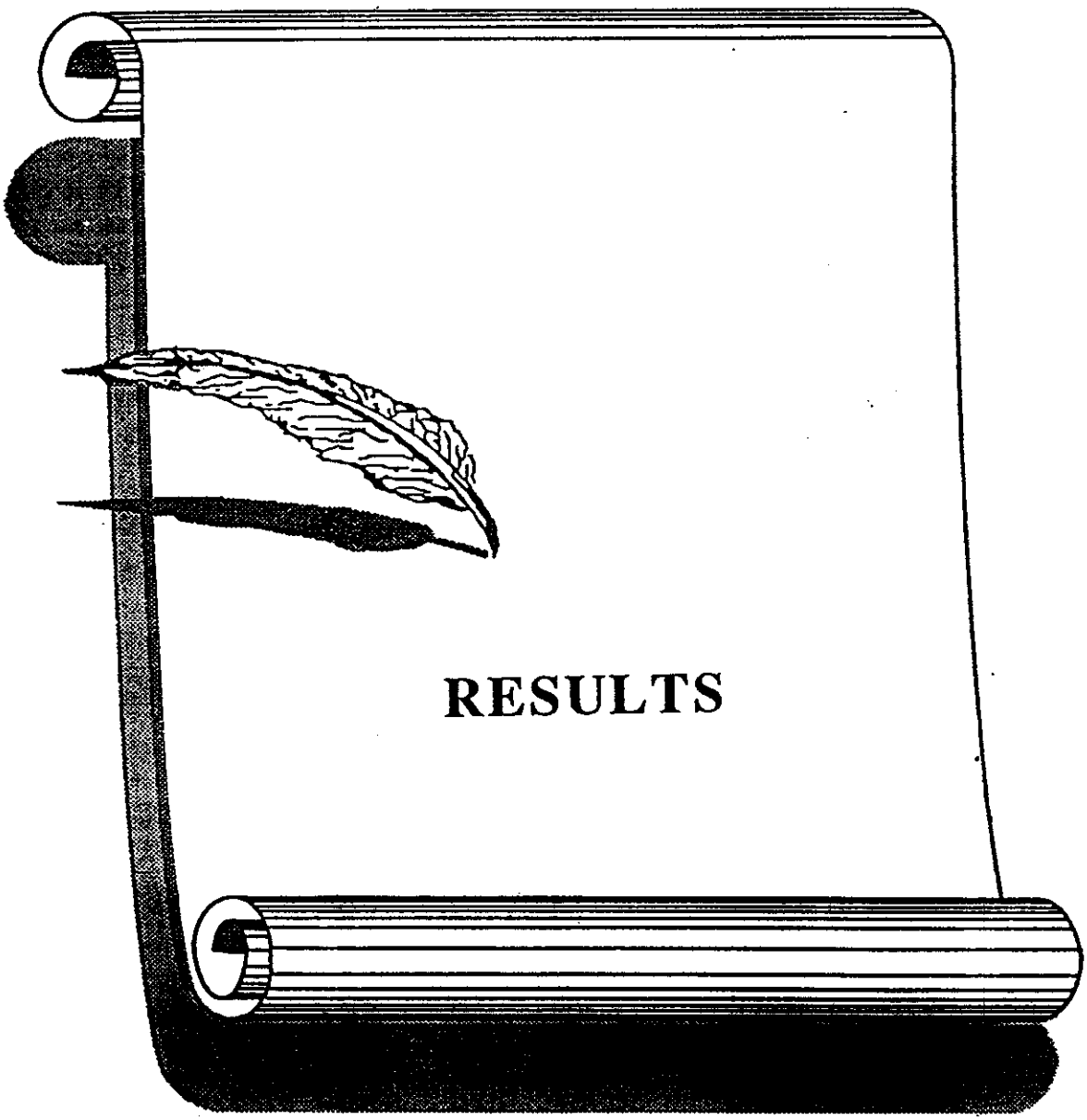

RESULTS

RESULTS

The results of all studied groups are illustrated in successive tables (1-15) in appendix.

The clinical and general characteristics for all studied groups are summarized in table (1). Liver function tests are illustrated in table (2) and presented as mean value ± SD for each studied group. The mean value of total leukocytic and monocytic counts for all studied groups are shown in table (4).

The data concerned with viral markers and autoantibodies for all patient groups are summarized in table (3). The data are shown as a percentage of patients with sero positive for viral markers and autoantibodies. The data concerned with viral markers are illustrated in figures (11-14). The control group of patients are not included in this percentage as they were selected from subjects with already sero negative for viral markers and auto antibodies.

Serum levels of TNF-α, IL-1β, IL-6 and CRP for all studied groups are illustrated in table (5). Cytokines and CRP serum levels are presented as mean value ± SD and illustrated in figures (15-18). The proinflammatory cytokines and CRP serum levels of cirrhotic group were elevated compared to other groups.

Our nermol ranges were as follows serum TNF-α (5.91-18.59 pg/ml) with mean value (11.54±2,7), serum IL-1B (0.0-11.0 pg/ml) with mean value (5.9±2.9), serum IL-6 (30-80

pg/ml) with mena vlaue (59.33±13.8) and Serum (CRP 0.0-1.0 mg/dl) with mean value 0.51 ± 0.29.

The comparative study and statistical variations between all studied groups are illurtrated in tables (6-9). Serum levels of TNF-α, IL-1β, IL-6, and CRP were significantly elevated in all patient groups compared to control. CRP showed no statistical vartions in group III compared to control ($P > 0.05$).

The intergroup study defined that cytokine and CRP serum levels were significantly increased with the severity of the disease which may be of prognostic value and explain the role of cytokines in chronic liver diseases. TNF-α, IL-1β, IL-6 and CRP serum levels of patients with sever chronic active hepatitis (group IIB) were significantly elevated compared to patients with mild degree (group IIA). Significant elevation of cytokines and CRP serum levels had been found in patients with decompensated cirrhosis (group IVB) compared to patients with compensated cirrhosis (group IVA). This data are illustrated in tables 10 and 11 respectively and presented as mean value ± SD, t value and its significance.

Analysis of relationships between individual cytokines and laboratory parameters was accomplished by grouping laboratory values into four categories including: (a) parameters of liver injury (AST, ALT and serum bilirubin concentrations), (b) hepatic synthetic function (prothrombin time and albumin serum concentrations), (c) total leukocytic and serum monocytic

counts as a source for these cytokines. The correlation coefficients between TNF-α, IL-1β, IL-6 or CRP and laboratory parameters are presented as r value and its significance in tables (12-15) and illustrated in figures (19-25). The group III showed no significant correlation (P> 0.05) which may be refered to the high critical value due to low number of patients included in this group.

Good positive correlation between cytokine/CRP and AST/ALT were found in group II, also a good negative correlations with prothrombin time and serum albumin were detected in group IV and group V. Serum total bilirubin showed well positive correlation in group IV. This data may reflect a good correlation with progress of the disease and its severity.

In group (II), a good positive correlation had been found between AST and TNF-α (r=0.33), IL-1β (r=0.37); IL-6 (r= 0.49) and CRP (r= 0.44). A significant positive correlation had been found also between ALT and TNF-α (r= 0.37), IL-1β (r=0.31) and IL-6 (r=0.31). No significnat correlation between CRP and ALT, but a significant negative correlation had been found between CRP and serum albumin (r = -0.33).

In group (IV);serum total bilirubin was positively correlated with TNF-α (r= 0.27), IL-1β (r = 0.32) ; IL-6 (r= 0.34) and CRP (r= 0.29). Serum albumin had a significant negative correlation with TNF-α. (r= -0.67), IL-1β (r= -0.65), IL-6 (r = -0.58) and CRP (r= -0.29). TNF-α, IL-1β , IL-6 and CRP were negatively

correlated with prothrombin time (r = -0.77, -0.66, -0.66, and 0.56 respectively).

In group (V), TNF-α had a well negative correlation with serum albumin (r = -0.36) and prthrombin time (r= -0.44). IL-1β was only negatively correlated with prthrombin time (r= 0.34). IL-6 had a good positive correlation with AST (r = 0.34), serum total bilirubin (r = 0.36) and a negative correlation with serum albumin (r=-0.38) and prothrombin time (r= -0.47). CRP was significantly correlated with prothrombin time (r= -0.37).

No significant correlation had been found between any of cytokines or CRP and total leukocytic or monocytic counts in all studied groups.

The correlation coefficients amonge cytokines and CRP were presented in tables (16-19) and illustrated in figures (26-34). The proinflammatory cytokines (TNF-α, IL-β and IL-6) and CRP were well correlated with each other in all studied group except in group III where elevated critical value and low number of patients included in this group result in insignificant correlation.

