— Regular Article —

Combined Impact of the Association between Prepregnancy Body Mass Index and Gestational Weight Gain on the Placental/Umbilical Cord Blood Volume Collected

Atsuko Omori,[a] Takako Chiba,[a] Kenji Takahashi,[a] Kanako Tanaka,[b] Mami Manabe,[b] and Ikuo Kashiwakura*,[a]

[a]*Department of Radiological Life Sciences, Division of Medical Life Sciences, Hirosaki University Graduate School of Health Sciences, 66–1 Hon-cho, Hirosaki 036–8564, Japan and* [b]*Department of Obstetrics and Gynecology, Hirosaki National Hospital, Tomino-cho, Hirosaki 036–8545, Japan*

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Placental/umbilical cord blood (CB) contains multipotent hematopoietic stem/progenitor cells and has been utilized worldwide both clinically and experimentally. Neonatal birth weight is positively correlated with CB volume and with the total number of nucleated cells associated with engraftment and survival after CB transplantation. Considering the recent increase in the frequency of low birth weight (< 2500 g) and the decline in body mass index (BMI) among Japanese women of childbearing age, the present study investigated the combined impact of the prepregnancy BMI and the gestational weight gain on CB volume. From 1998 to 2007, CB samples were obtained from 579 healthy women with singleton vaginal deliveries. The prepregnancy BMI was classified into the underweight, normal, overweight, or obese groups. According to the current gestational weight gain recommendations or other new optimum recommendations, the gestational weight gain was classified into below, within, or above recommendations in each prepregnancy BMI group. The neonatal weight and placental weight had significantly positive effects on the CB volume. Underweight pregnant women demonstrated significantly lower neonatal and placental weight. According to the current recommendations, no significant difference in the CB volume was observed. According to the new optimum recommendations for underweight pregnant women, a significantly higher CB volume was obtained from the group within the recommended weight gain range than from the group below the recommended range. In the underweight group, a higher CB volume could be obtained if the upper limit of the gestational weight gain increases by a few kilograms more than the current gestational weight gain recommendations.

Key words —— cord blood volume, neonatal weight, prepregnancy body mass index, gestational weight gain

INTRODUCTION

Placental/umbilical cord blood (CB) contains multipotent hematopoietic stem/progenitor cells.[1] CB can be harvested at no risk to the mother or infant and is increasingly used as an alternative source of hematopoietic stem cell transplantation for patients with diseases such as hematopoietic malignancies.[2,3] The total number of nucleated cells infused is a significant factor associated with engraftment and survival after transplantation.[4,5] However, it is difficult to predict the number of nucleated cells per CB sample prior to cell processing because of the extremely wide variations in individual samples. Although previous studies have reported the positive correlations between neonatal weight, placental weight, CB volume, and the total number of nucleated cells,[6–8] thus far very little has been elucidated regarding the combined impact of the association between prepregnancy body mass index (BMI) and gestational weight gain on the CB volume collected.

Recently, the increasing number of overweight and obese women has been a major public health problem worldwide,[9–11] and maternal obesity is as-

*To whom correspondence should be addressed: Department of Radiological Life Sciences, Division of Medical Life Sciences, Hirosaki University Graduate School of Health Sciences, 66–1 Hon-cho, Hirosaki 036–8564, Japan. Tel. & Fax: +81-172-39-5938; E-mail: ikashi@cc.hirosaki-u.ac.jp

sociated with a high risk of antenatal/intrapartum complications.[12–15] Meanwhile, in Japan a dramatic increase in thinness and a decline in BMI have emerged among Japanese women of childbearing age.[16] Furthermore, the increasing frequency of low birth-weight infants (< 2500 g) and the decline in the average birth weight have been a nationwide concern for the last few decades.[17] Both prepregnancy underweight and low gestational weight gain are associated with low birth weight.[18, 19]

Considering these issues, we hypothesized that gestational weight gain below the recommendations might be one of the factors that negatively affects the CB volume collected for hematopoietic stem cell transplantation. Over the past decade, we have investigated megakaryocytopoiesis/thrombopoiesis as well as the action of cytokines and their radiation sensitivity using cluster of differentiation 34 positive ($CD34^+$) cells prepared from more than 700 CB samples collected for basic research.[20–22] These investigations of hematopoietic progenitor cells have shown remarkably significant differences in both the CB volume and the number of nucleated cells. Moreover, the current criteria of CB banking are based on the CB volume and the total number of nucleated cells, and there is no other determining factor of CB collection from pregnant women who voluntarily offered their CB for donation thus far. Therefore, the present study retrospectively investigated whether weight control during pregnancy could influence the CB volume collected, based on the association between the prepregnancy BMI and gestational weight gain.

