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The Relationship Between Umbilical Cord Blood Interferon γ-Inducible Protein-10 (IP-10) Levels and Clinical and Laboratory Parameters in Preterm Infants

Preterm Bebeklerde Umbilikal Kord Kanı İnterferon γ ile Uyarılabilen Protein-10 (IP-10) Düzeyleri ile Klinik ve Laboratuvar Parametreleri Arasındaki İlişki

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Öz

İnterferon γ ile uyarılabilen protein-10 (IP-10) güçlü inflamatuar mediatörlerden biridir. Çalışmamızda antenatal erken membran rüptürü (PPROM), fetal inflamatuar yanıt sendromu (FIRS) ve prematüreliğe bağlı morbiditeleri olan veya olmayan prematüre bebeklerde kordon kanı IP-10 düzeylerinin karşılaştırılması amaçlanmıştır. 37. gebelik haftasının altında doğan 85 prematüre bebek çalışmaya dahil edildi. Doğum anında umbilikal korddan alınan kan örneklerinde ELİSA yöntemi ile interlökin (IL)-6 ve IP-10 düzeyleri ölçüldü. Tüm olgularda prematüreliğe bağlı gelişebilecek komplikasyonlar (respiratuar distres sendromu, erken ve geç başlangıçlı sepsis, nekrotizan enterokolit, intraventriküler kanama, prematüre retinopatisi, bronkopulmoner displazi) ve mortalite kaydedildi. Kordon kanında 11 pg/ml üzerinde olan IL-6 düzeyleri FIRS olarak kabul edildi. PPROM'lu grupta (n=27, %31.8) kordon kanında medyan IP-10 seviyesi diğer gruplara göre anlamlı derecede yüksek bulundu (IP-10=345.6 pg/ml vs. 28.3 pg/ml, p<0.001). FIRS saptanan olgularda (n=36, %42.4) kordon kanında medyan IP-10 düzeyi FIRS saptanmayanlara göre anlamlı derecede yüksek saptandı (p<0.001). Erken başlangıçlı sepsis gelişen olgularda da kordon kanında medyan IP-10 seviyesi anlamlı derecede yüksek idi (p=0.019). Prematüreliğe bağlı diğer morbiditeler ile kordon kanı IP-10 düzeyi arasında anlamlı bir ilişki bulunamadı. Çalışmamızda fetal inflamasyonu olan ve erken başlangıçlı sepsis gelişen prematüre bebeklerde kordon kanında IP-10 seviyelerinin yüksek olduğu saptanmıştır. Kordon kanında yüksek IP-10 seviyesi, neonatal sepsis gelişen/gelişecek prematüre bebeklerde intrauterin inflamasyonu göstermek için erken bir belirteç olarak kullanılabilir.

Anahtar Kelimeler: Fetal İnflamatuar Yanıt Sendromu, İnflamasyon, İnterferon γ ile Uyarılabilen Protein-10, Prematürite, Sepsis

Abstract

Interferon γ-inducible protein-10 (IP-10) is one of the potent inflammatory mediators. This research aims to compare cord blood IP-10 levels in preterm infants with or without antenatal preterm prelabor rupture of the membranes (PPROM), fetal inflammatory response syndrome (FIRS) and prematurity related morbidities. We enrolled 85 newborns with gestational age below 37 weeks. Umbilical cord blood samples were obtained at delivery and stored. Cord blood IP-10 and interleukin (IL)-6 levels measured with ELISA test. All enrolled preterm infants have been followed-up for prematurity related conditions including respiratory distress syndrome, early and late onset sepsis, necrotising enterocolitis, intraventricular haemorrhage, premature retinopathy, bronchopulmonary dysplasia and mortality. FIRS defined as IL-6 levels of umbilical cord above 11 pg/ml. Cord blood median IP-10 levels were significantly higher in PPROM group (n=27, 31.8%) than in the group without PPROM (IP-10=345.6 pg/ml vs. 28.3 pg/ml, p<0.001). Cord blood median IP-10 levels were significantly higher in preterm infants with FIRS (n=36, 42.4%) compared to infants without FIRS (p<0.001). Cord blood median IP-10 levels were also higher in preterm infants with early onset sepsis than those without early onset sepsis (p=0.019). We did not observe relationship between cord blood IP-10 levels and other prematurity-related complications. Increased cord blood IP-10 levels have been observed in preterm infants with fetal inflammation and who developed early onset sepsis. Cord blood IP-10 could be considered an early marker for intrauterine inflammation and its effect on fetal outcomes, such as the development of neonatal sepsis in preterm infants.

