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Abstract

Background: Early atherosclerotic change is found even in childhood, and there is an urgent need to clarify the factors causing childhood atherosclerosis and take preventive measures. Early detection of the contributing risk factors is crucial for preventive measures to be undertaken. Pulse wave velocity (PWV) is a widely used technique method for the assessment of atherosclerosis in children.

Methods: Lifestyle questionnaire, and baPWV values and anthropometric data were obtained from junior high school students in an urban area of Japan over a period of 3 years between 2006 and 2008.

Results: Mean baPWV values increased from 867.4 ± 99.5 m/s to 944.5 ± 117.5 m/s m/s in boys, and 864.0 ± 99.5 m/s to 923.0 ± 101.3 m/s in girls. Obese students had higher baPWV compared with non-obese students in both genders across each grade. Logistic regression analysis using third grade student data revealed that high baPWV was dependent on SBP, time for television (TV) and symptoms of depression and anxiety, whereas low baPWV was dependent on time for games, light exercise, sleep and indoor play, as well as good friendship and motivation.

Conclusion: SBP, time for watching TV, and symptoms of depression and anxiety may contribute to arterial stiffness and have relation with obesity in junior high school students.

Key words: pulse wave velocity, atherosclerosis, lifestyle, junior high school students, longitudinal study

Abbreviations

baPWV : brachio-ankle pulse wave velocity

cfPWV: carotid-femoral pulse wave velocity

BMI : body mass index

ABI : ankle brachial index

CIMT: carotid intima-media thickness

FMD: flow mediated dilatation

CAVI: cardio ankle vascular index

SBP: systolic blood pressure

DBP: diastolic blood pressure

Introduction

Childhood obesity is considered as "a crisis in public health"[1]; the prevalence of obesity and its comorbidities in children is expected to increase in the future[2]. Adult cardiovascular disease is likely to occur in individuals possessing risk factors for cardiovascular disease in their childhood[3][4]. Childhood obesity is one such risk factor and the target for preventive measures[5]. Recently, various non-invasive

techniques that determine carotid intima-media thickness (CIMT), flow mediated dilatation (FMD), and pulse wave velocity (PWV) have been developed to estimate atherosclerotic change[6]. Several studies have reported on the presence of atherosclerotic change in obese children[7)8)9)]. Although CIMT and FMD are reliable techniques for the detection of atherosclerosis in children, they require time and skilled experience. Since PWV is a relatively quick procedure that is easy to operate and reproduce, many studies have used PWV to assess atherosclerosis in children[10)11)12)13)14)15)].

Aging is an inevitable and integral factor of childhood; therefore, the results obtained from these studies should be interpreted carefully[16)]. Few longitudinal studies[17)18)19)] have reported on the risk factors of atherosclerotic change in children. Here we have the opportunity to conduct a longitudinal study on the effect of lifestyle on PWV in junior high school students over a period of 3 years.

Subjects and methods

Subjects: The study consisted of 1729 junior high school students (930 boys and 799 girls) in Okayama city, Japan. Written informed consent was obtained from the students and their guardians. This study was approved by the ethical committee of Kyoto Prefectural University of Medicine (ERB-C-624-1).

Anthropometric data, data from a lifestyle questionnaire and measurement of baPWV values were obtained annually over a period of 3 years at the same time in school hours of "integrated study".

Methods: baPWV, blood pressure (SBP, DBP) and heart rate were measured using an electromyography device (form BP-203RPEIII, Omron-Cohlin Co., Ltd., Tokyo, Japan). Following a resting period of 5 min, baPWV was measured in each arm simultaneously, and the mean baPWV value was recorded. Body fat percentage (%) was measured using a bioelectrical impedance analyzer (SS-103, Sekisui Chemical Co., Ltd., Tokyo, Japan). Percentage of overweight was calculated using the following equation[20]:

$$\text{Percentage of overweight (\%)} = (\text{measured weight} - \text{standard weight}) / \text{standard weight} \times 100$$

Standard weight was determined as the mean age and height of each gender, in accordance with the Japanese school health report in the year 2000. Obese students were defined as those having a percentage of overweight > 20%. The lifestyle questionnaire was designed according to the report of surveillance of health in school children and adolescents [21] conducted by Japan Society of School Health, to which psychological factors and motivational status were added. Time for indoor play means time spent on reading book or listening to music in the room after school, excluding the time for watching TV, and games. Vigorous exercise means exercise with lasting fatigue till the next day, and light exercise means exercise without fatigue. These comments had been described in the questionnaire. Data were obtained from the same students as they progress through school from the year 2006 (when

they are in the first grade) to 2008 (when they are in the third grade). None of the students moved or dropped out during the 3 years of the study.

