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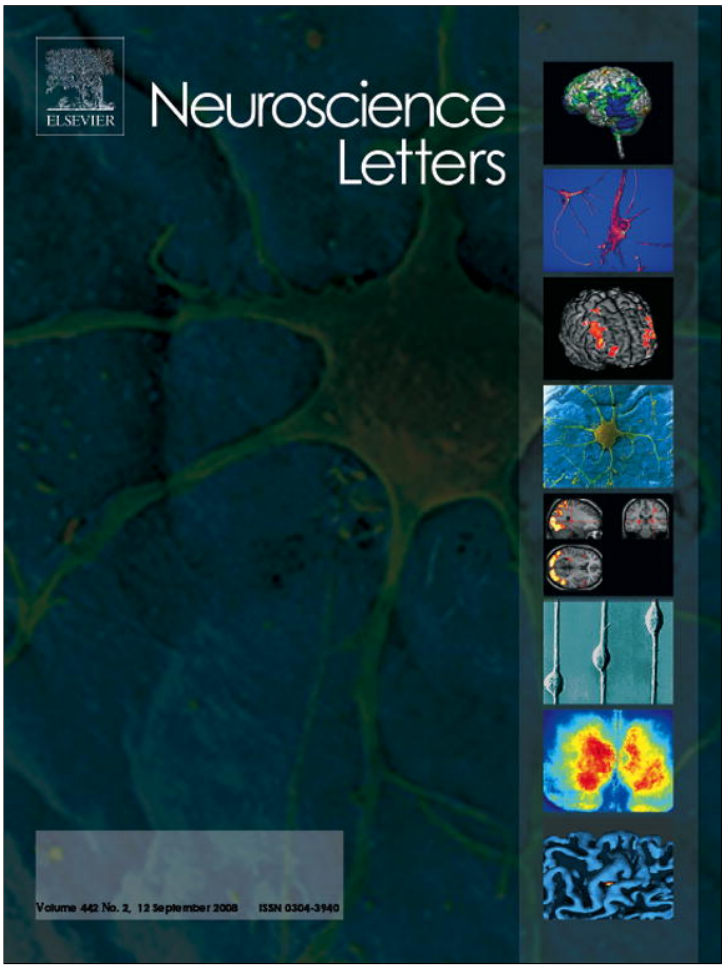

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Neuroscience Letters 442 (2008) 86–90

Contents lists available at ScienceDirect

Neuroscience Letters

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

TNF-alpha polymorphisms are associated with obsessive-compulsive disorder

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ARTICLE INFO

Article history:
Received 14 April 2008
Received in revised form 8 July 2008
Accepted 9 July 2008

Keywords:
Case-control study
Cytokine
Autoimmune
Molecular genetics
Rheumatic fever

ABSTRACT

Introduction: Several lines of evidence support an immunologic involvement in obsessive-compulsive disorder (OCD): the increased prevalence of OCD in patients with rheumatic fever (RF), and the aggregation of obsessive-compulsive spectrum disorders among relatives of RF probands. Tumor necrosis factor alpha is a proinflammatory cytokine involved in RF and other autoimmune diseases. Polymorphisms in the promoter region of the *TNFA* gene have been associated with RF. Given the association between OCD and RF, the goal of the present study was to investigate a possible association between polymorphisms within the promoter region of *TNFA* and OCD. *Materials and methods*: Two polymorphisms were investigated: −308 G/A and −238 G/A. The allelic and genotypic frequencies of these polymorphisms were examined in 111 patients who fulfilled DSM-IV criteria for OCD and compared with the frequencies in 250 controls. *Results*: Significant associations were observed between both polymorphisms and OCD. For −238 G/A, an association between the A allele and OCD was observed ($\chi^2 = 12.05$, $p = 0.0005$). A significant association was also observed between the A allele of the −308 G/A polymorphism and OCD ($\chi^2 = 7.09$, $p = 0.007$). Finally, a haplotype consisting of genotypes of these two markers was also examined. Significant association was observed for the A–A haplotype ($p = 0.0099$ after correcting for multiple testing). *Discussion*: There is association between the −308 G/A and −238 G/A *TNFA* polymorphisms and OCD in our Brazilian sample. However, these results need to be replicated in larger samples collected from different populations.
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Obsessive-compulsive disorder (OCD) is a psychiatric disorder characterized by unwanted thoughts (obsessions) and repetitive behaviors (compulsions) [2]. While the etiology of OCD is still unknown, the evidence of a genetic component is strong and has been supported by twin, family, linkage and genetic association studies [19].

