

IL-6, CRP, and Albumin Levels in Sera of Iraqi Patients With Inflammatory Bowel Diseases

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Abstract:

Background:

Inflammatory bowel diseases (IBD) are heterogeneous group of disease, which include ulcerative colitis (UC) and Crohn's disease (CD). The pathogenesis of the disease is related to infections, genetic and immunological factors.

Objectives: This case control study aims to investigate the serum levels of IL-6, CRP, and albumin in untreated IBD and to study correlations among these parameters.

Patients and Methods : The study covered 70 subjects. It comprised a total of 40 patients with IBD (24 with UC and 16 with CD), compared with 30 healthy individuals with no history of IBD. Serum IL-6 analysis was performed by ELISA and serum CRP was estimated by latex agglutination test whereas serum albumin was measured by Bromocresol green method.

Results: The present results revealed that there are no significant differences between UC and CD patients for each serum IL-6, CRP, and albumin, whereas the levels of IL-6 and CRP were significantly higher in patients as compared to healthy control $P < 0.001$. Furthermore serum albumin level was significantly reduced in Patients with IBD when compared to control group $P < 0.001$. Interestingly the present study showed linear correlation between IL-6 and CRP while inverse correlation was found between IL-6 and albumin.

Conclusions: These finding suggest that IL-6 play an important role in pathogenesis of IBD, and the disturbance in serum IL-6, CRP, and albumin in IBD patients as compared to control subjects confirm the disturbance of immune response in IBD patients.

Keywords: IBD , Cytokines, CRP, albumin.

Introduction:

Inflammatory bowel diseases are defined as heterogeneous group of diseases that have a common final manifestation, which is the mucosal inflammation. The pathogenesis of these diseases is related to several genetic, environmental and immunological factors (1). IBD include UC and CD, they differ sufficiently to be clearly distinguishable, however they have common features. Both have features of inflammation of the bowel, lack of a proven causal agent, patterns of familial occurrences and both create systemic manifestations (2).

Immunological abnormality has been demonstrated in IBD patients and imbalance between pro-inflammatory cytokines and anti-inflammatory cytokines play an important role in the initiation and regulation of the immune response (3). IL-6 is produced by a variety of cells, such as monocytes, macrophages, T lymphocytes, B lymphocytes, fibroblasts, and endothelial cells. It may affect the proliferation of epithelial cells and act as an autocrine growth factor for enterocytes. It has been shown to stimulate T cell and B cell activation and proliferation, and to increase immunoglobulin synthesis from epithelium of human intestine (4 & 5). It is interesting to know that IL-6 has been proposed as a marker of inflammation in IBD and its concentration was elevated in serum, peripheral blood mononuclear cells (PBMC), mucosa biopsy and lamina propria mononuclear cells (LPMC) in IBD patients (6 & 7). Some papers also show that IL-6 is increased in the systemic circulation in Crohn's disease and is not elevated in UC patients (8).

C-reactive protein (CRP) is synthesized in hepatocytes, and an increase in serum value of

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this acute phase protein indicates the existence of acute inflammation. The synthesis of CRP is regulated by IL-1, IL-6, TNF- α , which are pro-inflammatory cytokines (9). Hermanowicz and associates noted that serum elevation of CRP is found in UC and the activity of disease correlated with CRP level (10). CRP levels proved to be a useful objective indicator of disease activity and response to therapy and reflected changes in disease activity before the erythrocyte sedimentation rate (11).

Subjects and Methods:

Subjects:

Patients group:

The current study comprised of patients with IBD (21 females and 19 males; mean age 38 years, ranged between 18-65).

The patients group was classified in to two groups:-

1-Ulcerative colitis group included 24 patients (60%).

2-Crohn's disease group included 16 patients (40%).

The clinical examination and diagnosis were performed by physician specialized in Gastroenterology and Hepatology at Gastroenterology and Hepatology Teaching hospital.

Healthy control group:

This group included 30 subjects who had no history or clinical evidence of IBD or any chronic disease.

Methods:

Estimation of serum IL-6:

IL-6 was determined in serum using commercially available ELISA and performed as recommended in leaflet with kit. (Immunotech, Beckman Coulter, France).

Estimation of serum CRP:

Serum CRP levels were estimated to all study groups according to the procedure provided with kits by Spinreact, S.A.

Principle:-

The latex reagent is a suspension of polystyrene latex particles of uniform size coated with IgG fraction of an anti-human CRP specific serum. Latex particles allow visual observation of the Ag-Ab reaction. The reaction takes place when the CRP present in the serum, reacts with IgG coated latex particles, resulting in the

formation of a web between them. A clear agglutination become evident when the serum contains approximately more than 6 mg/l of CRP.