Keywords: Fetal Inflammatory Response Syndrome, Inflammation, Interferon γ-Inducible Protein-10, Prematurity, Sepsis

Introduction

Infants born before the 37th postconceptional week of pregnancy or before the 259th day from the mother's last menstrual period are named as "preterm infants". Approximately 10% of all births are preterm, and 1-2% of these infants are younger than the 32nd week of gestation and have a birth weight below 1500 grams (1,2). In many cases, the cause of preterm birth cannot be diagnosed exactly, but increasing risk factors increases the incidence of preterm birth. The most common pathological condition that causes preterm birth is inflammation of the maternal-fetal connection (3). Studies have shown that 40% of preterm births involve intrauterine inflammation and infection, and these infections are mostly subclinical. In particular, it has been found that more than 80% of women who give birth before the 28th week of gestation have

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intrauterine infection, and infection rates decrease as the gestational week progresses (4).

Survival rates of preterm infants have increased in recent years. This increase in survival rates has also brought about an increase in morbidity rates. In addition to the different biological structure and physiological characteristics of preterm infants, respiratory distress syndrome (RDS), necrotizing enterocolitis (NEC), intraventricular hemorrhage (IVH), retinopathy of prematurity (ROP), and in the long-term cerebral palsy (CP) and bronchopulmonary dysplasia (BPD) like serious morbidities affects the prognosis (5). These problems specific to premature infants are associated with fetal inflammation, and their incidence and severity increase in the presence of inflammation (3).

Interferon-γ inducible protein-10 (IP-10) or C-X-C motif chemokine ligand 10 (CXCL10) is a chemoattractant chemokine for T cells, exerting its effects through interaction with the cell surface chemokine (C-X-C motif) receptor 3 (CXCR3) (6-8). CXCR3, a cell-surface G protein-coupled receptor expressed mainly by T-helper (Th) 1 cells, cytotoxic T cells and natural killer cells that have a key role in immunity and inflammation. IP-10 is secreted from cells as a response to increased interferon (IFN)-γ. Expression of IP-10 is seen in many Th 1 type inflammatory diseases (e.g., multiple sclerosis, rheumatoid arthritis, psoriasis, systemic lupus erythematosus, graves disease, inflammatory bowel dissease, and type I diabetes), where it is thought to play an important role in recruiting activated T cells into sites of tissue inflammation (6). In these diseases, IP-10 levels correlate with tissue infiltration of T lymphocytes (6-9).

There are limited studies on IP-10 in newborn infants. Previous research suggests that serum IP-10 levels are significantly increased in birth-asphyxiated and perinatally infected neonates (10). In infants younger than four months (including newborns) with suspected serious bacterial infections, IP-10 assays might be predictive (11). A study in preterm lambs exposed to chorioamnionitis showed that IP-10 might contribute to lung injury and altered pulmonary vascular development (12).

Currently, cytokine levels in cord blood, placental pathological examination, clinical examination in the postnatal period, and imaging are the methods used in diagnosis to evaluate the fetal effects caused by intrauterine inflammation (2, 13). In this study, we evaluated the effects of IP-10 and interleukin (IL)-6 levels in the cord blood of preterm infants on clinical and laboratory parameters and their role in determining the severity of inflammation.

Material and Method

This prospective single-center cohort study was conducted at the Neonatology clinic of Osmangazi University Faculty of Medicine Hospital Center (a level III neonatal intensive care unit [NICU]) in Eskisehir, between 2010 June and 2011 July. All infants born less than 37 weeks of gestation and admitted to the NICU for any reason were included in the study. Infants with major congenital anomalies and those referred to other hospitals were excluded due to the potential for missing medical records. Ethical approval for the study was obtained from the institutional ethics committee (dated 21.05.2010, numbered 86).