OCD is a clinically heterogeneous disorder with several possible subtypes. It has been hypothesized that one of the subtypes is associated with autoimmune disorders triggered by streptococcal infections (e.g., rheumatic fever (RF) and Sydenham's chorea (SC) [33]. Children that develop acute OCD after a group A streptococcal infection were described by Swedo [46] who coined the acronym PANDAS for pediatric autoimmune neuropsychiatric disorders after streptococcal infection. These children developed OCD and tics but neither RF nor SC. It has also been reported that 4% of parents and grandparents of SC patients as well as 6.7% of the parents and grandparents of PANDAS developed RF when compared to parents and grandparents of controls (1.4%), suggesting a common liability between RF and OCD triggered by strep infections [46].

In Brazil, the incidence of acute RF has decreased by 75% over the last 10 years [16]; however, the rate is still very high, reaching 5000 new cases/year (Brazilian Health Ministry). Results from several studies conducted in Brazil suggest that OCD and related disorders are more frequent in RF patients. [31,22,1]. A family study also found higher rates of OCD and related disorders in first-degree relatives of RF patients than in first-degree relatives of controls, suggesting a familial association between RF and OCD [21]. While this is an interesting observation, further genetic studies are needed

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doi:10.1016/j.neulet.2008.07.022

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to support or refute this association. Nonetheless, the presence of auto-antibodies due to molecular mimicry mechanisms has been considered a possible explanation for the association between OCD and RF, following the autoimmune model for SC.

In order to understand this autoimmune hypothesis, several studies focusing on cytokines have been conducted. Cytokines are soluble glycoproteins of low molecular weight produced by different cellular types in all organs [3]. The imbalance in cytokine production can result in the loss of appropriate immune responses and excessive inflammation contributing to the severity of infectious diseases and progression of auto-immunity [28]. Tumor necrosis factor A (TNF-alpha) is a proinflammatory cytokine produced by monocytes, macrophages and T and B lymphocytes [6], and also by microglia in the central nervous system (CNS). It has been suggested that the polymorphism at −308 G/A in the *TNFA* gene affects its transcription. Studies with both increased [27,42] and decreased [9,48,49] TNF plasma levels associated with −308 A have been published. Though controversial, higher transcription of *TNFA* and consequent higher plasma levels is in accordance with a higher inflammatory response associated with the development of RF. Specific *TNFA* polymorphisms have been associated with RF in several studies [5,20,41,42,44].

TNFA has also been investigated in neuropsychiatric disorders due to the effect of TNF on the CNS. For example, *TNFA* polymorphisms have been associated with depression and schizophrenia [23,24,13,18,30]. More details about *TNFA* can be found at http://www.ncbi.nlm.nih.gov/entrez/dispomim.cgi?id=191160.

Several studies have compared plasma and cerebrospinal cytokine levels between OCD patients and controls [26,11,10,34,35,7]. The single previous study on a *TNFA* polymorphism and OCD found no association between the −308 G/A and OCD [50]. This previous study used a family based approach and evaluated only the −308 G/A polymorphism of the *TNFA* gene [50]. In the present study two polymorphisms in the promoter region of the *TNFA* were examined in a group of patients with OCD and in controls.

The project was approved by the Institutional Review Board of the University of São Paulo. Blood samples were obtained from OCD and control individuals after giving written informed consent. Some of the patients and controls were recruited for a family study. For a detailed description of the ascertainment see Hounie et al. [22,21]. Briefly, OCD patients were recruited through the Obsessive-Compulsive Spectrum Disorders Program (PROTOC-University of São Paulo). DNA samples of unselected healthy controls (unrelated bone marrow donors) were obtained from the Immunology Laboratory at the Heart Institute (InCor). They were screened for RF, which was an exclusion criterion but were not screened for psychiatric illnesses. Thus, it is possible that some may have OCD. The final sample consisted of 111 OCD patients and 250 controls. As this was an exploratory study, we did not calculate the sample power in advance and subjects were selected from available patients from PROTOC.

Among the 111 OCD patients 51 (46%) were females and 60 (54%) were males, while in the control group 138 (55.2%) were females and 112 (44.8%) were males ($\chi^2 = 2.28$, $p = 0.13$). The mean ages of the OCD and controls groups were, respectively, 30.8 (S.D. = 12.6; 95% CI 28.19–33.44) and 28.1 (S.D. = 13.8; 95%CI 26.25–29.99), not significantly different ($t = 11.64$; S.D. = 13.4; $p = 0.1$). The mean YBOCS score was 19 (S.D. = 11.6; 95% CI 16.45–21.63). Eight patients (7.2%) in the OCD group had a history of past RF. In the OCD group, 19 (17%) patients had a tic disorder.

