Print ISSN 1110-208X.
Online ISSN 2357-0016

Diagnostic and Prognostic Value of Serum IL-18 & IL-35 in Patients with Bacterial Infection with or without Sepsis

Fatma M. Abdel Salam [a], Amira M. Abdelrahman [b], Dalia G. Laban[a], Tamer E. Eleraky [a]

[a] Hepatology, Gastroenterology and Infectious Diseases Department, Faculty of Medicine Benha University, Egypt.

[b] Clinical and Chemical Pathology, Faculty of Medicine Benha University, Egypt.

Corresponding to:
Dr. Dalia G. Labana.
Hepatology, Gastroenterology and Infectious Diseases Department, Faculty of Medicine Benha University, Egypt.

Email:
dalialaban1983@gmail.com

Received: 29 September 2024

Accepted:4 December 2024

Abstract:

Background: IL-18 modulates innate & adaptive immunity and its dysregulation can cause autoimmune or inflammatory diseases. IL-35 is related to occurrence and progression of many diseases; it is mainly involved in immune response, autoimmune diseases, infections and inflammation, IL 35 can be used as a promising therapeutic target. This study aimed to detect the value of IL-18 and IL 35 in the serum of patients of bacterial infection with or without sepsis and evaluate prognostic value of IL-18 and IL-35 in bacterial infection with or without sepsis. **Methods:** This prospective study was done on 70 patients admitted by bacterial infection. Patients were divided into two groups: Group 1: Bacterial Infection with Sepsis, group 2: Bacterial Infection without Sepsis and detect the level and prognostic value of IL-18 & IL 35 in the serum of patients. **Results:** Significant differences were noted between studied groups regarding IL-18 and IL-35 (p=0.001 and 0.012 respectively). Significant differences were noted between studied groups regarding APACHE Score, SOFA Score and prognosis (p=0.001). In both groups: There was negative correlation between IL-18 and IL-35 ROC curve analyses were done for serum IL18 showed: - Cutoff 14 , AUC 0.804, Sensitivity 86%, Specificity 60%, PPV 68% ,NPV 81%and Accuracy 73%. ROC curve analyses for serum IL35 showed: - Cutoff 150, AUC 0.626, Sensitivity 66%, Specificity71%, PPV 70%, NPV 68%and Accuracy 69%. **Conclusion:** elevation of IL-18 supports its role in diagnosing and monitoring sepsis. The decrease of IL-35 may indicate a failure of anti-inflammatory responses during sepsis.

Keywords: Serum IL-18; Serum IL-35 Bacterial Infection; Sepsis.

Introduction

Sepsis refers to host's uncontrolled immune response to infection, which in turn affects organ functions and causes death of critically ill patients; it is also one of the main problems which face the global health care system [1, 2]

Sepsis: Suspected source of clinical infection and two or more systemic inflammatory response syndrome (SIRS) criteria [3].

In-hospital mortality of patients with sepsis exceeds 10% but may be up to 40% in severe cases which deteriorate into septic shock [4].

Sepsis is characterized by an aggravated, uncontrolled, and self-sustaining inflammation which spreads via the circulation. Pathogens and their toxic products contribute to this process, as endotoxins are found in the blood of patients and associated with shock and multiorgan dysfunction [5].

The main mechanisms involved include: cytopathic injury, which is mediated by direct cell injury by pro-inflammatory mediators and/or other products of inflammation, tissue ischemia due to insufficient oxygen supply, with alteration to process of apoptosis [6].

The Sequential Organ Failure Assessment (SOFA) score is a simple and objective score that allows for calculation of both the number and the severity of organ dysfunction in six organ systems respiratory [partial pressure and saturation of oxygen], coagulatory [platelet counts], liver[serum bilirubin], cardiovascular[mean arterial pressure], neurologic [Glasgow coma scale], and renal [serum creatinine and urine output] [7, 8].

A widely used ICU prognostic scoring model, the Acute Physiology and Chronic Health Evaluation II (APACHE II) scoring system has been recognized It has shown to be an accurate measurement of patient severity and correlates strongly with outcome in critical patients [9].

Interleukin-18 (IL18, also known as interferon-gamma inducing factor) is a protein which encoded by the IL18 gene [10]. It is a proinflammatory cytokine. Many cell types, both hematopoietic cells and non-hematopoietic cells, have the potential to produce IL-18. Originally, IL-18 production was recognized in Kupffer cells, liver macrophages. However, IL-18 is constitutively expressed in non-hematopoietic cells, such as intestinal epithelial cells, keratinocytes, and endothelial cells [11].

IL-18 can modulate both innate and adaptive immunity and its dysregulation can cause autoimmune or inflammatory diseases [12]. Studies have shown that the severity and prognosis of sepsis may be related to IL-18 [13, 14].

IL-35 is produced by regulatory T cells and is related to occurrence and progression of a variety of diseases; it is mainly involved in immune response, autoimmune diseases, infections and inflammation, as a new inflammatory factor, IL 35 may have role as a promising therapeutic target [15, 16].

Therefore, it has been speculated that IL-18 and IL-35 are involved in sepsis. However, there are currently few studies on the expression of IL-18 and IL-35 and its correlation between them and thrombocytopenia in patients with sepsis [17]. And this is under research yet.

The purpose of this study was to detect the value of IL-18 and IL 35 in the serum of patients of bacterial infection with or without sepsis and evaluate diagnostic and prognostic value of IL-18 and IL-35 in bacterial infection with or without sepsis.

Patients and methods

This prospective study was done on 70 patients admitted by bacterial infection to Tanta fever hospital intensive care unit from May 2022 to April 2024.

