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Correlation between Serum Interleukin- lß and Neonatal Sepsis in Neonatal Intensive Care Unit in Zagazig University Hospitals

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Abstract

Background: The role of interleukin-lß in the pathogenesis of sepsis is widely accepted, but less is known regarding its role in the newborn period.

Objective: The aim of the work was to detect relation between serum level of IL- lß and clinical presentation of neonatal sepsis.

Patients and Methods: This case control trial study included a total of thirty-six newborns, attending at the Neonatal Intensive Care Unit, Zagazig University Hospitals. They were categorized into two groups; 18 each: Group I: newborns with suspected clinical sepsis, and Group II (control group) healthy newborns with no sepsis. Interleukin- lß was assessed in all neonates.

Results: IL-lß showed significant increase in diseased versus control group (20.07±8.68 versus 1.21±0.48, respectively with p <0.001). IL-lß showed insignificant difference in preterm versus full term neonates. IL-lß showed significant difference in subgroup analysis. Full term neonates in patient group had the highest mean (23.811±10.55 pg/ml, p<0.001).

Conclusion: It could be concluded that premature babies have lower IL-1ß serum concentrations, while mature newborns with sepsis had higher IL-1ß serum concentrations than healthy newborns.

Keywords: Interleukin- lß, Intensive Care Unit, Neonatal Sepsis

INTRODUCTION

Every year, the WHO estimates that 130 million newborns are born around the world. More over 10 million of those infants die before they reach the age of five months, with about eight million dying before their first birthday [(1)]. Four million newborns die each year in the first month of their lives. The bulk of newborn deaths are reported in developing nations like Egypt. In the poor world, around 1.6 million newborns die each year because of infections, and infections are the leading cause of neonatal mortality [(2)].

In resource-limited nations, neonatal sepsis is a leading cause of infant morbidity and mortality. Hospitalized neonates are at danger for developing sepsis if they are not properly cared for in a neonatal intensive care unit (NICU) or a hospital nursery, which is why it's important to keep an eye out for signs of infection. The diagnosis of newborn sepsis in the absence of blood culture data is a matter of clinical judgement. Temperature instability, apneic episodes, food intolerance, poor blood perfusion, and shock are some of the symptoms. Detecting and treating sepsis in environments with limited clinical laboratory capacity, antibiotic medication, and supportive care can be particularly difficult [(3)].

Monocytes and macrophages are the primary producers of interleukin lß, a soluble protein. It is a member of the cytokine family (IL-6 as well as tumor necrosis factor (TNF)) with a wide range of biological characteristics. All three cytokines have the potential to stimulate T and B lymphocytes, enhance cell proliferation, and initiate or suppress gene production of a number of proteins. As a result of infection, microbial toxin, inflammatory agents, lymphocyte activation products, and clotting components, the production of IL-1 has been shown to increase dramatically. Acute phase reactions are linked to the systemic effects of IL-lß. Fever, the production of hepatic acute-phase proteins, and the release of neutrophils are all caused by it, and systemic hypotension occurs when it is produced in excess [(4)].

The role of IL- lß in the etiology of sepsis is widely accepted, but its role in the newborn period is unknown. Research on the ability of systemic infection to create IL-lß has given mixed results. Increased levels of IL- lß have been reported by several researchers [(5)], Others, however, have found that the levels remained the same [(6)] and even in lower levels [(7)] among neonates with sepsis.

Septicemia caused by bacteria is the leading cause of newborn death (mortality in the first 28 days of life) [(8)]. Neonatal infection is still a substantial cause of long-term illness and death despite advances in perinatal care, with an incidence of 1 to 10 per 1000 live births [(9)]. Sepsis is more common in newborns than in any other stage of life, and it varies greatly from place to place. Neonatal sepsis was found in 1–5 cases per 1,000 live births in some wealthy countries, whereas other population-based studies in developing countries revealed rates of 49–170 per 1,000 live births [(10)].

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In Asia, Africa, and Latin America and the Caribbean, the mortality rate for newborns is 34, 42, and 17 per 1000 live births. There is considerable variation in the prevalence of newborn bacterial sepsis between countries and even within countries. As a result, the death rate for newborns in Liberia and South Africa is 68 and 11 per 1000 live births, respectively. Neonatal intensive care unit reported infection rates range from 3.2 to 30 per 100 admissions or discharges, showing the considerable variation between hospitals. Surgery patients may be more likely to be admitted to neonatal critical care units **(11).**

Sepsis is the leading cause of newborn mortality and morbidity in underdeveloped countries, accounting for 1.6 million deaths per year. Sepsis is diagnostic from family history and examination with routine investigations (CBC, CRP, blood culture) for suspected cases. New specific investigation with serum IL-lß for evaluation of neonatal sepsis. As a little study were designed for this association of subject before. *(Write the reference of this paragraph)*

The aim of the current work was to detect relation between serum level of IL- lß and clinical presentation of neonatal sepsis.

