Increased C1_q binding and arthritis in primary biliary cirrhosis

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SUMMARY Using the C1_q binding assay, circulating immune complexes were detected in 31 of 50 (62%) patients with primary biliary cirrhosis and 17 of these had arthritis. This took the form of a seropositive inflammatory polyarthritis in 12 of 18 patients with C1_q binding $\geq 20\%$, whereas a milder seronegative arthritis associated with scleroderma and Raynaud's phenomenon was found in five of 13 patients with C1_q binding $< 20\%$. Only two of the 19 patients with normal binding had arthritis and this was of a mild and transient nature. There was a positive correlation between C1_q binding and the serum concentrations of IgG and IgM. Results also supported the hypothesis that circulating immune complexes may be involved in the development of arthritis in patients with primary biliary cirrhosis.

Although the aetiology of primary biliary cirrhosis (PBC) remains obscure, various immune mechanisms have been implicated in its pathogenesis. Deposits of immunoglobulins and complement have been found around the bile ducts¹ and complement activation is increased.² Increased catabolism and synthesis rates of complement have also been described in PBC³ and have been ascribed to complement fixation⁴ by the increased levels of circulating immune complexes present in up to 95% of such cases.^{5,6} It has been suggested that the locally formed complexes responsible for the tissue damage in the liver may, by spilling into the systemic circulation, also cause tissue damage elsewhere.⁴ To date, no correlation has been shown between the high levels of circulating immune complexes and extrahepatic manifestations in PBC such as arthritis, although immune complexes are known to be involved in the pathogenesis of rheumatoid arthritis⁷ and other conditions with arthropathy such as systemic lupus erythematosus.⁸

In the present study we have used the C1_q binding technique of Zubler⁹ for the demonstration

of circulating immune complexes in the serum of patients with PBC and have, for the first time, shown a relationship between the level of C1_q binding and the occurrence and type of arthritis.

Methods

Fifty patients were investigated, 38 of whom were female. The mean age was 55 years and the mean duration of disease from the onset of the first symptom was 52 months (range 2-210 months). All had serum alkaline phosphatase activity greater than twice the upper normal limit and in 76% the mitochondrial antibody test was positive. Liver histology was diagnostic of, or compatible with, PBC in each case and at the time of this study 14% were at stage I, 10% at stage II, 22% at stage III, and 46% at stage IV.¹⁰ Four patients did not have recent liver histology available for staging.

Each patient had a complete rheumatological assessment with particular note being made of the history of joint pain and stiffness as well as clinical evidence of arthritis—that is, tenderness, swelling, and reduced range of movement. Rheumatoid nodules, skin changes, and Raynaud's phenomenon were also noted. Lacrimal gland function was tested using a modified Schirmer test¹¹ and patients with less than 5 mm of wetting in five minutes were

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considered to have xerophthalmia. In 37 patients posteroanterior radiographs of the hands were obtained. The presence of erosions at the joint margins was used as the sole criterion of erosive arthritis, as the interpretation of juxta-articular osteoporosis is unreliable in PBC because of the demineralisation seen in these patients as a result of chronic cholestasis. Titres of rheumatoid factor were measured using the latex fixation and Rose Waaler tests, which were considered positive with titres at or above 1/40 and 1/32 respectively. Immunoglobulin concentrations were estimated by laser nephelometry.

$C1_q$ BINDING ASSAY

Sera were tested for the presence of immune complexes by the ^{125}I $C1_q$ binding assay of Zubler *et al.*⁹ as modified by Casali *et al.*¹² This included the use of Tween 20 to reduce background binding. The mean binding in 147 normal control sera was $6 \pm 2\%$ ($\pm$ SD). Values above 10% were considered raised.

Results

$C1_q$ binding was increased in 31 (62%) of the 50 patients, with a mean value of $21.5 \pm 10.5\%$ ($\pm$ SD) and range from 11–45%. Levels correlated positively with serum concentrations of IgG and IgM ($P < 0.05$) but not with duration of liver disease, liver function tests, or histological stage of the disease. When values for $C1_q$ binding were considered in relation to the occurrence of joint disease, 19 of 31 patients with increased binding had joint symptoms or signs, while such findings were present in only two of 19

patients with normal binding. These two patients, cases 1 and 2 (Table 1), had a mild transient non-erosive arthritis involving the interphalangeal joints predominantly. In case 2 the knee joints were also involved and the rheumatoid factor was positive. Seven patients with normal binding, including both with arthritis, had symptomatic xerophthalmia (Table 2, Figure). In contrast, 17 of 31 patients with increased $C1_q$ binding had arthritis and two had arthralgia. Patients in whom $C1_q$ binding was between 10 and 20% tended to have less severe joint-related symptoms than those with binding levels $>20\%$. Five of 13 patients in the former category had a mild synovitis involving the joints of the hands and wrists predominantly and four of these had scleroderma. One patient had arthralgia only and this occurred in association with the CRST syndrome. Only two patients in this category were seropositive for rheumatoid factor and none had radiological evidence of joint erosions.