An informed written consent was obtained from the patients. Every patient received an explanation of the purpose of the study and had a secret code number. The study was done after being approved by the Research Ethics Committee, Faculty of Medicine, Benha University.

Inclusion criteria were patients ≥ 18y, with sepsis caused by different pathogen infections (Gram negative, Gram positive, and anaerobic bacteria) and met the latest diagnostic criteria for sepsis.
For the diagnosis of sepsis, clinicians must obtain historical, clinical, laboratory, and radiographic data supportive of infection and organ dysfunction.
Symptoms includes fever which is the most common manifestation of sepsis. The absence of fever, however, does not exclude sepsis. Sepsis-induced hypothermia and the absence of fever are more likely in older adults and in people with chronic alcohol abuse or immunosuppression. Hypotension is the presenting abnormality in approximately 40% of patients with sepsis. In older adults, generalized weakness, agitation or irritation, or altered mental status may be the only manifestation [(30)].

Laboratory diagnosis
Laboratory testing should include a complete blood count with differential; basic metabolic panel; lactate, procalcitonin, and liver enzyme measurements; coagulation studies; and urinalysis. Arterial or venous blood sampling can determine the degree of acid-base abnormalities, which are common in sepsis and are likely secondary to tissue hypoperfusion (lactic acidosis) and renal dysfunction [(31)]**.**
Two sets of peripheral blood cultures were obtained (including a set from a central venous catheter, if present), as well as cultures of urine, stool (for diarrhea or recent antibiotic use), sputum (for respiratory symptoms), and skin and soft tissue (for skin abscess, ulceration, or drainage). Cerebrospinal, joint, pleural, and peritoneal fluid cultures are obtained as clinically indicated.[(29)]**.**

Imaging
Imaging studies included chest radiography, with additional studies as indicated (e.g., echocardiography for suspected endocarditis, computed tomography of the chest for empyema or parapneumonic effusion, computed tomography of the abdomen/pelvis for renal or abdominal abscess) [(32)]**.**

Exclusion criteria were below 18 y, females who are Pregnant and in puerperium, patients who refused testing, with a history of hematological malignancies, had a history of chemotherapy, received therapeutic anticoagulation or blood transfusion in the prior four weeks, died within 24 hours after they were hospitalized, advanced renal diseases and hemodynamically unstable patient.

Grouping: Patients (n=70) were selected and divided into two equal groups: **Group 1: (n=35)** Bacterial Infection with Sepsis. **Group 2: (n=35)** Bacterial Infection without Sepsis.
All studied cases were subjected to the following: Full history taking. Laboratory investigations, including **[**Complete blood count (CBC), C-reactive protein, Erythrocyte sedimentation rate (ESR), Blood culture, Liver function tests (Alanine amino transferase (ALT) , Aspartate amino transferase (AST) , Bilirubin), prothrombin time (PT), INR, Random blood glucose, Kidney function tests, Arterial blood gases, Serum blood level of sodium and potassium and detection of serum level of IL18 and IL35 by ELISA technique.]. **Abdominal pelvic ultrasound. Sequential Organ Failure Assessment (SOFA) wase carried out for all the patients within 24 hours after they were hospitalized.**

Sequential organ failure assessment (SOFA) is a scale widely used in emergencies, internal medicine, surgery, and ICU to evaluate the disease condition and prognosis of patients with multiple organ failure, which can dynamically reflect the changes of organ function [27]
SOFA measures the following:
Ratio of arterial oxygen tension to fraction of inspired oxygen (PaO2/FiO2)
Amount of vasoactive medication necessary to avoid hypotension
Bilirubin level
Platelet count
Glasgow coma score
Serum creatinine or urine output [28]

Acute Physiology and Chronic Health Evaluation (APACHE II) were carried out for all the patients within 24 hours after they were hospitalized.
A widely used ICU prognostic scoring model.
It has shown to be an accurate measurement of patient severity and correlates strongly with outcome in critical patients [25, 26].
It has many variables which are temperature; heart rate; respiratory rate; mean arterial blood pressure; oxygenation; arterial pH; serum potassium, sodium, and creatinine; hematocrit; white blood cell (WBC); and Glasgow Coma Scale

Blood Samples preparation:
Blood Samples preparation:
1. Serum preparation
After collection of the whole blood, the blood was allowed to be clotted by leaving it undisturbed at room temperature. This usually takes 10-20 minutes. the clot was removed by centrifuging at 2,000-3,000 rpm for 20 minutes. Then the supernatant (serum) was taken for liver enzymes , blood glucose, NA ,K ,CRP , IL18 and IL35.
2 ml of citrated blood 1-9 centrifuged for 15 minutes at 3000 rpm then the plasma was taken for PT, INR.
2 ml of EDTA blood was taken for C B C, IL 18 and IL 35 measurement**:**

Laboratory investigations include:
1. Complete blood count (CBC) with differential count performed on automated cell counter by swelab alpha plus apparatus manufactured in Sweden.
2. C-reactive protein is performed by latex agglutination method.
3. ESR by Westergren method.
4. Blood culture.
5. Liver function tests: - (ALT – AST) by enzymatic method by Respons 920 apparatus.
6. PT – INR by Sysmex apparatus.
7. Random blood glucose.
8. Kidney function tests: (blood Urea and Serum Creatinine)
9. Arterial blood gases.
10. Serum blood level of sodium and potassium.
11. Detection of serum level of IL18 and IL35 by ELISA technique.