Investigations comparing populations with different ethnic backgrounds have shown significant variations in the allelic frequency. The ability to detect a genetic association may be compromised in case-control studies when cases and controls do not come from the same ethnic population. One approach to minimize ethnic heterogeneity is to compare physical characteristics of the two samples. However, in Brazil, physical characteristics such as skin pigmentation, hair color and texture and shape of nose and lips are poor predictors of ethnic ancestry, which makes ethnic matching in our case-control studies difficult and unreliable [29,38,45]. A more reliable way to identify ethnic backgrounds is to genotype all individuals with the so-called ethnicity markers. It was beyond the scope of this study to obtain those data on these samples. However, given that both cases and controls were from the same catchment area in São Paulo and were seen in the same hospital, the two samples can be considered to be representative of the same geographical and socio-economic background.

Genomic DNA was extracted following the "salting out" method. The polymorphisms at *TNFA* −308 G/A (rs1800629) and *TNFA* −238 G/A (rs361525) in the promoter region of *TNFA* were typed by polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) as described elsewhere [32,41].

Allelic and genotypic distributions of the *TNFA* polymorphisms were compared between patients and controls. Genotypic distributions were compared using a χ^2-test with continuity correction. Allelic frequencies were also compared using standard χ^2 significance test. The expected frequencies were obtained using restricted maximum likelihood estimators. The R Package program version 2.6.1 was used for this analysis [40]. A test for deviations from the Hardy–Weinberg equilibrium (HWE) was performed using the HWE program [37]. Haplotype analysis was performed using the UNPHASED software [14] which performs permutation analyses and points to the haplotype that confers higher risk (best p-value) and the general risk of the haplotype that contains risk alleles (global significance). For all statistic tests, the critical level of significance was set at 0.05.

The genotypic distribution of the −308 G/A polymorphism was in HWE both in cases ($\chi^2 = 0.02$, $p = 0.86$) and in controls ($\chi^2 = 2.03$, $p = 0.15$) as well as the −238 G/A in the control group ($\chi^2 = 1.87$, $p = 0.17$). The −238 G/A polymorphism in the case group was not in HWE ($\chi^2 = 16.9$, $p = 0.00004$). The genotypic and allelic distributions of the −238 G/A polymorphism are displayed in Table 1. As can be seen, the frequency of the A allele of marker −238 G/A was signif-

Table 1
Distribution of the genotypes and alleles of the TNFA (−238 or rs361525) polymorphism between patients and controls

Polymorphism −238	Cases (%)	Controls (%)	χ^2 rates	d.f.	p
Genotype					
A/A	6 (5.4)	1 (0.4)			
A/G	14 (12.6)	15 (6.0)			
G/G	91 (82)	234 (93.6)			
Total	111 (100)	250 (100)	11.94	2	0.002
(AA + AG) × GG			10.29	1	0.001
AA × (AG + GG)			7.66	1	0.005
Alleles A × G	$A = 26$; $G = 196$	$A = 17$; $B = 487$	12.05	1	0.0005

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Table 2
Distribution of the genotypes and alleles of the TNFA-(−308 or rs1800629) polymorphism between patients and controls

Polymorphism −308	Cases (%)	Controls (%)	χ^2	d.f.	p
Genotype					
A/A	3 (2.7)	4 (1.6)			
A/G	32 (27.9)	38 (15.2)			
G/G	76 (69.4)	208 (83.2)			
Total	111 (100)	250 (100)	8.69	2	0.012
(AA + AG) × GG			9.08	1	0.002
AA × (AG + GG)			0.08	1	0.773
ALLELES A × G	A = 38; B = 184	A = 46; B = 454	7.09	1	0.007

icantly elevated in cases (11.7%) when compared to controls (3.4%) ($\chi^2_{(1d.f.)} = 12.05$, $p = 0.0005$), suggesting an association of this allele with OCD. As expected, the genotypic distributions also differed significantly between cases and controls ($\chi^2_{(2)} = 11.94$, $p = 0.002$). However, since the cases were not in HWE, the discrepancy may have been even more pronounced than expected. Of interest is that there are more A/A and fewer A/G case individuals over what would be expected.

