The 23K Variant of the R23K Polymorphism in the Glucocorticoid Receptor Gene Protects against Postnatal Growth Failure and Insulin Resistance after Preterm Birth

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Context: Preterm birth is associated with postnatal growth failure, abdominal fat accumulation, insulin resistance, and hypertension, resembling increased glucocorticoid bioactivity.

Objective: We tested the effects of the R23K and N363S polymorphisms in the glucocorticoid receptor gene, associated with decreased and increased sensitivity to cortisol, respectively, on linear growth and the adult metabolic profile in a cohort (n = 249) of men and women born less than 32 gestational weeks and followed up prospectively from birth until 19 yr of age.

Design and Participants: This was a birth cohort study that included 249 19-yr-old survivors born at a gestational age less than 32 wk from the Dutch Project on Preterm and Small-for-Gestational-Age Infants cohort.

Setting: This project was a nationwide multicenter follow-up study.

Main Outcome Measures: Linear growth and adult body composition, fasting cortisol, glucose, insulin, and cholesterol concentrations, and blood pressure were measured.

Results: The 23K variant (n = 24) was associated with lower fasting insulin levels (mean difference after log transformation: −0.09 [95% confidence interval −0.16, −0.01] mU/liter) and a lower homeostatic model assessment for insulin resistance index (mean difference after log transformation: −0.09 [95% confidence interval −0.16, −0.01]) as well as with a taller stature departing from the age of 1 yr onward. 23K carriers showed complete catch-up growth between the ages of 3 months and 1 yr, and attained height was similar to the population reference mean, whereas stature in noncarriers was on average 0.5 sd below this mean. In contrast, the N363S polymorphism was not associated with any of the outcomes.

Conclusions: Carriers of the 23K variant are, at least in part, protected against postnatal growth failure and insulin resistance after preterm birth. (J Clin Endocrinol Metab 92: 4777–4782, 2007)

FUNCTIONAL CHANGES IN the gene encoding the glucocorticoid receptor (GR) play an important role in glucocorticoid bioactivity. To date, several functional polymorphisms in the GR gene (NR3C1) have been identified, including R23K (ER22/23EK) and N363S. The 23K variant has been associated with decreased sensitivity to glucocorticoids and a beneficial metabolic health (1). Elderly persons with this variant had lower levels of fasting insulin, and of total and low-density lipoprotein (LDL) cholesterol (1). Thirty-six-year-old men with this variant had a taller stature, more lean body mass, and greater muscle strength, whereas their female contemporaries had a tendency toward a smaller waist circumference (2). Unfavorable effects were found with the 363S variant, which has been associated with increased sensitivity to glucocorticoids (3).

Subjects carrying this variant were predisposed to obesity and coronary artery disease (3–6).

A number of recent studies have elucidated the long-term metabolic consequences of preterm birth. These consequences include an increased risk of short stature (7, 8), abdominal fat accumulation (9), insulin resistance (10–12), and hypertension (12–14). The clustering of postnatal growth failure and metabolic risk factors in individuals born preterm is indicative for, and/or resembles, effects of increased glucocorticoid bioactivity. Indeed, in a small sample of young adults, it was found that basal cortisol levels were higher after preterm birth (15). It is unknown whether common variants in the GR gene could explain variations in the endocrine-metabolic state of adults born prematurely.

Therefore, we tested the effects of the R23K and N363S polymorphisms on linear growth, body composition, insulin resistance, the serum lipid profile, and blood pressure in a cohort of 19-yr-old men and women who were born very preterm (i.e. < 32 gestational weeks).

Subjects and Methods

Subjects

The Project on Preterm and Small-for-Gestational-Age Infants (POPS) study is a nationwide, multicenter, prospective follow-up study, com-

First Published Online September 11, 2007

Abbreviations: GR, Glucocorticoid receptor; HOMA-IR, homeostatic model assessment for insulin resistance index; HPA, hypothalamus-pituitary-adrenal; LDL, low-density lipoprotein; POPS, Project on Preterm and Small-for-Gestational-Age Infants; SDS, sd score; SNP, single-nucleotide polymorphism.