Antenatal histories, birth weight, and demographic findings of the cases were recorded. The infants were divided into four groups according to their antenatal history (Group 1: those with preterm prelabor rupture of membranes [PPROM], Group 2: those with preeclampsia, Group 3: those with gestational diabetes, Group 4: those without any risk factors).

The infants included in the study were divided into two groups according to the presence of PPROM in antenatal history, and early onset sepsis and IP-10 levels were compared between the groups.

IL-6 and IP-10 levels were measured in cord blood as markers of fetal inflammation in all preterm infants. Infants with IL-6 levels above 11 pg/ml in cord blood were classified as having “fetal inflammatory response syndrome” (FIRS) (13). The study group was then stratified by the presence or absence of FIRS. Neonatal morbidities, mortality and IP-10 levels were compared between these two groups.

During their NICU stay, all preterm infants were monitored for complications that may develop due to fetal inflammation and prematurity (RDS, early and late onset sepsis, NEC, IVH, ROP, BPD and mortality).

Sample collection

Blood samples were taken from the umbilical cord, centrifuged at 5000 rpm for 10 minutes, and the serum was separated and stored at -80 °C until analysis.

Measurement of cytokine levels

ELISA kits (Immunoassays Quantikine kits R&D Systems for human IL-6 and IP-10) were used for IL-6 and IP-10 measurements.

Clinical description

PPROM is defined as the rupture of fetal membranes before labor begins and failure to deliver within 18-24 hours following membrane rupture (14).

RDS was defined as respiratory distress with cyanosis on room air, tachypnea (respiratory rate

>60/min), intercostal retractions, and the persistence or progression of respiratory distress for 48-96 hours of life, along with a diffuse reticulogranular appearance on chest radiography and an air bronchogram (15).

Neonatal sepsis occurs in the presence of at least three of the following clinical findings, including: tachycardia (heart rate >200/min) (except in cases such as sleep, anemia, hypo/hyperthyroidism, pain, post-feeding) or bradycardia, hypotonia, hypotension, seizure, tachypnea, apnea, respiratory distress, cyanosis, impaired skin color and perfusion, malnutrition, lethargy, irritability. These clinical findings were evaluated as high acute phase reactants and/or accompanying blood culture positivity. If sepsis findings appeared within the first 72 hours of life, it was considered "early onset" sepsis, and if it appeared after the first 72 hours of life, it was considered "late onset" sepsis (16).

Diagnosis and staging in patients with clinical and radiological signs and symptoms suggestive of NEC were made according to modified Bell scoring (17).

Risky preterms (<34 weeks of gestation) and infants who were clinically considered to have IVH were evaluated using transfontanel ultrasonography (USG) (18).

BPD was diagnosed using criteria from the American National Public Health Institute and classified as mild, moderate, or severe (19).

ROP screening was performed on infants born <1500 g or <32 weeks of gestation. The screening program started at 4-6 weeks after birth or at 31-33 weeks postconceptionally. Staging was done according to vascular proliferation (20).

Statistical evaluation

'SPSS for Windows 27.0' package program was applied in statistical evaluation of the results. Whether the quantitative variables conformed to normal distribution was examined with the Kolmogorov-Smirnov test. Independent groups were compared for non-normally distributed variables using the Mann Whitney U test or Kruskal Wallis analysis of variance. The relationship between quantitative variables was determined by Pearson or Spearman correlation analysis; The relationship between qualitative variables was examined with chi-square analysis. Descriptive statistics of quantitative variables that conform to normal distribution are shown as mean ± standard deviation, and descriptive statistics of quantitative variables that are not normally distributed are shown as median (25 th-75 th percentiles). P<0.05 values were considered statistically significant.

Results

In total, 85 preterm infants were admitted to the NICU during the study period. Of these, 49.4% were female and 50.6% were male. The majority of infants (70.6%) were delivered via cesarean section. The median gestational age of the infants was 33 (30.5-35) weeks, and mean birth weight was 1924 ± 699 g. Epidemiological data of preterm infants included in the study are shown in Table 1.

Table 1. Demographic characteristics of preterm infants.