A more severe arthritis was present in 12 of 18 patients with $C1_q$ binding $>20\%$, 11 were seropositive for rheumatoid factor, and three had radiological evidence of joint erosions. One patient had arthralgia only. Each of this group of patients had required treatment with non-steroidal anti-inflammatory drugs at some stage. The criteria of the American Rheumatism Association for 'classical' rheumatoid arthritis¹³ were satisfied in case 20, 'probable' in cases 9, 10, 11, 14, and 17, and 'possible' in cases 13, 16, and 19 (Table 1). The remaining two patients did not satisfy these criteria: in case 21, who had bilateral knee joint effusions, an assay of aspirated synovial fluids revealed $C1_q$ binding of 5%

Table 1 Details of patients with primary biliary cirrhosis, $C1_q$ binding values and joint findings

Case no.	Age/sex	Duration (m)	Hist. stage	$C1_q$ % binding	Joint lesions	Hand x-rays	RA factor	AN factor	Skin	Schirmer test
1	58 F	26	1	6	Hands	Normal	—	—	R	+
2	56 F	60	NA	10	Polyarthritis	Normal	128	—	—	+
3	59 F	120	NA	11	Polyarthritis hands, wrists, shoulders	Normal	—	—	RS	—
4	57 F	27	2	11	Arthralgia hands	Normal	1024	—	RS	—
5	62 F	120	4	12	Arthritis hands	—	—	—	RS	—
6	58 F	14	2	12	Arthritis hands	Normal	—	—	RS	—
7	50 F	48	2	12	Arthritis hands	Normal	—	—	RS	+
8	70 M	22	4	13	Arthritis hands	Normal	—	—	—	—
9	62 F	40	4	20	Polyarthritis hands, wrists, elbows, knees	Normal	64	80	—	+
10	65 F	89	4	23	Polyarthritis hands, wrists, elbows, knees	Normal	40	—	R	—
11	44 F	60	3	23	Polyarthritis hands, elbows, knees	Normal	256	—	—	+
12	45 F	30	1	23	Arthralgia hands	NA	—	—	—	—
13	63 F	64	3	26	Arthritis hands	Normal	32	—	R	—
14	50 F	21	4	27	Polyarthritis hands, wrists, elbows, knees	Normal	32	60	—	—
15	65 F	60	4	28	Psoriatic arthritis	Erosions	—	—	P	—
16	57 F	6	1	29	Arthritis knees	NA	—	—	—	—
17	48 F	66	NA	30	Polyarthritis knees, ankles, feet	Erosions	32	—	R	—
18	56 F	210	3	31	Polyarthritis wrists, knees	Normal	1024	100	RS	—
19	54 F	84	4	36	Arthritis hands, knees	Normal	256	—	—	—
20	74 F	48	1	40	Nodular polyarthritis	Erosions	160	—	—	—
21	37 F	125	4	44	Arthritis knees	Normal	32	—	—	—

RA: rheumatoid factor. AN: anti-nuclear factor. NA: not available. R: Raynaud's phenomenon. RS: Raynaud's and scleroderma. P: psoriasis

Table 2 Frequencies of joint and joint-related abnormalities in patients with normal and increased binding*

		Joint involvement						
		Mild N=9	Moderate N=12	+ RA factor N=21	Sclerodactyly N=9	Raynaud's N=16	Xerophthalmia N=21	No abnormalities N=4
Normal binding	<10% N=19	2 (10.5%)	—	2 (10.5%)	—	1 (5%)	7 (36.8%)	12 (63%)
	<20% N=13	6 (46%)	—	5 (38.4%)	7 (53.8%)	9 (69%)	6 (46%)	2 (15%)
	20% N=18	1 (5.5%)	12 (66.6%)	14 (77.7%)	2 (11.1%)	6 (33.3%)	8 (44.4%)	—
Trend† P		<0.001	<0.001	<0.001	0.35	<0.06	=0.63	<0.001
Heterogeneity† P		<0.001	<0.001	<0.001	<0.001	<0.001	=0.84	<0.001

*The frequency of each abnormality is compared between the groups.

†Fischer's exact test.

