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IMMUNOLOGICAL FEATURES OF HLA-B27 ANTERIOR UVEITIS

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Abstract

Analysis of the immunological features of anterior uveitis (AU) revealed a dichotomy of abnormalities defined in terms of the HLA-B27 status of the patient. HLA-B27-positive AU was characterised by the occurrence of iris autoantibodies and an absolute T cell lymphopenia during active disease which returned to normal with recovery. This phenomenon was not observed in HLA-B27-negative AU or in controls and could not be attributed to antilymphocyte antibodies as these were not detected. Furthermore, there were no changes in T-cell subsets (helper and suppressor T lymphocytes). Compared with HLA-B27-positive AU patients, the HLA-B27-negative group demonstrated elevated IgE levels and increased prevalence of smooth muscle autoantibodies.

Key words: anterior uveitis, HLA-B27, immunological abnormalities, T lymphocytes, autoantibodies.

INTRODUCTION

Despite extensive investigation the aetiology and pathogenesis of most cases of anterior uveitis (AU) remain undetermined. The recent description of the association of the HLA-B27 antigen with AU[1] has renewed interest in this previously enigmatic disease and raised the possibility of an underlying immune pathogenesis. The value of immunological markers in classifying and understanding disease mechanisms is well illustrated in patients with the pauciarticular form of juvenile polyarthritis who develop a chronic AU characterised by the presence of antinuclear antibodies and HLA-DR5. Such knowledge not only has theoretical value, but also allows the recognition of children at risk of developing AU, and has prognostic significance.

Byrom *et al.*[2] described a T-cell lymphopenia during acute AU in HLA-B27-positive patients and attributed this to a presumed viral infection. The possibility of an infective aetiology is supported by a number of other studies[3,4] including our own, in which we demonstrated a significant association of a cellular immune response to *Chlamydia trachomatis* in patients with HLA-B27-positive AU.[5]

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TABLE 5

Mean Number and Standard Deviation of T-lymphocyte Subsets in Forty-one Patients with Anterior Uveitis

T Lymphocytes	Patients with anterior uveitis				Controls	
	Active uveitis		In remission			
	HLA-B27 positive ($n = 19$)	HLA-B27 negative ($n = 22$)	HLA-B27 positive ($n = 19$)	HLA-B27 negative ($n = 22$)	HLA-B27 positive ($n = 3$)	HLA-B27 negative ($n = 15$)
T cells	922 ± 70	1970 ± 310†	1215 ± 171*	2324 ± 516	2486 ± 612	2179 ± 392
Helper T cells	535 ± 52	1024 ± 135	892 ± 152	1231 ± 342	1566 ± 418	1176 ± 274
Suppressor T cells	286 ± 42	453 ± 210	514 ± 232	767 ± 287	596 ± 378	610 ± 237
Helper:Suppressor ratio	0.56 ± 0.18	0.44 ± 0.16	0.57 ± 0.22	0.62 ± 0.17	0.38 ± 0.16	0.52 ± 0.18

AU = anterior uveitis.
* $P < 0.01$ (HLA-B27-positive AU in remission vs HLA-B27-positive with active AU).
† $P < 0.01$ (HLA-B27-negative with active AU vs HLA-B27-positive with active AU).

suppressor T cells. Aberrations in this ratio are associated with a number of autoimmune diseases such as SLE;[9] however, our results indicated no such abnormalities in HLA-B27-positive AU. The reason for the T-cell lymphopenia is unknown; however, the possibility of an infective aetiology remains attractive and is supported by our recent observation on the association of chlamydial infection in the HLA-B27-positive group of patients.[5]

HLA-B27-negative AU, by contrast, was characterised by abnormalities in humoral immunity including raised IgE levels and smooth muscle autoantibodies. Elevated levels of IgE have previously been reported in AU;[4] however, previous studies did not relate this observation to the HLA-B27 phenotype of the patients. Such patients had idiopathic AU and did not have an increased incidence of atopy, parasitic infestation or eosinophilia. The increased smooth muscle autoantibodies in this group suggest another pathogenic mechanism which may involve a myositis of uveal smooth muscle. Alternatively, such antibodies may be an effect and not a cause of AU, an observation supported by the fact that HLA-B27-negative AU is characterised by a more prolonged and severe iris inflammation (probably involving iris smooth muscle) than that seen in HLA-B27-positive AU, which in contrast was characterised by an increased number and titre of iris autoantibodies.

