

Antinuclear Antibodies and Anti-DNA Antibodies in Scleroderma

A Possible Relationship between Joint Manifestations and Increased Antibodies in Localized Scleroderma

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Antinuclear antibodies (ANA), including anti-DNA antibodies, and rheumatoid factors (RAT, Waaler-Rose) were determined prospectively during a 3-year period in 40 patients with localized scleroderma (LS) compared with 77 patients with generalized scleroderma (GS). ANA were increased in 26% of patients with LS, and in 47% with GS, anti-DNA antibodies in 23% of patients with LS, and in 34% with GS. Thus, the anti-DNA antibody level was lower compared with the known level in systemic lupus erythematosus. Rheumatoid factors were present in 6-7% of patients with LS, and in 14-15% of patients with GS. Increased antinuclear antibodies were not associated with any specific type of localized scleroderma, nor with internal disorders, and no case of clinical overlap to discoid or systemic lupus erythematosus was observed. However, six patients with localized scleroderma and complaints of arthralgia all presented increased antibodies, and one patient showed overlap to rheumatoid arthritis. It is suggested that increased ANA and anti-DNA antibodies in localized scleroderma, associated with joint manifestations, represents a systemic component in this type of scleroderma, with activation of the immune system and similarities with generalized collagen diseases.

Key words: scleroderma; circumscribed; morphoea; antinuclear antibodies; anti-DNA antibodies; rheumatoid factors.

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Antinuclear antibodies (ANA) including antibodies to DNA have been demonstrated in many patients with generalized scleroderma (12). Reports on cases and smaller materials of patients with localized scleroderma have indicated an increase in antinuclear antibodies in linear types as well as morphoea, seemingly in some cases related to internal diseases or overlap to other collagen diseases (2, 12, 18).

In the present study of a large series of patients with localized scleroderma, ANA and anti-DNA antibodies were analysed together with rheumatoid factors, and the results compared with those in patients with generalized scleroderma.

MATERIAL AND METHODS

The study comprised 40 patients with a clinical and light-microscopical diagnosis of localized scleroderma admitted to the Department of Dermatology during the 3-year period February 1981 to February 1984. The type of localized scleroderma, the number of patients, sex, age and age at onset of scleroderma are shown in Table 1. No patients with previous or concurrently diagnosed rheumatological disease were included. As part of clinical study on localized scleroderma it had been registered if the patients had had arthralgia related to the onset and initial phase of disease.

ANA (IgG class) were determined at Statens Seruminstitut, Copenhagen, by indirect immunofluorescence technique using human fibroblast cultured *in vitro* (10, 15). Results were assessed from the presence of nuclear fluorescence. Values were read as the weakest dilution of serum (reciprocal value) to induce fluorescence. The specific pattern of nuclear fluorescence was not assessed. Analysis in 498 blood donors (298 females, 200 males, mean ages 35.2 and 34.4 years) had shown a titre ≥ 40 in 12 persons (2.4), and this titre was thus used to discriminate increased ANA.

Anti-DNA antibodies were determined at the Department of Clinical Chemistry CL by radioimmunoassay (modified Farr technique) as adopted in 1979 by this laboratory, using double-stranded ^{14}C -DNA as tracer and ammonium sulfate precipitation (16). In our set-up, values were slightly lower than those originally described by Manthorpe et al. (16), who defined values above 14% as pathological. To define our normal range we performed analyses in 40 healthy persons (26 females, 14 males, mean age 32.6 years and range 18-64 years). In these controls anti-DNA antibodies were 5.7% ex-

Table 1

Type of localized scleroderma, sex, age (mean and range) and age at onset (mean and range) in 40 patients

	Patients <i>n</i>	Sex		Age (years)	Age at onset
		Females	Males		
Morphoea					
localized plaque	14	9	5	30.7 (10-48)	27.2 (6-47)
generalized morphoea	12	10	2	31.6 (16-57)	52.3 (34-76)
Linear types					
extremity and trunk	8	7	1	31.6 (16-57)	15.5 (7-32)
<i>en coup de sabre</i>	6	5	1	26.2 (8-51)	16.0 (6-29)

Table 2

Antinuclear antibodies, anti-DNA antibodies and rheumatoid factors in 40 patients with localized scleroderma and 77 patients with generalized scleroderma

	Patients <i>n</i>	Antinuclear antibodies ^a		Anti-DNA antibodies ^b		Rheumatoid factors ^c		Waller-Rose test ^d	
		Samples	Increased	Samples	Increased	Samples	Increased	Samples	Increased
		<i>n</i>	<i>n</i>	<i>n</i>	<i>n</i>	<i>n</i>	<i>n</i>	<i>n</i>	<i>n</i>
Morphoea									
localized plaque	14	14	3	9	1	10	2	11	2
generalized morphoea	12	12	4	10	3	12	0	12	0
Linear types									
extremity and trunk	8	6	3	7	2	5	0	3	0
<i>en coup de sabre</i>	6	6	0	4	1	3	0	5	0
Localized scleroderma, total	40	38	10 (26%)	30	7 (23%)	30	2 (7%)	31	2 (6%)
Generalized scleroderma	77	74	35 (47%)	70	23 (34%)	26	4 (15%)	29	4 (14%)

^a Increased in 2.4% of healthy individuals (titre ≥ 40).

^b Increased in 2.5% of healthy controls ($\geq 10\%$).

^c Increased in 1.0% of healthy individuals (titre ≥ 20).

^d Increased in 3.4% of healthy individuals (titre ≥ 40).

pressed as mean value, range 2-11%. One person (2.5%) had anti-DNA antibodies $\geq 10\%$, and values over this percentage were defined as pathological.