Il18, ll35 assay was performed by using Nova kits by ELISA technique according to manufacturer's instructions.
Approval code: MD 8-9-2021

Statistical analysis
Statistical analysis was done by SPSS 24, IBM, Armonk, NY, United States of America. Quantitative variables were presented as mean and standard deviation (SD) and compared between the two groups utilizing unpaired Student's t- test and ANOVA (F) test. Qualitative variables were presented as frequency and percentage (%) and were analyzed utilizing the Chi-square test or Fisher's exact test when appropriate. Linear Correlation Coefficient [r] and Receiver Operating Characteristic curve analysis were used. A two tailed P value < 0.05 was considered statistically significant.

Results

No significant differences were noted between studied groups regarding age, sex, temperature, fever, dyspnea, cough, and neck rigidity. There were significant

differences regarding heart rate, respiratory rate, blood pressure and disturbed conscious level. **Table 1**

Non-significant differences were noted between studied groups regarding Hemoglobin (Hb), Red blood cells (RBCS), Hematocrit (HCT), Platelets (PLT), Neutrophils, lymphocyte, ESR, Ph and PO2 in ABG as p- value is (> 0.05) but significant difference were noted in White blood cells (WBCS), CRP as p-value is (0.001). There were significant differences noted between studied groups regarding liver functions and renal functions as p-value (<0.05), but non-significant in INR, Random Blood Sugar NA, K as P-value (>0.05). **Table 2**

Significant differences were noted between studied groups regarding IL-18 where ranged from [6.88: 35.27] with mean 16.66 ± 8.87 for group 1 and ranged from [6.54-37.7] with mean 9.74 ± 7.25 for group 2 as p- value (0.001). Also, significant differences were noted between studied groups regarding IL-35 [63.5-388.02] with mean 114.60 ± 78.15 for group 1 and [56.25 – 835.71 with mean 186.44± 143.92for group 2, as p- value (0.012). Significant differences were noted between studied groups regarding APACHE Score, SOFA Score and prognosis as p- value (0.001). Non-significant differences were noted between studied groups regarding blood culture, ultrasound finding, site of infections. **Table 3**

There are significant differences between studied groups as regard prognosis as p-value is 0.001

Table 1: Age, sex, vital signs and clinical manifestations of the studied groups.

		Range	**Mean ± S. D**	**p. value**
Age(years)	Group 1	19 – 93	59.40 ± 19.91	0.260
	Group 2	18 – 84	53.91 ± 20.52	
Gender (Sex)		**Group 1 (n=35)**	**Group 2 (n=35)**	**P-value**
Male	**N**	17	17	1.0
	%	48.6%	48.6%	
Female	**N**	18	18	
	%	51.4%	51.4%	
Vital signs				
Temperature (℃)		38.54 ± 0.69	38.35 ± 0.57	0.197
Heart rate (beat /min)		97.57 ± 11.46	79.80 ± 8.96	**0.001***
RR (Cycle/min)		21.43 ± 2.23	20.20 ± 1.75	**0.012***
Blood pressure (mmHg)	**Hypotensive**	28 80.0%	3 8.6%	**0.001***
Clinical manifestations				
Fever		34 97.1%	34 97.1%	1.0
DCL		42.9% 20	68.6% 11	**0.030***
Dyspnea		8 22.9%	6 17.1%	0.550
Cough		7 20.0%	12 34.3%	0.179
Neck rigidity		9 25.7%	3 8.6%	0.057

*: statistically significant as P value <0.05.
RR respiratory rate, DCL disturbed conscious level

Table 2: Complete blood count, ESR & CRP parameters, PH, po2 in ABG, Liver functions, coagulation profile (INR), renal functions and Electrolytes (Na- K) of the studied groups

		Range			Mean	±	S. D	p. value
HB(g/dl)	Group 1	6.9	–	16	11.20	±	2.40	0.214
	Group 2	8.8	–	15.1	11.84	±	1.83	
RBCS	Group 1	2.36	–	5.6	3.92	±	0.80	0.106
(mil/cmm)	Group 2	2.78	–	6.04	4.22	±	0.73	
HCT (%)	Group 1	20.7	–	48	33.34	±	7.24	0.427
	Group 2	23.1	–	45	34.56	±	5.44	
PLT (/cmm)	Group 1	51000	–	621000	223485.71	±	130541.04	0.744
	Group 2	46000	–	506000	233314.29	±	119551.56	
WBCs(/cmm)	Group 1	3500	–	56400	22057.19	±	9356.27	**0.001***
	Group 2	1600	–	23200	10082.86	±	4196.32	
Neutrophil (%)	Group 1	40.7	–	94.9	79.87	±	12.69	0.648
	Group 2	34.3	–	94.8	78.48	±	12.57	
Lymph(%)	Group 1	2.2	–	70.5	15.00	±	13.78	0.641
	Group 2	3.3	–	58	16.41	±	11.22	
ESR 1(mm)	Group 1	15	–	100	37.86	±	22.04	0.255
	Group 2	15	–	90	32.29	±	18.40	
ESR 2 (mm)	Group 1	20	–	90	55.00	±	20.38	0.159
	Group 2	20	–	110	47.57	±	22.83	
CRP(u/l)	Group 1	16.97	–	250	123.27	±	69.01	**0.001***
	Group 2	6	–	215	71.33	±	56.92	
PH	Group 1	7.26	–	7.59	7.39	±	0.08	0.575
	Group 2	7.35	–	7.52	7.40	±	0.05	
PO2(mmHg)	Group 1	35.38	–	173.9	79.66	±	25.95	0.773
	Group 2	39.2	–	115	78.21	±	14.32	
RBS (mg/dl)	Group 1	87.2	–	428	183.61	±	100.57	0.636
	Group 2	79	–	594	172.22	±	98.27	
Liver functions								
ALT (u/l)	Group 1	10.9	–	189.8	50.68	±	42.71	**0.003***
	Group 2	8.6	–	77	26.91	±	15.97	
AST (u/l)	Group 1	12.5	–	278.5	65.55	±	63.02	**0.001***
	Group 2	12	–	96	28.69	±	16.45	
Total	Group 1	0.1	–	5.67	1.69	±	0.99	**0.033***
bilirubin(mg/dl)	Group 2	0.6	–	5.4	1.22	±	0.78	
Direct	Group 1	0.3	–	3.79	1.10	±	0.69	**0.018***
bilirubin(mg/dl)	Group 2	0.3	–	3.2	0.75	±	0.52	
INR	Group 1	0.8	–	2.3	1.34	±	0.37	0.606
	Group 2	1	–	4.1	1.28	±	0.52	
Renal functions								
Urea(mg/dl)	Group 1	25	–	298	105.66	±	76.05	**0.031***
	Group 2	15	–	190	46.49	±	32.33	
Creatinine	Group 1	0.26	–	7.75	2.22	±	1.63	**0.026***
(mg/dl)	Group 2	0.4	–	2.6	1.08	±	0.41	
Electrolytes (Na- K)								
Na(mmol/l)	Group 1	128	–	151.7	137.85	±	5.10	0.711
	Group 2	129	–	171	138.38	±	6.56	
K(mmol/l)	Group 1	2.5	–	6.8	4.00	±	0.86	0.905
	Group 2	3.16	–	4.69	3.98	±	0.40	