Immune complexes have been extensively investigated in the sera and aqueous humor in AU,[18] and although the results of the present study failed to reveal increased levels of immune complexes, we have previously reported, using a more extensive battery of immune-complex assays, that such complexes are increased in the HLA-B27-negative subgroup.[5] Despite the lack of direct histological evidence for immune-complex deposition in man there is strong circumstantial and experimental evidence[19] that such complexes may play a pathogenic role in certain patients with this disease.

In summary, we hypothesise that the T-cell lymphopenia of HLA-B27-positive AU reflects an immune response to an infective agent, while HLA-B27-negative AU is a heterogenous disease group with a number of possible immune mechanisms including allergy, myositis and immune complex deposition.

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TABLE 1

Aetiology of Anterior Uveitis in Forty-one Patients

Aetiology	Number of patients	
	HLA-B27 positive ($n = 19$)	HLA-B27 negative ($n = 22$)
Idiopathic	12	19
Ankylosing spondylitis	4	0
Reiter's syndrome	3	0
Behcet's syndrome	0	1
Sarcoidosis	0	1
Herpes simplex virus	0	1

Previous studies have revealed a number of immunological abnormalities associated with AU including increased levels of IgE,[6] immune complexes[7,8] and autoantibodies.[6,9] Additionally, recent studies in other autoimmune diseases have described changes in T-lymphocyte numbers and subsets (helper and suppressor T lymphocytes) which may reflect abnormalities in immune regulation. In systemic lupus erythematosus (SLE)[10] a deficiency of suppressor T cells may be of fundamental importance in allowing B-cell hyperactivity with autoantibody production and hypergammaglobulinaemia.

In the present study the relationship between the HLA-B27 antigen and the immunological features of AU have been examined. Our results reveal a dichotomy of abnormalities in AU defined in terms of the HLA-B27 phenotype and may reflect pathogenic immune mechanisms involved in disease production.

METHODS

Patients and Controls

Forty-one consecutive AU patients referred to the Uveitis Research Clinic at Sydney Eye Hospital were investigated as part of an ongoing study. Normal healthy laboratory personnel served as controls. All patients were seen by an ophthalmologist and a physician, and investigation was undertaken on the basis of a careful history and physical examination. All patients were extensively investigated for possible immune abnormalities as outlined below.

Methods

HLA typing for the B27 antigen was performed by the N.S.W. Red Cross Blood Transfusion Service using the two-phase NIH microlymphocytotoxicity assay.[11] Laboratory investigations included: full blood count; erythrocyte sedimentation rate (ESR); serum levels of urea and electrolytes; liver function tests; serological tests for syphilis, toxoplasma, viruses and *Chlamydia*; chest and sacroiliac X-ray examinations; angiotensin converting enzyme estimation; and delayed-type hypersensitivity skin tests. Immunoglobulin (IgG, IgM, IgA) and complement (C3, C4) levels were determined by radioimmunodiffusion using commercial plates (Hyland). IgE levels were measured by radioimmunoassay (Pharmacia). CH50 and PH50 assays were performed using a standard haemolytic method.[12] Autoantibodies were screened for by indirect immunofluorescence using a composite tissue block consisting of rat stomach and liver and mouse stomach and kidney. Immune complexes were assessed by the Clq binding assay and quantitative cryoglobulin concentrations,[7] while T-cell numbers and subsets were analysed by indirect immunofluorescence using monoclonal antibodies[13] (OKT3, 4, 8, Ortho Diagnostic Systems). Antilymphocyte antibodies were analysed by the NIH method.[14] Antibodies to a crude extract of bovine iris were examined using a tanned sheep red blood cell haemagglutination method.[15] Titres greater than 1/8 were considered significant.

