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A cross-sectional study on upright heart rate and BP changing characteristics: basic data for establishing diagnosis of postural orthostatic tachycardia syndrome and orthostatic hypertension

Juan Zhao,1 Zhenhui Han,2 Xi Zhang,2 Shuxu Du,3 Angie Dong Liu,4 Lukas Holmberg,4 Xueying Li,5 Jing Lin,1 Zhenyu Xiong,2 Yong Gai,2 Jinyan Yang,1 Ping Liu,1 Chaoshu Tang,6 Junbao Du,1,7 Hongfang Jin1

ABSTRACT

Objective: We aimed to determine upright heart rate and blood pressure (BP) changes to suggest diagnostic criteria for postural orthostatic tachycardia syndrome (POTS) and orthostatic hypertension (OHT) in Chinese children.

Methods: In this cross-sectional study, 1449 children and adolescents aged 6–18 years were randomly recruited from two cities in China, Kaifeng in Henan province and Anguo in Hebei province. They were divided into two groups: 844 children aged 6–12 years (group I) and 605 adolescents aged 13–18 years (group II). Heart rate and BP were recorded during an active standing test.

Results: 95th percentile ($P_{95}$) of $\delta$ heart rate from supine to upright was 38 bpm, with a maximum upright heart rate of 130 and 124 bpm in group I and group II, respectively. $P_{95}$ of $\delta$ systolic blood pressure (SBP) increase was 18 mm Hg and $P_{95}$ of upright SBP was 132 mm Hg in group I and 138 mm Hg in group II. $P_{95}$ of $\delta$ diastolic blood pressure (DBP) increase was 24 mm Hg in group I and 21 mm Hg in group II, and $P_{95}$ of upright DBP was 89 mm Hg in group I and 91 mm Hg in group II.

Conclusions: POTS is suggested when $\delta$ heart rate is $\geq 38$ bpm (for easy memory, $\geq 40$ bpm) from supine to upright, or maximum heart rate $\geq 130$ bpm (children aged 6–12 years) and $\geq 125$ bpm (adolescents aged 13–18 years), associated with orthostatic symptoms. OHT is suggested when $\delta$ SBP (increase) is $\geq 20$ mm Hg, and/or $\delta$ DBP (increase) $\geq 25$ mm Hg (in children aged 6–12 years) or $\geq 20$ mm Hg (in adolescents aged 13–18 years) from supine to upright; or upright BP $\geq 130/90$ mm Hg (in children aged 6–12 years) or $\geq 140/90$ mm Hg (in adolescents aged 13–18 years).

INTRODUCTION

Orthostatic intolerance (OI) is a group of diseases induced by standing and relieved by recumbence, and includes symptoms such as dizziness, headache, palpitations, nausea, abdominal pain, concentration difficulties, hyperventilation, presyncope and even syncope.1 It is one of the most common causes for syncope in children and adolescents, seriously affecting their daily life and school studies,2 and places heavy mental and economic burdens on families.3–7 Postural orthostatic tachycardia syndrome (POTS), vasovagal syncope and orthostatic hypertension (OHT) make up the main components of OI.

The active standing test is one of the most important diagnostic methods for OI. During the test, clinical symptoms, heart rate and blood pressure (BP) changes can be

Strengths and limitations of this study

- Orthostatic intolerance is one of the most common causes for syncope in children and adolescents, seriously affecting their daily life and school studies.
- The distribution characteristics of upright heart rate and blood pressure in children and adolescents evidenced by large-sampled data was lacking.
- This study suggested distribution characteristics of heart rate and blood pressure changes during an active standing test in children and adolescents.
- The upright heart rate and blood pressure changes were useful to suggest diagnostic criteria for postural orthostatic tachycardia syndrome or orthostatic hypertension in children and adolescents.
- This study was conducted in the north of China. To ascertain whether the results are applicable to children and adolescents in other areas will need multicentre studies.
recorded from supine to upright position. However, the active standing test-based diagnostic criteria for children and adolescents with POTS or OHT have mainly been sourced from the data of adults due to the absence of large-sampled epidemiological data from children. In 1993, the POTS diagnostic criteria were reported as an increased heart rate of ≥30 bpm or a maximum heart rate of ≥120 bpm without BP decline within 10 min after the individual stands upright from a supine position. These diagnostic criteria were previously used in most studies on children and adolescents in this area. The OHT diagnostic criterion was proposed by Fessel and Robertson in 2006 as an increased systolic blood pressure (SBP) ≥20 mm Hg without requirements for diastolic blood pressure (DBP) after standing compared with BP at supine position. However, up to now, the OHT diagnostic criterion has not been used for diagnosing children.