Hb: haemoglobin,RBCs Red blood corpuscles, HCT Haematocrit value ,PLT: platelet count, WBCs: white blood cells, ESR1erythrocyte sedimentation rate first hour, ESR2erythrocyte sedimentation rate second hour , CRP C reactive protein ,PO2 Partial pressure of oxygen, RBS random blood sugar, Alt alanine amino transferase Alt aspartate amino transferase, K: potassium, Na: Sodium

*: statistically significant as P value <0.05

Table 3: Serum level IL-18 & IL-35, APACHE Score, SOFA score, ultrasound findings, site of infections of the studied groups.

		Range			**Mean**	**±**	**S. D**	**p. value**
IL-18 (pg/ml)	Group 1	6.88	–	35.27	16.66	±	8.87	**0.001***
	Group 2	6.54	–	37.7	9.74	±	7.25	
IL-35 (pg/ml)	Group 1	63.5	–	388.02	114.60	±	78.15	**0.012***
	Group 2	56.25	–	835.71	186.44	±	143.92	
APACHE score	Group 1	3	–	34	17.40	±	8.42	**0.001***
	Group 2	0	–	19	7.26	±	4.98	
SOFA score	Group 1	2	–	14	7.23	±	3.98	**0.001***
	Group 2	1	–	5	2.54	±	1.31	
				Group 1	**Group 2**			
Blood culture	**No growth**		**N**	22	23			0.364
			%	62.9%	65.7%			
	gram negative bacteria		**N**	10	6			
			%	28.6%	17.1%			
	gram positive bacteria		**N**	3	6			
			%	8.6%	17.1%			
No abnormality			**N**	11	10			0.794
			%	31.4%	28.6%			
Hepatomegaly			**N**	8	10			0.584
			%	22.9%	28.6%			
Splenomegaly			**N**	3	5			0.452
			%	8.6%	14.3%			
Cirrhotic			**N**	11	8			0.420
			%	31.4%	22.9%			
Ascites			**N**	3	1			0.303
			%	8.6%	2.9%			
Site of infection								
Chest infection			N	8	13			0.095
			%	22.9%	37.1%			
C N S infection			N	19	18			
			%	54.3%	51.4%			
UTI			N	3	4			
			%	8.6%	11.4%			
Others			N	5	0			
			%	14.3%	0.0%			
Prognosis								
ICU death			**N**	16	1			**0.001***
			%	45.7%	2.9%			
Improved			**N**	19	34			
			%	54.3%	97.1%			

*: statistically significant as P value <0.05

Il18 interleukin 18, il35 interleukin 35, APACHE Acute Physiology and Chronic Health Evaluation . SOFA Sequential Organ Failure Assessment, CNS infection central nervous system infection, UTI urinary tract infection

Table 4: **Correlation between serum IL-18 & IL-35 with all patients' parameters in group 1 and group 2.**

