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ARAŞTIRMA / RESEARCH

Serum vitamin D levels in newborn with clavicle fracture

Klavikula kırığı saptanan yenidoğan hastalarda serum D vitamini düzeyleri

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Abstract

Purpose: The aim of this study was to determine the possible role of vitamin D on the clavicle fracture in the newborn.

Materials and Methods: This retrospective cross-sectional study was conducted between 2018-2020. Infants who were admitted to neonatal intensive care unit due to the clavicle fracture were included. Infants who did not have a fracture, but were admitted to the hospital for reasons instead of exclusion criteria were included as the control group. Birth weight, height, head circumference, gender, gestational week, delivery type, other clinical findings such as brachial plexus paralysis in the infant, maternal age, number of pregnancies, risk factors time to diagnosis were recorded. According to the serum 25-OH vitamin D levels, newborns were classified as having normal vitamin D status (>30 ng/mL), vitamin D insufficient (between 20-30 ng/mL), or deficiency (<20 ng/mL). Groups were compared for the vitamin D levels, the demographics and clinical characteristics.

Results: A total of 48 infants were included. The groups did not differ in terms of demographics. Age of diagnosis ranged from postnatal 1 to 6 days of life. Serum calcium levels were significantly lower in the case group. Although phosphorus and alkaline phosphatase levels did not differ between groups, vitamin D levels were significantly lower in the case group.

Conclusion: This study shows that newborns with clavicle fracture had lower vitamin D levels. Cautious palpation of the clavicle can catch the fracture before the hospital discharge even there is no evident symptom and serum vitamin D level of those infants should be checked in order to detect the possible deficiency timely.

Keywords: Clavicle fracture, newborn, vitamin D

Öz

Amaç: Bu çalışmada yenidoğan döneminde klavikula kırığı üzerine D vitamininin olası rolünün belirlenmesi amaçlanmıştır.

Gereç ve Yöntem: Bu retrospektif kesitsel çalışma 2018-2020 yılları arasında yapılmıştır. Klavikula kırığı nedeniyle yenidoğan yoğun bakım ünitesinde izlenen bebekler çalışmaya alındı. Kontrol grubu kırığı olmayan ancak dışlama kriterleri dışındaki nedenlerle hastaneye başvuran bebeklerden oluşturuldu. Doğum ağırlığı, boyu, baş çevresi, cinsiyeti, gebelik haftası, doğum şekli, bebekte brakiyal pleksus felci gibi diğer klinik bulgular, anne yaşı, gebelik sayısı, tanı zamanı ve risk faktörleri kaydedildi. Yenidoğanlar serum 25-OH D vitamini düzeylerine göre D vitamini düzeyi normal (>30 ng/mL), D vitamini yetersiz (20-30 ng/mL) veya eksikliği olanlar (<20 ng/mL) olarak sınıflandırıldı. Gruplar D vitamini düzeyleri, demografik ve klinik özellikler açısından karşılaştırıldı.

Bulgular: Çalışmamıza 48 bebek dahil edildi. Gruplar demografik açıdan benzer bulundu. Klavikula kırığı için tanı yaşı doğum sonrası 1 ila 6 gündü. Serum kalsiyum düzeyleri vaka grubunda anlamlı olarak daha düşüktü. Fosfor ve alkalin fosfataz düzeyleri gruplar arasında farklılık göstermemesine rağmen, D vitamini düzeyleri vaka grubunda anlamlı derecede düşüktü.

Sonuç: Bu çalışma, klavikula kırığı olan yenidoğan bebeklerde D vitamini düzeylerinin düşük olduğunu göstermiştir. Klavikulanın dikkatli bir şekilde palpasyonu hastaneden taburcu olmadan önce kırığı yakalayabilir ve bu bebeklerde olası eksikliği zamanında saptamak için serum D vitamini düzeyine bakılmalıdır.