PATIENTS AND METHODS

This case control trial study included a total of thirty-six newborns, attending at the Neonatal Intensive Care Unit, Zagazig University Hospitals. This study was conducted between June 2019 to March 2020.

Newborns enrolled in the study were categorized into two groups; 18 each: Group I: newborns with suspected clinical sepsis, and Group II (control group) healthy newborns with no sepsis.

Ethical Consideration:

This study was ethically approved by Zagazig University's research ethics committee. Written informed consent of all the participants' parents was obtained and submitted them to Zagazig University (ZU-IRB#6475). We adhered to the Helsinki Declaration, the ethical guideline of the World Health Organization for human trials.

Inclusion criteria: (1) Preterm and full-term neonates. (2) Documented extremely probable sepsis criteria in accordance with the following: (a) Presence of any of these clinical signs: temperature instability, apnea, tachypnea needing supplemental oxygen, tachycardia, or bradycardia, feeding difficulties, poor perfusion, poor activity, hypotonia, hepatomegaly. (b) Altered blood parameters: leucocytic count with differential count, platelet count. (c) CRP >6 mg/dl. (3) Neonates with maternal risk factors as, maternal intrapartum fever, premature rupture of membrane for more than 18 hours before labor, chorioamnionitis as well as urinary tract infection.

Exclusion criteria: (1) Patient outside age group. (Must be age and sex matched). (2) Patient which receiving antibiotics. (3) Patient whose refused participating the study

This is what all of the participants in this research had to go through:

History: The patient's age, sex, gender, maternal risk factors, gestational age, prenatal, natal history were all recorded in a thorough medical history.

Clinical examination: Weight of a newborn Suckling and Moro reflexes (Moro) were performed on the newborns as well as vital parameters such as heart rate and respiration rate, Spotting the early indications of sepsis: Restlessness, sleepiness, pallor, and mottled skin characterize the infant's condition, and a fluctuation in temperature, either hyperthermia or hypothermia problem with the respiratory system.

Laboratory evaluation: At the onset of sepsis suspicion, blood samples were collected for determination of CRP, serum creatinine and for blood culture.

Quantitative C- reactive protein (CRP): Results that exceeded 6 mg/l were considered positive.

Specific investigation (il1ß assay**):** It was determined by using enzyme immune assay (EIA) technique (R&d system) using quantikine test kits

Statistical analysis

The independent t-test (t) and the Mann-Whitney (MW) tests were employed to compare parametric and non-parametric data respectively on SPSS version 23, in the analysis of the differences between the groups. When there was a difference between two groups of non-parametric data, Proportions were compared using the Chi-square test (x2). Diagnostic and prognostic utility in newborn sepsis were evaluated using Receiver Operating Characteristics (ROC) analysis. Cut-off points and their associated values. P value 0.05 was considered statistically significant (S). It was judged highly significant (HS) when the P value was 0.001 and non-significant (NS) when the P value was >0.05.

RESULTS

Controls and patients had no statistically significant differences in gender distribution. On the other hand; there was a statistically insignificant difference between study groups regarding maturity **(Table 1).**

Table 2 shows the different organisms that showed positive growth in blood cultures.

The following metrics showed statistically significant variations between study groups: white blood cells, lymphocytes, neutrophils, and platelets. Means of white blood cells, and neutrophils were

higher in the patients' group. The means of lymphocytes and platelets were lower in the patients' group on the other hand **(Table 2).**

N/L ratio showed significant difference respectively in diseased and control group (63.2±8.8 versus 83.6±44.6, respectively with p <0.001) **(Table 3).**

ROC curve showed that N/L ratio demonstrated fair diagnostic accuracy in predicting neonatal sepsis [AUC = 0.645 (95% CI 0.605 – 0.877)] with sensitivity of 82% and specificity of 55.1% **(Table 4).**

IL-lß showed significant increase in diseased versus control group (20.07±8.68 versus 1.21±0.48, respectively with p <0.001). IL-lß showed insignificant difference in preterm versus full term neonates **(Table 5).**

IL-lß showed significant difference in subgroup analysis. Full term neonates in patient group had the highest mean (23.811±10.55 pg/ml, p<0.001) **(Table 6).**

Table (1): Qualitative demographic data.