MATERIALS AND METHODS

CB Collection —— After the approval by the Committee of Medical Ethics of Hirosaki University Graduate School of Medicine, CB samples were collected at the Fukushi Birth Center located in Goshogawara-shi (Aomori, Japan). Informed consent was obtained from all the mothers prior to collection after an explanation by the midwife during the later gestational period. During the period from August 1998 until the end of September 2007, a total of 579 CB samples were collected from healthy women with singleton vaginal deliveries. Women who had cesarean sections and medical complications of pregnancy were excluded, as relatively healthy pregnant women with no severe diseases tend to give birth at birth centers under the Medical and Midwifery Law in Japan. According to the guidelines of the Tokyo Cord Blood Bank, CB was collected after the delivery of the placenta, drained by gravity in an adjacent room, and allowed to flow into a sterile collection bag containing 28 ml of citrate-phosphate-dextrose anticoagulant (CBC-20, Nipro, Osaka, Japan) until the flow ceased (approximately 5 min). One midwife was primarily responsible for collecting CB samples throughout the entire collection period for this study. Extensive data regarding the medical and family history were voluntarily collected and reported by the staff at the birth center. Of 579 CB samples, a total of 547 were available for analysis due to excluding the CB samples that had either unknown or missing obstetric factor data in the clinical charts.

BMI and Gestational Weight Gain Recommendations —— The obstetric factors extracted from the clinical charts were maternal age, paternal age, residential area, maternal occupation, the number of pregnancies, parity, self-reported maternal weight and height, prepregnancy BMI, gestational weight gain, maternal smoking status, gestational age, neonatal gender, duration of labor, neonatal weight and height, placental weight, cord length, and CB volume (CB net weight).

According to the World Health Organization (WHO)[23] and the Japan Society for the Study of Obesity,[24] prepregnancy BMI (kg/m^2) was classified into four groups: underweight (< 18.5), normal (18.5–< 25), overweight (25–< 30), and obese (≥ 30). According to the Ministry of Health, Labor and Welfare of Japan,[17] and the Japan Society of Obstetrics and Gynecology,[25] the current gestational weight gain recommendations are 9–12 kg for the underweight, 7–12 kg for normal, and 5–7 kg for overweight and obese groups. The gestational weight gain in each prepregnancy BMI was classified into three groups: below recommendations, within recommendations, and above recommendations.

Additional Analysis of the Underweight Group —— Another set of optimum gestational weight gain recommendations for underweight women has been recently proposed[26] after reconsidering the risks related to this group, based on the findings that neonatal weight tends to decrease in the case of gestational weight gain of less than 10 kg and that heavy for date (HFD) newborns or preeclampsia tend to increase in the case of gestational weight gain of more than 14 kg. Therefore, additional analyses of the underweight group were performed according to the new optimum gestational weight gain recommendations, including 10–14 kg for the underweight, 7–13 kg for normal, and < 7 kg for overweight and obese groups.

Statistical Analysis —— A univariate analysis was performed using Pearson's correlation coefficient. To determine significant differences among obstetric factors or the CB volume, a multiple-comparison test was performed using the Bonferroni-Dunn test between each BMI group or between each group of gestational weight gain (below recommendations, within recommendations, and above recommendations) of each BMI group. The statistical analysis was performed using Origin software (Origin Lab, Northampton, MA, U.S.A.) for Windows. A value of $p < 0.05$ was considered to be statistically significant.

