TRAF5 and TRAF3IP2 Gene Polymorphisms Are Associated with Behçet’s Disease and Vogt-Koyanagi-Harada Syndrome: A Case-Control Study

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Abstract

Background: TRAF5 and TRAF3IP2 have been reported to be associated with several autoimmune diseases. Behçet’s disease (BD) and Vogt-Koyanagi-Harada (VKH) syndrome are two autoimmune uveitis entities whereby both genetic and environmental factors are thought to be involved.

Objective: The role of TRAF5 and TRAF3IP2 in BD and VKH has not yet been reported and was therefore the subject of this study.

Methods: The study included 789 BD patients, 940 VKH patients and 1601 healthy unrelated individuals. Genotyping was performed by polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) or TaqMan® SNP Genotyping Assay. Real-Time PCR was used to detect mRNA expression from PBMCs obtained from healthy controls with (n = 22) or without (n = 79) stimulation. Levels of TNF-α, IL-6 and IL-8 in culture supernatants were measured by ELISA (n = 22).

Results: Three SNPs (rs6540679, rs12569232, rs10863888) of TRAF5 and rs13210247 of TRAF3IP2 were significantly associated with Behçet’s disease and VKH syndrome (corrected P values ranging from 9.45 × 10^{-12} to 0.027). TRAF3IP2 rs33980500 and rs13190932 were not polymorphic in Han Chinese. Following stimulation by lipopolysaccharide (LPS), carriers of the GG genotype of rs6540679/TRAF5 had a higher TRAF5 mRNA expression (p = 0.004) and an increased TNF-α (p = 0.0052) and IL-6 (p = 0.0014) level compared with AA and AG genotype carriers.

Conclusion: This study provides evidence that TRAF5 and TRAF3IP2 genes are involved in the development of BD and VKH syndrome. Functional research suggested that TRAF5 gene polymorphisms may regulate TRAF5 expression and downstream inflammatory cytokines such as TNF-α and IL-6.

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Introduction

Uveitis is an intraocular inflammation that is caused by either infectious or noninfectious mechanisms and which can lead to serious visual impairment. To unravel the immunological mechanisms that cause uveitis we have focused on two commonly occurring uveitis entities in China, namely Behçet’s disease (BD) and Vogt Koyanagi Harada (VKH) syndrome [1,2].

BD is an autoinflammatory disease characterized by a diverse spectrum of clinical manifestations including recurrent oral aphthae, uveitis, multiform skin lesions and genital ulceration [3,4]. It has a relatively high prevalence along the Silk Road countries such as Japan, China, Turkey, and the Mediterranean region [5]. The usual onset is between the age of twenty to forty. The cause of BD is still unknown but epidemiological studies have suggested that certain genetic factors play an important role in its development [6]. We and others have recently identified polymorphisms in multiple immunoregulatory genes as a risk factor for developing BD, such as CD40, STAT3, STAT4 and JAK2 [7,8,9]. These pathways are extremely complex and many other gene polymorphisms remain to be studied to unravel the exact genetic susceptibility to BD.
Patients and healthy controls study population

All subjects gave their written informed consent for this study, and the study protocol was approved by the Ethics Committee of the First Affiliated Hospital of Chongqing Medical University, Chongqing, China (Permit Number: 2009-201008). All procedures followed the tenets of the Declaration of Helsinki.

A case-control genetic study of two genes in individuals with or without uveitis was performed. 789 BD patients, 940 VKH patients and 1601 healthy unrelated individuals were included in this study. All subjects were of Han Chinese descent. Blood samples were obtained from the First Affiliated Hospital of Chongqing Medical University (Chongqing, China) through 2005 until 2013.

Figure 1. Schematic location of the chosen SNPs of TRAF5 and TRAF3IP2.

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DNA extraction and genotyping

Genomic DNA samples of BD, VKH patients and healthy controls were extracted by using the QIAamp DNA Blood MIni Kit (Qiagen, Valencia, CA). Rs6540679, rs33980500, rs13210247, rs13190932 were genotyped by polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP), the primers used to amplify the target DNA sequence by PCR are shown in Table S3. Digestion products were visualized on a 4% agarose gel and stained with GoldView™ (SBS Genetech, Beijing, China). Rs12569232 (TagMan assay ID: C_26176858_10) genotypes were investigated using the TaqMan® SNP Genotyping Assay (Applied Biosystems, Foster City, CA, USA) on the Applied Biosystems 7500 Real-Time PCR system. All the operations were performed according to manufacturers’ instructions. The analysis was performed using TaqMan® Genotyper Software. Direct sequencing was also performed by the Beijing Biomed Co., Ltd (Beijing, China) using randomly selected subjects (10% of all samples) to validate the accuracy of genotyping. The genotyping success rate of the various SNPs ranged between 93.6% and 100%.

