Journal of Tropical Pediatrics, 2019, 0, 1–8
doi: 10.1093/tropej/fmz021
Original paper

Role of Vitamin-D Deficiency in Term Neonates with Late-Onset Sepsis: A Case–Control Study

Amit Agrawal , MD, Aekta Gupta, MD *and* Jyotsna Shrivastava, MD

Department of Pediatrics, Gandhi Medical College, Hamidia Hospital, Bhopal, Madhya Pradesh 462030, India
Correspondence: Jyotsna Shrivastava, Department of Pediatrics, Kamla Nehru Hospital, Gandhi Medical College, Hamidia Hospital, 3rd Floor, Bhopal, Madhya Pradesh 462030, India. Tel: +91-9826023745. E-mail <shrivastav_rajendra@rediffmail.com>.

ABSTRACT

Objective: To find the relationship between vitamin-D levels and late-onset sepsis (LOS) in term neonates.
Methods: This case–control study was conducted in neonatal intensive care unit of a tertiary care teaching institution in central India. Full-term neonates with culture-proven LOS were taken as cases. Maternal and neonatal demography, clinical examination and investigations were recorded. Correlation of vitamin-D deficiency (<20 ng/ml) with LOS was assessed.
Results: Total 225 term neonates including 175 cases and 50 controls were included. Maternal and neonatal demographic profile was comparable. The mean vitamin-D level in cases (12.28 ± 6.11 ng/ml) was significantly lower than that in controls (14.88 ± 7.2 ng/ml) ($p = 0.002$). Total 151 (86.29%) neonates out of 175 cases and 37 (74%) out of 50 controls had the vitamin-D deficiency ($p = 0.00003$). On multiple regression analysis, neonatal sepsis ($p = 0.00003$) was found to be significantly associated with vitamin-D deficiency.
Conclusion: This study shows that vitamin-D deficiency in term neonates may predispose them to LOS.

KEYWORDS: neonate, vitamin-D, sepsis, late-onset sepsis

INTRODUCTION

Neonatal sepsis is a clinical syndrome characterized by signs and symptoms of infection with or without accompanying bacteremia in the first month of life. It encompasses various systemic infections of the newborn such as septicemia, meningitis, pneumonia, arthritis, osteomyelitis and urinary tract infections. The incidence of neonatal sepsis varies from 1 to 8 neonates per 1000 live births accounting for about 1 million or 26% of neonatal deaths, worldwide [1]. The incidence of neonatal sepsis in India is 38 per 1000 live births according to the data from National Neonatal Perinatal Database, 2014 [2]. Neonatal sepsis can be classified into early-onset sepsis (EOS) and late-onset sepsis (LOS). LOS occurs after 72 h of life [3].

Vitamin-D is a fat-soluble, steroid hormone which has well-known effects on bone metabolism. Recent studies have shown that vitamin-D also plays an important role in a wide variety of non-skeletal

conditions including cardiovascular diseases, insulin resistance, metabolic syndrome, allergy, autoimmune disorders and certain types of cancers [4–6]. Early vitamin-D deficiency has been shown to be associated with increased risk for fetal growth restriction, infections and poor neurodevelopment in later life [7, 8]. Studies have suggested that vitamin-D has effects on the functioning of the innate immune system by inducing antimicrobial peptides in epithelial cells, neutrophils and macrophages [8]. The epidemiological evidence indicates that low cord blood vitamin-D levels correlate with the increased susceptibility to infections in the newborns [9].

Recently, a few studies have clearly shown that the lower levels of vitamin-D increase the risk of early-onset neonatal sepsis in term infants [10–12]. There is a lack of sufficient data defining the role of vitamin-D in LOS and only a few studies have investigated the role of vitamin-D in neonates with LOS [13]. Therefore, we planned this study to find out the relationship between vitamin-D levels and LOS in term neonates.

MATERIALS AND METHODS

This case–control study was conducted in a neonatal intensive care unit (NICU) of a tertiary care teaching institution in central India from March 2016 to February 2017. Prior approval from the institutional ethical committee was obtained (14061-63/MC/IEC/2015 dated 17/06/2015).

