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Adult derived genetic blood pressure scores and blood pressure measured in different body postures in young children

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Abstract

Aims: Several genes are related to blood pressure (BP) levels in adults, but it is largely unknown whether these genes also determine BP early in life.

Methods: Systolic BP (SBP) and diastolic BP (DBP) were measured in 720 5-year-old children from the WHeezing-Illnesses-STudy-LEidsche-Rijn (WHISTLER) birth cohort in sitting and supine positions using a semi-automatic oscillometric device. Illumina chip technology was used to genotype 18, 19, 11 and 12 single nucleotide polymorphisms associated with adult SBP, DBP, mean arterial pressure (MAP) and hypertension, respectively, in the children's DNA and separate weighted genetic risk scores (GRSs) were constructed. The associations are reported as linear regression coefficients (mmHg BP in childhood/GRS score point) or odds ratios (highest childhood BP quintile/hypertension GRS score point).

Results: A higher GRS for SBP was related to higher supine SBP (0.37, 95% CI 0.01 to 0.7), but not to supine DBP (−0.05, 95% CI −0.4 to 0.3) or supine MAP (0.19, 95% CI −0.1 to 0.5). A higher GRS for DBP was related to a higher supine SBP (0.66, 95% CI 0.1 to 1.2), but not to supine DBP (−0.07, 95% CI −0.6 to 0.4) or supine MAP (0.28, 95% CI −0.2 to 0.7). With the sitting BP measurements, the GRSs for SBP and DBP were related to neither SBP nor DBP. No association was found between GRS for MAP and SBP, DBP or MAP. Hypertension GRS was not associated with a higher BP in children.

Conclusions: Higher scores for adult derived diastolic and systolic BP genes appear to be related to higher supine systolic BP at age 5 years.

Keywords

Blood pressure, genetics, sitting and supine body posture, children

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Introduction

Cardiovascular disease (CVD) is the leading cause of death worldwide and mortality from CVD is increasing.[1] Although CVD events almost exclusively occur later in life, many classical risk factors already exert an influence in early life. This is particularly true for blood pressure (BP),[2] which shows moderate tracking from childhood to adulthood.[3] Early and midlife BP trajectories are also related to CVD events later in life.[4] BP measurements can be performed in sitting, supine or standing postures.[5] A few studies in adults

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have shown contradictory results, but there is agreement that various postures lead to different BP values.[6–8] The differences in BP between different postures are usually attributed to different hydrostatic pressure patterns detectable in veins and capillaries.[9,10]

Both lifestyle and genetics affect the variance in BP.[11,12] It may be speculated that sitting BP is more influenced by factors reflecting both nature and nurture, such as body size, whereas supine BP might more reflect innate BP values predominantly driven by genes. Our previous study in healthy young children showed higher sitting BP values in children with relatively high levels of (intra-)abdominal body fat compared with supine BP values.[13]

Recently acquired knowledge of BP-related genetic variations now enables the study of the mutual relationships between genetic influences on BP variations in early life when environmental influences are still minimal. Genome-wide association studies (GWASs) have identified multiple single nucleotide polymorphisms (SNPs) that are associated with BP in European adults.[14–17] A study has demonstrated no association between adult derived genetic risk scores (GRSs) and sitting systolic BP (SBP), diastolic BP (DBP) or mean arterial pressure (MAP) in healthy young children.[18] Similar results within young children were described by Taal et al.[19] It is unknown to what extent supine BP is already reflected by genetic predispositions during childhood and the association between hypertension GRS and childhood BP has not been investigated. The purpose of this study was to examine the associations between various adult derived genetic BP scores and BP measured in both sitting and supine postures in healthy young children.

Methods

Study design and study population

This study was part of the WHeezing-Illnesses-STudy-LEidsche-Rijn (WHISTLER) prospective, open population birth cohort study. Participants in WHISTLER live in Leidsche Rijn, a new residential area under development near the city of Utrecht in the Netherlands. Almost 100,000 people of different ages, social, cultural and economic backgrounds live in this area. From 2001 onwards, WHISTLER has included 2468 healthy newborn infants. Exclusion criteria are gestational age <36 weeks, major congenital abnormalities and neonatal respiratory disease. From 2007, the cohort has focused on determinants of vascular development in early life. Newborn infants were recruited until 2014 and were actively followed throughout childhood. When the children reach the ages of 5 and 8–10 years, they are invited to our local outpatient clinic.[20,21]

