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Short Communication

The G allele in IL-10-1082 G/A may have a role in lowering the susceptibility to panic disorder in female patients

Kim H-J, Kim Y-K. The G allele in IL-10-1082 G/A may have a role in lowering the susceptibility to panic disorder in female patients.

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Background: Immune system activation is involved in the pathophysiology of panic disorder (PD). We investigated INF-γ + 874 A/T, TNF-α-308 G/A, and IL-10-1082 G/A single nucleotide polymorphisms (SNPs) to determine their association with PD.
Method: This study enroled 135 PD patients and 135 healthy controls. INF-γ + 874 A/T (rs2430561), TNF-α-308 G/A (rs1800629), and IL-10-1082 G/A (rs1800896) were genotyped.
Results: There were no differences in genotypes or allele frequencies between the patient and control groups, regardless of accompanying agoraphobia. However, for female patients, the G allele frequency in IL-10 SNP was higher in the control group than in the patient group. Additionally, the female control group had a higher frequency of the A/G and G/G genotype in the IL-10 SNP than the female patient group.
Conclusion: We suggest that the G allele in IL-10-1082 G/A might have a role in reducing the manifestations of PD in female patients. Further studies are needed to extend and confirm our findings.

Keywords: cytokine; IFN-r; IL-10; panic disorder; TNF-α

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Significant outcomes

- The G allele in IL-10-1082 G/A might have a role in reducing the manifestations of panic disorder in female patients.
- Gender differences in gene polymorphisms can alter the manifestations of panic disorder.

Limitations

- Our sample size was relatively small.
- Further studies are required to extend and confirm our preliminary findings in a larger sample of participants matched for age and gender.

Introduction

Panic disorder (PD) is classified as an anxiety disorder with repeated and unexpected panic attacks. Although PD is a common mental disorder with great disease burden, only a few convincing risk genes have been reported, with its genetic component proven by family and twin studies (1,2).

Previous studies have led to the increasing recognition that pro-inflammatory cytokines could

enhance the activity of enzymes related to the degradation of neurotransmitters, suggesting that immune system activation is involved in the pathophysiology of psychiatric disorders, including PD (3,4). Additionally, a broad spectrum of cytokine abnormalities, which suggest a generalised inflammatory state, may be present in individuals with PD (5). One study suggested that IL-1β levels are a marker of PD (6), while another suggested that IL-3 levels were sensitive to the presence of anxiety and stress (7). However, cytokine changes have not been found in experimentally provoked panic attacks (8).

Despite increasing interest in the biological factors underlying PD, few studies have been conducted on the associations between PD and its genetic aetiologies, with the exception of serotonin-related gene polymorphisms (9–11). Genetic polymorphisms of BDNF and the adenosine receptor have also been studied for their relationship with PD, but no association has been demonstrated (12,13). There has not been an association identified between IL-10 genotypes and PD (14).

Given insufficient data on the inflammatory pathophysiology of PD, we tried to detect a possible relationship between cytokine genotypes and allele frequencies in PD patients. We selected IFN-r + 874 A/T, TNF-α-308 G/A, and IL-10-1082 G/A single nucleotide polymorphisms (SNPs) due to their functionality in inflammation regulation (15–19), and hypothesised that these SNPs could be associated with PD.

Furthermore, as female patients have greater susceptibility to PD than male patients (20), we aimed to detect possible differences between subgroups divided by gender and the presence of accompanying agoraphobia. We believe that the identification of positive associations would aid in the understanding of the pathogenesis of PD.

Methods

Participants and assessments

A total of 135 patients were diagnosed with panic disorder with or without agoraphobia according to DSM-IV diagnostic criteria. Each patient was assessed by clinical interviews using the Structured Clinical Interview for DSM-IV (SCID) (21), and patients with premorbid schizophrenia, mood disorder, and other anxiety disorders were excluded from the study.

We enroled 135 healthy individuals in the normal control group who visited the Hospital for regular health screenings. Patients were randomly selected and excluded if they had any cancer, hypertension, diabetes mellitus, hepatitis, dyslipidemia, psychiatric family history, or a history of using psychotropic drugs. All patients enroled in the study were biologically unrelated native Koreans. The study protocol was approved by the Ethics Committee of Korea University, and written informed consent was obtained from all participants.

