T was neither significantly associated with BPD in recent meta-analyses by Cohen-Woods et al. (2010) using a total sample of 2584 subjects nor by Zintzaras (2006) in a sample of 1415 subjects. The meta-analysis by Gilbody et al. (2007) on a total sample of 1648 subjects did find a statistically significant association, which is concordant with the present findings (fixed effects OR_{TTvCC} 1.82, 95% CI: 1.22–2.70; OR_{TvC} = 1.41, 95% CI: 1.19–1.68).

Jonsson et al. (2008) reported in their meta-analysis on 2211 subjects, almost significant or borderline significant association between *MTHFR* C677T (fixed effects OR_{TvC} 1.20, 95% CI: 1.04–1.39; $OR_{TTvCT/CC}$ 1.29, 95% CI: 0.94–1.76; OR_{TTvCC} 1.35, 95% CI: 0.97–1.88; OR_{CTvCC} 1.21, 95% CI: 0.99–1.48; $OR_{TT/CTvCC}$ 1.25, 95% CI: 1.03–1.51).

4.4.3. Unipolar depressive disorder

Gilbody et al. (2007) (total sample 11,709 subjects: 1280 cases of UDD; 10,429 controls) found evidence of an association between *MTHFR* C677T and UDD (fixed effects OR_{TTvCC} = 1.36, 95% CI: 1.11–1.67; OR_{TvC} = 1.14, 95% CI: 1.04–1.26; OR_{CTvCC} = 1.10, 95% CI: 0.96–1.25). This was however not supported by the meta-analyses of Zintzaras (2006) with a total sample of 1604 subjects and the meta-analysis of Gaysina et al. (2008) with a total sample of 2566 subjects.

4.5. Common genetic vulnerability

As mentioned previously, shared genetic vulnerability for psychiatric disorders has been established by family and twin studies

Please cite this article in press as: Peerbooms, O.L.J., et al. Meta-analysis of MTHFR gene variants in schizophrenia, bipolar disorder and unipolar depressive disorder: Evidence for a common genetic vulnerability?. Brain Behav. Immun. (2011), doi:10.1016/j.bbi.2010.12.006

(Cardno et al., 2002; Huang et al., 2010). A recent report by the International Schizophrenia Consortium furthermore indicated that common genetic variations may account for one third of the total genetic liability of SZ and that this polygenetic risk is shared with BPD (Purcell et al., 2009). Similarly, a recent GWAS study using a cross-disorder design reported that common genetic variations in the *NPAS3* gene are shared between SZ and BPD (Huang et al., 2010). Similar cross-disorder designs can be applied to recently published GWAS datasets and may be used to replicate the current findings (Baum et al., 2008; Huang et al., 2010; Moskvina et al., 2009; Stefansson et al., 2009).

4.6. Biological mechanisms underlying common genetic vulnerability in MTHFR

MTHFR is a crucial enzyme involved in one-carbon metabolism (OCM), which is a folate- mediated pathway that is divided into a methylation cycle and a DNA synthesis cycle (Sugden, 2006). MTHFR catalyses the conversion of 5,10-methylenetetrahydrofolate (5,10-MTHF) to 5-MTHF, the predominant circulating form of folate (Frankenburg, 2007; Krebs et al., 2009; Sugden, 2006). 5,10-MTHF is involved in DNA synthesis as an essential donor molecule for purines synthesis and a substrate molecule for thymidine synthase, which is a rate limiting step in DNA biosynthesis (Frankenburg, 2007; Krebs et al., 2009; Sugden, 2006). In the methylation cycle, the methyl group of 5-MTHF is furthermore used for the re-methylation of homocysteine to methionine, and the conversion of methionine to S-adenosylmethionine (SAM), which is a major methyl donor to DNA, proteins, neurotransmitters, hormones and phospholipids (Frankenburg, 2007; Krebs et al., 2009; Sugden, 2006).

