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Measurement of serum proteins during attacks of ulcerative colitis as a guide to patient management

N. A. BUCKELL, J. E. LENNARD-JONES, M. A. HERNANDEZ, J. KOHN, P. G. RICHES, AND J. WADSWORTH

From St Mark's Hospital, London, The London Hospital, Whitechapel, London, and Supraregional Protein Reference Unit, Putney Hospital, London, and the Computing Unit for Medical Sciences, St Bartholomew's Hospital, London

SUMMARY Serial measurements of 11 serum proteins have been made throughout 39 admissions of 36 patients to hospital for the treatment of acute attacks of ulcerative colitis. There was a striking correlation between rapid changes in C-reactive protein and pre-albumin concentrations and the clinical response to medical treatment. Measurements of the α_1-acid glycoprotein, albumin, and total serum protein concentrations at the time of admission were found to correlate with the outcome of the attack. Measurement of these proteins provides a useful guide to the management of patients with attacks of ulcerative colitis.

Any protein concentration observed in the serum depends on synthesis, catabolism, loss, and distribution within the body compartments. The synthesis of several serum proteins, the 'acute-phase reactants', increases in response to many stimuli including inflammation (Koj, 1974). Increased serum concentrations of many of these proteins have been reported in active ulcerative colitis and their concentrations rise as the activity of the colitis increases (Cooke *et al.*, 1958; Dearing *et al.*, 1969; Weeke and Jarnum, 1971; Marner *et al.*, 1975; Jensen *et al.*, 1976). Conversely, the concentrations of the serum albumin and pre-albumin fall with increasing severity of ulcerative colitis (Weeke and Jarnum, 1971; Marner *et al.*, 1975). This decrease appears to be mainly the result of faecal protein loss (Bendixen *et al.*, 1970); constant or decreased synthesis and more rapid catabolism may also be factors.

There is a need for objective measurements to assess the severity of attacks of ulcerative colitis and their response to treatment. We have measured the serum concentrations of five acute-phase reactants, as well as albumin, pre-albumin, and immunoglobulin at intervals during such attacks to identify changes from normal which might give a useful guide to clinical management.

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Method

SELECTION OF PATIENTS

Thirty-nine consecutive admissions of 36 patients with attacks of ulcerative colitis to St Mark's Hospital, The London Hospital, and Enfield District Hospital, Chace Wing, were studied. Two patients with additional illnesses which could affect some serum protein levels were excluded from analysis. The diagnosis of ulcerative colitis was established in each case by the history, sigmoidoscopic and rectal biopsy findings, a barium enema examination, and the exclusion of infection except in patients undergoing emergency surgical treatment when the diagnosis was confirmed by examination of the resected specimen.

CLINICAL PROCEDURES

A detailed record of each patient's condition was kept daily on a specially designed form. The results of the haematological investigations and routine laboratory determinations of the serum albumin and total serum protein concentrations were known to the clinicians looking after the patients but the other measurements described in this paper were unknown to them during the patient's admission. Decisions about the failure of medical treatment and the need for urgent surgical treatment were made on the basis of symptoms, signs, the standard haematological and

biochemical findings, and the results of abdominal radiographs. The outcome of the attack was classified as a response to medical treatment, or failure to respond with need for urgent surgical treatment.

LABORATORY TECHNIQUES

Venous blood was taken shortly after admission and at least twice weekly thereafter. The total serum protein and albumin concentrations were measured in the routine clinical laboratory using the biuret method (Kachmar, 1970) and bromcresol green technique (Northam and Widdowson, 1967, adapted for use on a Vickers M300 autoanalyser) respectively. A sample was stored at −20°C until the following measurements were made. The serum albumin, IgG, IgM, and IgA were determined by immunoprecipitation using a Technicon autoanalyser. The α_1-antichymotrypsin was measured using the rocket immunoelectrophoresis technique (Laurell, 1966). The seromucoid was separated from the serum by the method of Mandel (Mandel *et al.*, 1955) and estimated using the Lowry technique (Lowry *et al.*, 1951). The α_1-acid glycoprotein, α_1-antitrypsin, C-reactive protein, and pre-albumin concentrations were measured by radial immunodiffusion (Mancini *et al.*, 1965). The haemoglobin concentration and erythrocyte sedimentation rate were also measured.

METHODS OF ANALYSIS

Not all the proteins studied were normally distributed and the analysis has been carried out using two statistical techniques. Differences in individual protein concentrations between the two clinical outcome groups have been analysed using the Wilcoxon rank sum test. In the second technique all the protein concentrations have been combined in several discriminant analyses to identify combinations of proteins which give useful separation of the outcome groups and allow prediction of the clinical result.

