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ASSOCIATION OF *GPX4*-718C/T POLYMORPHISM WITH TOURETTE SYNDROME IN A CHINA POPULATION

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Abstract: Tourette syndrome (TS) is an inherited neuro developmental disorder. However, it has no definite susceptibility genes. In order to research the genetic susceptibility, *GPX4*-718C/T was as a candidate single nucleotide polymorphism (SNP) to study. We recruited 100 TS nuclear family trios. Alleles of *GPX4*-718C/T were detected by TaqMan real-time PCR. We analyzed the tag SNP in *GPX4* using the transmission disequilibrium test (TDT). TDT shown no over-transmission in the *GPX4*-718C/T polymorphism ($\chi2$=0.910, P=0.397, OR=1.535, 95%CI=0.875-2.693). Haplotype relative risk (HRR) was used to ensure the accuracy of the TDT result and no statistical difference was found in the HRR result ($\chi2$=2.261, P=0.133, OR=0.635, 95%CI=0.350-1.150). Although this finding indicated an unlikely association between *GPX4*-718C/T and TS in a China population, a potential role for *GPX4* cannot be ruled out in TS genetic susceptibility.
Keywords: Tourette syndrome; *GPX4*; Transmission disequilibrium test; Haplotype relative risk; Single nucleotide polymorphism.

1. Introduction

Tourette syndrome (TS) is an early onset inherited neuropsychiatric disorder with an estimated prevalence of 0.1–1% in children and adolescents between 5 and 18 years old(Robertson 2008). Following decades of extensive studies, the exact susceptibility genes remain poorly understood. Numerous literature shown that *DRD1* (dopamine receptor D1) (Chou et al. 2004), *ADR* (adrenergic receptor) (Brett et al. 1997), *5-HT* (5-hydroxytryptamine receptor) (Niesler et al. 2005), *SLITRK1* (slit and trk-like family member 1) (Abelson et al. 2005), and *NLGN4* (neuroligin 4) (Lawson-Yuen et al. 2008) may be association with TS genetic susceptibility. The results of these studies, however, were not satisfactory. Children with Tourette syndrome have high levels of biochemical indices of oxidative stress in previous study (Landau et al. 2012). And Glutathione Peroxidase (GPX) 4 is a unique membrane-associated

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selenium-dependent antioxidant enzyme that can directly reduce phospholipid hydroperoxides and protect against oxidative stress (Imai and Nakagawa 2003). *GPX4*-718C/T polymorphism as a tag SNP was relation to certain disorders, such as Kashin-Beck disease (Du et al. 2012). Now we designed this experiment to study the relationship between *GPX4*-718C/T polymorphism and TS genetic susceptibility in a China population.

2. Methods and Statistical Analysis

We recruited 100 TS nuclear family trios (patients and their parents) from the Affiliated Hospital of Qingdao University. Clinical material including age and gender were collected in a clinical database. TS patients comprised 79 male and 21 female, aged between 5 and 21 years old. Mean ± SD age of patients was 12.8±17.1 years old. They were diagnosed based on the criteria of the Diagnostic and Statistical Manual of Mental Disorders, fourth edition, text revision (DSM-IV). Genomic DNA was extracted from peripheral blood using standard chloroform method. *GPX4*-718C/T polymorphism was conducted by real-time PCR. Taqman probe and primer were synthesized by Applied Biosystems of Life Technologies. Forward and reverse primers were 5-CCGCCCGAGCCCCTGCCCACGCCCT-3 and 5-GGAGCCTTCCACCGGCACTCATGAC-3. PCR mixture contained 1.25μl 20×SNP Genotyping Assay, 12.5μl 2×PCR Master Mix, and 11.25μlDNA and DNase-free water. The Amplifications were carried out in a C1000™ thermal cycler system and CFX96™ real-time system under the following conditions: 95°C for 3 min, followed by 45 cycles at 95°C for 15s and 60°C for 1 min. For each cycle, fluorescence signals ware detected by VIC/FAM-labeled probes. Genotype data of parents were tested by Hardy-Weinberg equilibrium (HWE). Then all the data was counted using statistical software package SPSS 21.0. TDT and HRR were used to test the transmission disequilibrium. Odds ratios (ORs) and 95% confidence intervals (CIs) were used to show the relative risk degree. All of the Statistical significance was set at $P<0.05$.

