Cell-mediated immunity in rheumatoid arthritis

ALEX A. ANDRIANAKOS*, JOHN T. SHARP, DONALD A. PERSON†, MARTIN D. LIDSKY, AND JOSEPH DUFFY‡

From the Rheumatic Disease Section, Department of Medicine, Baylor College of Medicine, Houston, Texas 77030, USA

SUMMARY Cell-mediated immunity in rheumatoid arthritis (RA) was assessed by skin testing with six antigens in 107 patients, 94 of whom were age, sex, and race-matched with healthy individuals or patients with diseases unrelated to immunological abnormalities. 20% of RA patients were anergic. Impaired cell-mediated immunity in the RA patients was manifested by a decrease in the magnitude of skin reactivity as well as a decrease in the incidence of positive reactions to multiple antigens. Depression in cell-mediated immunity was related to age but not to sex, duration of disease, or disease activity. A slight correlation was found between absolute peripheral lymphocyte counts and the number of positive skin tests, and was confirmed by finding an association between lymphocyte counts and the size of skin reactions. A correlation was also found between lymphocyte counts and disease activity.

Four explanations of the observed depression in cell-mediated immunity in RA were considered: (1) a preoccupation of the immune mechanism of the host with cell-mediated immunity reactions related to the pathogenesis of the disease; (2) a depression of cell-mediated immune reactivity by a virus infection; (3) depression of cell-mediated immunity by therapy; and (4) immune complex suppression of cell-mediated immunity. No effect of gold therapy was found. The near universal use of salicylates or other anti-inflammatory drugs did not permit investigation of the effect of these drugs on cell-mediated immunity.

Cell-mediated immune reactions may participate in the pathogenesis of rheumatoid arthritis (RA), although their precise role has not been elucidated. Several investigators have reported depressed skin reactivity to common antigens in RA patients (Wasz-Höckert, 1951; Houba *et al.*, 1964; Hayes *et al.*, 1970; Bacon, 1970; Bitter, 1971; Waxman, *et al.*, 1973; Toh *et al.*, 1973), and Epstein and Jessar (1959) have reported a decreased capacity for sensitization in these patients. A correlation between energy and long duration of disease has been reported (Waxman *et al.*, 1973), but whether there is any relation between skin reactivity and the course of the disease has not been determined. This study

was undertaken to gain further insights into the status of cell-mediated immunity in patients with RA and to ascertain whether there is a relation between cell-mediated immune reactions, disease activity, duration of disease, treatment, and response to treatment.

Materials and methods

RA PATIENTS

107 patients with RA from the Baylor College of Medicine affiliated hospitals and outpatient clinics (Ben Taub General Hospital, Veterans Administration Hospital, and Texas Institute for Rehabilitation and Research) were studied (Table 1). All had classical or definite RA (Ropes *et al.*, 1956, 1958). 84 (72 White and 12 Black) were male and 23 (15 White and 8 Black) were female; their ages ranged from 23 to 77 years, mean 54.2 years. 59 patients were being treated with gold salts for periods ranging from 6 to 25 months, with a mean of 16.8 months. Many of these patients were also

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Correspondence to Dr. J. T. Sharp, Department of Medicine, V.A. Hospital, Danville, Illinois 61832, U.S.A.

*Present address: University of Athens Medical School, 2nd Department of Medicine, Hippokraton General Hospital, Athens (610) Greece

†Senior Investigator, The Arthritis Foundation

‡Present address: Mayo Clinic and Foundation, Rochester, Minnesota 55901

taking salicylates, indomethacin or paracetamol. Of 48 patients not treated with gold, 42 were receiving only aspirin, indomethacin, or paracetamol. 6 patients were receiving prednisone in addition to anti-inflammatory drugs; 5 were on 5 mg/day and one was on 10 mg/day. 5 of the RA patients were seronegative and 2 had Felty's syndrome.

A history was taken and a physical examination was performed by the same physician on each patient at the time of skin testing. A complete blood count, Wintrobe erythrocyte sedimentation rate (ESR), anti-IgG titre, and serum protein electrophoresis were obtained in most patients at the same time. Absolute peripheral lymphocyte counts were calculated from the differential cell count and the total white blood cell count. The latex fixation test for anti-IgG activity was carried out on twofold serum dilutions of 1:40 through 1:81 920 using a technique described previously (Singer and Plotz, 1956). Serum protein electrophoresis was done in the individual hospital laboratories. Pretreatment clinical and laboratory data were available on the patients receiving gold therapy from the records of another study. Disease activity was assessed by the articular index (Lansbury and Haut, 1956).