Children and adolescents are at a special growth period. Their cardiovascular development is immature and presents different features compared with adults, such as a smaller stroke volume, thinner vessel walls, larger vessel diameter and incomplete nervous system regulation. Their heart rates are faster and their BPs are lower than in adults. In particular, the exact changes of heart rate and BP from supine to upright are larger than in adults and the normal range of heart rate and BP varies with ages. Therefore, the diagnostic criteria for adults are sometimes not suitable for children and adolescents. In a consensus statement, Freeman et al. indicated that increased heart rate should be raised by at least 40 bpm in POTS diagnosis of adolescents aged between 12 and 19 years. Singer et al. reported that the 95th percentile (P95) of heart rate change was 42.9 bpm in normal children and adolescents aged between 8 and 19 years, using head-up tilt test (HUTT). However, large sample-sized evidence for the criteria of POTS in children is lacking and OI diagnostic criteria for Chinese children have not been established. Therefore, the present study was designed to determine the normal-values of heart rate and BP changes during the standing test in children and adolescents in China, with a specific focus on the age-related and gender-related differences.

METHODS

Participants
In this cross-sectional study, we randomly selected five schools in Kaifeng, in Henan province, and Anguo, in Hebei province in China, from October 2012 to January 2013. We randomly recruited students from each grade of the schools (198, 113, 118, 158, 127, 130, 154, 97, 67, 107, 106 and 74 students from grades 1–6 of elementary schools and grades 1–6 of high schools, respectively). Totally, 1449 children and adolescents aged 6–18 years were sampled for this study. All students took the active standing test. They were divided into two groups according to age: 844 children aged between 6 and 12 years in group I (434 boys and 410 girls) and 605 adolescents aged between 13 and 18 years in group II (319 boys and 286 girls). Their medical history was taken and physical examination was carried out for the study participants. Those participants with heart and pulmonary vascular diseases or with a history of taking drugs in the prior 3 months were excluded through history of illness, BP measurement, physical examination and ECG.

Basic anthropometric indexes
Anthropometric study was performed using the same medical scales (Q241, Zhejiang Anbo Medical Devices Co, Ltd) according to a standard measurement method. The instrument was calibrated prior to application and its quality was controlled regularly. The weight and height were measured and the body mass index (BMI) was calculated as kg/m².

Protocol for the active standing test
The children were tested with an active standing test. A quiet environment with a room temperature of 22°C and dim light was required during the active standing test. The children layd quietly at supine position for at least 10 min before the basal BP and heart rate were recorded. Then, they were asked to stand upright on their own for 10 min, and their heart rate and BP were monitored during the whole test using a Dash 2000 Multi-Channel Physiologic Monitor (General Electric, Schenectady, New York, USA), which automatically and dynamically displayed heart rate using three-lead ECG and intermittent oscillometric BP. We used the most stable heart rate in a 60 s interval as the participant’s heart rate during this period. The heart rate was sampled every minute during the 10 min period. The sampling time of the heart rate was 1 min. The maximum upright heart rate was defined as the maximum heart rate during the 10 min of active standing. In the active standing test, we derived four BPs for each participant, one supine BP and three standing BPs at 3, 6 and 9 min, respectively, after standing. The children recovered recumbence immediately in those cases where dizziness, headache, nausea, paleness or other discomfort occurred while their heart rate and BP were being observed. Appropriate cuffs were used throughout the research.

Statistical methods
Measurement data were analysed using SPSS V.13.0 (SPSS, Chicago, Illinois, USA) for statistical analysis and expressed in mean±SD. After homogeneity test of variance in both groups, independent sample student t test was performed for analysing the differences between the two age groups. Repeated measure analysis was used to analysing the differences for heart rate and BP in different positions and times. χ² Test was performed for sex ratio comparison and p<0.05 was deemed as statistically significant. For data in skewed or unknown distribution, a percentile approach built on a confidence level was

used for determination of normal value range. We selected the P95 as a normal threshold for the change of heart rate and BP from supine to standing.18–20

RESULTS

Clinical features of participants

The study comprised of 1449 children and adolescents aged between 6 and 18 years. The mean height, weight and BMI in the children aged 6–12 years was higher than in the adolescents aged between 13 and 18 years (p<0.05, table 1). No participant with missing data was found in either index.