When data from all patient groups were analyzed using one-way ANOVA test, TNF-α serum level was highly significant in differentiation between different groups of patients with CLD (F ratio = 4.212) CRP and IL-6 represents the second and third after TNF-α to differentiate between studied groups. (F ratio = 3.3 for CRP, 3.1 for IL-6). Il-1β was not significnat (F ratio = 2.23), the details of ANOVA test are illustrated in table (20).

Table 1: Illustratrates clinical and general characteristics for all studied grups.

Group	Diagnosis	NO	Age(yr)	Sex M/F	Liver Biopsy
I	Control	30	32.0 ± 5.7	23/7	0/30
II	C.A.H	33	41.8 ± 8.4	28/5	30/30
III	C.P.H	8	35.3 ± 8.1	7/1	8/8
IV	Cirrhosis	50	47.6 ± 10.3	42/8	28/50
V	P.P.F	25	38.4 ± 7.2	19/6	5/25
	all subjects	146	39.0 ± 7.9	120/26	71/146

C.A.H: Chronic active hepatitis

C.P.H: Chronic persistant hepatitis

P.P.F : Periportal fibrosis.

CLD : Chronic liver disease.

Table 2: Shows liver function tests for all studied groups presented as mean value ± SD.

Group	AST 10-42 U/L	ALT 10-60 U/L	ALP 26-88 U/L	TB 0.2-1.0 mg/dl	DB 0-0.2 mg/dl	Alb 3.5-50 gm/dl	TP 6-8 gm/dl	γGT 7.64 U/L	PT 70-100%
I	26±6.9	21.8±7.5	57±20	0.7±0.19	0.02±0.05	4.1±0.35	7±0.5	22±11	98±2.9
II	124.5±75.9	136±50.8	108±54	1.9±1.9	0.4±0.9	3.4±0.6	6.9±0.6	132.6±176	77±15.7
III	55.7±16.8	59.6±25.5	56.7±16.4	1.4±0.5	0.3±0.4	3.7±0.5	7.0±0.5	36.0±21.8	78±8.3
IV	96.4±57.2	62.4±33.4	115.5±74.6	2.7±2.8	0.8±1.3	2.7±0.9	6.8±0.97	61.8±49	63.4±15.4
V	67.1±30.9	52.4±25.3	80.8±24.4	1.6±0.8	0.24±0.5	3.3±0.7	6.9±0.6	60.0±54.7	77.3±15.9

Table 3: Illustrates percentage of patients with seropositive for viral markers and autoantibodies in all patient groups

Group	VIRAL MARKERS			AUTOANTIBODIES		
	HBs Ag	HBc Ab	Anti-HCV	ANA	ASMA	AMA
II	18%	87%	75%	30%	54	0
III	75%	100%	37%	0%	12.5%	0%
IV	6%	74%	76%	4%	34%	2%
V	8%	52%	64%	0%	36%	0%
Total CLD	14.6%	75%	70.6%	10.3%	38%	0.8%

Table 4: Shows the mean value ±SD of total leukocytic and monocytic counts in all studied groups.

Item	I	II	III	IV	V
W.B.Cs	6232	6271	5925	5813	6144
	±	±	±	±	±
Cell/ul	1394	1553	1325	1424	1639
Monocytes	209	305	199	212	268
	±	±	±	±	±
cell/ul	81	177	67	100	172

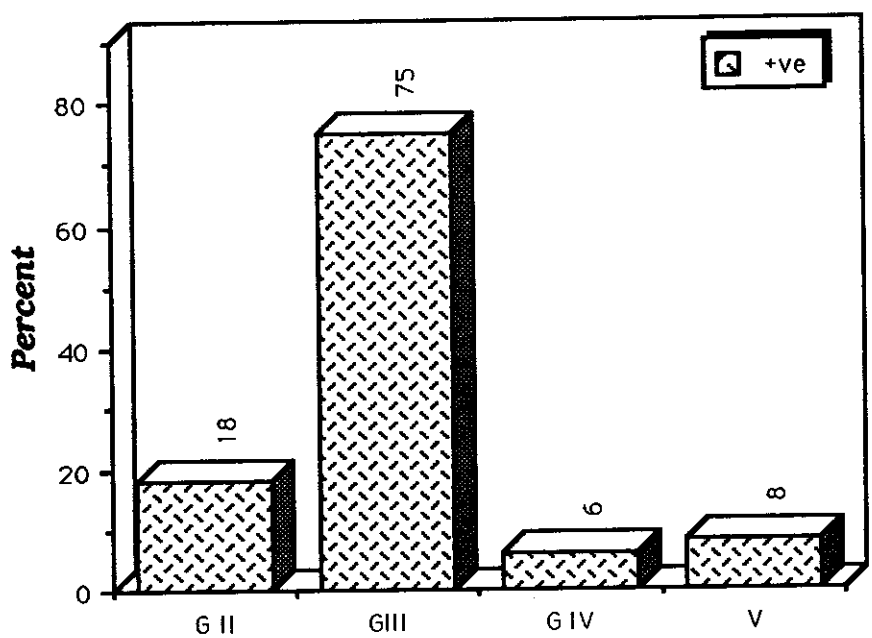

Fig. (11): Illustrates the percent of patients with seropositive for HBs Ag in all patient groups.

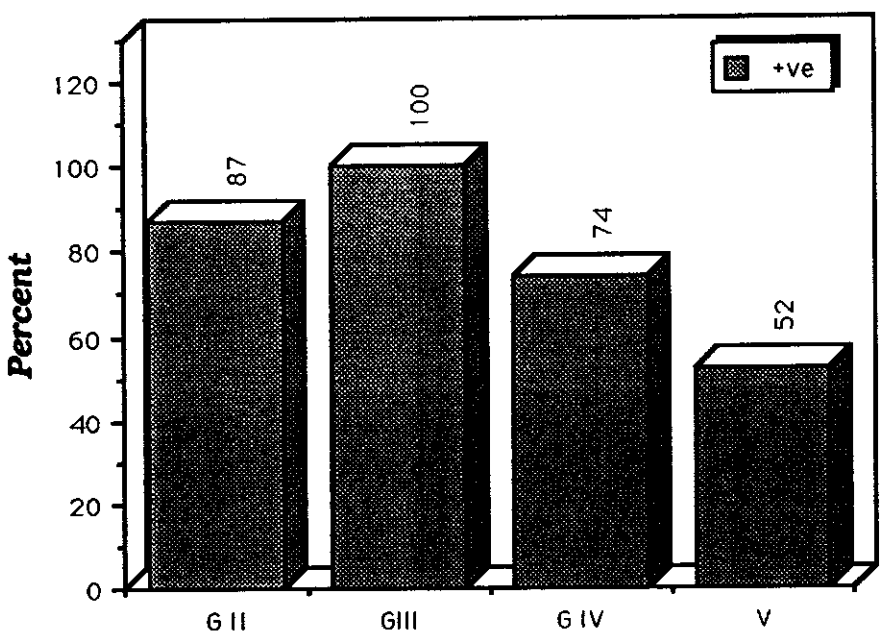

Fig. (12): Illustrates the percent of patients with seropositive for HBc Ab in all patient groups.

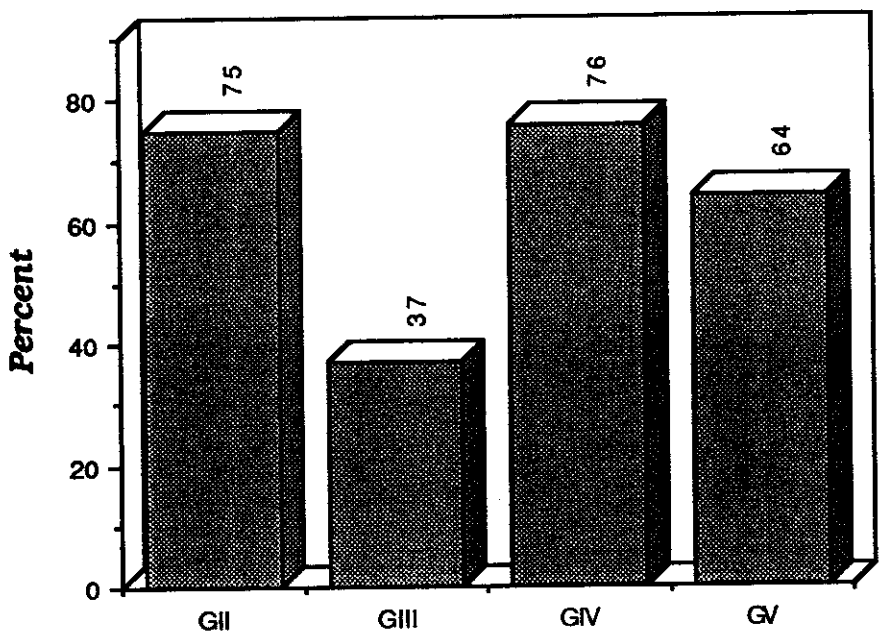

Fig. (13): Illustrates the percent of patients with seropositive for Anti-HCV in all patient groups.