Real-time PCR

Peripheral blood mononuclear cells (PBMCs) were prepared from heparinized blood by Ficoll-Hypaque density-gradient centrifugation. Isolated PBMCs were cultured in 24-well plates containing RPMI-1640 supplemented with 10% calf serum (Greiner, Wemmel, Belgium), 2 mM L-glutamine and 100 U/ml penicillin/streptomycin (Invitrogen, Carlsbad, CA). Cells were left unstimulated or were stimulated with a cocktail of anti-CD3 (5 µg/ml, eBioscience, San Diego, CA, USA) and anti-CD28 antibodies (5 µg/ml, eBioscience, San Diego, CA, USA) to mimic


Table 1. Polymorphisms of the TRAF5 and TRAF3IP2 genes in ocular Behçet’s Disease.

| Gene     | SNPs | Genotype | BD n (%) | Control n (%) | P value | Pc value | OR (95%CI) |
|----------|------|----------|----------|---------------|---------|----------|------------|
|          |      | Allele   | n (%)    | n (%)         |         |          |            |
| TRAF5    | rs10863888 | AA       | 74 (10)  | 170 (10.8)    | 0.566   | 5.094    | 0.919 (0.689–1.226) |
|          |      | AG       | 359 (48.6) | 650 (41.3) | 0.001   | 0.009   | 1.343 (1.127–1.601) |
|          |      | GG       | 306 (41.4) | 754 (47.9) | 0.003   | 0.027   | 0.769 (0.644–0.917) |
|          |      | A        | 507 (34.3) | 990 (31.4) | 0.053   | 0.477   | 1.138 (0.998–1.298) |
|          |      | G        | 971 (65.7) | 2158 (68.6) | 0.053   | 0.477   | 0.879 (0.771–1.002) |
| rs12569232 | CC     | 9 (1.2) | 20 (1.2) | 0.893 | 8.037 | 0.947 (0.429–2.090) |
|          |      | CG       | 96 (12.6) | 304 (19.0) | 1.20×10⁻⁴ | 1.08×10⁻³ | 0.617 (0.481–0.790) |
|          |      | GG       | 655 (86.2) | 1277 (79.8) | 1.56×10⁻⁴ | 1.40×10⁻³ | 1.583 (1.246–2.011) |
|          |      | C        | 114 (7.5) | 344 (21.0) | 4.34×10⁻⁴ | 1.30×10⁻⁴ | 0.674 (0.540–0.840) |
|          |      | G        | 1406 (92.5) | 2858 (89.3) | 4.34×10⁻⁴ | 1.30×10⁻⁴ | 1.484 (1.190–1.852) |
| rs6540679 | AA     | 58 (7.4) | 81 (5.2) | 0.036 | 0.324 | 1.449 (1.022–2.053) |
|          |      | AG       | 355 (45.0) | 497 (31.9) | 4.00×10⁻¹⁰ | 3.60×10⁻⁹ | 1.750 (1.467–2.086) |
|          |      | GG       | 376 (47.7) | 982 (62.9) | 1.35×10⁻¹² | 1.22×10⁻¹¹ | 0.536 (0.451–0.637) |
|          |      | A        | 471 (29.8) | 659 (21.1) | 3.86×10⁻¹¹ | 1.16×10⁻¹⁰ | 1.589 (1.384–1.824) |
|          |      | G        | 1107 (70.2) | 2461 (78.9) | 3.86×10⁻¹¹ | 1.16×10⁻¹⁰ | 0.629 (0.548–0.722) |
| TRAF3IP2  | rs13210247 | AA       | 539 (88.4) | 1513 (95.1) | 1.79×10⁻⁸ | 1.61×10⁻⁷ | 0.391 (0.280–0.548) |
|          |      | AG       | 71 (11.6) | 76 (4.8) | 7.83×10⁻⁹ | 7.05×10⁻⁸ | 2.626 (1.873–3.682) |
|          |      | GG       | 0 (0.0) | 2 (0.1) | 0 | 0 |
|          |      | A        | 1149 (94.2) | 3102 (97.5) | 6.91×10⁻⁸ | 2.07×10⁻⁷ | 0.417 (0.301–0.579) |
|          |      | G        | 71 (5.8) | 80 (2.5) | 6.91×10⁻⁸ | 2.07×10⁻⁷ | 2.396 (1.728–3.322) |

SNP, single-nucleotide polymorphism; VKH, Vogt-Koyanagi-Harada; BD, Behçet’s disease; OR, odds ratio; CI, confidence interval; Pc, Bonferroni corrected p value which p, equaled to a p value multiplied by 9 (for genotype) or 3 (for allele).