3. Results

Genotype of parents was consistent with HWE and can undertake the genetics research ($\chi2=2.143$, $P=0.143$). The transmitted and non-transmitted groups were not statistically different in the TDT ($\chi2=0.910$, $P=0.397$, OR=1.535, 95%CI=0.875-2.693) (Table 1). The HRR also indicated no statistically difference between transmitted and non-transmitted groups

($\chi2$=2.261, P=0.133, OR=0.635, 95%CI=0.350-1.150) (Table 2). Both the results shown the *GPX4*-718C/T polymorphism may be no relation with TS genetic susceptibility.

Table 1: The TDT result of the *GPX4*-718C/T polymorphism

Groups		Non-transmitted allele	
		C	T
Transmitted allele	C	47	40
	T	49	64
TDT result		$\chi2$=0.910, P=0.397, OR=1.535, 95%CI=0.875-2.693	

Table 2: The HRR result of the *GPX4*-718C/T polymorphism

Groups	C(+)	C(-)
Transmitted allele	62	38
Non-transmitted allele	72	28
HRR result	$\chi2$=2.261, P=0.133, OR=0.635, 95%CI=0.350-1.150	

4. Discussion

The factors that contribute to the pathogenesis of TS are poorly defined. Twins and families studies (Pauls et al. 1981, Price et al. 1985) provided strong evidence that genetic factors, particularly polygenic hereditary patterns, are involved in the vertical transmission of TS. More recent studies suggested that the pattern of inheritance is more complex and that some genes have a major effect while others act as modifiers (Pauls 2001). Through too many researches in the field of genetics, the investigators were still not clear about its main predisposing genes, so the follow-up study of the genetics is still highly necessary. Oxidative stress is suspected to be important in neurodegenerative diseases including Alzheimer's disease, Huntington's disease, Depression and Multiple sclerosis (Patel and Chu 2011). And TS, as a neurodevelopmental disorder, has studied the role of oxidative stress in its pathogenesis (Landau et al. 2012, Shen et al. 2014). The protein encoded by *GPX4* belongs to the glutathione peroxidase family, members of which catalyze the reduction of hydrogen peroxide, organic hydroperoxides and lipid hydroperoxides, and thereby protect cells against oxidative damage

(Bellinger et al. 2009, Brigelius-Flohe 1999). Mutations in this gene are associated with Sedaghatian type of spondylometaphyseal dysplasia (SMDS) (Smith et al. 2014). These fully show that the *GPX4* plays a fatal role in the process of oxidative stress. G*PX4*-718C/T (*GPX4* 3'untranslated region, chromosome 19) as a tag SNP was association with diverse disorders by changing glutathione peroxidase family enzyme activity, such as Kidney disease, Kashin-Beck disease, Colorectal cancer (Du et al. 2012, Monteiro et al. 2013, Meplan et al. 2010). We guessed that there was a relationship between G*PX4*-718C/T polymorphism and TS genetic susceptibility. We collected 100 nuclear family trios to study, which could be favorable to exclude irrelevant factors. TDT and HRR ensure the reliability of the experimental results. Nevertheless, our statistical results of TDT and HRR shown that the P values were greater than 0.05 (P_{TDT}=0.397, P_{HRR}=0.133). These demonstrated that the *GPX4*-718C/T polymorphism may be no relation with TS genetic susceptibility in a China population. Certainly, there are some limitations in our study. Small sample size, single race and sole SNP are likely to result in the deviation of the results. And we will make up for these limitations as far as possible in the future research.

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References

[1] ABELSON JF ET AL. 2005. Sequence variants in SLITRK1 are associated with Tourette's syndrome. Science 310: 317-320.

[2] BELLINGER FP, RAMAN AV, REEVES MA AND BERRY MJ. 2009. Regulation and function of selenoproteins in human disease. Biochem J 422: 11-22.