At the current time, 2156 WHISTLER-children are aged at least 5 years. Of these, 924 have visited our outpatient clinic; 7, 11 and 186 children were excluded based on gestational age <36 weeks, missing BP measurements and missing GRSs, respectively. The study population therefore consisted of 720 5-year-old children who had at least one BP measurement and GRS available. BP was measured in both sitting ($n = 719$) and supine ($n = 658$) postures (Supplementary data, available online). The paediatric medical ethics committee of the University Medical Centre Utrecht approved the study. Written informed consent was obtained from the children's parents.[20]

Neonatal visit and follow-up in infancy

The parents of eligible children were invited to visit our outpatient clinic when their infant was *c*. 4 weeks old. Anthropometric measurements were made, a buccal swab was collected and questionnaires were completed by the parents. After this visit the parents were asked to complete a questionnaire dealing with growth, development, respiratory symptoms and disease every month for a period of 12 months. Information about parental characteristics was obtained as part of the Utrecht Health Project, which has collected data on health and disease from all Leidsche Rijn inhabitants since 2000.[20] Informed consent included permission to use relevant child health information obtained from general practitioners in Leidsche Rijn[20] (see Supplementary data, available online, for details about DNA sampling and quality control).

Genetic risk scores

The SBP, DBP and hypertension GRSs were constructed using data from the latest GWAS conducted in 200,000 adults of European ancestry. This GWAS identified 29 independent SNPs at 28 loci that were associated with SBP, DBP, both, or hypertension.[16] Another GWAS performed in 120,000 adults of European ancestry identified 22 SNPs related to MAP.[17]

A total of 10 of the 29 SBP and DBP SNPs found at the genome-wide significance threshold ($p < 5 \times 10^{-8}$) were available on our chip and 10 further alternative BP SNPs were selected that were in strong linkage disequilibrium ($r^2 > 0.8$) with the corresponding GWAS-identified SNPs. Nine of the 29 hypertension SNPs were available on the genotyping array and three other SNPs were highly correlated ($r^2 > 0.8$) with the remaining GWAS-identified SNPs. Of the 22 MAP-related SNPs, we selected six SNPs and five other SNPs that were in strong linkage disequilibrium ($r^2 > 0.8$). If several correlated SNPs were reported, the SNPs with the

lowest reported p values were chosen. In total, 18, 19, 11 and 12 SNPs associated with SBP, DBP, MAP and hypertension ($p < 5 \times 10^{-8}$), respectively, were considered for inclusion and passed quality control tests.

The SNPs were used to calculate separate weighted GRSs by multiplying the reported effect size of each selected SNP with the child's genotype (coded 0, 1 or 2) and then summing all values. The SBP GRS ranged from −2.4 to 18.0 mmHg/point, the DBP GRS from −3.7 to 11.3 mmHg/point, the MAP GRS from −5.8 to 4.4 mmHg/point and the hypertension GRS from 0.4 to 1.5 mmHg/point. The weighting was based on the effect size reported for SBP, DBP, MAP or hypertension related SNPs in the most recent and largest GWASs.[16,17] Details of the SNPs and weights can be found in the Supplementary data, available online.

Follow-up visit in 5-year-old children

The children visited the outpatient clinic at the age of 5 years. Anthropometric measures were obtained by investigators and trained paediatric research nurses with the children wearing indoor clothes, without shoes and standing with their feet slightly apart. Body mass index was calculated as weight in kilograms divided by height in metres squared. Waist circumference was measured twice bare-chested at the level midway between the lowest rib border and the iliac crest. Hip circumference was also measured twice by measuring the smallest circumference of the hip near to the upper face of the hip joint. Abdominal fat measures were obtained ultrasonographically as described previously.[27,28] Information about child and parental characteristics in previous years was obtained by questionnaires.[29,30]

The children's SBP and DBP were measured three times at the right brachial artery, separately in (1) sitting and (2) supine postures, using a semi-automatic oscillometric device (DINAMAP, Critikon, Tampa, FL, USA). Appropriate cuff sizes relative to the diameter of the arm were used. The first measurement was made after 5 minutes rest and 2 minutes of quietness was held between measurements. The means of the SBP and DBP recordings were used for analyses. MAP was derived from the mean SBP and DBP values as follows: DBP+(SBP−DBP)/3.