Full-term neonates (gestational age >37 completed weeks) admitted to NICU with culture-proven sepsis were taken as cases. Only those neonates who presented between 72 h and 21 days of age and had blood culture-proven LOS were included as cases. The controls were full-term, healthy neonates delivered by cesarean section, aged 72 h–21 days and who stayed with their mother in the postnatal ward due to various maternal reasons. The sample size was calculated using the formula $4pq/E^2$ and the minimum recommended sample size was 175 cases and 50 controls. We could not take controls in a 1:1 ratio due to the financial constraints as the facility for vitamin-D assessment was not available in our institution. After explaining the nature, procedure and purpose of the study, written consent was obtained from the parents/legal guardians of the case and controls.

The exclusion criteria were (i) presence of co-morbidities like perinatal asphyxia, hyaline membrane disease, transient tachypnea of newborn, prematurity or post-maturity. (ii) Major congenital anomalies. (iii) Neonates with EOS or presence of chorioamnionitis/premature rupture of the membrane in mothers. (iv) Neonates of mothers who have received vitamin-D supplementation during pregnancy or neonates who have received vitamin-D supplementation after birth. (v) Neonates receiving formula feeding.

Quantitative estimation of vitamin-D was done by Maglumi fully auto chemiluminescence immunoassay analyzer. The results were expressed in ng/ml. Serum 25-hydroxyvitamin-D (25-OH-D) level was assessed in all the recruited subjects. Serum 25-OH-D levels <15 ng/ml was considered as a deficiency and >20 ng/ml as sufficiency, while severe deficiency was defined as a level <5 ng/ml [14].

The data obtained from the subjects was recorded in a pre-structured proforma. Maternal data including maternal age, religion, socio-economic status, last menstrual period, risk factors and drug intake were obtained from the mother/legal guardians of the baby and from the medical records of the mother. A detailed natal and postnatal history including age at admission, gestational age, gender, type of feeding, pre-lacteal feeds and presenting complaints of the neonates was also obtained. The thorough physical examination was done and the heart rate, respiratory rate, temperature, capillary refill time, color, saturation and systemic examination findings were noted for all the recruited neonates. The gestational age was assessed from the last menstrual period and from the New Ballard Score [15].

Relevant investigations were sent for all the neonates as per the hospital policy including hemoglobin, total leukocyte count, direct leukocyte count, C-reactive protein (CRP), blood culture, electrolytes and random blood sugar. Other investigations like chest X-ray, lumbar puncture, urine routine and microscopy, urine for fungal hyphae, urine culture and arterial blood gas analysis were done as and when required. The main outcome measure was the serum 25-OH-D levels in cases and controls. Correlation of vitamin-D deficiency (<20 ng/ml) with various factors was assessed.

Statistical analysis

Continuous variables were presented as mean for parametric data and as median for non-parametric or skewed data. Categorical variables were expressed as frequencies and percentages. The Student's '*t*'-test was applied to calculate the statistical significance of the data following normative distribution. The Mann–Whitney test was applied whenever data followed a non-normative distribution. Categorical data between the groups were compared using the Chi-square test or Fisher's exact test as appropriate. *p*-Value <0.05 was taken to indicate a statistically significant difference. Spearman correlation was used for establishing correlation wherever required. Multivariate regression analysis was done for analyzing multiple categorical variants. All the statistical analysis was performed using SPSS version 20 (IBM).

RESULTS

The study population consisted of a total 225 full-term neonates; out of whom 175 were cases and 50 were taken as controls as shown in the study flow-chart (Fig. 1). Out of 175 cases, 81 were males and 94 were females with a male:female ratio of 0.86:1. Out of 50 controls, 27 and 23 were males and females, respectively, with a male to female ratio of 1.17. Although all the controls were inborn, most of the cases (113, 64.57%) were outborn.

The mean age of cases was 9.5 ± 5.03 days; of these, 47.4% presented with sepsis within 72 h–7 days. The mean age of control was 6.74 ± 0.92 days. The mean weight of cases was 2.441 ± 0.67 kg and of controls was 2.630 ± 0.51 kg ($p = 0.066$). The median maternal age in the study group was 24 years [interquartile range (IQR) 21–27 years] and the control group was 24 years (IQR 22–27 years). The maximum number of mothers belonged to 20–25 years of age in both groups. As shown in Table 1, maternal and neonatal demographic factors were comparable.