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antigen presentation. Alternatively the cells were stimulated with LPS (1 mg/ml, Sigma-Aldrich Co. LLC), at 37°C in humidified 5% CO₂ for 72 hours at a density of 1×10⁶ cells/ml. Total RNA was extracted from the cultured PBMCs using TRIzol (Invitrogen), followed by reverse transcription using a transcriptase kit (Takara Biotechnology Co. Ltd., Dalian, China.). Real-time quantitative PCR was performed to compare the mRNA expression of TRAF3IP2 and TRAF5 gene, using the Applied Biosystem 7500 Real-time PCR System and was determined using the SYBR Green I Assay kit (Applied Biosystems) and normalized to β-actin mRNA. Relative expression levels were calculated using the 2^−ΔΔCt method. The sense and antisense primers used in this experiment are depicted in Table S4.

Cytokine Measurements

The concentration of IL-8, IL-6 and TNF-α in cell culture supernatants was measured with the human Duoset enzyme-linked immunosorbent assay (ELISA) Development kit (R&D System, Minneapolis, MN) according to the manufacturers’ instructions.

Statistical analysis

Hardy-Weinberg equilibrium was tested using the χ² test for goodness of fit and a p-value<0.05 was considered as a significant disequilibrium. The patterns of linkage disequilibrium (LD) of the tested SNPs were compared using Haploview (version 4.0, Broad Institute of MIT and Harvard, Cambridge, MA). Allele and genotype frequencies were compared between patients and controls by the χ² test or two-sided Fisher’ exact test using SPSS (version 13.0; SPSS Inc, Chicago, IL). The p values were corrected (pₐ) with the Bonferroni correction by multiplying with the number of analyses performed. For Table 1 and Table 2, pₐ equaled to a p value multiplied by 9 (for genotype analysis) or 3 (for allele analysis). A p <0.05 was considered significant.

Results

Six SNPs were successfully genotyped and confirmed to Hardy-Weinberg expectation in controls. A linkage disequilibrium analysis using Haploview software showed that the selected SNPs were not linked. Both rs33980500 and rs3190932 of the TRAF3IP2 gene were not polymorphic in Han Chinese and were therefore not analyzed further. Analysis of the clinical symptoms of our BD patients revealed six primary features, including oral ulcer (100.0%), skin lesions (70.0%), genital ulcer (57.9%), arthritis (39.1%), positive pathergy test (24.5%) and hypopyon (22.3%). The VKH patients could be subdivided according to seven clinical features, including nuchal rigidity (17.9%), headache (50.4%), scalp allergy (10.4%), tinnitus (40.5%), alopecia (39.8%), poliosis (36.6%) and vitiligo (21.5%).

Association of the TRAF5 and TRAF3IP2 gene polymorphisms with susceptibility to ocular Behçet’s disease and VKH syndrome

Significant differences between BD patients and controls were observed for four SNPs. The frequencies of the rs12569232/ TRAF5 CG genotype and C allele were significantly lower in BD patients (pₐ = 1.08×10⁻³, OR = 0.617; pᵣ = 1.30×10⁻³, OR = 0.674, respectively), whereas a higher frequency of the GG genotype was observed (pᵣ = 1.40×10⁻³, OR = 1.583) com-
pared with controls. As to rs10863888/TRAF5, an increased frequency of the AG genotype was observed in BD patients ($p_c = 0.009$, OR = 1.343), while a decreased GG frequency ($p_c = 0.027$, OR 0.769) was found. The frequency of the AG genotype and A allele of rs6540679/TRAF5 were remarkably increased ($p_c = 3.6 \times 10^{-10}$, OR = 1.750; $p_c = 1.16 \times 10^{-10}$, OR = 1.589 respectively), while the frequency of the homozygous GG genotype was markedly decreased ($p_c = 1.22 \times 10^{-11}$, OR = 0.536) in BD patients.

