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Original article

Zinc Protoporphyrin to Heme Ratio versus Ferritin as a Measure of Iron Sufficiency in Preterm Infants

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Abstract:

Background: Preterm babies with anemia (also known as anemia of prematurity) experience a bigger and quicker hemoglobin decline. As a result, blood transfusions and the administration of human synthetic erythropoietin may be required. The purpose of this research was to assess ferritin and zinc protoporphyrin to heme ratio (ZnPP/H) as biomarkers of iron status in preterm neonates and to ascertain the influence of particular clinical events on these parameters. **Methods:** This prospective cross-sectional research included 170 neonates with gestational ages < 34 weeks. They were chosen from the Neonatal Intensive Care Unit at Benha University Hospital. **Results:** Eighty-one of them (49.1%) had anemia. Patients with sepsis had higher CRP levels and lower platelet counts than individuals without the condition. Ferritin levels were noticeably lower in the anemic group than in the non-anemic group. After 4 weeks, there was a substantial rise in ZPP/H ratio. Also, ZPP/H ratio fell after blood infusion compared to baseline. **Conclusion:** Since sepsis and transfusion have greater impact on ferritin than on ZnPP/H, ZnPP/H may be a more accurate indicator of iron state in neonates than ferritin. In terms of the effects of clinical events, serum ferritin levels were substantially reduced in the anemic group compared to the non-anemic group, whereas ZPP and ZPP/H ratio levels were markedly higher. The septic group had noticeably greater serum ferritin values.

Keywords: Iron Deficiency in Preterm Babies, Zinc Protoporphyrin to Heme Ratio, Ferritin.

Introduction

The most prevalent micronutrient deficit in infants around the globe is iron deficiency (ID). Recent research indicates that ID without anaemia has comparable, possibly permanent effects on neurodevelopment as does ID with anaemia. The American Academy of Pediatrics (AAP) screened children for ID anaemia at around one year of age and for ID without anaemia in high-risk children, including those from poor income backgrounds. Dietary variables and prenatal ID have been linked to low-income children's increased risk for ID, though the exact causes are unknown (1).

In the lack of postnatal supplementation or materno-fetal iron transfer during the third trimester, preterm babies are innately iron deficient. Iron deficiency in full-term babies can also be caused by perinatal risk factors like intrauterine growth delay and maternal diabetes mellitus (2).

The American Academy of Pediatrics advises high-risk infants to take iron supplement of 2-4 mg/kg/day due to their vulnerability to iron shortage. These recommendations might not be sufficient to sustain iron sufficiency in NICU patients based on earlier research conducted at the University of Washington (UW) newborn intensive care unit (NICU). In order to supplement newborns effectively, the ability to judge iron adequacy is essential. Ferritin and zinc protoporphyrin to heme ratio (ZnPP/H) are two commonly used indicators of iron adequacy in newborns. Plasma ferritin has been used as a secondary indicator of iron adequacy because ferritin serves as an iron carrier in plasma as well as binding and storing iron intracellularly (3).

The optimal measurement during these periods may not be ferritin, as it is an acute phase reactant that increases with stress or inflammation. In red blood cells, where iron is preferred for erythropoiesis, ZnPP/H ratio assesses zinc in connection to iron absorption. When there is not enough iron available, more zinc is added, increasing ZnPP/H ratio. Although it has been shown that persistent inflammation in adult populations raises ZnPP/H levels, it is currently unclear how inflammation in neonates impacts ZnPP/H (4).

This study aimed to evaluate zinc protoporphyrin to heme ratio (ZnPP/H) and ferritin as biomarkers of iron status in preterm infants, and to determine how specific clinical events can affect these measures.

Patients and Methods:

This study was a cross sectional study which was conducted on preterm infants with gestational age of ≤ 34 weeks who were selected from the Neonatal Intensive Care Unit in Benha University Hospital during the period between January 2021and August 2022.

Type of the study: Cross sectional study

Patients:

This study included 170 Neonates born at ≤ 34 weeks gestation who were admitted to the Neonatal Intensive Care Unit in Benha University Hospitals.