[3] BRETT PM, CURTIS D, ROBERTSON MM AND GURLING HM. 1997. Neuroreceptor subunit genes and the genetic susceptibility to Gilles de la Tourette syndrome. Biol Psychiatry 42: 941-947.

[4] BRIGELIUS-FLOHE R. 1999. Tissue-specific functions of individual glutathione peroxidases. Free Radical Bio Med 27: 951-965.

[5] CHOU IC, TSAI CH, LEE CC, KUO HT, HSU YA, LI CI AND TSAI FJ. 2004.

Association analysis between Tourette's syndrome and dopamine D1 receptor gene in Taiwanese children. Psychiatric genetics 14: 219-221.

[6] DU XH, DAI XX, SONG RX, ZOU XZ, SUN WY, MO XY, BAI GL AND XIONG YM. 2012. SNP and mRNA expression for glutathione peroxidase 4 in Kashin-Beck disease. Brit J Nutr 107: 164-169.

[7] IMAI H AND NAKAGAWA Y. 2003. Biological significance of phospholipid hydroperoxide glutathione peroxidase (PHGPx, GPx4) in mammalian cells. Free radical biology & medicine 34: 145-169.

[8] LANDAU YE, STEINBERG T, RICHMAND B, LECKMAN JF AND APTER A. 2012. Involvement of immunologic and biochemical mechanisms in the pathogenesis of Tourette's syndrome. Journal of neural transmission 119: 621-626.

[9] LAWSON-YUEN A, SALDIVAR JS, SOMMER S AND PICKER J. 2008. Familial deletion within NLGN4 associated with autism and Tourette syndrome. European Journal of Human Genetics 16: 614-618.

[10] MEPLAN C, HUGHES DJ, PARDINI B, NACCARATI A, SOUCEK P, VODICKOVA L, HLAVATA I, VRANA D, VODICKA P AND HESKETH JE. 2010. Genetic variants in selenoprotein genes increase risk of colorectal cancer. Carcinogenesis 31: 1074-1079.

[11] MONTEIRO MB, PATENTE TA, MOHAMMEDI K, QUEIROZ MS, AZEVEDO MJ, CANANI LH, PARISI MC, MARRE M, VELHO G AND CORREA-GIANNELLA ML. 2013. Sex-specific associations of variants in regulatory regions of NADPH oxidase-2 (CYBB) and glutathione peroxidase 4 (GPX4) genes with kidney disease in type 1 diabetes. Free radical research 47: 804-810.

[12] NIESLER B, FRANK B, HEBEBRAND J AND RAPPOLD G. 2005. Serotonin receptor genes HTR3A and HTR3B are not involved in Gilles de la Tourette syndrome. Psychiatric genetics 15: 303-304.

[13] PATEL VP AND CHU CT. 2011. Nuclear transport, oxidative stress, and neurodegeneration. Int J Clin Exp Pathol 4: 215-229.

[14] PAULS DL. 2001. The genetics of Tourette syndrome. Current psychiatry reports 3: 152-157.

[15] PAULS DL, COHEN DJ, HEIMBUCH R, DETLOR J AND KIDD KK. 1981. Familial pattern and transmission of Gilles de la Tourette syndrome and multiple tics. Archives of general psychiatry 38: 1091-1093.

[16] PRICE RA, KIDD KK, COHEN DJ, PAULS DL AND LECKMAN JF. 1985. A twin study of Tourette syndrome. Archives of general psychiatry 42: 815-820.

[17] ROBERTSON MM. 2008. The prevalence and epidemiology of Gilles de la Tourette syndrome. Part 1: the epidemiological and prevalence studies. J Psychosom Res 65: 461-472.

[18] SHEN CP, CHOU IC, LIU HP, LEE CC, TSAI YH, WU BT, HSU BD, LIN WY AND TSAI FJ. 2014. Association of Glutathione S-Transferase P1 (GSTP1) Polymorphism with Tourette Syndrome in Taiwanese Patients. Genet Test Mol Bioma 18: 41-44.

[19] SMITH AC ET AL. 2014. Mutations in the enzyme glutathione peroxidase 4 cause Sedaghatian-type spondylometaphyseal dysplasia. J Med Genet 51: 470-474.