Heberden Society

Clinical meeting, Belfast, September, 1971

Enhanced Catabolic Rate and a Structural Anomaly in the Serum IgG of RA Patients. By J. WATKINS and A. J. SWANNELL (*M.R.C. Rheumatism Unit, Canadian Red Cross Memorial Hospital, Taplow*).

Studies of IgG catabolism in patients with RA and SLE have indicated that they may possess enhanced immunological mechanisms for the removal of this protein. This communication reports a preliminary study of IgG catabolism in RA and in health and a parallel study of the catabolism of human IgG proteins in mice.

Six patients with seropositive rheumatoid arthritis who were not receiving gold or corticosteroids, were studied. Immunologically pure IgG was isolated from their serum by column chromatography on DEAE-cellulose. Each immunoglobulin preparation was labelled with radioiodine and injected back intravenously into the patient. Some preparations were also injected intraperitoneally in a batch of mice (20 male; 20 female). The elimination of the isotope was followed by scintillation counting of serum samples (in man) and by whole radiation measurements (in mice). Biological half-lives were measured over 32 days in man and 14 days in the mouse.

Individual half-life values in man are shown in Table I.

TABLE I Individual half-life values

Subjects	Age (yrs)	Duration of disease	State of disease	Terminal biological half-life (days)
RA Patients				
M. N. (F)	42	5 yrs	'Inactive'	20.0
H. T. (M)	32	1 yr	'Inactive'	22.4
I. R. (F)	50	11 yrs	Flare	12.0
R. W. (M)	65	8 wks	Early acute	13.0
E. S. (F)	35	6 mths	Early acute	14.0
D. W. (M)	59	6 yrs	Flare	14.0
Healthy individuals				
3F, 2M	21-48	—	—	19-24

It appears significant that an increased catabolic rate is observed in those patients with early or active disease.

In mice human IgG is eliminated by either normal (log linear) mechanisms or by "immune type" mechanisms. With IgG obtained from healthy volunteers the log linear pattern predominates: only a small percentage of mice show immune phenomena (Table II). In contrast IgG samples from two out of three RA patients elicited an immune response in a significantly higher proportion of mice.

TABLE II Mode of catabolism in mice of IgG from normal and rheumatoid subjects

Source of IgG	Proportion of mice showing immune phenomenon (per cent).	
	Male	Female
Sex		
Healthy donors		
Single donor	7	4
Donor pool	0	7
RA patients		
Donor 1	22	44
Donor 2	10	53
Donor 3	0	11

These experiments indicate that rheumatoid IgG may be more immunogenic than IgG from healthy volunteers. This suggests a structural anomaly in the IgG of rheumatoid patients which may influence its rate of elimination from the body. The observation that female mice show greater immune awareness of rheumatoid IgG than male mice may imply that a sex linked factor influences antigenic recognition.

Discussion

DR. W. W. BUCHANAN (*Glasgow*) Could it be that the increased elimination of rheumatoid IgG in rheumatoid patients merely represents a hypermetabolic state? It is known, for instance, that there is an increased turnover of albumin in rheumatoid patients.

It would also be of interest to know whether your mice would show 'immune' elimination of rheumatoid albumin. Your results are interesting but the interpretation of their immunological significance requires caution.

DR. SWANNELL Our results show an increased catabolic rate for IgG in RA patients; this would certainly suggest an increased rate of metabolism. However, the increased rate of catabolism is not due to elevation of the serum IgG levels. Two of our healthy controls had higher IgG levels than the RA patients.

We are suggesting that a minor structural alteration of rheumatoid globulin makes the protein more antigenic in mice. It is unnecessary for such protein to bind with RF in order to produce pathological complexes.

DR. W. W. BUCHANAN (*Glasgow*) How would these mice handle another IgG antibody (say tetanus antibody). Would they regard rheumatoid albumen differently from normal?

DR. SWANNELL We have no data at present on the handling of such IgG antibodies in the mouse. It is surprising that the mouse will tolerate human IgG at all. We have no knowledge of its tolerance to other human serum proteins.

PROF. E. G. L. BYWATERS (*Taplow*) These studies are preliminary and, while the results are clear, their significance is not yet clear.

PROF. D. L. GARDNER (*London*) Why did you use twenty times the dose of labelled material in mice compared to man.

DR. SWANNELL The short (5-day) half-life of IgG in the mouse makes a large radioactive label desirable for accuracy of measurement.

DR. H. L. F. CURREY (*London Hospital*) Immune elimination of rheumatoid, as opposed to non-rheumatoid, IgG would raise the interesting possibility that rheumatoid IgG is more antigenic. Did you study the development of anti-globulins in the mouse?

DR. SWANNELL We attempted to verify apparent immune responses by a second challenge with human IgG.

Female mice classified as immune after the first exposure to the antigen almost invariably showed a secondary response, indicating the presence of antibodies. Results in male mice were more complex, suggesting an immune recognition without antibody production.

Serum and Synovial Fluid Complement in Rheumatoid and Other Arthritides. By J. WEBB and R. G. ROBINSON (*Sutton Rheumatism Research Laboratory, The Royal North Shore Hospital of Sydney, Australia*).