Inclusion criteria:

- Neonates of both sexes.
- Neonates born at ≤ 34 weeks gestation.

Exclusion criteria:

- Neonates with congenital malformations.
- Lack of parental consent.

Ethical consideration:

- Approval of the study protocol by the Ethical Scientific Committee of Benha University was obtained in the study {M.S:14.12.2020}.

- Informed verbal and written consents were obtained from the parents before enrollment in the study.

Methods:

The patients were subjected to the following:

- History taking including prenatal, neonatal, postnatal and family history.
- History of recent red blood cell transfusions.
- Clinical Examination.
- Investigation:
  - CBC with reticulocytic count and immature/total neutrophil count, CRP, and blood culture to assess for infection (sepsis will be confirmed by a positive blood culture 2 days before or 1 day after ZnPP/H and ferritin readings, whichever comes first).
  - Plain X-ray abdomen was performed when necessary to diagnose NEC (1 day or 2 days after ZnPP/H and ferritin readings).
  - At the moment of laboratory collection, paired ZnPP/H and ferritin readings as well as the infant's age and corrected gestational age were gathered.
  - Serum ferritin level

Principle

An automated instrument-based enzyme-linked fluorescent ELISA called the VIDAS Ferritin (FER) test is available (ELFA). The apparatus controls the assay's stages and temperature. For the test, a solid phase and a throwaway pipette-like implement called the Solid Phase Receptacle (SPR®) are used. Mouse polyclonal anti-ferritin antibodies are formed on the SPR during production. The VIDAS Ferritin assay's architecture prevents SPR random reactions. The chemicals for the test are located in the sealed Reagent Tubes. The substance is inserted into the well containing the antiferritin antibody attached to alkaline phosphatase. As the sample/conjugate mixture cycles in and out of the SPR, the ferritin will bind to antibodies coated on the SPR and to the conjugate, forming a "sandwich." Reverse is eliminated through cleaning. The 4-methylumbelliferyl phosphate, a luminescent material, is cycled through by the SPR. Any remaining enzyme on the SPR wall will catalyse the conversion of the substrate into the luminescent product, 4-methylumbelliferone. The optical scanner in the device detects fluorescence intensity, which is related to the ferritin concentration discovered in the sample. After the VIDAS Ferritin assay is complete, the instrument immediately analyses the results, and a summary is generated for each sample.

ELISA kit for human zinc protoporphyrin

Test Concept

This substance is used for an enzyme-linked immunosorbent test (ELISA). The apparatus has been pre-coated with human ZPP antibody. ZPP from the substance binds to antibodies that have been coated on the wells when it is inserted. After that, a biotinylated human ZPP antibody is applied, and it binds to the material's ZPP. Streptavidin-HRP is then added, which binds the biotinylated ZPP antibody. After fermentation, unbound streptavidin-HRP is eliminated through washing. When the substrate solution is added, the colour adjusts based on the Human ZPP content. The process is halted by introducing an acidic stop solution, and absorption is then measured at 450 nm.

Statistical Analysis

The statistical software for social sciences (SPSS 24.0, IBM/SPSS Inc., Chicago, IL)

was used to statistically evaluate the results. Two different kinds of statistical research were carried out:

Descriptive data analysis

It included figures for averaging the continuous data as mean and standard deviation (SD) for data that were regularly distributed or median and range for data that were skewed. The presentation of qualitative statistics used frequency with a number (%).

Analytical or inferential statistics.