Results

CLINICAL OUTCOME

Clinical details of the eight patients requiring surgical treatment are listed in the Table. The remaining 28 patients (31 admissions) responded to medical treatment. There was no directly related death but one patient aged 86 years died of acute myocardial ischaemia after remission of the colitis had been obtained with oral prednisolone.

CORRELATION OF PROTEIN CONCENTRATIONS ON ADMISSION WITH CLINICAL OUTCOME

The α_1-acid glycoprotein, albumin (determined immunochemically), and total protein concentrations on admission differed significantly between the two outcome groups (Fig. 1). There is much overlap between the groups, although an α_1-acid glycoprotein concentration of more than 2·5 g/l, an albumin concentration of less than 20 g/l, and a total protein concentration of less than 60 g/l were each associated with an operative rate of more than 60%. In addition, four of five patients with an albumin concentration (measured by the bromcresol green method) of less than 30 g/l required surgical treatment. There was no significant correlation between the seromucoid and immunoglobulin concentrations or the ESR and the outcome of the attack. The haemoglobin concentration was lower ($P < 0·05$) among the patients who required surgical treatment.

A discriminant analysis was performed to identify that combination of two protein concentrations

Table *Details of patients treated surgically*

Patient	*Sex*	*Age (yr)*	*Total length of history*	*Extent of disease*	*Urgency of operation*	*Days in hospital before operation*	*Reason for operation*
GL	F	57	5 yr	Total	Urgent	26	Only moderate improvement on ACTH 80 u, i.m. daily and deterioration on reducing dose
JD	F	71	5 yr 6 m	Total	Urgent	17	Failure to respond to intravenous hydrocortisone 400 mg daily
KF	F	61	8 yr	Total	Emergency	0	Toxic dilatation and perforation of colon
VJ	M	27	9 yr	Total	Urgent	8	Deterioration in spite of treatment with ACTH 80 u, i.m. daily
PM	M	48	5 yr 2 m	Mid-transverse colon	Semi-urgent	28	Failure to respond and finally deterioration on oral prednisolone 40 mg daily
HM	M	71	15 yr	Left-sided	Urgent	13	Marked colonic dilatation and imminent perforation
MN	F	63	2 m	Left-sided	Urgent	4	Toxic dilatation and perforation of colon
PR	M	37	3 yr 6 m	Total	Semi-urgent	15	Long-standing failure to respond to treatment with oral prednisolone

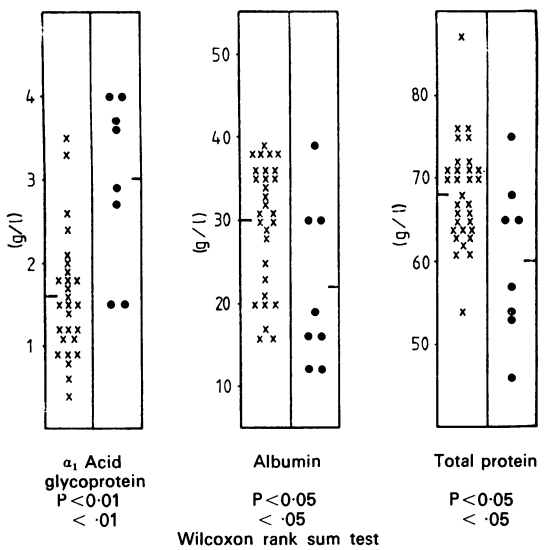

Fig. 1 *Comparison of initial protein concentrations in the two outcome groups.* × *Response to medical treatment.* ● *Surgical treatment required.*

which gave the best separation of the two outcome groups. The best combination was the albumin (determined immunochemically) and IgG, but the results given by the individual protein concentrations were more easily interpreted and gave almost as good separation.

RESPONSE TO TREATMENT

Changes in the concentrations of the C-reactive protein and pre-albumin showed a remarkable correlation with the clinical response to treatment (Fig. 2). The change in C-reactive protein concentration differed markedly between those patients who responded to medical treatment and those who did not ($P < 0.05$, Wilcoxon rank sum test). The majority of those patients responding to medical treatment showed a fall in C-reactive protein concentration from high to near normal levels. The rate of fall corresponded to the rate of clinical improvement and, in most cases, the concentration declined most markedly within the first four days. Two of the three patients with significantly raised levels at the time of discharge required further hospital admission and surgical treatment for active colitis. In patients requiring surgical treatment the C-reactive protein concentrations rose or remained raised during medical treatment.

The pre-albumin concentration (Fig. 2) tended to rise in those patients who responded to medical treatment and usually remained low or fell in those patients who required surgical treatment. The difference in response was significant ($P < 0.01$, Wilcoxon rank sum test), but these changes tended to occur within the range of normality.