Although obvious differences were found for the rs10863888 TRAF5 genotypes in VKH patients compared with healthy controls, they became non-significant after Bonferroni correction. The frequency of the CG genotype and C allele of rs12569232/TRAF5 was markedly lower in VKH patients as compared to controls. The frequency of the GG genotype was much higher ($p_c = 1.33 \times 10^{-9}$, OR = 0.446; $p_c = 9.45 \times 10^{-12}$, OR = 0.442; $p_c = 5.65 \times 10^{-12}$, OR = 2.339), whereas the AA genotype and A allele of rs66540679/TRAF5 were both decreased in VKH patients ($p_c = 2.90 \times 10^{-4}$, OR = 0.411; $p_c = 2.61 \times 10^{-3}$, OR = 0.972) (Table 2).

Relationship among genotypes, gene expression at the mRNA level and downstream inflammatory factors

Since the most significant association between TRAF gene polymorphisms and uveitis was found for TRAF5, we studied the effect of the different genotypes on the expression of TRAF5 under normal or inflammatory conditions. Real-Time PCR was performed to detect mRNA expression from PBMCs obtained from healthy controls. We genotyped 79 controls for the three SNPs of TRAF5 and then used Real-Time PCR to detect their expression.
TRAF5 expression at the mRNA level without stimulation. No difference could be detected under this condition (Figure S1). Following stimulation by LPS, carriers with the GG genotype in SNP rs6540679 had a higher TRAF5 mRNA expression compared with individuals carrying the AA and AG genotype (p = 0.004) (Figure 2). No effect on TRAF5 mRNA expression was observed for rs12569232 or rs10863888 (data not shown). Furthermore no effect on TRAF5 gene expression was observed when PBMCs were stimulated by a cocktail of anti-CD3/CD28 antibodies.

The aforementioned result showed that different genotypes of rs6540679/TRAF5 could affect TRAF5 expression and therefore a further study was designed to investigate if different genotypes of rs6540679/TRAF5 could also affect the cytokine response of PBMCs following LPS stimulation. We measured the level of TNF-α, IL-6 and IL-8 (n = 22), which are important TRAF5 downstream factors [31,32,33,34,35] in PBMC cultured supernatants by ELISA. Carriers of the GG genotype showed a higher TNF-α and IL-6 secretion, compared to AA and AG carriers (p = 0.0052, and p = 0.0014 respectively) (Figure 3). No effect of the various TRAF5 rs6540679 genotypes on IL-8 production could be detected.

Discussion

In this study, we show that TRAF5 and TRAF3IP2 gene polymorphisms are associated with VKH syndrome and ocular BD in a Han Chinese population. Highest p values were observed for the association between uveitis and TRAF5, and we therefore focused on this SNP to investigate a possible functional association. This approach revealed that carriers of the GG genotype in SNP rs6540679/TRAF5 had a higher TRAF5 mRNA expression level and enhanced TNF-α and IL-6 secretion, compared to AA and AG carriers. Expression was not altered for the other TRAF5 SNPs. We did not perform a haplotype analysis since the sample size was too small.

Our finding that the G allele of TRAF5 rs6540679 had a protective effect (OR = 0.629) in BD and a risk effect (OR = 1.235) in VKH suggests that the role of these factors depends on the
context whereby they are involved during the process of inflammation. Similarly TRAF rs12569232 is most significant in VKH while rs6540679 is the most significant in BD. The functional studies performed until now have only addressed the mRNA expression and as yet no studies have addressed the effect of the gene polymorphisms on the binding characteristics of the factors and their final effect on for instance NF-kappa-B activation.

Both uveitis entities we investigated in this study differ markedly as to the causative mechanisms and the role of TRAF family members for each disease might be different. BD is considered to be caused by an aberrant inflammatory response towards certain environmental triggers whereas VKH is an autoimmune disease directed against melanocyte antigens [36,37]. How the members of the TRAF family exactly influence the inflammatory response in these two diseases remains to be elucidated.

Taken together, the results confirm earlier studies showing a role for TRAF5 in the control of inflammatory responses and the increased risk of certain polymorphisms of this gene for autoimmune or autoinflammatory diseases (Table 3) [17,18,32,33,34]. Of the TRAF5 polymorphisms included in our study, only rs10863888 was included in earlier studies on the association with RA. Associations with the TRAF5 SNPs rs6540679 and rs12569232 have not yet been reported earlier. On the other hand we did not include TRAF5 rs7514863, which showed an association with RA, because we designed six PCR primers of this SNP for PCR-RFLP, but none of their PCR product was satisfactory to be included in a following digestion step. In addition, no TaqMan® SNP Genotyping Assay is available for rs7514863.