Although the exact role of OCM in neurodevelopment has not been well characterized, evidence from both experimental animal and human studies has shown that components of OCM (del Rio Garcia et al., 2009; Dror and Allen, 2008; Zeisel, 2009) influences brain development, brain maturation and function. Thus, given the crucial role of *MTHFR* in OCM, the biological mechanism underlying the common genetic vulnerability of *MTHFR* may result in (i) aberrant methylation (Zeisel, 2009), (ii) aberrant DNA synthesis (Greenblatt et al., 1994), and (iii) increased turnover of neurotransmitters (Bottiglieri et al., 2000; Greenblatt et al., 1994). The absence of clear evidence for major genetic effects, despite (high) estimates of heritability for major psychiatric disorders, together with evidence of 'causal' environmental exposures resulting in changes in gene expression is consistent with the concept that the biologic underpinnings of psychiatric disorders are epigenetic in form rather than DNA sequence-based (Dror and Allen, 2008; Zeisel, 2009). DNA methylation is a critical epigenetic modification of the genome that controls many biologic processes, including embryonic development, X-chromosome inactivation, imprinting, and gene expression (Bredy et al., 2010; Stahl, 2010). Although methylation patterns are established during early life, they are not fixed, and gradual hypomethylation of the genome is reported to occur with age, together with aberrant hypermethylation of gene promoter regions (Lahiri et al., 2009). Thus, the correct establishment of DNA methylation patterns is important not only during early life but also for long-term health benefits, including psychiatric and neurologic disease susceptibility (Lahiri et al., 2009).

To conclude, the results of this meta-analysis are consistent with the hypothesis that SZ, BPD and UDD share a common genetic vulnerability linked to the common MTHFR C677T polymorphism. Given the link between genetic variations of *MTHFR* and differential methylation potentials, these results support the epigenetic hypothesis of major psychiatric disorders.

Acknowledgments

The research leading to these results has received funding from the European Community's Seventh Framework Programme under Grant Agreement No. HEALTH-F2-2009-241909 (Project EU-GEI) and from the Netherlands Organisation for Scientific Research (NWO) for Bart P.F. Rutten (VENI Award No. 916.11.086).

References

Allen, N.C., Bagade, S., McQueen, M.B., Ioannidis, J.P., Kavvoura, F.K., Khoury, M.J., Tanzi, R.E., Bertram, L., 2008. Systematic meta-analyses and field synopsis of genetic association studies in schizophrenia: the SzGene database. Nat. Genet. 40, 827–834.

Almeida, O.P., Flicker, L., Lautenschlager, N.T., Leedman, P., Vasikaran, S., van Bockxmeer, F.M., 2005. Contribution of the MTHFR gene to the causal pathway for depression, anxiety and cognitive impairment in later life. Neurobiol. Aging 26, 251–257.

Almeida, O.P., McCaul, K., Hankey, G.J., Norman, P., Jamrozik, K., Flicker, L., 2008. Homocysteine and depression in later life. Arch. Gen. Psychiatry 65, 1286–1294.

Arinami, T., Yamada, N., Yamakawa-Kobayashi, K., Hamaguchi, H., Toru, M., 1997. Methylenetetrahydrofolate reductase variant and schizophrenia/depression. Am. J. Med. Genet. 74, 526–528.

Baum, A.E., Akula, N., Cabanero, M., Cardona, I., Corona, W., Klemens, B., Schulze, T.G., Cichon, S., Rietschel, M., Nothen, M.M., Georgi, A., Schumacher, J., Schwarz, M., Abou Jamra, R., Hofels, S., Propping, P., Satagopan, J., Detera-Wadleigh, S.D., Hardy, J., McMahon, F.J., 2008. A genome-wide association study implicates diacylglycerol kinase eta (DGKH) and several other genes in the etiology of bipolar disorder. Mol. Psychiatry 13, 197–207.

Betcheva, E.T., Mushiroda, T., Takahashi, A., Kubo, M., Karachanak, S.K., Zaharieva, I.T., Vazharova, R.V., Dimova, I.I., Milanova, V.K., Tolev, T., Kirov, G., Owen, M.J., O'Donovan, M.C., Kamatani, N., Nakamura, Y., Toncheva, D.I., 2009. Case-control association study of 59 candidate genes reveals the DRD2 SNP rs6277 (C957T) as the only susceptibility factor for schizophrenia in the Bulgarian population. J. Hum. Genet. 54, 98–107.

