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Original Article

Relation of Prenatal and Postnatal Status to Calcaneus Quantitative Ultrasound in Adolescents

Yoko Takahata[a,b]*, Da-Hong Wang[b], Takanobu Anai[c], and Keiki Ogino[b]

[a]*Faculty of Nursing, Hiroshima International University, Kure, Hiroshima 737-0112, Japan,*
[b]*Department of Public Health, Okayama University Graduate School of Medicine, Dentistry and Pharmaceutical Sciences, Okayama 700-8558, Japan, and*
[c]*Oita University Faculty of Medicine Graduate School of Medicine, Hazama, Oita 879-5593, Japan*

This study aimed to elucidate the relationship of prenatal and/or postnatal factors, including acquired factors, with the calcaneus stiffness index as measured by quantitative ultrasound (QUS-SI) in adolescents. We recruited 1,143 adolescents with a mean age of 14.8 ± 1.8 years (501 boys and 642 girls). The subjects' calcaneus QUS-SI was measured using an ultrasound bone densitometer. We also measured the subjects' height, weight, and grip strength. Data on prenatal and postnatal factors were obtained from maternal and child health handbooks. A self-reporting questionnaire was used to obtain information on subjects' secondary sexual characteristics and lifestyle factors. We found that maternal weight gain during pregnancy was independently associated with calcaneus QUS-SI in girls, and that grip strength was also significantly associated with calcaneus QUS-SI in both sexes. The present findings suggest that excessive restriction of maternal weight gain would have a negative effect on the calcaneus QUS-SI of girls, and that exercise and strength-building activities are likely to result in a higher calcaneus QUS-SI in both sexes of adolescents.

Key words: adolescents, calcaneus QUS-SI, prenatal and/or postnatal status, stiffness index, ultrasound bone densitometer

Bone formation starts as early as in the third trimester of pregnancy, after which the bone is broken down and rebuilt continuously throughout life. It has been reported that people usually reach their peak bone mass in their late teens [1]. Therefore, it is important to attain a high peak bone mass during late teenage years to reduce the risk of osteoporosis in later life. Body size, regular physical activities and diet have been proven to be helpful for acquiring sufficient peak bone mass in adolescents [2-9]. At the same time, congenital variables such as maternal diet, birth weight and birth height have been shown to influence bone mass [10-17].

Rohana *et al.* reported that parenteral nutrition for longer periods among very-low-birth-weight infants increased the risk of having a low bone mineral density (BMD) [10]. Other researchers have demonstrated that fetal growth patterns and birth weight affect bone mineral content (BMC) in childhood and adulthood [11-13], although a large cohort study found that birth weight had at most a weak effect on BMD, and only in men [14]. Some findings on the relationship between BMD or BMC and congenital variables also postulated that low birth weight and

Received August 25, 2011; accepted December 5, 2011.
*Corresponding author. Phone:+81-72-643-6221; Fax:+81-72-643-8473
E-mail:y-takahata@baika.ac.jp (Y. Takahata)

poor childhood growth correlate with a higher risk of hip fracture later in life [15]. Some researchers suggest that influential factors during the fetal period and infancy on later BMD or skeletal growth may be due to programming of the growth hormone [16-21].

Several studies have shown significant correlations between either BMD [22-25] or BMC [26] and the calcaneus stiffness index by quantitative ultrasound (QUS-SI) and have suggested that prenatal and postnatal status influence calcaneus QUS-SI during childhood and adulthood. However, it is unclear whether the prenatal and postnatal status affects the calcaneus QUS-SI during adolescence. This study, therefore, aimed to elucidate the relation of prenatal and/or postnatal status, as well as acquired factors and lifestyle factors on the calcaneus QUS-SI in adolescent subjects.

Materials and Methods

Subjects. This study recruited 1,183 adolescents and recorded their anthropometric data from September to December in 2008. Forty adolescents were excluded from the study because their maternal and child health handbooks were missing. The study subjects included 501 boys (320 in junior high school and 181 in senior high school) and 642 girls (371 in junior high school and 271 in senior high schools). All subjects were in good health and were free of chronic diseases affecting bone metabolism. Informed consent was obtained from all subjects, parents and guardians in accordance with the Helsinki Declaration. The study was approved by the Ethics Committee of the Prefectural University of Hiroshima.

Calcaneus bone density measurement. The speed of sound (SOS), broadband ultrasound attenuation (BUA) and the stiffness index of the calcaneus were measured using an ultrasound bone densitometer (Achilles InSight, GE Healthcare. Little Chalfont, UK). The stiffness index was used for evaluation of the bone mineral density in this study and was calculated by the formula: $(BUA-50) \times 0.67 + (SOS-1380) \times 0.28$. We measured the participants' stiffness index using the ultrasound bone densitometer. The reason why we employed the ultrasound bone densitometer was that it has no side effects and correlates well with the DXA-measured BMD [22-26, 27, 28] or BMC [28]. The densitometer was operated by one researcher who had practiced performing measurements repeatedly before starting the research, and the final intra-sample mean coefficient of variation for the stiffness index was 0.7%.

