

Rheumatoid arthritis, juvenile arthritis, iridocyclitis and the Epstein-Barr virus

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SUMMARY In order to examine the relation of Epstein-Barr virus (EBV) infection to chronic arthritis in children antibodies to EB virus capsid antigen (EBVCA) and rheumatoid arthritis nuclear antigen (RANA) were analysed in sera from 133 patients classified as juvenile rheumatoid arthritis (JRA) or pauciarticular, polyarticular, or systemic juvenile chronic arthritis. Except for an increased frequency in the systemic subgroup, the prevalence of antibodies to EBVCA and titres of anti-RANA antibodies was similar in patients and controls. These data do not support an aetiological role for EBV in chronic arthritis in children, including JRA, and suggest that the mechanisms which may account for the higher titres of anti-RANA antibodies in adult RA do not occur in children.

Key phrase: antibodies to rheumatoid arthritis nuclear antigen.

The possibility of an association between rheumatoid arthritis (RA) and the Epstein-Barr virus (EBV) excites interest for two reasons: firstly, as identifying a possible aetiological agent and secondly, because of evidence of an abnormal responsiveness to the virus. It is now clear that EBV cannot be the sole aetiological agent, since several studies¹⁻³ have reported RA patients without antibodies to EB viral capsid antigens (VCAs) which are regarded as a reliable marker of previous EBV infection. Evidence for an abnormal response to the virus is often stated to be the finding of higher titres of antibodies to EBVCA, EB nuclear antigen (EBNA), and early antigen (EA) in sera from RA patients, though these studies disagree on the specificity of the antibodies that are raised. Although these studies are controversial,¹⁻⁷ there is general agreement that RA patients have higher titres of antibodies to rheumatoid arthritis nuclear antigen (RANA), originally described as a soluble cellular protein induced by EBV. This relation to the virus is supported by a close correlation between

anti-EBVCA and anti-RANA in normal sera,⁸ but its significance in RA is questioned by the finding of anti-RANA in some RA sera that do not contain antibodies to EBVCA.⁷ However, the use of such seroepidemiological methods in adult disease is complicated by the fact that over 90% of normal individuals in urban areas have serological evidence of previous EBV infection by the age of 20.⁹

An approach is to examine children in whom the frequency of EBV infection is much lower.⁹ One study failed to show an increased prevalence of anti-VCA in juvenile chronic arthritis (JCA),¹⁰ but in this survey there was no analysis based on the classification into its subgroups: pauciarticular, polyarticular, systemic, and rheumatoid factor positive juvenile rheumatoid arthritis (JRA).¹¹ Since these may be different diseases with different aetiologies,¹² it is possible that this survey may have missed a higher frequency of anti-VCA in those resembling adult RA (the JRA groups).

In this study we have compared the frequency of antibodies to EBVCA in 133 patients with JCA with that expected for this age group. The population was subdivided to see if there was an increased prevalence of EBV infection in any of the four subgroups, in particular those with JRA. Antibodies to RANA were also measured in 53 of these patients plus a

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further 20 to see if the prevalence and titre were comparable with those seen in adult RA or merely reflected the low frequency of EBV infection in childhood.

Patients and methods

Sera were collected from 133 patients with JCA and divided into subgroups by criteria previously described.¹¹ Their ages and classification are summarised in Table 1.

The 14 patients with rheumatoid factor positive JCA (JRA) fulfilled the American Rheumatism Association criteria for the diagnosis of classical or definite RA and were all aged between 10 and 16. IgG antibodies to VCA were detected by indirect immunofluorescence at a screening dilution of 1:8.¹³

Anti-RANA antibodies were examined in 73 patients and 20 young controls by immunodiffusion against a soluble extract of Wil 2 cells as previously described.⁷ The antigen source was from the same batch as that reported in the previous study, and analysed results are compared with these findings. All sera were tested blind after only one freeze thawing, without knowledge of diagnosis or VCA antibody status.

STATISTICS

The prevalence of antibodies in each of the groups was compared with each other and with a similar historical control group described in a previous study.⁹ Significance was determined by the χ^2 test.