IgG levels (Fig. 2) tended to be lower in the surgically treated group but there was no consistent change in concentration in either group during medical treatment.

Two examples of C-reactive protein and pre-albumin measurements during medical treatment of attacks of ulcerative colitis are shown in Fig. 3. In the first case the fall in C-reactive protein and rise of pre-albumin were associated with a rapid clinical response to treatment with intravenous prednisolone. In the second case the falling pre-albumin and maintained high level of C-reactive protein were associated with worsening of the patient's condition and surgical treatment was required.

The results of the highest and lowest serum protein concentrations observed during each patient's admission were analysed and correlated with the outcome. Response to medical treatment always occurred if the C-reactive protein concentration did not exceed 2·5 mg/dl or the total serum protein concentration remained above 60 g/l throughout the admission.

Discriminant analyses were performed on the protein results at increasing time intervals after admission. When all proteins were included separation of up to 85% of patients in the two outcome groups was possible but such an analysis is impractical for routine use. There was no combination of two or three protein measurements which gave consistently good separation of the two groups, though albumin (determined immunochemically) was always selected in these combinations.

Discussion

The C-reactive protein concentration starts to rise eight hours after surgical trauma and begins to decline on the third day when the concentrations of other acute phase reactants are still maximal (Aronsen *et al.*, 1972). As normal C-reactive protein concentrations are very low (<0·7 mg/dl) the synthesis of small quantities of this protein may give rise to a several-fold increase in the serum concentration. Pre-albumin also occurs in low concentration (10-35 mg/dl) in normal serum and has a short half-life of 1·9 days (Oppenheimer *et al.*, 1965) compared with 15 days for albumin (Beeken *et al.*, 1962). Changes in the concentrations of these proteins may therefore rapidly reflect the clinical response of patients with attacks of ulcerative colitis to medical treatment.

The correlations between the clinical response to treatment and the changes of C-reactive protein and pre-albumin concentrations were considerably better

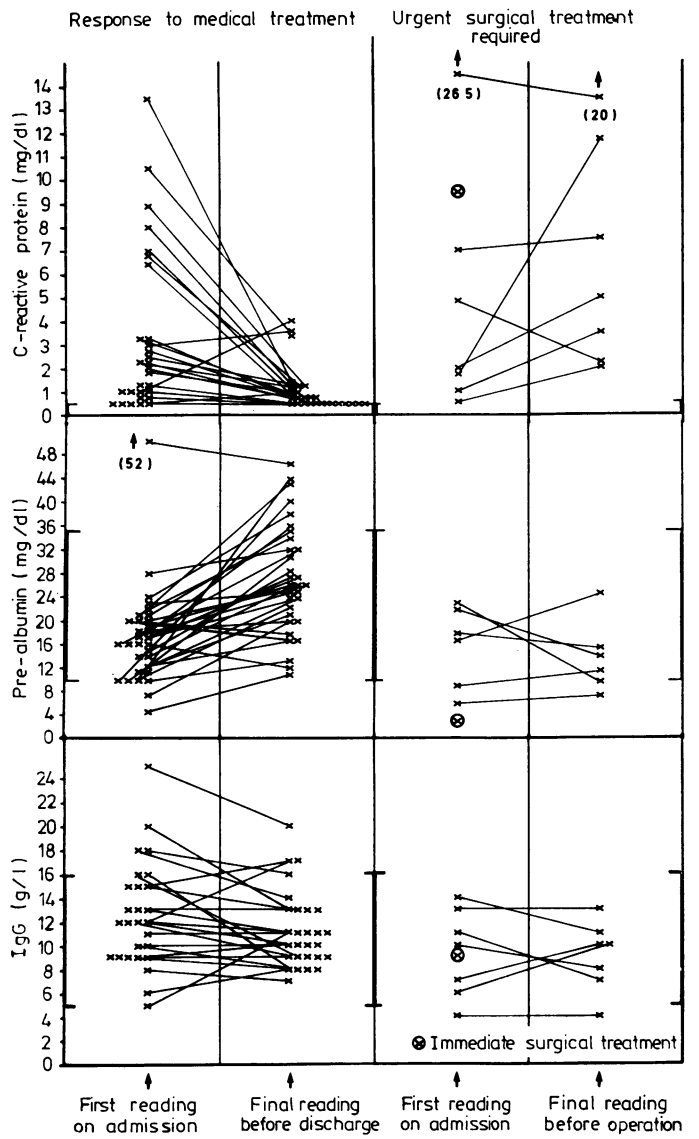

Fig. 2 *Changes in protein concentrations in the two outcome groups during medical treatment. (The normal ranges of the protein concentrations are indicated by the thickened bars on each scale.)*

than that shown by any other clinical or metabolic parameter. The sustained fall of a previously raised C-reactive protein concentration to near normal levels always coincided with a favourable response to medical treatment and no patient in whom the concentration remained below 2·5 mg/100 ml required surgical treatment. A rising or persistently raised C-reactive protein level was associated with urgent surgical treatment in six of eight cases.