Group 1	**IL-18**		**IL-35**	
	r	**P value**	**r**	**P value**
IL-35 (pg/ml)	-0.102	0.004*		
Age (years)	-0.460	0.345	0.072	0.679
Temperature (℃)	0.026	0.882	0.210	0.226
Heart rate (b/min)	0.255	0.045*	0.182	**0.029***
R R(cycle/min)	0.230	0.033*	0.128	**0.042***
HB (g/dl)	0.070	0.688	0.356	**0.036***
RBCS (mil/cmm)	0.090	0.608	0.256	0.137
HCT (%)	0.076	0.663	0.340	**0.046***
PLT (/cmm)	-0.084	0.047*	-0.201	0.022
WBCs (/cmm)	0.045	0.001*	0.099	**0.021***
Neutrophil (%)	0.278	0.010*	0.198	**0.028***
Lymph (%)	-0.280	0.103	0.124	0.479
ESR 1 (mm)	-0.044	0.801	0.071	0.684
ESR 2 (mm)	0.011	0.950	-0.127	0.473
CRP (U/L)	0.213	0.025*	0.058	**0.041***
PH	-0.076	0.664	0.046	0.792
PO2 (mmhg)	-0.235	0.174	-0.038	0.829
ALT (U/L)	0.344	0.043*	0.075	0.670
AST (U/L)	0.143	0.413	-0.044	0.802
Total bilirubin (mg/dl)	0.216	0.521	0.089	0.610
Direct bilirubin (mg/dl)	0.252	0.314	0.108	0.537
INR	0.216	0.212	-0.035	0.841
Urea (mg/dl)	0.125	0.476	0.042	0.809
Creatinine (mg/dl)	0.144	0.040*	0.213	**0.029***
Na (mmol/l)	0.015	0.934	-0.024	0.892
K(mmol/l)	0.029	0.869	0.174	0.319
APACHE score	0.114	0.001*	-0.157	**0.021***
SOFA score	0.029	0.002*	-0.331	**0.042***
RBS(mg/dl)	0.087	0.621	0.183	0.293
Group 2	**IL-18**		**IL-35**	
	r	**P value**	**r**	**P value**
IL-35 (pg/ml)	-0.056	0.013*		
Age (years)	-0.051	0.770	-0.162	0.353
Temperature (℃)	-0.187	0.283	-0.074	0.674
Heart rate (b/min)	0.323	0.049*	0.158	**0.030***
R R(cycle/min)	0.004	0.005*	0.201	**0.024***
HB (g/dl)	-0.076	0.662	0.060	0.734
RBCS (mil/cmm)	-0.132	0.450	0.256	0.137
HCT (%)	-0.100	0.566	0.105	0.548
PLT (/cmm)	-0.265	0.124	-0.022	0.045
WBCs (/cmm)	0.044	0.018*	0.084	**0.031***
Neutrophil (%)	0.050	0.027*	0.048	**0.024***
Lymph (%)	0.010	0.953	0.029	0.870
ESR 1 (mm)	-0.171	0.327	-0.042	0.809
ESR 2 (mm)	-0.245	0.156	0.023	0.895
CRP (U/L)	0.031	0.041*	0.082	**0.031***
PH	-0.162	0.353	-0.068	0.698
PO2 (mmhg)	0.069	0.695	0.050	0.776
ALT (U/L)	0.026	0.884	0.102	**0.042***
AST (U/L)	0.166	0.340	-0.109	0.533
Total bilirubin (mg/dl)	0.110	0.621*	0.035	0.723
Direct bilirubin (mg/dl)	0.130	0.507*	0.106	0.643
INR	-0.125	0.473	0.009	0.958
Urea (mg/dl)	-0.042	0.809	-0.063	0.721
Creatinine (mg/dl)	0.008	0.002*	0.081	**0.032***
Na (mmol/l)	-0.147	0.398	-0.090	0.608
K(mmol/l)	-0.065	0.710	-0.124	0.476
APACHE score	0.067	0.003*	-0.213	**0.021***
SOFA score	0.220	0.004*	-0.033	**0.031***
RBS(mg/dl)	-0.186	0.291	-0.226	0.198

r: Pearson correlation, * Significant p value < 0.05

RR Respiratory rate Hb: haemoglobin, PLT: platelet count, WBCs: white blood cells APACHE Acute Physiology and Chronic Health Evaluation. SOFA Sequential Organ Failure Assessment RBS random blood sugar, ESR1erythrocyte sedimentation rate first hour, ESR2 erythrocyte sedimentation rate second hour, CRP C reactive protein ,PO2 Partial pressure of oxygen, RBS random blood sugar, Alt alanine amino transferase Alt aspartate amino transferase, K: potassium, Na: Sodium

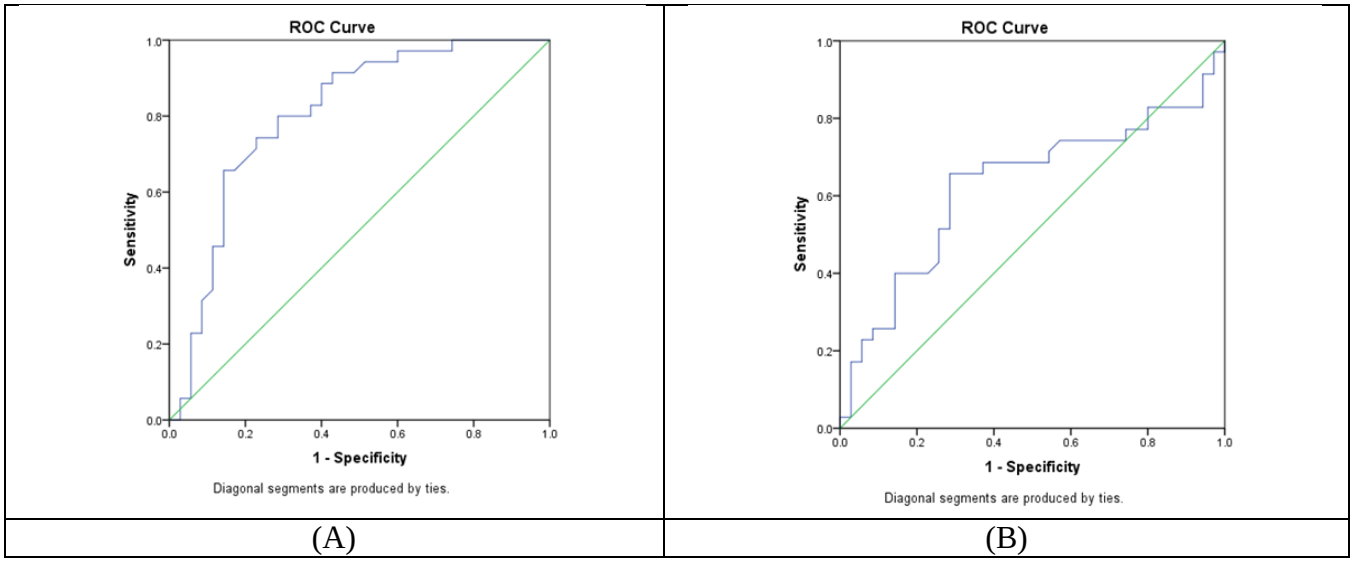
As regarding correlation between IL-18 and all patient parameters in group 1 showed that :-

Positive correlation between IL-18 and (Heart Rate – Respiratory Rate – WBCS – Neutrophils – CRP –creatinine). positive correlation between IL-18 and (APACHE score and SOFA score). negative correlation between IL-18 and platelets. negative correlation between IL-18 and IL-35.