The genotypic and allelic distributions of the −308 G/A polymorphism are displayed in Table 2. Similar to the results of −238 G/A, the frequency of the A allele of marker −308 G/A was also significantly elevated in cases (17.1%) when compared to controls (9.2%) ($\chi^2_{(1)} = 7.09$, $p = 0.007$). Furthermore, given the allele frequency differences the genotypic differences were also significantly different ($\chi^2_{(2)} = 8.69$, $p = 0.012$). No significant differences between female and male patients regarding the two SNPs studied were observed (data not shown). There was an association with the haplotype compounded by the alleles A–A of the investigated polymorphisms (best p-value = 0.00003; global significance = 0.0099).

To our knowledge, this is the first study to report an association between a cytokine genetic polymorphism and OCD.

Few studies have assessed TNF-alpha levels in OCD patients. As noted above, the majority of these studies have only evaluated plasma, blood cultured cells or cerebrospinal cytokine levels. Conclusions from those studies have been controversial. Denys et al. [11] studied the secretion of TNF-alpha in cultures of cells of 50 OCD non-medicated patients and 25 controls. They found a significant decrease in the production of TNF-alpha ($p < 0.0001$) in OCD compared to controls. Furthermore, studies of adults with OCD found smaller concentrations of TNF-alpha in the peripheral blood [7,11,35]. Mittleman et al. [34] did not find different levels of TNF-alpha in pediatric OCD compared to attention deficit/hyperactivity disorder or schizophrenia patients. On the other hand, Konuk et al. [26] evaluated plasma levels of TNF-alpha in 31 drug-free OCD patients and 31 controls. They found significant increases in the levels of TNF-alpha in OCD patients. In addition, the age of onset was negatively correlated with TNF-alpha level. It is difficult to compare results from different studies on TNF-alpha plasma levels due to their methodological differences. TNF-alpha plasma levels vary with age, body mass index, gender, time of the day, medicine intake, and several other factors [17,35,26]. Although studying TNF polymorphisms does not answer the question of the existence of an association between OCD and high TNF plasma levels, it may shed light to an immune component in some OCD subgroups. It is noteworthy that the myelin oligodendrocyte glycoprotein (MOG) gene has been reported to be associated with OCD. MOG is involved in the autoimmune response that characterizes multiple sclerosis, playing an important role in mediating the complement cascade [51].

The −308 *TNFA* polymorphism has been studied in RF patients in at least five independent samples from Turkey [42,5], Mexico [20], Brazil [41], and Egypt [44]. In all but one study [5], the A allele of the −308 G/A polymorphism was found to be associated with rheumatic heart disease. The A allele of the −238 G/A polymorphism was found to be associated with RF by Ramasawmy et al. [41] although in the Mexican study [20] the association was with the G allele. The A allele of the −308 G/A polymorphism has also been associated to other autoimmune diseases with neuropsychiatric manifestations such as lupus erythematosus [53]. The only previous study with OCD probands was a family based study evaluating *TNFA* −308 G/A in 127 OCD probands and their parents, which found negative results [50].

It has been suggested that the polymorphism at −308G/A affects *TNFA* transcription. Studies with both positive [27,42] and negative [49,9,48] findings correlating *TNFA* transcription with *TNFA* polymorphisms have been published. Results from a recent study suggest that the *TNFA* −308 G allele cosegregates with high serum TNF-alpha level in family studies [17]. Similarly, controversial reports concerning the *TNFA* −238 G/A polymorphism are also found in the literature [15,4,12,39,25] with the suggestion that the presence of the A allele may lead to increased transcription of *TNFA*.

In the current study, we observed an association between the A alleles of the two polymorphisms studied. Unfortunately, it was not possible to examine TNF plasma levels in our sample. On the other hand, considering that TNF plasma levels vary with several factors, it is possible that the presence of a polymorphism per se may not be sufficient to explain TNF plasma levels which are the result of a complex chain of interconnected genetic and environmental factors. Furthermore, changes in behavior may probably be related to changes in the CNS through local production of cytokines such as TNF and not to levels of TNF-alpha in the plasma [43].

Understanding the interaction between the CNS and the immune system is of great importance. It has been known for quite some time that serotonergic pathways are implicated in the pathophysiology of OCD [52]. Furthermore, it has been demonstrated that TNF-alpha may influence serotonin transporter activity. Conversely, it has been reported that the serotonin transporter may induce TNF-alpha expression [36,8,26]. Thus, further study of this and other genes is warranted.