RESULTS

Table 1 shows a summary of the characteristics of the 547 CB samples. The mean net weight of the CB collected was 54.8 ± 21.6 g. The mean maternal age was 26.6 ± 4.6 years, with a range of 17–41 years. The mean prepregnancy BMI (kg/m^2) was 21.2 ± 3.3, which was less than the standard BMI of 22. The distribution of prepregnancy BMI was underweight (< 18.5) 17.4%, normal (18.5–< 25) 70.4%, overweight (25–< 30) 9.3%, and obese (≥ 30) 2.9%. The mean neonatal birth weight was 3216.3 ± 375.3 g, including low birth weight (< 2500 g) 1.7% and macrosomia (≥ 4000 g) 2.9%. The mean gestational age was 39.5 ± 1.2 weeks, with a range of 35–43 weeks. Regarding the CB net weight of each BMI group such as underweight (< 18.5), normal (18.5–< 25), overweight (25–< 30), and obese (≥ 30), the data were 54.7 ± 19.9 g (range 20–237 g), 54.1 ± 21.0 g (range 10–195.0 g), 59.6 ± 29.2 g (range 20–210 g), and 55.8 ± 15.0 g (range 35.2–80.0 g), respectively. Consequently, no significant difference in the CB net weight was observed between any of the prepregnancy BMI groups.

Table 1. Characteristics of CB and Obstetric Factors

	n (%)	Mean ± S.D.	Range
CB net weight[a] (g)	547	54.8 ± 21.6	10 – 210
Maternal age (years)	547	26.6 ± 4.6	17 – 41
Parity			
Nulliparous	211 (38.6)		
Multiparous	336 (61.4)		
Maternal height (cm)	547	158.7 ± 5.6	143.5 – 177
Prepregnancy weight (kg)	547	53.5 ± 9.0	37 – 100
Prepregnancy BMI[b]	547	21.2 ± 3.3	15.0 – 35.4
Underweight (< 18.5)	95 (17.4)		
Normal (18.5–< 25)	385 (70.4)		
Overweight (25–< 30)	51 (9.3)		
Obese (≥ 30)	16 (2.9)		
Gestational weight gain (kg)	547	11.9 ± 4.4	(−) 2 – 28
Gestational age (weeks)	547	39.5 ± 1.2	35 – 43
Maternal smoking status			
Nonsmokers	442 (80.8)		
Smokers	105 (19.2)		
Neonatal gender			
Male	275 (50.3)		
Female	272 (49.7)		
Neonatal weight (g)	547	3216.3 ± 375.3	2160 – 4540
Neonatal height (cm)	547	49.7 ± 1.8	44 – 55
Placental weight (g)	547	516.8 ± 86.0	290 – 840
Cord length (cm)	547	60.0 ± 13.2	18 – 125

a) CB net weight indicates CB volume excluding anticoagulant (CPD). *b*) BMI: weight in kilograms divided by the square of the height in meters (kg/m^2). Prepregnancy BMI was classified into four groups: underweight (< 18.5), normal (18.5–< 25), overweight (25–< 30), and obese (≥ 30). *n*: the number of CB samples. S.D.: standard deviation. Prepregnancy weight was self-reported.