Statistical analysis: Statistical analyses were performed using SPSS 22.0 (IBM). The impact of gender differences on baPWV was addressed by assessing the measured variables separately for each gender type. Mean values were compared using either t-test or analysis of variance (ANOVA) and the Bonferroni post-hoc test. Difference of means among each grade was tested by repeated measures ANOVA. Significance was defined as $p < 0.05$. Logistic regression analyses were performed to assess the relationship between baPWV and the parameters measured in this study. The variable reduction method was applied to all of the variables measured.

Results

Table 1 summarizes the subject characteristics obtained. There were significant gender differences in height, weight, waist size, waist/height ratio, obesity index, body fat %, SBP, heart rate and sleep time (duration) across each grade. However, gender differences in BMI, DBP, and baPWV were only found for third grade students. There were differences in height, weight, waist circumference, percent of overweight, body fat %, BMI, heart rate, baPWV, ABI and time for sleep between genders across each grade. For both genders, decreased sleeping time, time spent on vigorous exercise, time for game, and

increase in the time for studying were found from the second grade to the third grade. Significant correlations were found in time for studying vs time for games and time for watching TV (time for studying vs time for games: boys r = −0.139** and girls r = −0.155**; time for studying vs time for watching TV: boys r = −0.091** and girls r = −0.117**) in the third grade. Increase in the time for studying was inversely related to the increase in time for games (boys r = −0.168** and girls r = −0.128**).

BaPWV values were higher in obese students compared to non-obese students in each gender and grade (Table 2). BaPWV data were grouped by percentile and categorized into the following: G1 (baPWV below the 25th percentile, low), G2 (baPWV between the 25th and 75th percentiles, normal) and G3 (baPWV above the 75th percentile, high). In boys, high baPWV was associated with increases in weight, percent of overweight, body fat %, blood pressure, and heart rate (Table 3). In girls, high baPWV was associated with increases in body fat %, blood pressure and heart rate (Table 3).

Depending on the change in baPWV over the three year period, students were divided into four groups: group A (normal to normal), group B (normal to high), group C (high to normal), and group D (high to high), whereby “normal” was defined as having a baPWV below the 75th percentile, and “high” was defined as having a baPWV above the 75th percentile. As shown in Table 4, SBP, DBP, and heart rate

were significantly higher in group D compared to group A for each gender and grade. In boys, time for watching TV was significantly higher in group D compared to group A.

Logistic regression analyses revealed that (1) baPWV was inversely associated with baPWV increment from the first grade to the third grade (Table 5), and, (2) that baPWV in the first and second grade was predictive of baPWV in the third grade. Logistic regression analysis using baPWV data from students across all grades revealed that the odds ratio (OR) was highest for baPWV in first grade students in both genders (boys: $OR^{\dagger} = 3.24$, girls: $OR^{\dagger} = 3.51$) (Table 5). In boys, this was followed by weight (OR = 1.85), body fat% ($OR^{\dagger} = 1.76$) , and BMI (OR = 1.68) in second grade students, and, TV viewing duration (OR = 1.54), orthostatic dysfunction (OR = 1.52) , and general fatigue(OR = 1.50) in first grade students; In girls, this was followed by study duration in first grade students (OR = 1.45), degree of concentration (OR = 1.42), motivation ($OR^{\dagger} = 1.41$) , and weight (OR = 1.31) in second grade students, and having a pale face ($OR^{\dagger} = 1.35$) and time spent on vigorous exercise (OR = 1.33) in first grade students (Table 5).

Logistic regression analysis was performed using the objective variables as baPWV value at the third grade (Table 6). In both boys and girls, high odds ratios were found in baPWV at the second grade (boys: OR = 3.90 and girls: OR = 3.98) and baPWV at the first grade (boys: OR = 2.64 and girls: OR = 3.71). In boys, they were followed by feel sick at bathing at the second grade (OR = 1.80), time for

games at the second grade (OR† = 1.78), time for light exercise at the first grade (OR† = 1.62), SBP at the first and second grade (OR = 1.62, 1.60), sleep time at the first grade (OR† = 1.58), headache (OR = 1.56), time for watching TV (OR = 1.51) at the first grade, dizziness (OR† = 1.46), general fatigue (OR = 1.44), time for indoor play (OR† = 1.40), anxiety for action at the second grade (OR = 1.40), and good friendship at the first grade (OR† = 1.39). In girls, they were followed by SBP at the second grade (OR = 1.96), having breakfast at the first grade (OR = 1.89), lack of sleep at the second grade (OR = 1.89), emotional ups and downs at the first grade (OR = 1.71), ABI at the first grade (OR = 1.60), concentration at the second grade (OR = 1.60), having motivation at the second grade (OR† = 1.54), feel sick during morning (OR = 1.52), anxiety for action (OR = 1.44), and time for games at the first grade (OR† = 1.39).