The main limitation of the present study is the sample size and the fact that it was not possible to accurately control for possible population stratification. It is also somewhat concerning that the case group was not in HWE for the −238 G/A polymorphism. On the other hand, deviations from HWE can be very informative. In control subjects, deviations could indicate that one or more of the model assumptions have been violated or that a genotyping error has occurred. In case subjects, deviation from HWE, assuming sources of error have been eliminated, may indicate the association of a locus with disease [47]. Other explanation could be ethnic differences between subjects and consequent sample stratification. It is generally accepted that Hardy–Weinberg disequilibrium may be secondary to ethnic stratification. As noted, this could be the case in the current study, since we did not adequately control for ethnic background. However, the lack of HWE occurred only with the

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−238 G/A and not with the −308 G/A polymorphism, which was also associated with OCD, although less significantly, suggesting that the association is real.

These findings should be replicated in larger samples and in populations from different ethnic backgrounds. In addition, it is possible that the polymorphisms examined here are in linkage disequilibrium with non-identified genes that are in fact those contributing to the pathogenesis of OCD or even with other polymorphisms within the *TNFA* gene. The SNPs studied were a reasonable starting point to explore the hypothesis of this study. Our results suggest that the evaluation of this gene is important to clarify its role in OCD. However, a more comprehensive SNP coverage within the *TNFA* is warranted.

Future studies which correlate *TNFA* plasma level and *TNFA* polymorphisms in OCD are also warranted.

In conclusion, we observed an association of the A alleles of two *TNFA* polymorphisms with OCD. Of note is that the same alleles that been reported to be associated with RF. This finding further supports the idea that OCD and RF may share common genetic susceptibility. The replication of these findings in an independent sample will give support to the autoimmune theory for OCD and open new possibilities for its treatment.

Acknowledgments

This work was supported by grants from CAPES/PRODOC, FAPESP (grants 98/15-013-9 and 2005/55628-8), and CNPq.

We thank Maria Alice and Maria Eugênia De Mathis, Juliana B Diniz, Pedro G Alvarenga, André A Seixas, Fernando Akkerman, Marcos S Vasconcelos, Maria C Bravo, Priscila Chacon, Sérgio Broto, Yara Garzuzi, Rosemar Prota and Daniela Dourado for conducting the clinical evaluations. We also thank Maria Lúcia Carnevale Marin, Lea Oliveira, Kellen Faé and Gabriel Victora for helping in the genotyping. We also thank Dr. Rajendranath Ramasawmy for his help and support.