Physical examination. The subjects' height (by DST-210N, Muratec-KDS Corp. Kyoto, Japan) and weight (by TBF-546, Tanita Corp. Tokyo, Japan) were measured without shoes. A Smedley type dynamometer was used to measure the grip strength to represent muscle strength. All measurements were performed twice for each hand and the highest result for each hand was used to calculate the mean grip strength.

Questionnaires. A self-reporting questionnaire was used including variables of sex, age, secondary sexual characteristics, bed time, average duration of sleep, exercise habits in junior high school, and calcium-rich food intake, such as milk, dairy products, fermented soybeans, bean curds, and fish. The age of secondary sexual characteristics was determined based on the age when pubic hair started to appear for boys and the age when menstruation started for girls. The researcher explained the questions to the subjects if any item on their questionnaires was left blank.

Birth weight, birth height, pre-pregnancy weight, maternal height, maternal weight gain, neonatal weight gain at 1 month, neonatal weight gain at 4 months, and gestational length were estimated using the subjects' maternal and child health handbooks.

Statistical analysis. The subjects' characteristics were compared using the unpaired *t*-test to find if any significant difference existed between sexes. Pearson's correlation coefficient was used to find the relationship between the calcaneus QUS-SI and other factors. A simple linear regression was used to determine the factors having significant correlation with the calcaneus QUS-SI for the next step analysis. Then a multiple regression analysis was used to determine the effect of prenatal and/or postnatal factors, and lifestyles on the calcaneus QUS-SI in adolescents. One-way analysis of variance covariance (ANOVA) followed by post-hoc Tukey's compromise test was used to find the difference of calcaneus QUS-SI for various lifestyle factors in each sex. In order to find the approximate value of the suitable gestational weight gain for future intervention trials, ANOVA followed by post-hoc Tukey's compromise tests was used to determine the difference of calcaneus QUS-SI in girls among 4 maternal weight-gain groups. Dietary guidelines for pregnant women by Ministry of Health, Labor and Welfare (2006) recommend that the lower limit of gestational weight gain is around 5 kg [29]; we therefore used less than 6 kg as the minimum group, and sought to determine how calcaneus QUS-SI changes for 5-kg increments in gestational weight. A p value < 0.05 was considered to be statistically significant. Statistical analyses were performed using the SPSS 19.0 software program.

Results

Subjects' characteristics. The subjects' characteristics are listed in Table 1 and Table 2. No significant differences were observed between sexes regarding the mean value of the calcaneus QUS-SI. Mean values of weight, height, secondary sex characteristics, grip strength, birth weight, birth height, neonatal weight gain at 1 month, and neonatal weight gain at 4 months were significantly higher in boys while only the mean value of gestational length was significantly higher in girls (Table 1). The correlation coefficients between prenatal factors, postnatal factors, acquired gain factors, lifestyle status and calcaneus QUS-SI in both genders are also shown in Table 1. Almost all factors were significantly associated with the calcaneus QUS-SI. In addition, the results on how these factors correlated with calcaneus QUS-SI are shown in Table 1 by the regression coefficients of the simple linear regression analysis. All factors showed significant relationships with the calcaneus QUS-SI of the girls.

The survey showed that 85.2% of boys went to bed before 12 a.m. and so did 74.8% of girls (Table 2). A higher proportion of boys (more than 45%) consumed milk, fermented soybeans, bean curds, and fish once a week or more, and a higher proportion of boys (89.8%) had engaged in regular exercise at the junior-high age (Table 2). Differences in calcaneus QUS-SI for various lifestyle factors in both sexes are shown in Table 2. The calcaneus QUS-SI classified by bed time and milk intake in boys showed significant differences among the categorized groups.

The effect of prenatal and/or postnatal factors, acquired factors, and lifestyle factors on

Table 1 Subjects' characteristics 1 — The effect of prenatal and postnatal factors, and acquired factors on the stiffness index —

