THE RELATIONSHIP OF RHEUMATOID SERUM FACTOR TO RHEUMATOID ARTHRITIS

A 5-YEAR FOLLOW UP OF A POPULATION SAMPLE

BY

J. BALL AND J. S. LAWRENCE

*From the Rheumatism Research Centre and the Empire Rheumatism Council Field Unit,
University of Manchester*

It is known from population surveys that only some 30 per cent. of individuals whose serum has a positive sheep cell agglutination titre of 32 or more (expressed as a reciprocal value) actually have any evidence of arthritis or other "collagen disease", and similar proportions have been encountered using the F II-latex particle and Bentonite flocculation tests (Aho, Julkunen, Laine, Ripatti, and Wager, 1961; Ball and Lawrence, 1961; Mikkelsen, Dodge, Duff, Epstein, and Napier, 1962; Burch, Bloch, and Valkenburg, personal communications). This might mean that a non-rheumatoid factor having similar agglutinating properties to the rheumatoid serum factor is present in the serum of a proportion of healthy members of the population. Family studies have seemed to confirm this, since relatives of asymptomatic sero-positive individuals are themselves more likely to be asymptomatic and have a positive serum test (Lawrence and Ball, 1958). The alternative hypothesis, that the serum factor is identical in rheumatoid and asymptomatic individuals, appears to us more probable. If this is so it would have to be assumed that the factor is associated with a predisposition to the disease which only appears when some other cause becomes operative. To test this hypothesis, a group of sero-positive individuals from random population samples of the town of Leigh have been re-examined after an interval of 5 years. This is an extension of an earlier study which suggested that persons with a positive sheep-cell agglutination test were more likely to develop rheumatoid arthritis (Ball and Lawrence, 1961). The present survey confirms this conclusion and amplifies it.

Method

All those whose serum was positive (titre 32 or more) by the sheep cell agglutination test in population samples examined in 1954 and 1956 were revisited in 1959 and 1961 respectively and questioned about rheumatic and other symptoms and infections since the previous examination. Those seen in 1954 were aged 55-64; in 1956 all ages from 15-54 and 65 onwards were included. In the 1959 and 1961 reviews the joints were examined clinically and blood was taken. X rays of the hands, feet, and cervical spine were also obtained from those seen in 1961.

Samples of individuals with a titre of <4 and of those with titres of 4 to 16, matched by age and sex with the sero-positive group, were drawn at random from the 1954 and 1956 surveys, and were followed up in the same way. In the 1961 follow-up, clinical, serological, and radiological assessments were made without knowledge of the previous sheep cell agglutination titre. Since essentially the same conclusions emerged from the 1959 and 1961 follow-ups, the two separate studies have been grouped together to facilitate analysis.

Results

(1) Composition of Initial Samples and Completion Rate

In the 1954 sample, there were in each of the three groups nineteen individuals, giving a total of 57, of whom four had died, 51 were re-examined, and two had moved out of the area or refused to co-operate further.

In the 1956 sample, there were 26 individuals in each of the three groups, giving a total of 78, of whom thirteen had died and eight had moved out of the area or did not co-operate. Thus, of the 135 under review, 108 persons were re-examined, 51

having a clinical examination, x rays, and a blood test, 46 a clinical examination and blood test, and eleven a clinical examination only. The death and refusal rates were similar in the sero-positive and sero-negative groups (Table I).

Of the 45 initially sero-positive individuals (Table II), seven had both clinical and radiological evidence of rheumatoid arthritis, three had clinical disease only, and nine had radiological changes only. This is a higher proportion of persons with rheumatoid arthritis than is usually found amongst sero-positive individuals in population samples, and must be attributed to the age distribution, 57 of the 135 individuals being between the ages of 55 and 64. At this age an association between rheumatoid factor and arthritis is more likely to be found. Of the seven who died in the sero-positive group, only one had had clinical arthritis. None of the sero-negative arthritics died. The mortality was 6 per cent. in those with clinical arthritis, 14 per cent. in those with radiological disease, and 13 per cent. where neither clinical nor radiological signs were present. None

of those with clinical arthritis refused re-examination or were not available, but one with radiological disease and nine with no disease were not re-examined. This is in conformity with our previous experience of random samples that those without rheumatic complaints are more likely to refuse.