References

[1] P.G. Alvarenga, A.G. Hounie, M.T. Mercadante, J.B. Diniz, M.S. Vasconcelos, G. Spina, E.C. Miguel, Obsessive-compulsive symptoms in heart disease patients with and without history of rheumatic fever, J. Neuropsychiatry Clin. Neurosci. 18 (3) (2006) 405–408.
[2] American Psychiatric Association, Diagnostic and Statistical Manual of Mental Disorders, fourth edition, American Psychiatric Association, Washington, DC, 1994, p. 886.
[3] F.R. Balkwill, F. Burke, The cytokine network, Immunol. Today 10 (9) (1989) 299–304.
[4] J.P. Bayley, H. de Rooij, P.J. van den Elsen, T.W. Huizinga, C.L. Verweij, Functional analysis of linker-scan mutants spanning the −376, −308, −244, and −238 polymorphic sites of the TNF-alpha promoter, Cytokine 14 (2001) 316–323.
[5] A. Berdeli, Y. Tabel, H.A. Celik, R. Ozyurek, B. Dogrusoz, H.H. Aydin, Lack of association between TNFalpha gene polymorphism at position −308 and risk of acute rheumatic fever in Turkish patients, Scand. J. Rheumatol. 35 (2006) 44–47.
[6] B. Beutler, A. Cerami, The biology of cachectin/TNF-a primary mediator of the host response, Annu. Rev. Immunol. 7 (1989) 625–655.
[7] F. Brambilla, G. Perna, L. Bellodi, C. Arancio, A. Bertani, G. Perini, C. Carraro, F. Gava, Plasma interleukin-1 beta and tumor necrosis factor concentrations in obsessive-compulsive disorders, Biol. Psychiatry 42 (11) (1997) 976–981.
[8] K. Brebner, S. Hayley, R. Zacharko, Z. Merali, H. Anisman, Synergistic effects of interleukin-1beta, interleukin-6, and tumor necrosis factor-alpha: central monoamine, corticosterone, and behavioral variations, Neuropsychopharmacology 22 (6) (2000) 566–580.
[9] B.M. Brinkman, D. Zuijdeest, E.L. Kaijzel, F.C. Breedveld, C.L. Verweij, Relevance of the tumor necrosis factor alpha (TNF alpha) −308 promoter polymorphism in TNF alpha gene regulation, J. Inflamm. 46 (1995) 32–41.
[10] L.L. Carpenter, G.R. Heninger, C.J. McDougle, A.R. Tyrka, C.N. Epperson, L.H. Price, Cerebrospinal fluid interleukin-6 in obsessive-compulsive disorder and trichotillomania, Psychiatry Res. 112 (3) (2002) 257–262.
[11] D. Denys, S. Fluitman, A. Kavelaars, C. Heijnen, H. Westenberg, Decreased TNF-alpha NK activity in obsessivecompulsive disorder, Psychoneuroendocrinology 29 (7) (2004) 945–952.
[12] C. Drouet, A.N. Shakhov, C.V. Jongeneel, Enhancers and transcription factors controlling the inducibility of the tumor necrosis factor-alpha promoter in primary macrophages, J. Immunol. 147 (1991) 1694–1700.
[13] S. Duan, Y. Xu, W. Chen, Z. Liu, T. Guo, J. Gao, L. Bian, Y. Zhen, X. Li, X. Zhang, Y. Pan, N. Gu, G. Feng, L. He, No association between the promoter variants of tumor necrosis factor alpha (TNF-alpha) and schizophrenia in Chinese Han population, Neurosci. Lett. 366 (2) (2004) 139–143.
[14] F. Dudbridge, Pedigree disequilibrium tests for multilocus haplotypes, Genet. Epidemiol. 25 (2003) 115–221.
[15] C.L. Fong, A.H. Siddiqui, D.F. Mark, Identification and characterization of a novel repressor site in the human tumor necrosis factor alpha gene, Nucl. Acids Res. 22 (1994) 1108–1114.
[16] L. Guilherme, R. Ramasawmy, J. Kalil, Rheumatic fever and rheumatic heart disease: genetics and pathogenesis, Scand. J. Immunol. 66 (2–3) (2007) 199–207.
[17] N. Haddy, C. Sass, S. Maumus, B. Marie, S. Droesch, G. Siest, D. Lambert, S. Visvikis, Biological variations, genetic polymorphisms and familial resemblance of TNF-alpha and IL-6 concentrations: STANISLAS cohort, Eur. J. Hum. Genet. 13 (2005) 109–117.
[18] H.Y. Handoko, D.J. Nancarrow, N.K. Hayward, J.U. Ohaeri, H. Aghanwa, J.J. McGrath, D.F. Levinson, C. Johns, M.K. Walters, D.A. Nertney, T.N. Srinivasan, R. Thara, B.J. Mowry, Tumor necrosis factor haplotype analysis amongst schizophrenia probands from four distinct populations in the Asia-Pacific region, Am. J. Med. Genet. B: Neuropsychiatr. Genet. 121 (1) (2003) 1–6.
[19] M.J. Hemmings, D.J. Stein, The current status of association studies in obsessive-compulsive disorder, Psychiatr. Clin. N. Am. 29 (2006) 411–444.