	Boys	Correlation coefficient for boys	β of the simple linear regression for boys	Girls	Correlation coefficient for girls	β of the simple linear regression for girls
N	501			642		
Stiffness index*1	107.8 ± 18.6			107.5 ± 15.4		
Age (years)	14.7 ± 1.7	0.418**	0.42***	14.9 ± 1.8	0.204**	0.20***
Weight (kg)†	54.7 ± 11.7	0.276**	0.28***	49.3 ± 8.9	0.269**	0.27***
Height (cm)†	164.1 ± 8.9	0.247**	0.25***	155.3 ± 5.6	0.187**	0.19***
Secondary sexual characteristics (years)*2†	12.5 ± 1.1	0.015	0.05	11.9 ± 1.1	−0.074*	−0.08*
Grip strength (kg)†	33.3 ± 9.2	0.371**	0.37***	23.9 ± 5.1	0.247**	0.25***
Pre-pregnancy weight (kg)	50.5 ± 6.9	0.041	0.07	50.8 ± 6.8	0.041	0.10*
Maternal height (cm)	156.9 ± 5.1	−0.074*	−0.04	157.0 ± 5.0	0.069*	−0.08*
Maternal weight gain (kg)	10.6 ± 3.7	−0.077*	−0.09	10.7 ± 3.8	0.109**	0.15**
Gestational length (days)†	273.3 ± 12.3	−0.067*	−0.12**	275.1 ± 13.0	−0.064*	0.11**
Birth weight(g)†	3,099.0 ± 475.0	0.054	0.08	3,005.0 ± 427.0	0.074*	0.12**
Birth height (cm)†	49.7 ± 2.7	0.044	0.07	49.1 ± 2.6	−0.054	−0.10*
Neonatal weight gain at 1 month (g)†	1,227.8 ± 336.1	0.047	0.10*	1,047.9 ± 316.3	0.071*	0.14**
Neonatal weight gain at 4 months (g)†	3,773.8 ± 645.6	0.041	0.09	3,418.5 ± 661.9	−0.067*	0.20***

means ± SD
*1: The stiffness index was used as subjects' bone mineral density.
*2: For Boys: The age when pubic hair started to appear.
For Girls: The age when menstruation started.
† The significant differences of stiffness index were shown between both sexes.
*$p < 0.05$, **$p < 0.01$, ***$p < 0.001$

Table 2 Subjects' characteristics 2 — The effect of lifestyle factors on the stiffness index —

	Boys (n = 501)	Average stiffness index for boys	Girls (n = 642)	Average stiffness index for girls
Bed time				
Go to bed between 9 p.m. and 10 p.m	56 (11.2%)	101.5	36 (5.6%)	107.4
Go to bed between 10 p.m. and 11 p.m.	209 (41.7%)	106.2 * * *	209 (32.6%)	105.1
Go to bed between 11 p.m. and 12 a.m.	162 (32.3%)	109.5	235 (36.6%)	108.9
Go to bed between 12 a.m. and 1 a.m.	55 (11.0%)	114.4	140 (21.8%)	109.0
Go to bed on 1 a.m. or later	19 (3.8%)	109.8	22 (3.4%)	105.9
Average duration of sleep				
Under 5 hours	10 (2.0%)	110.0	14 (2.2%)	106.9
5 to 6 hours	125 (25.0%)	110.6	242 (37.7%)	107.2
7 to 8 hours	331 (66.1%)	107.3	358 (55.8%)	108.1
9 to 10 hours	32 (6.4%)	102.4	27 (4.2%)	103.2
Over 10 hours	3 (0.6%)	94.0	1 (0.2%)	121.6
Calcium-rich food intake				
Milk intake				
Every day	319 (63.7%)	106.3 *	311 (48.4%)	106.9
Once every 2 or 3 days	68 (13.6%)	115.9	85 (13.2%)	108.4
Once a week	31 (6.2%)	105.7 *	57 (8.9%)	106.7
Once a month	29 (5.8%)	103.7	73 (11.4%)	108.4
Never	54 (10.8%)	109.8	116 (18.1%)	108.2
Dairy products intake				
Every day	61 (12.2%)	107.6	58 (9.0%)	105.4
Once every 2 or 3 days	118 (23.6%)	108.8	176 (27.4%)	107.3
Once a week	149 (29.7%)	109.1	207 (32.2%)	107.8
Once a month	140 (27.9%)	106.6	168 (26.2%)	106.9
Never	33 (6.6%)	103.2	33 (5.1%)	109.9
Fermented soybeans intake				
Every day	40 (8.0%)	107.9	19 (3.0%)	105.1
Once every 2 or 3 days	59 (11.8%)	110.8	66 (10.3%)	107.8
Once a week	126 (25.1%)	105.2	176 (27.4%)	109.2
Once a month	156 (31.1%)	108.3	223 (34.7%)	107.6
Never	120 (24.0%)	108.3	158 (24.6%)	105.7
Bean curds intake				
Every day	55 (11.0%)	107.5	60 (9.3%)	108.5
Once every 2 or 3 days	138 (27.5%)	108.4	160 (24.9%)	108.7
Once a week	184 (36.7%)	108.7	218 (34.0%)	108.0
Once a month	102 (20.4%)	106.6	157 (24.5%)	105.5
Never	22 (4.4%)	102.3	47 (7.3%)	106.6
Fishes intake				
Every day	27 (5.4%)	110.1	20 (3.1%)	110.7
Once every 2 or 3 days	208 (41.5%)	107.8	275 (42.8%)	107.5
Once a week	197 (39.3%)	107.6	243 (37.9%)	107.9
Once a month	54 (10.8%)	105.4	93 (14.5%)	106.0
Never	15 (3.0%)	113.6	11 (1.7%)	106.0
Exercise experience in junior high school	450 (89.8%)		431 (67.1%)	

*The significant differences by one-way ANOVA followed by post-hoc Tukey's compromise tests and $p < 0.05$.

the calcaneus QUS-SI in adolescents. The relationships of prenatal and/or postnatal factors and lifestyle status with the calcaneus QUS-SI of subjects of both sexes were analyzed using a multiple regression analysis after adjustment for age, bed time, average duration of sleep, and calcium-rich food intake. The results of this analysis are displayed in Table 3.