Discussion

In this study, statistical analysis was conducted by gender as there is much difference in physical growth, blood pressure, and baPWV between boys and girls[10)22)].No significant differences in baPWV were found between genders in the first and second grade. However, third grade data showed significant gender differences in baPWV, and blood pressure, suggesting the effect of sex hormones on vascular elasticity. Estrogen may inhibit atherosclerosis by increasing nitric oxide production and suppressing pathological proliferation of vascular smooth muscle[23)]. The mean age of menarche is reported to be

12.2 years in Japan[24)] however, the pubertal stage including the age of menarche was not checked in this study. Therefore we could not estimate the effect of estrogen on PWV in this study.

The increase in baPWV between the second and third grade was larger than that of the first to second grade. This may be explained by increased stress and anxiety from the high school entrance test, and decreased sleeping time and time spent on vigorous exercise resulted from increased time for studying as shown in Table1.

In the first grade, baPWV was inversely associated with baPWV increment in both genders. In the second grade, weight was positively associated with baPWV increment in both genders. These results suggest that weight gain from the first grade to the second grade may be associated with baPWV increment. In boys, time for watching TV was associated with an OR of 1.54 for baPWV increment. Increased time for watching TV may result in reduced sleep and exercise, and an increase in sedentary time. In girls, time for studying, concentration and time spent on vigorous exercise were all positively associated with baPWV increment. These activities require mental strain, which may result in increased sympathetic activity and consequentially result in baPWV increment. There is a possibility that many of the students spent a lot of time spent on vigorous exercise at the first grade continued their sports activity until the third grade. Being successful at both study and club activities might require reduction in time for games, indoor play, and sleep.

It is well known that hypertension and dyslipidemia are key atherosclerosis risk factors in children[25)26)]. These risk factors tend to be present in obese children. In this study, obese students had higher baPWV than non-obese students, suggesting that obesity may play an important role in the development of atherosclerosis in children. Our results were consistent with studies by Cote et al.[13)] and Hudson et al.[12)], who similarly concluded that obese children had higher PWV compared to non-obese children.

Our results also showed that there was a positive association between baPWV in the first and second grade and baPWV in the third grade; Therefore, students with high baPWV in the third grade were likely to have had high baPWV in the first and second grade (and vice versa). SBP was also associated with baPWV, particularly in boys; SBP in the first and second grade was predictive of baPWV in the third grade. Interestingly, although the obese students had higher baPWV than non-obese students. Logistic regression analysis revealed that obesity itself was not a significant contributor toward baPWV. Taken together, these results suggest that perhaps it is not obesity itself, but rather the factors associated with obesity such as hypertension, reduced time for exercise, increased time for watching TV, and, symptoms of depression, that have an impact on baPWV.

Hypertension is an important risk factor for high baPWV in both boys and girls[27)28)29)]. Continued hypertension may result in pathophysiological changes such as thickening of the intima and media, which can result in functional changes and PWV elevation[26)]. It is important to note that although measurement of PWV is a widely accepted technique used to assess for arterial stiffness, its accuracy is influenced by changes in blood pressure. As a result, the use of the CAVI, which is based on the

stiffness parameter and is theoretically independent of blood pressure, has been favored by some investigators[30)31)]. However, some studies using the CAVI in children have reported lower CAVI values in obese children compared to non-obese children, proposing that vascular adaptation to obesity had occurred[30)].

Obesity and hypertension are independent risk factors of atherosclerosis[26)27)]; Importantly, childhood hypertension has been associated with atherosclerosis in adulthood[25)28)]. Similarly, the relationship between PWV and obesity in children has been well studied[10)11)15)]. In our study, we assessed the contribution of various parameters toward the increased baPWV observed in junior high school students. Our results revealed that high baPWV was associated with reduced time for light exercise and increased time for watching TV; These factors are consistent with reduced activity, and promote obesity and arterial stiffness[32)33)]. Additionally, reduced time for sleep, anger and symptoms of anxiety may contribute to sympathetic excitation, resulting in elevation of blood pressure and heart rate, promoting arterial stiffness. In children, reduced time for sleep has been associated with obesity and atherosclerosis[34)35)]. In this study, many psychological symptoms such as feeling sick during bath time, headaches, general fatigue, anxiety, and emotional ups and downs were associated with increased baPWV. Psychological stresses have been previously reported to induce transient endothelial dysfunction[36)]. Symptoms of depression and anxiety have also been reported to mediate subclinical atherosclerosis[37)38)]. In young females, anger has been shown to raise blood pressure and have an impact

on cardiotoxic autonomic and hemodynamic profiles[39]. These investigators reported that trait forgiveness was associated with a more cardioprotective profile, and proposed that decreasing anger while increasing forgiveness may have clinical relevance; this suggests that stress-coping education and the implementation of interventional programs aimed at suppressing anger may be useful in the prevention of atherosclerosis in children.