Figure Relationship between severity of C1_q binding, joint disease, extra-articular manifestations, and rheumatoid factor.

which contrasted with a serum value of 44%; case 15 had psoriatic arthritis with sacroiliitis. The patients with the more marked joint disease had significantly higher levels of C1_q binding (29.75 ± 7.13) than those with mild disease (12.22 ± 4.52) (Table 3, Figure). Similarly, they exhibited a significantly higher frequency of positive rheumatoid factor and radiological evidence of joint erosions than in those with mild or absent symptoms.

Xerophthalmia did not have a statistically significant association either with joint disease or the level of C1_q binding, and, although lacrimal gland dysfunction was common in those with mild arthropathy, these differences did not reach statistical significance (Tables 2 and 3). In contrast, there were highly significant associations between scleroderma and mild arthropathy of the hand and C1_q binding levels between 11 and 19% (Tables 2 and 3).

Discussion

The findings with respect to the C1_q binding assay in the present series are similar to those of Thomas *et al.*,^{4,5} although they found a higher frequency and level of C1_q binding in primary biliary cirrhosis. Both groups found a positive correlation between the level of C1_q binding and serum IgG and IgM but not with disease activity of histological stage. The C1_q binding assay can detect both small

Table 3 Relationship between severity of joint disease, C1_q binding, rheumatoid factor, and extra-articular manifestations

Joint disease	C1 _q binding % M ± SD	+ RA factor N=21	Sclerodactyly N=9	Raynaud's N=16	Xerophthalmia N=21
None N=29	12.96 ± 8.91% (6–43%)	8 (27.5%)	3 (10.3%)	7 (24%)	10 (34%)
Mild N=9	12.22 ± 4.52 (6–23%)	3 (33%)	5 (55%)	5 (62%)	5 (55%)
Moderate N=12	29.75 ± 7.13 (20–44%)	10 (83%)	1 (8%)	4 (33%)	6 (50%)
Heterogeneity P	= 3×10^{-7}	<0.003	<0.014	=0.23	=0.43
Trend P	= 8×10^{-7}	<0.002	=0.64	=0.38	=0.28

(8–14 s) and large (19–20 s) circulating immune complexes in PBC but, for very large complexes, the Raji cell radioimmunoassay is more effective, although only in 15% of PBC patients are these the only complexes present.^{5,6} It has not been previously possible to show a correlation between increased levels of circulating immune complexes and extra-hepatic manifestations of PBC using either of these techniques, as we have in this study for arthritis.

The frequency of inflammatory polyarthritis in the present series (24%) falls midway between that reported by Clark (5%)¹⁴ and by Childs *et al.* (42%)¹⁵ in this disease. As in rheumatoid arthritis, where rheumatoid factors are the most consistent serological markers and show a strong correlation with C1_q binding,¹⁶ the patients with the most severe articular lesions in the present series had significantly higher levels of C1_q binding and frequency of positive rheumatoid factor than in those without joint lesions.

Increased concentrations of circulating immune complexes have also been shown to be associated with arthralgia and arthritis in chronic active hepatitis¹⁷ and hepatitis B virus infection.¹⁸ Likewise, there is now clear evidence that immune complexes are present in both the serum and the synovial fluid of patients with rheumatoid arthritis and that they play an important part in the pathogenesis of the chronic joint inflammation and the systemic vascular lesions.¹⁹

The results of the present series suggest that two separate forms of inflammatory arthritis occur in primary biliary cirrhosis. In the more severe form the rheumatoid factor is positive, with joint erosions seen in up to 25% of cases. Of the patients with erosions, one (case 20) had classical rheumatoid arthritis, while another patient (case 15) had psoriatic arthritis. The other patient with minimal erosions had arteritis of the legs and bowel. Although the criteria of the American Rheumatism Association for 'probable' rheumatoid arthritis were satisfied in cases 9, 10, 11, and 17 and 'possible' in cases 13, 16, and 19 (Table 1), clinically these patients did not have rheumatoid arthritis. All of these patients had C1_q binding greater than 20% and it seems likely that circulating immune complexes are important in the pathogenesis of this type of arthritis. The finding of normal levels of C1_q binding in synovial fluid and increased levels in the serum of one patient suggests that the joint lesion is secondary to the circulating complexes, not the cause, and in this respect differs from rheumatoid arthritis when synovial fluid complex concentrations are high.

The second form of arthritis—seronegative, milder, and usually transient—appears to be specifically associated with scleroderma and

Raynaud's phenomenon. It is not characterised by marked increases in circulating immune complexes and therefore the likelihood of aetiological association is less strong. This arthritis is typical of that described in scleroderma.²⁰

Finally, it is known that penicillamine causes a reduction in circulating immune complexes in primary biliary cirrhosis²¹ with improvement in the hepatic lesion, but it remains to be seen whether the joints will be favourably affected by this treatment.

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