

Fibroblast Growth Factor 19 as a Predictor for Gastrointestinal -Liver Dysfunction in Neonatal Sepsis

Ola G. Behairy ^a, Abdulhameed A. Abdulhameed ^a, Mahmoud M. Hamed ^a, Dina S. Abdelmotaleb ^b

^a Department of Pediatrics,
Faculty of Medicine Benha
University, Egypt.

^b Department of Clinical and
Chemical Pathology, Faculty of
Medicine Benha University,
Egypt.

Corresponding to: Mahmoud M.
Hamed, Department of Pediatrics,
Faculty of Medicine Benha
University, Egypt.

Email: alaaashafy2017@gmail.com

Received: 29 April 2023

Accepted: 19 August 2023

Abstract

Background: Fibroblast growth factor 19 is a protein which is encoded on FGF19 gene and acts as a hormone, which regulates bile acid synthesis, with effects on glucose and lipid metabolism. **This study aimed to** measure serum level of FGF19 to investigate its potential role as a diagnostic biomarker for gastrointestinal -liver dysfunction in neonatal sepsis. **Methods:** This case-control study was conducted on 100 full term neonates from the Neonatal Intensive Care Unit at Benha University Hospitals. They were divided into 2 equal groups: Sepsis group: included 50 full term neonates diagnosed as sepsis (clinically and laboratory). Control group: included 50 full term neonates matched sepsis group in age and sex. All newborns were subjected to full history taking, complete clinical examination, laboratory investigations and specific laboratory investigations (FGF19 using ELISA). **Results:** FGF19 level was significantly lower in sepsis group compared to control group (231.36 ± 66.7 vs. 349.49 ± 87.81 Pg/ml, P value <0.001). FGF19 level was significantly higher in survived than non-survived patients. There was a significant relationship between FGF 19 level and constipation and hepatomegaly (P value= 0.006 , 0.030 respectively). There was an insignificant correlation between FGF 19 level and Hb, PLT, Urea and day of admission. FGF 19 level can significantly predict the gastrointestinal-liver dysfunction in neonatal sepsis with AUC 0.872, at cut off value \leq

282.8 Pg/ml, with 82 % sensitivity, 80 % specificity, 80.4% PPV and 81.6 % NPV.

Conclusion: we found that serum FGF19 levels are correlated with the occurrence of GI dysfunction and could be used as a potential biomarker for GI dysfunction-associated liver injury.

Keywords: Fibroblast Growth Factor 19; Predictor; Gastrointestinal-Liver Dysfunction; Neonatal Sepsis.

Introduction

Sepsis is a clinical syndrome that complicates severe infection and is characterized by the systemic inflammatory response syndrome (SIRS), immune dys-regulation, microcirculatory derangements, and end-organ dysfunction (1).

The clinical manifestations of neonatal sepsis are non-specific and have varied clinical features. The various manifestations includes decreased acceptance of feed, respiratory distress, pneumonia, apnea, delayed capillary refill time, cold peripheries, mottling, temperature instability including hypothermia and hyperthermia, hypotonia, seizures, bulging fontanels, disseminated intravascular coagulation (2).

Acute gastrointestinal injury (AGI) is common in intensive care unit and associated with worse prognosis in critically ill patients. The Gastrointestinal (GI) plays a crucial role in the pathophysiology of sepsis (3). The signs and symptoms about AGI include abnormal bowel sounds, abdominal distention, diarrhea, constipation, vomiting, inability to tolerate enteral feeding, and GI hemorrhage, prolonged jaundice, necrotizing enterocolitis, cholestasis and rarely acute liver failure, gastrointestinal barrier dysfunction provides an outlet for intestinal flora moving to other locations, leading to the aggravation of sepsis with multiple organ dysfunction syndrome (4).

Fibroblast growth factor 19 (FGF19) is secreted from intestinal epithelial cells. It is involved in the feedback regulation of bile acid synthesis mediating the communication between the small intestine and the liver (5). Also, it functions as a hormone, with effects on glucose and lipid metabolism (6).

Therefore, this study aimed to measure serum level of FGF19 to investigate its potential role as a diagnostic biomarker for gastrointestinal -liver dysfunction in neonatal sepsis.