JCEM is published monthly by The Endocrine Society (http://www.endo-society.org), the foremost professional society serving the endocrine community.
TABLE 1. Perinatal characteristics of participants by GR genotype

| Characteristic                      | Genotype                      | P value | 23K vs. noncarriers | 363S vs. noncarriers |
|-------------------------------------|-------------------------------|---------|---------------------|----------------------|
|                                     | R23/23K | N363/363S | Noncarriers |                     |                      |
| n                                   | 24      | 15        | 210          |                      |                      |
| Males (%)                           | 12      | 6         | 102          | 0.89                 | 0.52                |
| Obstetric                           |         |           |              |                      |                      |
| Maternal age (yr)                   | 28.1 ± 4.5 | 27.5 ± 3.3 | 27.2 ± 5.4 | 0.44                 | 0.82                |
| Purity > 0 (%)                      | 12 (50.0%) | 6 (40.0%) | 100 (47.6%) | 0.83                 | 0.57                |
| Part of multiple pregnancy (%)      | 2 (8.3%) | 2 (13.3%) | 54 (25.7%) | 0.06                 | 0.48                |
| Hypertension during pregnancy (%)   | 8 (33.3%) | 1 (6.7%)  | 35 (16.7%) | 0.06                 | 0.48                |
| Drugs and alcohol intoxication (%)  | 9 (37.5%) | 7 (46.7%) | 113 (53.8%) | 0.13                 | 0.59                |
| Prolonged rupture of membranes (%)  | 2 (8.3%) | 4 (26.7%) | 51 (24.3%) | 0.08                 | 0.76                |
| Maternal glucocorticoid treatment (%)| 7 (29.2%) | 4 (26.7%) | 42 (20.0%) | 0.30                 | 0.52                |
| Body proportions at birth           |         |           |              |                      |                      |
| Gestational age (wk)                | 30 ± 1.4 | 30.0 ± 1.5 | 29.9 ± 1.5 | 0.34                 | 0.73                |
| Birth weight (grams)                | 1321 ± 268 | 1449 ± 411 | 1335 ± 327 | 0.83                 | 0.20                |
| SDS                                 | -0.33 ± 0.93 | 0.19 ± 0.91 | -0.13 ± 1.03 | 0.37                 | 0.24                |
| Postnatal clinical course           |         |           |              |                      |                      |
| Respiratory distress syndrome (%)   | 10 (41.7%) | 7 (46.7%) | 102 (48.6%) | 0.52                 | 0.89                |
| Intracranial hemorrhage (%)         | 5 (20.8%) | 2 (13.3%) | 36 (17.1%) | 0.58                 | 1.00                |
| Sepsis (%)                          | 12 (50.0%) | 4 (26.7%) | 70 (33.5%) | 0.11                 | 0.59                |
| Postnatal glucocorticoid treatment (%)| 0 (0%)   | 4 (26.7%) | 14 (6.7%)  | 0.37                 | 0.02                |

Values represent mean ± SD or percent. Continuous variables were compared with the unpaired t test. Dichotomous variables were compared by the χ² test or Fisher’s exact test where appropriate.

praising 94% of all live-born very preterm (<32 wk gestation) and/or very low-birthweight (<1500 g) infants born in The Netherlands in 1983 and has documented birth, growth, and a number of other characteristics from birth onward (16, 17). At the follow-up visits (at the age of 3 months, 6 months, 1 yr, and 2 yr postterm and at the chronologic age of 5 yr), length/height was measured. Length until the age of 2 yr was measured to the nearest 1 cm in supine position, fully extended with the heels in contact with a baseboard. Standing height at the age of 5 yr was measured to 1-mm accuracy. At 19 yr of age, all 637 living subjects born with a gestational age less than 32 wk who were free from congenital skeletal deformations, Down syndrome, chromosomal abnormalities, multiple congenital deformations, or inborn errors of metabolism and who were not born to mothers with gestational diabetes were approached by mail to participate in the POPS-19 study. Of these subjects, 395 consented to participate (62% response rate). Subjects with diabetes mellitus or on thyroid hormone or systemic corticosteroids as well as non-Caucasian subjects and pregnant women were excluded for this specific study. The data of nonfasted subjects were also not analyzed. The approval of the medical ethical committees of all participating centers was obtained for the POPS-19 study.