(2) Composition of Samples at Follow-up

Sheep Cell Agglutination Test.—Of the 35 persons with a positive sheep cell agglutination test who were re-examined, six refused to have a second blood test and in one it was not possible to obtain a sample. Of the remaining 28, 21 remained positive and seven became negative, four dropping to a titre of <4 (Table III, opposite). These four all had minimal positive titres initially. Only one with an initial titre of more than 32 became negative and this one fell insignificantly to a borderline titre of 16. Of the 28 positives the titre fell significantly (two tubes or more) in nine and rose in six.

This behaviour contrasts with the negative group.

TABLE I
COMPLETION RATE, BY INITIAL SHEEP CELL AGGLUTINATION TITRE

Initial Sample		Completion at 5-Year Follow-up				
Sheep Cell Agglutination Titre	No. of Cases	Re-examined Clinically, by X ray and Blood Test	Clinical and Blood Test Only	Clinical Only	Died	Refused or Not Available
32+	45	16	12	7	7	3
4 to 16	45	18	16	2	5	4
<4	45	17	18	2	5	3
Total	135	51	46	11	17	10

TABLE II
INITIAL SAMPLES BY CLINICAL AND RADIOLOGICAL CRITERIA

Sheep Cell Agglutination Titre	Signs of Rheumatoid Arthritis	No. of Cases	Follow-up		
			Died	Refused or Not Available	Re-examined
32+	Clinical and Radiological	7	1	0	6
	Clinical Only	3	0	0	3
	Radiological Only	9	2	0	7
	None	26	4	3	19
	Total	45	7	3	35
4-16	Clinical and Radiological	1	0	0	1
	Clinical Only	2	0	0	2
	Radiological Only	6	1	1	4
	None	36	4	3	29
	Total	45	5	4	36
<4	Clinical and Radiological	1	0	0	1
	Clinical Only	4	0	0	4
	Radiological Only	4	0	0	4
	None	36	5	3	28
	Total	45	5	3	37
Total No. of Cases		135	17	10	108

TABLE III
COMPARISON OF INITIAL SHEEP CELL AGGLUTINATION TITRES WITH THOSE FOUND AFTER 5 YEARS*
Sheep Cell Agglutination Titre, 1959-61

	<4	4	8	16	32	64	128	256	512	1,024	Total
<4	34			1							35
4	9	4	1	1							15
8	10	1	1	1	2						15
16	2		2								4
32	4	1	1		2		1				9
64						4		2			6
128				1	1	1	1	2	1	1	8
256							1	2		1	4
512								1			1
1,024											—
Total	59	6	5	4	5	5	4	6	1	2	97

* Figures outside dotted lines refer to cases with a 4-fold or greater change in titre.

Only two of 69 whose test was initially negative became positive, the titre rising from eight in the initial test to 32 after 5 years. Amongst the 34 persons with initial titres of 4 to 16, there was a significant fall in titre in twelve and a significant rise in three after 5 years; in all, 21 of them had dropped to <4. Of the 35 with an initial titre of <4, all but one had remained at <4.

Clinical Rheumatoid Arthritis.—Of the 35 persons with a positive sheep cell agglutination test who were re-examined, nine had clinical evidence of rheumatoid arthritis initially (Table IV). On follow-up at 5 years all of these still had the disease and four others had developed it in the intervening

period. A fifth developed signs of lupus erythematosus. Of 36 persons with a sheep cell agglutination titre of 4 to 16 who were re-examined, three had clinical disease initially. On follow-up, two of these still had the disease and one had recovered. Of the 37 with a sheep cell agglutination titre of <4, five had clinical arthritis initially. On the 5-year follow-up two of these five still had definite disease, the other three having recovered. In addition, one person with an initial titre of <4 without any clinical disease had developed it. Of the above three who had recovered, one had developed generalized osteo-arthritis, one was graded as having doubtful arthritis on follow-up, and the third had recovered completely. Of the seven in