Patients and methods

This comparative cross-sectional study was conducted on 100 neonates admitted to Neonatal Intensive Care Unit at Benha University Hospitals 6 months since its acceptance from February 2022 to July 2022. The study was approved by the Ethics Committee of Faculty of Medicine, Benha University Hospitals (Approval code: MS 13-12-2021). Informed written consent was obtained from the patients' guardians or parents.

Patients were classified into two groups: **Sepsis group:** 50 newborns diagnosed with sepsis (clinically and laboratory) and **Control group:** 50 newborns as a control group matched sepsis group in age and sex.

Inclusion criteria were: neonates who are full term gestational age, neonates diagnosed as sepsis (clinically and laboratory), positive clinical signs of

sepsis which were defined as two or more of the following clinical signs: respiratory compromise (tachypnea, grunting, intercostal retractions, apnea, and need to ventilation), gastrointestinal compromise (feeding intolerance and abdominal distension), neurological changes (seizure and irritability), cardiovascular compromise (hypotension and cyanosis), general signs (fever and lethargy). Sepsis was defined as Systemic inflammatory response syndrome (SIRS) and evidence of infection (positive microbiological culture and clinical symptoms).

Exclusion criteria were: newborns with multiple congenital anomalies or neonates with hypoxic ischemic encephalopathy.

All neonates were subjected to full history taking and clinical examination with stress on (maternal history for (PROM, UTI, and fever during pregnancy), clinical features of sepsis and GI-liver dysfunction and presence of comorbidities. **Neonatal investigations included** complete blood count, C reactive protein and blood culture, liver function test bilirubin (Total and direct), ALT, AST albumin), kidney function test (blood urea, serum creatinine), coagulation function (activated partial thromboplastin time, APTT; international normalized ratio, INR), Fibroblast growth factor 19 (FGF19) levels was determined using (ELISA)].

Blood sample collections:

Venous blood samples were withdrawn under complete aseptic conditions at the time point of patient's admission to the NICU:

1. One milliliter of whole blood was taken in an EDTA vacutainer and mixed gently; this sample was used to measure the complete blood count (CBC).
2. Two milliliters of blood into 3.2% sodium Citrate vacutainer for coagulation function [Activated Partial Thromboplastin Time (APTT) and International Normalized Ratio (INR)].
3. Three milliliters for blood culture.
4. Five milliliters of blood were taken in plain test tube and left until coagulation. After coagulation, the samples were centrifuged at 1,500 rpm for 15 minutes. The separated serum was used for the assay of:
 - Liver Function test: alanine aminotransferase (ALT), aspartate aminotransferase (AST), albumin, bilirubin (total and direct). Kidney Function Tests: urea and creatinine and C-Reactive Protein

The rest was stored at -20 C until the assay of FGF19. CBC was carried out for all samples using the automated hematology analyzer XS series, SN 12526, SYSMEX Corporation, Kobe,

Japan. CRP was estimated photometrically using Biomed-CRP Latex, Biomed Diagnostics.

Biochemical liver and kidney function tests were assessed by DIALAB, 13771103, Thermo Company, USA.

Coagulation Function (APT and INR): using automated blood coagulation analyzer CS-1600, SN 12058, Sysmex Corporation, Kobe, Japan.

Blood Culture: Using automated microbial detection system (BACT/ALERT 3D60, Ref 248009, SN 304BS4322, bioMérieux, Inc, USA).

Serum FGF19 levels were determined using Enzyme linked immunosorbent assays (ELISA) (MultiScience [LIANKE] Biotech, CO., LTD, Hangzhou, China, Catalog No: DL-FGF19-Hu), ELISA kits 96T.

Statistical analysis

Statistical analysis was done by SPSS v27 (IBM©, Armonk, NY, USA). Shapiro-Wilks test and histograms were used to evaluate the normality of the distribution of data. Quantitative parametric data were presented as mean and standard deviation (SD) and were analyzed by unpaired student t-test. Quantitative non-parametric data were presented as the median and interquartile range (IQR) and were analyzed by Mann Whitney-test. Qualitative variables were presented as frequency and percentage (%) and analyzed using the Chi-square test or Fisher's exact test when

appropriate. Pearson correlation was done to estimate the degree of correlation between two quantitative variables. Evaluation of Diagnostic Performance was performed using diagnostic sensitivity, specificity, PPV and NPV. A two tailed P value < 0.05 was considered statistically significant.