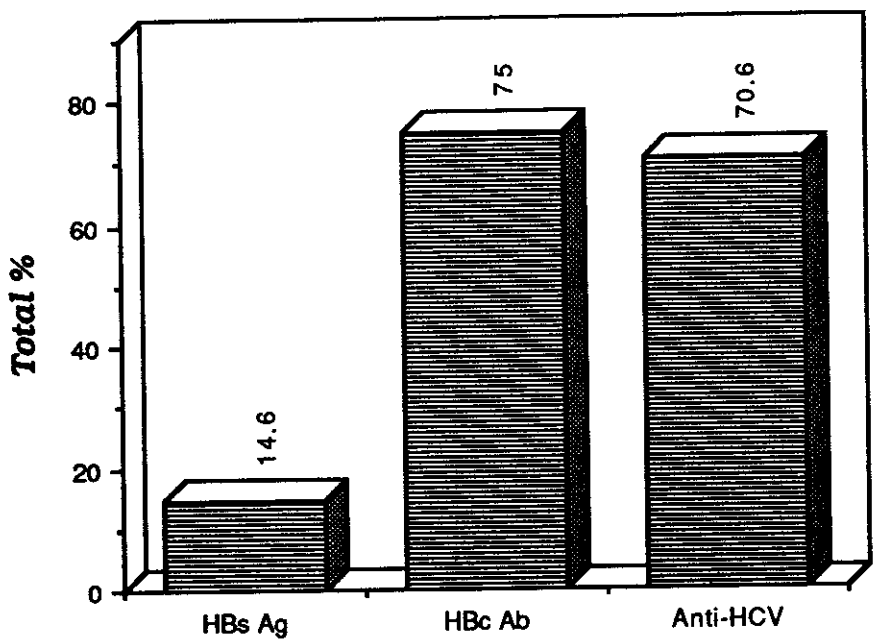

Fig. (14): Illustrates the percent of patients with seropositive for viral markers in all patient groups.

Table 5: Illustrates cytokines and CRP serum levels in all studied groups presented as mean value ± SD.

Group	TNF-α pg/ml	IL-Iβ pg/ml	IL-6 pg/ml	CRP mg/dl
GI	11.54 ± 2.7	5.96 ± 2.9	59.33 ± 13.8	0.51 ± 0.29
GII	40.64 ± 18	9.54 ± 2.8	187.58 ± 60.9	1.25 ± 0.59
GIII	27.13 ± 6	8.12 ± 1.6	143.75 ± 36.2	0.75 ± 0.23
GIV	44.19 ± 21.4	10.35 ± 3.4	192.85 ± 80.8	1.35 ± 0.9
GV	30.35 ± 15.43	8.78 ± 2.6	148.2 ± 59.5	0.88 ± 0.54

Table 6: Comparative study and statistical variations of TNF-α serum levels between all studied groups.

Group		GI	GII	GIII	GIV	GV
GI	t p	====== ======	8.7 <0.001	10.7 <0.001	8.3 <0.001	6.6 <0.001
GII	t p	8.7 <0.001	====== ======	2.1 <0.05	0.75 >0.05	2.4 <0.05
GIII	t p	10.7 <0.001	2.1 <0.05	===== =====	2.2 <0.5	0.57 >0.05
GIV	t p	8.3 <0.001	0.75 >0.05	2.2 <0.05	===== =====	2.9 <0.01
GV	t P	6.6 <0.001	2.4 <0.05	0.57 >0.05	2.9 <0.01	===== =====

P < 0.05 : Significant
P < 0.01 : Highly significant
P > 0.05 : Not significnt

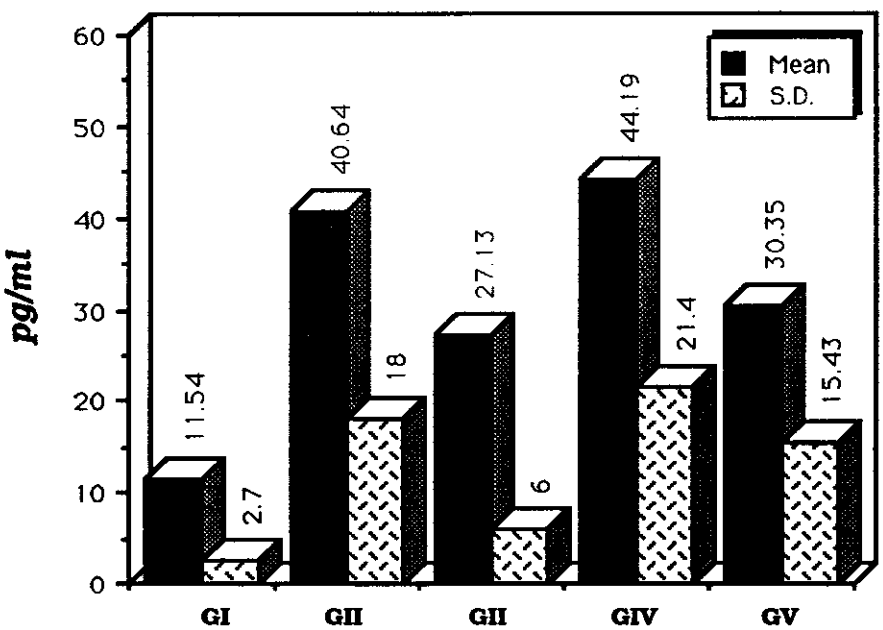

Fig. (15): Comparative study for TNF-α among all studied groups.

Table 7: Comparative study and statistical variations of IL-1β serum levels between all studied groups.

Group		GI	GII	GIII	GIV	GV
GI	t p	====== ======	4.9 <0.001	2.1 <0.05	5.8 <0.001	3.7 <0.001
GII	t p	4.9 <0.001	====== ======	1.3 >0.05	1.08 >0.05	1.0 >0.05
GIII	t p	2.1 <0.5	1.3 >0.05	===== =====	1.8 <0.05	0.65 >0.05
GIV	t p	5.8 <0.001	1.08 >0.05	1.8 <0.05	===== =====	1.9 <0.05
GV	t P	3.7 <0.001	1.0 >0.05	0.65 >0.05	1.9 <0.05	===== =====

P < 0.05 : Significant
P < 0.01 : Highly significant
P > 0.05 : Not significnt

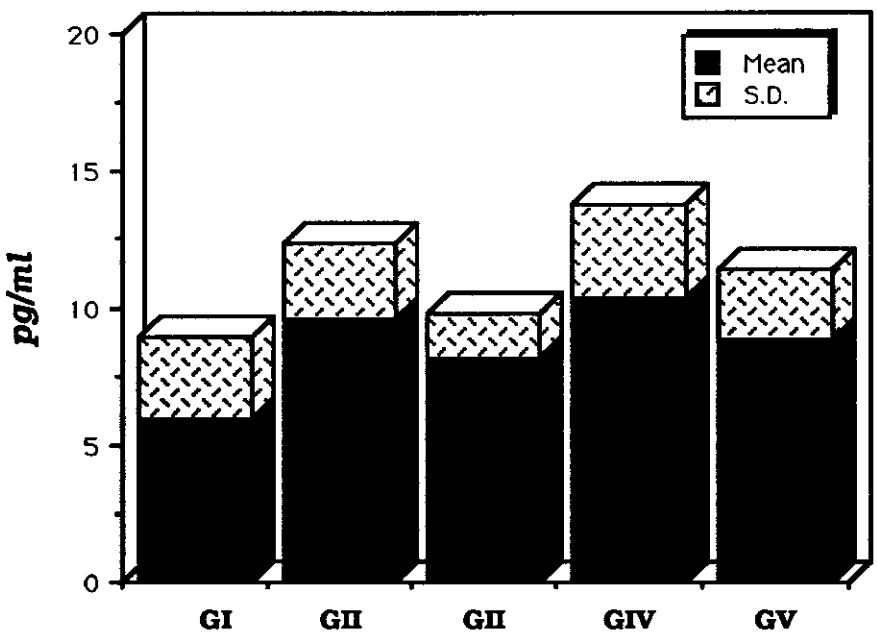

Fig. (16): Comparative study for IL-1β among all studied groups.

Table 8: Comparative study and statistical variations of IL-6 serum levels between all studied groups.