**Table 3. TRAF family gene polymorphisms in autoimmune diseases.**

| Gene       | SNP          | Autoimmune | Race         | P value | OR (95%CI) |
|------------|--------------|------------|--------------|---------|------------|
| TRAF1 [51,52,53] | rs3761847    | SLE        | Japanese     | 0.016   | 1.36 (1.05–1.76) |
|            |              | RA         | North American & Swedish | 4 x 10^{-14} | 1.30 (1.23–1.42) |
|            | rs7021206    | RA         | Japanese     | 0.031   | 1.18 (1.01–1.39) |
| TRAF6 [54] | rs5030445    | SLE        | African American | 0.00173 | 0.68 (0.53–0.87) |
|            | rs5030437    | SLE        | African American | 0.00456 | 0.70 (0.54–0.90) |
|            | rs5030472    | SLE        | European     | 0.003342 | 0.65 (0.49–0.87) |
|            | rs5030470    | SLE        | European     | 0.000179 | 0.55 (0.40–0.76) |
| TRAF3IP2 [23,24,55,56] | rs33980500   | SLE        | Italian     | 0.021   | 1.71 |
|            | rs13210247   | psoriasis  | German       | 2.36 x 10^{-10} | * |
|            | rs13190932   | PsA        | German       | 5.76 x 10^{-7} | 2.08 (1.61–2.69) |
|            |              | BD         | Chinese      | 7.05 x 10^{-8} | 2.63 (1.87–3.68) |
|            | rs13196377   | PsA        | German       | 9.36 x 10^{-7} | 2.17 (1.66–2.88) |
|            |              | UC         | Italian      | 0.02     | 5.05 (1.12–2.83) |
|            | rs10863888   | RA         | UK Caucasian | 0.003   | 1.39 (1.11–1.75) |
| TRAF5 [18] | rs7514863    | RA         | UK Caucasian | 0.005   | 1.2 (1.06–1.36) |

*The data not stated in paper.

SLE, Systemic Lupus Erythematosus; RA, Rheumatoid Arthritis; PsA, Psoriatic Arthritis; UC, Ulcerative Colitis; CD, Crohn’s Disease.

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VKH syndrome and BD are two of the most common uveitis entities in China [38,39]. Both diseases account for approximately one-third of the uveitis prevalence in our country [39]. The genetic background of both diseases is not yet completely clear. In recent years, we have reported on the association of various immune response related genes with the susceptibility to both BD and VKH including STAT4, STAT3, JAK2, CD40 [7,8,9,40]. In the present study, the selection of the candidate locus/gene was primarily based on GWAS data in patients with psoriasis, psoriatic arthritis (PsA) [24] and psoriasis vulgaris (PsV) [23], previously described associations for inflammatory diseases [17,25,41,42], as well as the involvement of the gene products in the control of the immune and inflammatory response [43,44,45].

TRAF5 and Act1, encoded by the TRAF3IP2 gene, have been implicated in the control of immune and inflammatory responses [44,45,46]. Both proteins act as regulators of NF-kappa-B activation. During recent years, evidence is mounting that they are involved in the pathogenesis of various autoimmune and autoinflammatory disorders [17,32,33,42,47]. In view of the fact
that inflammation can induce Act1-TRAF2-TRAF5 complex formation, and in turn, can stimulate a downstream stress reaction, we aimed to assess whether these two genes were involved in the susceptibility to develop BD and VKH syndrome. The polymorphisms we used were based on earlier reports showing significant associations with RA, psoriatic arthritis, psoriasis and Inflammatory Bowel Disease [10,23,24,25] and included three SNPs (rs6540679, rs12569232, rs10863888) of TRAF5 and three SNPs (rs13210247, rs33980509, rs13190932) of TRAF3IP2 (Table 3).

To ensure the validity of our data we took the following measures. First, the sample sizes of VKH patients, BD patients and normal controls were large enough to ensure an association analysis, and our sample was larger than previous reports [7,8,10,23,32,48,49,50]. Second, the controls were strictly selected according to their birth origin to obtain a comparable immuno-genetic background. Additionally, we randomly selected 10% of the samples to undergo direct sequencing to validate the results of genotyping by PCR-RFLP.

Although our results suggested that TRAF5 and TRAF3IP2 are associated with the development of BD and VKH syndrome, it is still unknown how these SNPs exert their roles in these two diseases. The complex and redundant function of the members of the TRAF family and the context they are involved in may dictate their possible divergent roles in immune mediated diseases.