As regarding correlation between IL-35 and all patient parameters in group2 showed that: -

Positive correlation between IL-35 and (Heart Rate – Respiratory Rate – WBCS – Neutrophils – CRP – creatinine). negative correlation between IL-35 and (APACHE score and SOFA score). negative correlation between IL-35 and platelets. negative correlation between IL-35 and IL-18. **Table 4**

ROC curve analyses were done for serum IL18 showed: - Cutoff 14, AUC 0.804, Sensitivity 86%, Specificity 60%, PPV 68%, NPV 81%, Accuracy 73%. ROC curve analyses were done for serum IL35 showed: - Cutoff 150, AUC 0.626, Sensitivity 66%, Specificity71%, PPV 70%, NPV 68%, Accuracy 69%. **Figure 1**



(A) (B)

Figure 1: ROC curve analysis of IL18 (A).
ROC curve analysis of IL35 (B).

ROC curve analyses were done for serum IL18 showed :-

Cutoff 14 AUC0.804 Sensitivity 86% Specificity 60% PPV 68% NPV 81% Accuracy 73%.

ROC curve analyses were done for serum IL35 showed :-

Cutoff 150 AUC 0.626 Sensitivity 66% Specificity71%PPV 70% NPV 68% Accuracy 69%.

Discussion

The current study shows that, no significant differences were noted between studied groups regarding age and sex as p-values were 0.260 and 1.0. this comes in agreement with, Zhu et al., who conducted a study about IL-18 and IL-35 in the serum of patients with sepsis thrombocytopenia and the clinical significance.

One hundred and sixty-six patients with sepsis and 80 healthy subjects were included. They reported that no significant differences were noted between studied groups regarding age and sex as p- values were 0.34 and 0.65 [17].

In the present study, no significant differences were noted between studied groups regarding temperature as the p-value is (0.197), but heart rate and respiratory rate were significantly increased in group 1 compared to group 2 as p-values respectively (0.001) and (0.012). Significant differences were noted between studied groups regarding blood pressure as the p-value is 0.001, with higher incidence of hypotension in group 1. In partial accordance with these results, Li et al., conducted a study about the clinical value of serum interleukin-18 in neonatal sepsis diagnosis and mortality prediction. They prospectively enrolled 91 septic neonates and 31 non-septic neonates. They found no significant difference between studied groups regarding temperature and respiratory rate, however, heart rate was significantly higher in patients with sepsis compared to the healthy control (p=0.018) [18].

According to the current study, non-significant differences were noted between studied groups regarding random blood sugar, hemoglobin (Hb), RBCS, hematocrit (HCT), platelets (PLT), neutrophils, and lymphocytes as p-value is (> 0.05) but WBCS were significantly increased in group 1 (p-value=0.001). In partial agreement with the current study, Zhu et al., reported that no significant differences were noted between patients with sepsis and healthy subjects regarding platelet count with a significant increase in WBCS in patients with sepsis (p<0.01). However, they disagreed with us in reporting that hemoglobin levels was significantly higher in the healthy control group [17].

As regards our results, non-significant differences were noted between studied groups regarding ESR (1st and 2nd hours) as the p-value (0.159) but CRP was significantly increased in group 1 (p= 0.001). According to our findings, the liver functions including ALT, AST, total bilirubin, and direct bilirubin were significantly increased in group 1 compared to group 2 (p=0.05), but a non-significant difference was reported in INR as P-value (0.606). In accordance with the present study, Li et al., found that CRP was significantly higher in patients with sepsis compared to the healthy control (p<0.05). ALT and AST were significantly higher in patients with sepsis compared to the healthy control (p<0.05) [18].

Regarding the present study, the renal functions including urea and creatinine were significantly increased in group 1 compared to group 2 as p-values (0.031) and (0.026) respectively. Non-significant differences were noted between studied groups regarding electrolytes (Na- K) as p-value (>0.05). In agreement with us, Zhu et al., come in agreement as regard the serum creatinine was significantly higher in patients with sepsis compared to the healthy control group (p<0.01) [17].

In the present study, IL-18 was significantly increased in group 1 compared to group 2 (p=0.001). IL-35 was significantly decreased in group 1 compared to group 2 (p=0.012). The range of IL-18 was with a mean of 16.66 ± 8.87 for group 1 and was with a mean of 9.74 ± 7.25 for group 2. The range of IL-35 was with a mean of 114.60 ± 78.15 for group 1 and was with a mean of 186.44± 143.92 for group 2. In accordance with the present study , Li et al., declared that IL-18 level was significantly higher in patients with sepsis compared to the healthy control (p<0.05), reaching the highest levels in the non-survival sepsis group (P< 0.001) [18].

In parallel with the present study, Zhixia et al., investigated the application value of peripheral blood IL-18/IL-35 in the evaluation of sepsis and its prognosis. They included 120 patients and reported that the concentration of IL-18 and IL-35

were significantly increased in patients with sepsis ($p<0.05$) [19].

According to our results, APACHE and SOFA scores were significantly increased in group 1 compared to group 2 ($p=0.001$).