Study protocol

Subjects who gave written informed consent to participate were seen after an overnight fast between 0830 and 1000 h at one of the outpatient clinics of the 10 participating centers. Assessors were blinded with respect to the perinatal characteristics of the subjects.

After 30 min in supine position, systolic and diastolic blood pressure was measured three times consecutively with an automatic blood pressure device (Dinamap; Critikon, Norderstedt, Germany) on the non-dominant arm. The mean values of these measurements were used in the statistical analyses. The cuff size was adjusted to fit arm length and circumference. Venous blood was subsequently obtained in supine position. Thereafter anthropometry was performed, for which assessors had received extensive training before the study and retaining during the entire study period at 2-month intervals. Subjects were measured barefoot while wearing underwear only. Weight was measured to the nearest 0.1 kg on a balance scale and height to the nearest 0.1 cm with a fixed stadiometer. Waist and hip circumferences were measured at 0.1 cm accuracy using standard methods (18). Four skinfold thickness measurements were made on the left side of the body at triceps, biceps, subscapular, and iliacal regions. From these measurements, fat mass was calculated using the equations of Durnin and Rahaman (19). A more detailed description of skinfold thickness measurements obtained in the POPS-19 study has been published elsewhere (9).

Laboratory analyses

Blood samples were stored at −80 C and thawed only once, immediately before analysis. Glucose and total cholesterol were measured in a fully automated computerized laboratory system with a Hitachi 747 chemistry analyzer (Hitachi, Tokyo, Japan). High-density lipoprotein

TABLE 2. Length/height SDS by GR genotype

| Follow-up visit | Genotype (mean ± SD) | Comparisons | Mean difference (95% CI) |
|-----------------|----------------------|-------------|-------------------------|
|                 | R23/23K              | N363/363S   | Noncarriers             |
| Birth           | -0.45 ± 0.55         | -0.12 ± 0.71| -0.46 ± 0.99            | -0.10 ± 1.14 | -0.05 ± 1.29 | 0.01 (0.86, 0.44) | -0.09 (0.89, 0.71) |
| 3 months        | -0.74 ± 0.92         | -1.21 ± 0.94| -1.70 ± 0.59            | -0.99 ± 1.41 | -0.77 ± 1.33 | 0.09 (0.69, 0.50) | 0.23 (0.51, 0.97) |
| 6 months        | -0.01 ± 0.16         | -0.57 ± 0.97| -1.46 ± 0.98            | -0.66 ± 1.13 | -0.83 ± 1.26 | 0.44 (0.12, 0.99) | 0.15 (0.80, 0.50) |
| 1 yr            | 0.69 ± 1.10         | -0.19 ± 0.72| -0.74 ± 0.76            | -0.53 ± 0.84 | -0.51 ± 1.12 | 0.84 (0.30, 1.38) | 0.02 (0.59, 0.63) |
| 2 yr            | 0.49 ± 0.81         | -0.11 ± 0.68| -0.45 ± 0.62            | -0.60 ± 1.22 | -0.44 ± 1.32 | 0.68 (0.29, 1.07) | 0.19 (0.47, 0.85) |
| 5 yr            | 0.51 ± 0.81         | -0.19 ± 0.62| -0.65 ± 0.53            | -0.62 ± 0.97 | -0.39 ± 1.00 | 0.66 (0.25, 1.08) | 0.0 (0.52, 0.51) |
| 19 yr           | 0.14 ± 0.64         | -0.31 ± 0.60| -0.66 ± 0.44            | -0.47 ± 1.03 | -0.48 ± 1.11 | 0.39 (0.08, 0.70) | 0.15 (0.41, 0.70) |

CI, Confidence interval.