Data analysis

The means and variances of parent and child characteristics were calculated by the median SBP GRS. Pearson correlation coefficients were obtained for the associations between sitting and supine BP. We considered the correlation moderate to be when $0.50 < r < 0.70$ and high when $r > 0.70$. Associations between SBP, DBP and MAP parameters and GRSs were assessed with multivariate general linear modelling, with SBP, DBP and MAP as simultaneous dependent variables, thus accounting for their mutual correlation, and GRSs for SBP, DBP and MAP as independent variables. Separate models were run for BP measurements in the sitting and supine positions. To test whether sex acted as an effect modifier, an interaction term was added to the models.

The association between hypertension GRS and normal vs. high BP was investigated. First, the children were categorized into different height percentiles according to standard height charts corrected for age and sex. Second, the children were categorized in normal or high BP categories based on their BP values corrected for height percentiles and sex. We calculated for each child whether or not it had normal BP based on classified blood pressure tables, separately for sex and age. Standard height charts were used to categorize a child in a height percentile. Then, a child was categorized in the normal BP group if SBP and DBP values were equal to standardized values of the 90th percentile or lower; and categorized in the higher BP group if SBP or DBP values were higher than the 90th percentile. We have choosen this cut-off point as in the clinic prehypertension is defined as BP values >90 th percentile and hypertension is defined as BP values equal to the 95th percentile or higher[31] Logistic modelling was performed to assess whether hypertension GRS was associated with being in the higher BP group based on sitting or supine measurements.

As height and weight are important determinants for BP and possibly affect BP via growth-related SNPs, additional explanatory analyses aimed to investigate whether associations between GRSs and particularly sitting BP depended on adjustment for height and weight. To that end, general linear regression models were performed simultaneously for SBP, DBP and MAP with height and weight as covariates. The residuals were saved, thus removing the variation in BP due to height and weight influences. Subsequently, the residuals were related to GRS.

The results are expressed as linear regression coefficients with 95% confidence intervals (95% CIs) and p values and odds ratios (95% CIs, p values) for the GRS hypertension analyses. Confidence intervals not including zero (or one for GRS hypertension), corresponding to two-sided $p < 0.05$, were considered to be statistically significant. All analyses were performed using SPSS version 21.0 for Windows.

Results

Table 1 shows the general characteristics of the children. The mean ± SD GRSs were 8.1 ± 1.6 for SBP and 4.4 ± 1.0 for DBP. In the sitting posture, the

Table 1. Baseline characteristics of WHISTLER offspring ($n = 720$).

	N	Low SBP GRS (≤8.1 points) (N = 362)	High SBP GRS (>8.1 points) (N = 358)	p
WHISTLER offspring at birth				
Male sex	720	49.4	48.6	0.82
Birth weight (g)	694	3528 ± 512	3540 ± 493	0.76
Birth height (cm)	616	50.9 ± 2.4	51.1 ± 2.2	0.18
Mother's age (years)	632	32.8 ± 3.7	32.6 ± 3.5	0.57
% NOT exposed to smoke during pregnancy	695	78.0	80.3	0.46
WHISTLER follow-up age 5 years				
Age (years)	719	5.4 ± 0.5	5.4 ± 0.3	0.14
Height (cm)	720	115 ± 5.1	115 ± 4.4	0.72
Body mass index (kg/m^2)	720	15.2 ± 1.4	15.2 ± 1.3	0.75
Waist circumference (cm)	714	53.1 ± 3.5	52.9 ± 3.5	0.52
Hip circumference (cm)	713	58.8 ± 3.9	58.8 ± 3.5	0.84
No exposure to smoke during infancy	705	94.9	92.3	0.15

Data are presented as mean ± SD values or percentages.
GRS: genetic risk score; SBP: systolic blood pressure.

mean ± SD SBP was 105 ± 7.5 mmHg, the mean ± SD DBP was 54 ± 7.2 mmHg and the mean ± SD MAP was 73.3 ± 6.6 mmHg. In the supine posture the values were 103 ± 7.1, 53 ± 6.3 and 70.9 ± 5.8 mmHg, respectively. The correlations between sitting and supine SBP ($r = 0.60$, $p < 0.001$) and DBP ($r = 0.50$, $p < 0.001$) were moderate.