Group		GI	GII	GIII	GIV	GV
GI	t p	====== ======	14.5 <0.001	8.7 <0.001	12.1 <0.001	9.8 <0.001
GII	t p	14.5 <0.001	====== ======	1.9 <0.05	0.15 >0.05	2.6 <0.1
GIII	t p	8.7 <0.001	1.9 <0.05	===== =====	1.8 >0.05	0.1 >0.05
GIV	t p	12.1 <0.001	0.15 >0.05	1.8 <0.05	===== =====	2.2 <0.05
GV	t P	9.8 <0.001	2.6 <0.01	0.1 >0.05	2.2 <0.05	===== =====

P < 0.05 : Significant
P < 0.01 : Highly significant
P > 0.05 : Not significnt

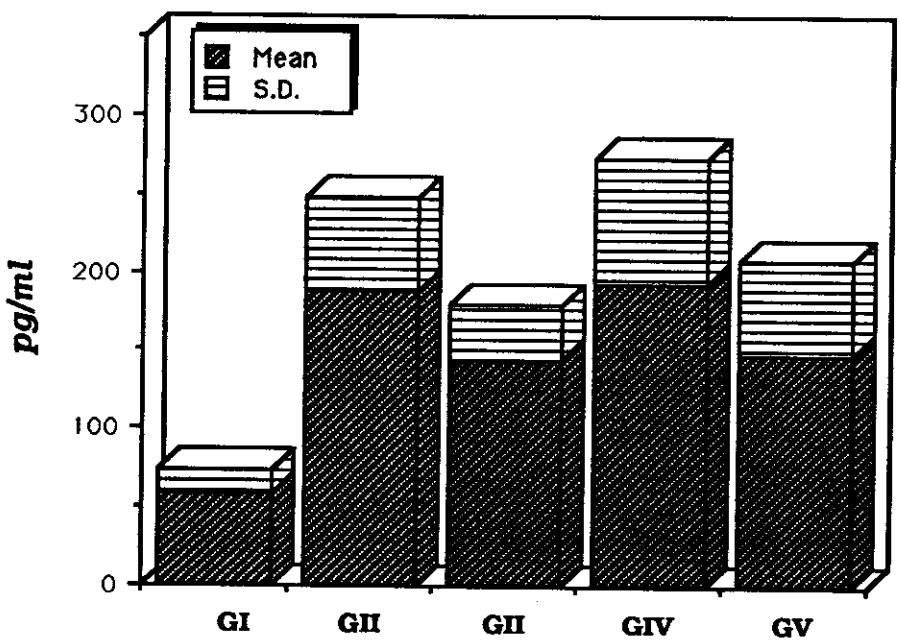

Fig. (17): Comparative study for IL-6 among all studied groups.

Table 9: Comparative study and statistical variations of CRP serum levels between all studied groups.

Group		GI	GII	GIII	GIV	GV
GI	t p	====== ======	5.7 <0.001	1.6 >0.05	5.4 <0.001	2.34 <0.05
GII	t p	5.7 <0.001	====== ======	2.3 <0.05	0.19 >0.05	2.4 <0.05
GIII	t p	1.6 >0.05	2.3 <0.05	===== =====	2.0 <0.05	0.32 >0.05
GIV	t p	5.4 <0.001	0.19 >0.05	2.0 <0.05	===== =====	2.5 <0.01
GV	t P	2.34 <0.05	2.4 <0.05	0.32 >0.05	2.5 <0.01	===== =====

P < 0.05 : Significant
P < 0.01 : Highly significant
P > 0.05 : Not significnt

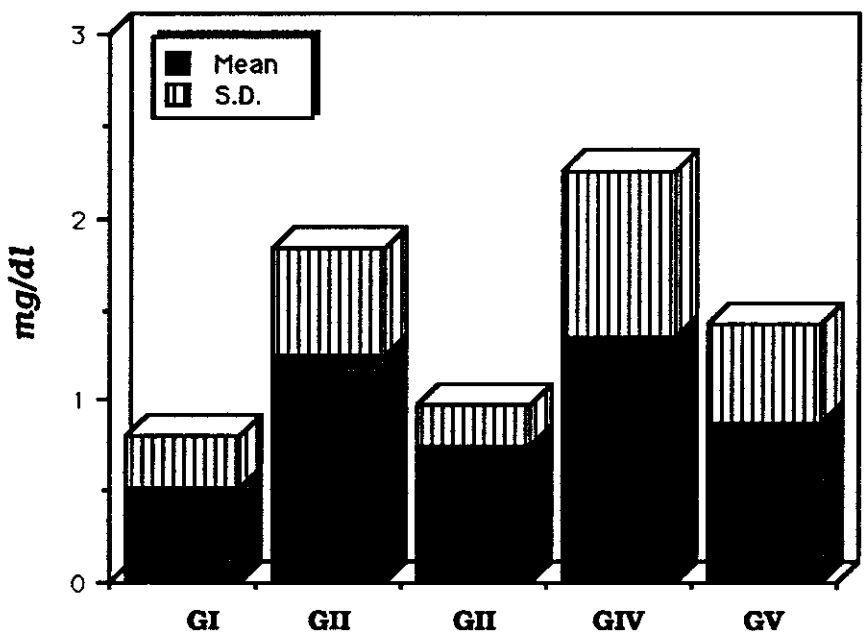

Fig. (18): Comparative study for CRP among all studied groups.

Table 10: Illustrates cytokines and CRP serum levels in relation to stage of liver disease in group II. Statistical variations between mild and severe chronic active hepatitis.

Item	Group (II A)		Group (II B)		t	P	Sig
	Mean	SD	Mean	SD			
TNF-α	22.5	4.7	50	18.3	6.7	<0.001	HS
IL-Iβ	7.1	0.8	10.8	3.1	4.5	<0.001	HS
IL-6	131.6	39.2	216.9	60.6	4.7	<0.001	HS
CRP	0.72	0.27	1.6	0.6	5.8	<0.001	HS

G (II A) Mild chronic active hepatitis
G (II B) severe chronic active hepatitis

Table 11: Illustrates cytokines and CRP serum levels in relation to stage of liver disease in group (IV) Comparative study between compensated and decompensated cirrhotic patients.

Item	Group (IV A)		Group (IV B)		t	P	Sig
	Mean	SD	Mean	SD			
TNF-α	26.69	8.7	56.7	18.9	7.5	<0.001	HS
IL-Iβ	7.7	2.25	12.3	2.9	6.25	<0.001	HS
IL-6	135.2	61.85	236	65.39	5.6	<0.001	HS
CRP	0.85	0.47	1.7	0.98	4.25	<0.001	HS

G (IV A) Compensated cirrhosis
G (IV B) Decompensated cirrhosis

Table 12: Correlation between biochemical / laboratory parameters and cytokine / CRP serum levels in group II

Item	TNF-α	IL-Iβ	IL-6	CRP
AST	0.33*	0.37*	0.49*	0.44*
ALT	0.37*	0.31*	0.31*	NS 0.16
TB	NS 0.07	NS 0.13	NS - 0.24	NS - 0.28
Alb	NS - 0.28	NS - 0.17	NS - 0.30	- 0.33*
PT	NS - 0.22	NS - 0.07	NS 0.15	NS - 0.06
W.B.Cs count	NS - 0.11	NS 0.03	NS - 0.11	NS - 0.16
Monocytic count	NS - 0.29	NS - 0.25	NS - 0.26	NS -0.22

* = *Significant (P< 0.05)*

NS = Not significnat (P > 0.05)

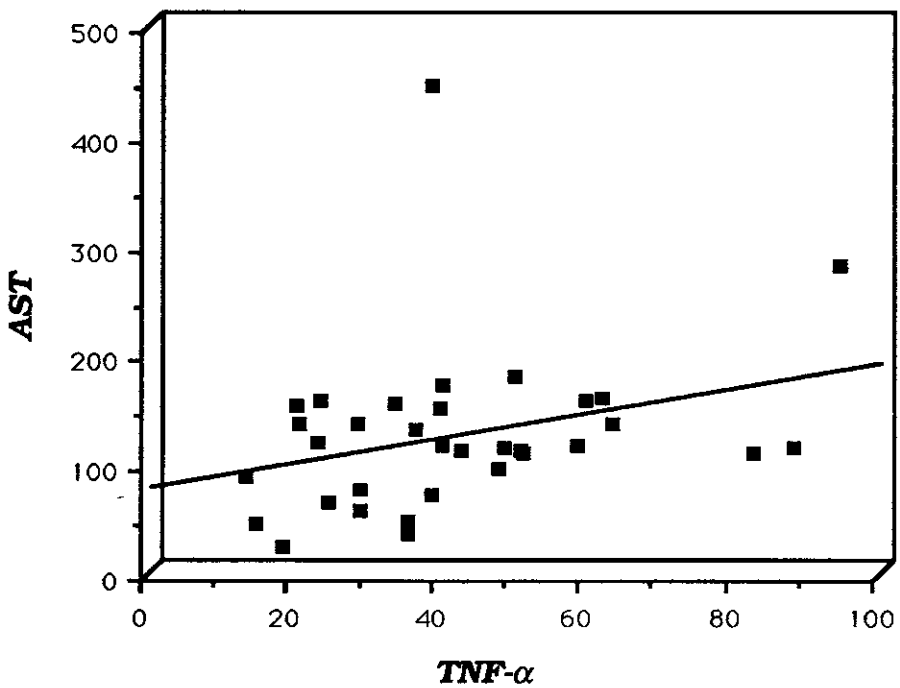

Fig. (19): Illustrates the correlation between TNF-α and AST in group II.

