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Genetic Markers Linked to Rheumatoid Arthritis Are also Strongly Associated with Articular Manifestations in Ulcerative Colitis Patients

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ABSTRACT: Ulcerative colitis is often accompanied by the development of extraintestinal, mainly articular, manifestations. Genetic differences could be underlying that clinical heterogeneity. We performed a case–control study to determine whether TNFab microsatellites or HLA-DR alleles were associated with the development of articular manifestations in patients with ulcerative colitis. With that aim, a total of 84 ulcerative colitis patients with articular manifestations and 172 without them were genotyped for TNFab microsatellites and HLA-DR. A healthy control sample ($n = 595$) was also included for comparative purposes. Haplotypes were inferred with the Arlequin software. The influence of HLA-DRB1*0103 and HLA-B27, factors previously known to be associated with extraintestinal manifestations, was specifically addressed. We observed that TNFa6b5 minihaplotype increases the susceptibility to developing articular manifestations in ulcerative colitis patients ($p = 0.003$, OR = 2.39). The locus HLA-DR does not appear to be involved in these extraintestinal manifestations by itself; however, the frequency of subjects carrying TNFa6b5 in combination with DR1, DR7, or DR11 is very significantly increased in patients with articular manifestations ($p = 3.9\times10^{-8}$). The associations found were independent of DRB1*0103 and HLA-B27. Thus, it seems that the development of articular manifestations in ulcerative colitis patients appears to be influenced by some genetic factor(s) present in some major histocompatibility complex haplotypes. *Human Immunology 67, 324–330 (2006).* © American Society for Histocompatibility and Immunogenetics, 2006. Published by Elsevier Inc.

KEYWORDS: HLA; TNF; ulcerative colitis; articular manifestations; rheumatoid arthritis

ABBREVIATIONS

HLA	human leukocyte antigen
IBD	inflammatory bowel disease
LTA	lymphotoxin alpha
MHC	major histocompatibility complex
MICA	MHC class I polypeptide-related sequence A
OR	odds ratio
TNF	tumor necrosis factor
UC	ulcerative colitis
RA	rheumatoid arthritis

INTRODUCTION

Articular manifestations are the most common extraintestinal manifestations in ulcerative colitis (UC), ranging from peripheral arthritis [1] to spondylitis and sacroiliitis [2]. Despite the clear definition of the arthropathies in inflammatory bowel disease (IBD), there are important discrepancies related to their prevalence. The prevalence of peripheral arthritis ranges from 5 to 20% but this range results mainly from the difficulty of distinguishing between peripheral arthritis and arthralgias. Axial arthropathies occur in 3 to 5% of patients, although frequencies as high as 25% have also been reported [3]. It has been postulated that the genetic constitution of the patient partially determines the clinical

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manifestations of the disease, but specific genetic determinants contributing to specific phenotypes remain poorly elucidated [4].

Extraintestinal manifestations (cutaneous, ocular, articular, hepatobiliary, etc. [5]) have occasionally been grouped together when genetic factors have been sought [6], although there are reasons to suspect genetic heterogeneity. We reasoned that each group of patients showing the same type of extraintestinal manifestation might constitute a genetically more homogeneous group. Thus, some alleles have been associated with specific complications of the disease. In particular, the rare MHC class II allele HLA-DRB1*0103 [7], which is also associated with the severity of the disease as measured by intractability and need of surgery [8, 9], has been associated with arthritic manifestations. The HLA-B27 allele has also been repeatedly associated with spondylitis- and uveitis-like complications [7, 10].

In addition to HLA-presenting alleles, the MHC contains a plethora of genes related to the immune and inflammatory responses, which are good candidates to be relevant to the pathogenesis of distinct clinical manifestations. Several of the most obvious inflammation-related genes are located in the TNF/LTA gene region or in its vicinity. The TNFab microsatellites, located in that region, are very informative because of their extensive polymorphism and because they occasionally characterize specific ancestral extended haplotypes in combination with DRB1 alleles. Moreover, some TNFab alleles are related, by means of their strong linkage disequilibrium with promoter single nucleotide polymorphisms, to the transcription rate of the TNFA gene [11]. Thus, the study of TNFab microsatellites seemed an *a priori* interesting choice because of their double role as haplotype "tag" and as surrogate marker of TNF promoter polymorphisms. However, the main attraction of these markers was the fact that they had already been associated with the major autoimmune disorder involving joints, rheumatoid arthritis, a disease linked to TNFa6b5 in European populations [12, 13].

Earlier studies described several associations between TNFab alleles and susceptibility to autoimmune diseases [14], although some of them most likely result from linkage disequilibrium with other loci at the MHC. Additionally, several TNFab minihaplotypes have been related to clinically defined subgroups. Susceptibility to Crohn's disease has been associated with TNFa2b1 [15], but for ulcerative colitis the information available is scarce [16], perhaps because of the absence of positive results (publication bias).

Since cytokines such as TNFα and other immune-related proteins whose genes are located in the central portion of the MHC are not antigen specific as are the typical HLA class II and class I alleles, it would not be surprising to find the same TNFab alleles associated with different pathologies. For instance, the TNFa2b3 minihaplotype and its tightly linked TNF-308A promoter allele are associated with, among other diseases, diabetes, lupus, and myasthenia gravis. As a matter of fact, it has been postulated that the proinflammatory nature of the DR3-B8 extended HLA haplotype might result from in part the high production of TNF marked by that promoter allele. Similarly, taking into account that articular manifestations are sometimes present when a (otherwise variable in origin) systemic activation of the immune system exists, it could be argued that some of the non-antigen-specific genetic determinants modulating the involvement of the joint might be shared between distinct pathological entities. In the present study our primary aim was to analyze the relationship between the TNFab alleles and the presence of articular manifestations in ulcerative colitis in Spanish patients, adding information provided by the DRB1 locus and trying to elucidate which genetic combinations influence those manifestations. Specifically, due to the previously found association between minihaplotype TNFa6b5 and rheumatoid arthritis [12, 13], we wanted to test the hypothesis that shared genetic elements might underlie common inflammatory processes resulting in articular affectation.

MATERIALS AND METHODS

Subjects

A total of 256 Spanish Caucasian patients with ulcerative colitis from the Madrid region were studied and classified into two separate groups according to the presence or not of articular manifestations: 84 (49% women) and 172 (36% women), respectively. Diagnosis of UC was based on standard clinical, radiologic, endoscopic, and histologic criteria. The median duration of follow-up was 9 years (mean 11, range 2–41). Patients thought to present articular manifestations were referred to the rheumatologist to confirm the diagnosis and exclude rheumatoid arthritis. The rheumatological disorders associated with UC found in our patients were peripheral arthropathies (arthralgias, pauciarticular or type I arthritis and polyarticular or type II arthritis, 26%), axial arthropathies (including spondylitis and sacroiliitis, 4%), and both (2%). Arthralgias and arthritis were included when they were flare dependent or when they were a specific UC complication as assessed by the rheumatologist. A group of 595 ethnically matched healthy controls from the same region was also included in the study. All the patients and controls were recruited from a single Spanish center after obtaining the patient's informed consent. Patients and controls were specifically asked about their ancestry to exclude non-Spanish de-