Total 188 out of 225 neonates had a vitamin-D deficiency (<20 ng/ml) giving a high incidence (81%) of neonatal vitamin-D deficiency. One hundred and fifty-one (86.28%) out of 175 cases and 37 (74%) neonates out of 50 controls had a vitamin-D deficiency. The cases (12.28 ± 6.11 ng/ml) had significantly lower vitamin-D levels as than the controls (14.88 ± 7.2 ng/ml) ($p = 0.002$). In the present study, 36 cases (20.57%) had severely vitamin-D deficiency (<5 ng/ml), 86 (49.14%) cases had deficiency (between 5 and 15 ng/ml), 29 (16.57%) cases had insufficiency (between 15 and 20 ng/ml) and only 24 (13.7%) had vitamin-D levels of >20 ng/ml. Out of the 50 controls, 7 (14%) were severely deficient, 15 (30%) were deficient, 15 (30%) had insufficiency and 13 (26%) had vitamin-D sufficiency.

The most common presenting complaint of recruited cases was respiratory distress (34.9%), followed by lethargy (18.9%) and not accepting feeds (8.1%) and the most common system involved was the respiratory system (29.1%). The most common organism grown in blood culture was *E. coli* (57, 32.5%), followed by *Klebsiella* (42, 24%), *Pseudomonas* (23, 13.14%), Group B streptococcus (17, 9.7%), *Staphylococcus aureus* (11, 6.2%) and Coagulase negative *Staphylococcus* (8, 4.5%). Twelve percent of the cases had culture-proven meningitis and the most common organisms grown in CSF cultures were *E. coli* (6.2%) followed by *Klebsiella* (1.7%) and CONS (1.7%).

In our study, 91% of the neonates were CRP positive; however, no significant correlation was found between CRP and vitamin-D levels. Similarly, vitamin-D deficiency has no correlation with the bacteriological culture profile of cases. Also, no significant difference in vitamin-D levels was found between Gram-positive (13.47 ± 10.04 ng/ml) and Gram-negative (12.35 ± 6.17 ng/ml) sepsis ($p = 0.579$).

Multivariate regression analysis was done to assess the association of vitamin-D deficiency with covariables like age, sex, weight, maternal age, sepsis and religion (Table 2). Only sepsis and religion (or sun exposure) were found to have a strong association with vitamin-D levels ($p = 0.00463$ and 0.00037, respectively). Significantly ($p = 0.002$) higher cases (85, 48.57%) belonging to the Muslim community had vitamin-D deficiency than the controls (19, 38%) from the same community. The mean vitamin-D level of the cases (10.61 ± 5.43 ng/ml, $n = 93$) was significantly lower ($p = 0.001$) than the controls (14.94 ± 8.10 ng/ml, $n = 28$) belonging to the Muslim community. This implies that though

