PPAR-γ Gene Polymorphisms and Psoriatic Arthritis

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ABSTRACT. ***Objective.*** Peroxisome proliferator-activated receptor-γ (PPAR-γ) activation has been shown to play a role in suppressing angiogenesis and inflammation, both important pathological features of psoriatic arthritis (PsA). Given the potential physiological role for PPAR-γ in PsA, we examined known coding polymorphisms in the *PPAR-γ* gene in a Caucasian population.

Methods. PsA was diagnosed as an inflammatory arthritis in patients with psoriasis, in the absence of other etiologies for inflammatory arthritis. Control subjects were ascertained from the same population and were all Caucasian. DNA samples were genotyped for 4 *PPAR-γ* variants by time-of–flight mass spectrometry using the Sequenom platform. All 4 single-nucleotide polymorphisms (SNP) were previously-reported coding variations, 3 of which caused an amino acid change: Pro12Ala (rs1801282), Pro40Ala (rs1805192), and Pro115Gln (rs1800571); the fourth SNP, C161T (rs3856806), was synonymous. All primers were designed using Sequenom SpectroDesigner software, and scanned using a mass spectrometry workstation.

Results. Of the 4 SNP examined, Pro40Ala and Pro115Gln were found to be nonpolymorphic in our population. Minor allele frequency for patients with PsA and controls for Pro12Ala (G) were 9.0% vs 13.8% (p = 0.017) and for C161T (T) 10.7% vs 12.0% (p = 0.56), respectively. All genotypes satisfied Hardy-Weinberg equilibrium.

Conclusion. An association between PsA and a known coding SNP of the *PPAR-γ* gene was observed in our Caucasian population. Further studies are now warranted for validation of our findings in an independent cohort. (First Release June 15 2006; J Rheumatol 2006;33:1631–3)

Key Indexing Terms:
PSORIATIC ARTHRITIS PEROXISOME PROLIFERATOR-ACTIVATED RECEPTOR-γ
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Recently, Bongartz, *et al* reported on the use of a novel class of insulin-sensitizing drugs (thiazolidinediones) in the treatment of active psoriatic arthritis (PsA)[1]. Specifically, in this open label trial, 6 out of 10 patients with active PsA improved sufficiently to satisfy the Psoriatic Arthritis Response Criterion (PsARC) after pioglitazone was instituted. Pioglitazone was developed for the treatment of type II diabetes mellitus (DM) and acts as a ligand for the peroxisome proliferator-activated receptor-γ (PPAR-γ). The *PPAR-γ* gene is expressed in many human tissues and its activation has been shown to play a role in suppressing both angiogenesis and inflammation[2,3]. Specifically, PPAR-γ activation can decrease proinflammatory cytokine expression and suppress neoangiogenesis in models of inflammatory disease[2].

PPAR-γ is a potential candidate gene for susceptibility in PsA, as Mingrone, *et al* demonstrated that abnormal lipid metabolism often coexists with glucose intolerance in patients with psoriasis[4]. Epidemiological studies have noted increased prevalence of type I DM in patients with psoriasis[5]. Also, the Pro12Ala single-nucleotide polymorphism (SNP) of *PPAR-γ* has been associated with type II DM[6] and biomarkers of systemic inflammation[7]. Finally, several animal models have shown that peroxisome PPAR-γ agonists significantly reduced the progression of experimentally induced osteoarthritis in guinea pigs[8] and clinical disease activity scores as well as histological scores of joint destruction in a mouse model of collagen-induced arthritis[9].

Given the potential physiological role of PPAR-γ activation and the apparent success of PPAR-γ-agonist in the treatment of active PsA and in animal models of arthritis, we examined 4 known coding polymorphisms in the *PPAR-γ* gene in our PsA population: Pro12Ala (rs1801282), Pro40Ala (rs1805192), Pro115Gln (rs1800571), and C161T (rs3856806). The PPAR-γ Pro12Ala SNP has been associated with reduced transcriptional and receptor activity of PPAR-γ[10], and the presence of the Ala isoform has been linked to higher insulin sensitivity and both higher and lower body mass index[11,12]; whereas the Pro12Pro isoform has been associated with higher levels of biomarkers of inflammation as well as shorter survival times in patients with endstage renal disease[7]. The Pro40Ala, Pro115Gln, and C161T polymorphisms have all been inconsistently associated with diabetes, insulin sensitivity, and obesity[10,12,13].