Estimation of serum albumin:

Serum albumin levels were estimated in all study groups according to the procedure provided with kits by Randox Company.

Principle:-

Measurement of serum albumin is based on its quantitative binding to the indicator 3,3',5,5'-tetrabromo-gesol salphan-thalein (Bromocresol green Bcg). The albumin- Bcg complex absorbs light at 600-650 nm.

Statistical analysis:

It was assessed using P(Kruskall-Wallis-test) and P(Mann-Whitney- test), as well as P(ANOVA) test and P(Bonferroni-test).

Results:

Serum IL-6, CRP, and albumin levels were estimated in 40 patients with IBD, (24 UC & 16 CD) compared with 30 healthy control group, age and sex matched.

Table-1 demonstrate the age and gender of the patients with IBD, which showed slight female predominance.

The present study revealed a significant elevation in serum levels of IL-6 among UC and CD patients (10.6 pg/ml & 7.9 pg/ml) respectively in comparison to that of healthy control (3.3 pg/ml) $P < 0.001$, Table-2. An anticipated, this study showed significant differences in the median levels of CRP between patients and control as clearly shown in Table-3. Regarding the concentration of serum albumin Table-4 pointed out a significant reduction in the mean of serum albumin in both groups of patients (UC=2.9 \pm 0.8g/L and CD=3 \pm 0.7g/L) when compared to healthy control group (4.1 \pm 0.5g/L), $P < 0.001$.

It is worthy to mention that there was positive linear correlation between serum IL-6 level and serum CRP level, whereas inverse correlation was found between serum albumin concentration and each of IL-6 and CRP $P < 0.001$ as shown in Table-5. Figure-1 demonstrate inverse correlation between IL-6 and albumin.

Table-1: Ages and gender distribution of the studied groups

		Ulcerative colitis		Crohn's disease		Healthy control	
Age in year	Minimum	18		21		20	
	Maximum	65		50		63	
	Mean	40		37		38	
	SD	13		12.7		15.7	
	Total	24		16		30	
Gender							
	Female	42	42%	16	45.7%	22	44%
	Male	58	58%	19	54.3%	28	56%
	Total	100	100	35	100	50	100

Table- 2: The difference in median levels of IL-6 (pg/ml) among the three studied groups

	Ulcerative colitis	Crohn's disease	Healthy control	P (Kruskall-Wallis)
Serum IL-6				
Minimum	2.1	1.9	1.8	
Maximum	40.5	23.4	5.6	
Median	10.6	7.9	3.3	<0.001
NO.	24	16	30	
P (Mann-Whitney)				
UC X HC <0.001				
CD X HC <0.001				
UC X CD= NS				

Table-3: The difference in median levels of CRP (mg/dl) among the three studied groups

	Ulcerative colitis	Crohn's Disease	Healthy control	P (Kruskall-Wallis)
Serum CRP				
Minimum	<6	<6	<6	
Maximum	48	48	<6	
Median	6	6	<6	<0.001
No.	24	16	30	
P (Mann-Whitney)				
UC X HC < 0.001				
CD X HC < 0.001				
UC X CD = NS				

Table-4: The difference in mean levels of albumin (g/l) among the three studied groups

	Ulcerative colitis	Crohn's disease	Healthy controls	P (ANOVA)
Serum albumin				
Minimum	1.5	3.2	1.8	
Maximum	5	5.2	4.6	
Mean	2.9	3	4.1	<0.001
SD	0.82	0.70	0.5	
No.	40	16	30	
P (Bonferroni t-test)				
UC X HC < 0.001				
CD X HC < 0.001				
UC X CD = NS				

Table-5: The correlation of serum IL-6 level with other variables (CRP and albumin)