Taken together, our study, for the first time, provides evidence for a role of TRAF5 and TRAF3IP2 polymorphisms in the development of BD and VKH syndrome.

References

1. Yang P, Fang W, Meng Q, Ren Y, Xing L, et al. (2008) Clinical features of Chinese patients with Behc¸et’s disease. Ophthalmology 115: 312–318. e314.
2. Yang P, Ren Y, Li R, Fang W, Meng Q, et al. (2007) Clinical characteristics of Vogt-Koyanagi-Harada syndrome in Chinese patients. Ophthalmology 114: 696–614. e603.
3. Zhang Z, Peng J, Hou X, Dong Y (2006) Clinical manifestations of Behc¸et’s disease in Chinese patients. APLAR Journal of Rheumatology 9: 244–247.
4. Ideguchi H, Suda A, Takeno M, Ueda A, Ohno S, et al. (2011) Behc¸et disease: evolution of clinical manifestations. Medicine (Baltimore) 90: 125–132.
5. Keino H, Okada AA (2007) Behc¸et’s disease: global epidemiology of an Old Silk Road disease. Br J Ophthalmol 91: 1573–1574.
6. Meguro A, Inoko H, Ota M, Katsuyama Y, Oka A, et al. (2010) Genetics of Behc¸et disease inside and outside the MHC. Annals of the rheumatic diseases 69: 747–754.
7. Chen F, Hou S, Jiang Z, Chen Y, Kijlstra A, et al. (2012) CD40 gene polymorphisms confer risk of Behc¸et’s disease but not of Vogt-Koyanagi-Harada syndrome in a Han Chinese population. Rheumatology (Oxford) 51: 47–51.
8. Hou S, Yang Z, Du J, Jiang Z, Shu Q, et al. (2012) Genome-wide association study identifies susceptible locus in STAT4 for Behc¸et’s disease in Han Chinese. Arthritis Rheum.
9. Hu K, Hou S, Jiang Z, Kijlstra A, Yang P (2012) JKAR and STAT3 polymorphisms in a Han Chinese population with Behc¸et’s disease. Invest Ophthalmol Vis Sci 53: 530–541.
10. Read RW, Holland GN, Rao NA, Tabbara KE, Ohno S, et al. (2001) Revised diagnostic criteria for Vogt-Koyanagi-Harada disease: report of an international committee on nomenclature. American journal of ophthalmology 131: 647–652.
11. Davis J, Mittal K, Freidlin V, Mellov S, Optican D, et al. (1990) HLA associations and ancestry in Vogt-Koyanagi-Harada disease and sympathia- ophthalias. Ophthalmology 97: 1137.
12. Zhao M, Jiang Y, Abrahams IW (1991) Association of HLA antigens with Vogt-Koyanagi-Harada syndrome in a Chinese population. Archives of ophthalmology 109: 360–369.
13. Jiang Z, Yang P, Hou S, Li F, Zhou H (2010) Polymorphisms of IL23R and Vogt-Koyanagi-Harada syndrome in a Chinese Han population. Human Immunology 71: 414–417.
14. Shu Q, Yang P, Hou S, Li F, Chen Y, et al. (2010) Interleukin-17 gene polymorphism is associated with Vogt-Koyanagi-Harada syndrome but not with Behc¸et’s disease in a Chinese Han population. Human Immunology 71: 988–991.
15. Chou M, Yang P, Hu R, Hou S, Li F, et al. (2011) Elevated serum osteopontin levels and genetic polymorphisms of osteopontin are associated with Vogt-Koyanagi-Harada disease. Invest Ophthalmol Vis Sci 52: 7084–7089.
16. Au PY, Yeh WC (2007) Physiological roles and mechanisms of signaling by TRAF2 and TRAF5. Adv Exp Med Biol 597: 32–47.
17. Bulek K, Liu C, SwaiDasi S, Wang L, Page RC, et al. (2011) The inductive kinase IKK is required for IL-17-dependent signaling associated with neutrophilia and pulmonary inflammation. Nat Immunol 12: 834–832.
18. Potter C, Eyre S, Cope A, Worthington J, Barton A (2003) Investigation of association between the TRAF family genes and RA susceptibility. Ann Rheum Dis 66: 1322–1326.
19. Ma AI (2012) The ubiquitous nature of IL-17. Nat Immunol 13: 1034–1035.
20. Doyle MS, Collins ES, Fitzgerald OM, Pennington SR (2012) New insight into the functions of the interleukin-17 receptor adaptor protein Act1 in psoriatic arthritis. Arthritis Res Ther 14: 296.
21. Sun D, Novosny M, Bulek K, Liu C, Li X, et al. (2011) Treatment with IL-17 prolongs the half-life of chemokine CXCL1 mRNA via the adaptor TRAF5 and the splicing-regulatory factor NF2 (ASF). Nature immunology 12: 853–860.
22. Rassi IM, Juntla CM, Fachin AL, Sandrin-Guercio P, Mello S, et al. (2000) Gene expression profiles stratified according to type 1 diabetes mellitus susceptibility regions. Annals of the New York Academy of Sciences 1150: 282–289.
23. Ellingshaus E, Ellingshaus D, Stuart PE, Nair RP, Debrus S, et al. (2010) Genome-wide association study identifies a psoriasis susceptibility locus at TRAF3IP2. Nat Genet 42: 991–995.
24. Huflinier U, Uebe S, Elciki AB, Bowes J, Giardina E, et al. (2010) Common variants at TRAF3IP2 are associated with susceptibility to psoriatic arthritis and psoriasis. Nat Genet 42: 996–999.
25. Capacci C, Biancone L, Di Fusco D, Ranieri M, Condino G, et al. (2012) TRAF3IP2 gene is associated with cutaneous extraintestinal manifestations in Inflammatory Bowel Disease. Journal of Crohn’s and Colitis.
26. Berghold R, Brousson C, Paljea A, Berchold LA, Floydl T, et al. (2012) Identification of novel type 1 diabetes candidate genes by integrating genome-wide association data, protein-protein interactions, and human pancreatic islet gene expression. Diabetes 61: 954–962.
27. O’Rielly DD, Rahman P (2011) Genes of susceptibility and treatment response in psoriatic arthritis. Nat Rev Rheumatol 7: 710–732.
28. Chi W, Zhu X, Yang P, Liu X, Liu X, et al. (2008) Upregulated IL-23 and IL-17 in Behc¸et patients with active uveitis. Investigative ophthalmology & visual science 49: 3058–3064.
29. Chi W, Yang P, Li B, Wu C, Jin H, et al. (2007) IL-23 promotes CD4+ T cells to produce IL-17 in Vogt-Koyanagi-Harada disease. Journal of allergy and clinical immunology 119: 1218–1224.