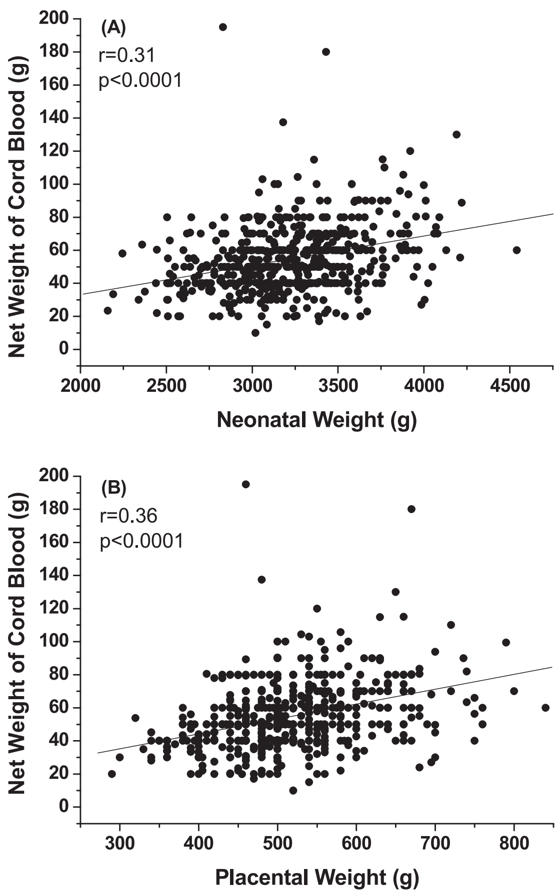

Fig. 1. Correlations between the Obstetric Factors of CB Samples

Significant positive correlations were observed (A) between neonatal birth weight and CB net weight (n = 547), and (B) between placental weight and CB net weight (n = 547). Pearson's correlation coefficient: $^{*}p < 0.01$.

As shown in Fig. 1, the CB net weight had significantly positive correlations with birth weight and placental weight (r = 0.31, p < 0.01; r = 0.36, p < 0.01, respectively). A significantly higher gestational weight gain was observed in the underweight group compared with the other BMI groups (Fig. 2a). In contrast, a significantly lower gestational age, neonatal weight, and placental weight were observed in the underweight group (Fig. 2b, 2c, and 2d).

According to the current gestational weight gain recommendations, no significant difference in the CB weight was observed between any groups (Fig. 3). On the other hand, according to the new optimum gestational weight gain recommendations (10–14 kg) recently proposed for the underweight group, a significantly higher CB weight was obtained from the group within the recommended weight gain range (10–14 kg) compared with the group below the recommended range (< 10 kg), but

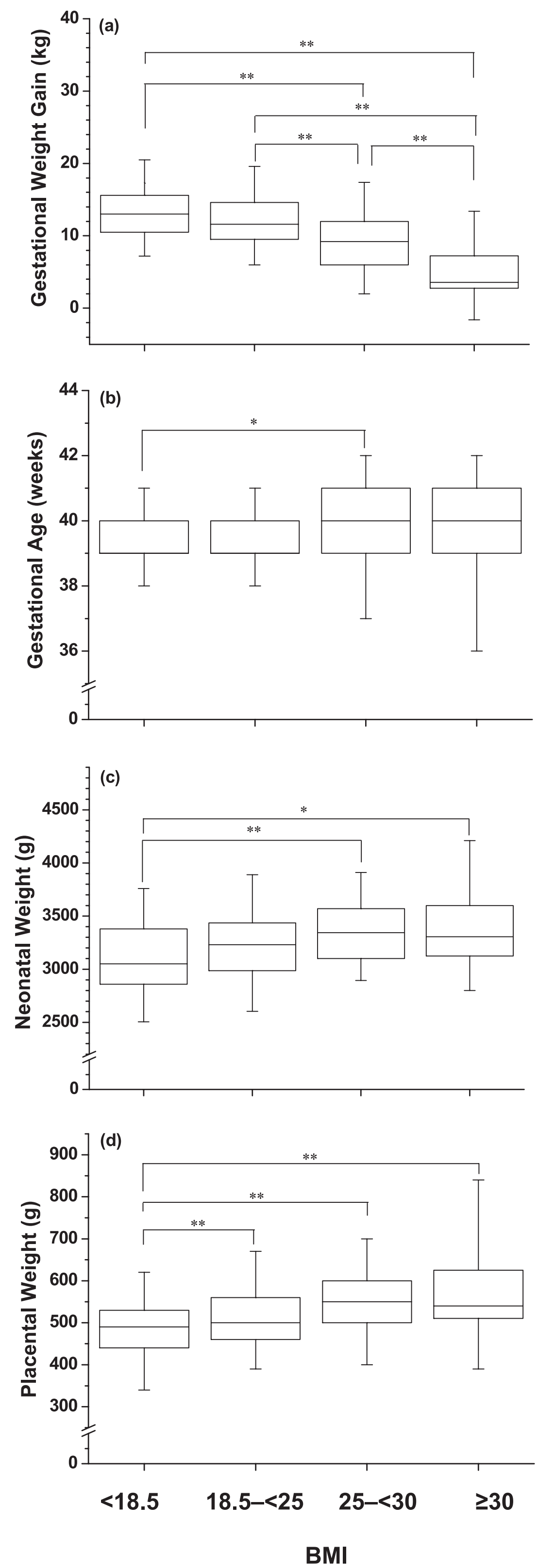

Fig. 2. Multiple-Comparison Test of Obstetric Factors among the Prepregnancy BMI Groups