1. Model I : There was no significant association between any prenatal or postnatal factor and the calcaneus QUS-SI in boys. However, a high maternal weight gain and a short gestational length were both significantly associated with high calcaneus QUS-SI in girls analyzed by both the simple regression analysis ($p = 0.001$, $p = 0.006$, respectively) and multiple regression analysis ($p = 0.014$, $p = 0.042$, respectively.). The multiple regression models explained about 19% and 7% of the variance of boys and girls,

Table 3 The effect of prenatal and/or postnatal factors, acquired gain factors, and lifestyle status on calcaneus QUS-SI in both genders by a multiple regression analysis

Boys n = 501

	Factors	β	*p* values
Model Ⅰ	Birth weight(g)	0.11	0.154
	Birth height(cm)	−0.05	0.512
	Pre-pregnancy weight (kg)	−0.01	0.867
	Maternal height (cm)	−0.07	0.118
	Maternal weight gain (kg)	−0.06	0.139
	Neonatal weight gain at 1 month (g)	0.05	0.256
	Neonatal weight gain at 4 months (g)	0.01	0.913
	Gestational length (days)	−0.06	0.307
Model Ⅱ	Birth weight(g)	0.09	0.240
	Birth height(cm)	−0.05	0.517
	Pre-pregnancy weight (kg)	−0.02	0.737
	Maternal height (cm)	−0.08	0.101
	Maternal weight gain (kg)	−0.07	0.115
	Neonatal weight gain at 1 month (g)	0.05	0.297
	Neonatal weight gain at 4 months (g)	0.00	0.966
	Gestational length (days)	−0.03	0.571
	Secondary sexual characteristics (years)*	−0.07	0.102
	Grip strength (kg)	0.21	**<0.0001**
	Weight (kg)	−0.03	0.615
	Height (cm)	−0.04	0.535
Model Ⅲ	Birth weight(g)	0.11	0.170
	Birth height(cm)	−0.06	0.409
	Pre-pregnancy weight (kg)	−0.01	0.792
	Maternal height (cm)	−0.06	0.258
	Maternal weight gain (kg)	−0.09	0.053
	Neonatal weight gain at 1 month (g)	0.02	0.661
	Neonatal weight gain at 4 months (g)	0.01	0.915
	Gestational length (days)	−0.01	0.840
	Secondary sexual characteristics (years)*	−0.08	0.079
	Grip strength (kg)	0.19	**0.001**
	Weight (kg)	−0.00	0.955
	Height (cm)	−0.06	0.373
	Exercise habits in junior high school	0.08	0.067

Model Ⅰ : Adjusted $R^2 = 0.19$ ($p < 0.0001$), Model Ⅱ : Adjusted $R^2 = 0.23$ ($p < 0.0001$), Model Ⅲ : Adjusted $R^2 = 0.25$ ($p < 0.0001$)
Model Ⅰ, Model Ⅱ : Adjusted for age
Model Ⅲ : Adjusted for age, bed time, average duration of sleep and calcium-rich food intake
*The age when pubic hair started to appear.

Girls n = 642

	Factors	β	*p* values
Model Ⅰ	Birth weight(g)	0.12	0.061
	Birth height(cm)	−0.10	0.103
	Pre-pregnancy weight (kg)	0.03	0.493
	Maternal height (cm)	0.05	0.228
	Maternal weight gain (kg)	0.10	**0.014**
	Neonatal weight gain at 1 month (g)	0.06	0.178
	Neonatal weight gain at 4 months (g)	0.02	0.642
	Gestational length (days)	−0.10	**0.042**
Model Ⅱ	Birth weight(g)	0.07	0.301
	Birth height(cm)	−01.0	0.135
	Pre-pregnancy weight (kg)	−0.02	0.612
	Maternal height (cm)	0.02	0.714
	Maternal weight gain (kg)	0.10	**0.021**
	Neonatal weight gain at 1 month (g)	0.04	0.409
	Neonatal weight gain at 4 months (g)	0.01	0.851
	Gestational length (days)	−0.10	0.059
	Secondary sexual characteristics (years)*	−0.07	0.082
	Grip strength (kg)	0.15	**0.001**
	Weight (kg)	0.12	**0.013**
	Height (cm)	0.02	0.639
Model Ⅲ	Birth weight(g)	0.05	0.506
	Birth height(cm)	−0.09	0.142
	Pre-pregnancy weight (kg)	−0.02	0.701
	Maternal height (cm)	0.04	0.441
	Maternal weight gain (kg)	0.10	**0.026**
	Neonatal weight gain at 1 month (g)	0.03	0.492
	Neonatal weight gain at 4 months (g)	0.00	0.999
	Gestational length (days)	−0.09	0.094
	Secondary sexual characteristics (years)*	−0.08	**0.047**
	Grip strength (kg)	0.11	**0.013**
	Weight (kg)	0.14	**0.005**
	Height (cm)	0.01	0.791
	Exercise habits in junior high school	0.15	**<0.0001**