- The Pearson Chi-square (2) test was used to evaluate one qualitative measure between two or more groups.
- Student test was used to determine whether there was a substantial variation between two groups with normally distributed data. With the help of the Shapiro-Wilk test and Levine's test, the assumptions of normalcy in each group and uniformity of variances were correspondingly confirmed.
- The non-parametric counterpart of the Student test is the Mann-Whitney U test.
- Analysis of variance (ANOVA or F test) was used to determine whether there was a meaningful variation between more than two normally distributed groups in the continuous data. With the help of the Shapiro-Wilk test and Levine's test, the assumptions of normalcy in each group and uniformity of variances were correspondingly confirmed.
- When ANOVA conditions were broken and there were more than two sets of skewed data, the Kruskal-Wallis test was used to analyze the data.
- In data with a normal distribution, a paired test was used to assess the means of two groups at the time of admittance and four weeks later.
- When data had an abnormal distribution, the Wilcoxon signed-rank test was used to assess the means of two groups at admittance and after four weeks.

(Importance level) P-value

The significance threshold at which the null-premise (the hypothesis of no difference) was denied was fixed at 0.05 for all applied tests, meaning that P-values below 0.05 are statistically non-significant, while P-values above 0.05 are significant.

Results:

The studied group consists of 88 males (51.8%) and 82 females (48.2%), their mean gestational age was 32.5±1.9 weeks and their mean age on admission was 1.3±0.65 days. The weight of 16.5% of was < 1500 kg, and 4.7% of the patients were small for gestational age Table 1.

Regarding the maternal history, 120 neonates (70.6%) had history of maternal prenatal steroid intake, 2 neonates (1.2%) had history of maternal fever, 10 neonates (5.9%) were infants of diabetic mothers, 11 neonates (6.5%) had history of maternal preeclampsia, 4 neonates (6.5%) had mothers with hypertension, 3 (1.8%) systemic lupus erythematosus (SLE), 12 (7.1%) urinary tract infection, and 2 neonates (1.2%) had a maternal history of COVID-19. Premature rupture of membranes occurred in 75 neonates (44.1%). One hundred thirty-one neonates (77.1%) were delivered by Cesarean section, while 39 neonates (22.9%) were delivered vaginally.

On resuscitation, 27 neonates had respiratory distress (RD) grade 2. They needed suction, warming and nasal O_2. Fifteen neonates had RD grade 3 and needed suction, warming and nasal O_2. Forty-nine neonates had RD grade 3 and needed suction, warming and neonatal

CPAP. Twenty-five neonates had RD grade 4 and were intubated in the delivery room and put on mechanical ventilation (MV) in the NICU Table 2.

The average systolic blood pressure in the study group was 80 mmHg, the average diastolic blood pressure was 42 mmHg, the average heart rate was 123 bpm, the average body temperature was 36.9 C^{o}, and the average respiratory rate was 60.3 bpm. Ninety percent of the newborns had capillary renewal times under three seconds, and 76.8% of them were pink. The examined neonates' average weight was 1929426 grammes, and their average length was 48.81.2 cm. Table 3.

CNS examination revealed that 21 neonates (12.4%) had good reflexes, consciousness and anterior fontanel (AF) size at level with no abnormal fits. Ninety neonates (53%) had moderate reflexes, consciousness, AF at level with no abnormal fits. Thirty-nine neonates (22.9%) had poor reflexes, AF size at Level and were sedated on MV and 20 neonates (11.7%) had poor reflexes, consciousness and AF at Level with no abnormal fits. On chest examination; 138 neonates (81.2%) had diminished air entry bilaterally, chest in-drawing and grunting. On cardiac examination, 33 neonates (19.4%) had murmurs. On abdominal examination, all the enrolled neonates had no organomegaly Table 4.

Table 1: Demographic data of the studied group.

Demographic data		**Studied group**	
		N=170	**%**
Sex	Males	88	51.8%
	Females	82	48.2%
Postnatal age (days)	Mean ±SD	1.3±0.65	
	Range	1-3	
Gestational age (weeks)	Mean ±SD	32.5±1.9	
	Range	28-34	
Body weight (grams)	<1500	28	16.5%
	>1500	142	83.5%
Small for gestational age	Yes	8	4.7%
	No	162	95.3%

Table 2: Perinatal history of the studied group.