		Controls	**Patients**	**p-value**
Variables	**Attributes**	**n (%)**	**n (%)**	
Gender	Male	9(50)	10 (55.6)	0.631
	Female	9(50)	8 (44.4)	
	Total	18(100)	18 (100)	
Maturity	Pre-term	9(50)	9(50)	-
	Full-term	9(50)	9(50)	
	Total	18(100)	18 (100)	
χ^2 (Chi-squared) test				

Table (2): Blood culture results of study population.

Group		**Patients**
Variables	**Attributes**	**n (%)**
Organism	Klebsiella pneumoniae	8 (44.5)
	MRSA	2 (11.1)
	Coagulase negative Staphylococci	2 (11.1)
	Escherichia coli	2 (11.1)
	Staphylococcus aureus	2 (11.1)
	Streptococcus pneumoniae	2 (11.1)
	Total	18 (100)

Table (3): Means and standard deviations of some hematological parameters of study population.

Group	**Controls**	**Patients**	**Test**	**p-value**
Variables	**Mean ± SD**	**Mean ± SD**		
COMPLETE BLOOD COUNT				
Hemoglobin (g/dl)	14.95 ± 2.22	13.91 ± 2.44	t	0.066
Red blood cells (million/mm³)	4.48 ± 0.68	4.16 ± 0.89	t	0.092
White blood cells (1000/mm³)	9.29 ± 2.91	13.39 ± 3.06	t	**0.001**
Lymphocytes (%)	49.27 ± 4.64	36.23 ± 5.02	M	**0.002**
Neutrophils (%)	42.65 ± 3.05	56.43 ± 5.42	t	**<0.001**
Platelets (1000/mm³)	276.69 ± 7.19	177.46 ± 19.16	t	**<0.001**

Table (4): Relation between N/L ratio and P/L ratio in both groups.

Group	**Controls**	**Patients**	**Test**	**p-value**
Variables	**Mean ± SD**	**Mean ± SD**		
N/L ratio	1.3±0.6	3.2±2.3	M	**<0.001**
P/L ratio	83.6±44.6	63.2±8.8	M	0.06
M = Mann-Whitney U test.				