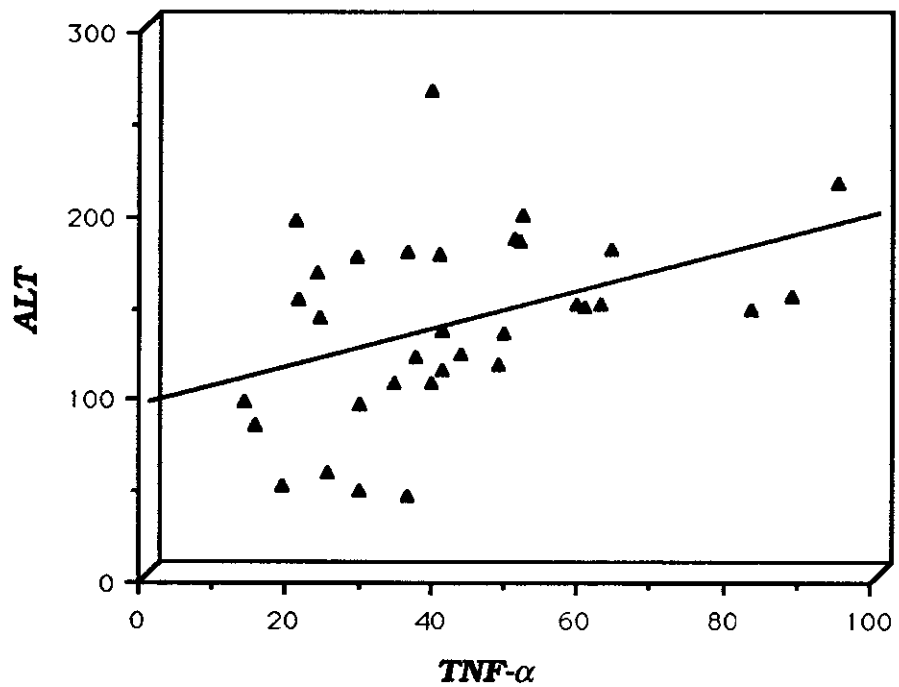

Fig. (20): Illustrates the correlation between TNF-α and ALT in group II.

Table 13: Correlation between biochemical / laboratory parameters and cytokine/ CRP serum levels in group (III).

Item	TNF- α	IL-Iβ	IL-6	CRP
AST	NS 0.52	NS 0.02	NS -0.30	NS -0.24
ALT	NS 0.59	NS 0.09	NS 0.52	NS - 0.19
TB	NS 0.38	NS 0.27	NS - 0.33	NS - 0.29
Alb	NS - 0.03	NS 0.26	NS - 0.05	NS - 0.48
PT	NS 0.47	NS 0.16	NS 0.17	NS - 0.43
W.B.Cs count	NS 0.30	NS 0.23	NS - 0.16	NS - 0.26
Monocytic count	NS 0.02	NS - 0.16	NS - 0.42	NS 0.24

* = *Significant (P< 0.05)*

NS = Not significnat (P > 0.05)

Table 14: Correlation between biochemical / laboratory parameters and cytokine / CRP serum levels in group(IV)

Item	TNF- α	IL-Iβ	IL-6	CRP
AST	NS - 0.15	NS - 0.07	NS -0.2	NS - 0.06
ALT	NS - 0.05	NS - 0.04	NS -0.16	NS -0.02
TB	0.27*	0.32*	0.34*	0.29*
Alb	- 0.67*	- 0.65*	- 0.58*	- 0.49*
PT	- 0.77*	- 0.66*	0.66*	- 0.56*
W.B.Cs count	NS - 0.02	NS - 0.09	NS - 0.02	NS - 0.06
Monocytic count	NS - 0.03	NS - 0.15	NS - 0.15	NS - 0.02

* = *Significant (P< 0.05)*

NS = Not significnat (P > 0.05)

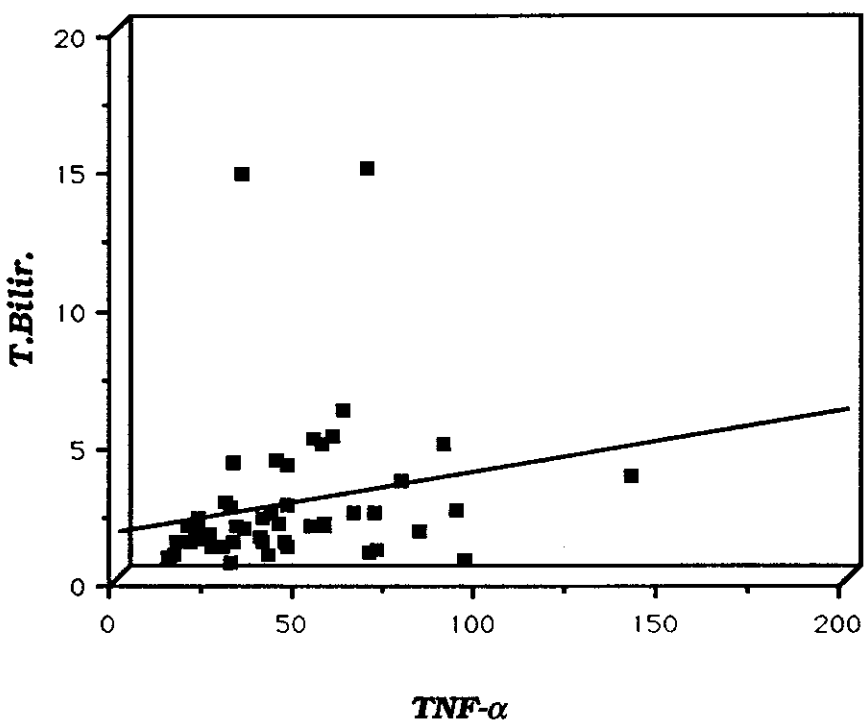

Fig. (21): Illustrates the correlation between TNF-α and T. bilirubin in group IV.

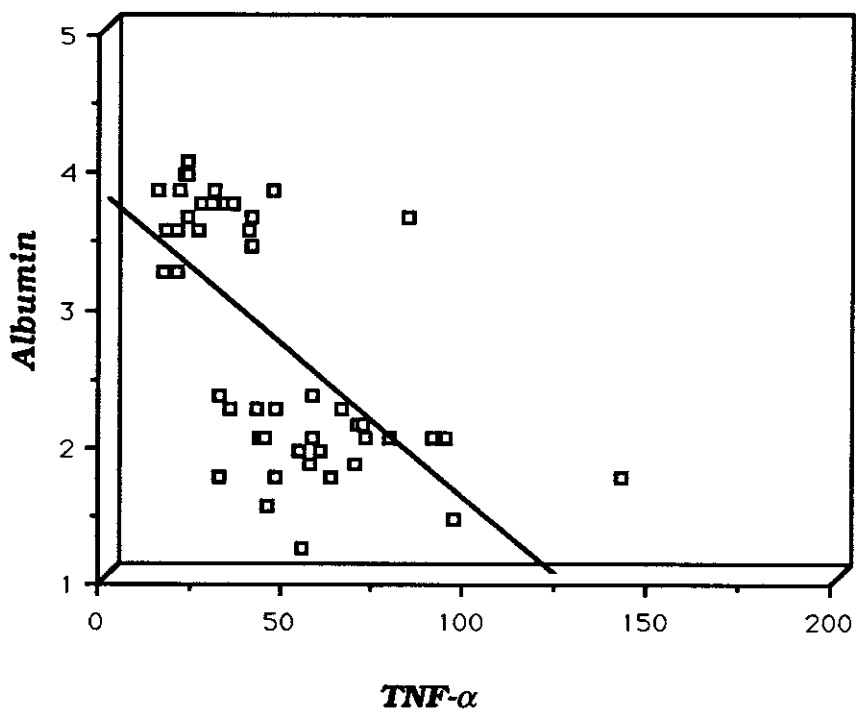

Fig. (22): Illustrates the correlation between TNF-α and albumin in group IV.