30. International Study Group for Behcet’s Disease (1990) Criteria for diagnosis of Behcet’s disease. Lancet 335: 1078–1080.

31. Kraus ZJ, Nakano H, Bishop GA (2009) TRAF5 is a critical mediator of in vitro signals and in vivo functions of LMP1, the viral oncopgenic mimic of CD40. Proceedings of the National Academy of Sciences 106: 17140–17145.

32. Cerhan JR, Liu-Mares W, Frederickson ZS, Novak AJ, Cunningham JM, et al. (2012) Genetic and functional effects of membrane metalloendopeptidase on diabetic nephropathy development. Am J Nephrol 34: 483–490.

33. Snell LM, Liu GH, McPherson AJ, Moraes TJ, Watts TH (2011) T-cell intrinsic effects of GITR and 4-1BB during viral infection and cancer immunotherapy. ImmunoL Rev 244: 197–217.

34. Zhang D, Gu T, Forsberg E, Efendic S, Brismar K, et al. (2011) Genetic variation in tumor necrosis factor and the nuclear factor-kappaB canonical pathway and risk of non-Hodgkin’s lymphoma. Cancer Epidemiol Biomarkers Prev 17: 3161–3169.

35. Zirlik A, Bavendiek U, Libby P, MacFarlane L, Gerdes N, et al. (2007) TRAF-1, -2, -3, -5, and -6 are induced in atherosclerotic plaques and differentially mediate proinflammatory functions of CD40L in endothelial cells. Arterioscler Thromb Vasc Biol 27: 1101–1107.

36. Sugita S, Takase H, Taguchi C, Imai Y, Kamoi K, et al. (2006) Ocular infiltrating CD4+ T cells from patients with Vogt-Koyanagi-Harada disease recognize human melanocyte antigens. Invest Ophthalmol Vis Sci 47: 2547–2554.

37. Ohno S (1981) Immunological aspects of Behc¸et’s disease. Trans Ophthalmol Soc UK 101: 335–341.

38. Fang W, Yang P (2008) Ocular inflammation in dermatitis herpetiformis skin lesions. Curr Eye Res 33: 517–523.

39. Yang P, Zhang Z, Zhou H, Li B, Huang X, et al. (2005) Clinical patterns and characteristics of uveitis in a tertiary center for uveitis in China. Curr Eye Res 30: 943–948.