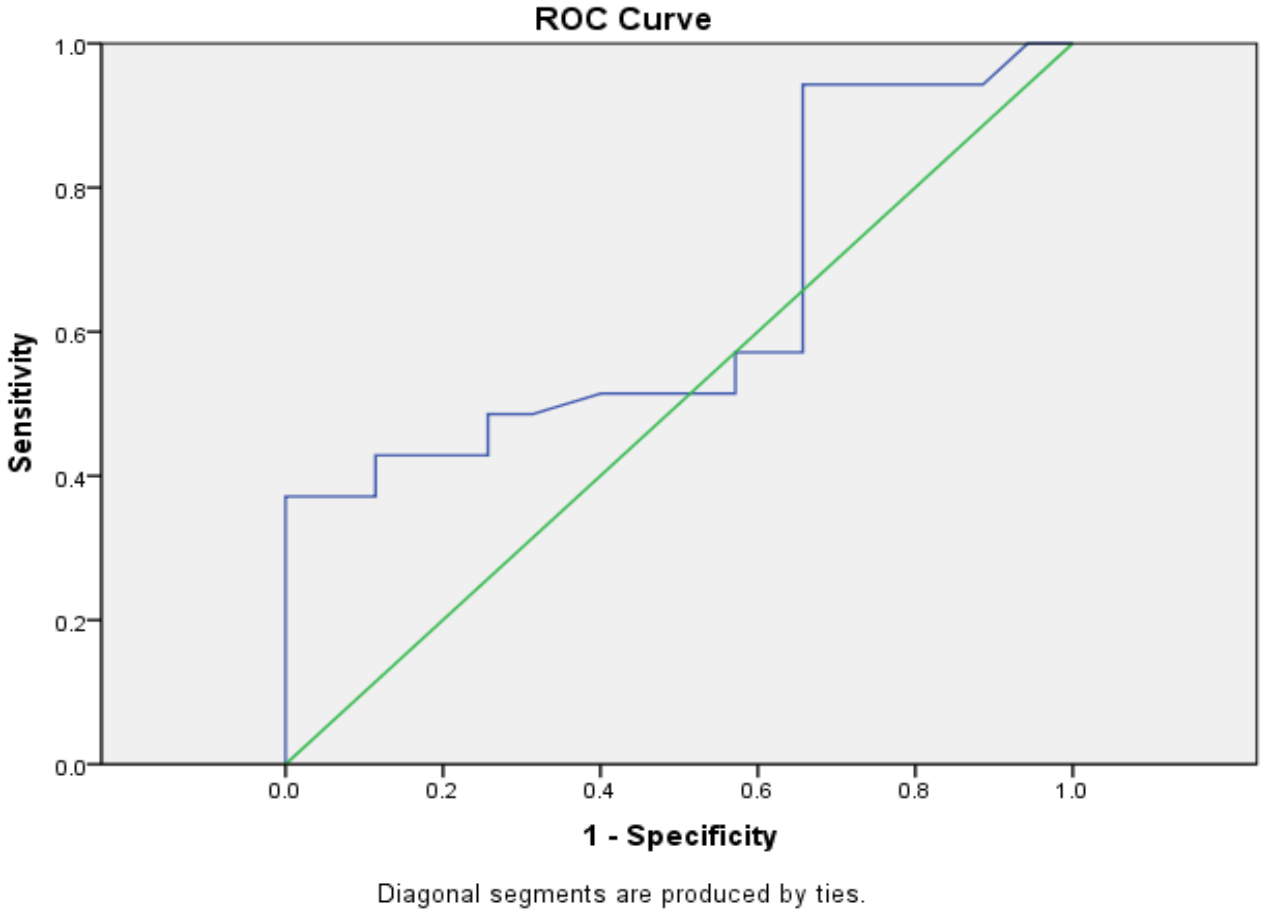

Fig. (2): Receiver-operator characteristics (ROC) curve for N/L ratio in diseased group.

Table (5): Ratio of N/L Area Under Curve.

Area	S.E	P value	95% Confidence Interval		Cut off value	Sensitivity	Specificity
			Lower Bound	Upper Bound			
0.645	0.068	0.045*	0.605	0.877	1.46	82.0%	55.1%

Table (6): Interleukin-1ß in cases-control comparison, and according to maturity in both groups.

Group	Controls	Patients	Test	p-value
Variables	Mean ± SD	Mean ± SD		
IL-lß (pg/ml)	1.21±0.28	20.07±3.68	M	**<0.001**
M = Mann-Whitney U test.				
Group	Preterm	Full term	Test	p-value
Variables	Mean ± SD	Mean ± SD		
IL-lß (pg/ml)	8.61±1.44	12.58±1.64	M	0.164
M = Mann-Whitney U test.				

Table (7): Interleukin-1ß in subgroups.

Group	Patients		Controls		Test	p-value
Variables	Preterm	Full term	Preterm	Full term		
IL-lß (pg/ml)	16.33±3.14	23.811±1.55	0.886±0.18	1.36±0.3	K	**<0.001**
K = Kruskal wallis test.						

DISCUSSION

Gender distribution did not differ statistically between controls and patients according to this study's findings. There were substantial disparities in gestational age, weight, and entry age between the research groups. The control group's mean gestational ages and weights were higher than those of the study participants, and the reverse was true for the participants' admission ages.

A study conducted by **Omar *et al.*** [12] found no differences in mean (SD) age between male and female neonates of 48.25 hours, with 27 (45.0 percent) male neonates and 33 (55.0 percent) female neonates. Also, low birth weight was significantly higher among study group.

Hematological parameters in this study groups showed statistically significant differences regarding means of the following measurements: white blood cells, lymphocytes, neutrophils, and platelets. Means of white blood cells, and neutrophils were higher in the patients' group. The means of lymphocytes and platelets were lower in the patients' group on the other hand.

This is similar to **Maamouri *et al.*** [13] study infants with high clinical suspicion of sepsis had significantly higher white blood cell counts than those in the control group (P=0.000). A substantial difference (P =0.000) existed between neonates with a strong clinical suspicion of sepsis and control groups in terms of neutrophil percentage.

In our study, ROC curve showed that N/L ratio demonstrated fair diagnostic accuracy in predicting neonatal sepsis [AUC = 0.645 (95% CI 0.605 – 0.877)] with sensitivity of 82% and specificity of 55.1%.