Approval Code:Ms.13.12.2021

Results

Regarding the maternal risk factors, UTI and PROM were significantly different between both groups (P value<0.001) as occurred only in the case group whereas other factors (Gestational HTN, DM, fever and mode of delivery) were insignificantly different between both groups (P-values = 1.39, 0.268, 1,0.078 respectively). Gestational age, sex, height, weight and consanguinity were insignificantly different between both groups (P-values = 0.498, 0.218, 0.590, 0.089, 0.241, respectively).

Mottling, hypotension, temperature instability, respiratory distress grades, Oxygen requirement and heart rate were statistically significant between both case and control groups (P value<0.05). Regarding the neurological manifestations, hypo-activity, poor suckling and weak motor reflex were statistically different between both case and control groups as occurred only in the case group (P value <0.001). In sepsis group, the onset of sepsis ranged from 1-26 days with a mean of 4.12 ± 7.41 days. 42 (84%) patients had early

onset sepsis and 8 (16%) patients had late onset sepsis.

Regarding jaundice, hepatomegaly, feeding intolerance and vomiting were significantly different between both groups (P value <0.001) as these symptoms were presented only in the case group. Ascites, constipation and NEC were insignificantly different between both groups. Diarrhea did not occur in both groups. **Figure 1**

Regarding the results of blood culture in sepsis group, there was no growth in 21 (42%) patients, *Staphylococcus epidermidis* was found in 7 (14%) patients, CONS was found in 6 (12%) patients, Gram +ve bacilli was found in 5 (10%) patients, klebsiella was found in 4 (8%) patients, *Staphylococcus hominis* was found in 2 (4%) patients, Gram +v streptococci was found in 2 (4%) patients, Gram -ve bacilli was found in 1 (2%) patient, Acinetobacter was found in 1 (2%) patient and MRSA was found in 1 (2%) patient.

Regarding the laboratory investigations, TLC, total serum bilirubin, direct serum bilirubin, serum ALT, serum AST, APTT, INR, blood urea, serum creatinine and CRP were significantly higher in case group compared to control group (P value <0.05). PLT and serum albumin level, were significantly lower in case group compared to control group (P value <0.001).

Regarding the final diagnosis of patients in sepsis group, congenital pneumonia

occurred in 35 (70%) patients, community acquired pneumonia occurred in 6 (12%) patients, CNS infection occurred in 1 (2%) patient, septicemia occurred in 8 (16%) patients.

Regarding the Griffin neonatal sepsis score, lethargy or hypotonia occurred in 49 (98%) patients, Temperature instability occurred in 6 (12%) patients, serum glucose (>180) in 3 (6%) patients, WBC count (leukocytosis or leukopenia) in 12 (24%) patients, feeding intolerance occurred in 18 (36%) patients, respiratory affection occurred in 25 (50%) patients. The Griffin neonatal sepsis score ranged from 1-6 with a mean of 3.18±1.06].

FGF19 level was significantly lower in case group compared to control group (231.36 ± 66.7 vs. 349.49 ± 87.81 pg/ml, P value <0.001). **Figure 2.**

In case group, there was a significant negative correlation between FGF 19 level and Griffin neonatal sepsis score, onset of sepsis, Day of discharge, Duration of admission, and outcome, total and direct bilirubin, ALT, AST, APTT, INR, creatinine , TLC and CRP. There was a significant positive correlation between FGF 19 level and Albumin (r=0.519, p<0.001). There was an insignificant correlation between FGF 19 level and Hb, PLT count, blood urea level and day of admission. **Table 1.**

FGF 19 level can significantly predict the Gastrointestinal-Liver Dysfunction in Neonatal Sepsis with AUC 0.872 and P

value <0.001, at cut off value \leq 282.8 Pg/ml, with 82 % sensitivity, 80 %

specificity, 80.4% PPV and 81.6 % NPV. **Table 2, Figure 3.**

Table 1: Correlation between FGF 19 level and different parameters in sepsis group (N=50)