In our study, increased game and indoor play was associated with low baPWV. This was in contrast to previous reports that time for watching TV was associated with high baPWV[40]. Time for watching TV, much like time for games and indoor play, is considered as sedentary time[41]. However, in our study, time for watching TV was twice the length of time for games or indoor play, compromising time for sleep and exercise. In contrast, there are many elements of game and indoor play that may have a positive influence on mood[42], and be considered not only as an effective relaxation tool, but also as a useful means for establishing good friendship.

Taken together, our results demonstrate that continued sympathetic stress through elevation of blood pressure, increased in heart rate, symptoms of nervousness, anxiety, and anger, may contribute toward high baPWV. Activities which promote parasympathetic activity such as adequate time for sleep, light exercise, and, game and indoor play with friends may lower baPWV, and consequently reduce the risk for atherosclerosis risk. We therefore propose that implementation of preventive measures such as

regular measurement of blood pressure, adequate sleep[43)44)], lifestyle changes, and the introduction of stress-coping educational and interventional programs, may reduce the risk of early development of atherosclerosis in children.

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Disclosure

The authors declare no conflict of interest.

Author contribution

HF contributed to the conception and design of this study; HA and KY performed the statistical analysis. FI drafted the manuscript. HN and KK critically reviewed the manuscript and supervised the whole study process. All authors read and approved the final manuscript.

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Table 1: Subject characteristics.

		1st grade				2nd grade			3rd grade			Repeated measures anova (ranova)			
	Gender	N	Mean	SD	Boys vs girls	Mean	SD	Boys vs girls	Mean	SD	Boys vs girls	p value	1st vs 2nd	1st vs 3rd	2nd vs 3rd
Age (years)	Boys	930	12.5	.5	ns	13.5	.5	ns	14.5	.5	ns	**	✓	✓	✓
	Girls	799	12.6	.5		13.6	.5		14.6	.5		**	✓	✓	✓
Height (cm)	Boys	930	154.9	8.2	**	161.9	7.4	**	166.3	6.3	**	**	✓	✓	✓
	Girls	799	152.9	5.7		155.5	5.3		156.6	5.3		**	✓	✓	✓
Weight (kg)	Boys	930	45.5	10.4	**	51.3	11.2	**	56.4	11.4	**	**	✓	✓	✓
	Girls	799	43.6	7.5		47.1	7.4		49.2	7.4		**	✓	✓	✓
Waist circumference(cm)	Boys	928	66.8	8.8	**	69.1	9.1	**	71.5	9.1	**	**	✓	✓	✓
	Girls	798	65.0	6.8		67.9	7.0		68.6	6.8		**	✓	✓	✓
Waist/height ratio	Boys	928	.431	.051	**	.427	.051	**	.430	.051	**	**	✓		✓
	Girls	798	.425	.041		.437	.043		.438	.042		**	✓	✓	
Percent of overweight(%)	Boys	930	-1.9	15.8	**	1.3	16.7	**	3.2	17.4	**	**	✓	✓	✓
	Girls	799	-3.8	13.6		-1.2	13.6		-1.1	13.1		**	✓	✓	
Body fat(%)	Boys	928	15.9	5.8	**	15.1	6.0	**	15.3	6.3	**	**	✓	✓	
	Girls	797	20.8	4.9		21.6	4.3		22.1	4.8		**	✓	✓	✓
BMI (kg/m^2)	Boys	930	18.8	3.2	ns	19.5	3.4	ns	20.3	3.5	*	**	✓	✓	✓
	Girls	799	18.6	2.6		19.4	2.7		20.0	2.7		**	✓	✓	✓
SBP (mmHg)	Boys	930	110.3	10.8	**	112.0	10.6	**	117.5	10.9	**	**	✓	✓	✓
	Girls	799	107.4	9.4		107.6	9.5		109.6	9.4		ns(0.051)			
DBP (mmHg)	Boys	930	58.1	6.5	ns	58.6	6.2	ns	62.4	7.0	**	**		✓	✓
	Girls	799	58.3	6.4		58.4	6.3		60.4	6.3		ns		✓	✓
Heart rate (bpm)	Boys	930	71.9	10.7	**	69.4	11.1	**	72.2	11.6	**	**	✓	✓	
	Girls	799	74.8	10.8		72.9	11.2		73.9	11.2		ns(0.060)			
BaPWV (cm/s)	Boys	930	867.4	96.8	ns	879.2	111.9	ns	944.5	117.7	**	**	✓	✓	✓
	Girls	799	864.2	99.5		876.2	101.3		923.0	101.3		**	✓	✓	✓
ABI	Boys	930	1.023	.085	**	1.024	.086	**	1.011	.083	ns	**		✓	✓
	Girls	799	.991	.080		1.004	.082		1.005	.077		*	✓	✓	
Sleeping time (min)	Boys	917	459.1	66.3	**	459.4	66.1	**	442.4	70.4	**	**		✓	✓
	Girls	792	442.5	67.0		451.2	61.8		432.2	66.6		**	✓	✓	✓
Time spent on vigorous exercise(min)	Boys	930	70.4	84.2	**	74.4	81.2	**	39.7	60.7	**	**		✓	✓
	Girls	799	50.9	72.8		52.0	72.9		19.0	39.9		**		✓	
Light exercise (min)	Boys	930	17.4	34.3	ns	19.8	37.2	*	19.9	35.3	*	ns			
	Girls	799	15.3	32.3		16.6	29.7		16.7	30.1		ns			
Time for indoor play (min)	Boys	930	57.8	74.4	ns	58.2	70.0	ns	58.3	69.7	ns	ns			
	Girls	799	60.1	73.9		64.5	78.2		58.2	73.6		ns			
Time for games (min)	Boys	930	58.9	74.4	**	68.8	75.7	**	61.7	77.2	**	*	✓		
	Girls	799	40.1	73.9		46.2	71.8		39.9	69.4		ns			
Time for watching TV (min)	Boys	930	100.5	85.1	**	102.6	81.2	*	92.2	81.7	*	**		✓	✓
	Girls	799	120.7	97.9		111.1	90.7		100.3	83.0		**	✓	✓	✓
Time for studying (min)	Boys	930	60.3	64.9	ns	53.2	54.5	**	77.0	69.1	**	**	✓	✓	✓
	Girls	799	64.5	57.5		61.4	57.2		92.0	74.2		**		✓	✓