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MATERIALS AND METHODS

PsA was diagnosed as an inflammatory arthritis in patients with psoriasis in the absence of other etiologies for inflammatory arthritis. All PsA probands were Caucasians from the Newfoundland population, and were unrelated to each other. Information was collected on a standardized protocol for age at onset of psoriasis, PsA, and disease pattern. Control subjects were healthy volunteers from Newfoundland who responded to an advertisement seeking controls for genetic studies. As the controls were examined we verified that they did not have PsA or a history of psoriasis or inflammatory arthritis. This study was approved by the local ethics committee of Memorial University of Newfoundland. Informed consent was obtained from all subjects.

The population of the island of Newfoundland consists mainly of descendants of English and Irish settlers who arrived in the 17th and 18th centuries. The geographic and social isolation of this island has ensured very little inward migration for several hundred years. The population (530,000 individuals; Statistics Canada 2001) has grown mostly through internal expansion, with a relatively homogeneous genetic background, ideal for the study of complex disease.

Whole-blood samples were obtained from PsA probands and controls. DNA was extracted using the Promega Wizard Genomic DNA purification kit. Detection of SNP was performed by analysis of primer extension products generated from previously amplified genomic DNA using a Sequenom chip-based MALDI-TOF mass spectrometry platform. In brief, polymerase chain reactions (PCR) and extension reactions were designed using MassArray software, and were carried out using 2.5 ng of template DNA. Unincorporated nucleotides in the PCR product were deactivated using shrimp alkaline phosphatase. Amplification of the SNP site was carried out using the MassExtend primer and specific d/ddNTP termination mixes, which were also determined using MassArray software. The primer extension products were then cleaned and spotted onto a SpectroChip. The chips were scanned using a mass spectrometry workstation (Bruker), and the resulting spectra were analyzed and genotypes were determined using Sequenom SpectroTyper-RT software.

Samples were genotyped for all 4 *PPAR-γ* variants by time-of–flight mass spectrometry using the Sequenom platform. All 4 SNP were previously-reported coding variations, 3 of which caused an amino acid change: Pro12Ala, Pro40Ala, and Pro115Gln; whereas the fourth SNP, C161T, was synonymous, resulting in no amino acid change in the final protein. All primers were designed using Sequenom SpectroDesigner software, scanned using a mass spectrometry workstation (Bruker), and analyzed using Sequenom SpectroTyper-RT software. Statistical analysis of all PPAR-γ variants was performed using chi-square tests.

RESULTS

Of the 4 *PPAR-γ* SNP examined, 2 (Pro40Ala and Pro115Gln) were found to be nonpolymorphic in our population. For the Pro12Ala (rs1801282) SNP, an association was noted for the minor allele between PsA cases and controls (9.0% vs 13.8%, respectively; p = 0.017, OR 0.62, 95% CI 0.45–0.93), and this association remained significant (p = 0.034) in multiple testing using Bonferroni's correction (Table 1). Meanwhile, no association was noted for the C161T (rs3856806) SNP (10.7% vs 12.0%, respectively; p = 0.56). No haplotype associations were noted, and there was no correlation between the genotypes and age at onset of psoriasis, age at onset of PsA, or disease pattern.

Table 1. Genotype frequencies in PPAR-γ in PsA.

PPAR-γ	Genotype	PsA Patients, N = 251 (%)	Controls, N = 235 (%)	p
Pro12Ala	CC	208 (82.9)	177 (75.3)	
rs1801282	GC	41 (16.3)	51 (21.7)	
	GG	2 (0.8)	7 (3.0)	
G allele frequency, %		9.0	13.8	0.017
		N = 256		
C1431T	CC	207 (80.9)	185 (79.4)	
rs3856806	TC	43 (16.8)	40 (17.2)	
	TT	6 (2.3)	8 (3.4)	
T allele frequency, %		10.7	12.0	0.56

DISCUSSION

In our study a modest association was noted for the Pro12Ala (rs1801282) SNP, which results in an amino acid change from proline to alanine. To our knowledge, cases and controls were not related to each other. Even though selection of cases and controls was not limited to one geographic region, it is conceivable that cryptic relatedness exists in the Newfoundland population, resulting in a selection bias.

Interestingly, a previous study noted no association between *PPAR-γ* polymorphisms and uncomplicated psoriasis[14]. This would appear to suggest that the Pro12Ala variant of the *PPAR-γ* gene confers susceptibility specifically to PsA rather than psoriasis. However, we would use caution in interpreting this result, as 2 separate populations were studied in the psoriasis and PsA study. Further investigation of this gene from patients with PsA and psoriasis from our population and other independent populations is warranted.

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