Bjelland, I., Tell, G.S., Vollset, S.E., Refsum, H., Ueland, P.M., 2003. Folate, vitamin B12, homocysteine, and the MTHFR 677C->T polymorphism in anxiety and depression: the Hordaland Homocysteine Study. Arch. Gen. Psychiatry 60, 618–626.

Bottiglieri, T., Laundy, M., Crellin, R., Toone, B.K., Carney, M.W., Reynolds, E.H., 2000. Homocysteine, folate, methylation, and monoamine metabolism in depression. J. Neurol. Neurosurg. Psychiatry 69, 228–232.

Bredy, T.W., Sun, Y.E., Kobor, M.S., 2010. How the epigenome contributes to the development of psychiatric disorders. Dev. Psychobiol. 52, 331–342.

Cardno, A.G., Rijsdijk, F.V., Sham, P.C., Murray, R.M., McGuffin, P., 2002. A twin study of genetic relationships between psychotic symptoms. Am. J. Psychiatry 159, 539–545.

Carmichael, S.L., Yang, W., Correa, A., Olney, R.S., Shaw, G.M., 2009. Hypospadias and intake of nutrients related to one-carbon metabolism. J. Urol. 181, 315–321. discussion 321.

Chen, Z., Liu, Y., Zhang, D., Liu, Z., Wang, P., Zhou, D., Zhao, T., Wang, T., Xu, H., Li, S., Feng, G., He, L., Yu, L., 2009. C677T methylenetetrahydrofolate reductase gene polymorphisms in bipolar disorder: an association study in the Chinese population and a meta-analysis of genetic association studies. Neurosci. Lett. 449, 48–51.

Cohen-Woods, S., Craig, I., Gaysina, D., Gray, J., Gunasinghe, C., Craddock, N., Elkin, A., Jones, L., Kennedy, J., King, N., Korszun, A., Knight, J., Owen, M., Parikh, S., Strauss, J., Sterne, A., Tozzi, F., Perry, J., Muglia, P., Vincent, J., McGuffin, P., Farmer, A., 2010. The Bipolar Association Case-Control Study (BACCS) and meta-analysis: no association with the 5, 10-methylenetetrahydrofolate reductase gene and bipolar disorder. Am. J. Med. Genet. B Neuropsychiatr. Genet. 153 (7), 1298–1304.

Crisan, C.C., 2006. P.1.a.009 Methylenetetrahydrofolate reductase gene polymorphisms (C677T and A1298C) in patients with schizophrenia. Eur. Neuropsychopharmacol. 16, S213.

de Jonge, R., Tissing, W.J., Hooijberg, J.H., Jansen, G., Kaspers, G.J., Lindemans, J., Peters, G.J., Pieters, R., 2009. Polymorphisms in folate-related genes and risk of pediatric acute lymphoblastic leukemia. Blood 113, 2284–2289.

del Rio Garcia, C., Torres-Sanchez, L., Chen, J., Schnaas, L., Hernandez, C., Osorio, E., Portillo, M.G., Lopez-Carrillo, L., 2009. Maternal MTHFR 677C>T genotype and dietary intake of folate and vitamin B(12): their impact on child neurodevelopment. Nutr. Neurosci. 12, 13–20.

Dror, D.K., Allen, L.H., 2008. Effect of vitamin B12 deficiency on neurodevelopment in infants: current knowledge and possible mechanisms. Nutr. Rev. 66, 250–255.

Dudbridge, F., Gusnanto, A., 2008. Estimation of significance thresholds for genomewide association scans. Genet. Epidemiol. 32, 227–234.

Feng, L.G., Song, Z.W., Xin, F., Hu, J., 2009. Association of plasma homocysteine and methylenetetrahydrofolate reductase C677T gene variant with schizophrenia: A Chinese Han population-based case-control study. Psychiatry Res. 168, 205–208.

Frankenburg, F.R., 2007. The role of one-carbon metabolism in schizophrenia and depression. Harv. Rev. Psychiatry 15, 146–160.