Table 2 shows that a higher GRS for SBP seems to be related to a 0.37 mmHg/point higher supine SBP, but not to supine DBP or MAP. Each point increase in the DBP GRS was associated with a 0.66 mmHg higher supine SBP, but not to the supine DBP or MAP. The GRS for SBP and DBP were not related to the sitting BP nor to the sitting MAP. The GRS for MAP was not related to the sitting or supine BP or MAP. There was no effect modification by sex. After eliminating BP variance due to height and weight, the estimates remained almost similar for GRSs (Supplementary data, available online). No association was found between hypertension GRS and being in the >90th percentile BP group in children (Supplementary data, available online).

Discussion

Adult derived genetic BP scores can explain part of the variation in supine SBP already present in young children. A higher BP GRS seems to be associated with a higher systolic childhood BP when measured in the supine posture, but not when measured in a sitting posture. The GRS for MAP was not related to the sitting BP, the supine BP or the MAP. Hypertension GRS was not associated with hypertension in children with BP values >90th percentile.

To our knowledge, our measurements of BP in different body positions in a large group of healthy, young children are unique. With our sample size of *c*. 700 children we were able to show the statistically significant associations presented, but we acknowledge that we may have lacked the statistical power to detect other associations. We used weighted GRSs rather than the less precise unweighted scores.[32] We performed multivariate modelling, including SBP, DBP and MAP, thus reducing the numbers of statistical tests. A limitation is that our GRSs were based on only a part of the GWAS-identified SNPs. However, we included alternative highly correlated SNPs instead of the original GWAS-identified SNPs. Associations might have been stronger or more precise if we could have included all 29 original genome-wide BP associated SNPs. Despite the use of fewer SNPs, we detected a relation between adult derived SBP SNPs and supine SBP in children. Our BP measurements were performed in the same order, but we feel that this will not have changed the rankings of children.

Although genomic DNA or BP measurements were not collected in all children, the exclusion of these participants did not cause selection bias as it is unlikely that exclusion was based on the GRS on BP. However, these results are only applicable to children of European ancestry according to our selection criteria. In addition, the baseline characteristics of the excluded children were comparable with the included children. Both participants and researchers were unaware of the risk SNPs and therefore BP measurement

Table 2. Associations between genetic risk scores and different measures of blood pressure ($n = 720$).

	Children in sitting posture						Children in supine posture					
	Systolic blood pressure (mmHg)		Diastolic blood pressure (mmHg)		Mean arterial pressure (mmHg)		Systolic blood pressure (mmHg)		Diastolic blood pressure (mmHg)		Mean arterial pressure (mmHg)	
	β (95% CI)	*p*	β (95% CI)	*p*	β (95% CI)	*p*	β (95% CI)	*p*	β (95% CI)	*p*	β (95% CI)	*p*
Adult derived genetic risk score for:												
Systolic blood pressure (points)	−0.031 (−0.4; 0.3)	0.86	−0.146 (−0.5; 0.2)	0.42	−0.024 (−0.4; 0.3)	0.89	0.374 (0.01; 0.7)	0.05	−0.047 (−0.4; 0.3)	0.78	0.185 (−0.1; 0.5)	0.23
Diastolic blood pressure (points)	−0.008 (−0.6; 0.5)	0.98	−0.369 (−0.9; 0.2)	0.18	−0.128 (−0.6; 0.4)	0.62	0.655 (0.1; 1.2)	0.02	−0.073 (−0.6; 0.4)	0.77	0.275 (−0.2; 0.7)	0.25
Mean arterial pressure (points)	−0.046 (−0.8; 0.7)	0.90	−0.309 (−1.0; 0.4)	0.39	−0.130 (−0.8; 0.5)	0.70	0.101 (−0.6; 0.8)	0.78	0.016 (−0.6; 0.7)	0.96	0.085 (−0.5; 0.7)	0.78

errors based on the determinant is implausible. Because we measured BP in early childhood and used genomic DNA for analyses, confounding is hardly conceivable. Our findings by median GRSs did not show evidence of confounding.

Systolic blood pressure

We showed that higher GRSs appear to be associated with higher supine SBP, whereas no association was found with regard to sitting SBP. Although speculative, this may suggest that BP level is a polygenic trait from childhood onwards and supine SBP may better represent the innate BP value.