Model Ⅰ: Adjusted $R^2 = 0.07$ ($p < 0.0001$), Model Ⅱ: Adjusted $R^2 = 0.10$ ($p < 0.0001$), Model Ⅲ: Adjusted $R^2 = 0.14$ ($p < 0.0001$)
Model Ⅰ, Model Ⅱ: Adjusted for age
Model Ⅲ: Adjusted for age, bed time, average duration of sleep and calcium-rich food intake
*The age when menstruation started.

respectively.

2. Model Ⅱ: A simple regression analysis showed a significant relationship of grip strength to calcaneus QUS-SI among boys ($p < 0.0001$). In model Ⅱ, the age of development of secondary sexual characteristics, grip strength, height, and weight were further included. The model for boys indicated that grip strength was a strong correlative of calcaneus QUS-SI ($p < 0.0001$), which explained 23% of the variation of calcaneus QUS-SI. The model for girls revealed that maternal weight gain during pregnancy, grip strength, and body weight were important correlative factors of calcaneus QUS-SI (10%) ($p = 0.021$, $p = 0.001$, and $p = 0.013$, respectively.). These results agreed with those by the simple regression analysis ($p = 0.006$, $p < 0.0001$, and $p < 0.0001$, respectively).

3. Model Ⅲ: Regular exercise was further included in this model in addition to the variables in model Ⅰ and Ⅱ. The model for boys again indicated grip strength was a strongly correlative factor of calcaneus QUS-SI ($p = 0.001$), which agreed with the result by the simple regression analysis ($p < 0.0001$). The model for girls showed that maternal weight gain during pregnancy, early menstruation, grip strength, body weight, and exercise habits in junior high school were strongly correlative factors for calcaneus QUS-SI ($p = 0.026$, $p = 0.047$, $p = 0.013$, $p = 0.005$, and $p < 0.0001$, respectively.). These results were consistent with those by the simple regression analysis ($p = 0.001$, $p = 0.048$, $p < 0.0001$, $p = 0.005$, and $p < 0.0001$, respectively). This multiple regression model explained about 25% and 14% of the variance of calcaneus QUS-SI in boys and girls, respectively.

The relation between the specific maternal weight gain during pregnancy and the calcaneus QUS-SI in girls. One-way ANOVA with post-hoc Tukey's compromise tests indicated that, in girls, the groups with maternal weight gain of 11 kg-15.9 kg or over 16 kg had a significantly higher QUS-SI than those of under 6 kg or 6 kg-10.9 kg (Fig. 1).

Discussion

The present study demonstrates that maternal weight gain during pregnancy, one of several prenatal factors tested, was positively associated with the calcaneus QUS-SI in adolescent girls, even after adjustment for age, bed time, average duration of sleep, and calcium-rich food intake. Such an association was independent of height, weight, other prenatal (maternal height, pre-pregnancy weight, and gestational length) and postnatal factors (neonatal weight gain, grip strength, secondary sex characteristic, *etc.*), and exercise. In addition, we found that maternal weight gain of more than 11 kg had a significantly higher QUS-SI compared to that of less than 11 kg gain. Most existing literature on childhood BMC or BMD has focused on the effects of birth weight and height [10–17], while observational studies regarding the effects of maternal weight gain during pregnancy on adolescents' bone health are limited. The present findings imply that excessive restriction of maternal weight gain might have a negative effect on the calcaneus QUS-SI of the girls. However, we did not find any significant association between maternal weight gain during pregnancy and calcaneus QUS-SI in boys. Such a gender-based difference may be partially explained by the different timing in boys and girls of peak QUS-SI [30]. The study by Magarey *et al.* showed that peak bone mineral density can be acquired by 17 years of age in girls but 1–2 years later in boys, and that the prolonged response to testosterone in boys might be the source of this difference [31]. The second explanation is based on another work of Magarey *et al.* showing that by age 11 to 17 years, girls had attained over 90% of their mothers' BMC in forearm bone, whereas the same age-range boys had attained only 80% or more of their fathers' BMC [32]. Information on the fathers' genetic factors might be helpful when evaluation of adolescent boys' bone mass. Unfortunately, the fathers' data were not available in the present study.