CONTROLS

94 of the RA patients were matched for age (± 2.5 years), sex, and race with 94 control subjects, 14 of whom were healthy individuals and 80 were patients with diseases unrelated to known immunological abnormalities. The healthy individuals were spouses of RA patients. Patients in the control group included 40 with gout, 13 with hypertensive cardiovascular disease, 9 with degenerative joint disease, and 18 with other conditions (Table 1). 71 of the control group were male, 59 Whites and 12 Blacks, and 23 were female, 15 Whites and 8 Blacks. Their ages ranged from 26 to 76 years, mean 55.1 years.

Results were analysed comparing either the matched or the total patient groups to the control subjects and no differences were found. The data presented here are from the total patient group.

SKIN TESTING

All RA patients and controls received 0.1 ml intradermal injections of the following six antigens: (1) Purified protein derivative (PPD) (Connaught, Lot 15021-1), 50 IU/ml; (2) mumps skin test antigen (Eli Lilly & Co., Lot DNC 2-V24-1); (3) streptokinase-streptodornase (SK-SD) (Varidase, Lederle, Lot 5-2201-66), 400 units of streptokinase and 100 units of streptodornase per ml; (4) dermatophytin 'O' (Hollister-Stier, Lot H50637 M), 1:100 dilution; (5) mixed monilia antigens (Hollister-Stier, Lot F4454710), 1:100 dilution; (6) dermatophytin (Trichophytin) (Hollister-Stier, Lot E40098 M), 1:100 dilution.

The injections were made into the anterior surface of the forearms and skin reactions were read 48 hours later. Two perpendicular diameters of the induration were measured and the mean of these measurements was recorded. Induration with a mean diameter of 5 mm or more was considered a positive reaction for all antigens except PDD in which instance a mean diameter of 10 mm or more was considered positive (Shaffer and Goldin, 1969). Individuals who had no reaction of these magnitudes were considered to be anergic.

STATISTICAL ANALYSIS

χ^2 analysis, mean values, and standard errors, the coefficient of correlation, and Student's 't' test were used to evaluate the data (Hill, 1961).

Results

ANERGY AND POSITIVE SKIN REACTIONS

20 RA patients (18.7%), including the 2 with Felty's

Table 1 Rheumatoid patients and control subjects tested

Group	No.	Male		Female		Age (yr)	
		White	Black	White	Black	Range	Mean
RA patients	107	72	12	15	8	23-77	54.2
Gold therapy	59	41	8	8	2	23-68	52.1
Other treatment	48	31	4	7	6	39-77	56.9
Controls	94	59	12	15	8	26-76	55.1
Healthy	14	2	0	12	0	26-59	50.7
Gouty	40	31	9	0	0	35-76	55.8
DJD	9	7	1	0	1	46-66	57.6
HCVD	13	8	1	1	3	40-71	57.0
Trauma	6	6	0	0	0	35-54	45.2
Nonspecific arthralgias	4	3	1	0	0	39-62	52.7
Diabetes mellitus	3	1	0	0	2	51-71	62.6
Osteopenia	3	0	0	2	1	59-69	65.0
Paget's disease	2	1	0	0	1	58-59	58.5

DJD = degenerative joint disease; HCVD = hypertensive cardiovascular disease.

syndrome, were anergic to all antigens, while only 2 controls (2.1%), one healthy female and one male with gout, were anergic (Table 2). The difference was significant even when the 2 patients with Felty's syndrome were excluded from the analysis. Sex and race were examined for an effect on skin reactivity and none was found. None of the seronegative RA patients and none of the patients taking prednisone was anergic.

Positive skin reactions were more frequent among the control subjects than in RA patients for all antigens and this difference was significant except for trichophytin (Table 3). SK-SD and mumps antigens gave the highest frequency of positive skin reactions. Of the 87 RA patients who reacted to one or more antigens, 85 reacted to either SK-SD, or mumps, or both. 11 of these patients reacted only to SK-SD and 7 only to mumps. Similarly, among the 92 controls who had positive reactions to one or more antigens, 91 were sensitive to either SK-SD, or mumps, or both. The frequency of positive reactions to one or more, 2 or more, 3 or more,

Fig. 1 Incidence of positive skin reactions to multiple skin test antigens in RA patients and control subjects.

and 4 or more skin test antigens was found to be significantly higher among the control group than in RA patients (Fig. 1).

