Seasonal and meteorological associations of vitamin K-dependent coagulation factors in 1-month-old infants: assessment of Normotest values

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Short communication

Seasonal and meteorological associations of vitamin K-dependent coagulation factors in 1-month-old infants: Assessment of Normotest values

Short Title: Seasonal variation in Normotest values

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Abstract

Objective: We aimed to determine the presence of seasonal and meteorological associations of the activity of vitamin K-dependent coagulation factors to explain the seasonal variation in vitamin K deficiency-related bleeding.

Methods: Seasonal and monthly changes in Normotest values in 1,759 healthy 1-month-old infants were retrospectively accessed, and the impact of meteorological parameters on Normotest values was analyzed.

Results: Normotest values peaked in winter and were the lowest in summer, with statistically significant differences among the seasonal values ($p<0.001$). Comparing monthly variations, the values peaked in January and were the lowest in August ($p<0.001$). Only the average daily air temperature significantly correlated with the Normotest values on multiple linear regression ($p<0.001$) and with low Normotest values on multiple logistic regression analysis (odds ratio, 1.023; $p=0.002$).

Conclusion: Seasonal and monthly variations in Normotest values were observed in 1-month-old infants, possibly due to fluctuations in daily air temperature.

Keywords:

Infant; Normotest; Seasonal variation; Temperature; Vitamin K deficiency bleeding

Introduction

Vitamin K deficiency bleeding (VKDB) is an acquired coagulopathy that develops in early infants who do not have sufficient vitamin K (VK) stores to support production of coagulation factors II, VII, IX, and X [1]. Especially in late VKDB, which occurs during 2–12 weeks after birth, bleeding predominantly occurs within the brain; the prevalence and mortality rates of intracranial hemorrhage are as high as 60–80% and 20–50%, respectively, and the associated morbidity is characterized by severe neurological disability [2,3].

Japan has a temperate climate with four unique seasons (spring, summer, autumn, and winter). A previous nationwide survey in Japan demonstrated that VKDB in early infancy develops more frequently in summer and early autumn; this was also the trend in the southern part of Japan [4-6]. However, physiological mechanisms explaining these variations are still poorly understood. In a previous study, we described seasonal variations in the international normalized ratio among neonates at 4 days after birth, and hypothesized that the differences were related to ambient temperature [7]. However, the role of VK-dependent coagulation factors remains unclear.

In Japan, oral VK is routinely administered thrice to every infant. A dose of 2 mg is administered on the day of birth, upon discharge from the maternity ward, and at 1-month after birth [8]. In addition, to detect a latent hemorrhagic state due to VK deficiency, screening with Normotest (NT; also known as hepaplastin test) is performed in limited regions among infants of approximately 1-month of age; this corresponds with the age of peak incidence of VKDB [4]. The NT system developed by Owren et al. [9] is used to measure VK-dependent coagulation factors II, VII, and X as part of the extrinsic coagulation pathway [4,10]; the value in normal adults is 100 ± 30% [9,11].

In the present study, we conducted a retrospective review of data from a large sample of infants, collected over a long time period, and analyzed the NT results to determine the presence of seasonal variations and the association between meteorological parameters and VK-dependent coagulation status in 1-month-old infants.

Methods

Subjects and data collection

A retrospective chart review was conducted among consecutive healthy infants who were born in the Hamamatsu University Hospital and underwent the routine 1-month check-up between January 1, 2012 and January 31, 2016. According to records, healthy infants born at ≥36 weeks of gestation with birth weight ≥2,300 g received care in the neonatal unit and were administered 2 mg of VK syrup (Menatetrenone Kaytwo Syrup; Eisai Co., Ltd., Tokyo, Japan) orally between 6–12 hours and 4 days after birth. To evaluate the NT values in healthy infants receiving routine VK prophylaxis, those with low birthweight (<2,500 g), macrosomia (≥4,000 g), those delivered preterm (<37 weeks) or post-term (≥42 weeks), and those admitted to the neonatal intensive care unit were excluded. Infants with underlying diseases likely to cause secondary VKDB and with mothers who had coagulation disorders and/or took medications likely to affect fetal coagulation were also excluded. Based on findings from previous studies [4,8,10,12], data on sex, gestational age, birth weight, nutrition, age in days at 1-month check-up, and body weight at 1-month check-up were obtained as plausible predictors of NT values.