Perinatal history		**Studied group**	
		N=170	**%**
Prenatal steroids	Yes	120	70.6%
	No	50	29.4%
Maternal fever	No	168	98.8%
	Yes	2	1.2%
Infant of diabetic mother	Yes	10	5.9%
	No	160	94.1%
Maternal Diseases	No	131	77.1%
	Preeclampsia	11	6.5%
	HTN	4	2.4%
	SLE	3	1.8%
	UTI	12	7.1%
	COVID-19	2	1.2%
PROM	No	95	55.9%
	Yes	75	44.1%
Mode of delivery	Cesarean section	131	77.1%
	Vaginal delivery	39	22.9%
Resuscitation on delivery room	Not Attend	54	31.8%
	RD grade 2, suction, warming, put on Nasal O_2	27	15.9%
	RD grade 3, suction, warming, put on Nasal O_2	15	8.8%
	RD grade 3, suction, warming, put on N. CPAP	49	28.8%
	RD grade 4, intubated in delivery room and put on MV	25	14.7%

Table 3: Vital signs and anthropometric measurements in the studied group.

Vital signs and anthropometric measurements		**Studied group**	
		N=170	**%**
Systolic blood pressure (mmHg)	Mean ±SD	80±7	
	Range	66-95	
Diastolic blood pressure (mmHg)	Mean ±SD	42.1±6.9	
	Range	24-58	
HR (beat/min.)	Mean ±SD	123±7.2	
	Range	110-144	
Temperature (°C)	Mean ±SD	36.9±0.3	
	Range	36.2-37.8	
Respiratory rate/min	Mean ±SD	60.3±5.5	
	Range	41-77	
Capillary refill time	< 3 sec	154	90.6%
	> 3 sec	16	9.4%
Body weight (gram)	Mean ±SD	1929±426	
	Range	880-2900	
Length (cm)	Mean ±SD	48.8±1.2	
	Range	45-50	

Table 4: Clinical examination in the studied group

Clinical data		Studied group N=170	%
CNS examination	Good Reflexes, Conscious, AF at level, No abnormal fits	21	12.4%
	Moderate Reflexes, Conscious, AF at Level, No abnormal fits	90	53%
	Poor Reflexes, Sedated on MV, AF at Level	39	22.9%
	Poor Reflexes, Conscious, AF at Level, No abnormal fits	20	11.7%
Chest examination	Fair air entry bilateral, no adventitious sounds	32	18.8%
	Diminished air entry bilateral, retraction, grunting	138	81.2%
Heart examination	S1+S2+0	134	78.8%
	S1+S2+Murmur	33	19.4%
	S1+S2 (Bounding Heart Sounds)	3	1.8%
Abdominal examination	Organomegaly	0	0.0%
	No organomegaly	170	100.0%

Laboratory investigations in the studied group reveled that the mean hemoglobin concentration was 15.8±2 g/dl, the mean platelet count was 228±98 103/l, the mean leucocytic count was 12.02±4.9/L and the mean reticulocyte count was 3.7±1.9 %. Table 5

Serum ferritin level showed significant elevation in infants with history of maternal fever, and those who needed resuscitation for grade IV respiratory distress. Howevwr, there was no significant difference in ferritin level as regards to sex, history of PROM, mode of delivery, oxygen support on admission, or cause of admission. Table 6

Zinc protoporphyrin to heme (ZPP/H) ratio showed significant decrement in the patients with history of maternal fever, and those who needed resuscitation for grades II and IV RD, those who needed oxygen support on admission and those admitted for neonatal problems other than jaundice. However, there was no significant difference in ZPP level as regards to sex, history of PROM, or mode of delivery. Table 7

ROC analysis was done to assess the performance of serum levels of ferritin and ZPP as well as ZPP/H ratio to predict anemia in preterm infants. Regarding ferritin; AUC was 0.582 (95% confidence interval: 0.0.495-0.669), p=0.065. Regarding ZPP; AUC was 0.876 (95% confidence interval: 0.823-0.928), p<0.001. At a cutoff point > 13.2 µg/dl, the sensitivity was 79% and specificity was 79.8%. Regarding ZPP/H ratio; AUC was 0.993 (95% confidence interval: 0.984-1), p<0.001. At a cutoff point > 26.09, the sensitivity was 100% and specificity was 95.5%. Table 8

Table 5: Laboratory results in the studied group.