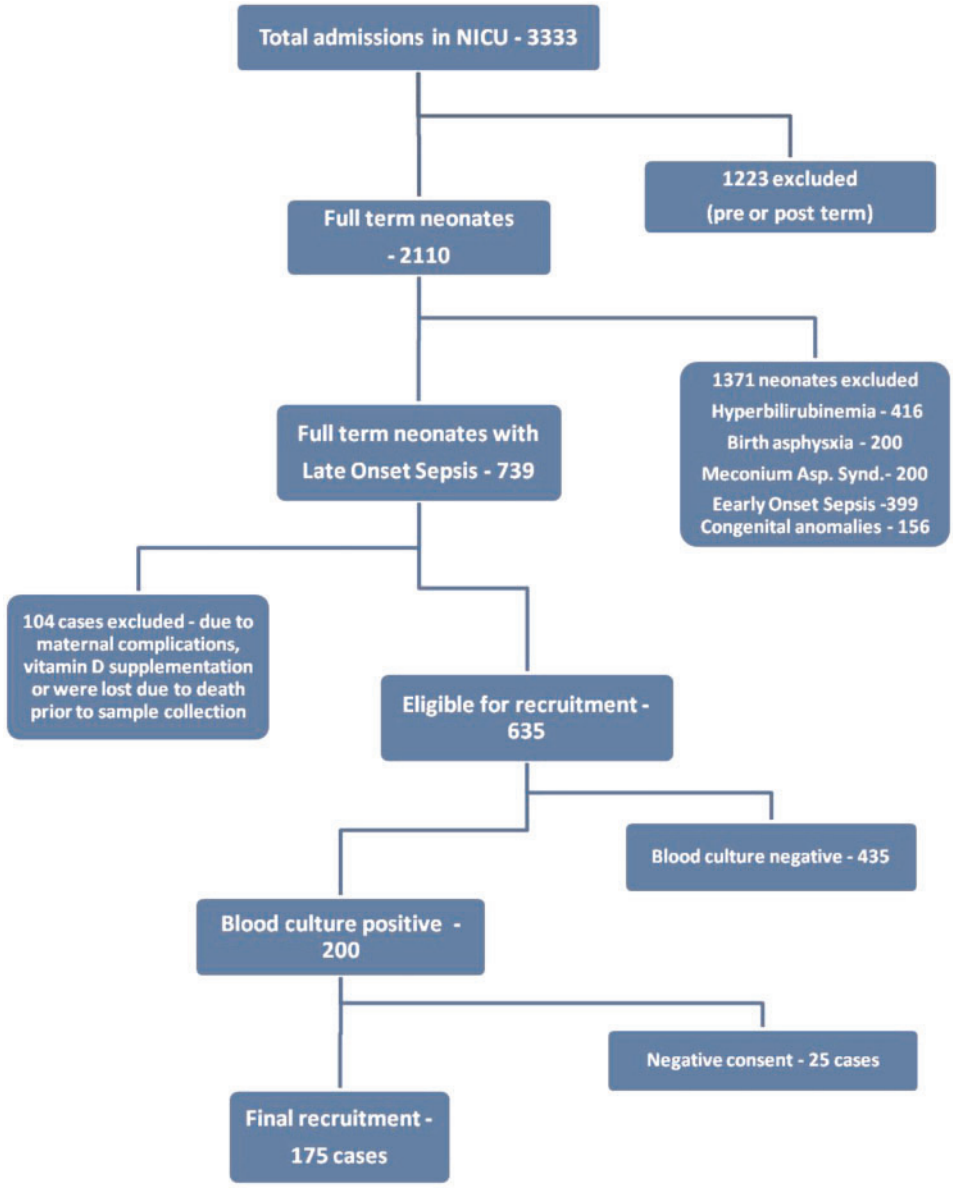

Fig. 1. Study flowchart.

religion was related to vitamin-D level, sepsis was a strong co-variant influencing vitamin-D level in this community.

DISCUSSION

In this study, we had shown that the vitamin-D was significantly lower in the neonates with LOS than in the healthy controls suggesting a possible role of vitamin-D deficiency in the pathogenesis of LOS. We could not assess the maternal vitamin-D levels of the recruited neonates due to logistic problems. However, we had excluded neonates presenting with EOS so as to rule out the impact of maternal/perinatal factors on the development of sepsis and investigate the independent association of vitamin-D levels with sepsis. Second, maternal vitamin-D

Table 1. Demographic profile of the study population

Demographic factors	Cases ($n = 175$), *N* (%)	Control ($n = 50$), *N* (%)	Total ($n = 225$), *N* (%)	*p*-Value
Maternal factors				
Residence				
Rural	104 (59.43)	22 (44.00)	126 (56.00)	0.075
Urban	71 (40.57)	28 (56.00)	99 (44.00)	
Maternal age (years)				
<20	25 (14.29)	5 (10.00)	30 (13.33)	0.842
20–25	85 (48.57)	26 (52.00)	111 (49.33)	
25–30	56 (32.00)	17 (34.00)	73 (32.44)	
>30	9 (5.14)	2 (4.00)	11 (4.89)	
Religion				
Hindu	81 (46.30)	21 (42.00)	102 (45.30)	0.632
Muslim	94 (53.10)	29 (56.00)	121 (53.70)	
Presence of maternal risk factors				
Antepartum hemorrhage	7 (4.00)	2 (4.00)	9 (4.00)	0.419
Pre-eclampsia	16 (9.10)	3 (4.00)	19 (7.40)	
Diabetes mellitus	3 (1.60)	1 (2.00)	3 (2.10)	
Severe anemia	6 (3.40)	1 (2.00)	7 (3.10)	
None	123 (70.10)	45 (90.00)	168 (75.40)	
Neonatal factors				
Birth weight (kg)				
<2.5	132 (75.43)	32 (64.00)	150 (66.67)	0.148
≥2.5	43 (24.57)	18 (36.00)	75 (33.33)	
Age at admission (days)				
<7	83 (47.43)	32 (64.00)	125 (55.55)	0.054
>7	92 (52.57)	18 (36.00)	100 (44.46)	