The present study protocol was designed in accordance with the Declaration of Helsinki and was approved by the ethics committee of the Hamamatsu University School of Medicine (Approval No: 17-317). Written informed consent was obtained from the parents of each study participant via a signed consent form.

Blood sampling and Normotest measurement

Blood sampling and NT were performed as described in a previous study [13]. Capillary whole blood samples for NT were routinely obtained from a heel prick during the 1-month check-up. A single heel prick was performed, and the first drop of capillary blood obtained was collected in a 10-μL pipette for NT measurement. All blood samples were drawn by a single dedicated and experienced neonatologist (SI: corresponding author); two laboratory experts performed the NT measurement. All the evaluations began at 13:00, immediately after the infants arrived at the hospital, in an examination room at the pediatric outpatient clinic. NT was performed soon after sampling using a commercial kit, Hepaplastin Test (Eisai Co., Tokyo, Japan). After the 10 μL of whole blood samples were pipetted and mixed with the 250 μL of reagent, the clotting time was measured using a coagulometer (Thrombotrack Solo; Axis-Shield, Norway). The values of NT (%) were obtained using reference correlation curves prepared by diluting standard plasma for every lot of the kit.

Data collection of seasons and meteorological parameters

Summer was considered to range from June 1 to August 31, autumn from September 1 to November 30, winter from December 1 to February 28, and spring from March 1 to May 31. The meteorological data (air temperature, diurnal air temperature, change in mean temperature from the day before, relative humidity, barometric pressure, total precipitation amount, and sunshine duration) were obtained from the website of the Japan Meteorological Agency on the day the blood samples were taken. In addition, it is known that air temperatures usually peak at 13:00–14:00 throughout the year at Hamamatsu; this is when the NT values were measured in the present study. Therefore, the daily ambient temperature data collected included readings of maximum and mean air temperatures. These were measured at the Hamamatsu Local Meteorological Observatory, which is located approximately 1-mile from the hospital. Japan comprises a chain of long islands running from north to south, and Hamamatsu is a city located in the central region. The climate is relatively mild with an annual average temperature of 16–17°C.

Statistical analysis

The data have been presented as medians with interquartile range for continuous variables, and as counts and percentages for categorical variables. Non-parametric methods were used (Spearman correlation coefficient and Mann-Whitney U test) to assess the influence of participant characteristics on NT values. The Kruskal-Wallis test was used to compare clinical and meteorological variables according to different seasons and months. A linear regression model was constructed using meteorological parameters with significant seasonal variation and NT values as independent and dependent variables, respectively. The Statistical Package for Social Sciences (SPSS version 25, Tokyo, Japan) and XLSTAT 2019 statistical software (Addinsoft, NY, USA) for Windows were used to manage and analyze the data. All statistical tests were two-sided, and the level of statistical significance was set at a p-value <0.05.

Results

Samples

During the study period, a total of 2,611 infants underwent a 1-month check-up. After excluding those meeting the exclusion criteria and outliers, 1,759 infants were enrolled for analyses. In the enrolled infants (male/female ratio, 905:854), the median gestational age was 39.4 weeks and median birth weight was 3,028 g; these infants underwent the 1-month check-up at a median 31 days of life, and their median body weight at the check-up was 4,135 g. The median NT value was 91%, and the interquartile range was between 81% and 101%. The variables sex, gestational age, birth weight, nutrition, age in days at 1-month check-up, and body weight at check-up did not significantly differ between the 4 seasons (Table 1).

Seasonal and monthly variations in Normotest values

Over the four seasons, NT values peaked in the winter (median, 92%) and were lowest in summer (median, 87%). Differences in NT values among the seasons were all statistically significant ($p<0.001$; Table 1). The NT also significantly varied on a monthly basis ($p<0.001$), with the values peaking in January (median, 93%) and lowest in August (median, 87%).

