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Studies on cold insoluble globulin

I. Concentrations in citrated plasma in rheumatic disorders

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SUMMARY Cold insoluble globulin (CIG) is a normal glycoprotein of human serum and plasma. The physiological significance of this protein is unknown, but it shows a temperature-dependent relation to fibrinogen and fibrin. It is possible that it represents a substrate for activated fibrin-stabilising factor in the polymerisation of fibrin. CIG is found on the surface of fibroblasts. In the present study CIG was estimated in citrated plasma in 115 patients with rheumatic diseases. Increased amounts were found in patients with systemic lupus erythematosus, secondary amyloidosis in classical and definite rheumatoid arthritis, and in male patients with juvenile rheumatoid arthritis.

Cold insoluble globulin (CIG) is a normal glycoprotein of human serum and plasma (Blombäck and Blombäck, 1956; Mosesson and Umfleet, 1970). In gel electrophoresis it moves between the $alpha_2$- and the $beta_1$- globulin bands (Laurell, 1972). It consists of at least 2 polypeptide chains bound together with disulphide bridges and with a molecular weight of about 440 000 daltons (Mosesson *et al.*, 1975; Fyrand and Solum, 1976b). CIG is found on the surface of fibroblasts and glial cells (Ruoslahti and Vaheri, 1975). It represents the main non-clottable protein of heparin precipitable fraction (Fyrand and Solum, 1976b). The physiological significance of CIG is unknown. Only a few clinical studies on CIG have been published, most of them reporting a fairly constant plasma level of this protein (Aronsen *et al.*, 1972; Johansson *et al.*, 1972; Häller and Laurell, 1972; Fyrand, 1977).

Because of the relationship between CIG and fibroblasts, studies of CIG in connective tissue diseases may be important. In this study, plasma concentrations of CIG in patients with different rheumatic disorders have been investigated.

Material

One hundred and fifteen inpatients (67 women and 48 men) with various rheumatic disorders have been investigated. The diagnoses were made according to the definitions of the American Rheumatism Association (Bennett and Burch, 1967).

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At the time of investigation, all patients were receiving various medications (analgesics, anti-inflammatory and cytostatic agents). Electroimmunoassay was performed for the estimation of the levels of CIG in plasma. The reference and the control groups for this assay are described elsewhere (Fyrand and Solum, 1976a).

Methods

The production of citrated plasma and the procedure of electroimmunoassay is described elsewhere (Fyrand and Solum, 1976a). Blood haematocrit was measured in all patients, and in 7 patients with low values a correction of the CIG concentrations was made to the lowest level of the normal haematocrit distribution (females 35–44%, males 39–48%). Student's t test was used for statistical evaluation. In larger groups of patients (classical and definite rheumatoid arthritis), sex- and age-related control groups were used. In smaller groups, only sex-related control groups were used, as in juvenile rheumatoid arthritis no corresponding age groups were available in the control material.

Results

The largest group investigated was that of typical rheumatoid arthritis. No significant increase in plasma CIG concentration was found in this disease compared to that of our normal material. Because of the significantly lower CIG concentration in the female control group below 50 years the females

Table *CIG in citrated plasma in 115 patients with rheumatic diseases*

	Women				*Men*			
	No.	*Age*	*CIG (U/ml)*	*Significance*	*No.*	*Age*	*CIG (U/ml)*	*Significance*
		Mean ± SD	*Mean ± SD*			*Mean ± SD*	*Mean ± SD*	
Rheumatoid arthritis (classical and definite)								
Women < 50 years	15	36·7 ± 10·3	112·5 ± 29·7	NS				
> 50 years	21	60·0 ± 5·2	120·9 ± 34·7	NS				
Men All age groups					24	52·0 ± 14·8	127·4 ± 35·4	NS
Secondary amyloidosis in rheumatoid arthritis (classical and definite)	3	52·7 ± 9·0	194·0 ± 69·1	$P < 0·001$	1	23·0	197·0	
Spondylitis ankylopoetica					5	50·2 ± 10·7	138·4 ± 29·7	NS
Juvenile rheumatoid arthritis	14	17·7 ± 14·0	110·3 ± 19·1	NS	6	9·1 ± 3·7	136·0 ± 16·8	$P < 0·025$
Psoriatic arthritis	4	44·3 ± 14·2	101·5 ± 33·0	NS	9	52·4 ± 10·3	131·0 ± 31·2	NS
Systemic lupus erythematosus	6	44·7 ± 16·0	140·0 ± 25·5	$P < 0·01$	1	26·0	140·0	
Progressive systemic sclerosis	4	44·8 ± 16·4	106·5 ± 13·9	NS	2	23·0	101·2 ± 33·5	NS

with typical rheumatoid arthritis were sub-divided by age (below and over 50). Still no significant deviation from normal values was observed.

Significantly raised levels of CIG were found in the male group of juvenile rheumatoid arthritis ($P < 0.025$) in contrast to the corresponding female group. An increase was also found in female patients with secondary amyloidosis in rheumatoid arthritis ($P < 0.001$) and in females with systemic lupus erythematosus (SLE) ($P < 0.01$). The corresponding male groups were too small for statistical evaluation, but the single male patient in each group demonstrated a raised level of CIG (197 U/ml and 140 U/ml, respectively). Plasma CIG levels in ankylosing spondylitis, psoriatic arthritis, and progressive systemic sclerosis did not differ significantly from the control groups.

Discussion

Concentrations of CIG in healthy persons is dependent upon age and sex (Fyrand and Solum, 1976a). Women below 50 years demonstrate significantly lower levels of CIG compared with women over 50 ($P < 0.01$) and with all male groups ($P < 0.02$). In men increased concentrations are found in higher age groups but the difference is not statistically significant. In women over 50 plasma CIG levels are not different from those of the corresponding male group (Fyrand and Solum, 1976a).

CIG is present on the surface of fibroblasts (Ruoslahti and Vaheri, 1975). When the cells are cultured *in vitro*, CIG is found in the medium. When these fibroblasts are transformed by addition of Rous' sarcoma virus, CIG disappears from the surface (Vaheri and Ruoslahti, 1974). It may be that fibroblasts represent the source of CIG in blood (Ruoslathi and Vaheri, 1975). Studies on CIG in diseases affecting connective tissue may therefore be of importance, as indicated by increased amounts of CIG in plasma reported in this study. As to increased amounts of plasma CIG in secondary amyloidosis, recent reports demonstrate that fibroblast may be responsible for the production of amyloid (Natvig *et al.*, 1977; Runne and Orefanos, 1977).

Lower CIG values in the female group of juvenile rheumatoid arthritis may be explained by a higher age level of the patients in this group compared with the male group, and may therefore represent a higher incidence of 'burned out' cases. Levels of CIG in plasma are considerably lower in newborns, but have not been reported in healthy children, and this may also influence the statistical significance of patients with juvenile rheumatoid arthritis. As there is a tendency towards lower levels of CIG in plasma in lower age groups (Fyrand and Solum, 1976a/b; Ganrot, 1972), adequate control groups for juvenile patients would probably give lower P values than those indicated in this study in juvenile rheumatoid arthritis.

Further studies on CIG in plasma in these subgroups (SLE, juvenile rheumatoid arthritis, secondary amyloidosis) compared with corresponding age and sex control groups should be undertaken.

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