A gender comparison of subject characteristics in each grade. t-tests were used to assess gender differences for each characteristic. Difference in means among each grade was tested by repeated measures ANOVA.
ns: not significant, * $p < 0.05$, ** $p < 0.01$

Table 2: Effect of obesity on baPWV(cm/s) in each gender.

		1st grade				2nd grade				3rd grade			
Gender	Obesity	N (%)	Mean	SD	Obese vs non-obese	N (%)	Mean	SD	Obese vs non-obese	N (%)	Mean	SD	Obese vs non-obese
Boys	Non-obese	855 (91.9)	864.7	94.6	**	842 (90.5)	873.6	108.9	**	822 (88.4)	941.7	117.5	*
	Obese	75 (8.1)	897.5	114.9		88 (9.5)	932.6	126		108 (11.6)	966	117.5	
Girls	Non-obese	752 (94.1)	861.3	98.6	**	749 (93.7)	872.8	97.4	**	753 (94.2)	920.7	99.6	*
	Obese	47 (5.9)	910.2	103.6		50 (6.3)	927.4	138.9		46 (5.8)	959.6	121.3	

A comparison of the effect of obesity on baPWV in each gender and grade. t-tests were used to assess for differences in baPWV between obese and non-obese students.

*p < 0.05, ** p < 0.01

Table 3: A comparison of various parameters in grouped third grade baPWV data.