Anahtar kelimeler: D vitamini, klavikula kırığı, yenidoğan

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INTRODUCTION

Vitamin D has an important place in both skeletal and extraskeletal health. Its fundemental role is regulatory in the intestinal absorption of calcium and phosphorus. Lower levels of vitamin D will result in elevations of parathyroid hormone due to the decreased intestinal absorption of calcium and phosphorus[1]. Hence, the parathyroid hormone causes the resorption of calcium from bone which ends in impaired bone mineralization and higher risk of osteomalacia, osteoporosis, and bone fractures[1,2]. Vitamin D also ranks among various extraskeletal entities including malignity, infection, autoimmune disorders, diabetes, and cardiac diseases[3,4]. Limited studies evaluated the association between vitamin D, or its main determinants, and fracture risk in children[5]. The clavicle fracture (CF) is observed during vaginal delivery with an estimated frequency of 1-1,5% associated with birth trauma[6]. The etiological factors for CF were defined as procedural induction of labor, prolonged labor, macrosomia, a narrow cervix, and use of delivery instruments and shoulder dystocia[1,4,7]. Although birth weight is the most common and well-known etiology for CF, maternal vitamin D deficiency is also shown to be an independent risk factor for CF of newborn, recently[8]. Besides, maternofetal vitamin D deficiency is proven to be associated with long bone fractures in newborn infants[9]. Despite vitamin D's crucial and having a well-established role in bone health, its association to CF of newborns is not yet studied. To the best of our knowledge this is the first study, in literature, showing newborn infants with clavicle fracture had lower vitamin D levels. Therefore, the hypothesis of this study is that there is an association between lower levels of vitamin D and CF in newborn infants.

MATERIALS AND METHODS

Study design and participants

This research was conducted as a retrospective cross-sectional study. Medical records of newborns were recorded and analyzed at the tertiary referral center of Tekirdag Namık Kemal University between Jan 2018-July 2020 by the same neonatologist. Infants who were admitted to level-I neonatal intensive care unit (NICU) due to the clavicle fracture, were included in this study. The study is approved by the institutional ethical committee of Tekirdag Namık Kemal University (15.01.2021/E-46048792-050.01.04-3525). Informed consent was obtained from parents at the first outpatient clinic control of infants following the ethics committee approval. The inclusion criteria were newborns who were diagnosed with CF based on clinical examination (skin discoloration, swelling, tenderness, and crepitus over the clavicle, etc.) and radiographic findings (X-ray) within the postnatal first week of life. Infants who were younger than 35 gestational weeks of age (>35 gestational weeks) were excluded to avoid the effect of prematurity-related metabolic derangements such as osteopenia. Newborns who have additional medical problems including trauma, congenital abnormalities, metabolic abnormalities, maternal or neonatal bone disorders (such as osteogenesis imperfecta, metabolic bone disease) were excluded.Use of instrumentation during delivery, such as forceps or vacuum extraction, and neonates with shoulder dystocia were also excluded. Perinatal conditions such as gestational DM, preeclampsia may disrupt the placental transmission, so pregnancies have with these conditions been excluded. Birth weight (g), height (cm), head circumference (cm), gender, gestational week, delivery type, other clinical findings such as brachial plexus paralysis in the infant, maternal age, number of pregnancies, CF risk factors (large baby, presentation anomaly, shoulder dystocia, etc.), time to diagnosis were recorded. Infants who did not have CF but were admitted to the hospital for reasons such as neonatal jaundice, transient tachypnea of the newborn instead of exclusion criteria were included as the control group.

For the comparison, age- and gender-matched control were enrolled into the study by using random numbers. Serum 25-OH vitamin D, calcium, phosphorus, and alkaline phosphatase were measured within the first postnatal week (48-72 hours of life) in both groups.

Biochemical analysis

According to the serum 25-OH vitamin D levels, newborns were classified as having normal vitamin D status (>30 ng/mL), vitamin D insufficient (between 20-30 ng/mL) or deficiency (<20 ng / mL)[10]. The serum concentration of 25-OH vitamin D was measured using an Electro Chemo Luminescence Immuno Assay from Roche Diagnostics.