The degree of skin reactivity as measured by induration was found to be significantly lower in RA patients than in the control group for four antigens: PPD, SK-SD, dermatophytin 'O', and mixed monilia antigens (Fig. 2). The results were examined by applying different rules for selecting subgroups. First, reactive patients were defined as those who reacted to at least one antigen. Second, all RA patients and controls who failed to manifest any induration for a specific antigen were excluded when considering tests with that antigen. Using these subgroups the reactive RA patients exhibited significantly smaller reactions than the reactive control group to 4 antigens. Even when all individuals who failed to manifest induration to a specific antigen were excluded, reactions among RA patients were fewer than among the control group for 2 antigens (Table 4).

Fig. 2 Magnitude of skin reactions to six antigens is shown in RA patients and control subjects. Derm 'O' = Dermatophytin 'O'; SK-SD = streptokinase-streptodornase; PPD = purified protein derivative.

Table 2 Incidence of anergy

	No. reactive to one or more antigens	No. anergic	Total	P value
RA patients				
Total group	87 (81.3%)	20 (18.7%)	107	<0.0005*
Excluding patients with Felty's syndrome	87 (82.9%)	18 (17.1%)	105	<0.0005*
Male	71 (85.5%)	13 (15.5%)	84	NS
Female	16 (69.6%)	7 (31.4%)	23	
White	72 (82.8%)	15 (17.2%)	87	NS
Black	15 (75%)	5 (25%)	20	
Gold therapy	49 (83.1%)	10 (16.9%)	59	NS
Other treatment	38 (79.2%)	10 (20.8%)	48	
Controls	92 (97.9%)	2 (2.1%)	94	

*RA patients were compared with the controls. Other comparisons were intragroup—e.g. male patients were compared to female patients, etc. NS = not significant.

Table 3 *Incidence of positive reactions to individual antigens*

Antigens	RA patients % positive	Controls % positive	P value
PPD	2.8	12.8	<0.01
Mumps	57.0	72.3	<0.02
SK-SD	61.7	86.2	<0.0005
Dermatophytin 'O'	29.0	48.9	<0.02
Mixed monilia antigen	31.8	48.9	<0.02
Trichophytin	13.1	16.0	NS

PPD=purified protein derivative; SK-SD=streptokinase-streptodornase.

Table 4 *Mean diameter of skin-induration*

Antigens	First subgroup (mean $\pm$ SEM)*			Second subgroup (mean $\pm$ SEM)†			
	RA patients	Controls	P value	RA patients	n	Controls	n
PPD	0.79 $\pm$ 0.28	2.03 $\pm$ 0.46	<0.025	6.32 $\pm$ 1.42	11	9.38 $\pm$ 0.99	21
Mumps	10.43 $\pm$ 1.07	10.23 $\pm$ 0.89	NS	12.46 $\pm$ 1.08	75	12.56 $\pm$ 0.90	75
SK-SD	11.1 $\pm$ 0.98	15.49 $\pm$ 0.97	<0.005	13.23 $\pm$ 0.97	74	17.59 $\pm$ 0.87	81
Dermatophytin 'O'	3.34 $\pm$ 0.54	5.01 $\pm$ 0.61	<0.05	7.29 $\pm$ 0.74	42	9.04 $\pm$ 0.69	51
Mixed monilia	3.31 $\pm$ 0.45	5.14 $\pm$ 0.66	<0.025	6.23 $\pm$ 0.51	49	9.09 $\pm$ 0.81	52
Trichophytin	1.45 $\pm$ 0.27	1.15 $\pm$ 0.25	NS	5.04 $\pm$ 0.41	25	5.3 $\pm$ 0.48	20

*Mean induration to individual antigens for 87 reactive RA patients and 92 reactive controls in mm $\pm$ SEM.

†Mean induration for each antigen was calculated by excluding all reactions with no induration. Numbers (n) are different for each antigen and for patient and control groups.

Table 5 *Comparison of several clinical and laboratory indexes between anergic and reactive RA patients*