		PWV			Significance			
		G1(low)	G2(normal)	G3(high)				
Boys	N	232	466	232	P	G1 vs G2	G1 vs G3	G2 vs G3
	BaPWV (cm/s)	806.3 ± 43.9	934.7 ± 45.0	1102.5 ± 72.4	**	✓	✓	✓
	Height (cm)	165.8 ± 6.2	166.5 ± 6.4	166.5 ± 6.2	ns			
	Weight (kg)	55.1 ± 12.9	56.4 ± 10.7	58.1 ± 11.2	*		✓	
	Waist circumference (cm)	70.7 ± 10.0	71.5 ± 8.5	72.5 ± 9.2	ns			
	Waist/height	.426 ± 0.055	.429 ± .048	.435 ± .052	ns			
	Percent of overweight(%)	1.2 ± 19.5	2.7 ± 5.8	5.8 ± 17.2	*		✓	
	Fat %	14.6 ± 15.2	15.2 ± 6.0	16.2 ± 6.4	*		✓	
	BMI	19.9 ± 4.0	20.2 ± 3.2	20.9 ± 3.5	**		✓	
	SBP (mmHg)	110.6 ± 8.8	117.2 ± 10.1	125.1 ± 9.4	**	✓	✓	✓
	DBP (mmHg)	57.9 ± 5.6	62.2 ± 6.3	67.3 ± 6.6	**	✓	✓	✓
	Heart rate (bpm)	68.6 ± 9.9	72.0 ± 11.4	76.3 ± 12.3	**	✓	✓	✓
	ABI	1.024 ± 0.09	1.009 ± .079	1.002 ± .088	*		✓	
	Sleeping time (min)	442.5 ± 66.6	441.9 ± 71.3	440.6 ± 72.1	ns			
	Vigorous exercise (min)	42.8 ± 64.0	39.6 ± 58.3	36.8 ± 62.3	ns			
	Light exercise (min)	20.0 ± 35.7	21.5 ± 38.5	16.6 ± 27.0	ns			
	Time for indoor play (min)	52.6 ± 61.0	59.3 ± 72.5	62.0 ± 71.9	ns			
	Time for games (min)	63.7 ± 80.7	62.1 ± 76.5	58.8 ± 75.2	ns			
	Time for watching TV (min)	82.2 ± 71.6	93.4 ± 84.3	99.7 ± 85.1	ns(0.064)			
	Time for studying (min)	79.6 ± 69.2	76.3 ± 66.7	76.0 ± 73.8	ns			
Girls	N	199	400	200	P	G1 vs G2	G1 vs G3	G2 vs G3
	BaPWV (cm/s)	802.1 ± 40.1	916.2 ± 39.2	1056.7 ± 61.5	**	✓	✓	✓
	Height (cm)	156.5 ± 5.4	156.9 ± 5.2	156.9 ± 5.6	ns			
	Weight (kg)	48.4 ± 7.0	49.3 ± 6.9	49.7 ± 8.6	ns			
	Waist circumference (cm)	67.8 ± 6.7	68.6 ± 6.4	69.4 ± 7.6	ns			
	Waist/height	.434 ± .420	.438 ± .040	.443 ± .046	ns			
	Percent of overweight(%)	-2.6 ± 12.1	-1.2 ± 12.5	-.3 ± 14.9	ns			
	Fat %	21.4 ± 4.4	22.2 ± 5.0	22.7 ± 4.8	*		✓	
	BMI	19.7 ± 2.4	20.0 ± 2.5	20.2 ± 3.0	ns			
	SBP (mmHg)	104.0 ± 7.3	108.8 ± 8.1	116.7 ± 9.5	**	✓	✓	✓
	DBP (mmHg)	56.5 ± 4.7	60.0 ± 5.6	65.2 ± 6.3	**	✓	✓	✓
	Heart rate (bpm)	70.5 ± 9.3	72.8 ± 10.3	79.4 ± 12.7	**		✓	✓
	ABI	1.009 ± .081	1.007 ± .074	.995 ± .077	ns			
	Sleeping time (min)	420.0 ± 67.7	434.3 ± 66.9	438.7 ± 64.2	*	✓	✓	
	Vigorous exercise (min)	18.4 ± 40.9	18.5 ± 39.6	20.6 ± 39.8	ns			
	Light exercise (min)	17.9 ± 26.9	16.8 ± 33.2	15.2 ± 26.3	ns			
	Time for indoor play (min)	57.2 ± 77.3	56.9 ± 74.0	61.9 ± 68.9	ns			
	Time for games (min)	41.5 ± 74.6	39.3 ± 69.4	39.3 ± 64.3	ns			
	Time for watching TV (min)	96.7 ± 88.2	101.9 ± 85.8	101.0 ± 71.5	ns			
	Time for studying (min)	94.7 ± 77.9	91.5 ± 75.8	90.3 ± 67.3	ns			

Third grade baPWV were grouped by percentile into: G1 (low), G2 (normal) and G3 (high). Values are expressed as mean±SD. This table compares grouped baPWV with the various parameters measured in each gender. Differences between groups were examined using one way ANOVA and the Bonferroni post-hoc test.

ns: not significant, * $p < 0.05$, ** $p < 0.01$

Table 4: A comparison of various parameters with the pattern of change in baPWV.