		FGF19 level		
		r	P	
Griffin neonatal sepsis score		-0.557	< 0.001*	
Onset of sepsis		-0.249	0.012*	
Day of admission		-0.155	0.123	
Day of discharge		-0.345	< 0.001*	
Duration of admission		-0.452	< 0.001*	
Outcome		-0.281	0.005*	
Total Bilirubin (mg/dl)		-0.436	< 0.001*	
Direct bilirubin (mg/dl)		-0.413	< 0.001*	
Albumin (mg/dl)		0.519	< 0.001*	
ALT (U/L)		-0.456	< 0.001*	
AST (U/L)		-0.458	< 0.001*	
APTT (s)		-0.427	< 0.001*	
INR		-0.438	< 0.001*	
Urea (mg/dl)		-0.024	0.813	
Creatinine (mg/dl)		-0.215	0.032*	
Hb (g/dl)		0.020	0.840	
TLC (*10³ cells/μL)		-0.407	< 0.001*	
PLT (*10³ cells/μL)		0.080	0.431	
CRP (mg/L)		-0.332	0.001*	
GIT & Liver dysfunction	Ascites	NAD	223.9 \pm 69.9	0.058
		Mild	324.0 \pm 35.5	
		Mild to moderate	127.8 \pm 0.0	
	Jaundice		221.98 \pm 74.3	0.598
	Vomiting		239.1 \pm 83.1	0.154
	Constipation		152.5 \pm 65.1	0.006*
	Feeding intolerance		231.8 \pm 81.5	0.474
	Hepatomegaly		210.9 \pm 67.2	0.030*

Hb: hemoglobin, PLT: platelet count, TLC: total leukocyte count, ALT: Alanine transaminase, AST: aspartate aminotransferase, APTT: Activated Partial Thromboplastin Clotting Time, INR: international normalized ratio, CRP: C - reactive protein, FGF19: Fibroblast growth factor 19, r: coefficient correlation, *: statistically significant as P value <0.05.

Table 2: Diagnostic performance of FGF 19 level ($\mu\text{g/ml}$) for prediction of Gastrointestinal-Liver Dysfunction in Neonatal Sepsis

	Cut off	Sensitivity %	Specificity %	PPV	NPV	AUC	P value
FGF 19 level ($\mu\text{g/mL}$)	≤ 282.8	82	80	80.4	81.6	0.872	<0.001*

FGF19: Fibroblast growth factor 19, PPV: positive predictive value, NPV: negative predictive value, AUC: area under the curve, CI: confidence interval, *: statically significant as P value <0.05