Preterm infants (n=85)	
Gestational age (week)#	33 (30.5-35)
Birth weight (g)*	1924±699
Cesarean birth n (%)	60 (70.6)
Female n (%)	42 (49.4)
Apgar score 1th min#	5 (3-7)
Apgar score 5th min#	8 (6-9)
Maternal age (years)*	28.7±5.0

*Mean ± Standard deviation, #Median (25 th-75 th percentiles)

Antenatal history and IP-10

The maternal risk factors of the infants are shown in Table 2. There was a significant difference in IP-10 and IL-6 levels between the four groups (p<0.001 and p<0.001, respectively) (Table 2). In the post-hoc analysis, cord blood median IP-10 and IL-6 levels were found to be significantly higher in group 1 than in groups 2, 3 and 4 (Figure 1). There was no significant difference in C-reactive protein (CRP) levels among the four groups (p=0.399) (Table 2).

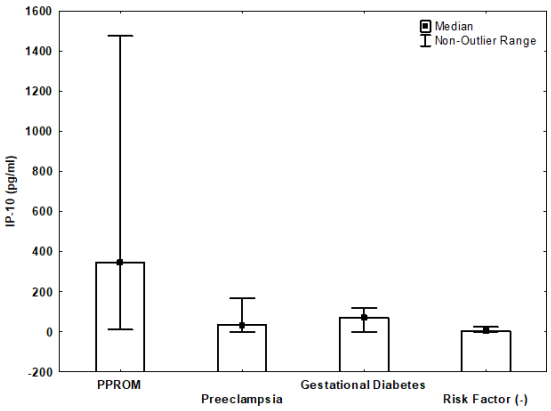

Figure 1. Cord blood IP-10 levels of the neonates according to maternal risk factors. PPROM: preterm prelabor rupture of the membranes, IP-10: interferon γ-inducible protein-10

In our study, six of 17 preterm infants who developed sepsis (five early onset sepsis, one late onset sepsis) had PPROM in their antenatal history. The median IP-10 level (345.6 pg/ml) in the group with antenatal PPROM was significantly higher than in the group without PPROM (28.3 pg/ml) (p<0.001). While the rate of early onset sepsis was twice as high in the PPROM group (18.5%) compared to the non-PPROM group (8.6%), there was no significant statistically difference between two groups (p=0.277). These findings are summarized in Table 3.

Table 2. Cord blood IP-10, IL-6 and CRP levels of the infants according to maternal risk factors

	Group 1 (n=27)	Group 2 (n=20)	Group 3 (n=15)	Group 4 (n=23)	p
IP-10 (pg/ml)	345.6 (141.2-924.2)**	33.3 (0.72-111.7)	69.7 (34.5-101.6)	4.8 (0.76-24.9)	**<0.001**
IL-6 (pg/ml)	14.6 (12.0-26.6)**	4.28 (1.1-16.7)	3.56 (1.8-6.3)	1.65 (0.6-6.4)	**<0.001**
CRP (mg/dl)	0.50 (0.2-3.08)	0.37 (0.15-16.7)	1 (0.1-3)	2.1 (0.3-3.08)	0.399

IP-10: interferon γ-inducible protein-10, IL-6: interleukin-6, CRP: C-reactive protein. Data are presented as median (25 th-75 th percentiles). **Group 1 is statistically different from group 2, group 3 and group 4 (p<0.001, p=0.008 and p<0.001 for IP-10 respectively and p=0.009, p=0.010 and p=0.010 for IL-6 respectively).

Morbidity and IP-10

Early onset sepsis developed 10 (11.8%) and late onset sepsis developed 7 (8.2%) of the infants. In our study, 20 (23.5%) of the infants had RDS; 4 (4.7%) had NEC (stage 1B in one, stage 2A in two, and stage 3A in one infant); 7 (8.2%) had IVH (stage 1 IVH in two, stage 2 IVH in four, and stage 4 IVH in one infant); 5 (5.9%) had ROP (stage 3 ROP in four, stage 4 ROP in one infant); and 9 (10.6%) had BPD (mild BPD in three, moderate BPD in four, and severe BPD in two infants).

The cord blood median IP-10 level of infants who developed early onset sepsis was significantly higher than that of infants who did not develop early onset sepsis (p=0.019), but no significant difference was found in terms of IL-6 levels (p=0.350) (Table 4). No significant statistical difference was detected between the cord blood median IP-10 and IL-6 levels of infants who developed and did not develop late onset sepsis, RDS, NEC, IVH, ROP, or BPD (p>0.05) (Table 4).