TABLE 2

Parameters of Humoral Immunity in Forty-one Patients with Anterior Uveitis

Parameter	Concentration		Normal range
	Patients with HLA-B27	Patients without HLA-B27	
C3 (g/L)	1.4 ± 0.2	1.3 ± 0.2	0.8 – 1.2
C4 (g/L)	0.4 ± 0.1	0.3 ± 0.2	0.2 – 0.4
CH50 (units)	339 ± 66	350 ± 50	159 – 387
PH50 (units)	20.0 ± 3.4	21.5 ± 5.6	10.2 – 27.6
IgG (g/L)	8.9 ± 2.8	9.7 ± 2.5	8.0 – 20.0
IgM (g/L)	1.3 ± 0.5	1.2 ± 0.4	0.5 – 1.5
IgA (g/L)	2.2 ± 0.9	1.9 ± 0.9	0.9 – 2.5
IgE (g/L)	102 ± 41	238 ± 70*	0 – 150

*$P < 0.01$.

TABLE 3

Autoantibodies in Forty-one Patients with Anterior Uveitis

Autoantibody	Patients with anterior uveitis % (number)		Controls % (number)	
	HLA-B27 positive ($n = 19$)	HLA-B27 negative ($n = 22$)	HLA-B27 positive ($n = 4$)	HLA-B27 negative ($n = 15$)
SMA	15.8% (3)	36.4% (8)†	0	0
ANA	10.5% (2)	13.6% (3)	25% (1)	0
IRIS	57.9% (11)*	22.7% (5)	0	0
RETIC	10.5% (2)	9.1% (2)	0	7% (1)

SMA = Smooth muscle autoantibody, ANA = antinuclear antibody, IRIS = iris antibody, RETIC = reticulin autoantibody.
* $P < 0.01$. † $P < 0.05$.

RESULTS

Patients consisted of 18 females (6 of whom were HLA-B27-positive) with a mean age of 38.5 ± 21 years, and 23 males (13 HLA-B27-positive) with a mean age of 37.1 ± 15 years. Table 1 summarises the aetiology in each group. The only major disease association was with the seronegative arthropathies (ankylosing spondylitis and Reiter's syndrome) in males with HLA-B27-positive AU. Complement levels (C3, C4) were elevated in the majority of patients, but not significantly above the levels in controls. The only abnormality in immunoglobulins was the increased IgE level in HLA-B27-negative AU (Table 2). Autoantibodies were increased in both groups of AU patients (Table 3), with smooth muscle antibodies (SMA) and iris antibodies increased in the HLA-B27-negative and HLA-B27-positive patients respectively. Immune complexes were normal in all patients studied (Table 4). Table 5 outlines the data for T-cell subsets and reveals the T-cell lymphopenia in the patients with HLA-B27-positive AU. The helper/suppressor (OKT4/OKT8) cell ratio and absolute numbers showed no differences between groups.

DISCUSSION

In agreement with a number of previous studies[1,16,17] we found that 46% (19/41) of the patients were HLA-B27-positive. The vast majority of patients had idiopathic AU, the only exception being the association of ankylosing spondylitis and Reiter's syndrome in HLA-B27-positive males.

The presence or absence of the HLA-B27 antigen divided AU patients into two groups which differed in both clinical and immunological features. HLA-B27-positive AU was characterised by an absolute T-cell lymphopenia during active disease; this returned to normal during remission. This phenomenon was not observed in HLA-B27-negative AU or controls and could not be attributed to antilymphocyte antibodies, as these were not detected. Byrom *et al.*[2] described a similar finding in HLA-B27-positive AU and in the HLA-B27-positive, but not the HLA-B27-negative, relatives of AU patients with active disease and postulated the lateral transmission of a virus as the aetiological factor. Such a T-cell lymphopenia may be associated with an imbalance in the normal ratio of helper to

TABLE 4

Immune Complexes in Forty-one Patients with Anterior Uveitis

Assay	Patients with anterior uveitis		Controls	
	HLA-B27-positive ($n = 19$)	HLA-B27-negative ($n = 22$)	HLA-B27-positive ($n = 4$)	HLA-B27-negative ($n = 15$)
Clq binding (%)	5.0 ± 1.8	5.3 ± 2.3	3.4 ± 3.4	6.1 ± 1.7
Cryoglobulin (g/L)	0.2 ± 0.3	0.2 ± 0.2	0.3 ± 0.2	0.3 ± 0.3