Table 2. Multivariate linear regression for vitamin-D levels in sepsis

Model	Standardized coefficients	*T*	Sig. (*p*-value)	95% Confidence interval for B	
	Beta			Lower bound	Upper bound
Age	0.057	0.836	0.404	−0.103	0.256
Gender	−0.070	−1.080	0.281	−2.462	0.719
Residence	0.062	0.950	0.343	−0.832	2.382
Maternal age	0.061	0.998	0.344	−0.110	0.315
Religion	−0.208	−3.217	**0.001**	−4.157	−0.999
Weight	−0.121	−1.606	0.110	−4.042	0.412
Sepsis	−0.287	−3.754	**0.000**	−6.508	−2.027

deficiency is common in our country and studies have shown a linear correlation between maternal and neonatal vitamin-D levels [16–18]. Therefore, it would not have affected our study objective as other maternal demographic factors were comparable in both groups.

25-OH-D induces more than 2000 genes, many of which have a role in fetal development [19].

Therefore, 1,25(OH)2 D may be relevant to the 'fetal programing hypothesis' in which environmental factors such as 1,25(OH)2 D influence the genomic programing of the fetus and neonate and influence disease risk in later life. It has been experimentally proven that 1,25-(OH)D3 alone or in conjunction with lipopolysaccharide can induce antimicrobial peptide gene expression through toll like receptors (TLR) and cause the release of antimicrobial activity in neonates [20]. Recent experimental studies have also shown the diminished TLR induced expression of cathelicidin activity by the monocytes cultured from vitamin-D deficient plasma [9]. Another study has found significantly high cathelicidin and low 25-OH-D levels in neonates with congenital pneumonia suggesting a possible role of fetal 25-OH-D deficiency as a predisposing factor for congenital pneumonia [21]. In addition to the systemic inflammatory response modulation, vitamin-D also has a role in the local control of pathogens as it has been reported to inhibit the growth of or killed strains of *S. aureus*, *Streptococcus pyogenes*, *Klebsiella pneumonia* and *E. coli* [22].

Recently, few studies have shown the association of vitamin-D deficiency with EOS in neonates [10–12]. First time in 2015, Cetinkya *et al.* had shown that maternal and neonatal vitamin-D levels were significantly lower in neonates with EOS ($p < 0.001$). In their study, the mean neonatal vitamin-D levels were 8.6 ± 3.1 and 19.0 ± 4.8 ng/dl in the study and control group, respectively [10]. Cizmeni *et al.*, from Turkey, found that cord blood vitamin-D levels of infants with EOS were significantly lower than the controls (median 12.6 vs. 21 ng/ml, $p = 0.038$). They have included a total of 83 (40 cases and 43 controls) full-term normal birth weight neonates [11]. These results are corroborative with the results of our study. Another recent study from China also showed that low serum level of 25-OH-D is associated with the development of EOS in full-term neonates [12].

However, there is a lack of sufficient data defining the role of vitamin-D in LOS and only a few authors have studied the association of vitamin-D in neonates with LOS [13]. Dhandai *et al.* [13] conducted a prospective study in term and preterm neonates with LOS and found that neonates with vitamin-D deficiency are at greater risk of LOS than those with sufficient vitamin-D levels. In their study, cases had significantly lower mean vitamin-D levels (15.37 ± 10.0 ng/ml) than the control group (21.37 ± 9.53 ng/ml) ($p = 0.001$).

In the present study, most of the subjects (i.e. 83.5%) were found to be vitamin-D deficient. In a study done by Sachan *et al.* [18] on pregnant women and neonates in northern India, 95.7% neonates were found to have hypovitaminosis D (vitamin-D < 20 ng/ml) with mean cord blood 25-OH-D level of 8.4 ± 5.7 ng/ml. In the present study, it was observed that significantly higher cases (86.29% vs. 74%) had vitamin-D deficiency than the controls ($p = 0.0003$). Similar results were observed in other studies also [10–12]. In the present study, 36 cases (20.57%) had severely vitamin-D deficiency (<5 ng/ml), 86 (49.14%) cases had deficiency (between 5 and 15 ng/ml), 29 (16.57%) cases had insufficiency (between 15 and 20 ng/ml) and only 24 (13.7%) had vitamin-D levels of >20 ng/ml. Out of the 50 controls, 7 (14%) were severely deficient, 15 (30%) were deficient, 15 (30%) had insufficiency and 13 (26%) had vitamin-D sufficiency.