In agreement with us, Lei., investigated the value of interleukin-35 (IL-35) in the diagnosis of sepsis patients. A total of 110 patients with confirmed sepsis (sepsis group) and 110 patients with systemic inflammatory response syndrome (SIRS) were selected as the control group. They showed that APACHEII and SOFA scores in the sepsis group were higher than those in the control group, and the difference was statistically significant ($P<0.05$) [20].

According to the current results, significant differences were noted between studied groups regarding prognosis as p-value (0.001), with poor prognosis in group 1. There were 16 (45.7%) dead patients and 19 (54.3%) improved patients in group 1, however, there were 1 (2.9%) dead patient and 34 (97.1%) improved patients in group 2.

In agreement with our results, Pairattanakorn et al., performed a prospective cohort study evaluated various scoring systems for predicting mortality in sepsis patients. They reported significant differences in mortality rates between those classified as having sepsis versus controls, with a p-value of 0.001 for mortality prediction using the SOFA score [21].

In the present study, regarding the correlation between IL-18 and IL-35 with all patient's parameters in both groups, positive correlations were found between (IL-18 and IL-35) with (heart rate, respiratory rate, WBCS, neutrophils, CRP, and creatinine,). There were negative correlations between (IL-18 and IL-35) with platelets, between IL-35 and (APACHE, and SOFA scores), and between IL-35 and IL-18.

In accordance with these results , Li et al., declared that IL-18 level was positively correlated with heart rate, respiratory rate, and CRP level ($p<0.05$), but no correlation was found with neutrophil count [18].

In consistent with the present findings, Zhiyong., reported that IL-35 level was positively correlated with the scores of CRP and WBC, but negatively correlated with APACHE II ($P<0.05$) [22].

In the present study, for the prediction of sepsis, serum IL18 at a cutoff point =14 and an AUC of 0.804 showed a sensitivity of 86%, specificity of 60%, PPV of 68%, NPV of 81%, and accuracy of 73%. Serum IL35 at a cutoff point =150 and an AUC of 0.626 showed a sensitivity of 66%, specificity of 71%, PPV of 70%, NPV of 68%, and accuracy of 69%.

These results were in agreement with, Yucang et al., who revealed that the area under the ROC curve (AUC) of IL-35 for diagnosing infection was 0.76. When the cut-off value of IL-35 was 41.97 ng, the sensitivity and specificity were 94.00% and 60.00% [23]. Additionally, Lin et al., found that serum IL-18 had a significant role in predicting short-term prognosis in critically ill patients with acute kidney injury. The area under the curve (AUC) for IL-18 was reported as 0.872, with a sensitivity of 80% and specificity of 95% at an optimal cut-off point [24].

Conclusion

The elevation of IL-18 supports its role in diagnosing and monitoring sepsis, while the decrease of IL-35 may indicate a failure of anti-inflammatory responses during septic conditions. Serum IL-18 is a superior biomarker for predicting sepsis compared to serum IL-35, demonstrating higher sensitivity and accuracy. While IL-35 may still provide some utility in clinical settings, its lower diagnostic performance suggests it should not be relied upon as the primary marker for sepsis detection.