In **Martins *et al.*** [14] study, it was estimated according to ROC curve that the neutrophil-lymphocyte ratio was 0.62, that the number of circulating blood bands was 0.98, and that the overall neutrophil count was 0.51. More than a 5.0 neutrophil-to-lymphocyte ratio, a leukocyte count of more than 12,000 mm^3/mL, and a band neutrophil percentage of more than 10% were all associated with an increased risk of sepsis.

In an Egyptian study CBC was done for 319 cases. Abnormalities in the CBC were found in 213 (66.8%) neonates with 22 (6.9%) having leucopenia (WBC < 5,000/mm^3), 71 (22.3%) leukocytosis (WBC > 20,000/mm^3), 74 (23.2%) neutropenia, and 145 (45.5%) thrombocytopenia (platelets < 140,000/mm^3) [15].

In another Egyptian case–control study from April 2015 to October 2016, at the neonatal intensive care unit of Menoufia University Hospital. As a control group, there were 20 healthy outpatient neonates whose clinical and hematological scores indicated that they were free of neonatal sepsis. There was a significant statistical difference in CBC (hemoglobin, hematocrit value, white blood cells, total leukocytic count, immature leukocytes, immature to total leukocytes ratio, immature to mature leukocytes) between healthy and diseased children [16].

In the present study, controls cultures showed no growth. Klebsiella pneumonia was the most prevalent organism among our patient group 8 (44.5), MRSA 2 (11.1), Coagulase negative, Staphylococci 2 (11.1%), Escherichia coli 2(11.1%), Staphylococcus aureus 2 (11.1%) and Streptococcus pneumonia 2 (11.1%).

In Ethiopia, **Eyesus *et al.*** [17] did their study, which included 251 people. They have found 117 (46.6 percent) had signs of growth of bacteria, and 120 different bacteria were isolated. Bacteria of the Gram positive kind were frequently found 81. (67.5 percent). Bacteria of the S. aureus 49 (40.8 percent) and coagulase-negative Staphylococci 26 (21.6 percent) and K. pneumoniae 19 species were most frequently isolated (15.8 percent). Overall, there were 78 isolates that were multidrug resistant (65 percent: 95% CI: 56.7–72.5 percent). Gram-positive bacteria had a multidrug resistance rate of 69%, whereas Gram-negative bacteria had an MDR rate of 22%.

In **Park *et al.*** [18] study, in 18 of the cases, bacteria could be grown. Bacteria found on blood cultures were Enterococcus faecium and Streptococcus (4 cases, 22 percent).

Il-1ß levels differed statistically significantly between groups, with a greater mean value in the sepsis group, according to this research.

The IL-1ß levels in non-surviving septic patients were shown to be lower in comparison to either controls or survivors, according to **Luger *et al.*** [19].

Sullivan *et al.* (4) showed that children with sepsis had somewhat increased (but not statistically significant) levels of IL-1ß. Serum IL-1ß levels were greater in neonates with septic conditions, according to de **Bont and colleagues** [5] and **Ozdemir *et al.*** [6].

In contrast to **Atici *et al.*** [20] study in which Sepsis-affected newborns' IL-I ß levels are much lower (median 0.01 ng/ml) than those of normally developing newborns (median 27.9 ng/ml). This is statistically significant with a p value of 0.001.

A variety of factors, such as discrepancies in the procedures utilized or the timeframes at which samples were collected, could account for the inconsistent results regarding IL-1ß levels in septic newborns.

In our study, IL-lß showed significant difference in subgroup analysis. Full term neonates in patient group had the highest mean (23.811±10.55 pg/ml, p<0.001).

Premature neonates' IL-Iß cellular production has been studied only in a few cases. Our findings

reveal that IL-Iß production is inhibited in both healthy mature and preterm neonates in the presence of severe neonatal infection, IL-lß production by monocytes and macrophages may be reduced, suggesting this. Infection-induced prostaglandin E, on the other hand, may decrease the synthesis of IL-lß. IL-6 may also impede IL-ability Iß's to transcribe proteins [(21)].

In agreement with **Atici *et al.*** [(20)] study that showed that neonatal septicaemia can be diagnosed early using serum IL-1ß levels, which were significantly lower in preterm and term neonates with severe systemic infection.

CONCLUSION

It could be concluded that premature babies have lower IL-1ß serum concentrations, while mature newborns with sepsis had higher IL-1ß serum concentrations than healthy newborns. Studies including repeated measurements of IL-Iß at intervals of a few days in infants who have been treated for septicemia are needed and may be more informative.

The utilization of more longitudinal studies to examine the efficacy of various markers in the early detection of newborn sepsis is encouraged.