Indexes	Anergic patients (mean $\pm$ SEM)	Reactive patients (mean $\pm$ SEM)
Duration of disease (yr)	7.4 $\pm$ 1.3	10.2 $\pm$ 0.9
Articular index	52.9 $\pm$ 9.6	51.8 $\pm$ 3.9
Subcutaneous nodules (%)	45	36
Age (yr)	56.4 $\pm$ 2.08	53.7 $\pm$ 0.9
ESR (mm/h)	30 $\pm$ 2.7	26.5 $\pm$ 1.7
Anti-IgG (tube)	8.6 $\pm$ 0.48*	8.3 $\pm$ 0.24*
Absolute peripheral lymphocytes (/mm ³)	1485 $\pm$ 173 (1.5 $\pm$ 0.17 $\times$ 10 ⁹ /l)	1803 $\pm$ 75 (1.8 $\pm$ 0.75 $\times$ 10 ⁹ /l)
Serum proteins (g/l)		
Albumin	41 $\pm$ 1.6	42.3 $\pm$ 0.72
α_1 -globulin	2.7 $\pm$ 0.21	3.01 $\pm$ 0.10
α_2 -globulin	8.7 $\pm$ 0.44	8.31 $\pm$ 0.21
β -globulin	10.8 $\pm$ 0.56	10.4 $\pm$ 0.20
γ -globulin	16.2 $\pm$ 1.09	14.3 $\pm$ 0.61

*Geometric mean titre of 8.6 is 1:7746, 8.3 is 1:6296.

Table 6 *Effect of age on induration of skin reactions*

		Induration (mm) (mean $\pm$ SE)					
		PPD	Mumps	SK-SD	Derm-'O'	Monilia	Trichophytin
(1) RA patients <55 yr	n=4	n=35	n=40	n=15	n=16	n=9	
	8.25 $\pm$ 1.18	13.24 $\pm$ 1.68	15.23 $\pm$ 1.50	8.07 $\pm$ 1.50	6.25 $\pm$ 0.99	5.17 $\pm$ 0.67	
(2) RA patients 55 yr or older	n=7	n=40	n=34	n=27	n=33	n=16	
	5.21 $\pm$ 2.09	11.78 $\pm$ 1.40	10.88 $\pm$ 1.03	6.87 $\pm$ 0.80	6.23 $\pm$ 0.60	4.97 $\pm$ 0.54	
(3) Control subjects <55 yr	n=8	n=33	n=36	n=23	n=22	n=12	
	11.5 $\pm$ 1.89	12.70 $\pm$ 1.40	19.11 $\pm$ 1.40	9.57 $\pm$ 1.13	10.82 $\pm$ 1.50	5.58 $\pm$ 0.74	
(4) Control subjects 55 yr or older	n=12	n=42	n=45	n=28	n=30	n=8	
	7.92 $\pm$ 1.06	12.45 $\pm$ 1.18	16.38 $\pm$ 1.07	8.61 $\pm$ 0.86	7.83 $\pm$ 0.82	4.88 $\pm$ 0.44	
(1) v. (2)							
t	1.03	0.68	2.31	0.77	0.02	0.22	
P	>0.10	>0.10	<0.0125	>0.10	>0.10	>0.10	
(3) v. (4)							
t	1.78	0.14	1.58	0.68	1.86	0.72	
P	<0.05	>0.10	>0.10	>0.10	<0.05	>0.10	
(1) v. (3)							
t	1.13	0.25	1.88	0.81	2.33	0.40	
P	>0.10	>0.10	<0.05	>0.10	<0.025	>0.10	
(2) v. (4)							
t	1.29	0.37	3.60	1.47	1.60	0.12	
P	>0.10	>0.10	<0.0005	>0.10	>0.10	>0.10	

n=no. of reactors.

Table 7 *Anergy in relation to inactive disease and therapy*

RA patients	Active	Inactive	Total	P value
Gold therapy				
Anergic	7	3	10	<0.025
Reactive	47	2	49	
Other therapy				
Anergic	9	1	10	NS
Reactive	36	2	38	
Total				
Anergic	16	4	20	<0.01
Reactive	83	4	87	

SKIN REACTIVITY AND DISEASE ACTIVITY

Several factors reflecting the severity and course of arthritis were examined (Table 5). These included the duration of the disease, articular index, incidence of subcutaneous nodules, age, ESR, anti-IgG titres, serum proteins, and absolute peripheral lymphocyte counts. None was significantly related to anergy. However, when the effect of age on induration of skin test responses among patients and controls reacting to a given antigen was examined, it was found that older individuals in both groups had smaller mean reactions for every antigen, although differences for specific antigens were significant in only three instances (Table 6).

An apparent relationship was found between anergy and inactive disease, defined as an articular index of 0 (Table 7). Among the anergic patients 20% had inactive disease while only 5% of the reactive patients had inactive disease. This difference was largely accounted for by the anergic group on gold therapy, among whom 30% had inactive disease. Although this difference is significant ($P<0.01$) it should be noted that the number of patients with inactive disease was small and a shift of one patient from the inactive, anergic group would change the P value from <0.01 to >0.05 .