[20] G. Hernandez-Pacheco, C. Flores-Dominguez, J.M. Rodriguez-Perez, N. Perez-Hernandez, J.M. Fragoso, A. Saul, E. Alvarez-Leon, J. Granados, P.A. Reyes, G. Vargas-Alarcon, Tumor necrosis factor-alpha promoter polymorphisms in Mexican patients with rheumatic heart disease, J. Autoimmun. 21 (2003) 59–63.
[21] A.G. Hounie, D.L. Pauls, M.C. do Rosario-Campos, M.T. Mercadante, J.B. Diniz, M.A. De Mathis, M.E. De Mathis, P. Chacon, R.G. Shavitt, M. Curi, L. Guilherme, E.C. Miguel, Obsessive-compulsive spectrum disorders and rheumatic fever: a family study, Biol. Psychiatry 61 (3) (2007) 266–272.
[22] A.G. Hounie, D.L. Pauls, M.T. Mercadante, M.C. Rosario-Campos, R.G. Shavitt, M.A. de Mathis, P.G. de Alvarenga, M. Curi, E.C. Miguel, Obsessive-compulsive spectrum disorders in rheumatic fever with and without Sydenham's chorea, J. Clin. Psychiatry 65 (7) (2004) 994–999.
[23] T.Y. Jun, C.U. Pae, J.H. Hoon-Han, W.M. Chae, K.S. Bahk, A. Kim, Serretti, Possible association between-G308A tumor necrosis factor-alpha gene polymorphism and major depressive disorder in the Korean population, Psychiatr. Genet. 13 (3) (2003) 178–179.
[24] T.Y. Jun, C.U. Pae, J.H. Chae, W.M. Bahk, K.S. Kim, C.W. Pyo, H. Han, Tumor necrosis factor-beta gene polymorphism may not be associated with major depressive disorder in the Korean population, Psychiatry Clin. Neurosci. 57 (1) (2003) 31–35.
[25] W. Kaluza, E. Reuss, S. Grossmann, R. Hug, R.E. Schopf, P.R. Galle, M.E. Hermann, T. Hoehler, Different transcriptional activity and in vitro TNF-alpha production in psoriasis patients carrying the TNF-alpha 238A promoter polymorphism, J. Invest. Dermatol. 114 (2000) 1180–1183.
[26] N. Konuk, I.O. Tekın, U. Ozturk, L. Atik, N. Atasoy, S. Bektas, A. Erdogan, Plasma levels of tumor necrosis factor-alpha and interleukin-6 in obsessive compulsive disorder, Hindawi Publ. Corp. Med. Inflamm. 27 (2007) 1–5.
[27] K.M. Kroeger, K.S. Carville, L.J. Abraham, The −308 tumor necrosis factor-alpha promoter polymorphism effects transcription, Mol. Immunol. 34 (1997) 391–399.
[28] T.A. Luger, T. Schwarz, Evidence for an epidermal cytokine network, J. Invest. Dermatol. 95 (Suppl. 6) (1990) 100S–104S.
[29] P.M. McKeigue, J.R. Carpenter, E.J. Parra, M.D. Shriver, Estimation of admixture and detection of linkage in admixed populations by a Bayesian approach: application to African American populations, Ann. Hum. Genet. 64 (2000) 171–186.
[30] I.V. Meira-Lima, A.C. Pereira, G.F. Mota, M. Floriano, F. Araújo, A.J. Mansur, J.E. Krieger, H. Vallada, Analysis of a polymorphism in the promoter region of the tumor necrosis factor alpha gene in schizophrenia and bipolar disorder: further support for an association with schizophrenia, Mol. Psychiatry 8 (2003) 718–720.
[31] M.T. Mercadante, G.F. Busatto, P.J. Lombroso, H. Prado, M.C. Rosário-Campos, R. do Valle, M.J. Marques-Dias, M.H. Kiss, J.F. Leckman, E.C. Miguel, The psychiatric symptoms of rheumatic fever, Am. J. Psychiatry 157 (12) (2000) 2036–2038.
[32] F. Migot-Nabias, L.E. Mombo, A.J. Luty, B. Dubois, R. Nabias, C. Bisseye, P. Millet, C.Y. Lu, P. Deloron, Human genetic factors related to susceptibility to mild malaria in Gabon, Genes Immun. 1 (2000) 435–441.
[33] E.C. Miguel, J.F. Leckman, S. Rauch, M.C. do Rosario-Campos, A.G. Hounie, M.T. Mercadante, P. Chacon, D.L. Pauls, Obsessive-compulsive disorder phenotypes: implications for genetic studies, Mol. Psychiatry 10 (3) (2005) 258–275.
[34] B.B. Mittleman, F.X. Castellanos, L.K. Jacobsen, J.L. Rapoport, S.E. Swedo, G.M. Shearer, Cerebrospinal fluid cytokines in pediatric neuropsychiatric disease, J. Immunol. 159 (6) (1997) 2994–2999.
[35] P. Monteleone, F. Catapano, M. Fabrazzo, A. Tortorella, M. Maj, Decreased blood levels of tumor necrosis factor alpha in patients with obsessive-compulsive disorder, Neuropsychobiology 37 (4) (1998) 182–185.
[36] R. Mössner, A. Heils, G. Stöber, O. Okladnova, S. Daniel, K.P. Lesch, Enhancement of serotonin transporter function by tumor necrosis factor alpha but not by interleukin-6, Neurochem. Int. 33 (3) (1998) 251–254.
[37] J. Ott, Methods of analysis and resources available for genetic trait mapping, J. Heredity 90 (1999) 68–70.