Correlation between Normotest values and meteorological parameters

Simple linear regression analysis showed that there were weakly negative, but significant correlations between NT values and daily maximum air temperature ($p<0.001$), daily mean air temperature ($p<0.001$), daily mean relative humidity ($p<0.001$), and daily sunshine duration ($p=0.015$; Table 2). Stepwise multiple linear regression analysis was performed to evaluate the impact of the independent variables (daily maximum and mean air temperature, daily mean relative humidity, and daily sunshine duration) on the NT values. Only the daily mean air temperature was found to significantly influence the NT values ($p<0.001$). In addition, multiple logistic regression analysis was performed to identify the significant meteorological predictors of a low NT value. For this analysis, a low NT value was defined less than the 25th percentile, and all meteorological parameters used in the present study were considered for the multivariable model. This analysis revealed that only daily mean air temperature (adjusted odds ratio, 1.02; 95% confidence interval, 1.01–1.04, $p=0.002$) was independently associated with a low NT value.

Discussion

Our results show that the NT values decreased significantly during summer compared to that in the other seasons. Moreover, the effects of daily mean air temperature on the NT values and low NT values were noted. These findings indicate that in early infancy, VKDB occurs more frequently during the warm seasons. With respect to the relationship between seasons and the NT value, a slightly negative but significant correlation was observed between daily mean air temperature and NT values, on examining the meteorological data potentially affecting seasonal variation.

Seasonal changes in physiological mechanisms, such as disrupted hemostasis, are not well recognized; however, there seems to be a prothrombic state during winter. Several studies have shown seasonal variations in fibrinogen in the adult population, in which a winter-peak and summer-low have been statistically confirmed [14-16], and a strong negative relation has also been found between ambient temperature and plasma fibrinogen levels [15]. However, fibrinogen is not a VK-dependent coagulation factor, and the NT assay is not sensitive to its variation.

To our knowledge, there are no reports demonstrating the association between seasonal or ambient temperature and factor II or IX, among VK-dependent coagulation factors. There were no significant changes in factor X plasma levels, another VK-dependent coagulation factor measured by NT, during exposure to cold [17]. Conversely, the levels of factor VII (FVII) have been shown to increase in winter months [14,18], and a positive correlation has also been shown between ambient temperature and FVII [19]. NT has been reported to be useful for screening abnormal FVII activity [20]. There were no significant changes in factor X plasma levels, another VK-dependent coagulation factor measured by NT, during exposure to cold [17]. Based on these findings, we hypothesize that changes in the season and ambient temperature could lead to variations in the procoagulant blood marker, FVII, and could consequently influence the NT values in early infants.

The present study has several limitations. First, NT is not a standard test in most parts of the world and only provides multi-factorial results. To identify a stronger association with VKDB, a specific factor, such as FVII, must be considered. However, during the measurement of the activities of such specific factors, two challenges are encountered during sample collection in early infancy, namely, venous access and the requirement of a comparatively larger volume of blood. In contrast, only 10 μL of capillary blood is required for the NT and is easily obtained through a single heel prick [9,13]. Second, the retrospective study design limited the appropriate assessment of confounding factors. Third, the meteorological data included in the statistical analyses were not available at the time of blood sampling. Individual actual exposures to meteorological parameters may not always be consistent with the recorded data of a specific geographical region. This may introduce a certain level of evaluation error. In addition, data on the indoor temperature and humidity were not available. It is possible that even short-term exposure to indoor temperature and humidity may influence NT values.

The present study also has certain strengths. Information on seasonal variation in VK-dependent coagulation factors is also limited among adults, with most studies based on a small sample size, a limited number of variables, a short time period, and/or biased selection of parameters [14,15,18]. This is the first study evaluating the association between seasonal and meteorological data in early infancy with NT, a diagnostic test that evaluates VK-dependent coagulation factors comprehensively; the sample size was also relatively large.

Conclusion

A seasonal variation was possibly observed in the NT values in 1-month-old infants due to variations in daily air temperature. The impact of seasonal and ambient temperature variations on NT values, probably characterized by the seasonal variation in FVII, could explain the frequent late onset of VKDB in warm seasons and warm regions in Japan.

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References

[1] Pichler E, Pichler L. The neonatal coagulation system and the vitamin K deficiency bleeding-a mini review. *Wien Med Wochenschr* 2008; 158:385–395.