TABLE IV
PRESENCE OF CLINICAL RHEUMATOID ARTHRITIS INITIALLY AND AFTER 5 YEARS

Initial Assessment			Re-assessment After 5 Years					
Sheep Cell Agglutination Titre	Clinical Rheumatoid Arthritis	Total Re-assessed	S.C.A.T. Positive		S.C.A.T. Negative		S.C.A.T. Not Tested	
			Clinical Rheumatoid Arthritis		Clinical Rheumatoid Arthritis		Clinical Rheumatoid Arthritis	
			Present	Absent	Present	Absent	Present	Absent
Positive 32+	Present ..	9	6	0	1	0	2	0
	Absent ..	26	3	12	1	5	0	5
Negative 4-16	Present ..	3	1	0	1	1	0	0
	Absent ..	33	0	1	0	30	0	2
Negative <4	Present ..	5	0	0	2	3	0	0
	Absent ..	32	0	0	1	29	0	2
Total No. of Cases ..		108	10	13	6	68	2	9

whom the sheep cell agglutination titre reverted from positive to negative, one continued to show evidence of rheumatoid arthritis, five remained free of it, and one (who previously was considered to have only generalized osteo-arthritis) was graded on follow-up as having minimal rheumatoid arthritis as well.

Radiological Evidence.—Of the sixteen persons with a positive sheep cell agglutination test who were re-examined radiologically, only three initially had erosive changes (in the feet in two and in the hands, feet, and neck in the other). After 5 years these three still had erosions and five others had developed them. Of these eight persons, four had erosions in the hands, six in the feet, and three in the neck (Table V). None of the five in this radiological series whose sheep cell agglutination titre became negative developed radiological changes. Of the eighteen persons with sheep cell agglutination titres of 4 to 16 who were re-examined radiologically, one had erosive changes in the hands and one in the feet. After 5 years, four persons in all were affected. One still had changes in the hands but two had them in the feet, and one now had changes in the cervical spine. Of the seventeen persons with a sheep cell agglutination titre of <4 one initially had changes in the feet and one in the cervical spine. After 5 years the latter had developed changes also in the hands; one other had developed changes in the hands and three had developed them in the cervical spine. Thus six persons were affected at the end of 5 years.

Taking clinical and radiological disease together, the sero-positive group included seven new cases of rheumatoid arthritis in nineteen who had not

previously had either clinical or radiological rheumatoid arthritis, one new case in a person with a sheep cell agglutination titre of 4 to 16 out of 29 who had not previously had either clinical or radiological rheumatoid arthritis, and four new cases amongst the 28 persons with a sheep cell agglutination titre of <4 who had previously no evidence of rheumatoid arthritis. The difference between sero-positives and sero-negatives is highly significant ($\chi^2 = 6.4$ $P \approx 0.01$). Of the seven sero-positives who developed the disease during the 5-year period, two had developed clinical disease and five only radiological disease. In addition, one who had previously had only radiological disease (in one-third metatarsalphalangeal joint) developed clinical disease during the 5-year period. The patient with a sheep cell agglutination titre of 4 to 16 who developed arthritis developed only radiological disease. Of the four new cases with a sheep cell agglutination titre of <4, three had radiological signs only, and one both clinical and radiological.

Other Diseases.—If genetic factors play a part in determining the sheep cell titre, it may be expected, in view of the manifest disadvantage of possessing a positive test, that some advantage also accrues. If dominant genes are involved this advantage might apply to homozygotes or to heterozygotes. A careful history was therefore taken of illness occurring during the 5-year follow-up, particular attention being paid to infections, in view of the known association of the sheep-cell factor with γ globulin. The history before the first test might also be expected to be important but, as this was not realized in 1954-56, no special care was taken to