DNA analysis

INF-γ + 874 A/T (rs2430561), TNF-α-308 G/A (rs1800629), and IL-10-1082 G/A (rs1800896) were genotyped. The genotyping of the SNPs was conducted based on methods reported in previous studies (22–24).

Statistical analysis

Demographic and clinical variables between the patient group and control group were analysed using the χ^2 test, Fisher's exact test, and independent *t*-test. Deviations from a theoretical distribution of genotypes were analysed using Hardy–Weinberg equilibrium, which was tested using the χ^2 test for goodness of fit. Statistical analysis was performed using SPSS version 12.0 for Windows. The significance level was set at $p < 0.05$ for all statistical analyses.

Results

The gender distribution between the PD patients and healthy controls was M : F = 71 : 64 and M : F = 54 : 81, respectively, and the mean ages of each group were 38.56 ± 8.58 and 37.02 ± 6.97 years, respectively. There were no significant differences in the gender distribution ($\chi^2 = 4.31$, $p = 0.051$) and mean age ($t = 1.619$, $p = 0.107$) between the two groups.

The distributions of the three polymorphisms in PD patients and controls were in agreement with Hardy–Weinberg equilibrium. The Hardy–Weinberg equilibria of the three genes were as follows: INF-γ + 874 A/T (rs2430561) (PD, $\chi^2 = 1.285$, df = 1, $p = 0.257$; controls, $\chi^2 = 0.077$, df = 1, $p = 0.781$); TNF-α-308 G/A (rs1800629) (PD, $\chi^2 = 0.645$, df = 1, $p = 0.422$; controls, $\chi^2 = 0.789$, df = 1, $p = 0.374$); and IL-10-1082 G/A (rs1800896) (PD, $\chi^2 = 1.394$, df = 1, $p = 0.2377$; controls, $\chi^2 = 0.004$, df = 1, $p = 0.953$).

Comparisons between the PD and control groups in terms of INF-γ and TNF-α polymorphisms are summarised in Tables 1 and 2. There were no significant differences in genotype or allele frequency between the two groups. Male and female subgroups revealed no significant differences with either genotype. Other subgroups, regardless of the presence

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Table 1. Genotype and allele frequencies of IFNr874A/T in panic disorder (PD) patients and controls

	Genotypes					Allele frequencies			
	A/A	A/T	T/T	χ^2	*P*	A	T	χ^2	*P*
PD patients	91	42	2	3.80	0.15	224	46	2.94	0.11
Controls	105	28	2			238	32		
Male PD patients	53	17	1	1.36	0.51	123	19	0.02	0.43
Male controls	44	10	0			98	10		
Female PD patients	38	25	1	4.89	0.09	101	27	2.88	0.11
Female controls	61	18	2			140	22		
PD with agoraphobia	68	32	2	0.69	0.71	168	36	0.22	0.71
PD without agoraphobia	23	10	0			56	10		

Table 2. Genotype and allele frequencies of TNF-α G/A in panic disorder (PD) patients and controls

	Genotypes					Allele frequencies			
	G/G	G/A	A/A	χ^2	*P*	G	A	χ^2	*P*
PD patients	119	16	0	0.30	0.72	254	16	3.93	0.07
Controls	116	19	0			151	19		
Male PD patients	61	10	0	0.01	1.00	132	10	0.01	1.00
Male controls	46	8	0			100	8		
Female PD patients	58	6	0	0.61	0.60	122	6	0.57	0.62
Female controls	70	11	0			151	11		
PD with agoraphobia	90	12	0	<0.01	1.00	192	12	<0.01	1.00
PD without agoraphobia	29	4	0			62	4		

Table 3. Genotype and allele frequencies of IL10A/G in panic disorder (PD) patients and controls

	Genotypes					Allele frequencies			
	A/A	A/G	G/G	χ^2	*P*	A	G	χ^2	*P*
PD patients	124	10	1	3.13	0.21	258	12	2.61	0.15
Controls	115	19	1			249	21		
Male PD patients	64	6	1	1.18	0.55	134	8	1.19	0.36
Male controls	51	3	0			105	3		
Female PD patients	60	4	0	6.42	0.04	124	4	6.51	0.01
Female controls	64	16	1			144	18		
PD with agoraphobia	95	6	1	1.71	0.43	196	8	0.54	0.50
PD without agoraphobia	29	4	0			62	4		

of agoraphobia, were compared, but no statistically significant differences were observed.