Total haemolytic complement in serum and synovial fluid samples, which were paired whenever possible, was estimated by a method modified from Kabat and Mayer (1961). A normal serum range was established in 42 healthy persons of 185 to 345 C'H₅₀ units/ml (mean 265 ± 2 SD). The rank sum test was applied to the results for statistical evaluation (Table).

TABLE Mean serum and synovial fluid complement levels with standard deviations (SD) and numbers in each of the various disease groups

Disease group	Haemolytic complement levels					
	Serum			Synovial fluid		
	No.	Mean	S.D.	No.	Mean	S.D.
Normal	42	265	40			
Osteoarthritis				30	60	
Acute gout	4	400	55	7	170	45
Reiter's disease	16	390	60	16	190	60
Polyarthritis nodosa	9	405	60			
Progressive systemic sclerosis	10	290	60			
Ankylosing spondylitis	10	285	65	8	70	35
Systemic lupus erythematosus				7	10	5
Rheumatoid arthritis						
Total	107	310	85	81	65	50
(i) seronegative	25	335	75	20	90	55
(ii) seropositive	64	315	75	48	65	45
(iii) with arteritis	18	245	85	13	25	35

SERUM The mean levels in ankylosing spondylitis (285) and progressive systemic sclerosis (290) did not differ from those in normal subjects (260). There were significant elevations in gout (400), Reiter's disease (390), polyarthritis nodosa (405), and rheumatoid arthritis (310). Within the group of rheumatoid arthritides, the levels in those with complicating arteritis (245) were significantly lower than in other seropositive (315) or seronegative (335) cases.

SYNOVIAL FLUID The mean levels in inflammatory rheumatoid disease (65) and ankylosing spondylitis (70) effusion did not differ from those in non-inflammatory osteoarthritis (60). Levels were high in gout (170) and Reiter's diseases (190) and very low in systemic lupus erythematosus (10). When arteritis complicated rheumatoid arthritis (25), the levels were significantly lower than in cases of uncomplicated rheumatoid arthritis (75).

RHEUMATOID PLEURAL EFFUSION Very low complement levels were found in three cases (5, 10, and 15).

There was a significant correlation between complement levels in paired serum and synovial fluid in cases of rheumatoid arthritis (rank correlation coefficient, $R = 0.658$). However, the strength of this correlation is not great because of wide scatter and is by no means predictive. There was no correlation between complement and

rheumatoid factor levels in either serum or synovial fluid.

These findings support the evidence for local auto-immune mechanisms in rheumatoid synovitis, and provide some evidence for more general autoimmune disease when systemic arthritis complicates rheumatoid disease.

Reference

Kabat, E. A., and Mayer, M. M. (1961) 'Experimental Immunology', 2nd ed., p. 149. Thomas, Springfield, Ill.

Discussion

DR. A. G. S. HILL (*Stoke Mandeville*) Was the synovial fluid complement level lower than the serum level in all groups?

DR. WEBB Yes.

DR. A. G. S. HILL (*Stoke Mandeville*) Presumably this is unlikely to be due to consumption of complement from the joint fluid: there is a diffusion barrier?

DR. WEBB This is probably related to lower protein levels in synovial effusions. We have not correlated protein and complement levels but they do correlate in normal subjects.

Calcium Pyrophosphate Deposits in Synovial Membrane. By E. G. L. BYWATERS (*M.R.C. Rheumatism Unit, Canadian Red Cross Memorial Hospital, Taplow*).

Crystals of calcium pyrophosphate are known to occur in chondrocalcinosis or pseudogout (Žitnan and Šit'aj, 1963), in articular cartilage, synovial fluid, and synovial membrane (McCarty and Hollander, 1961), in hypophosphatasemia (O'Duffy, 1970), in haemochromatosis in similar locations (Atkins, McIvor, Smith, Hamilton, and Williams, 1970) as well as in the intervertebral discs in that condition and in hyperparathyroidism, and in ochronotic synovial membrane (Bywaters, unpublished data).

We now record calcium pyrophosphate crystal deposition in synovial membranes of three out of 43 consecutive rheumatoid arthritis patients removed during arthroplasties, and in two more since then. Each of the three patients had had chronic progressive erosive seropositive polyarthritis for over 12 years and each had shown the changes expected of rheumatoid arthritis in the tissues removed at arthroplasty. What was not expected was the presence of pyrophosphate. In one case, this was detected only microscopically: positively birefringent crystals were embedded in a basophil matrix and enclosed in giant cells, multinucleate and with eosinophil cytoplasm. In the second case, these deposits were numerous and associated with hydroxyapatite deposition in the synovial membrane. In the third case (who had had rheumatoid arthritis for 40 years, since the age of 30 with erosions, nodules, a raised erythrocyte sedimentation rate, and a positive latex test) arthroplasty showed an eroded joint covered with calcific deposits, visible grossly and also radiologically and microscopically. She had had neither steroid, gold, nor local cortisone injections. Serum calcium, phosphorus, and phosphatase were normal. Such crystal deposits occurred mainly in avascular villi and in joints with a considerable accumulation of necrotic bone fragments.

The wide spectrum of diseases with calcium pyrophosphate crystal deposition seems to imply an innate tendency for this to occur in the synovial milieu.

While in particular patients this might be due to hereditarily low phosphatase (O'Duffy, 1970), in others it