Statistical analysis

Statistical analyses performed with GraphPad Prism (ver. 7.00) Software. Results presented as mean ±

SEM (mean ± standard error of the means). The minimum sample size of the study was calculated by using Cohen's high effect size (d=0.8) over the difference between the means of two independent groups. Accordingly, the minimum number of individuals to be included in the study at a power of 0.80 and an error probability of α=0.05 was calculated as 42 participants (21 newborn infant per group). Descriptive statistics (arithmetic mean, median, standard deviation, minimum-maximum values, etc.) were used in expressing the variables. Whether the data complies with the normal distribution was evaluated using the Shapiro-Wilk test.

After checking whether the variables meet the normality condition, t-test was used in case of normal distribution. Therefore, demographic features of newborn including gestational age, birth weight, maternal age were compared with Student's t test. with CF and the control group infants (Table 1). Also, mean vitamin D, Ca, P, ALP values of newborns with and without CF were compared with Student's t test (Table 2,4). P value <0,05 was considered statistically significant.

RESULTS

A total of 24 infants were found to be eligible for the study. Figure-1 demonstrates the flow diagram of the study (Figure 1). Fifteen of the participants were male and nine of them were female. Age of diagnosis ranged from postnatal 1 to 6 days of life. The gestational age ranged from 36 weeks to 40 weeks in the CF group with an average of 39 weeks. We included 24 healthy newborns of same age (1 to 6 days of life) and gender (15 male and 9 female) who were born at or after 35 weeks of gestation and did not have any medical issues including CF. In the control group, gestational age ranged from 35 to 41 weeks with an average of 39 weeks. Group 1 included 24 infants with CF and group 2 had 24 healthy infants. A comparison of demographic characteristics of groups is presented in Table 1 (Table 1).

The characteristics clinical findings of newborns with CF at the time of diagnosis are presented in Table 2 (Table 2). Although the birth weight was higher in CF group than the control group, the difference was not statistically significant (Table 1).

Table 1. Comparison of demographic features of newborn with clavicle fracture and the control group infants.

			Clavicula Fracture (+) (n=24)				Clavicle Fracture (-) (n=24)			
			Mean±SEM/n-%				Mean±SEM/n-%			
Maternal Age		26.18	±	1.13			31.17	±	1.05	**0.002**
Sex	Female		9	37.5%				9	37.5%	0.9
	Male		15	62.5%				15	62.5%	
Birth Weight		3789	±	85.25			3548	±	118	0.11
Gestational Age		37.2	±	2.3			37.4	±	3.8	0.16
Maternal Parity		2.52	±	0.28			1.79	±	0.18	0.58
Apgar Score 1st Min		7.05		0.38			7.96	±	0.21	0.61
Apgar Score 5th Min		8.47	±	0.23			9.05	±	0.10	0.37

SEM: Standard error of the mean

Table 2. Comparison of serum vitamin D levels between newborn with clavicle fracture and the control group.

Serum Vitamin D	Control (n=24)	Clavicle Fracture (n=24)	*P*
Mean, ng/ml	12.3±1.55	8.78±0.76	**0.047**

Table 3. Clinical characteristics of newborns with clavicle fracture at the time of diagnosis

Clinical finding	n
Reduced arm movements	21(87.5%)
Crepitation at the fracture site	3 (12.5%)
Skin discoloration	2 (8.33%)
Erb-Duchenne palsy	9 (37.5%)
Klumpke's palsy	1 (4.16%)