Table 1 JCA patients studied

Subgroup	No	Age range (years)	Mean age (years)
Pauciarticular	42	2-12	6.3
Polyarticular	25	2-14	9.6
Systemic	52	2-16	9.7
Juvenile RA	14	10-16	13

Table 2 Prevalence of antibodies to EBVCA in JCA

Age (years)	Pauci		Poly		Systemic		JRA	
	VCA+/No	%	VCA+/No	%	VCA+/No	%	VCA+/No	%
0-5	2/20	10	0/3	0	5/11	45	0/0	0
5-10	7/17	41	2/11	18	13/20	65	0/0	0
10-16	2/5	40	7/11	64	9/21	43	9/14	64
Total	11/42	26	9/25	36	27/52	52	9/14	64

Results and discussion

The prevalence of antibodies to VCA in the whole population was 43.1%. The relation of antibodies to EBVCA to age and diagnostic subgroups is seen in Table 2. The percentage of children with this marker of previous EBV infection was less than 50% in all groups with three exceptions. The 64% in the whole JRA group and the 64% in the 10-16-year-old polyarticular group can be accounted for by age, since these percentages are similar to those of normal children of the same age.⁹ The 65% prevalence in the 5-10-year-old systemic group is higher than expected.

The 52% in the whole systemic group is significantly increased ($p > 0.05$) compared with the pauci- and polyarticular groups combined. This can be accounted for by the high prevalence in the under 10 year olds which could be due to the increased exposure of these young children to the EB virus during their relatively frequent hospital admissions, a phenomenon previously described by Pereira *et al.*¹⁴ On the other hand, it is conceivable that EBV could be an initiating agent in some patients with systemic JCA, though the normal frequency in the older patients would argue against this. In addition if the historical control group⁹ is used for comparison the prevalence is not significantly increased, though the pauciarticular group has a significantly decreased frequency. The low prevalence of anti-VCA antibodies in pauci- and polyarticular JCA again reflects their age and is strong evidence against EBV being of any significance in these subgroups. A similar conclusion can be drawn for the iridocyclitis in JCA, as four patients with this complication had no serological evidence of previous EBV infection.

Anti-RANA antibodies were detected in 17 out of 73 patients with JCA (23%) and six out of 20 controls (30%) (Table 3). Twenty nine patients were VCA positive and of these 15 (52%) had anti-RANA antibodies, similar to the findings for the adult controls where, among VCA positive sera, 63% were anti-RANA positive. The titres were also

Table 3 Relationship of antibodies to EBVCA with anti-RANA titres

Anti-RANA titre	Controls					
	JCA (n=73)		Children (n=20)		Adults (n=93)	
	VCA+	VCA-	VCA+	VCA-	VCA+	VCA-
Neg	14	42	6	8	27	17
Neat	9	2	2	0	16	3
1/4	4	0	3	0	28	0
1/16	2	0	1	0	2	0
Total	29	44	12	8	73	20

very similar, the majority being 1/4 or less in both disease and control groups. Thus the titre of anti-RANA and its relation to EBV infection is the same in JCA as it is in normal controls, and the lower overall frequency of anti-RANA merely reflects the lower frequency of EBV infection in children. This is in contrast with adult RA, where not only are the antibodies more frequent (86%) but they are also present in higher titres.⁷ No difference could be shown in the titres of anti-RANA between the subgroups, though the numbers became small with subclassification. Out of eight JRA sera tested only two (both from VCA positive individuals) contained anti-RANA, at titres of neat and 1/4. The frequency of anti-RANA antibodies amongst those with systemic JCA was the highest: seven out of 15 (47%), probably reflecting the relatively high frequency of anti-EBVCA antibodies in this group.

This study has shown that the frequency of anti-VCA closely correlates with age in JRA, and the lack of a significant increase in frequency in juvenile rheumatoid arthritis provides further evidence against an aetiological role for the virus in RA. The surprising finding of normal levels of anti-RANA antibodies, which were closely related to EBV infection in these children, suggests that the relatively high levels found in adult RA are due to disease mechanisms not found in children and which remain unknown.

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