It was found that heart rate after standing was significantly greater than that at supine position in both groups (figure 1, p<0.001). BP after standing was also higher than BP at supine position in both groups (figure 2, p<0.001).

Features of haemodynamic changes

In all of the participants, the supine heart rate was 84 ±12 bpm, maximum upright heart rate 107±14 bpm and δ heart rate 22±9 bpm. The supine heart rate and maximum upright heart rate significantly differed between the two groups (p<0.001) but the δ heart rate did not (p=0.632) (figure 1 and table 2).

The supine SBP was 110±10 mm Hg and upright SBP, 118±10 mm Hg, and δ SBP increase was 8±7 mm Hg in all of the participants. The supine DBP was 65±9 mm Hg and upright DBP, 76±9 mm Hg, and δ DBP increase was 11±7 mm Hg in all the children and adolescents. There were significant differences in supine, upright and δ DBP between both groups (p<0.001) (figure 1 and table 2).

The appearance of OI symptoms in the active standing test

During the active standing test, there were 269 (18.6%) children and adolescents reporting about OI symptoms, and they recovered recumbence immediately (light headedness 130 (9.0%), headache 130 (9.0%), fatigue 125 (8.5%), long breath 74 (5.1%), vertigo 69 (4.8%), blurred vision 64 (4.4%), palpitations 56 (3.9%) and other discomfort 68 (4.7%)). No one fainted during the test. From the analysis, we found

Table 1 Baseline characteristics of children

<table>
<thead>
<tr>
<th>Group</th>
<th>n</th>
<th>Age, years</th>
<th>Male/female</th>
<th>Height, cm</th>
<th>Weight, kg</th>
<th>Body mass index, kg/m²</th>
</tr>
</thead>
<tbody>
<tr>
<td>6–12 years</td>
<td>844</td>
<td>±2</td>
<td>434/410</td>
<td>141±12</td>
<td>36±11</td>
<td>17.65±3.21</td>
</tr>
<tr>
<td>13–18 years</td>
<td>605</td>
<td>±2*</td>
<td>319/286</td>
<td>163±8*</td>
<td>53±10*</td>
<td>20.11±3.13*</td>
</tr>
</tbody>
</table>

Values are mean±SD.
*Denotes p value <0.01.
that the children with symptoms during the standing test were older than those without symptoms (12±2 vs 10±2 years, t=−9.667, p<0.001). Also, children with symptoms during the standing test had more frequent previous OI symptoms (103/269 vs 277/1180, χ²=24.852, p<0.001), higher supine heart rate (88±11 vs 86±10 bpm, t=2.258, p=0.024), faster heart rate maximum (111±11 vs 109±11 bpm, t=2.274, p=0.023) and greater BMI (18.973±3.662 vs 17.876±3.221 kg/m², t=−4.571, p<0.001), than those without symptoms during the standing test.

In our study participants, 287 children had an OI history. Forty-two children among them had a syncope history. The children who had an OI history were older than those without a history (12±2 vs 11±2 years, t=−3.824, p<0.001). The differences in supine heart rate (85±12 vs 88±11 bpm, t=−1.199, p=0.231), standing heart rate maximum (108±12 vs 110±11 bpm, t=1.059, p=0.290), supine BP (SBP 110±9 vs 109±10 mm Hg, t=−0.491, p=0.624), supine DBP (65±11 vs 64±9 mm Hg, t=−1.048, p=0.295) and upright BP (SBP 120±15 vs 117±10 mm Hg, t=−1.141, p=0.260), supine DBP (77±17 vs 76±9 mm Hg, t=−0.658, p=0.514) between the two groups were not significant.

Percentile values of maximum heart rate, δ heart rate, BP and δ BP increase in the active standing test

As shown in table 3, P95 of maximum upright heart rate was 130 bpm in group I and 124 bpm in group II. Since δ heart rate did not differ between the two groups, we combined the data of the two groups in analysing δ heart rate from supine to upright position and showed that P95 of δ heart rate was 38 bpm.