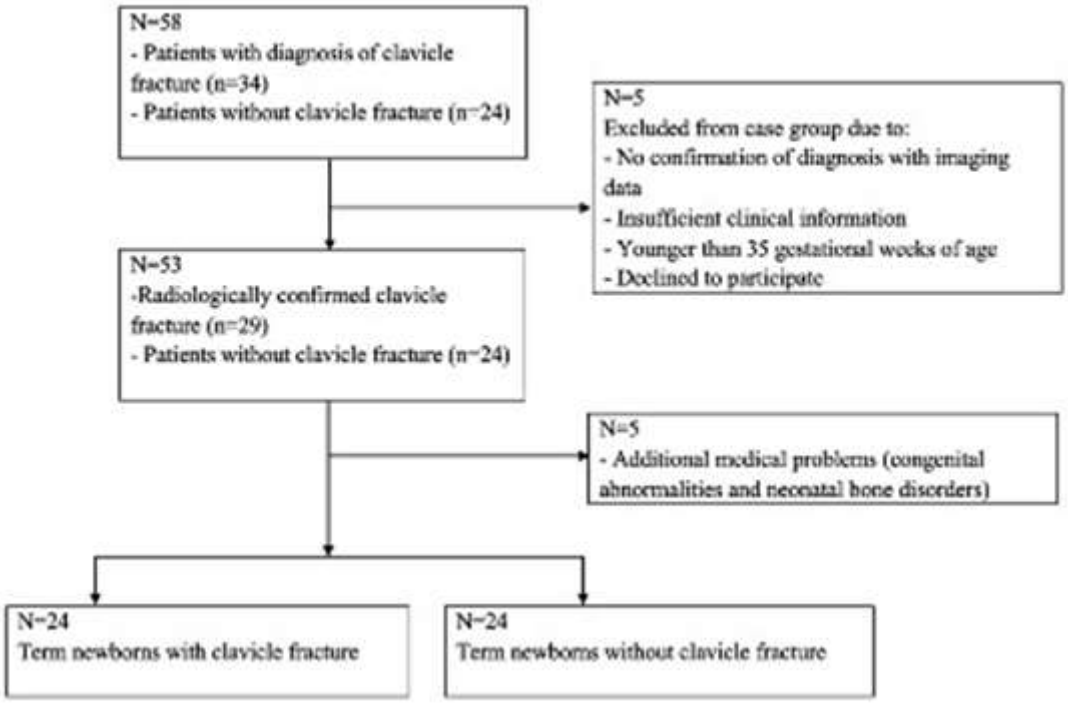

Figure 1. Flow chart of neonatol clavicle fracture

The maternal age of the infants with CF was significantly younger than the control group ($p<0,05$) (Figure 2 and Table 1). Serum calcium levels were significantly lower in infants with CF than the healthy controls ($p<0,05$) (Figure 3 and Table 2). The number of pregnancies ranged from 1to 5 in the CF group with an average of 2,52 $\pm$ 1 SEM. In the control group number of pregnancies ranged from 1 to 4 with an average of 1,792 $\pm$ 0,18 SEM. So, the parity rate did not differ between the groups.

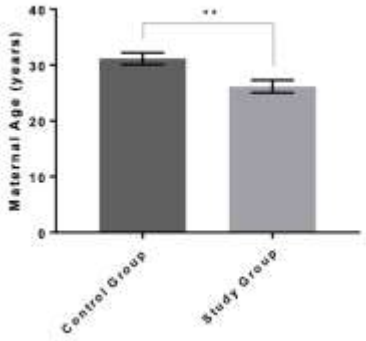

Figure 2. Comparison of maternal age between newborn with clavicle fracture and the control group.