The present findings also revealed that high grip strength was the only factor significantly associated with a high calcaneus QUS-SI in both boys and girls. This result is consistent with the research by Proctor *et al.*, who found a positively significant association between grip strength and total body bone mineral [33]. Aoyagi *et al.* observed that changes of BMD and grip strength or other physical performance measures tended to be correlated [34]. Rauch and Schoenau have pointed out that both longitudinal growth and muscle forces are the primary factors to which bone adapts its strength based on a functional model of bone development [35]. A 7-year longitudinal study by Xu *et al.* implied the possibility of the muscle-bone rela-

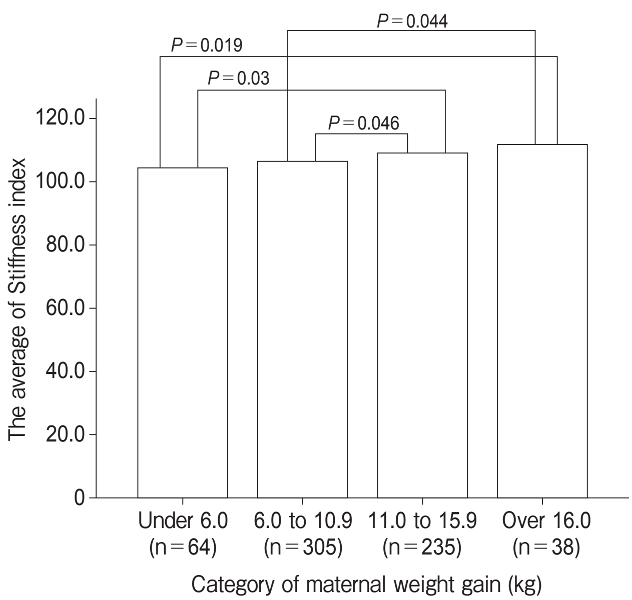

Fig. 1 The effects of the specific maternal weight gain on calcaneus QUS-SI of girls determined by a one-way ANOVA with post-hoc Tukey compromise test.

tionship during puberty increasing endosteal bone formation and bone tissue mineralization [36]. These lines of evidence indicate how exercise can improve muscle strength and benefit later bone development. In fact, a regular exercise habit in junior high school was significantly associated with a high calcaneus QUS-SI in girls (Table 3) even though the association was not significant in boys. However, exercise habit during adolescence and acquisition of muscle strength are important for both genders to gain higher calcaneus QUS-SI. A recent study showed that the effect of physical activity on bone mass could be mediated more by the kind of physical activity than by the total amount [37]. High-intensity, high-impact and weight-bearing physical activities are considered as "osteogenic activities", having more positive influence on bone mass in adolescents [38]. Kato and Umemura also reported that weight-bearing exercise before and in early puberty is helpful in strengthening the proximal femur of young adult women [39]. Since we had no information on the type of exercise of the subjects, further study is needed to confirm the effect of exercise on adolescent boys' calcaneus QUS-SI.

We also found that the mean age at menarche was 11.9 ± 1.1 years in adolescent girls and early menstruation was associated with high calcaneus QUS-SI, suggesting that female hormones affect the calcaneus QUS-SI, which was in agreement with the previous studies [36, 40].

The effect of body weight in girls was found to be an important determinant of calcaneus QUS-SI in our study. A similar result was found by Babaroutsi *et al.* [4], indicating that load stress on bone is important to gain calcaneus QUS-SI [4].

In addition, measuring the calcaneus QUS-SI is important for estimating the growth of the long bones, which is related to the expansion of body size [30]. The lengths of the long bones reach their peak in the late twenties, and the growth of the long bones is correlated with the calcaneus QUS-SI. This means if we help adolescents reach a higher peak bone mass on the calcaneus QUS-SI, it may be helpful for the growth of the long bones and/or reduce the potential for bone fracture.

The present study was subject to several limitations. First, we chose an ultrasound bone densitometer, the least invasive technique, for measurement of bone mass instead of DXA scan (a preferred technique for measuring BMD) since the participants were still immature. Second, we were not able to prove a cause-and-effect relation between pre- and postnatal factors, lifestyle factors and calcaneus QUS-SI because of the cross-sectional nature of our study. Third, some information such as genetic factors of the fathers, type of exercise by the subjects, and maternal smoking were not available. Further studies with the addition of these data may help to confirm the importance of maternal weight gain and early physical activity to adolescent calcaneus QUS-SI found in the present study.

In conclusion, we found an independent effect of maternal weight gain during pregnancy on calcaneus QUS-SI in adolescent girls, and a positive association between grip strength and calcaneus QUS-SI in both sexes. These findings imply excessive restriction of maternal weight gain would have a negative effect on the calcaneus QUS-SI of offspring bone health and that early physical activity/exercise helps to maximize peak bone mass and possibly reduce the risk of osteoporosis in later life. There is a need for further longitudinal studies to provide supportive evidence.