Frosst, P., Blom, H.J., Milos, R., Goyette, P., Sheppard, C.A., Matthews, R.G., Boers, G.J., den Heijer, M., Kluijtmans, L.A., van den Heuvel, L.P., et al., 1995. A candidate

genetic risk factor for vascular disease: a common mutation in methylenetetrahydrofolate reductase. Nat. Genet. 10, 111–113.
Gaysina, D., Cohen, S., Craddock, N., Farmer, A., Hoda, F., Korszun, A., Owen, M.J., Craig, I.W., McGuffin, P., 2008. No association with the 5,10-methylenetetrahydrofolate reductase gene and major depressive disorder: results of the depression case control (DeCC) study and a meta-analysis. Am. J. Med. Genet. B Neuropsychiatr. Genet. 147B, 699–706.
Gilbody, S., Lewis, S., Lightfoot, T., 2007. Methylenetetrahydrofolate reductase (MTHFR) genetic polymorphisms and psychiatric disorders: a HuGE review. Am. J. Epidemiol. 165, 1–13.
Greenblatt, J.M., Huffman, L.C., Reiss, A.L., 1994. Folic acid in neurodevelopment and child psychiatry. Prog. Neuropsychopharmacol. Biol. Psychiatry 18, 647–660.
Hernandez Sanchez, M.D.C., Gomez-Reino Rodriguez, I., Cuello Hormigo, L., 2009. Research on C677T polymorphism of the metiltetrahidrofolareductasa (MTHFR) in hospitalized patients due to greater depression in Unit of Psychiatry of Orense. Anales de psiquiatria 25 (2), 53–63 (Spanish).
Hickie, I., Naismith, S., Ward, P.B., Scott, E., Mitchell, P., Wilhelm, K., Parker, G., 2005. Vascular risk and low serum B12 predict white matter lesions in patients with major depression. J. Affect. Disord. 85, 327–332.
Huang, J., Perlis, R.H., Lee, P.H., Rush, A.J., Fava, M., Sachs, G.S., Lieberman, J., Hamilton, S.P., Sullivan, P., Sklar, P., Purcell, S., Smoller, J.W., 2010. Cross-disorder genomewide analysis of schizophrenia, bipolar disorder, and depression. Am. J. Psychiatry. 167, 1254–1263.
Iwamoto, K., Kato, T., 2009. Epigenetic profiling in schizophrenia and major mental disorders. Neuropsychobiology 60, 5–11.
Jonsson, E.G., Larsson, K., Vares, M., Hansen, T., Wang, A.G., Djurovic, S., Ronningen, K.S., Andreassen, O.A., Agartz, I., Werge, T., Terenius, L., Hall, H., 2008. Two methylenetetrahydrofolate reductase gene (MTHFR) polymorphisms, schizophrenia and bipolar disorder: an association study. Am. J. Med. Genet. B Neuropsychiatr. Genet. 147B, 976–982.
Kelly, C.B., McDonnell, A.P., Johnston, T.G., Mulholland, C., Cooper, S.J., McMaster, D., Evans, A., Whitehead, A.S., 2004. The MTHFR C677T polymorphism is associated with depressive episodes in patients from Northern Ireland. J. Psychopharmacol. 18, 567–571.
Kempisty, B., Bober, A., Luczak, M., Czerski, P., Szczepankiewicz, A., Hauser, J., Jagodzinski, P.P., 2007. Distribution of 1298A>C polymorphism of methylenetetrahydrofolate reductase gene in patients with bipolar disorder and schizophrenia. Eur. Psychiatry 22, 39–43.
Kempisty, B., Mostowska, A., Gorska, I., Luczak, M., Czerski, P., Szczepankiewicz, A., Hauser, J., Jagodzinski, P.P., 2006. Association of 677C>T polymorphism of methylenetetrahydrofolate reductase (MTHFR) gene with bipolar disorder and schizophrenia. Neurosci. Lett. 400, 267–271.