The Waaler-Rose test, based on serum activating the specific agglutination of sheep red blood cells, was performed at Statens Serum-institut, Copenhagen, according to the classical methodology (21, 29). This laboratory has found a positive Waaler-Rose test (titre ≥ 40) in 3.4% of 365 blood donors examined. Analyses also included determination of rheumatoid factors by the latex fixation test of Singer & Plotz (24). According to these authors 1% of 200 blood donors exhibited a positive RAT (titre ≥ 20).

Results obtained from patients with localized scleroderma were compared with analyses in 77 patients with generalized scleroderma (64 females, 13 males, mean age 52.7 years and range 28-73 years) admitted to the Department of Dermatology during the 3-year period February 1981 to February 1984, and meeting the American Rheumatism Association criteria for a definite diagnosis.

Statistical analysis was carried out by the Chi-square test.

RESULTS

It appears from Table 2 that ANA were increased in 26% of patients with localized scleroderma and in 47% of patients with generalized scleroderma ($P < 0.05$). These frequencies of increased ANA were both significant compared with those in blood donors. ($P < 0.001$, $P < 0.001$).

Anti-DNA antibodies were increased in 23%

of patients with localized scleroderma, and in 34% of patients with generalized scleroderma (Table 2). These frequencies of increased anti-DNA antibodies were both significant compared with the control group ($P < 0.01$, $P < 0.001$). Although there was a tendency for patients with localized scleroderma to have lower frequency of increased antibodies than patients with generalized scleroderma, the difference was not significant. Mean values and ranges of anti-DNA antibodies in the groups were: localized scleroderma 8.7% (2-30%), generalized scleroderma 8.9% (2-37%), and healthy controls 5.7% (3-11%).

Rheumatoid factors were only positive in two patients with localized scleroderma (Table 2 - the same patients presented positive RAT and Waaler-Rose). One of these patients showed clinical (hypertrophic synovium) and radiological (juxtaarticular bony erosions) evidence of early state rheumatoid arthritis, seemingly becoming manifest during the same period as plaques of morphoea had appeared. This patient probably represented an overlap between rheumatoid arthritis and localized scleroderma. The other patient had presented no clinical and radiological evidence of rheumatoid arthritis. As shown in Table 3, arthralgia had been noted in a total of six patients with localized scleroderma, all presenting increased anti-DNA antibodies (in 5 patients ANA were also analysed and found increased). The mean percentage of antibodies in patients with arthralgia was 18.8% (9-30%), while only 9.6% (2-12%) in non-arthralgic patients.

Evidence of internal diseases in localized scleroderma

Of 28 oesophagus X-rays three were abnormal (1 slight dilatation of the distal oesophagus, 1

Table 3

Antinuclear antibodies and rheumatoid factors in six patients with localized scleroderma who had presented arthralgia during the initial phase

Antinuclear antibodies		Anti-DNA antibodies		Rheumatoid factors		Waaler-Rose test	
Samples	Increased	Samples	Increased	Samples	Increased	Samples	Increased
<i>n</i>	<i>n</i>	<i>n</i>	<i>n</i>	<i>n</i>	<i>n</i>	<i>n</i>	<i>n</i>
5	5	6	6	5	2	5	2

reduced peristalsis and hiatal hernia, 1 possible reduction of peristalsis). Chest X-rays in 27 patients showed one case of pulmonary fibrosis (female aged 76). Of 21 determinations of pulmonary function, three indicated a slight reduction in diffusion capacity, one associated with restrictive insufficiency (a girl aged 17 with linear scleroderma). Analyses of serum creatinine concentration and urine protein and blood levels showed no evidence of renal disease. Thus, only the girl aged 17 presented a probable internal manifestation of scleroderma; however, ANA and antibodies were normal in this patient. No cases of overlap to other connective tissue diseases were found, except for the patient with evidence of rheumatoid arthritis described above.

DISCUSSION

The present study confirms previous findings of increased ANA and anti-DNA antibodies in localized scleroderma. In accordance with previous findings (1, 4, 5, 13, 22, 27) the frequency of increased ANA was lower in localized than in generalized scleroderma. Anti-DNA antibodies, on the other hand, showed no significant difference between localized and generalized scleroderma, as stated by others (8, 11, 17). The anti-DNA antibody level found in scleroderma was lower than the level previously reported in systemic lupus erythematosus by Manthorpe et al. (16).

In localized scleroderma increased ANA and anti-DNA antibodies have been reported in association with linear type (12, 20, 23, 26, 25) with internal disorders (2, 4), and with overlap to discoid and systemic lupus erythematosus (7, 15, 18, 28). In the present study these findings could not be confirmed to be common. However, six patients in our series who had presented arthralgia, all had positive ANA or anti-DNA antibodies (Table 3); two patients also had positive rheumatoid factors, one showing overlap to rheumatoid arthritis. Hanson et al. (9) reported on positive rheumatoid factor in seven of 21 children with localized scleroderma. Two children with high titres had evidence of arthritis. Arthralgia during the initial phase of

localized scleroderma is not uncommon; noted in 40% of patients in the extensive material of Christiansen et al. (6). In generalized scleroderma, the majority of patients complained of arthralgia, and joint effusion, inflammation of synovium and juxtaarticular bony erosions are not exceptional (3, 19). It is known that arthralgia and arthritis are common in systemic lupus erythematosus and other collagen diseases.

Thus, there is some evidence that increased ANA and anti-DNA antibodies in localized scleroderma, seemingly associated with joint manifestations, represent a systemic component of this type of scleroderma with activation of the immune system and similarities with generalized collagen diseases.

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