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References

1. Go G: The value of bone mass screening in schoolhood and adolescence under ultrasound bone densitometer. The Cent Jpn J Orthop Surg Traumatol (2000) 43: 843–853.
2. Yung PS, Lai YM, Tung PY, Tsui HT, Wong CK, Hung VW and Qin L: Effects of weight bearing and non-weight bearing exercises on bone properties using calcaneal quantitative ultrasound. Br J Sports Med (2005) 39: 547–551.
3. Babaroutsi E, Magkos F, Manios Y and Sidossis LS: Body mass index, calcium intake, and physical activity affect calcaneal ultrasound in healthy Greek males in an age-dependent and parameter-specific manner. J Bone Miner Metab (2005) 23: 157–166.
4. Babaroutsi E, Magkos F, Manios Y and Sidossis LS: Lifestyle factors affecting heel ultrasound in Greek females across different life stages. Osteoporos Int (2005) 16: 552–561.
5. Hirota T, Kusu T and Hirota K: Improvement of nutrition stimulates bone mineral gain in Japanese school children and adolescents. Osteoporos Int (2005) 16: 1057–1064.
6. Rautava E, Lehtonen-Veromaa M, Möttönen T, Kautiainen H, Heinonen OJ and Viikari J: Association of reduced physical activity and quantitative ultrasound measurements: a 6-year follow-up study of adolescent girls. Calcif Tissue Int (2006) 79: 50–56.
7. Robinson ML, Winters-Stone K, Gabel K and Dolny D: Modifiable