TABLE V
PRESENCE OF RADIOLOGICAL EROSION ARTHRITIS INITIALLY AND AFTER 5 YEARS*

Initial Assessment			Re-assessment After 5 Years					
Sheep Cell Agglutination Titre	Radiological Erosive Arthritis	Total Re-assessed	S.C.A.T. Positive		S.C.A.T. Negative		S.C.A.T. Not Tested	
			Radiological Erosive Arthritis		Radiological Erosive Arthritis		Radiological Erosive Arthritis	
			Present	Absent	Present	Absent	Present	Absent
Positive 32+	Present	3	3	0	0	0	0	0
	Absent	13	5	3	0	5	0	0
Negative 4-16	Present	2	0	0	2	0	0	0
	Absent	16	1	1	1	13	0	0
Negative <4	Present	2	0	0	2		0	0
	Absent	15	0	0	4	11	0	0
Total No. of Cases			9	4	9	29	0	0

* Only individuals x rayed on both occasions are included in this Table.

elicit such a history. Information on infections before the first test is therefore of necessity incomplete and in fact showed no significant differences between sero-positive and sero-negative individuals. During the 5-year follow-up period, three of those with a titre of 4 to 16 and two of those with a titre of <4 had an attack of influenza, but none of the sero-positive persons gave such a history. Three of the sero-positives had an eruption suggestive of lupus erythematosus. One of these had developed a recurrent peeling eruption on the fingers which was photosensitive. The second had had an eruption on the forehead and cheeks for 1 month which resembled lupus erythematosus. The third had had since the age of 12 a dry eczema of the hands and feet which was photosensitive.

Discussion

The present larger study confirms our tentative suggestion (Ball and Lawrence, 1961) that persons with a positive sheep cell agglutination test are more likely to develop rheumatoid arthritis. This is, however, contrary to the findings of Aho, Kirpilä,

and Wager (1959). Of seven patients with false positive sheep cell agglutination test followed for 9 years by these workers, none developed rheumatoid arthritis. Four, however, became sero-negative.

In the second half of this follow-up the use of x rays both initially and after 5 years proved particularly revealing and demonstrated that erosions frequently develop though they are always small in the absence of clinical manifestations. This could be observed with great clarity in the heads of the metatarsal bones. It explains the frequent finding of small erosions in persons without a history of polyarthritis in population samples and indicates that these lesions are probably rheumatoid in nature. Examples of erosions developing in a sero-positive female are shown in Fig. 1, and in Fig. 2 (overleaf).

The high incidence of rheumatoid arthritis in sero-positive individuals could be explained in two ways:

- (1) The positive sheep cell agglutination test is associated in some way with a predisposition to the disease;

Fig. 1.—X rays of fifth metacarpal joint, (a) initially and (b) at 5-year follow-up, showing development of erosion at the articular margin. There was no clinical evidence of rheumatoid arthritis initially, but 3 years later the patient had definite symptoms and signs.

Fig. 2.—X rays of wrist of same patient as in Fig. 1, (a) initially and (b) at 5-year follow-up, showing loss of joint space, development of bone cyst in radius and osteophyte on lunate.

- (2) In those who “developed” the disease during the 5-year period, it was actually present on the occasion of the first survey, but was not recognized.

If the second explanation is correct it would be expected that the titre would rise in those thirteen who apparently developed the disease. In fact the titre went up in four and down in four, remaining unchanged in five. A two-tube difference upwards occurred in one and downwards in three. In none did the test change from negative to positive with development of the disease. In one case it went from positive to negative. This person, a female aged 72, had generalized osteo-arthritis with Heberden's nodes and this may have obscured the rheumatoid disease on the first assessment. In the remainder, however, it would seem unlikely that a positive sheep cell agglutination test was due to unrecognized disease. It must therefore be concluded that the high incidence in sero-positives, indicating a predisposition in those with positive tests.