* For gender-genotype interaction, P < 0.05.
and LDL were measured with a turbidimetric assay on a Hitachi 911. Cortisol was measured with a fluorescence polarization immunoassay on an Abbott TDX (Abbott Laboratories, Abbott Park, IL). The sensitivity of this assay is 20 nmol/liter and the interassay coefficient of variation ranges from 3.1 to 6.4% at different levels. Insulin and C-peptide were measured with highly sensitive RIAs (Linco, St. Charles, MO). The detection levels of these assays are 0.1 mU/liter and 0.03 nmol/liter, respectively, and the interassay coefficients of variation range from 4.7 to 12.2% and 3.2 to 9.3% at different levels, respectively. Homeostatic model assessment for insulin resistance index (HOMA-IR) was calculated (20). Insulin and C-peptide levels and HOMA-IR were used as parameters of insulin resistance. Fasting insulin levels and HOMA-IR correlate strongly with insulin sensitivity assessed by the frequently sampled iv glucose tolerance test in young persons (21, 22).

For both the R23K (rs6190) and N363S (rs6195) single-nucleotide polymorphisms (SNPs), PCRs were performed using 2.5 ng of genomic DNA and standard reagents. SNPs were subsequently genotyped by mass spectrometry (homogeneous mass array system; Sequenom Inc., San Diego, CA), using standard conditions. Genotypes were analyzed by using Genotyper 3.0 software (Sequenom). We identified 24 subjects (9.6%) who were heterozygous for the 23K variant (12 men and 12 women) and 15 (6.0%) who were heterozygous for the 363S variant (six men and nine women). None of the subjects was carrier of both variants. The corresponding allele frequencies of 4.8 and 3.0%, respectively, were reasonably well in range with the allele frequencies observed in healthy Dutch populations, ranging from 3 to 4.5% for the 23K variant and from 3 to 5% for the 363S variant (1–3, 23, 24). For both SNPs, the genotype distribution was in agreement with the distribution predicted by the Hardy-Weinberg equilibrium ($P_0 = 0.42$ for the R23K polymorphism and $P_0 = 0.62$ for the N363S polymorphism).

Statistical analysis

Auxological data at birth and on subsequent occasions were converted to sd scores (SDSs) to correct for (gestational) age and sex, using Swedish references for preterm infants (25) and recently collected Dutch references (18, 26, 27), respectively. Comparisons were made between minor allele carriers and noncarriers, using the independent-samples t test. Outcomes with skewed distributions (cortisol, insulin, and HOMA-IR) were log transformed before statistical comparison. Analyses were repeated with adjustment for perinatal factors (obstetric characteristics, gestational age, and postnatal clinical course) using linear regression analysis. Modification by gender of the effect of genotype on outcomes was tested by first including the variables genotype (in which minor allele carrier = 1 and noncarrier = 0) and gender (in which male = 1 and female = 0) in a linear regression analysis followed by the inclusion of their product.

Results

Table 1 lists the perinatal characteristics of the 249 participants, showing that, apart from an unequal distribution in the numbers who had been treated with glucocorticoids as neonates, there were no statistically significant differences between the GR genotypes.

Table 2 summarizes the growth patterns of the groups up to adult height. 23K carriers and noncarriers showed a similar degree of catch-down growth between birth and the age of 3 months. Between the ages of 3 months and 1 yr, 23K carriers showed more rapid catch-up growth than noncarriers. Stature at 1 yr and beyond was greater than or similar to the population reference mean in carriers of the 23K variant, whereas in noncarriers it was on average 0.5 sd below this mean. Correction for perinatal factors only slightly reduced the strength of these associations (data not shown). Figure 1, A–C, shows that the difference in linear growth between 23K carriers and noncarriers was more pronounced in men, although the direction of association was similar for women. Despite these sex-specific observations, the test for interaction showed that the association between the R23K polymorphism and stature was dependent on gender only at 5 yr of age. Linear growth of 363S carriers did not differ from noncarriers.