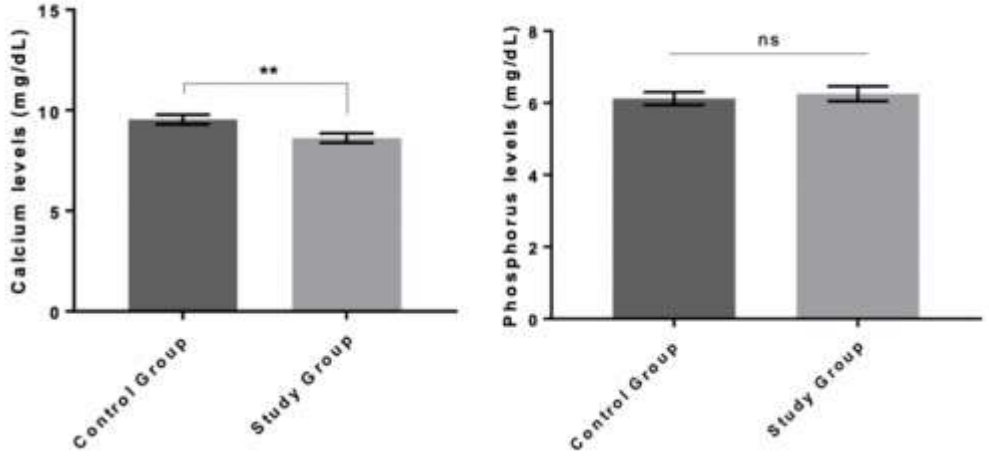

Figure 3. Comparison of serum calcium and phosphorus levels between newborn with clavicle fracture and the control group.

The groups did not differ in terms of the birth weight of infants (Table 2). The birth weights of infants ranged from 2900 g to 4500 g in the case group with an average of 3789 g ± 85,25 SEM. In the control group birth weight ranged from 2495 g to 4800 g with an average of 3548 g ± 118 SEM. Clinical characteristics of newborns with clavicle fracture at the time of diagnosis were shown in table 3 (Table 3). Apgar scores were not different between the groups. The first minute Apgar scores ranged from 4 to 9 in the patient group with an average of 7,05 ± 0,38 SEM. In the control group first minute Apgar scores ranged from 5 to 9 with an average of 7,95 ± 0,21 SEM. The fifth minute Apgar scores ranged from 5 to 9 in the patient group with an average of 8,47 ± 0,23 SEM. In the control group fifth minute Apgar scores ranged from 9 to 10 with an average of 9 ± 0,10 SEM. The mean vitamin D concentrations within the serum of fractured and unfractured newborns were 8,8 and 12,3 ng/mL, respectively. Vitamin D levels were significantly lower ($P = 0.047$) in the CF group compared to healthy newborns (Figure 4 and Table 1). Serum phosphorus and alkaline phosphatase levels were not significantly different between patients and clavicle fracture (p=0,63, p=0,53, respectively) (Table 4).

Table 4. Calcium, phosphorus and alkaline phosphatase comparison between the two groups

	Clavicle Fracture (-) (n=24) Mean±SEM	**Clavicle Fracture (+) (n=24) Mean±SEM**	**P**
Calcium	9.54±0.23 mg/dL	8,61±0.23 mg/dL	**0.008**
Phosphorus	6.12±0.18 mg/dL	6.26±0.20 mg/dL	0.63
Alkaline Phosphatase	186.2±12.86 U/L	197.5±12.38 U/L	0.53

SEM: Standard error of the mean

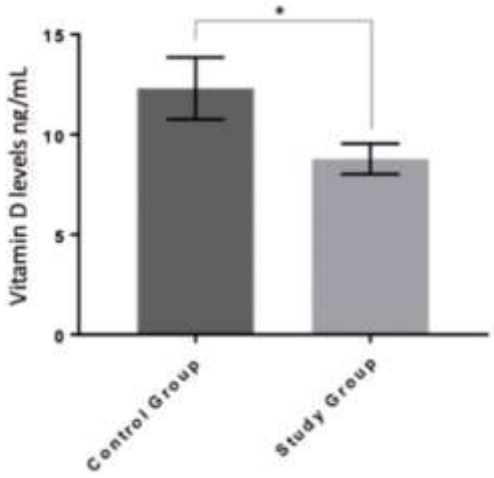

Figure 4. Comparison of serum vitamin D levels between newborn with clavicle fracture and the control group.