The obstetric factors are (a) gestational weight gain, (b) gestational age, (c) neonatal weight, and (d) placental weight. Prepregnancy BMI (kg/m^2) was classified into four groups: underweight (< 18.5, n = 95), normal (18.5–< 25, n = 385), overweight (25–< 30, n = 51), and obese (≥ 30, n = 16). Bonferroni-Dunn test: $^{*}p < 0.05$, $^{**}p < 0.01$.

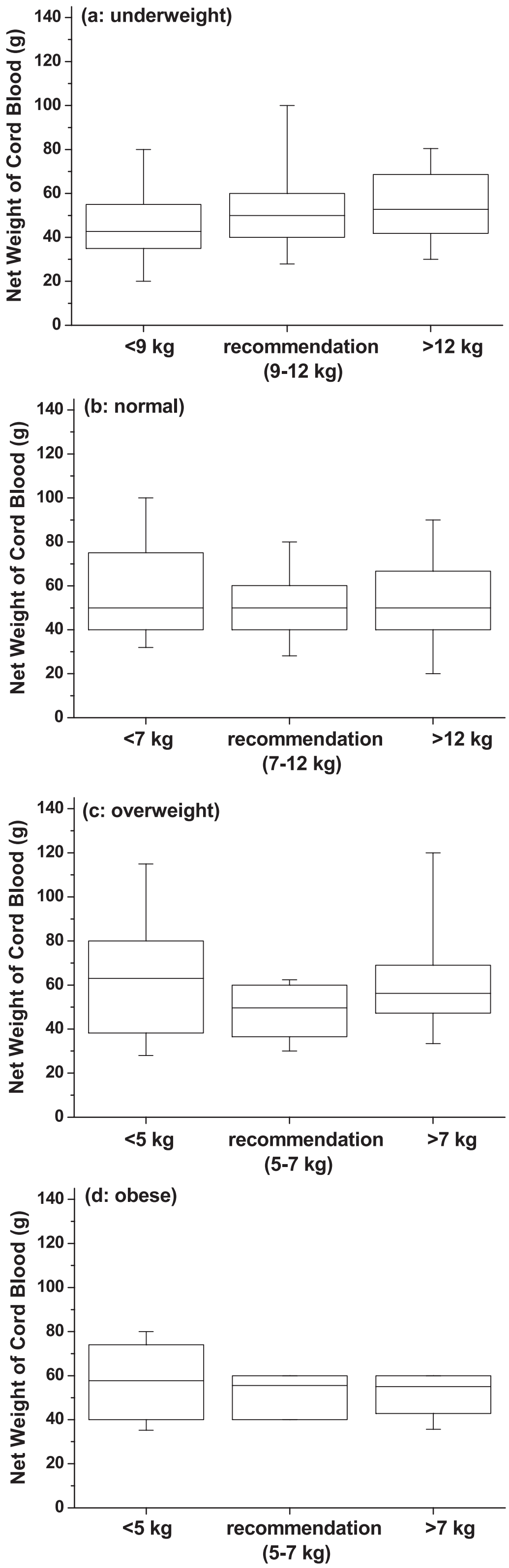

Fig. 3. Multiple-Comparison Test of CB Net Weight According to the Current Gestational Weight Gain Recommendations for Each Prepregnancy BMI

(a: underweight) < 9 kg (n = 12), 9–12 kg (n = 31), and > 12 kg (n = 52), (b: normal) < 7 kg (n = 31), 7–12 kg (n = 184), and > 12 kg (n = 170), (c: overweight) < 5 kg (n = 10), 5–7 kg (n = 7), and > 7 kg (n = 34), (d: obese) < 5 kg (n = 9), 5–7 kg (n = 3) and > 7 kg (n = 4). Bonferroni-Dunn test: no significance between any groups.