Mortality and IP-10

Ten (11.8%) of 85 preterm infants in the study group died. The cord blood median IP-10 level of these infants was 81.3 pg/ml (6.3-201), and the IL-6 level was 6.4 pg/ml (1.5-17.3). When the cord blood median IP-10 and IL-6 levels of these infants and the surviving preterm infants were compared, significant statistically difference was not detected between them (p=0.817 and p=0.859 respectively) (Table 4). Since nine out of 10 infants who developed mortality died before the 28th day, ROP and BPD were not evaluated in these infants.

FIRS and IP-10

FIRS was present in 36 (42.4%) of the preterm infants in the study group. The cord blood median IP-10 level of infants with FIRS (202.5 pg/ml [range=119.4-873.6 pg/ml]) was significantly higher than those without FIRS (11.8 pg/ml [range=0.81-62.1 pg/ml]) (p<0.001). From the perspective of perinatal morbidity, there was no statistical difference in terms of RDS, late onset sepsis, BPD, NEC, IVH, ROP frequency, and mortality between preterms with and without FIRS (p>0.05). Although the risk of early onset sepsis was higher in those with FIRS, it was not statistically significant (p=0.088) (Table 5).

Other Findings

A positive significant correlation was detected between cord blood IP-10 level and cord blood IL-6 level (r=0.80, p<0.001).

In Table 3, the resulting effect size of the posterior power analysis performed in the GPower program through the IP-10 descriptive statistics of those with and without PPROM was calculated as 2.512 and the obtained power was calculated as 99%.

Discussion

In our study, significant data were evaluated on the relationship between high cord blood IP-10 levels and morbidity, mortality, and other laboratory parameters in the neonatal period. This relationship has been recently researched, but data are limited. This is the first study to show that IP-10 levels are elevated in the cord blood of preterm infants with a history of PPROM in the antenatal period, FIRS, and early onset sepsis.

Table 3. Presence of early onset sepsis and IP-10 level according to the presence of PPROM

	PPROM (n=27)	No PPROM (n=58)	p
Early onset sepsis			
Present	5 (18.5)	5 (8.6)	0.277
Absent	22 (81.5)	53 (91.4)	
IP-10 (pg/ml)#	345.6 (141-924)	28.3 (0.92-99.1)	**<0.001**

PPROM: preterm prelabor rupture of the membranes, IP-10: interferon γ-inducible protein-10. #Median (25 th-75 th percentiles).

Table 4. Distribution of cord blood IP-10 and IL-6 levels according to neonatal morbidity and mortality

		IP-10 (pg/ml)	p	IL-6 (pg/ml)	p
RDS	Yes (n=20)	29.7 (2.1-164.2)	0.315	2.2 (1.1-13.2)	0.147
	No (n=65)	96.7 (11.8-208.7)		9.7 (1.9-18.4)	
Early onset sepsis	Yes (n=10)	173.4 (89.3-1236)	**0.019**	12.6 (5.0-18.2)	0.350
	No (n=75)	54.6 (4.8-145.6)		4.7 (1.4-14.6)	
Late onset sepsis	Yes (n=7)	11.2 (0.8-145.6)	0.350	1.6 (1.4-14.3)	0.554
	No (n=78)	77.1 (11.1-178.0)		6.4 (1.7-15.7)	
NEC	Yes (n=4)	72.3 (1.7-143.6)	0.501	7.0 (1.0-14.0)	0.638
	No (n=81)	69.8 (10.3-179.6)		6.4 (1.6-16.2)	
IVH	Yes (n=7)	9.5 (0.2-1234)	0.678	1.8 (1.0-13.1)	0.482
	No (n=78)	77.1 (11.5-171.9)		6.4 (1.6-15.7)	
ROP	Yes (n=5)	137.9 (0.6-690)	0.903	12.9 (0.6-17.7)	0.919
	No (n=71)	69.7 (9.5-168.9)		5.6 (1.4-17.3)	
BPD	Yes (n=9)	1.2 (0.1-410.7)	0.364	1.8 (0.6-17.3)	0.399
	No (n=67)	69.8 (11.7-168.9)		6.3 (1.5-17.3)	
Mortality	Yes (n=10)	81.3 (6.3-201)	0.817	6.4 (1.5-17.3)	0.859
	No (n=75)	69.8 (9.5-168.9)		6.4 (1.6-13.6)	