			Pattern of PWV change				Significance						
			A	B	C	D							
		N	591	105	107	127	P	A vs B	A vs C	A vs D	B vs C	B vs D	C vs D
Boys	1st grade	BaPWV (cm/s)	819.4 ±65.1	851.4 ±53.5	987.9 ±51.3	1002 ±53.9	**	✓	✓	✓	✓	✓	
		SBP (mmHg)	107.4 ± 9.5	114.1 ± 10.2	113.7 ± 12.4	117.8 ± 10.0	**	✓	✓	✓		✓	✓
		DBP (mmHg)	56.2 ± 5.4	59.3 ± 5.5	61.5 ± 7.7	63.2 ± 6.4	**	✓	✓	✓	✓	✓	
		Heart rate (bpm)	70.4 ± 10.0	72.7 ± 10.6	75.8 ± 11.7	75.2 ± 11.4	**		✓	✓			
		Sleeping time (min)	459.0 ± 66.0	456.9 ± 70.1	459.6 ± 66.9	457.0 ± 64.7	ns						
		Vigorous exercise (min)	71.9 ±81.0	71.3 ±91.6	57.6 ±73.5	73.5 ±94.5	ns						
		Light exercise (min)	17.4 ± 32.8	12.1 ± 19.3	21.4 ± 45.0	18.4 ± 43.0	ns						
		Time for games (min)	59.5 ± 78.6	58.6 ± 75.7	53.0 ± 72.0	61.3 ± 75.0	ns						
		Time for watching TV (min)	97.7 ± 85.4	104.5 ± 79.7	88.4 ± 89.4	120.1 ± 82.1	*			✓			✓
	2nd grade	BaPWV (cm/s)	837.7 ±85.7	903.2 ±90.8	920.6 ±94.4	1017.5 ±119.6	**	✓	✓	✓		✓	✓
		SBP (mmHg)	109.7 ± 9.4	114.8 ± 9.1	112.8 ± 12.7	120.1 ± 10.8	**	✓	✓	✓		✓	✓
		DBP (mmHg)	57.3 ± 5.6	59.4 ± 6.4	59.4 ± 6.6	63.3 ± 6.2	**	✓	✓	✓		✓	✓
		Heart rate (bpm)	68.2 ± 10.2	69.8 ± 11.0	71.4 ± 11.3	73.4 ± 13.0	**		✓	✓			
		Sleeping time (min)	459.2 ± 63.7	453.8 ± 70.7	460.1 ± 73.7	458.9 ± 71.1	ns						
		Vigorous exercise (min)	75.9 ±81.4	65.7 ±77.8	67.1 ±74.7	80.7 ±88.0	ns						
		Light exercise (min)	22.0 ± 41.7	16.0 ± 29.1	13.3 ± 20.1	18.1 ± 30.6	ns						
		Time for games (min)	69.5 ± 77.4	65.0 ± 68.4	71.7 ± 72.6	66.7 ± 76.6	ns						
		Time for watching TV (min)	94.4 ± 78.3	111.7 ± 76.3	117.0 ± 87.2	121.1 ± 87.8	*		✓	✓			
		N	591	105	107	127	P	A vs B	A vs C	A vs D	B vs C	B vs D	C vs D
Girls	1st grade	BaPWV (cm/s)	814.2 63.4	854.6 48.6	973.6 ±51.8	1013.9 ±78.4	**	✓	✓	✓	✓	✓	✓
		SBP (mmHg)	104.5 ± 7.7	108.1 ± 9.9	112.4 ± 8.8	116.2 ± 8.9	**	✓	✓	✓	✓	✓	✓
		DBP (mmHg)	56.1 ± 5.3	59.3 ± 5.7	61.3 ± 5.8	65.2 ± 6.0	**	✓	✓	✓		✓	✓
		Heart rate (bpm)	73.4 ± 9.9	74.3 ± 10.9	76.2 ± 12.1	80.9 ± 11.5	**			✓		✓	✓
		Sleeping time (min)	444.1 ± 65.0	440.5 ± 64.9	445.7 ± 63.1	438.2 ± 67.9	ns						
		Vigorous exercise (min)	50.5 ±71.7	56.6 ±79.8	53.6 ±79.8	44.9 ±68.7	ns						
		Light exercise (min)	14.9 ± 32.2	11.9 ± 17.5	17.1 ± 24.7	18.5 ± 46.4	ns						
		Time for games (min)	41.2 ± 69.6	32.0 ± 62.5	47.3 ± 66.9	35.4 ± 64.2	ns						
		Time for watching TV (min)	117.2 ± 97.5	118.8 ± 95.1	117.5 ± 84.9	141.7 ± 110.9	ns						
	2nd grade	BaPWV (cm/s)	841.6 ±85.5	910.1 ±95.8	907.5 ±70.8	983.4 ±105.8	**	✓	✓	✓		✓	✓
		SBP (mmHg)	105.7 ± 8.7	109.9 ± 8.9	108.3 ± 9.0	113.8 ± 11.0	**	✓		✓		✓	✓
		DBP (mmHg)	57.2 ± 5.9	59.3 ± 6.2	58.8 ± 6.1	62.7 ± 6.9	**	✓		✓		✓	✓
		Heart rate (bpm)	71.5 ± 10.3	72.6 ± 12.1	73.6 ± 10.7	79.0 ± 11.6	**			✓		✓	✓
		Sleeping time (min)	446.0 ± 81.1	451.9 ± 59.4	460.7 ± 59.4	456.2 ± 69.2	ns						
		Vigorous exercise (min)	53.9 ±72.9	66.1 ±51.5	70.5 ±30.4	30.4 ±52.1	ns						
		Light exercise (min)	16.2 ± 30.7	17.0 ± 25.2	18.4 ± 24.9	16.4 ± 32.8	ns						
		Time for games (min)	46.0 ± 71.3	43.8 ± 74.5	54.5 ± 76.7	41.6 ± 67.3	ns						
		Time for watching TV (min)	106.8 ± 92.7	115.0 ± 82.8	116.2 ± 87.1	123.5 ± 90.7	ns						