References

1. Dewitte A, Lepreux S, Villeneuve J, Rigothier C, Combe C, Ouattara A, et al. Blood platelets and sepsis pathophysiology: A new therapeutic prospect in critically ill patients? Ann Intensive Care. 2017;7:115.
2. Larkin CM, Santos-Martinez MJ, Ryan T, Radomski MW. Sepsis-associated thrombocytopenia. Thrombosis Research. 2016;141:11-6.
3. Dugar S, Choudhary C, Duggal A. Sepsis and septic shock: Guideline-based management. Cleve Clin J Med. 2020;87:53-64.
4. Singer M, Deutschman CS, Seymour CW, Shankar-Hari M, Annane D, Bauer M, et al. The Third International Consensus Definitions for Sepsis and Septic Shock (Sepsis-3). Jama. 2016;315:801-10.
5. Mayr FB, Yende S, Angus DC. Epidemiology of severe sepsis. Virulence. 2014;5:4-11.
6. Pena OM, Hancock DG, Lyle NH, Linder A, Russell JA, Xia J, et al. An Endotoxin Tolerance Signature Predicts Sepsis and Organ Dysfunction at Initial Clinical Presentation. EBioMedicine. 2014;1:64-71.
7. Vincent JL, Moreno R, Takala J, Willatts S, De Mendonça A, Bruining H, et al. The SOFA (Sepsis-related Organ Failure Assessment) score to describe organ dysfunction/failure. On behalf of the Working Group on Sepsis-Related Problems of the European Society of Intensive Care Medicine. Intensive Care Med. 1996;22:707-10.
8. Jones AE, Trzeciak S, Kline JA. The Sequential Organ Failure Assessment score for predicting outcome in patients with severe sepsis and evidence of hypoperfusion at the time of emergency department presentation. Crit Care Med. 2009;37:1649-54.
9. Akavipat P, Thinkhamrop J, Thinkhamrop B, Sriraj W. Acute physiology and chronic health evaluation (APACHE) II score–the clinical predictor in neurosurgical intensive care unit. Acta Clinica Croatica. 2019;58:50.
10. Okamura H, Tsutsi H, Komatsu T, Yutsudo M, Hakura A, Tanimoto T, et al. Cloning of a new cytokine that induces IFN-gamma production by T cells. Nature. 1995;378:88-91.
11. Yasuda K, Nakanishi K, Tsutsui H. Interleukin-18 in Health and Disease. Int J Mol Sci. 2019;20.
12. Baker KJ, Houston A, Brint E. IL-1 Family Members in Cancer; Two Sides to Every Story. Frontiers in Immunology. 2019;10:1197.
13. Eidt MV, Nunes FB, Pedrazza L, Caeran G, Pellegrin G, Melo DA, et al. Biochemical and inflammatory aspects in patients with severe sepsis and septic shock: The predictive role of IL-18 in mortality. Clinica Chimica Acta. 2016;453:100-6.
14. Okuhara Y, Yokoe S, Iwasaku T, Eguchi A, Nishimura K, Li W, et al. Interleukin-18 gene deletion protects against sepsis-induced cardiac dysfunction by inhibiting PP2A activity. International Journal of Cardiology. 2017;243:396-403.
15. Li MF, Li XL, Fan KL, Yu YY, Gong J, Geng SY, et al. Platelet desialylation is a novel mechanism and a therapeutic target in thrombocytopenia during sepsis: an open-label, multicenter, randomized controlled trial. Journal of Hematology & Oncology. 2017;10:104.
16. Gao P, Su Z, Lv X, Zhang J. Interluekin-35 in Asthma and Its Potential as an Effective Therapeutic Agent. Mediators of Inflammation. 2017;2017:5931865.
17. Zhu M, Rong X, Li M, Wang S. IL-18 and IL-35 in the serum of patients with sepsis thrombocytopenia and the clinical significance. Expirmental and Therapeutic Medicine. 2020;19:1251-8.
18. Li X, Li T, Dong G, Wei Y, Xu Z, Yang J. Clinical Value of Serum Interleukin-18 in Neonatal Sepsis Diagnosis and Mortality Prediction. Journal of Inflammation Research. 2022;15:6923-30.
19. Zhixia Z, Zhenwei. X, Yanlong. H, Zhiming. Y, Jianli. X. Clinical value of peripheral blood IL-18/IL-35 combined with procalcitonin in the evaluation of sepsis disease and prognosis. Chinese Emergency Medicine. 2018;38:1065-9.
20. Lei Z. Serum amyloid A, interleukin-35, and soluble myeloid cell trigger receptor-1 in the diagnosis of sepsis. Heilongjiang Medicine. 2021;45:2035-7.
21. Pairattanakorn P, Angkasekwinai N, Sirijatuphat R, Wangchinda W, Tancharoen L, Thamlikitkul V. Diagnostic and Prognostic Utility Compared Among Different Sepsis Scoring Systems in Adult Patients With Sepsis in Thailand: A Prospective Cohort Study. Open Forum Infect Dis. 2021;8:ofaa573.
22. Zhiyong P. Diagnostic effect of plasma IL-35 in patients with sepsis caused by intra-abdominal infection. Medical Laboratory and Clinical. 2020;31: 18-21.
23. Yucang M, Changtai. F, Lijuan. H, . CY, Gaoxiang. C. Changes in interleukin-35 in sepsis and its correlation with disease severity. Anhui Medicine. 2018;22:1905-8.
24. Lin C-Y, Chang C-H, Fan P-C, Tian Y-C, Chang M-Y, Jenq C-C, et al. Serum Interleukin-18 at Commencement of Renal Replacement Therapy Predicts Short-Term

Prognosis in Critically Ill Patients with Acute Kidney Injury. PloS one. 2013;8:e66028.

25. Knaus WA, Draper EA, Wagner DP, Zimmerman JE. APACHE II: a severity of disease classification system. Crit Care Med. 1985;13:818–29. 10.1097/00003246-198510000-00009
26. Moon BH, Park SK, Jang DK, Jang KS, Kim JT, Han YM. Use of APACHE II and SAPS II to predict mortality for hemorrhagic and ischemic stroke patients. J Clin Neurosci. 2015;22:111–5. 10.1016/j.jocn.2014.05.031
27. Zhang L., Qiu C., Yang L., et al. GPR18 expression on PMNs as biomarker for outcome in patient with sepsis. Life Sciences . 2019;217:49–56. doi: 10.1016/j.lfs.2018.11.061.
28. Vincent JL, de Mendonça A, Cantraine F, Moreno R, Takala J, Suter PM, Sprung CL, Colardyn F, Blecher S. Use of the SOFA score to assess the incidence of organ dysfunction/failure in intensive care units: results of a multicenter, prospective study. Working group on "sepsis-related problems" of the European Society of Intensive Care Medicine. Critical Care Medicine 1998, 26 (11): 1793-800
29. Rello, J., Valenzuela-Sánchez, F., Ruiz-Rodriguez, M. & Moyano, S. 2017. Sepsis: A Review of Advances in Management. Adv Ther, 34, 23-36.
30. Rumbus, Z., Matics, R., Hegyi, P., Zsiboras, C., Szabo, I., Illes, A., et al. 2017. Fever Is Associated with Reduced, Hypothermia with Increased Mortality in Septic Patients: A Meta-Analysis of Clinical Trials. PLoS One, 12, 23-35.
31. White, H. D., Vazquez-Sandoval, A., Quiroga, P. F., Song, J., Jones, S. F. & Arroliga, A. C. 2018. Utility of venous blood gases in severe sepsis and septic shock. Proc (Bayl Univ Med Cent), 31, 269-75.
32. Gauer, R., Forbes, D. & Boyer, N. 2020. Sepsis: diagnosis and management. Am Fam Physician, 101, 409-18.

To cite this article: Fatma M. Abdel Salam , Amira M. Abdelrahman , Dalia G. Laban, Tamer E. Eleraky. Diagnostic and Prognostic Value of Serum IL-18 & IL-35 in Patients with Bacterial Infection with or without Sepsis. BMFJ 2025;42(4):455-467.