Kim, J.M., Kim, S.W., Shin, I.S., Yang, S.J., Park, W.Y., Kim, S.J., Shin, H.Y., Yoon, J.S., 2008. Folate, vitamin b(12), and homocysteine as risk factors for cognitive decline in the elderly. Psychiatry Investig. 5, 36–40.
Kim, J.M., Stewart, R., Kim, S.W., Yang, S.J., Shin, I.S., Yoon, J.S., 2009a. Modification by two genes of associations between general somatic health and incident depressive syndrome in older people. Psychosom. Med. 71, 286–291.
Kim, S., Jung, D.C., Ahn, Y.M., Kim, Y.S., 2009b. P.1.a.021 Genetic association of MTHFR gene with schizophrenia and bipolar disorder. Eur. Neuropsychopharmacol. 19, S237.
Kim, Y.I., 1999. Folate and carcinogenesis: evidence, mechanisms, and implications. J. Nutr. Biochem. 10, 66–88.
Krebs, M.O., Bellon, A., Mainguy, G., Jay, T.M., Frieling, H., 2009. One-carbon metabolism and schizophrenia: current challenges and future directions. Trends Mol. Med. 15, 562–570.
Kronenberg, G., Colla, M., Endres, M., 2009. Folic acid, neurodegenerative and neuropsychiatric disease. Curr. Mol. Med. 9, 315–323.
Kunugi, H., Fukuda, R., Hattori, M., Kato, T., Tatsumi, M., Sakai, T., Hirose, T., Nanko, S., 1998. C677T polymorphism in methylenetetrahydrofolate reductase gene and psychoses. Mol. Psychiatry 3, 435–437.
Laanpere, M., Altmae, S., Stavreus-Evers, A., Nilsson, T.K., Yngve, A., Salumets, A., 2010. Folate-mediated one-carbon metabolism and its effect on female fertility and pregnancy viability. Nutr. Rev. 68, 99–113.
Lahiri, D.K., Maloney, B., Zawia, N.H., 2009. The LEARn model: an epigenetic explanation for idiopathic neurobiological diseases. Mol. Psychiatry 14, 992–1003.
Lee, Y.S., Han, D.H., Jeon, C.M., Lyoo, I.K., Na, C., Chae, S.L., Cho, S.C., 2006. Serum homocysteine, folate level and methylenetetrahydrofolate reductase 677, 1298 gene polymorphism in Korean schizophrenic patients. Neuroreport 17, 743–746.
Levine, A.J., Figueiredo, J.C., Lee, W., Conti, D.V., Kennedy, K., Duggan, D.J., Poynter, J.N., Campbell, P.T., Newcomb, P., Martinez, M.E., Hopper, J.L., Le Marchand, L., Baron, J.A., Limburg, P.J., Ulrich, C.M., Haile, R.W., 2010. A candidate gene study of folate-associated one carbon metabolism genes and colorectal cancer risk. Cancer Epidemiol. Biomarkers Prev. 19, 1812–1821.
Lewis, S.J., Lawlor, D.A., Davey Smith, G., Araya, R., Timpson, N., Day, I.N., Ebrahim, S., 2006. The thermolabile variant of MTHFR is associated with depression in the British Women's Heart and Health Study and a meta-analysis. Mol. Psychiatry 11, 352–360.
Lewis, S.J., Zammit, S., Gunnell, D., Smith, G.D., 2005. A meta-analysis of the MTHFR C677T polymorphism and schizophrenia risk. Am. J. Med. Genet. B Neuropsychiatr. Genet. 135B, 2–4.
Lichtenstein, P., Yip, B.H., Bjork, C., Pawitan, Y., Cannon, T.D., Sullivan, P.F., Hultman, C.M., 2009. Common genetic determinants of schizophrenia and bipolar disorder in Swedish families: a population-based study. Lancet 373, 234–239.