BaPWV data were grouped into four groups: Group A (normal to normal), Group B (normal to high), Group C (high to normal), Group D (high to high). Normal and high baPWV were defined as having a baPWV below and above the 75th percentile, respectively. This table compares grouped baPWV data with the various parameters measured. Values are expressed as mean±SD. Differences between groups were examined using one way ANOVA and the Bonferroni post-hoc test.

ns: not significant, * $p < 0.05$, ** $p < 0.01$

Table 5: Logistic regression analysis comparing baPWV increment from the first grade to the third grade with the various parameters measured.

	Variables	OR	95% confidence interval of OR		Remark
			Lower limit	Upper limit	
Boys	BaPWV 1st	3.24	2.48	4.24	†
	Weight 2nd	1.85	1.27	2.68	
	Body fat % 2nd	1.76	1.33	2.34	†
	BMI 2nd	1.68	1.14	2.48	
	Time for watching TV 1st	1.54	1.18	2.02	
	Orthostatic dysfunction 1st	1.52	1.13	2.03	
	General fatigue 1st	1.50	1.14	1.98	
Girls	BaPWV 1st	3.51	2.65	4.63	†
	Time for studying 1st	1.45	1.09	1.92	
	Concentration 2nd	1.42	1.04	1.95	
	Having motivation 2nd	1.41	1.06	1.87	†
	Weight 2nd	1.39	1.05	1.83	
	Pale face 1st	1.35	1.00	1.83	†
	Time spent on vigorous exercise 1	1.33	1.00	1.76	

This table shows the odds ratio and 95% confidence intervals for the various parameters measured and the likelihood of baPWV increment in boys and girls. Only significant variables are listed. For odds ratios < 1, the inverse odds ratio was used and marked with † in the remarks column.

Table 6: Logistic regression analysis comparing third grade baPWV and the various parameters measu

	Variables	OR	95% confidence interval of OR		Remark
			Lower limit	Upper limit	
Boys	BaPWV 2nd	3.90	2.82	5.41	
	BaPWV 1st	2.64	1.91	3.65	
	Feel sick at bathing 2nd	1.80	1.19	2.71	
	Time for games 2nd	1.78	1.31	2.40	†
	Time spent on light exercise 1st	1.62	1.20	2.18	†
	SBP 1st	1.62	1.16	2.26	
	SBP 2nd	1.60	1.15	2.23	
	Sleeping time 1st	1.58	1.18	2.12	†
	Headache 1st	1.56	1.16	2.10	
	Time for watching TV 1st	1.51	1.10	2.08	
	Dizziness 2nd	1.46	1.05	2.02	†
	General fatigue 2nd	1.44	1.03	2.00	
	Time for indoor play 2nd	1.40	1.03	1.91	†
	Anxiety for action 2nd	1.40	1.03	1.89	
	Good friendship 1st	1.39	1.03	1.89	†
Girls	BaPWV 2nd	3.98	2.78	5.71	
	BaPWV 1st	3.71	2.61	5.27	
	SBP 2nd	1.96	1.39	2.77	
	Having breakfast 1st	1.89	1.25	2.87	
	Lack of sleep 2nd	1.84	1.33	2.55	
	Emotional ups and downs 1st	1.71	1.21	2.42	
	ABI 1st	1.66	1.21	2.29	
	Concentration 2nd	1.60	1.11	2.32	
	Having motivation 2nd	1.54	1.10	2.15	†
	Feel sick during morning 1st	1.52	1.09	2.11	
	Anxiety for action 1st	1.44	1.02	2.02	
	Time for games 1st	1.39	1.01	1.93	†

This table shows the odds ratio and 95% confidence intervals for the various parameters measured and baPWV in the third grade. Only significant variables are listed. For odds ratios < 1, the inverse odds ratio was used and marked with † in the remarks column.