No previous study has been conducted with respect to supine SBP measurements. A few studies determined the influence of GRSs on SBP measured in a sitting posture in children.[18,19,33,34] Our results of an absence of association between GRSs and sitting SBP are in line with the results obtained by Oikonen et al.[34] with their GRS construction of five SBP-related SNPs, as well as with the results of Punwasi et al.[18] and Taal et al.[19] in children of comparable ages to our cohort. However, higher estimates were described in studies using a combination of GRSs.[33,34] The discrepancy is probably due to the inclusion of children with larger age ranges (6–17 and 3–18 years, respectively) because in older children, in particular, associations may be expected to more directly reflect derivation findings in adults. Discrepancy may also be due to different constructions of the GRSs using different SNPs with other effect sizes. The BP GRS of Howe et al.[33] was based on all 29 SNPs identified in the same GWAS. Oikonen et al.[34] constructed a combination GRS with 13 SBP- or DBP-related SNPs, which were identified by another older GWASs.[14,15]

Diastolic blood pressure

We found no statistically significant association between GRSs and DBP, neither in a sitting nor a supine posture. There is no prior study of supine BP measurements, but we could not detect any significant associations with sitting DBP, in agreement with other findings.[18,19] By contrast, Oikonen et al.[34] found a higher sitting DBP in relation to higher GRSs. Again, this is probably explained by their different GRS construction and particularly by the fact that they included both children and adolescents. Although there was no statistically significant association, both SBP and DBP GRS seemed to indicate an inverse association with DBP, particularly in a sitting position. In our view, these findings need further exploration, probably in a larger cohort, as the combined findings could point to a relation between adult derived GRS and early life pulse pressure as an indicator of early life vascular wall

elasticity.[35] We demonstrated no association between GRSs and MAP. This was in line with the results of Punwasi et al.[18]

Weight and height are well known determinants of BP.[36–39] BP SNPs are possibly associated with height- and weight-related SNPs, thus growth-related genes might also affect BP. However, our explanatory analyses with additional adjustment for weight and height demonstrated findings consistent with our original analyses.

We have demonstrated that known hypertension alleles have no detectable effect on relatively high BP already present in early childhood. BP was only measured at one point in time, which may result in a misclassification of BP values, particularly of high BP. We acknowledge that we may lack the statistical power for each of these associations and further research is needed.

In our study, sitting and supine BP were moderately correlated and the average SBP, DBP and MAP were 2, 1 and 2 mmHg, respectively, higher in a sitting than in a supine position. Our findings suggest that genetic BP variation particularly explains supine BP. The main relevance is that posture does make a difference in childhood BP and that this difference is important in revealing the genetic origins of childhood BP. It was noted that individual genes that each have very limited sized associations with BP in adulthood do, in combination, play a detectable part from very early life onwards. This indicates that there is a constitutional BP level in young childhood with a substantial environmental variation around it. The effect sizes of the risk SNPs in childhood are modest and do not in our view directly support individual children's risk assessments. However, our findings could be relevant from a population point of view, as small increases in adult BP result in an increased vascular mortality risk.[40] BP shows moderate tracking from childhood to adulthood[3] and early and midlife BP trajectories are related to CVD events later in life.[4]

Our findings thus provide evidence in the direction of the view that higher scores for adult derived SBP and DBP genes are associated with higher supine systolic BP in childhood as early as 5 years of age.

Perspectives

GWASs have identified several genetic variations associated with BP in adults. Genetic risk models based on these SNPs were associated with BP in adults. The aim of current study was to determine whether these GRSs also predict BP in healthy young children. This study contributes to the understanding of the aetiology of BP early in life. We demonstrated that higher GRSs for BP result in higher supine SBP. As small increases in BP already give a tendency to higher vascular mortality, these findings are important from a population point of view. Associations were only found in supine measurements, indicating that body posture does make a difference in childhood BP and that this difference is important in revealing the genetic origins of childhood BP. BP values are influenced by both nature and nurture and information from GRS on BP could be beneficial in classifying children into a (pre)hypertension category. It is conceivable that GRS, although not a perfect predictor of individual BP, could, in conjunction with other risk factors, be useful in improving the classification of children as having normal BP or hypertension. However, further dedicated research in this field is necessary.

Author contribution

The authors' responsibilities were as follows: MACJ, GWD, and CSPMU contributed to the study conceptualization and design. ML, NCO and AHM were responsible for quality control and GRSs construction. MACJ analysed the data and wrote the manuscript. MACJ, GWD and CSPMU contributed in the interpretation of the results. CSPMU, FLJV, DEG, CKE and GWD provided constructive feedback on each draft of the manuscript. All authors read and approved the final manuscript.

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