P95 of upright SBP was 132 mm Hg in group I and 138 mm Hg in group II. Since δ SBP increase from supine to upright position did not differ between the two groups, we combined the data of the two groups in analysing δ SBP increase P95 and showed that δ SBP increase P95 was 18 mm Hg. P95 of upright DBP was 89 mm Hg in group I and 91 mm Hg in group II, and P95 of δ DBP increase from supine to upright position

Figure 2  The distribution chart of time required for reaching the maximum heart rate after standing of all participants.

Table 2  The haemodynamic changes in children and adolescents

<table>
<thead>
<tr>
<th>Group</th>
<th>Supine heart rate, bpm</th>
<th>Heart rate max*, bpm</th>
<th>δ Heart rate†, bpm</th>
<th>Supine SBP, mm Hg</th>
<th>Upright SBP, mm Hg</th>
<th>δ SBP‡, mm Hg</th>
<th>Supine DBP, mm Hg</th>
<th>Upright DBP, mm Hg</th>
<th>δ DBP§, mm Hg</th>
</tr>
</thead>
<tbody>
<tr>
<td>6-12 years</td>
<td>88±11</td>
<td>10±12</td>
<td>22±8</td>
<td>116±10</td>
<td>116±10</td>
<td>6±8</td>
<td>63±9</td>
<td>75±9</td>
<td>12±8</td>
</tr>
<tr>
<td>Boys</td>
<td>87±11</td>
<td>10±12</td>
<td>22±8</td>
<td>115±10</td>
<td>115±10</td>
<td>6±8</td>
<td>63±9</td>
<td>75±9</td>
<td>12±8</td>
</tr>
<tr>
<td>Girls</td>
<td>89±11</td>
<td>10±12</td>
<td>22±8</td>
<td>116±10</td>
<td>116±10</td>
<td>6±8</td>
<td>63±9</td>
<td>75±9</td>
<td>12±8</td>
</tr>
<tr>
<td>P Value</td>
<td>0.147</td>
<td>0.074</td>
<td>0.005</td>
<td>&lt;0.001</td>
<td>0.051</td>
<td>0.012</td>
<td>0.884</td>
<td>0.047</td>
<td>0.004</td>
</tr>
</tbody>
</table>

Values are mean±SD.
*Heart rate max is the maximum heart rate during 10 min of standing.
†δ Heart rate is the increased heart rate during the active standing test.
‡δ SBP is the increased SBP during the active standing test.
§δ DBP is the increased DBP during the active standing test.

DBP, diastolic blood pressure; SBP, systolic blood pressure.
was 24 mm Hg in group I and 21 mm Hg in group II (table 3).

**Table 3** The percentage of heart rate and blood pressure in the active standing test

<table>
<thead>
<tr>
<th>Indices</th>
<th>Percentile</th>
<th>5th</th>
<th>10th</th>
<th>50th</th>
<th>90th</th>
<th>95th</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heart rate max*, bpm</td>
<td></td>
<td>92</td>
<td>96</td>
<td>110</td>
<td>125</td>
<td>130</td>
</tr>
<tr>
<td>6–12 years</td>
<td></td>
<td>76</td>
<td>85</td>
<td>101</td>
<td>118</td>
<td>124</td>
</tr>
<tr>
<td>13–18 years</td>
<td></td>
<td>8</td>
<td>12</td>
<td>23</td>
<td>33</td>
<td>38</td>
</tr>
<tr>
<td>δ Heart rate†, bpm</td>
<td></td>
<td>100</td>
<td>103</td>
<td>116</td>
<td>128</td>
<td>132</td>
</tr>
<tr>
<td>Upright SBP, mm Hg</td>
<td></td>
<td>105</td>
<td>108</td>
<td>119</td>
<td>133</td>
<td>138</td>
</tr>
<tr>
<td>6–12 years</td>
<td></td>
<td>–4</td>
<td>–1</td>
<td>8</td>
<td>15</td>
<td>18</td>
</tr>
<tr>
<td>13–18 years</td>
<td></td>
<td>–1</td>
<td>2</td>
<td>12</td>
<td>21</td>
<td>24</td>
</tr>
<tr>
<td>Upright DBP, mm Hg</td>
<td></td>
<td>60</td>
<td>64</td>
<td>75</td>
<td>86</td>
<td>89</td>
</tr>
<tr>
<td>6–12 years</td>
<td></td>
<td>65</td>
<td>68</td>
<td>78</td>
<td>87</td>
<td>91</td>
</tr>
<tr>
<td>13–18 years</td>
<td></td>
<td>–1</td>
<td>2</td>
<td>10</td>
<td>18</td>
<td>21</td>
</tr>
</tbody>
</table>

*Heart rate max is the maximum heart rate during the 10 min of standing.
†δ Heart rate is the increased heart rate during the active standing test.
‡Upright SBP is the increased SBP during the active standing test.
§Upright DBP is the increased DBP during the active standing test.
DBP, diastolic blood pressure; SBP, systolic blood pressure.