Laboratory results		**Studied group**	
		N=170	**%**
Hemoglobin (g/dl)	Mean ±SD	15.8±2	
	Range	11-20.5	
HTC (%)	Mean ±SD	47.5±5.6	
	Range	36.9-62	
Platelets (10^3/L)	Mean ±SD	228±98	
	Range	40-495	
Total leucocyte count (10^3/L)	Mean ±SD	12.02±4.9	
	Range	2.6-29.6	
Total Neutrophils (%)	Mean ±SD	48.3±17.2	
	Range	17-83	
Staff Neutrophils (%)	Mean ±SD	2.1±1.9	
	Range	0-10	
Segmented Neutrophils (%)	Mean ±SD	46.2±17.3	
	Range	13-73	
I/T Ratio (%)	Mean ±SD	0.05±0.05	
	Range	0-0.26	
Reticulocyte count (%)	Mean ±SD	3.7±1.9	
	Range	0.5 -11.9	

HTC: hematocrit, I/T: immature/total neutrophils

Table 6: Serum ferritin according to clinical data in the studied group.

Clinical data		**Ferritin (ng/ml)**		**Test**	**P value**
		Mean±SD	**Range**		
Sex	Male	873±975	168-4837	U=0.58	0.56
	Female	671±504	168-2217		
Maternal fever	No	763±785	168±4837	U=1.96	**0.040***
	Yes	1401±323	1101-1801		
PROM	No	710±596	168-2463	U=0.47	0.63
	Yes	858±977	178-4837		
Mode of delivery	Caesarian section	754±807	168-4837	U=0.65	0.51
	Vaginal delivery	846±725	196-2463		
Resuscitation	Not Attend	784±602	168-2463	K=12.3	**0.007***
	RD II	384±260	168-1024		
	RD III	708±549	178-2217		
	RD IV	1264±1500	232-4837		
Oxygen support on admission	No need	818±384	272-1117	K=4.3	0.23
	Nasal O2	644±398	168-1319		
	CPAP	621±556	178-2217		
	MV	1266±1358	204-4837		
Cause of admission	RD	774±814	168-4837	K=0.87	0.64
	Jaundice	800±367	272-1074		
	Poor feeding, vomiting, ..	783±500	168-1272		

U; Mann-Whitney U test, K; Kruskal-Wallis test; * significant, DM; Diabetes mellitus, HTN: hypertension, SLE; systemic lupus erythematosus, UTI: urinary tract infection, RD; respiratory distress, MV: mechanical ventilator, N.CPAP; neonatal continuous positive airway pressure.

Table 7: Zinc Protoporphyrin to Heme ratio (ZPP/H ratio) according to clinical data in the studied group.

Clinical data		ZPP/H ratio		Test	P value
		Mean±SD	Range		
Sex	Male	21.2±4.5	10.8-32.03	t=1.3	0.18
	Female	20.4±2.7	15.7-25.6		
Maternal fever	No	20.9±3.6	15.3-32.1	t=3.9	<0.001*
	Yes	18.9±4.2	10.5-27.5		
PROM	No	20.9±3.8	15.35-32.1	t=0.25	0.80
	Yes	20.76±3.6	10.8-28.7		
Mode of delivery	CS	20.7±3.4	10.8-32.1	t=0.77	0.51
	NV	21.2±4.7	15.97-30.3		
Resuscitation	Not Attend	20.5±4.2	15.3-32.1	F=5.8	0.001*
	RD II	19.47±2.5	16.9-22.96		
	RD III	20.4±3.2	15.7-28.7		
	RD IV	19.77±3.8	10.8-26.3		
Oxygen support on admission	No need	29.2±2.6	26.4-32.1	F=15.4	<0.001*
	Nasal O2	20.2±3.1	15.3-30.1		
	CPAP	20.8±3.5	15.97-28.7		
	MV	20.48±3.5	10.8-26.3		
Cause of admission	RD	20.6±3.5	10.8-30.1	F=8.8	<0.001*
	Jaundice	26.4±6	19.9-32.1		
	Poor feeding, vomiting	20±1.3	18.9-21.96		