Table 3 shows the adult metabolic profile for the GR genotypes. 23K carriers had lower fasting insulin levels and a lower HOMA-IR than noncarriers. These differences became somewhat larger after correction for perinatal factors (data not shown). In addition, 23K carriers had a lower waist-to-hip ratio, but this observation did not reach statistical significance. Interaction between the R23K polymorphism and gender on total and LDL cholesterol levels was observed, which was explained by opposite influences in men and women of
the 23K variant on cholesterol levels. The adult metabolic profile of 363S carriers did not differ from noncarriers.

**Discussion**

In this prospective study in subjects who were born very preterm (i.e. < 32 gestational weeks) and followed up until 19 yr of age, we found that the 23K variant in the GR gene was associated with lower fasting insulin levels and a lower HOMA-IR as well as with a shorter stature departing from the age of 1 yr. It was also associated with a smaller waist to hip circumference, although this observation was not statistically significant. Carriers of the 23K variant showed complete catch-up growth between the ages of 3 months and 1 yr and attained height was similar to the population reference mean. The N363S polymorphism was not associated with any of these outcomes.

In our study, we found that mean adult stature in carriers of the 23K variant was similar to the population reference mean, whereas mean height in the noncarriers was approximately 0.5 sd below this mean. Previous studies in healthy men of different ages found that 23K carriers were on average 4 cm taller than noncarriers (2, 24). This difference in final height was for an important part attributed to the pubertal growth spurt (2). In contrast, we found in our specific population of very preterm subjects that the growth pattern differed significantly between carriers and noncarriers already by the age of 1 yr. However, the difference (in SDS) between 23K carriers and noncarriers did not further increase after puberty. Furthermore, a large number of genome-wide linkage scans have been performed aiming at the detection of new chromosomal loci influencing the quantitative trait height (28). Notably, the chromosomal region of the GR gene on chromosome 5q31 is one of the regions that have been implicated in more than one of such studies (29, 30). Possibly, functional genetic variation at the GR gene is explaining these linkages and may be considered a strong positional candidate gene.

In addition, in this specific cohort, we found that the 23K variant was associated with lower fasting insulin levels and a lower HOMA-IR at only 19 yr of age, in line with findings from others in elderly people (1). Observations in other cohorts of effects of the R23K polymorphism on body composition and the serum lipid profile (1, 2) were not confirmed by our data. Furthermore, we found no statistically significant relations with blood pressure.

Experiments in rats have shown that nonhandling during early postnatal development permanently increases hypothalamus-pituitary-adrenal (HPA) axis activity (31). Similar effects in offspring were observed with naturally low-grooming mothers (32). Furthermore, it has been indicated that the extent of grooming in rat mothers specifically alters the methylation at the GR gene promoter in the hippocampus, thereby explaining how the effect of maternal care might persist into adulthood (33). During their neonatal course, preterm newborns are to a large extent devoid of maternal care and, instead, are subject to many stressful and sometimes even critical events, including, for example, respiratory distress, intubation and mechanical ventilation, and frequent blood sampling. Therefore, it could be possible that adverse postnatal circumstances in humans may also result in life-long activation of the HPA axis. This is supported by data from a small study in young adults, showing that basal cortisol levels are elevated after preterm birth (15). The current findings suggest that the 23K variant protects, at least in part, against postnatal growth failure and insulin resistance after preterm birth. We speculate that an extreme stressful event such as preterm birth may induce hypermethylation of the GR promoter, leading to less GR expression in central feed-back regions and hence enhanced stress responsiveness. GR expression in carriers of the 23K variant may be less vulnerable to alterations in DNA methylation. Indeed, SNPs have been shown to be associated with methylation of neighboring Cpg sites (34).