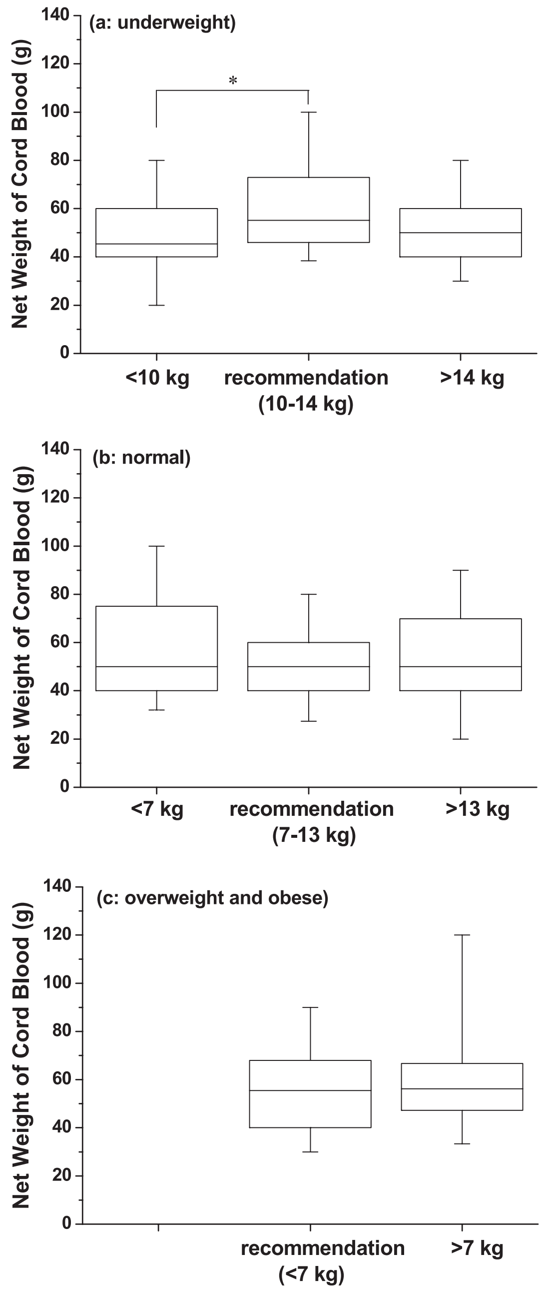

Fig. 4. Multiple-Comparison Test of CB Net Weight According to the New Optimum Gestational Weight Gain Recommendations for Prepregnancy BMI (Underweight)

(a: underweight) < 10 kg (n = 19), 10–14 kg (n = 42), and > 14 kg (n = 34), (b: normal) < 7 kg (n = 31), 7–13 kg (n = 211), and > 13 kg (n = 143), (c: overweight and obese) < 7 kg (n = 29) and > 7 kg (n = 38). Bonferroni-Dunn test: $^{*}p < 0.05$.

no significant differences were observed between any other groups (Fig. 4). Furthermore, Table 2 illustrates the results of multiple-comparison tests of the CB net weight, neonatal weight, and placental weight among the recommended weight gain ranges for the underweight group. Despite the higher neonatal weight in the group above recommenda-

Table 2. Multiple-Comparison Test of the CB Net Weight, Neonatal Weight and Placental Weight According to the New Optimum Gestational Weight Gain Recommendations for the Prepregnancy BMI Underweight Group

Gestational weight gain (kg)	*n*	CB net weight (g)	Neonatal weight (g)	Placental weight (g)
< 10 kg	19	46.23 ± 17.04 (45.4)	2848.95 ± 291.17 (2910)	431.83 ± 57.11 (440)
10 – 14 kg	42	60.87 ± 21.81 (55.1)*	3135.60 ± 327.03 (3120)	504.52 ± 74.65 (500)
> 14 kg	34	51.90 ± 16.86 (50)	3222.50 ± 425.22 (3250)	495.15 ± 79.22 (490)

CB net weight indicates CB volume excluding anticoagulant (CPD). CB net weight, neonatal weight, and placental weight data indicate the mean ± standard deviation and (median). Bonferroni-Dunn test: CB net weight within recommendations (10 – 14 kg) *vs.* below recommendations (< 10 kg). $^{*}p < 0.05$.

tions (> 14 kg), the CB volume was relatively lower in comparison with the group within recommendations (10–14 kg).