t: Student t-test; F: F value of one way ANOVA, *; significant, DM; Diabetes mellitus, HTN: hypertension, SLE; systemic lupus erythematosus, UTI: urinary tract infection, RD; respiratory distress, MV: mechanical ventilator, N.CPAP; neonatal continuous positive airway pressure.

Table 8: Performance of serum ferritin, ZPP, and ZPP/H ratio to predict anemia in preterm infants.

Variables	AUC	94% CI	Cut-off value	Sensitivity	Specificity	P value
Ferritin (ng/ml)	0.582	0.495-0.669	-	-	-	0.065
ZPP (µg/dl)	0.876	0.823-0.928	>13.25	79%	79.8%	<0.001*
ZPP/H ratio	0.993	0.984-1	>26.09	100%	95.5%	<0.001*

Discussion

In the current research, 81 patients (49.1%) of the examined neonates had anemia. This supports the findings of the research done earlier (5), which revealed that anemia usually does not develop in all the children who are early delivered with gestational ages between 33 and 37 weeks.

In another study the prevalence of iron deficiency (ID) was 10% and the prevalence of iron deficiency anemia (IDA) was 6%. Seventy percent of the participants in their research were anemic. The prevalence iron deficiency anemia

was found in 26.5% in Brazil, 9.9% in Sweden and 42.8% in Turkey. (7-9) Different subjects' traits, ages at sampling, nutrition, iron supplementation plans, and the ferritin cut-off points could all account for the variations (6),

In the present study, platelet count was significantly decreased in sepsis patients than in non-sepsis patients, this goes with other researchers (10) who tested platelets in neonatal sepsis. Comparing the sepsis group with the control group, they discovered a statistically significant decline. Also, some other scientists (11) discovered that the platelet count was noticeably decreased in the patients compared to the controls. The causes of thrombocytopenia can be platelet retention, consumption coagulopathy, and bone marrow depression. In the septic group, CRP levels were noticeably higher (12). Similarly, others (13 ,10) stated that septic patients had higher CRP levels.

Some studies indicated that CRP is still a significant, sensitive, and specific acute-phase protein for the prognosis of sepsis even though a number of novel indicators of infection have been researched lately, particularly in developing nations (14). It has been reported that CRP is not necessarily diagnostic for sepsis as elevations may elevate due to the physiologic rise after birth or non-infection associated conditions (15).

The present study showed that, I/T ratio was significantly elevated in neonatal sepsis. This agrees with those who reported that neutrophil left shift (I/T ratio $\geq$ 0.2) showed a significant correlation with early clinical signs of sepsis ($P < .001$), and higher incidence of late-onset sepsis (LOS) ($P = 0.001$) (16). Others revealed that the I/T ratio was higher in culture positive cases (17).

In our study, serum ferritin was significantly elevated in the sepsis patients. This came in agreement with what was reported as a relation between high levels of ferritin and the severity of sepsis (18). In their study the highest values of serum ferritin occurred in the multiple organ dysfunction syndrome (MODS) stage of neonatal sepsis (18). Serum ferritin is an acute phase protein, which increases during infection or inflammation (19).

The current study showed that, ferritin was significantly lower in the anemic than in the non-anemic patients.

Cohen et al., 2010 (20) stated that serum ferritin comprises 689 iron atoms per ferritin molecule while liver ferritin comprises 2074 iron atoms per molecule. Serum ferritin is a major source of iron as it engages in iron delivery, despite the frequent tendency to refer to it as "iron poor" that one frequently encounters in the literature. By shifting the ferritin cargo to the plasma membrane for secretion, it was further explained as how this secretion process depends on the tripartite motif-containing 16 (TRIM16) secretory autophagy receptor (21).