Fig. 3 Incidence of positive skin reactions is shown in RA patients divided into four groups representing progressively more disease activity.

To test further the significance of this finding the incidence of positive skin reactions was examined for a relationship to disease activity as measured by articular index (Fig. 3). Mumps, SK-SD, and any one of the other antigens were included in the three major antigens. The patients were divided into four equal groups with group I being the least active. The incidence of positive skin reactions in group III was lower than in all other groups, but the significance of this observation is doubtful since there was no scaling from group I through group IV.

The mean diameter of skin induration induced by SK-SD increased progressively from group I through III, but the differences were not significant (Fig. 4).

Fig. 4 Magnitude of skin reactions to SK-SD is shown in RA patients divided into four groups representing progressively more disease activity.

Fig. 5 Absolute peripheral lymphocyte counts (mm^3) are shown as they relate to the number of positive skin reactions in RA patients. Shaded areas show the mean $\pm$ SE. Lymphocyte counts were significantly lower in those with negative reactions and only one positive reaction when compared with those patients with two or more positive reactions. Conversion factor: Traditional units to SI—Lymphocytes: $1000/\text{mm}^3 \approx 1.0 \times 10^9/\text{l}$.

The magnitude of skin induration to all other antigens was nearly the same among the four subgroups.

LYMPHOCYTE COUNTS, SKIN REACTIVITY, AND DISEASE ACTIVITY

Absolute peripheral lymphocyte counts in the RA patients ranged from 430 to 3500/mm³ (0.4–3.5 × 10⁹/l). Patients with RA who had positive skin reactions to multiple antigens tended to have higher lymphocyte counts (Fig. 5). A slight correlation was also found between number of lymphocytes in the peripheral blood and the induration seen with

Fig. 6 Absolute peripheral lymphocyte counts (/mm³) are shown in RA patients divided into four groups representing progressively more disease activity. ○ = patients on gold; ● = patients on other treatment. Shaded areas show the mean ± SE. There are significant differences between lymphocyte counts in groups I and IV, II and III, II and IV with *P* values <0.05, 0.01, and 0.005, respectively. Also the lymphocyte counts were significantly lower in the combined groups I and II compared to groups III and IV when *P* < 0.001.

SK-SD (*r* = 0.21, ±SE_r = 0.09). Higher lymphocyte counts were related to greater disease activity (Fig. 6).

EFFECT OF TREATMENT

The incidence of positive tests and the mean diameters of skin reactions in patients receiving gold salts were not different from the entire group in spite of striking differences in disease activity, lymphocyte counts, serum protein, and anti-IgG titres (Table 8). Although the patients who were receiving gold were not matched or paired with the other patients with RA, the differences shown in Table 8 were believed to be due to treatment as shown by pretreatment measurements in the gold-treated patients.

Discussion

Previous studies on cell-mediated immunity in RA patients have led to conflicting conclusions (Wasz-Höckert, 1951; Epstein and Jessar, 1959; Houba *et al.*, 1964; Block *et al.*, 1968; Bacon, 1970; Hayes *et al.*, 1970; Bitter, 1971; Müller, 1971; Serre *et al.*, 1972; Curtis *et al.*, 1973; Griswold and McIntosh, 1973; Toh *et al.*, 1973; Waxman *et al.*, 1973). Some investigators have reported intact cell-mediated immune responses in RA patients (Block *et al.*, 1968; Müller, 1971; Curtis *et al.*, 1973) but others have found depressed cell-mediated immunity (Wasz-Höckert, 1951; Epstein and Jessar, 1959; Houba *et al.*, 1964; Bacon, 1970; Hayes *et al.*, 1970; Bitter, 1971; Griswold and McIntosh, 1973; Toh *et al.*, 1973; Waxman *et al.*, 1973). In studies reported here cell-mediated immunity, assessed by testing with 6 antigens, was found to be impaired in RA patients, 20% of whom were anergic compared with only 2% of the control group. Not only was anergy more frequent in the group with RA, but the magnitude of skin reactivity was decreased.