Lok, A., Assies, J., Koeter, M.W., Bockting, C.L., Visser, I., Moeton, M., 2008. Interaction of methylenetetrahydrofolate reductase (MTHFR) C677T polymorphism and folic acid levels in recurrent depression. J. Affect. Disord. 107, S75.
Mavros, M., Chirita, V., Popescu, O., Ferencz, B., 2008. The genetic polymorphism of MTHFR gene in schizophrenia. Rev. Med. Chir. Soc. Med. Nat. Iasi. 112, 76–82.
McGuffin, P., Rijsdijk, F., Andrew, M., Sham, P., Katz, R., Cardno, A., 2003. The heritability of bipolar affective disorder and the genetic relationship to unipolar depression. Arch. Gen. Psychiatry 60, 497–502.
Moskvina, V., Craddock, N., Holmans, P., Nikolov, I., Pahwa, J.S., Green, E., Owen, M.J., O'Donovan, M.C., 2009. Gene-wide analyses of genome-wide association data sets: evidence for multiple common risk alleles for schizophrenia and bipolar disorder and for overlap in genetic risk. Mol. Psychiatry 14, 252–260.
Muntjewerff, J.W., Kahn, R.S., Blom, H.J., den Heijer, M., 2006. Homocysteine, methylenetetrahydrofolate reductase and risk of schizophrenia: a meta-analysis. Mol. Psychiatry 11, 143–149.
Narayan, P., Dragunow, M., 2010. Pharmacology of epigenetics in brain disorders. Br. J. Pharmacol. 159, 285–303.
O'Donovan, M.C., Craddock, N.J., Owen, M.J., 2009. Genetics of psychosis; insights from views across the genome. Hum. Genet. 126, 3–12.
Ozbek, Z., Kucukali, C.I., Ozkok, E., Orhan, N., Aydin, M., Kilic, G., Sazci, A., Kara, I., 2008. Effect of the methylenetetrahydrofolate reductase gene polymorphisms on homocysteine, folate and vitamin B12 in patients with bipolar disorder and relatives. Prog. Neuropsychopharmacol. Biol. Psychiatry 32, 1331–1337.
Pan, C.C., McQuoid, D.R., Taylor, W.D., Payne, M.E., Ashley-Koch, A., Steffens, D.C., 2009. Association analysis of the COMT/MTHFR genes and geriatric depression: an MRI study of the putamen. Int. J. Geriatr. Psychiatry 24, 847–855.
Pasca, S.P., Dronca, E., Kaucsar, T., Craciun, E.C., Endreffy, E., Ferencz, B.K., Iftene, F., Benga, I., Cornean, R., Banerjee, R., Dronca, M., 2009. One carbon metabolism disturbances and the C677T MTHFR gene polymorphism in children with autism spectrum disorders. J. Cell. Mol. Med. 13, 4229–4238.
Perez, C.E., Ramos, V.C., Alcorta, A., Martinez, L.E., 2009. Association between folic acid, B12 vitamin, homocysteine and C677T polymorphism of MTHFR, in Mexican women with major depressive disorder. Biol. Psychiatry 65, 99.
Philibert, R., Gunter, T., Hollenbeck, N., Adams, W.J., Bohle, P., Packer, H., Sandhu, H., 2006. No association of the C677T methylenetetrahydrofolate reductase polymorphism with schizophrenia. Psychiatr. Genet. 16, 221–223.
Purcell, S.M., Wray, N.R., Stone, J.L., Visscher, P.M., O'Donovan, M.C., Sullivan, P.F., Sklar, P., 2009. Common polygenic variation contributes to risk of schizophrenia and bipolar disorder. Nature 460, 748–752.
Reif, A., Pfuhlmann, B., Lesch, K.P., 2005. Homocysteinemia as well as methylenetetrahydrofolate reductase polymorphism are associated with affective psychoses. Prog. Neuropsychopharmacol. Biol. Psychiatry 29, 1162–1168.