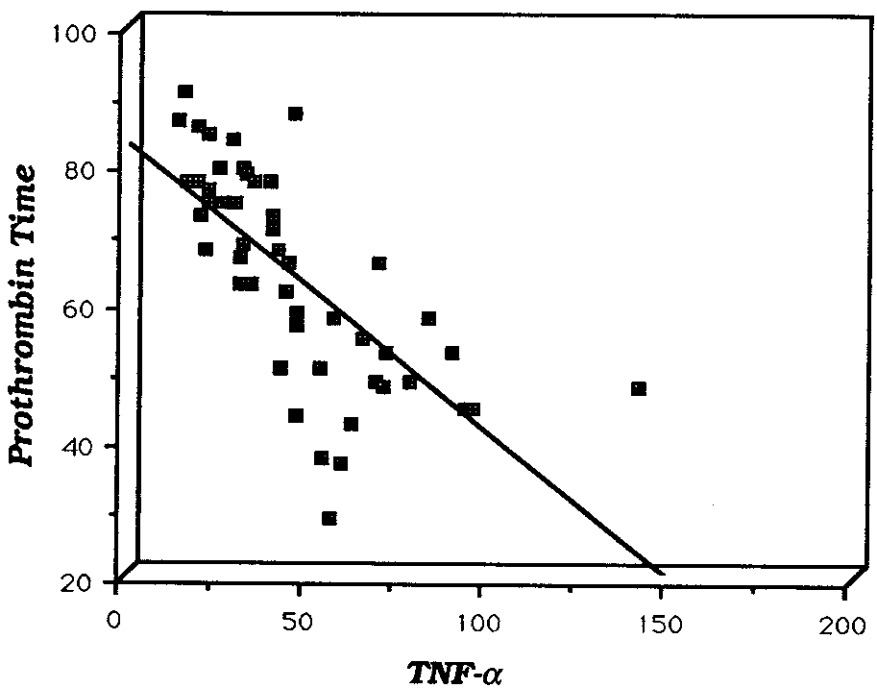

Fig. (23): Illustrates the correlation between TNF-α and prothrombin time in group IV.

Table 15: Correlation between biochemical / laboratory parameters and cytokine / CRP serum levels in group (V)

Item	TNF- α	IL-Iβ	IL-6	CRP
AST	NS 0.18	NS 0.25	0.34*	NS 0.15
ALT	NS - 0.07	NS 0.02	NS 0.22	NS - 0.06
TB	NS 0.08	NS 0.09	0.36*	NS - 0.11
Alb	- 0.36*	- 0.26*	- 0.38*	NS - 0.14
PT	- 0.44*	- 0.34*	0.47*	- 0.37*
W.B.Cs count	NS - 0.24	NS - 0.16	NS - 0.18	NS - 0.10
Monocytic count	NS 0.1	NS - 0.12	NS - 0.14	NS - 0.13

* = *Significant (P< 0.05)*

NS = Not significnat (P > 0.05)

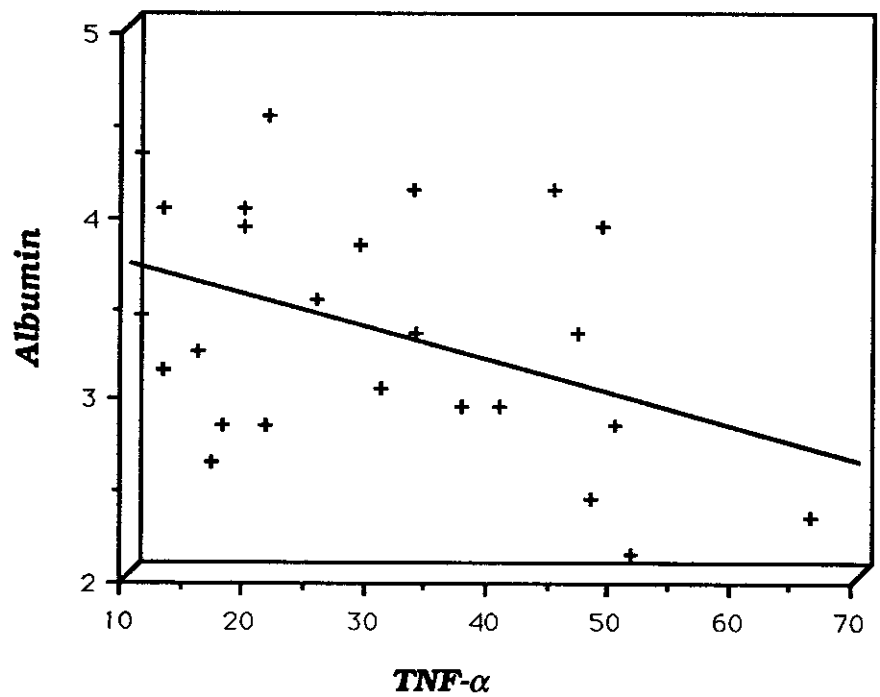

Fig. (24): Illustrates the correlation between TNF-α and albumin in group V.

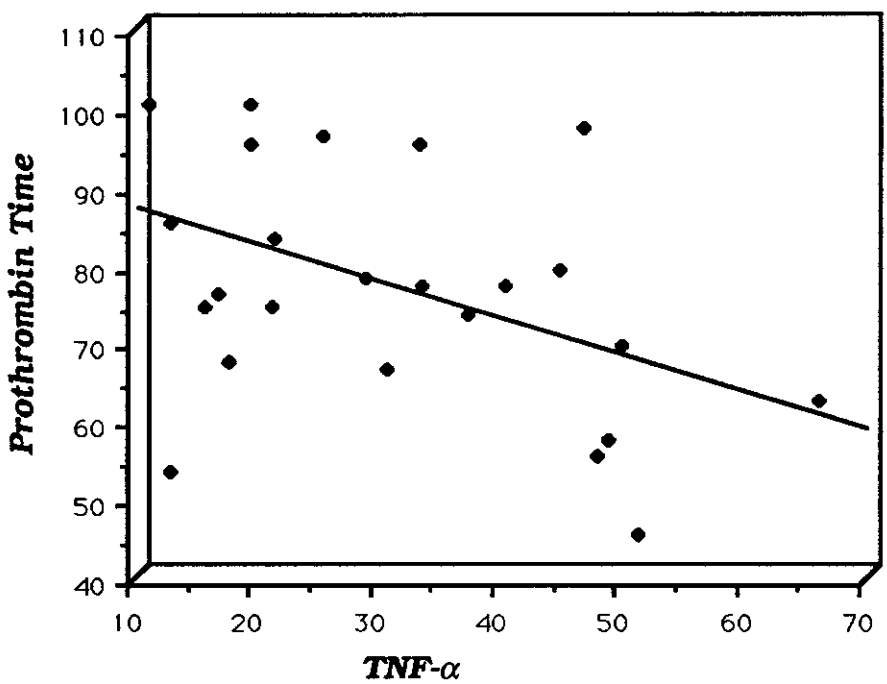

Fig. (24): Illustrates the correlation between TNF-α and prothrombin time in group V.

Table 16: Correlation among different cytokines and between cytokines and CRP in group (II)

Item	TNF- α	IL-IB	IL-6	CRP
TNF- α	======	0.84*	0.54*	0.56*
IL-Iβ	0.84*	======	0.49*	0.60*
IL-6	0.54*	0.49*	======	0.71*
CRP	0.56*	0.60*	0.71	======

* = *Significant (P< 0.05)*

NS = Not significant (P >0.05)

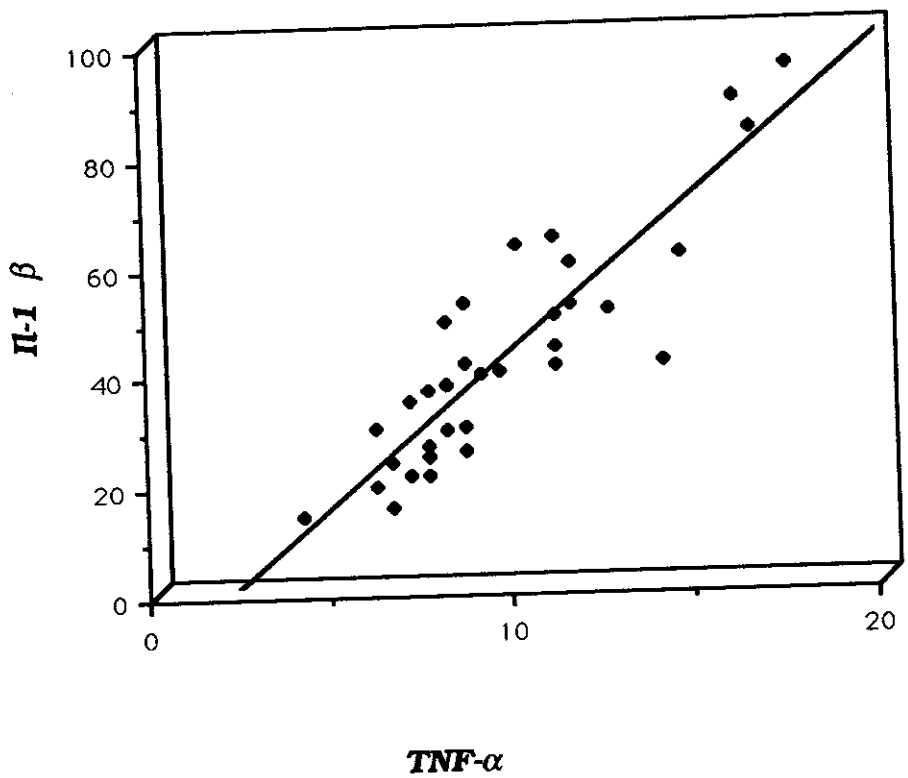

Fig. (25): Illustrates the correlation between TNF-α and IL-1β in group II.