Variables	IL-6	P (T-test)
CRP	0.684	<0.01
Albumin	-0.525	<0.01

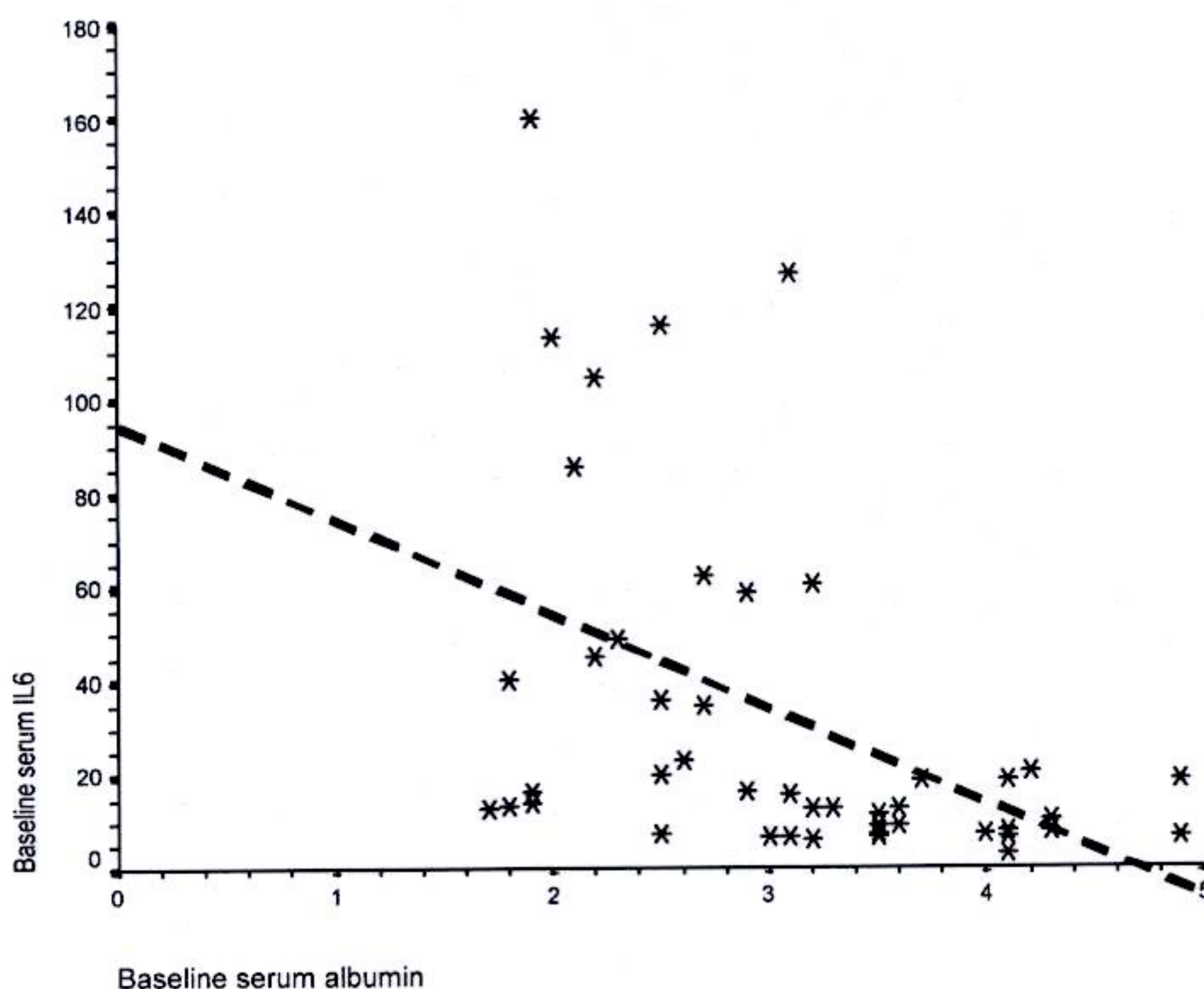


Fig. 1 Inverse correlation between serum levels of IL-6 and albumin.

Discussion:

The high level of IL-6 in IBD patients observed in this study was comparable with other studies (6 & 12) who reported similar increase of this cytokine. Bing-Xia and associates (12) interestingly pointed out that spontaneous production of IL-6 from peripheral blood mononuclear cells (PBMC) or production of IL-6 by stimulation of PBMC with lipopolysaccharid in patients was higher than

that in healthy controls. The serum elevation may quite well be due to the production of this cytokine by the infiltrating immune cells.

The result of CRP was also comparable to other studies (10 & 11) which collectively showed that a high level of serum CRP in active IBD patients in comparison to patients with inactive disease and healthy control group. It is generally accepted that IL-6 play a central role in the stimulation of CRP production, so the

increase of this protein level may be secondary to the IL-6 release which is brought about by the inflammatory cells (9). This may be one explanation for the positive linear correlation between serum levels of CRP and IL-6 in patients.

The mean serum albumin concentration was found to be less than that of the control group in both groups of patients in this study in agreement with others studies (13&14). The lower serum albumin concentrations may be due to the production of cytokines such as IL-6, which modulate the production of albumin by the hepatocytes (15).

In conclusion these findings suggest that IL-6 plays an important role in the pathogenesis of IBD, and the disturbance in serum IL-6, CRP, and albumin levels in patients compared to healthy subjects confirm the disturbance of immune response in those patients.

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