RDS: respiratory distress syndrome, NEC: necrotizing enterocolitis, IVH: intraventricular haemorrhage, ROP: retinopathy of prematurity, BDP: bronchopulmonary dysplasia, IP-10: interferon γ-inducible protein-10, IL-6: interleukin-6. Data are presented as median (25 th-75 th percentiles). *Since nine out of 10 infants who developed mortality died before the 28th day, ROP and BPD were not evaluated in these infants.*

Preterm births are the most significant cause of perinatal morbidity and mortality (1). The most common cause of preterm birth is inflammation and infection of the maternal-fetal junction. This inflammation and infection are mostly subclinical. Clinical and subclinical chorioamnionitis constitute 50% of preterm births, especially below the 30th week of gestation (21, 22). Similar to other studies, our study found that predisposing factors for preterm birth include pregnancy morbidities that initiate an inflammatory response, such as PPROM, preeclampsia, and gestational diabetes. It is not surprising that PPROM was found to be the most common cause in the prenatal history in our study, as it is the most common detectable factor associated with preterm birth and is present in approximately one-third of preterm births (23, 24). In our study, we found higher IP-10 and IL-6 levels in the cord blood of preterm infants with an antenatal history of PPROM compared to those without any antenatal disease.

The relationship between the increase in pro-inflammatory cytokines and preterm birth was first reported by Gomez et al. (13). IL-6 value of fetal plasma above 11 pg/ml obtained by cordocentesis in 105 pregnant women with preterm labor and 152 pregnant women with PPROM can be considered as the 'cut-off' value of the fetal inflammatory response, and IL-6 values above this figure are associated with increased neonatal morbidity. In other studies, it has been shown that there is a positive correlation between increased pro-inflammatory mediators in the amniotic fluid of pregnant women with preterm birth and amniotic fluid and fetal membrane culture results (25, 26). In our study, FIRS was more common in preterms with lower gestational age, in line with literature data. Additionally, the cord blood IP-10 level in patients with FIRS was higher than in those without FIRS. This finding suggested the pro-inflammatory role of IP-10 in the inflammation mechanism. Especially preterm infants with identified fetal inflammation face serious morbidities. RDS, sepsis, NEC, IVH and BPD are the most common causes of morbidity and mortality in these infants (27). While the incidence of early onset sepsis was higher in our cases who developed FIRS compared to those who did not, we found no significant relationship between the presence of FIRS and other neonatal morbidities, contrary to some data in the literature (27).

Neonatal sepsis is the leading cause of mortality and morbidity in preterm and very low birth weight infants (28, 29). As gestational age and birth weight decrease, the risk of developing sepsis increases (21). PPROM increases fetal infection risk, especially when chorioamnionitis is present, and the risk of neonatal sepsis rises (30, 31). In our study, six of the seven infants who developed early onset sepsis had PPROM in their antenatal history, and the rate of early onset sepsis in cases with PPROM was found to be twice as high as in cases without PPROM.

Interest in inflammatory mediators has recently focused on a group of small molecular weight cytokines known as chemokines. Chemokines are mostly secreted from inflamed or infected tissues and play significant role in different stages of the inflammatory pathways. There is a strong relationship between the amount of chemokine release and the severity of the inflammatory response (11). There are a few studies in the literature on IP-10 and other chemokine production and circulating concentrations in preterm infants (10, 11, 32, 33). In our study, the cord blood median IP-10 level of the preterm infants who developed early onset sepsis was significantly higher than those who did not develop sepsis. This finding shows that IP-10 is a valuable marker in detecting early onset sepsis. The CRP level, which is the most commonly used laboratory parameter in the early diagnosis of severe bacterial infections that cannot be detected clinically, was positive in only two of our patients

who developed early onset sepsis. Thus, it was shown once again that CRP is not a very sensitive marker of infection. Similar to our study, a study conducted on infants under four months of age, including newborns, showed that plasma IP-10 level was superior to white blood cell count and CRP levels in determining serious bacterial infections (11, 34).