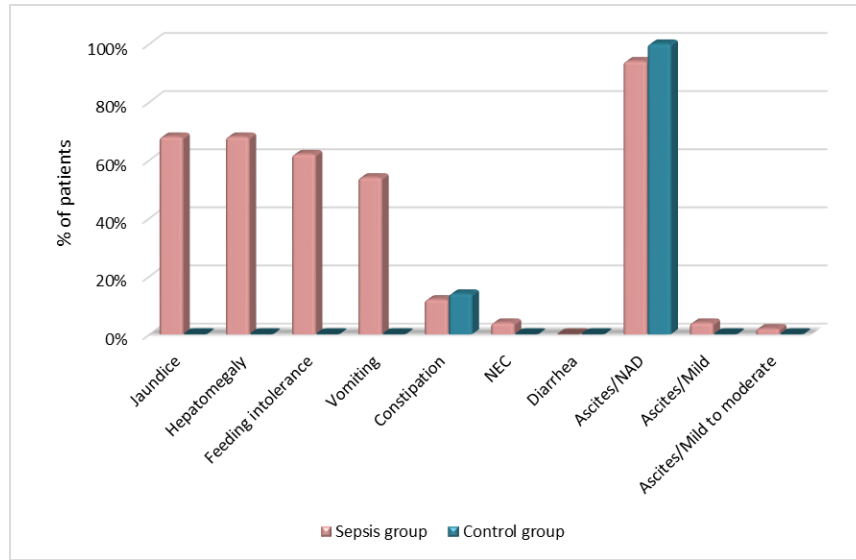


Figure 1: Gastrointestinal and Liver manifestations of the studied groups

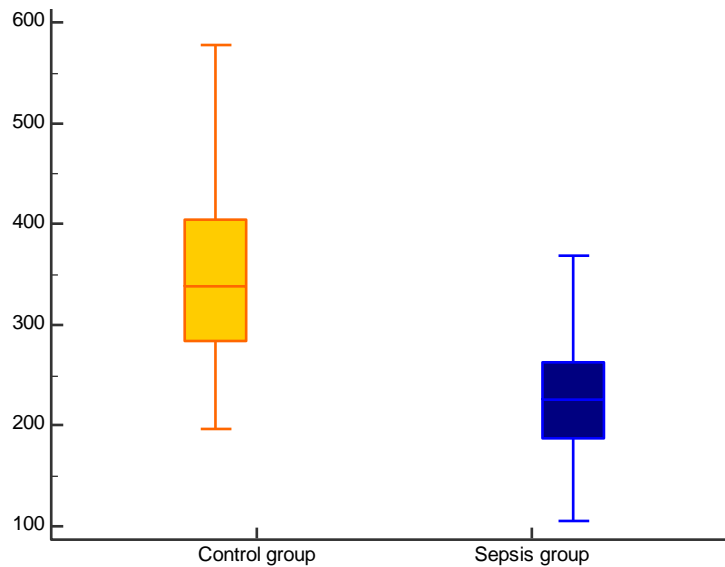


Figure 2: FGF19 ($\mu\text{g/mL}$) level of the studied groups

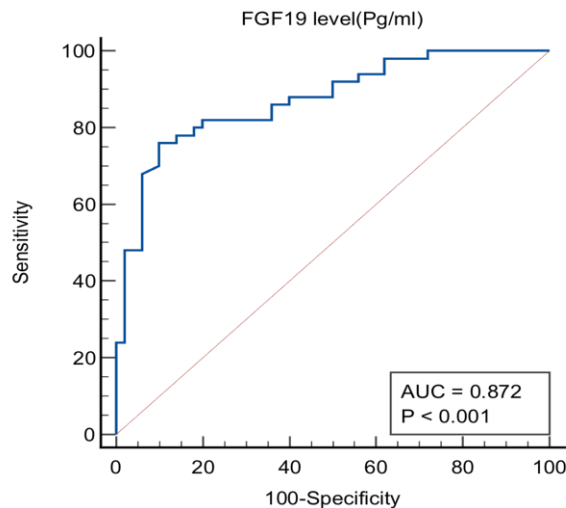


Figure 3: ROC curve analysis of FGF 19 level (FGF 19 level ($\mu\text{g/ml}$)) for prediction of Gastrointestinal-Liver Dysfunction in Neonatal Sepsis

Discussion

Acute gastrointestinal injury (AGI) is common in intensive care unit and associated with worse prognosis in critically ill patients. The Gastrointestinal injury (GI) plays a crucial role in the pathophysiology of sepsis. FGF19 secrets from intestinal epithelial cells and involves in the feedback regulation of bile acid synthesis mediating the communication between the small intestine and the liver (7).

Limited studies indicated that FGF19 levels are associated with ileal resection, diarrhea and disease activity (8).

This study aimed to measure serum level of Fibroblast growth factor 19 (FGF19) to investigate its potential role as a diagnostic biomarker for Gastrointestinal-liver dysfunction in neonatal sepsis.

Regarding clinical presentation, respiratory distress grades and oxygen requirement: mottling, hypotension,

temperature instability and heart rate were significantly increased in sepsis group ($P\text{-value} < 0.05$).

There have been several studies investigating the clinical presentation of sepsis in neonates, which align with the results of our study. A study found that neonates with sepsis had higher rates of respiratory distress, apnea, hypotension, and tachycardia compared to controls (9).

Similarly, another study reported that hypotension, tachypnea, and abnormal peripheral perfusion were more common in neonates with sepsis (10).

Regarding jaundice, hepatomegaly, feeding intolerance and vomiting were significantly different between both groups ($P\text{ value} < 0.001$) as these symptoms were presented only in the case group. Ascites, constipation and NEC were insignificantly different between both groups. Diarrhea did not occur in both groups.