40. Hu K, Yang P, Jiang Z, Hou S, Du L, et al. (2010) STAT4 polymorphism in a Chinese Han population with Vogt-Koyanagi-Harada syndrome and Behcet’s disease. Human Immunology 71: 723–726.

41. Jørgensen TN, Giltiay NV, Johnson A, Li X (2009) The role of Act1 in the control of autoimmunity. The Epigenetics of Autoimmune Diseases: 55–74.

42. Dolcino M, Cozzani E, Riva S, Parodi A, Tinazzi E, et al. (2012) Gene expression profiling in dermatitis herpetiformis skin lesions. Clin Dev Immunol 2012: 198956.

43. Kitamei H, Isabuchi K, Namba K, Yoshida K, Yanagawa Y, et al. (2006) Amelioration of experimental autoimmune uveoretinitis (EAU) with an inhibitor of nuclear factor-κB (NF-κB), pyrrolidine dithiocarbamate. Journal of leukocyte biology 79: 1193–1201.

44. Nicroli L, Naumni C, Benucci M, Chãndamo D, Cassara E, et al. (2007) Long-term efficacy of infliximab in refractory posterior uveitis of Behcet’s disease: a 24-month follow-up study. Rheumatology (Oxford) 46: 1161–1164.

45. Nishimoto K, Kochi Y, Ikari K, Yamamoto K, Suzuki A, et al. (2010) Association study of TRAF1-C5 polymorphisms with susceptibility to rheumatoid arthritis and systemic lupus erythematosus in Japanese. Annals of the rheumatic diseases 69: 368–373.

46. Ploeg R, Swielitat M, Padyukov L, Lee AT, Remmers EF, et al. (2007) TRAF1–C5 as a risk locus for rheumatoid arthritis—a genomewide study. New England Journal of Medicine 357: 1199–1209.

47. Han TU, Bang SY, Kang C, Bae SC (2009) TRAF1 polymorphisms associated with rheumatoid arthritis susceptibility in Asians and in Caucasians. Arthritis & Rheumatism 60: 2577–2584.

48. Perricone C, Ciccacci C, Ceccarelli F, Di Fusco D, Spadulli FR, et al. (2013) TRAF3IP2 gene and systemic lupus erythematosus: association with disease susceptibility and pericarditis development. Immunogenetics 65: 703–709.

49. Kitamei H, Isabuchi K, Namba K, Yoshida K, Yanagawa Y, et al. (2006) Amelioration of experimental autoimmune uveoretinitis (EAU) with an inhibitor of nuclear factor-κB (NF-κB), pyrrolidine dithiocarbamate. Journal of leukocyte biology 79: 1193–1201.

50. Jiang Z, Yang P, Hou S, Du L, Xie L, et al. (2010) IL-23R gene confers susceptibility to Behcet’s disease in a Chinese Han population. Ann Rheum Dis 69: 1323–1328.

51. Nishimoto K, Kochi Y, Ikari K, Yamamoto K, Suzuki A, et al. (2010) Association study of TRAF1-C5 polymorphisms with susceptibility to rheumatoid arthritis and systemic lupus erythematosus in Japanese. Annals of the rheumatic diseases 69: 368–373.

52. Plenge RM, Seielstad M, Padyukov L, Lee AT, Remmers EF, et al. (2007) TRAF1–C5 as a risk locus for rheumatoid arthritis—a genomewide study. New England Journal of Medicine 357: 1199–1209.

53. Han TU, Bang SY, Kang C, Bae SC (2009) TRAF1 polymorphisms associated with rheumatoid arthritis susceptibility in Asians and in Caucasians. Arthritis & Rheumatism 60: 2577–2584.

54. Perricone C, Ciccacci C, Ceccarelli F, Di Fusco D, Spadulli FR, et al. (2013) TRAF3IP2 gene and systemic lupus erythematosus: association with disease susceptibility and pericarditis development. Immunogenetics 65: 703–709.

55. Perricone C, Ciccacci C, Ceccarelli F, Di Fusco D, Spadulli FR, et al. (2013) TRAF3IP2 gene and systemic lupus erythematosus: association with disease susceptibility and pericarditis development. Immunogenetics 65: 703–709.

56. Perricone C, Ciccacci C, Ceccarelli F, Di Fusco D, Spadulli FR, et al. (2013) TRAF3IP2 gene and systemic lupus erythematosus: association with disease susceptibility and pericarditis development. Immunogenetics 65: 703–709.