Roffman, J.L., Gollub, R.L., Calhoun, V.D., Wassink, T.H., Weiss, A.P., Ho, B.C., White, T., Clark, V.P., Fries, J., Andreasen, N.C., Goff, D.C., Manoach, D.S., 2008. MTHFR 677C- -> T genotype disrupts prefrontal function in schizophrenia through an interaction with COMT 158Val- -> Met. Proc. Natl. Acad. Sci. USA 105, 17573–17578.
Salanti, G., Sanderson, S., Higgins, J.P., 2005. Obstacles and opportunities in meta-analysis of genetic association studies. Genet. Med. 7, 13–20.
Sazci, A., Ergul, E., Guzelhan, Y., Kaya, G., Kara, I., 2003. Methylenetetrahydrofolate reductase gene polymorphisms in patients with schizophrenia. Brain Res. Mol. Brain Res. 117, 104–107.
Sazci, A., Ergul, E., Kucukali, I., Kara, I., Kaya, G., 2005. Association of the C677T and A1298C polymorphisms of methylenetetrahydrofolate reductase gene with schizophrenia: association is significant in men but not in women. Prog. Neuropsychopharmacol. Biol. Psychiatry 29, 1113–1123.
Shi, J., Gershon, E.S., Liu, C., 2008. Genetic associations with schizophrenia: meta-analyses of 12 candidate genes. Schizophr. Res. 104, 96–107.
Slopien, R., Jasniewicz, K., Meczekalski, B., Warenik-Szymankiewicz, A., Lianeri, M., Jagodzinski, P.P., 2008. Polymorphic variants of genes encoding MTHFR, MTR, and MTHFD1 and the risk of depression in postmenopausal women in Poland. Maturitas 61, 252–255.
Smulders, Y.M., Stehouwer, C.D., 2005. Folate metabolism and cardiovascular disease. Semin. Vasc. Med. 5, 87–97.
Stahl, S.M., 2010. Methylated spirits: epigenetic hypotheses of psychiatric disorders. CNS Spectr. 15, 220–230.
Stefansson, H., Ophoff, R.A., Steinberg, S., Andreassen, O.A., Cichon, S., Rujescu, D., Werge, T., Pietilainen, O.P., Mors, O., Mortensen, P.B., Sigurdsson, E., Gustafsson, O., Nyegaard, M., Tuulio-Henriksson, A., Ingason, A., Hansen, T., Suvisaari, J., Lonnqvist, J., Paunio, T., Borglum, A.D., Hartmann, A., Fink-Jensen, A., Nordentoft, M., Hougaard, D., Norgaard-Pedersen, B., Bottcher, Y., Olesen, J., Breuer, R., Moller, H.J., Giegling, I., Rasmussen, H.B., Timm, S., Mattheisen, M., Bitter, I., Rethelyi, J.M., Magnusdottir, B.B., Sigmundsson, T., Olason, P., Masson, G., Gulcher, J.R., Haraldsson, M., Fossdal, R., Thorgeirsson, T.E., Thorsteinsdottir, U., Ruggeri, M., Tosato, S., Franke, B., Strengman, E., Kiemeney, L.A., Melle, I., Djurovic, S., Abramova, L., Kaleda, V., Sanjuan, J., de Frutos, R., Bramon, E., Vassos, E., Fraser, G., Ettinger, U., Picchioni, M., Walker, N., Toulopoulou, T., Need, A.C., Ge, D., Yoon, J.L., Shianna, K.V., Freimer, N.B., Cantor, R.M., Murray, R., Kong, A., Golimbet, V., Carracedo, A., Arango, C., Costas, J., Jonsson, E.G., Terenius, L., Agartz, I., Petursson, H., Nothen, M.M., Rietschel, M., Matthews, P.M., Muglia, P., Peltonen, L., St Clair, D., Goldstein, D.B., Stefansson, K., Collier, D.A., 2009. Common variants conferring risk of schizophrenia. Nature 460, 744–747.
Sterne, J.A.C., 2009. Meta-Analysis in Stata: An Updated Collection from the Stata Journal. Stata Press.