[2] Misirlioğlu ED, Aliefendioglu D, Bademci G, Baydar Z, Köse G, Çakmak FN. Intracranial hemorrhage due to vitamin K deficiency in infancy: clinical and radiological findings. *J Neurol Sci Turk* 2009; 26:18–25.

[3] Santorino D, Siedner MJ, Mwanga-Amumpaire J, Shearer MJ, Harrington DJ, Wariyar U. Prevalence and predictors of functional vitamin K insufficiency in mothers and newborns in Uganda. *Nutrients* 2015; 7:8545–8552.

[4] Hanawa Y, Maki M, Murata B, Matsuyama E, Yamamoto Y, Nagao T, et al. The second nation-wide survey in Japan of vitamin K deficiency in infancy. *Eur J Pediatr* 1988; 147:472–477.

[5] Nakayama K. The etiology of vitamin K deficiency in infants. *Perinat Med* 1982; 12:1029.

[6] Anai T, Matsu T, Oga M, Yoshimatsu J, Miyakawa I. Seasonal incidence of subclinical vitamin K deficiency during early newborn period. *Acta Obst Gynaec Ppn*

1991; 43:342–346.

[7] Iijima S, Sekii K, Baba T, Ueno D, Ohishi A. Seasonal variation in the international normalized ratio of neonates and its relationship with ambient temperature. *BMC Pediatr* 2016; 16:97.

[8] Takahashi D, Shirahata A, Itoh S, Takahashi Y, Nishiguchi T, Matsuda Y. Vitamin K prophylaxis and late vitamin K deficiency bleeding in infants: fifth nationwide survey in Japan. *Pediatr Int* 2011; 53:897–901.

[9] Owren PA, Strande OK. Normotest. *Farmakoterapi* 1969; 25:14–26.

[10] Hanawa Y, Maki M, Matsuyama E, Tada H, Urayama T, Yamada K, et al. The third nation-wide survey in Japan of vitamin K deficiency in infancy. *Acta Paediatr Jpn* 1990; 32:51–59.

[11] Shearer MJ, Rahim S, Barkhan P, Stimmler L. Plasma vitamin K1 in mothers and their newborn babies. *Lancet* 1982; 2:460–463.

[12] Rossi R, Albrecht O, Pollmann H, Jorch G, Harms E. Effect of a reduced vitamin K supplementation on prothrombin time in prematures and high-risk neonates. *Acta Paediatr* 1996; 85:747–749.

[13] Iijima S, Baba T, Ueno D, Ohishi A. International normalized ratio testing with a point-of-care coagulometer in 1-month-old infants: a comparison with Normotest.

Thromb. Res 2016; 145:72–77.

[14] Woodhouse PR, Khaw KT, Plummer M, Foley A, Meade TW. Seasonal variations of plasma fibrinogen and factor VII activity in the elderly: winter infections and death from cardiovascular disease. *Lancet* 1994; 343:435–439.

[15] Stout RW, Crawford V. Seasonal variations in fibrinogen concentrations among elderly people. *Lancet* 1991; 338:9–13.

[16] Van der Bom JG, de Maat MP, Bots ML, Hofman A, Kluft C, Grobbee DE. Seasonal variation in fibrinogen in the Rotterdam study. *Thromb Haemost* 1997; 78:1059–1062.

[17] Neild PJ, Syndercombe-Court D, Keatinge WR, Donaldson GC, Mattock M, Caunce M. Cold-induced increases in erythrocyte count, plasma cholesterol and plasma fibrinogen of elderly people without a comparable rise in protein C or factor X. *Clin Sci (Lond)* 1994; 86:43–48.

[18] Stout RW, Crawford VL, McDermott MJ, Rocks MJ, Morris TC. Seasonal changes in haemostatic factors in young and elderly subjects. *Age Aging* 1996; 25:256–258.

[19] Bull GM, Brozovic M, Chakrabarti R, Meade TW, Morton J, North WR, et al. Relationship of air temperature to various chemical, haematological, and haemostatic variables. *J Clin Pathol* 1979; 32:16–20.