Time to reach maximum heart rate in the active standing test

The median time to reach maximum heart rate was 5 (1–10) min, where maximum heart rate occurred in 255 children within 1 min after standing, accounting for 17.6% of the participants (figure 2).

**DISCUSSION**

Currently, great attention is being paid to OI in children and adolescents. Although some scholars believe that the HUTT is the golden standard examination to evaluate an individual’s response to postural changes, the active standing test has its own advantages and is preferred in many clinical settings since it is easy, practicable and reliable. Comparative studies on the active standing test and HUTT have discovered that both methods have similar sensitivity, and that the active standing test has even higher specificity than HUTT (79% vs 23%) when increased heart rate is ≥30 bpm, as a POTS diagnostic criterion. Thus, in the present study, we investigated the normal values of heart rate and BP changes during the active standing test and explored the POTS and OHT diagnostic criteria for children and adolescents in China.

Since adult POTS criteria were used by Schondrof and Low in 1993, the increased value of heart rate ≥30 bpm or maximum heart rate ≥120 bpm within 10 min from supine to standing or from supine to head-up tilt has been applied in many studies for children and adolescents. However, children and adolescents are in a special growth and development period. Their cardiovascular development is immature and they need an increased heart rate to ensure enough haemoperfusion. In addition, heart rate is mainly controlled by the nervous system. The excitement of the sympathetic nervous system allows rapid heart beat and excitement of the parasympathetic nervous system allows slow heart beat. Normally, the sympathetic and parasympathetic nervous systems are balanced, but in children and adolescents, they are not yet fully developed, and the dominant sympathetic nervous system leads to an increased heart rate. This physiologically fast heart beat, however, will gradually be reduced to the level of a normal adult. Heart rate and BP are related to age, sex, height, weight and race. Long-term observations have shown that BP varies with age, and that before puberty, BP in boys and girls increases rapidly over time.

Recent studies have shown that the diagnostic specificity was reduced when adult POTS and OI diagnostic criteria were applied on children and adolescents. Some studies showed that the change in heart rate from supine to upright position in children and adolescents was up to 40–50 bpm. For instance, Skinner et al. showed that 307 adolescents aged between 15 and 17 years had a heart rate change from –1 to +48 bpm at 5 min standing. Furthermore, Yamaguchi et al. found that the development maturity influenced heart rate change from supine to upright position. 173 school children aged between 6 and 18 years had greater heart rate change than non-pubertal children from supine to upright position (34 (9) bpm vs 29 (8) bpm, p<0.001). Therefore, an increase in heart rate ≥35 bpm as diagnostic criterion for children and adolescents with POTS is recommended. Recently, Freeman et al. and Singer et al. recommended 40 bpm as a POTS diagnostic criterion in children and adolescents by taking into account the variability of heart rate. Singer et al. studied 106 children and adolescents aged between 8 and 19 years with HUTT for 5 min and showed that maximum heart rate ≥130 bpm in children aged ≤13 years, and ≥120 bpm in adolescents aged ≥14 years, were the diagnostic criteria of maximum heart rate in children and adolescents with POTS. Up to now, large sample-sized studies of heart rate and BP change during the standing test are still needed.

We found that 18.6% of children and adolescents reported about OI symptoms during the standing test and they recovered recumbence immediately. Also, children with symptoms during the standing test were older than those without symptoms, which was in accordance with the fact that OI was common in older children. We noticed that children with symptoms during the standing test had faster supine heart rates than those without symptoms during the standing test, suggesting that the underlying mechanisms for OI symptoms likely involve the increased sympathetic nervous system excitability.
Our study results showed that the maximum change of heart rate from supine to upright was 52 bpm, and the change in the two groups did not differ (p=0.632). However, we found that the change of heart rate was not affected by age, sex, height, weight and BMI, in children and adolescents. This suggested that the same diagnostic criterion could be applied on children and adolescents with a large span of age variation. Our results showed that POTS should be suggested in children and adolescents when a heart rate change from supine to upright position ≥38 bpm (for easy memory, ≥40 bpm); and/or maximum heart rate ≥130 bpm at upright position within 10 min in group I and ≥125 bpm at upright position in group II, when there is a history of clinical orthostatic symptoms.