On multivariate analysis, only sepsis and religion of the recruited patients were found to have a strong correlation with vitamin-D deficiency. Here, we would like to highlight one fact that the majority of the Muslim women were wearing the Burka; so, this effect essentially denotes the effect of less sun exposure in these women. Collecting information on the duration of sun exposure would have been more appropriate; however, we did not collect the information on the duration of sun exposure and it was a limitation of our study. Still, the religion was comparable in both the groups with 53.1% and 56% of Muslim patients being in cases and controls, respectively. The mean vitamin-D level in neonate belonging to Muslim (10.76 ± 7.37 ng/ml) was significantly lower than those belonging to the Hindu community (14.77 ± 7.37 ng/ml) ($p = 0.000056$). This finding can be attributed to the lesser duration of sun exposure in mothers of this community because of their customs and clothing; thus leading to decreased vitamin-D levels in the females and hence their neonates. On further subgroup analysis, mean vitamin-D levels of cases from

the Muslim community (10.60 ± 5.43) was significantly lower than that in controls (14.94 ± 8.09) from the same community ($p = 0.001$). By keeping this in mind, it can be concluded that LOS has a positive association with vitamin-D deficiency; though, religion also influences the vitamin-D levels. Our inclusion of neonates with LOS removed the influence of maternal factors on the occurrence of sepsis and thus established vitamin-D deficiency as an independent risk factor to be investigated in the development of neonatal sepsis.

In our study, the mean vitamin-D level of the urban population (12.35 ± 6.66 ng/ml) was not significantly lower ($p = 0.464$) than that of the rural population (12.96 ± 5.8 ng/ml). This is in contrast to previous Indian studies where the vitamin-D status of the urban population was found to be lower than the rural population which is attributable to the dress code, occupation and to the lesser duration of exposure to sunlight [23, 24]. However, this is similar to the results of a study done by Sachan *et al.* [18], where they did not find a significant difference between vitamin-D levels of the urban and rural population. This can be explained by the poor dietary calcium intake among poorer rural parts of India leading to a vitamin-D deficiency in them similar to their urban counterparts. Adequate sun exposure and supplementation are known sources of vitamin-D. These, however, must be persuasively stressed on in the pre-conceptional and perinatal period by health care practitioners to reinforce community awareness and to decrease vitamin-D deficiency in mothers and their children.

Study limitations

The present study had certain limitations, i.e. the data on maternal 25-OH-D levels could not be collected due to financial constraints. This would have been more helpful and would have given the information about the requirement of vitamin-D supplementation during pregnancy and the correlation between maternal and neonatal vitamin-D levels. We did not collect the information on the duration of sun exposure and it was also a limitation of our study. Further randomized controlled trials are required to evaluate the role of vitamin-D in the prevention of sepsis in neonates.

CONCLUSION

In this study, we found that the neonates with vitamin-D deficiency have more chances of development of LOS than the neonates having vitamin-D sufficiency.

What is already known on this topic?

1. Low cord blood vitamin-D levels correlate with increased susceptibility to infections in newborns.
2. Early vitamin-D deficiency has been shown to increase the risk for fetal growth restriction and infections.
3. Few recent studies have shown that the lower vitamin-D levels increase the risk of early-onset neonatal sepsis in term infants.

What does this study add?

1. In this study, we assessed the vitamin-D levels in term neonates with LOS.
2. Serum vitamin-D levels were significantly lower in the term neonates with LOS than in the healthy controls suggesting a possible role of vitamin-D deficiency in the pathogenesis of LOS.