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[38] F.C. Parra, R.C. Amado, J.R. Lambertucci, J. Rocha, C.M. Antunes, S.D. Pena, Color and genomic ancestry in Brazilians, Proc. Natl. Acad. Sci. 100 (2003) 177–182.
[39] F. Pociot, S. D'Alfonso, S. Compasso, R. Scorz, P.M. Richiardi, Functional analysis of a new polymorphism in the human TNF alpha gene promoter, Scand. J. Immunol. 42 (1995) 501–504.
[40] R. Development Core Team, R: A language and environment for statistical computing, R Foundation for Statistical Computing, Vienna, Austria, 2007, ISBN 3-900051-07-0, URL http://www.R-project.org.
[41] R. Ramasawmy, K.C. Faé, G. Spina, G.D. Victora, A.C. Tanaka, S.A. Palácios, A.G. Hounie, E.C. Miguel, S.E. Oshiro, A.C. Goldberg, J. Kalil, L. Guilherme, Association of polymorphisms within the promoter region of the tumor necrosis factor-alpha with clinical outcomes of rheumatic fever, Mol. Immunol. 44 (8) (2007) 1873–1878.
[42] N. Sallakci, G. Akcurin, S. Koksoy, F. Kardelen, A. Uguz, M. Coskun, H. Ertug, O. Yegin, TNF-alpha G-308A polymorphism is associated with rheumatic fever and correlates with increased TNF-alpha production, J. Autoimmun. 25 (2005) 150–154.
[43] M. Sawada, K. Imamura, T. Nagatsu, Role of cytokines in inflammatory process in Parkinson's disease, J. Neural Transm. Suppl. (70) (2006) 373–381.
[44] A. Settin, H. Abdel-Hady, R. El-Baz, I. Saber, Gene polymorphisms of TNF-alpha (−308), IL-10(−1082), IL-6(−174), and IL-1Ra (VNTR) related to susceptibility and severity of rheumatic heart disease, Pediatr. Cardiol. 28 (5) (2007) 363–371.
[45] G. Suarez-Kurtz, D.D. Vargens, C.J. Struchiner, L. Bastos-Rodrigues, S.D. Pena, Self-reported skin color genomic ancestry and the distribution of GST polymorphisms, Pharmacogenet. Genom. 17 (2007) 765–771.
[46] S.E. Swedo, Pediatric autoimmune neuropsychiatric disorders associated with streptococcal infections (PANDAS), Mol. Psychiatry 7 (2002) S24–S25.
[47] J.S. Szeszko, J.M. Howson, J.D. Cooper, N.M. Walker, R.C. Twells, H.E. Stevens, S.L. Nutland, J.A. Todd, Analysis of polymorphisms of the interleukin-18 gene in type 1 diabetes and Hardy–Weinberg equilibrium testing, Diabetes 55 (2) (2006) 559–562.
[48] A.M. Uglialoro, D. Turbay, P.A. Pesavento, J.C. Delgado, F.E. McKenzie, J.G. Gribben, D. Hartl, E.J. Yunis, A.E. Goldfeld, Identification of three new single nucleotide polymorphisms in the human tumor necrosis factor-gene promoter, Tissue Antigens 52 (1998) 359–367.
[49] R.G. Westendorp, J.A. Langermans, T.W. Huizinga, C.L. Verweij, A. Sturk, Genetic influence on cytokine production in meningococcal disease, Lancet 349 (1997) 1912–1913.
[50] G. Zai, P.D. Arnold, E. Burroughs, M.A. Richter, J.L. Kennedy, Tumor necrosis factor-alpha gene is not associated with obsessive-compulsive disorder, Psychiatr. Genet. 16 (1) (2006) 43–45.
[51] G. Zai, Y.B. Bezchlibnyk, M.A. Richter, P. Arnold, E. Burroughs, C.L. Barr, J.L. Kennedy, Myelin oligodendrocyte glycoprotein (MOG) gene is associated with obsessive-compulsive disorder, Am. J. Med. Genet. B: Neuropsychiatr. Genet. 129 (1) (2004) 64–68.
[52] J. Zohar, J.L. Kennedy, E. Hollander, L.M. Koran, Serotonin-1D hypothesis of obsessive-compulsive disorder: an update, J. Clin. Psychiatry 65 (Suppl. 14) (2004) 18–21.
[53] J. Zúñiga, G. Vargas-Alarcón, G. Hernández-Pacheco, C. Portal-Celhay, J.K. Yamamoto-Furusho, J. Granados, Tumor necrosis factor-alpha promoter polymorphisms in Mexican patients with systemic lupus erythematosus (SLE), Genes Immun. 2 (7) (2001) 363–366.