Regarding the laboratory investigations, TLC, total bilirubin, direct bilirubin, ALT, AST, APTT, INR, urea, creatinine and CRP were significantly higher in case group compared to control group (P-value <0.05). PLT count and albumin were significantly lower in sepsis group compared to control group (P-value <0.001).

In agreement with our study, Tang et al. reported higher PLT count and albumin in GI dysfunction group when compared to non GI dysfunction group (11).

FGF19 is expressed at birth in preterm infants and decreases over time, even as enteral feeds increase. These findings differ from those in adults, where FGF19 is physiologically induced in enterocytes by exposure to BAs after eating, providing negative feedback to decrease further hepatic bile acid synthesis (12).

The enterocyte damage occurring in early stage of sepsis affects the secretion of enterocyte-derived factors. These bio-factors may help to identify GI dysfunction in the early stage(13).

FGF19, is an enterocyte-derived factor, is significantly correlated with GI dysfunction in patients with sepsis, which gave a new insight into assessment of GI dysfunction. The feasibility of serum FGF19 as a common indicator of GI dysfunction still needs to be confirmed in large size samples. (14).

In our study, FGF19 level was significantly different in sepsis group as it was significantly higher in survived than non-survived patients (P-value = 0.003). FGF19 level was significantly lower in sepsis group compared to control group (226.03 ± 72.18vs. 349.49±87.81, P value <0.001). Parallel

with our findings, a statically significance difference was found between survivors and non-survivors patients (15).

Regarding FGF19 level, in line with our study, a study found that serum FGF19 levels were significantly decreased in patients with sepsis-associated gastrointestinal dysfunction compared with patients without gastrointestinal dysfunction [48.4 (27.7, 95.6) µg/mL vs 77.6 (45.8, 151.2) µg/mL, P=0.046] (11)..

In our study, there was a significant negative correlation between FGF 19 level and Griffin neonatal sepsis score, onset of sepsis, day of discharge, duration of admission, outcome, total and direct bilirubin, ALT, AST, APTT, INR, blood urea, serum creatinine, TLC count and CRP. There was a significant positive correlation between FGF 19 serum level and albumin (r=0.519, p<0.001). Also, there was a significant relationship between FGF 19 levels and constipation and hepatomegaly.

In a study done it was reported that a significant negative correlation between FGF 19 level and onset of sepsis, Day of discharge, Duration of admission, bilirubin, ALT, AST, creatinine and CRP. Also, a significant positive correlation between FGF 19 level and albumin (r=0.056, P = 0.024) (15).

Our results are compatible with Tang et al. who found that FGF19 can significantly predict gastrointestinal dysfunction in pediatric patients with sepsis was 0.636 (95%CI 0.515–0.757). Also they found by using multivariate logistic regression analysis indicated that low FGF19 level was independently risk factor for the occurrence of GI

dysfunction in pediatric patients with sepsis (OR: 0.992, 95% confidence interval [CI]: 0.984–0.999, P = 0.046) (11). Also, a study documented that FGF19 serum levels displayed the diagnostic accuracy for GI dysfunction with a sensitivity of 62.5% and a specificity of 55.2 % with the cutoff value of 60 µg/mL (16).

FGF19 was proved to be correlated with the development and progression of alcoholic steatohepatitis. Interestingly, a study found that the lower serum FGF19 level was correlated with GI dysfunction complicated by ALI, and lower FGF19 than 52 µg/ml suggested a higher risk of occurrence of ALI. The difference was not statistical significance possibly due to a small sample size (11).

Our results merely revealed that FGF 19 level can significantly predicts the gastrointestinal-Liver dysfunction in neonatal sepsis with AUC 0.872 and P-value <0.001, at cut off value ≤ 282.8 µg/mL, with 82 % sensitivity, 80 % specificity, 80.4% PPV and 81.6 % NPV.

Conclusion

In the present study, we found that serum FGF19 levels are correlated with the occurrence of GI dysfunction, which also is a potential biomarker for GI dysfunction-associated liver injury. Therefore, serum FGF19 could be considered as a novel predictor for GI dysfunction or GI-liver dysfunction in neonatal sepsis.