lifestyle factors affecting bone health using calcaneus quantitative ultrasound in adolescent girls. Osteoporos Int (2007) 18: 1101–1107.
8. Laabes EP, Vanderjagt DJ, Obadofin MO, Sendeht AJ and Glew RH: Assessment of the bone quality of black female athletes using quantitative ultrasound. J Sports Med Phys Fitness (2008) 48: 502–508.
9. Cooper C, Cawley M, Bhalla A, Egger P, Ring F, Morton L and Barker D: Childhood growth, physical activity, and peak bone mass in women. J Bone Miner Res (1995) 10: 940–947.
10. Rohana J, Hasmawati J and Zulkifli SZ: Risk factors associated with low bone mineral content in very low birth weight infants. Singapore Med J (2007) 48: 191–194.
11. Beltrand J, Alison M, Nicolescu R, Verkauskiene R, Deghmoun S, Sibony O, Sebag G and Lévy-Marchal C: Bone mineral content at birth is determined both by birth weight and fetal growth pattern. Pediatr Res (2008) 64: 86–90.
12. Fricke O, Semler O, Stabrey A, Tutlewski B, Remer T, Herkenrath P and Schoenau E: High and low birth weight and its implication for growth and bone development in childhood and adolescence. J Pediatr Endocrinol Metab (2009) 22: 19–30.
13. Cooper C, Fall C, Egger P, Hobbs R, Eastell R and Barker D: Growth in infancy and bone mass in later life. Ann Rheum Dis (1997) 56: 17–21.
14. Dennison EM, Syddall HE, Sayer AA, Gilbody HJ and Cooper C: Birth weight and weight at 1 year are independent determinants of bone mass in the seventh decade: the Hertfordshire cohort study. Pediatr Res (2005) 57: 582–586.
15. Cooper C, Harvey N, Cole Z, Hanson M and Dennison E: Developmental origins of osteoporosis: the role of maternal nutrition. Adv Exp Med Biol (2009) 646: 31–39.
16. Tobias JH, Steer CD, Emmett PM, Tonkin RJ, Cooper C and Ness AR; ALSPAC study team: Bone mass in childhood is related to maternal diet in pregnancy. Osteoporos Int (2005) 16: 1731–1741.
17. Jones G and Dwyer T: Birth weight, birth length, and bone density in prepubertal children: evidence for an association that may be mediated by genetic factors. Calcif Tissue Int (2000) 67: 304–308.
18. Gluckman PD and Pinal CS: Regulation of fetal growth by the somatotrophic axis. J Nutr (2003) 133: 1741S–1746S.
19. Tobias JH and Cooper C: PTH/PTHrP activity and the programming of skeletal development in utero. J Bone Miner Res (2004) 19: 177–182.
20. Kapoor A, Dunn E, Kostaki A, Andrews MH and Matthews SG Fetal programming of hypothalamo-pituitary-adrenal function: prenatal stress and glucocorticoids. J Physiol (2006) 572: 31–44.
21. Davies JH, Evans BA and Gregory JW: Bone mass acquisition in healthy children. Arch Dis Child (2005) 90: 373–378.
22. Greenspan SL, Bouxsein ML, Melton ME, Kolodny AH, Clair JH, Delucca PT, Stek M Jr, Faulkner KG and Orwoll ES: Precision and discriminatory ability of calcaneal bone assessment technologies. J Bone Miner Res (1997) 12: 1303–1313.
23. Díez-Pérez A, Marín F, Vila J, Abizanda M, Cervera A, Carbonell C, Alcolea RM, Cama A, Rama T, Galindo E and Olmos C: Evaluation of calcaneal quantitative ultrasound in a primary care setting as a screening tool for osteoporosis in postmenopausal women. J Clin Densitom (2003) 6: 237–245.
24. López-Rodríguez F, Mezquita-Raya P, de Dios Luna J, Escobar-Jiménez F and Muñoz-Torres M: Performance of quantitative ultrasound in the discrimination of prevalent osteoporotic fractures in a bone metabolic unit. Bone (2003) 32: 571–578.
25. Ahuja SP, Greenspan SL, Lin Y, Bowen A, Bartels D and Goyal RK: A pilot study of heel ultrasound to screen for low bone mass in children with leukemia. J Pediatr Hematol Oncol (2006) 28: 427–432.
26. Pinheiro MM, Castro CM and Szejnfeld VL: Low femoral bone mineral density and quantitative ultrasound are risk factors for new osteoporotic fracture and total and cardiovascular mortality: a 5-year population-based study of Brazilian elderly women. J Gerontol A Biol Sci Med Sci (2006) 61: 196–203.
27. Bauer DC, Ewing SK, Cauley JA, Ensrud KE, Cummings SR and Orwoll ES: Osteoporotic Fractures in Men (MrOS) Research Group., Quantitative ultrasound predicts hip and non-spine fracture in men: the MrOS study. Osteoporos Int (2007) 18: 771–777.
28. Trimpou P, Bosaeus I, Bengtsson BA and Landin-Wilhelmsen K: High correlation between quantitative ultrasound and DXA during 7 years of follow-up. Eur J Radiol (2010) 73: 360–364.
29. Ministry of Health, Labor and Welfare: Dietary guidelines for pregnant women (2006) (in Japanese).
30. Kozuka K, Nagatsuka M, Moro N, FUjihara T, Shirato N, Okuyama D, Chiba H, Saito H and Yanaihara T: Changes in Bone Mineral Density in Normal Pubertal Boys and Girls. Acta Obst Gynaec Jpn (2000) 52: 669–675. (in Japanese)
31. Magarey AM, Boulton TJ, Chatterton BE, Schultz C, Nordin BE and Cockington RA: Bone growth from 11 to 17 years: relationship to growth, gender and changes with pubertal status including timing of menarche. Acta Paediatr (1999) 88: 139–146.
32. Magarey AM, Boulton TJ, Chatterton BE, Schultz C and Nordin BE: Familial and environmental influences on bone growth from 11-17 years. Acta Paediatr (1999) 88: 1204–1210.
33. Proctor DN, Melton LJ, Khosla S, Crowson CS, O'Connor MK and Riggs BL: Relative influence of physical activity, muscle mass and strength on bone density. Osteoporosis Int (2000) 11: 944–952.
34. Aoyagi K, Ross PD, Hayashi T, Okano K, Moji K, Sasayama H, Yahata Y and Takemoto T: Calcaneus bone mineral density is lower among men and women with lower physical performance. Calcif Tissue Int (2000) 67: 106–110.
35. Burr DB: Muscle Strength, Bone Mass, and Age-Related Bone Loss. J Bone Miner Res. (1997) 12: 1547–1551.
36. Xu L, Nicholson P, Wang Q, Alén M and Cheng S: Bone and muscle development during puberty in girls: a seven-year longitudinal study. J Bone Miner Res (2009) 24: 1693–1698.
37. Vicente-Rodríguez G: How does exercise affect bone development during growth? Sports Med (2006) 36: 561–569.
38. Gracia-Marco L, Moreno LA, Ortega FB, León F, Sioen I, Kafatos A, Martinez-Gomez D, Widhalm K, Castillo MJ and Vicente-Rodríguez G; HELENA Study Group: Levels of physical activity that predict optimal bone mass in adolescents: the HELENA study. Am J Prev Med (2011) 40: 599–607.
39. Kato T and Umemura Y: Childhood Sports Activity Induces Bone Strength in Young Premenopausal Women. School Health (2011) 7: 8–15.
40. Vicente-Rodríguez G, Urzanqui A, Mesana MI, Ortega FB, Ruiz JR, Ezquerra J, Casajús JA, Blay G, Blay VA, Gonzalez-Gross M and Moreno LA; AVENA-Zaragoza Study Group: Physical fitness effect on bone mass is mediated by the independent association between lean mass and bone mass through adolescence: a cross-sectional study. J Bone Miner Metab (2008) 26: 288–294.