In another study investigating chemokine levels to detect sepsis-induced disseminated intravascular coagulation in preterm infants at an early stage, it was found that IP-10 and other chemokines (monocyte chemoattractant protein-1 [MCP-1], IL-8, and monokine induced by interferon-γ [MIG]) were increased in infants with NEC and septicemia. This study demonstrated that preterm infants have the ability to mount strong cytokine and chemokine responses against pathogens (32).

In a large study evaluating inflammatory mediators as diagnostic markers in preterm infants with late onset bacterial infection, several cytokines and chemokines, including IP-10, IL-8, IL-6, MCP-1, MIG, regulated upon activation normal T cell expressed and secreted (RANTES), IL-1β, IL-10, and tumor necrosis factor-α (TNF-α), were studied at 0 and 24 hours. It was found that IP-10 is the best diagnostic marker of infection with the highest cut-off value at 0 and 24 hours (33). In our research, no significant difference was detected in cord blood IP-10 levels between preterms who developed late onset sepsis and those who did not. The difference between these results and ours is likely related to the very small number of infants with late onset sepsis examined.

Table 5. Distribution of neonatal morbidity, mortality and IP-10 levels according to the presence of FIRS

		FIRS (n=36)	No FIRS (n=49)	p
RDS	Yes (n=20)	7 (19.4)	13 (26.5)	0.615
	No (n=65)	29 (80.6))	36 (73.5)	
Early onset sepsis	Yes (n=10)	7 (19.4)	3 (6.1)	0.088
	No (n=75)	29 (80.6)	46 (93.9)	
Late onset sepsis	Yes (n=7)	3 (8.3)	4 (8.2)	>0.999
	No (n=78)	33 (91.7)	45 (91.8)	
NEC	Yes (n=4)	2 (5.6)	2 (4.1)	>0.999
	No (n=81)	34 (94.4)	47 (95.9)	
IVH	Yes (n=7)	3 (8.3)	4 (8.2)	>0.999
	No (n=78)	33 (91.7)	45 (91.8)	
ROP	Yes (n=5)	3 (9.4)	2 (4.5)	0.644
	No (n=71)	29 (90.6)	42 (95.5)	
BPD	Yes (n=9)	4 (12.5)	5 (11.4)	>0.999
	No (n=67)	28 (87.5)	39 (88.6)	
Mortality	Yes (n=10)	4 (11.1)	6 (12.2)	>0.999
	No (n=75)	32 (88.9)	43 (87.8)	
IP-10 (pg/ml)[#]		202.5 (119.4-873.6)	11.8 (0.81-62.1)	**<0.001**

FIRS: fetal inflammatory response syndrome, RDS: respiratory distress syndrome, NEC: necrotizing enterocolitis, IVH: intraventricular haemorrhage, ROP: retinopathy of prematurity, BDP: bronchopulmonary dysplasia, IP-10: interferon γ-inducible protein-10. Data are presented as number (%) or [#]median (25 th-75 th percentiles). *Since nine out of 10 infants who developed mortality died before the 28th day, ROP and BPD were not evaluated in these infants.*

In our study, contrary to studies in the literature, no significant relationship was found between mean cord blood IP-10 and IL-6 levels and the development of neonatal morbidities such as IVH, NEC, ROP, and BPD (12, 32, 35, 36). Studies have mostly associated IP-10 with infection and inflammation. Infection and inflammation are not the only causes of neonatal morbidity; the etiology is multifactorial. Factors such as respiratory support applications, oxygen applications, birth asphyxia, invasive interventions, mechanical ventilation, and inability to breastfeed may contribute to the development of morbidities (21, 29). For this reason, IP-10 levels may not have been found to be high in those who developed morbidity in our study.

There were some limitations regarding our study. First, the study was single-centered. Second, the sample size was small.

Conclusion

Since cord blood IP-10 levels are detected to be higher in preterm infants who had PPROM and developed FIRS during the antenatal period, it is thought that this chemokine may cause preterm birth by triggering systemic inflammation. IP-10 may be favorable as an early indicator in determining the presence and degree of inflammation in preterm infants.

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