Of the sixteen with sero-negative rheumatoid arthritis who were re-examined (Table II), only one (6 per cent.) developed a positive test. This one was more severe than the others and had initially a borderline sheep cell titre of 1 : 16. Of the 55 sero-negative non-rheumatoids who were re-tested

serologically, one (2 per cent.) had developed a positive test but continued to show no evidence of the disease. This may be compared with a follow-up of 50 hospital patients with sero-negative rheumatoid arthritis by Dixon (1960), in which twelve (24 per cent.) became positive. Since all these sera were tested in the same laboratory, the results should be comparable. It is clear that conversion to positive occurs more often in rheumatoid subjects than in non-rheumatoids and that it is more likely to happen in patients ill enough to require hospital treatment, though within the hospital group Dixon found no obvious relationship to severity. Indeed, he found a persistently negative sheep cell agglutination test compatible with a progressive course and fatal outcome. Dixon's results would suggest that the sheep cell factor may sometimes be produced by the rheumatoid process. This is in accordance with the findings of de Forest, Mucci, and Boisvert (1958), who reported conversion of positive to negative in patients undergoing remission. It may be concluded that the sheep cell factor is both associated with a predisposition to, and results from, the disease. In confirmation of this it may be noted that, whereas there was an excess of positive tests in the families of sero-positive individuals taken from the random sample in Leigh, no such familial aggregation was found in the families of

sero-positive hospital patients with Still's disease (Lawrence and Ball, 1958; Ansell, Bywaters, and Lawrence, 1961). Other evidence in favour of this conception of the sheep cell factor as a biological palindrome has been produced elsewhere (Lawrence, 1962).

Of the sixteen sero-negative rheumatoids who were re-examined (Table II), eight had clinical disease initially and four of these went into remission. In none of the sero-positive persons with clinical rheumatoid arthritis did the disease go into remission. This confirms the findings of others that the prognosis is worse in rheumatoid subjects with a positive sheep cell agglutination test.

When the sheep cell agglutination test is converted from positive to negative, the prognosis seems to be improved. Of the seven who did so, five remained free of the disease both clinically and radiologically, one continued to have both clinical and radiological signs, but clinically was improved, and one had generalized osteo-arthritis with superadded arthritis and showed no evidence of improvement.

Though the sheep cell agglutination test has been found positive more often in the older age groups in population samples, there is no evidence from this study that the sheep cell titre increases with age in the individual. Indeed, the general trend was towards a lower titre on follow-up, fourteen showing a higher and 35 a lower titre ($\chi^2 = 8$; $P < 0.01$).

These results support the hypothesis of a secular change in the sheep cell titre. This would bring the sheep cell factor into line with streptococcal and tuberculous infections which have also declined during the 20th century. It is perhaps pertinent that certain streptococci have been found capable of inducing a positive sheep cell agglutination test in experimental animals (Svartz, 1961).

Summary

A series of 45 persons with a positive sheep cell titre (1 : 32 or over) taken from a random population sample has been re-examined after a lapse of 5 years. Comparison has been made with 45 persons with a titre of 4 to 16 and with 45 others having a titre of <4 from the same population sample matched by age and sex to the positive group.

Among those re-examined, clinical or radiological evidence of rheumatoid arthritis was present originally in sixteen of the sero-positive group and in seven and nine respectively of the sero-negative groups. Seven of the sero-positive and five of each of the sero-negative groups had died during the 5-year period.

Of nineteen sero-positive subjects who initially had neither clinical nor radiological evidence of rheumatoid arthritis, seven were found to have developed evidence of the disease on follow-up compared with five of the 57 sero-negative subjects initially without disease.

Of 97 persons in whom the sheep cell agglutination test was repeated after 5 years, ten had a higher titre, and 21 a lower titre by two or more tubes. Seven converted from positive to negative and two from negative to positive.