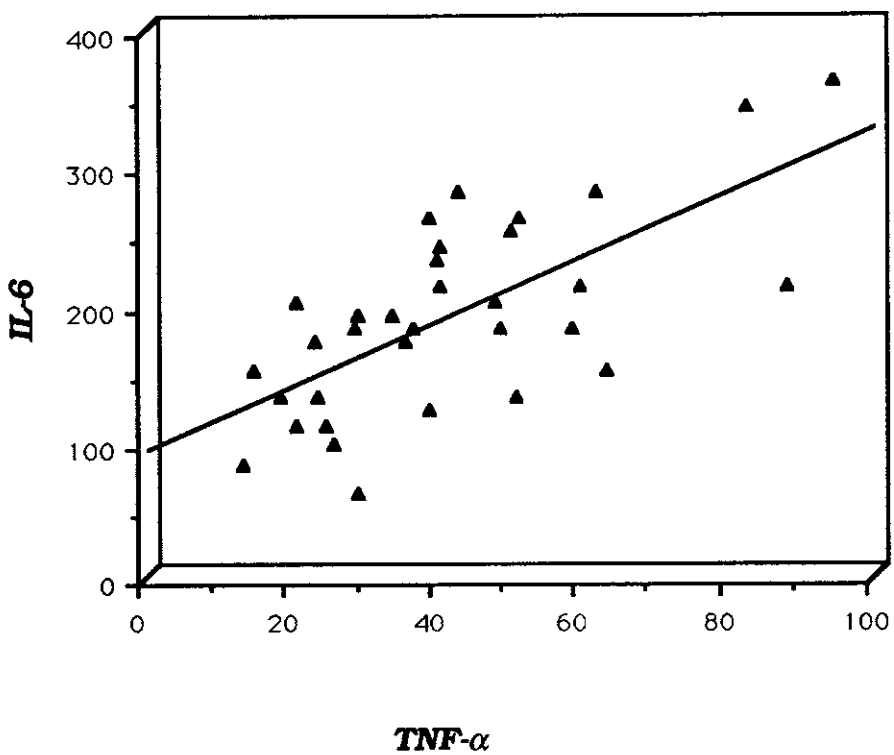

Fig. (26): Illustrates the correlation between TNF-α and IL-6 in group II.

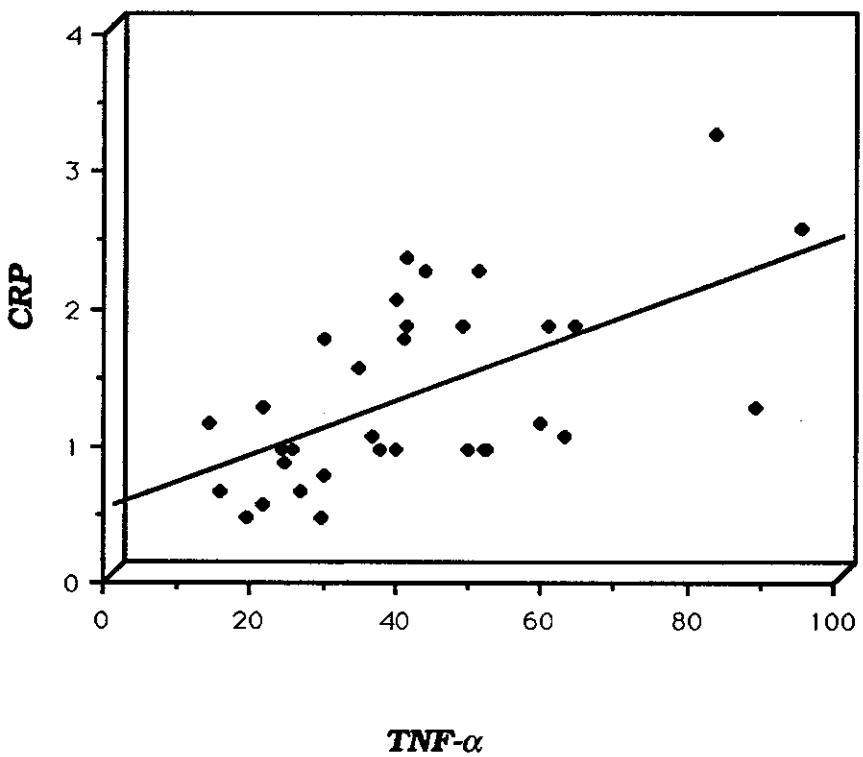

Fig. (27): Illustrates the correlation between TNF-α and CRP in group II.

Table 17: Correlation among different cytokines and between cytokines and CRP in group (III).

Item	TNF- α	IL-Iβ	IL-6	CRP
TNF- α	======	NS	NS	NS
	======	0.30	0.23	0.12
IL-Iβ	NS	======	NS	NS
	0.30	======	0.45	0.39
IL-6	NS	NS	======	NS
	0.23	0.45	======	0.45
CRP	NS	NS	NS	======
	0.12	0.39	0.45	======

* = *Significant (P< 0.05)*

NS = Not significant (P >0.05)

Table 18: Correlation among different Cytokines and between cytokines and CRP in group (IV).

Item	TNF-α	IL-Iβ	IL-6	CRP
TNF- α	======	0.79*	0.78*	0.65*
IL-Iβ	0.79*	======	0.75*	0.64*
IL-6	0.78*	0.75*	======	0.83*
CRP	0.65*	0.64*	0.83*	======

* = *Significant (P< 0.05)*

NS = Not significant (P >0.05)

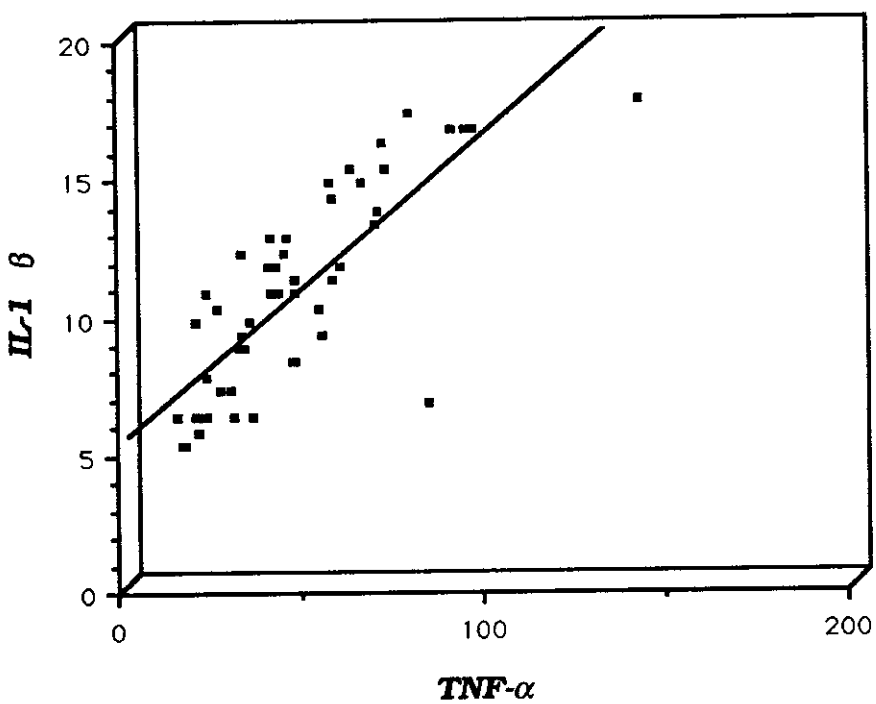

Fig. (28): Illustrates the correlation between TNF-α and IL-1β in group IV.