DISCUSSION

Vitamin D is known to be crucial for a healthy growing skeleton, but whether vitamin D is associated with CF risk in newborns yet unclear. To our knowledge, this study was the first to evaluate the association between serum levels of vitamin D and the risk of CF in a Turkish newborn cohort.

Vitamin D is essential in enhancing the bone health of the newborn. A wealth of literature on this subject released the association between 25(OH)D levels and fracture risk in pediatric population of all ages, races, and with a variety of fractures[4,6,11,12]. Pediatric fractures were found to be strongly related to their serum vitamin D levels in a study by Olney et al[13]. In that study, children who had lower levels of serum vitamin D had a higher risk of fracture due to the impaired bone mineral density (BMD)[13]. Maternal and neonatal vitamin D deficiency is a serious and common issue in developing countries including Turkey. The incidence of neonatal vitamin D deficiency is reported as 97,2% in a recent study from Turkey[14]. Therefore, the lower rate of vitamin D levels in the control group can be associated with the study population localized in a rural region of Turkey. Even though a high prevalence of vitamin D deficiency, ranging from 8% to 59%, was stated among children with fractures, none of these studies included a control group of children without fractures[11,12]. Since the studies were involved in different age groups, had seasonal and geographical differences, results cannot be generalized as some of them did not specify the skin type. A similar study revealed that the CFs were associated with poor vitamin D intake in children younger than six years and normal serum levels of vitamin D reduced the risk of CF by about 58%[5]. Nevertheless, though not conclusive, higher levels of vitamin D were suggested to reduce the risk of bone fractures[5]. However, there is no data regarding serum vitamin D levels in newborn with CF. In our study comparing the data presented in figure 2, it is evident that those newborns who had CFs had lower serum vitamin D levels. Hence, practitioners should include the deficiency of vitamin D in the differential diagnosis of bone fractures[15]. In contrast, Contreras et al. found no relationship between vitamin D deficiency and the risk of bone fracture in their study population[16]. But this study included only the cases who were admitted to the emergency department of a single center which limits the ability to generalize the results[16]. A Mendelian randomization study reported no beneficial effect of vitamin D on fracture, but the study was conducted in a population with lower overall fracture risk and the data were not sufficiently reliable[17]. The diagnosis of CFs is generally made by detecting tenderness around the clavicle region and reduced arm movements. Most of our patients (91.6%) were diagnosed on the first day. Only 2 of