There was also no significant difference between the patient and control groups in terms of the genotype or allele frequency of the IL-10 gene (Table 3). In the female subgroup, the G allele frequency was higher in the control group than in the PD group ($\chi^2 = 6.51$, $p = 0.01$), and the proportions of A/G and G/G were higher in the female control group than in the female PD patients ($\chi^2 = 6.42$, $p = 0.04$).

The age of onset of panic disorder was 33.0 ± 10.2 years, and the duration of panic disorder was 22.0 ± 9.3 months. There was no significant association between these polymorphisms and the age of onset or duration of illness in patients with panic disorder.

Discussions

To evaluate the pathophysiology of PD, we studied polymorphisms of INF-γ + 874 A/T, TNF-α-308 G/A, and IL-10-1082 G/A based on molecular evidence for their functionality in inflammation (25).

These three SNPs were previously studied in patients with suicidal attempts, and the INF-γ + 874 A/A, TNF-α-308 G/G, and IL-10-1082 A/A genotypes were demonstrated to show an association with suicide attempts (26). Another study investigated the association between these SNPs with suicide attempts in patients with major depressive disorder (MDD), which showed that the TNF-α-308 G/A polymorphism was an independent risk factor for suicide attempts in patients with MDD (27).

In comparison to previous reports, this study did not find significant differences in the genotype and allele frequencies of these 3 SNPs between the PD groups and the control groups; however, the G allele frequency in IL-10 was higher in the female control group than in female PD patients.

Some studies have reported that gender-specific gene promoter polymorphisms of the serotonin transporter disturb the HPA axis by controlling cortisol secretion (28), which is reflected in the manifestation of PD (29). These gender differences in gene polymorphisms, which can alter the manifestations of PD, were the reason we subdivided our study groups by gender. Although these results are limited by the small sample size due to the use of subgroups, which is a common methodological difficulty for such investigations, we suggest that the G allele in IL-10-1082 G/A might have a role in reducing the manifestations of PD in female patients.

As an anti-inflammatory cytokine, the association between IL-10 and MDD has been previously studied (30). IL-10 inhibits the nearly ubiquitous expression of indoleamine 2,3-deoxygenase (IDO), an enzyme responsible for directing tryptophan degradation, which is directly related to the 'neurodegeneration hypothesis' of psychiatric disorders (4,31). However, only a few studies have been performed investigating the relationship between IL-10 and PD. One study showed no significant association between SNPs in the IL-10 family of genes and PD patients (14). This study examined the rs1554286, rs2243176, rs1890866 SNPs of IL-10, which did not include our IL-10 SNP has limitation for including MDD.

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Stress results in lipopolysaccharide (LPS) – induced IL-10 production, and IL-10 is known to inhibit the production of INF-γ and TNF-α (32). Although the normalisation of IL-10 does not reverse the inhibition of cytokine regulation, we assumed that the negative results for INF-γ and TNF-α in our study could be due to the secondary effects of IL-10.

We suggest that our observed genotype and allele frequencies should be taken in context of our relatively small sample sizes. Further studies are required to extend and confirm our preliminary findings in a large sample of participants matched for age and gender.

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Authors' Contributions: Han-Joon Kim and Yong-Ku Kim designed the study and wrote the protocol. Han-Joon Kim wrote the first draft of the manuscript. All authors contributed to and have approved the final manuscript. This paper was carried out as the Master's thesis of Han-Joon Kim. We thank Jung-A Hwang for the laboratory work.

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Conflicts of Interest

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

Ethical Standards

All authors assert that all procedures contributing to this work complied with ethical standards of the relevant national and institutional committees on human experimentation and with the Helsinki Declaration of 1975, as revised in 2008.

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