The present research demonstrated that after 4 weeks, the ZPP/H ratio significantly increased. The laboratory's reference ratios based on corresponding newborn-corrected gestational age to guide iron supplementation were used (22). The highest ZnPP/H ratios are found in the most premature newborns, possibly due to their greater fetal growth velocity, greater relative increase in blood volume, and greater iron accretion needs (22).

As shown before, ZnPP/H ratios in premature infants at or after discharge from the neonatal intensive care unit were higher than the more stringent normal

limits for the postnatal age, but normal for the corresponding postconceptional age (23).

It has been reported previously (3) that nearly half of the children studied had an abnormal baseline ZPP, most of whom did not have anemia. Their second important finding was that although ZPP and other markers of iron status improved in their study population regardless of iron prescription, those prescribed iron had significantly greater improvements in ZPP and Hb at the time of follow-up. Their results suggest that ZPP could be useful in monitoring the response to therapy after iron is initiated. In a smaller study (24), the prevalence of abnormal ZPP found in African American infants in Detroit was 52%.

The current study showed that, serum ferritin was significantly increased after 4 weeks. It was stated that high ferritin level suggests inflammation, and low level confirms iron deficiency (25).

The current study showed that, no significant differences between both groups were detected as regards ZPP/H ratio. The results of (3) declared that common clinical inflammatory conditions, with instances of culture-positive sepsis, had less impact on ZnPP/H than ferritin.

The current study showed that serum ferritin was significantly elevated in sepsis patients. In agreement with the current research, it was noted that clinical inflammatory states, such as sepsis with a positive culture greatly impacted ferritin levels (3). The mean log values of ferritin changed considerably during sepsis, with an adjusted differential of 1.19 ng/mL (adjusted P=0.001).

A research highlights the significance of H-ferritin in the reaction to microbial infection. The authors infected either wild-type or H-ferritin knockout mice with Mycobacterium tuberculosis. They showed that knocking out H-ferritin specifically in myeloid cell populations decreased survival after infection, increased the overall inflammatory response, and increased the total bacterial loads (26).

According to the present research, serum ferritin significantly increased following blood transfusion compared to baseline. Our results were consistent with those of who claimed that red blood cell transfusions and other clinical inflammatory conditions had a substantial impact on ferritin (3). With a modified difference in mean log values of 1.03 ng/mL (adjusted P = 0.001), red blood cell transfusion administered within 7 days of the laboratory test substantially impacts ferritin levels.

Our study showed that, ZPP/H ratio decreased significantly after blood transfusion. This goes with the study which reported that, within 7 days of red blood cell transfer, a slight drop in ZnPP/H was noticed. The dilution impact of transfusing adult red blood cells, which have a smaller ZnPP/H ratio than newborns, may be the cause of this decline (3).

With instances of red blood cell transfusion, it was found that ZnPP/H was less influenced by prevalent clinical inflammatory conditions than ferritin. Infants with a positive blood culture within 2 days of or 1 day after ZnPP/H measurement showed no discernible change in ZnPP/H levels (3).

When adult RBCs are infused, the infant's iron status and the iron status of the adult who donated the cells combine to produce a ZnPP/H. Fifteen to eighteen percent of the circulating RBCs are supplied by a 15

mL/kg infusion. The typical adult ZnPP/H varies from 30 to 80, so one would anticipate a sharp decline in ZnPP/H after transfusion (27).

Conclusion:

Since sepsis and transfusion have greater impact on ferritin than on ZnPP/H, ZnPP/H may be a more accurate indicator of iron state in these patients than ferritin.

Regarding the impacts of clinical events, serum ferritin levels were considerably lower in the anaemic patients than in the non-anemic group, whereas ZPP and ZPP/H ratio levels were significantly greater in the anaemic group. In the septic group, serum ferritin levels were considerably higher.

Conflict of interest:

None of the contributors declared any conflict of interest.

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