REFERENCES

- Aho, K., Julkunen, H., Laine, V., Ripatti, N., and Wager, O. (1961). *Acta rheum. scand.*, 7, 201.
 —, Kirpilä, J., and Wager, O. (1959). *Ann. Med. exp. Biol. Fenn.*, 37, 377.
 Ansell, B. M., Bywaters, E. G. L., and Lawrence, J. S. (1961). "Atti del X Congresso della Lega Internazionale contro il Reumatismo", vol. 2, p. 263.
 Ball, J., and Lawrence, J. S. (1961). *Ann. rheum. Dis.*, 20, 235.
 Dixon, A. St. J. (1960). *Ibid.*, 19, 209.
 de Forest, G. K., Mucci, M. B., and Boisvert, P. L. (1958). *Arthr. and Rheum.*, 1, 387.
 Lawrence, J. S. (1962). *Amer. J. publ. Hlth*, 52, 1689.
 — and Ball, J. (1958). *Ann. rheum. Dis.*, 17, 160.
 Mikkelsen, W. M., Dodge, H. J., Duff, I. F., Epstein, F. H., and Napier, J. A. (1962). *Arthr. and Rheum.*, 5, 117.
 Svartz, N. (1961). "Atti del X Congresso della Lega Internazionale contro il Reumatismo", vol. 1, p. 57.

Rapport du facteur rhumatismal sérique à l'arthrite rhumatismale—un échantillon d'une population re-examiné au bout de cinq ans

RÉSUMÉ

Un groupe de 45 personnes donnant une réaction de Waaler-Rose positive (à partir du titre 1/32) et appartenant à un échantillon d'une population, fut re-examiné après un intervalle de 5 ans. On compara ce groupe à deux autres, aussi de 45 personnes et appartenant au même échantillon, mais ajustés en sa composition de manière à faire correspondre les trois groupes en ce qui concerne l'âge et le sexe des sujets. Dans l'un groupe témoin les sujets donnaient la réaction de Waaler-Rose aux titres de 1/4 à 1/16 et dans l'autre ils la donnaient à une dilution inférieure à 1/4.

Des preuves cliniques et radiologiques d'arthrite rhumatismale furent présentes au premier examen chez 16 sujets du groupe séropositif et chez 7 et 9 sujets respectivement des groupes séronégatifs. Sept personnes du groupe séropositif et cinq de chaque groupe séronégatif décédèrent au cours de la période de 5 ans.

Sur 19 sujets séropositifs qui n'avaient pas présenté à l'examen initial de signes cliniques ou radiologiques d'arthrite rhumatismale, sept d'entre eux ont accusé de tels signes cinq ans plus tard. Sur 57 sujets séronégatifs sans signes de maladie au début, 5 sujets l'ont présenté au second examen.

Parmi 97 personnes chez qui la réaction de Waaler-Rose a été répétée après cinq ans, chez dix d'entre elles l'agglutination s'est produite à une dilution supérieure et chez 21 d'entre elles à une dilution deux ou plus fois inférieure. Une conversion de la réaction positive à la négative s'est produite chez sept sujets et dans le sens inverse chez deux sujets.

Relación del factor reumatoide sérico con la artritis reumatoide—muestra de una población re-examinada después de 5 años

SUMARIO

Un grupo de 45 personas con reacción de Waaler-Rose positiva (desde una dilución de 1/32), perteneciendo a una muestra de una población, fué re-examinado cinco años más tarde. Se comparó este grupo a otros dos también de 45 personas y de la misma muestra, pero ajustados en su composición para que correspondan

entre sí por la edad y el sexo. En un grupo testigo los sujetos daban una reacción de Waaler-Rose en diluciones de 1/4 a 1/16 y en el otro en dilución inferior a 1/4.

Evidencia clínica y radiológica de artritis reumatoide fué presente en el examen original en 16 sujetos del grupo seropositivo y en 7 y 9 sujetos respectivamente de los grupos seronegativos. Siete personas del grupo seropositivo y cinco de cada grupo seronegativo murieron durante el período de 5 años.

De 19 sujetos seropositivos que inicialmente no presentaron indicios clínicos o radiológicos de artritis reumatoide, siete evidenciaron tales síntomas cinco años más tarde. De 57 sujetos seronegativos sin síntomas de esta enfermedad al principio, cinco los presentaron al segundo examen.

Al repetir la reacción de Waaler-Rose en 97 personas después de cinco años, se notó que en diez de ellas la aglutinación se produjo en una dilución superior y en veintiuno de ellas en una dilución dos o más veces inferior. Una conversión de la reacción positiva a la negativa sobrevino en siete sujetos y lo contrario en dos.