Table 8 Comparison of various clinical and laboratory indexes in RA patients

Indexes	Patients on other treatment (I) (mean ± SEM)	Patients on gold therapy		<i>P</i> values*	
		Before gold (2) (mean ± SEM)	At time of skin testing (3) (mean ± SEM)	1 v. 3	2 v. 3
Articular index	62.02 ± 5.7	77.37 ± 5.0	43.65 ± 4.5	<0.02	<0.001
Lymphocyte counts (/mm ³)	1888 ± 112 (1.89 ± 0.11 × 10 ⁹ /l)	2110 ± 85 (2.11 ± 0.09 × 10 ⁹ /l)	1635 ± 91 (1.64 ± 0.091 × 10 ⁹ /l)	NS	<0.001
Serum protein (g/l)					
Albumin	38.3 ± 0.95	39.2 ± 0.82	44.7 ± 0.76	<0.001	<0.001
α ₁ globulin	3.17 ± 0.16	3.61 ± 0.15	2.79 ± 0.12	NS	<0.001
α ₂ globulin	9.08 ± 0.32	8.99 ± 0.24	7.89 ± 0.25	<0.005	<0.005
β globulin	10.5 ± 0.31	10.48 ± 0.24	10.33 ± 0.26	NS	NS
γ globulin	15.88 ± 1.04	17.25 ± 0.76	13.94 ± 0.67	NS	<0.005
Anti-IgG titres (tube)	8.97 ± 0.32†	9.13 ± 0.27	8.0 ± 0.27	<0.025	<0.005

*Significance calculated by dividing difference of two means by the standard error of the difference of two means.

†Tube dilution of 8.97 is a titre of 1:10 024; 9.13 is 1:11 196; and 8.0 is 1:5120.

In each of the previous negative studies either the number of the patients studied was less than 25 (Block *et al.*, 1968; Curtis *et al.*, 1973) or only one skin test antigen was used (Müller, 1971), thus limiting the possibility of detecting impaired cell-mediated immunity. We therefore conclude that studies finding a general depression in cell-mediated immunity are more credible than those reporting no defect. What significance can we attribute to this finding? Four possibilities should be considered. First, cellular immune mechanisms may be pre-occupied with the reactions in the synovial membrane and other tissues which are involved in the disease. Second, if RA is an infectious disease, depressed cell-mediated immunity may be a consequence of that infection. Third, depressed cell-mediated immunity in RA may be a consequence of therapy. Fourth, the presence of immune complexes, which suppress a number of lymphocyte functions, may be responsible for the observed depression of skin reactivity.

Concerning the first, several investigators have suggested that cell-mediated immunopathological responses may play a role in the pathogenesis of RA. Multz and his colleagues (1968) have shown that RA patients were less reactive to common skin test antigens, but more reactive to autologous synovial tissue than nonrheumatoid patients. This suggests a cell-mediated hypersensitivity reaction to an antigen in the rheumatoid synovium. Others have reported that rheumatoid synovial homogenates caused inhibition of migration of leucocytes from RA patients in a lower concentration than was inhibitory to leucocytes from control individuals (Thiele and Rothenberger, 1971; Bacon *et al.*, 1973). Nonrheumatoid synovial extracts failed to inhibit leucocyte migration (Bacon *et al.*, 1973).

In other studies aggregated IgG has been reported to inhibit leucocyte migration in RA patients, but not in control groups (Fröland and Gaarder, 1973; Brostoff *et al.*, 1973), although it does not induce lymphocyte blastogenesis (Kacaki *et al.*, 1969). Since aggregated IgG might be extracted from rheumatoid synovial tissue the latter observation could account for leucocyte migration inhibition by homogenates of rheumatoid synovium. More recently, Person *et al.* (1976) have found that lymphocytes from RA patients are cytotoxic for rheumatoid synovial cells *in vitro*, an observation that suggests a cell-mediated immune response to an antigen in the rheumatoid synovial cells.

Other evidence that cellular mechanisms may be involved in the pathogenesis of RA was provided by the observation that prolonged thoracic duct lymphocyte drainage induced clinical improvement or remission of RA (Wegelius *et al.*, 1970) and also

suppressed cell-mediated immunity as assessed by skin testing (Paulus *et al.*, 1972). Ziff has inferred that both T and B lymphocytes are present in the synovial tissue (Ziff, 1974). T-cells are the predominant lymphocyte in rheumatoid synovial fluid (Fröland *et al.*, 1973). Finally, synovitis has been induced in rabbits by intra-articular injections of lymphokines (Ishikawa *et al.*, 1973), and recently lymphokine-like substances have been found in synovial fluid from RA patients (Stastny *et al.*, 1975). Other evidence for a role for cell-mediated immunity in RA is provided in this study by positive correlations between lymphocyte counts and disease activity and between lymphocyte counts and the number and magnitude of skin reactions.

Although at present there is no convincing evidence that RA is an infectious disease it should be noted that impaired cell-mediated immunity has been reported in a number of viral, bacterial, fungal, and parasitic infections (Salaman, 1969).