REFERENCES

1. Lawn JE, Cousens S, Zupan J; For the Lancet Neonatal Survival Steering Team. Neonatal survival 4 million neonatal deaths: when? Where? Why? Lancet 2005;365: 891–900.
2. National Neonatal Perinatal Database 2002-2003. www.newbornwhocc.org (20 Feb 2016, date last accessed).
3. Puopolo KM. Bacterial and fungal infections. In: Cloherty JP, Eichenwald EC, Hansen AR, *et al.* (eds). Manual of Neonatal Care. 7th edn. Philadelphia, PA, USA: Lippincott Williams & Wilkins, 2012, 624–655.
4. Camargo CA, Jr, Ingham T, Wickens K, *et al.* Cord blood 25hydroxyvitamin-D levels and risk of respiratory infection, wheezing, and asthma. Pediatrics 2011;127:180–7.
5. Shin YH, Shin HJ, Lee YJ. Vitamin-D status and childhood health. Korean J Pediatr 2013;56:417–23.
6. Liu PT, Stenger S, Li H, *et al.* Toll-like receptor triggering of a vitamin-D mediated human antimicrobial response. Science 2006;311:1770–3.
7. Thorne-Lyman A, Fawzi WW. Vitamin D during pregnancy and maternal, neonatal and infant health outcomes: a systematic review and meta-analysis. Paediatr Perinat Epidemiol 2012;26(Suppl 1):75–90.

8. Strom M, Halldorsson TI, Hansen S, *et al*. Vitamin-D measured in maternal serum and offspring neurodevelopmental outcomes: a prospective study with long-term follow-up. Ann Nutr Metab 2014;64:254–61.
9. Walker VP, Zhang X, Rastegar I, *et al*. Cord blood vitamin-D status impacts innate immune responses. J Clin Endocrinol Metab 2011;96:1835–43.
10. Cetinkaya M, Cekmez F, Buyukkale G, *et al*. Lower vitamin-D levels are associated with increased risk of early-onset neonatal sepsis in term infants. J Perinatol 2015;35: 39–45.
11. Cizmeci MN, Kanburoglu MK, Akelma AZ, *et al*. Cord-blood 25-hydroxyvitamin-D levels and risk of early-onset neonatal sepsis: a case-control study from a tertiary care center in Turkey. Eur J Pediatr 2015;174:809–15.
12. Yang LR, Li H, Yang TY, *et al*. Relationship between vitamin-D deficiency and early-onset neonatal sepsis. Zhongguo Dang Dai Er Ke Za Zhi 2016;18:791–5. [Article in Chinese].
13. Dhandai R, Jajoo M, Singh A, *et al*. Association of vitamin-D deficiency with an increased risk of late-onset neonatal sepsis. Paediatr Int Child Health 2018;38:193–7.
14. Misra M, Pacaud D, Petryk A, *et al*. Vitamin-D deficiency in children and its management: a review of current knowledge and recommendations. Pediatrics 2008;122: 398–417.
15. Ballard JL, Khoury JC, Wedig k, *et al*. New Ballard score, expanded to include extremely premature infants. J Pediatr 1991;119:417–23.
16. Kumar P, Shenoi A, Kumar RK, *et al*. Vitamin-D deficiency among women in labor and cord blood of newborns. Indian Pediatr 2015;52:530–1.
17. Hollis BW, Johnson D, Hulsey TC, *et al*. Vitamin-D supplementation during pregnancy: a double-blind, randomized clinical trial of safety and effectiveness. J Bone Miner Res 2011;26:2341–57.
18. Sachan A, Gupta R, Das V, *et al*. High prevalence of vitamin-D deficiency among pregnant women and their newborns in northern India. Am J Clin Nutr 2005;81:1060–4.
19. Adams JS, Hewison M. Update in vitamin-D. J Clin Endocrinol Metab 2010;95:471–8.
20. Wang TT, Nestel FP, Bourdeau V, *et al*. Cutting edge: 1,25-dihydroxyvitamin-D_3 is a direct inducer of antimicrobial peptide gene expression. J Immunol 2004;173:2909–12.
21. Gad GI, Abushady NM, Fathi MS, *et al*. Diagnostic value of anti-microbial peptide, cathelicidin in congenital pneumonia. J Matern Fetal Neonatal Med 2015;28:2197–200.
22. Youssef DA, Miller CW, El-Abbassi AM, *et al*. Antimicrobial implications of vitamin-D. Dermatoendocrinol 2011;3:220–9.
23. Harinarayan CV, Ramalakshmi T, Prasad UV, *et al*. High prevalence of low dietary calcium, high phytate consumption, and vitamin-D deficiency in healthy south Indians. Am J Clin Nutr 2007;85:1062–7.
24. Harinarayan CV, Ramalakshmi T, Venkataprasad U. High prevalence of low dietary calcium and low vitamin-D status in healthy south Indians. Asia Pac J Clin Nutr 2004;13: 359–64.