Please cite this article in press as: Peerbooms, O.L.J., et al. Meta-analysis of MTHFR gene variants in schizophrenia, bipolar disorder and unipolar depressive disorder: Evidence for a common genetic vulnerability?. Brain Behav. Immun. (2011), doi:10.1016/j.bbi.2010.12.006

Sugden, C., 2006. One-carbon metabolism in psychiatric illness. Nutr. Res. Rev. 19, 117–136.

Tan, E.C., Chong, S.A., Lim, L.C., Chan, A.O., Teo, Y.Y., Tan, C.H., Mahendran, R., 2004. Genetic analysis of the thermolabile methylenetetrahydrofolate reductase variant in schizophrenia and mood disorders. Psychiatr. Genet. 14, 227–231.

Ueland, P.M., Hustad, S., Schneede, J., Refsum, H., Vollset, S.E., 2001. Biological and clinical implications of the MTHFR C677T polymorphism. Trends Pharmacol. Sci. 22, 195–201.

van der Put, N.M., Gabreels, F., Stevens, E.M., Smeitink, J.A., Trijbels, F.J., Eskes, T.K., van den Heuvel, L.P., Blom, H.J., 1998. A second common mutation in the methylenetetrahydrofolate reductase gene: an additional risk factor for neural-tube defects? Am. J. Hum. Genet. 62, 1044–1051.

van der Put, N.M., van Straaten, H.W., Trijbels, F.J., Blom, H.J., 2001. Folate, homocysteine and neural tube defects: an overview. Exp. Biol. Med. (Maywood) 226, 243–270.

Van Snellenberg, J.X., de Candia, T., 2009. Meta-analytic evidence for familial coaggregation of schizophrenia and bipolar disorder. Arch. Gen. Psychiatry 66, 748–755.

Vilella, E., Virgos, C., Murphy, M., Martorell, L., Valero, J., Simo, J.M., Joven, J., Fernandez-Ballart, J., Labad, A., 2005. Further evidence that hyper-rhomocysteinemia and methylenetetrahydrofolate reductase C677T and A1289C polymorphisms are not risk factors for schizophrenia. Prog. Neuropsychopharmacol. Biol. Psychiatry 29, 1169–1174.

Wani, N.A., Hamid, A., Kaur, J., 2008. Folate status in various pathophysiological conditions. IUBMB Life 60, 834–842.

Wiemels, J.L., Smith, R.N., Taylor, G.M., Eden, O.B., Alexander, F.E., Greaves, M.F., 2001. Methylenetetrahydrofolate reductase (MTHFR) polymorphisms and risk of molecularly defined subtypes of childhood acute leukemia. Proc. Natl. Acad. Sci. USA 98, 4004–4009.

Yoshimi, A., Aleksic, B., Kawamura, Y., Takahashi, N., Yamada, S., Usui, H., Saito, S., Ito, Y., Iwata, N., Inada, T., Noda, Y., Yamada, K., Ozaki, N., 2010. Gene-wide association study between the methylenetetrahydrofolate reductase gene (MTHFR) and schizophrenia in the Japanese population, with an updated meta-analysis on currently available data. Schizophr. Res. 124, 216–222.

Yu, L., Li, T., Robertson, Z., Dean, J., Gu, N.F., Feng, G.Y., Yates, P., Sinclair, M., Crombie, C., Collier, D.A., Walker, N., He, L., St Clair, D., 2004. No association between polymorphisms of methylenetetrahydrofolate reductase gene and schizophrenia in both Chinese and Scottish populations. Mol. Psychiatry 9, 1063–1065.

Yuan, Y.G., Zhang, Z.J., Li, J.J., 2008. Plasma homocysteine but not MTHFR gene polymorphism is associated with geriatric depression in the Chinese population. Acta Neuropsychiatrica 20 (5), 251–255.

Zeisel, S.H., 2009. Importance of methyl donors during reproduction. Am. J. Clin. Nutr. 89, 673S–677S.

Zhang, H.Y., Luo, G.A., Liang, Q.L., Wang, Y., Yang, H.H., Wang, Y.M., Zheng, X.Y., Song, X.M., Chen, G., Zhang, T., Wu, J.X., 2008. Neural tube defects and disturbed maternal folate- and homocysteine-mediated one-carbon metabolism. Exp. Neurol. 212, 515–521.

Zintzaras, E., 2006. C677T and A1298C methylenetetrahydrofolate reductase gene polymorphisms in schizophrenia, bipolar disorder and depression: a meta-analysis of genetic association studies. Psychiatr. Genet. 16, 105–115.

Please cite this article in press as: Peerbooms, O.L.J., et al. Meta-analysis of MTHFR gene variants in schizophrenia, bipolar disorder and unipolar depressive disorder: Evidence for a common genetic vulnerability?. Brain Behav. Immun. (2011), doi:10.1016/j.bbi.2010.12.006