The role of therapy was evaluated by examining subgroups of patients. Since the frequency of anergy was similar in patients treated with gold as compared with those not receiving gold, this medication would not appear to be a likely cause for the observed depression of delayed reactions. Recently aspirin and several other anti-inflammatory drugs have been reported to depress lymphocyte reactivity *in vitro* (Pachman *et al.*, 1971; Opelz *et al.*, 1973). Since nearly all the patients in this study were receiving aspirin or indomethacin it was not possible to evaluate the role of these agents in the depressed cell-mediated immunity skin reactivity observed.

Neither the data from this study nor those published provide convincing evidence in favour of any of the possible explanations for depressed cell-mediated immunity in RA, and additional studies will be required to ascertain the exact significance of this observation.

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References

- Bacon, P. A. (1970). *Impaired Cell-Mediated Hypersensitivity in Man*, p. 49. Ed. by J. F. Jennings and D. J. Ward. The Robert Jones and Agnes Hunt Orthopaedic Hospital Management Committee, Oswestry.
- Bacon, P. A., Cracchiolo, A., Bluestone, R., and Goldberg, L.'S. (1973). Cell-mediated immunity to synovial antigens in rheumatoid arthritis. *Lancet*, 2, 699-702.

- Bitter, T. (1971). A clinical investigation of the pathogenesis of autoimmunity. *Rheumatoid Arthritis*, p. 305. Ed. by W. Müller, H.-G. Harwerth and K. Fehr. Academic Press, London and New York.
- Block, S. R., Gibbs, C. B., Stevens, M. B., and Shulman, L. E. (1968). Delayed hypersensitivity in systemic lupus erythematosus. *Annals of the Rheumatic Diseases*, 27, 311-318.
- Brostoff, J., Howell A., and Roitt, I. M. (1973). Leucocyte migration inhibition with aggregated gammaglobulin in patients with rheumatoid arthritis. *Clinical and Experimental Immunology*, 15, 1-7.
- Curtis, J. E., Sharp, J. T., Lidsky, M. D., and Hersh, E. M. (1973). Immune response of patients with rheumatoid arthritis during cyclophosphamide treatment. *Arthritis and Rheumatism*, 16, 34-42.
- Epstein, W. L., and Jessar, R. A. (1959). Contact-type delayed hypersensitivity in patients with rheumatoid arthritis. *Arthritis and Rheumatism*, 2, 178-181.
- Fröland, S. S., and Gaarder, P. I. (1973). The effect of IgG in vitro on leukocytes from patients with rheumatoid arthritis. *Scandinavian Journal of Immunology*, 2, 385-393.
- Fröland, S. S., Natvig, J. B., and Husby, G. (1973). Immunological characterization of lymphocytes in synovial fluid from patients with rheumatoid arthritis. *Scandinavian Journal of Immunology*, 2, 67-73.
- Griswold, W. R., and McIntosh, R. M. (1973). Lymphocyte responses to phytohemagglutinin in rheumatoid arthritis and glomerulonephritis and the effects of immunosuppression. *Experientia*, 29, 606-607.
- Hayes, J. R., Ward, D. J., and Jennings, J. F. (1970). Studies on cell-mediated hypersensitivity responses in rheumatoid arthritis. *Impaired Cell-Mediated Hypersensitivity in Man*, p. 37. Ed. by J. F. Jennings and D. J. Ward. The Robert Jones and Agnes Hunt Orthopaedic Hospital Management Committee, Oswestry.
- Hill, A. B. (1961). *Principles of Medical Statistics*, 9th ed. Oxford University Press, New York.
- Houba, V., Adam, M., Maleček, J., and Tesárek, B. (1964). Delayed hypersensitivity and antibody response in rheumatoid arthritis. *Experientia*, 20, 522-523.
- Ishikawa, H., Andreis, M., Stastny, P., and Ziff, M. (1973). Lymphokine-induced synovitis in the rabbit: an electron microscopic study. (Abst.) *Arthritis and Rheumatism*, 16, 121.
- Kačák, J. N., Bullock, W. E., and Vaughan, J. H. (1969). Failure of lymphocyte transformation in response to gammaglobulins in rheumatoid arthritis. *Lancet*, 1, 1289-1290.