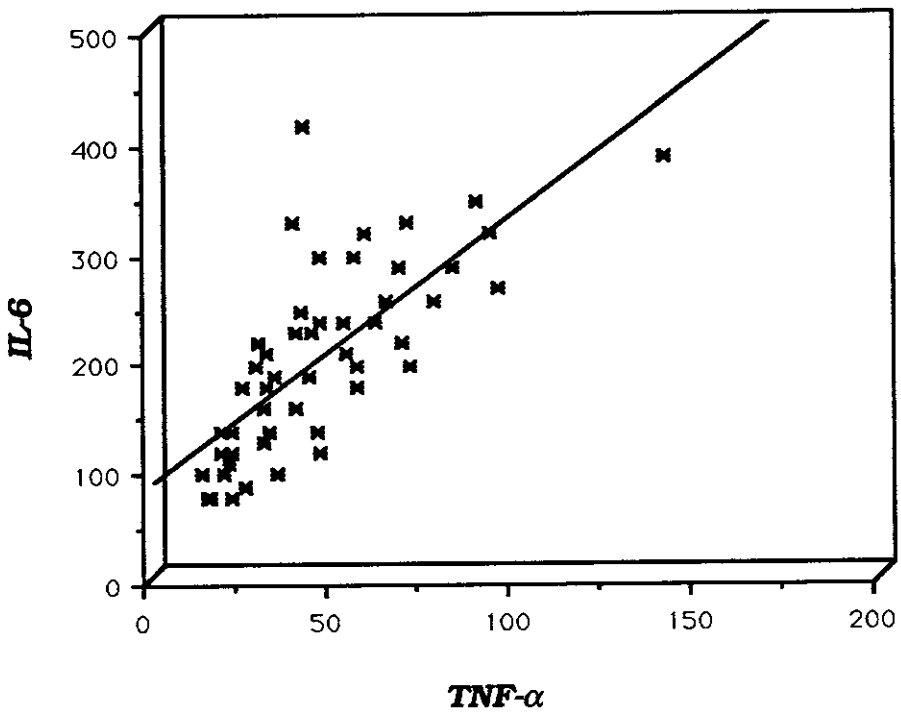

Fig. (29): Illustrates the correlation between TNF-α and IL-6 in group IV.

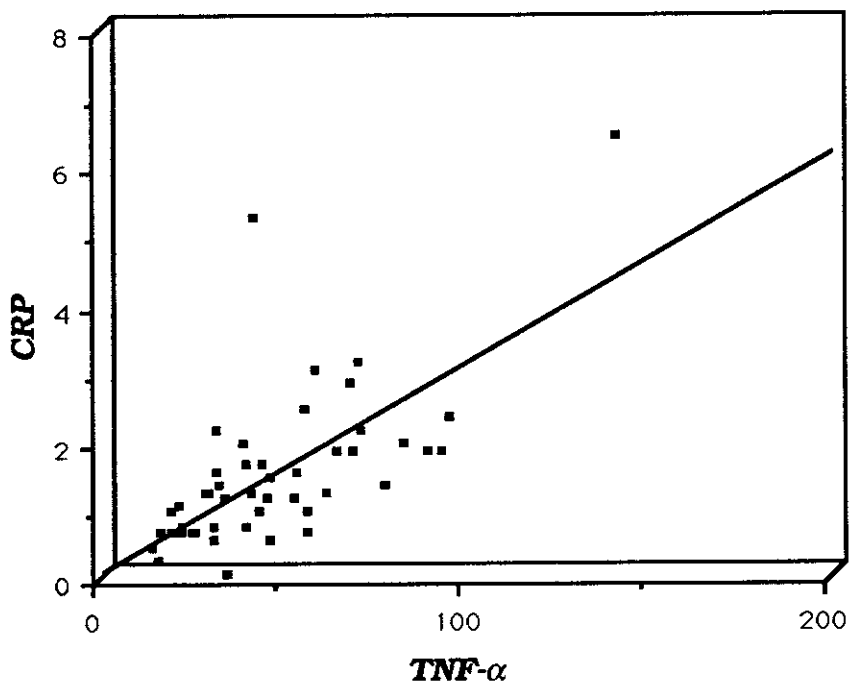

Fig. (30): Illustrates the correlation between TNF-α and CRP in group IV.

Table 19: Correlation among different cytokines and between cytokines and CRP in group (V).

Item	TNF- α	IL-I β	IL-6	CRP
TNF- α	======	0.79*	0.63*	0.46*
IL-I β	0.79*	======	0.50*	0.35*
IL-6	0.63*	0.50*	======	0.66*
CRP	0.46*	0.35*	0.66*	======

* = *Significant (P< 0.05)*

NS = Not significant (P >0.05)

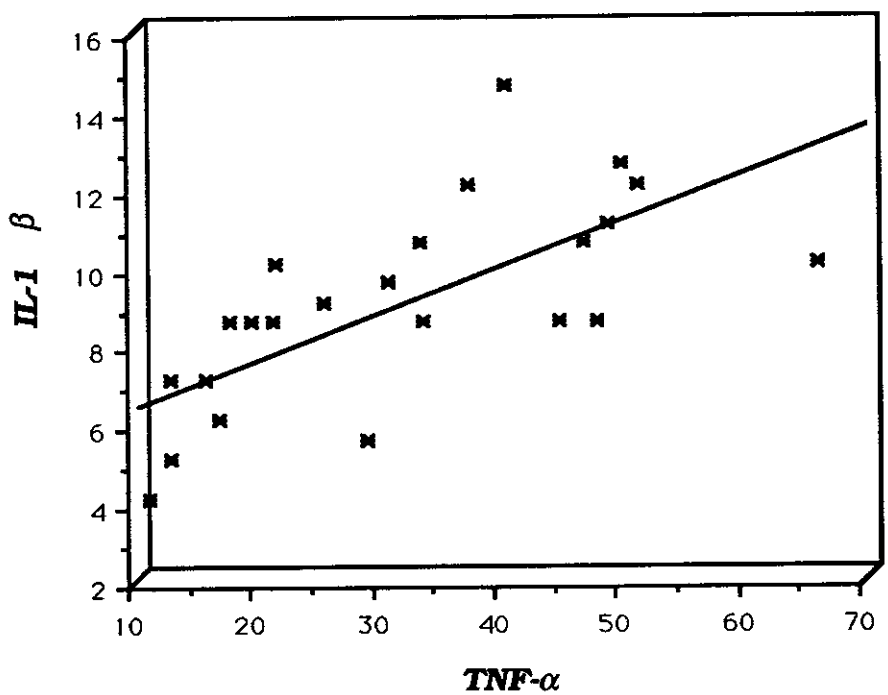

Fig. (31): Illustrates the correlation between TNF-α and IL-1β in group V.

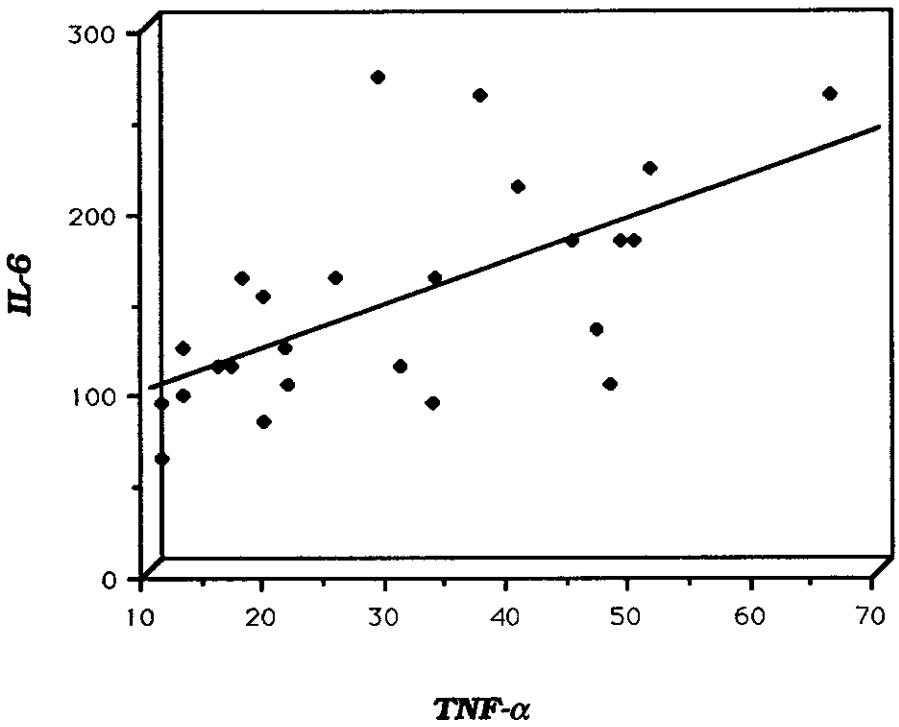

Fig. (32): Illustrates the correlation between TNF-α and IL-6 in group V.