DISCUSSION

Neonatal birth weight is influenced by obstetric factors, such as prepregnancy BMI, gestational weight gain, gestational age, gravida status, maternal smoking, and maternal age (juvenile or elderly primigravida),[27,28] but the placental weight is the strongest determinant of birth weight.[29] Various risks are not only associated with maternal obesity but also with underweight women.[18,19] Therefore, pregnant women should maintain a normal BMI to achieve a healthy pregnancy outcome.[30] In addition, a low weight gain is associated with an increased risk for all of the preterm birth subtypes in comparison with a normal weight gain and low weight gain is an independent and much stronger risk factor for early preterm birth than obesity, although the underlying mechanisms are poorly understood.[31]

In the present study, neonatal weight and placental weight had significantly positive correlations with the CB volume, which was consistent with previous reports indicating that bigger is better.[6–8] Considering the dramatic increase in thinness among Japanese women of childbearing age,[16] along with the decline in the BMI and the average birth weight,[17] careful control of the gestational weight gain in underweight women is important for both mothers and neonates. We found that the neonatal weight and placental weight in the underweight group were significantly lower than those in any other BMI group (Fig. 2c and 2d). However, according to the current gestational weight gain recommendations for each BMI group, no significant difference in the CB volume was observed between any groups (Fig. 3). This could be attributed to relatively healthy women giving birth at birth centers in Japan, thus indicating that a weight gain below, within, or above the recommendations does not seem to have a significant impact on the CB volume in women with uncomplicated pregnancies. However, the detailed mechanisms of the combined impacts of the prepregnancy BMI and gestational weight gain on CB quality still remain unclear. Further studies will be needed to clarify the associations between them.

Interestingly, according to the recently proposed new optimum gestational weight gain recommendations for the underweight group,[26] which is slightly higher than the current recommendations for underweight women (10–14 kg *vs.* 9–12 kg), a higher CB volume was obtained in comparison with the current recommendations (60.9 ± 21.8 g *vs.* 56.8 ± 24.1 g). As shown in Table 2, despite the higher neonatal weight in the above recommendations, the CB volume was relatively lower in comparison with the group within recommendations. To some extent, this is probably caused by a lower placental weight in the above recommendations, because no significant differences were observed in smoking status, maternal age, or gestational age between the below recommendations and within the recommendations (data not shown). Consequently, this outcome may be supported by the previous studies reporting that underweight women appeared to benefit from gaining more weight than recommended,[32] and that severely thin mothers with very low or very high pregnancy weight gains were at the greatest risk for spontaneous preterm birth.[33]

There are limitations in the present study due to collecting CB samples at a birth center. Potential confounders such as severe complications could be excluded. Moreover, CB samples were obtained from a regional birth center located in the northern part of Japan. Furthermore, despite the relatively large total number of CB samples, the small group size for underweight pregnant women may have limited the power to detect a difference between groups. However, the well-conducted pregnancy and gestational weight gain controls in each prepregnancy BMI group might strongly reflect the good-quality CB samples collected for donations and possibly lead to a decrease in the low birthweight ratios as well. Regarding the technical factors, even though a working environment may differ from the hospitals affiliated with CB banks, one expert midwife voluntarily performed the CB collection over a decade and her solid experience successfully contributed to consistent, stable CB collection without any problems.

Taken together, in the underweight group, a significantly higher CB volume could be obtained if the upper limit of the gestational weight gain increases by a few kilograms more than the current gestational weight gain recommendations for underweight pregnant women. Therefore, the upper limit of the gestational weight gain recommendations for underweight pregnant women should be considered as one of the critical factors for CB collection, providing that a healthy pregnancy is maintained. Our findings may thus give new insight into obstetric factors and provide useful information for improving efficient CB collection from underweight pregnant women.

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