- Lansbury, J., and Haut, D. D. (1956). Quantitation of the manifestations of rheumatoid arthritis. 4. Area of joint surfaces as an index to total joint inflammation and deformity. *American Journal of the Medical Sciences*, 232, 150-155.
- Müller, W. (1971). Immunological reactivity in rheumatoid arthritis. *Rheumatoid Arthritis*, p. 297. Ed. by W. Müller, H.-G. Harwerth and K. Fehr. Academic Press, London and New York.
- Multz, C. V., Kamin, E. J., McDowell, M. K., and Butler, B., Jr. (1968). Delayed allergic reactions to homogenized synovial tissue. (Abst.) *Arthritis and Rheumatism*, 11, 110.
- Opelz, G., Terasaki, P. I., and Hirata, A. A. (1973). Suppression of lymphocyte transformation by aspirin. *Lancet*, 2, 478-480.
- Pachman, L. M., Esterly, N. B., and Peterson, R. D. A. (1971). The effect of salicylate on the metabolism of normal and stimulated human lymphocytes in vitro. *Journal of Clinical Investigation*, 50, 226-230.
- Paulus, H. E., Machleder, H., Bangert, R., Stratton, J. A., Peter, J. B., Goldberg, L., Whitehouse, M. W., Yu, D., and Pearson, C. M. (1972). Effects of removing thoracic duct lymphocytes from a patient with severe rheumatoid arthritis. (Abst.) *Arthritis and Rheumatism*, 15, 122.
- Person, D. A., Sharp, J. T., and Lidsky, M. D. (1976). The cytotoxicity of leukocytes and lymphocytes from patients with rheumatoid arthritis for synovial cells. *Journal of Clinical Investigation*, 58, 690-698.
- Ropes, M. W., Bennett, G. A., Cobb, S., Jacox, R. F., and Jessar, R. A. (1956). Proposed diagnostic criteria for rheumatoid arthritis. *Bulletin on Rheumatic Diseases*, 7, 121-124.
- Ropes, M. W., Bennett, G. A., Cobb, S., Jacox, R. F., and Jessar, R. A. (1958). 1958 revision of diagnostic criteria for rheumatoid arthritis. *Bulletin on Rheumatic Diseases*, 9, 175-176.
- Salaman, M. H. (1969). Immunodepression by viruses. *Antibiotica et Chemotherapia*, 15, 393-406.
- Serre, H., Simon, L., Mandin, J., Sany, J., and Clot, J. (1972). Approche immunologique de la polyarthrite chronique rhumatismale: premiers résultats. *Revue de Rhumatisme et des Maladies Ostéo-articulaires*, 39, 91.
- Shaffer, J. G., and Goldin, M. (1969). Vaccines and diagnostic skin tests. *Clinical Diagnosis by Laboratory Methods*, 14th ed., p. 1069. Ed. by I. Davidsohn and J. B. Henry. Saunders, Philadelphia.
- Singer, J. M., and Plotz, C. M. (1956). The latex fixation test. I. Application to the serologic diagnosis of rheumatoid arthritis. *American Journal of Medicine*, 21, 888-892.
- Stastny, P., Rosenthal, M., Andreis, M., and Ziff, M. (1975). Lymphokines in the rheumatoid joint. *Arthritis and Rheumatism*, 18, 237-243.
- Thiele, H. G., and Rothenberger, W. (1971). Experiences with the leucocyte migration inhibition test in rheumatoid arthritis. *Rheumatoid Arthritis*, p. 463. Ed. by W. Müller, H.-G. Harwerth, and K. Fehr. Academic Press, London and New York.
- Toh, B. H., Roberts-Thomson, I. C., Mathews, J. D., Whittingham, S., and Mackay, I. R. (1973). Depression of cell-mediated immunity in old age and the immunopathic diseases, lupus erythematosus, chronic hepatitis and rheumatoid arthritis. *Clinical and Experimental Immunology*, 14, 193-202.
- Wasz-Höckert, D. (1951). Tuberculin sensitivity in rheumatoid arthritis. *Acta Pathologica et Microbiologica Scandinavica*, Suppl. 91, 135-138.
- Waxman, J., Lockshin, M. D., Schnapp, J. J., and Doneson, I. N. (1973). Cellular immunity in rheumatic diseases. I. Rheumatoid arthritis. *Arthritis and Rheumatism*, 16, 499-506.
- Wegelius, O., Laine, V., Lindström, B., and Klockars, M. (1970). Fistula of the thoracic duct as immunosuppressive treatment in rheumatoid arthritis. *Acta Medica Scandinavica*, 187, 539-544.
- Ziff, M. (1974). Relationship of cellular infiltration of rheumatoid synovial membrane to its immune response. *Arthritis and Rheumatism*, 17, 313-319.