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REFERENCES

1. **World Health Organization (2006):** World health report 2006: make every mother and child count. Geneva؛ https://www.who.int/whr/2006/whr06_en.pdf
2. **Vergnano S, Sharland M, Kazembe P *et al.* (2005):** Neonatal sepsis: an international perspective. Arch Dis Child Fetal Neonatal Ed؛, 90(3):220–4.
3. **Polin R, Randis T (2010):** Biomarkers for late-onset neonatal sepsis. Genome Med., 2(9):58-62.
4. **Sullivan J, Kilpatrick L, Costarino A *et al.* (1992):** Correlation of plasma cytokine elevations with mortality rate in children with sepsis. J Pediatr., 120: 510-15.
5. **de Bont E, Martens A, van Raan J *et al.* (1993):** Tumor necrosis factor-alpha, interleukin-I beta, and interleukin-6 plasma levels in neonatal sepsis. Pediatr Res., 33:380-83.
6. **Ozdemir A, Oygiir N, Gultekin M *et al.* (1994):** Neonatal tumor necrosis factor, interleukin-(2, interleukin-l/j and interleukin-6 response to infection. Am J Perinatol., l 1:282-85.
7. **Dinarello C (1991):** The proinflamatory cytokines interleukin-l and tumor necrosis factor and treatment of the septic shock syndrome. J Infect Dis., 163:1177-84.
8. **Movahedian A, Moniri R, Mosayebi Z (2006):** Bacterial culture of neonatal. Iranian Journal of Public Health, 35(4):84-89.
9. **Shim G, Kim S, Kim H *et al.* (2011):** Trends in epidemiology of neonatal sepsis in a tertiary center in Korea: a 26-year. J Korean Med Sci., 26(2):284-89.
10. **Thaver D, Zaidi A (2009):** Burden of neonatal infections in developing countries: a review of evidence from community-based studies. The Pediatric Infectious Disease Journal, 28:1-7.
11. **Moore D (1999):** Nosocomial infections in newborn nurseries and neonatal intensive care clinics; In Hospital epidemiology and infection control, Mayhall C, ed., Lippincott Williams & Wilkins, Philadelphia, PP. 665-694.
12. **Omar J, Isa S, Salwani T *et al.* (2019):** Procalcitonin as an Early Laboratory Marker of Sepsis in Neonates: Variation in Diagnostic Performance and Discrimination Value. The Malaysian Journal of Medical Sciences, 26: 61-69.
13. **Maamouri G, Hassan B, Azghandi M *et al.* (2017).** The Evaluation of Serum Procalcitonin Levels in Neonatal Infections. International Journal of Pediatrics, 5: 5287-5294.
14. **Martins E, Silveira L, Viegas K *et al.* (2019):** Neutrophil-lymphocyte ratio in the early diagnosis of sepsis in an intensive care unit: a case-control study. Revista Brasileira De Terapia Intensiva, 19: 1-5.
15. **El-Din S, Rabie E, El-Sokkary M *et al.* (2015):** Epidemiology of neonatal sepsis and implicated pathogens: a study from Egypt. BioMed Research International, 14: 132-136..
16. **Elgendy F, Khatab A, Badr H *et al.* (2018):** Evaluation of hepcidin as a biomarker for neonatal sepsis. Menoufia Med J., 31: 977-82.
17. **Eyesus T, Moges F, Eshetie S *et al.* (2017):** Bacterial etiologic agents causing neonatal sepsis and associated risk factors in Gondar, Northwest Ethiopia. BMC Pediatr., 17(1):137-42.
18. **Park I, Lee S, Yu S *et al.* (2014):** Serum procalcitonin as a diagnostic marker of neonatal sepsis. Korean Journal of Pediatrics, 57(10): 451-55.
19. **Luger A, Graf H, Schwarz H *et al.* (1986):** Decreased serum interleukin-l activity and iiionocytc IL- I production in patients with Fatal sepsis. Crit Care Med., 14: 458-61.
20. **Atici A, Satar M, Alparslan N (1996):** Serum interleukin-lp in neonatal sepsis. Acta Paediatr., 85: 371-74.
21. **Schindler R, Mancilla J, Endres S *et al.* (1990):** Correlations and interactions in the production of interleukin-6 (IL-6). 1L-l and tumor necrosis factor (TNF) in human blood mononuclear cells: IL-6 suppresses IL-l and TNF. Blood, 76: 40-44.