[20] Dalaker K, Janson TL, Johnsen B, Skartlien AH, Prydz H. A simple method for determination of the factor VII-phospholipid complex using Normotest. *Thromb Res* 1987; 47:287-293.

Table 1. Demographic data of the study population and climatic data by season

	All (n=1759)	Spring (n=367)	Summer (n=449)	Autumn (n=486)	Winter (n=457)	*P* value
Male sex, n (%)	905 (51.4)	184 (50.1)	219 (48.8)	258 (53.0)	244 (53.4)	0.43
Gestational age, weeks	39.4 (38.4–40.3)	39.4 (38.4–40.3)	39.3 (38.4–40.1)	39.4 (38.5–40.3)	39.4 (38.6–40.1)	0.46
Birth weight, g	3028 (2820–3274)	3036 (2820–3276)	3036 (2812–3271)	3020 (2820–3260)	3026 (2830–3290)	0.84
Breastfeed, n (%)	1113 (63.3)	246 (67.0)	270 (60.1)	304 (62.6)	293 (64.1)	0.22
Age in days*, d	31 (28–33)	31 (29–33)	31 (28–33)	31 (28–33)	31 (28–33)	0.21
Body weight*, g	4135 (3812–4440)	4135 (3795–4400)	4144 (3830–4450)	4144 (3823–4480)	4075 (3799–4390)	0.22
Normotest value, (%)	91 (81–103)	91 (83–104)	87 (78–99)	89 (81–100)	92 (82–103)	<0.001
Meteorological parameters						
Daily max. air T, ℃	21.8 (14.4–27.9)	18.6 (14.4–23.2)	28.7 (27.7–32.1)	24.3 (21.5–28.5)	12.2 (10.0–14.1)	<0.001

Daily mean air T, °C	17.6 (10.2–23.3)	13.0 (10.7–17.7)	25.9 (22.9–27.8)	19.7 (16.0–23.6)	7.0 (5.1–9.7)	<0.001
Diurnal air T, °C	8.5 (6.6–10.0)	9.2 (7.3–10.3)	7.1 (6.0–8.3)	8.8 (7.3–9.9)	9.1 (6.7–10.7)	<0.001
Change in mean T from the day before, °C	0.2 (-0.6–1.4)	0.5 (-1.1–1.9)	0.1 (-1.1–1.3)	0.4 (-0.4–1.5)	0.0 (-0.4–1.2)	0.12
Daily mean relative humidity, (%)	72 (60–81)	62 (45–77)	80 (75–84)	71 (68–78)	60 (46–71)	<0.001
Daily mean barometric pressure, hPa	1009 (1004–1014)	1010 (1007–1013)	1004 (1002–1006)	1010 (1007–1014)	1012 (1009–1016)	<0.001
Daily total precipitation, mm	0.0 (0.0–2.5)	0.0 (0.0–3.0)	0.0 (0.0–2.8)	0.0 (0.0–1.0)	0.0 (0.0–3.0)	0.08
Daily sunshine duration, h	7.2 (2.2–9.5)	8.8 (3.2–11.1)	6.1 (1.0–10.0)	6.5 (0.6–8.8)	7.9 (2.2–9.3)	<0.001

*, at 1-month checkup; T, temperature. Values are presented as median (interquartile range) unless otherwise indicated. A *p*-value of <0.05 was considered to be statistically significant.

Table 2. Simple and multiple regression analyses between Normotest values and meteorological parameters

	Simple regression analysis				Multiple regression analysis			
			95% confidence interval				95% confidence interval	
	β value	*P* value	Lower	Upper	β value	*P* value	Lower	Upper
Daily maximum air temperature	-0.127	<0.001	-0.064	-0.030		0.396		
Daily mean air temperature	-0.135	<0.001	-0.046	-0.022	-0.135	<0.001	-0.46	-0.022
Diurnal air temperature	0.028	0.232	-0.008	0.034	NA	NA	NA	NA
Daily mean relative humidity	-0.089	<0.001	-0.083	-0.026		0.757		
Daily mean barometric pressure	0.018	0.446	-0.73	1.66	NA	NA	NA	NA
Daily sunshine duration	0.058	0.015	0.001	0.01		0.076		

NA, not applicable. A *P* value of less than 0.05 was considered to be statistically significant.