Since adult OHT was proposed by Fessel and Robertson in 2006, it has been mostly applied in analysing recurrent cardiovascular risk in elderly patients with hypertension. Pathophysiologically speaking, OHT reflects sympathetic activity and microvascular and cardiac remodelling, and it has become a new risk factor for cardiovascular disease. Thomas et al. showed that risks of hypertension were increased substantially within eight follow-up years in young adults with increased BP ≥5 mm Hg after standing. However, there has been no definitive diagnostic criterion for OHT.

In our study, the BP was reduced only in a small number of cases, but was stable or increased in most children and adolescents after standing. The SBP change from supine to upright position did not differ significantly between the groups, but the DBP change did. Based on our present results, with BP at supine position as baseline BP and BP at 3 min after standing as orthostatic BP, and according to the percentile analysis of the BP data, OHT needs to be suggested when (1) δSBP increase from supine to upright position within 3 min ≥20 mm Hg, or δDBP increases ≥25 mm Hg in children aged 6–12 years and ≥20 mm Hg in adolescents aged 13–18 years; or (2) an increase in BP from supine to upright position ≥130/90 mm Hg (for children aged 6–12 years) or ≥140/90 mm Hg (for adolescents aged 13–18 years) within 3 min. For convenience in clinical use, we suggested 130 (P91.5)/90 (P95.2) and 140 (P96.6)/90 (P93.8) mm Hg instead of 132/89 and 138/91 mm Hg as the normal threshold for disease was used. With those references, we also used the P95 in our present study. More importantly, the OHT diagnosis should depend on the comprehensive situation of the patients including the upright change of BP, the upright absolute BP and clinical OI symptoms, as previously indicated. Further investigations need to be conducted to confirm previous findings and discover new insights of the situation as a BP measurement result.

In addition, recent studies recommend that observation time limit of HUTT should be shorter than 5 min after postural change. The reason is that changes in heart rate and BP have explicit performance within 5 min after standing, but lack significance between 6 and 10 min after standing. Nevertheless, in some studies, at least 2, 5 and 10 min after standing were included in the HUTT because some patients showed a delayed tachycardia. The results of our study found that the median time of reaching maximum heart rate was 5 (1–10) min in the active standing test in children and adolescents. Therefore, we recommend the orthostatic heart rate and BP to be measured within 10 min in children.

Limitations
Our present study still has some limitations. For example, though the study sample was large, the study was from the north of China. Whether the results are suitable to children and adolescents in other areas still needs validation through multicentre studies. As for diagnostic criterion, we still need a derivation cohort followed by a validation cohort to confirm these findings. The criteria of upright heart rate and BP came from the active standing test, and whether they fit HUTT or not also needs further research. Up to now, the significance of OHT in childhood is still incompletely understood.

CONCLUSIONS
Children have significant distribution characteristics of heart rate and BP during standing tests, which would be useful for the consideration of POTS when δ heart rate ≥38 bpm (for easy memory, ≥40 bpm), or maximum heart rate ≥130 bpm for children aged 6–12 years and ≥125 bpm for adolescents aged 13–18 years within 10 min from moving from supine to upright position, together with clinical orthostatic symptoms. OHT should be defined as a SBP increase ≥20 mm Hg, and/or DBP increase ≥25 mm Hg (children aged 6–12 years) or ≥20 mm Hg (adolescents aged 13–18 years), within 3 min from supine to upright position; or BP ≥130/90 mm Hg (children aged 6–12 years) or ≥140/90 mm Hg (children aged 6–12 years) after standing.

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Contributors
HJ, JD, CT, ZH and JZ designed the research. JZ, ZH, JY, JL, PL, XZ, ZX, YG, SD, XL and JZ participated in the acquisition of the data. ZH, JL, JD, ADL, LH and JZ analysed the data. JZ, ZH, JL, HJ and JD interpreted the results of the experiments. JZ, ZH, JD, CT, ADL, JY, JL, PL, XZ, ZX, YG, SD, XL and JZ contributed equally to this study.
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