Changes in the C-reactive protein and pre-albumin concentrations do not appear to precede the clinical response to treatment but more frequent measurements may detect earlier changes. The serial measurements of C-reactive protein reflect the response to treatment rather better than those of the pre-albumin. The fact that C-reactive protein falls rapidly and pre-albumin rises during successful medical treatment and that these changes occur for different reasons means that the information gained from both readings is complementary. Where immunochemical facilities for the measurement of proteins are not available the C-reactive protein may be determined semi-quantitatively[1] and inspection of a stained cellulose acetate electrophoretic strip will usually

[1]Wellcotest, obtainable from Wellcome Reagents Ltd.

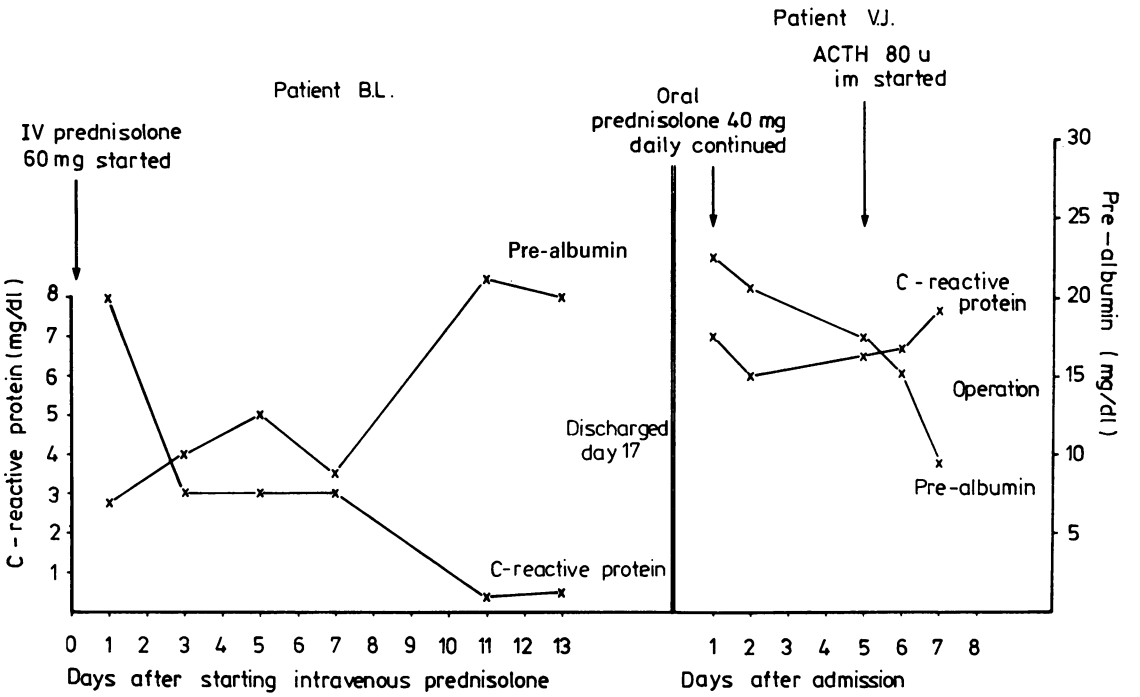

Fig. 3 *Serial measurements of C-reactive protein and pre-albumin in two patients during medical treatment of acute ulcerative colitis.*

indicate whether the pre-albumin concentration is within normal limits or not (Harris and Kohn, 1974).

The tendency to a lower IgG concentration among those patients treated surgically agrees with the reduced gamma globulin fraction (which consists mainly of IgG) found in surgically treated patients (de Dombal, 1968). However, changes in the concentration during medical treatment which correlated with the outcome were not seen in this study.

Single measurements of serum albumin, total protein, and α_1-acid glycoprotein, none of which change so rapidly as C-reactive protein and pre-albumin, give some guide to the duration, severity, and therefore the outcome of an acute attack of colitis. Measurement of the serum albumin by an immunochemical method gave more prognostic information than measurement by the bromcresol green method because the latter may over-estimate low serum albumin concentrations (McPherson and Everard, 1972; Beng *et al.*, 1974) and it thus gave higher readings among the patients who failed to respond to medical treatment. Immunochemical facilities for the measurement of C-reactive protein, pre-albumin, albumin, and α_1-acid glycoprotein are therefore helpful in the management of acute attacks of colitis.

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