References

1. Xu JL, Yu XL, Yao XQ, Xu LH, Jing M. Diagnostic Value and Prognostic Evaluation of Autophagy-Related Protein Expression Level in Sepsis Complicated with Acute

- Respiratory Distress Syndrome. *Dis Markers*. 2022;2022:8920926.
2. Sharma D, Sharma P, Soni P, Gupta B. *Ralstonia picketti* neonatal sepsis: a case report. *BMC Res Notes*. 2017;10:28-35.
3. Yu Y, Yang Y, Bian Y, Li Y, Liu L, Zhang H, et al. Hydrogen Gas Protects Against Intestinal Injury in Wild Type But Not NRF2 Knockout Mice With Severe Sepsis by Regulating HO-1 and HMGB1 Release. *Shock*. 2017;48:364-70.
4. Klingensmith NJ, Coopersmith CM. The Gut as the Motor of Multiple Organ Dysfunction in Critical Illness. *Crit Care Clin*. 2016;32:203-12.
5. Tang X, Ren Y, Wang C, Cui Y, Xiong X, Miao H, et al. Serum fibroblast growth factor 19 as a predictor for early stage of gastrointestinal-liver dysfunction in pediatric patients with sepsis. 2019.
6. Gadaleta RM, Moschetta A. Metabolic Messengers: fibroblast growth factor 15/19. *Nat Metab*. 2019;1:588-94.
7. Johansson H, Mörk LM, Li M, Sandblom AL, Björkhem I, Höijer J, et al. Circulating Fibroblast Growth Factor 19 in Portal and Systemic Blood. *J Clin Exp Hepatol*. 2018;8:162-8.
8. Nolan JD, Johnston IM, Pattni SS, Dew T, Orchard TR, Walters JR. Diarrhea in Crohn's disease: investigating the role of the ileal hormone fibroblast growth factor 19. *J Crohns Colitis*. 2015;9:125-31.
9. Wynn JL, Wong HR. Pathophysiology and treatment of septic shock in neonates. *Clin Perinatol*. 2010;37:439-79.
10. Lara B, Enberg L, Ortega M, Leon P, Kripper C, Aguilera P, et al. Capillary refill time during fluid resuscitation in patients with sepsis-related hyperlactatemia at the emergency department is related to mortality. *PLoS One*. 2017;12:e0188548.
11. Tang X, Ren Y, Wang C, Cui Y, Xiong X, Miao H, et al. Clinical value of fibroblast growth factor 19 in predicting gastrointestinal dysfunction in children with sepsis. *J Shanghai Jiaotong Uni*. 2020;40:1236-42.
12. Memon N, Griffin IJ, Lee CW, Herdt A, Weinberger BI, Hegyi T, et al. Developmental regulation of the gut-liver (FGF19-CYP7A1) axis in neonates. *J Matern Fetal Neonatal Med*. 2020;33:987-92.
13. Kokesova A, Coufal S, Frybova B, Kverka M, Rygl M. The intestinal fatty acid-binding

- protein as a marker for intestinal damage in gastroschisis. PLoS One. 2019;14:e0210797.
14. Gadaleta RM, Garcia-Irigoyen O, Cariello M, Scialpi N, Peres C, Vetrano S, et al. Fibroblast Growth Factor 19 modulates intestinal microbiota and inflammation in presence of Farnesoid X Receptor. EBioMedicine. 2020;54:102719.
 15. Chunxia X, Ren Y, Wang C, Cui Y, Xiong X, Miao H, et al. Serum fibroblast growth factor 19 as a predictor for early stage of gastrointestinal-liver dysfunction in pediatric patients with sepsis. 2019.
 16. Tang X, Ren Y, Wang C, Cui Y, Xiong X, Miao H, et al. Serum fibroblast growth factor 19 as a predictor for early stage of gastrointestinal-liver dysfunction in pediatric patients with sepsis. Research Square; 2019.

To cite this article: Ola G. Behairy, Abdulhameed A. Abdulhameed, Mahmoud M. Hamed, Dina S. Abdelmotaleb. Fibroblast Growth Factor 19 as a Predictor for Gastrointestinal - Liver Dysfunction in Neonatal Sepsis. BMFJ 2023;40(2):474-484.