our patients were diagnosed on the 4th and 6th day after discharge. The newborn with CF may have minimal symptoms or signs, and the diagnosis can be retrospective with palpation of the callus in the second week of life[6]. Also, diagnosis of CF can be delayed or overlooked in especially asymptomatic newborns at birth[18]. Given the physical examinations of the infants with delayed diagnosis, were found to be normal during the hospital stay, crepitation at the affected site and skin discoloration led to the diagnosis at physical examination as late findings of the fracture following discharge. Therefore, we suggest that cautious palpation of the clavicle region should be performed and noted before the hospital discharge. Whether clavicles cannot be palpated easily and/or crepitus is felt over, the infant may have a CF. Birth weight is reported as an important factor for CF in newborns. For example, a birth weight of more than 4,000 g was pointed as an independent risk factor for clavicle fracture[19]. Unlike previous studies, the mean birth weight was higher in infants with CF than in the control group, but the difference was not statistically significant in this study. This can be explained by the limited number of participants either the ones with birth trauma were excluded. There are studies showing that CF is associated with maternal parity. However, the results of studies in the literature vary. In the study of Brown et al., the risk of developing neonatal CF in nulliparous patients was found to be higher than in multiparas[20]. Conversely, other researchers found that multiparity is a significant risk factor for CF[20]. Similar to our study, no difference was found between the CF and parity status of the mother in the study of Mehta et al[21]. There was no association between the CF and parity in this study. We suggest that this result of our study is due to the low number of cases since we exclude those with birth trauma. Vitamin D helps the body of the newborn absorb sufficient calcium for the ossification of bones such as the clavicle. Comparing the calcium levels for the newborns with CF and the control group, the former exhibits a lower calcium absorption rate of about 8.6 at an average. On the contrary, the control group indicates higher serum calcium levels. However, this difference was not found to be statistically significant. The serum level of vitamin D in the newborn determines his/her calcium absorption from bone and/or intestine. Therefore, it is plausible that lower calcium levels contribute to weakened bones which enhances the risk of CF. However, since the serum calcium level in the newborn in the first days of life is related to the calcium of the mother, it would not be correct to make a clear comment on this issue. The serum calcium level in the first days of life may not be an accurate marker because of the calcium passed from the mother to the fetus. An important condition in which vitamin D insufficiency is more central to the etiology of hypocalcemia are cases mostly reported in newborns after the first week of life[22]. Since the calcium level was examined only once in our study, all these variables should be considered when evaluating the results. Since serum calcium levels are measured after the fracture detected in cases, we do not know exactly how dynamic changes affect their serum calcium. In the literature, studies on adult patients shown that serum calcium levels are elevated after fractures. This can be explained by bone resorption and decreased bone formation[23,24]. The results of studies showing the relationship between phosphorus level and bone fractures are contradictory[25-28]. There are no studies on newborns. In our study, no statistical difference was found between the serum phosphorus levels between the groups. Phosphorus is the main anion in the cell. In cases of hemolysis, the serum phosphorus level is measured high. Blood collection from newborns is difficult and likely to be hemolyzed. Therefore, it would not be accurate to assess serum phosphorus levels alone.

Our study had several limitations to be noted. First, the calcium level was measured only once at admission, and this may not be representative of each individual's baseline status. Second, ionized calcium and parathyroid hormone measurements were not available to determine the underlying mechanism of lower calcium levels in our study. On the other hand, the maternal serum vitamin D, calcium and parathyroid hormone measurements were not included. Particularly for the infants with delayed diagnosis, breastfeeding status was not involved in this study. Vitamin D sources in newborns depend on transplacental stores and breast milk. Maternal level of vitamin D is fundemental determinant for the vitamin D supply either at birth and early infancy. Therefore, maternal vitamin D deficiency during pregnancy and lactation is proven to act in the patogenesis of vitamin D deficiency in newborn infants[29,30].

This study revealed that there is an association between the CF and serum vitamin D level in new

born. Further studies are needed to elucidate the cause-and-effect relationship of the serum vitamin D levels and CF in newborn.

Yazar Katkıları: Çalışma konsepti/Tasarımı: EA, ST, SYS; Veri toplama: EA; Veri analizi ve yorumlama: EA, ST, SYS; Yazı taslağı: EA, ST; İçeriğin eleştirel incelenmesi: EA, ST, SYS; Son onay ve sorumluluk: EA, ST, SYS; Teknik ve malzeme desteği: EA,ST, SYS; Süpervizyon: EA, SYS; Fon sağlama (mevcut ise): yok.
Etik Onay: For this study, a decision was taken by the Ethics Committee of Tekirdag Namık Kemal University Rectorate Faculty of Medicine Deanery of Non-Interventional Clinical Research dated 29.12.2020 and research protocol number 2020.273.12.18.
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Author Contributions: Concept/Design : EA, ST, SYS; Data acquisition: EA; Data analysis and interpretation: EA, ST, SYS; Drafting manuscript: EA, ST; Critical revision of manuscript: EA, ST, SYS; Final approval and accountability: EA, ST, SYS; Technical or material support: EA, ST, SYS; Supervision: EA, SYS; Securing funding (if available): n/a.
Ethical Approval: For this study, a decision was taken by the Ethics Committee of Tekirdag Namık Kemal University Rectorate Faculty of Medicine Deanery of Non-Interventional Clinical Research dated 29.12.2020 and research protocol number 2020.273.12.18.
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