The functionality of the studied variants has been elucidated previously. The 23K variant has been associated with higher circulating cortisol levels after overnight dexamethasone suppression (1), whereas the 363S variant has been associated with lower postdexamethasone cortisol (3). These findings were subsequently confirmed by transfection study.

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**Table 3. Metabolic profile at 19 yr of age by GR genotype**

| Outcome                  | Genotype (mean ± sd) | Noncarriers (n = 102) | Men (n = 10) | Women (n = 92) |
|--------------------------|----------------------|-----------------------|--------------|----------------|
|                          | R23K                 | N363S/363S            | Men (n = 6)  | Women (n = 9)  |
| BMI (SDS)                | −0.44 ± 1.04         | 0.05 ± 0.55           | 0.07 ± 1.26  | 0.15 ± 1.01    |
| Waist (SDS)              | 0.20 ± 1.05          | 0.71 ± 0.50           | 0.57 ± 1.14  | 0.84 ± 0.97    |
| WHR (SDS)                | 0.53 ± 0.90          | 0.57 ± 0.41           | 0.66 ± 0.67  | 0.85 ± 0.85    |
| Absolute fat mass (kg)   | 11.1 ± 5.3           | 19.2 ± 3.1            | 11.8 ± 6.8   | 20.5 ± 6.7     |
| Fat percentage (%)       | 15.1 ± 4.7           | 31.2 ± 3.7            | 15.7 ± 6.1   | 31.2 ± 5.3     |
| Insulin (mU/liter)       | 0.82 ± 0.13          | 0.91 ± 0.16           | 0.80 ± 0.17  | 0.91 ± 0.19    |
| C-peptide (nmol/liter)   | 0.59 ± 0.15          | 0.66 ± 0.19           | 0.48 ± 0.13  | 0.68 ± 0.16    |
| HOMA-IR                  | 0.17 ± 0.16          | 0.24 ± 0.18           | 0.23 ± 0.19  | 0.23 ± 0.19    |
| Total cholesterol (mmol/liter) | 3.61 ± 0.58     | 4.85 ± 0.58           | 4.13 ± 0.50  | 4.59 ± 0.96    |
| HDL (mmol/liter)         | 1.27 ± 0.25          | 1.37 ± 0.22           | 1.40 ± 0.37  | 1.54 ± 0.35    |
| LDL (mmol/liter)         | 2.06 ± 0.42          | 3.02 ± 0.61           | 2.34 ± 0.67  | 2.68 ± 0.84    |
| Systolic BP (mm Hg)      | 128 ± 18             | 118 ± 12              | 123 ± 5      | 122 ± 12       |
| Diastolic BP (mm Hg)     | 63 ± 9               | 66 ± 9                | 65 ± 6       | 69 ± 9         |
| Cortisol (nmol/liter)    | 2.62 ± 0.12          | 2.92 ± 0.12           | 2.60 ± 0.12  | 2.88 ± 0.20    |

**Comparisons, Mean difference (95% CI)**

- 23K vs. noncarriers
- 363S vs. noncarriers

| Outcome                  | Genotype (mean ± sd) | Noncarriers (n = 108) | Men (n = 10) | Women (n = 98) |
|--------------------------|----------------------|-----------------------|--------------|----------------|
|                          | R23K                 | N363S/363S            | Men (n = 6)  | Women (n = 92) |
| BMI (SDS)                | −0.20 ± 1.06         | 0.14 ± 1.13           | 0.20 ± 1.01  | 0.78 ± 0.92    |
| Waist (SDS)              | 0.20 ± 1.03          | 0.84 ± 0.97           | 0.20 ± 1.01  | 0.84 ± 0.92    |
| WHR (SDS)                | 0.73 ± 0.96          | 0.94 ± 0.92           | 0.30 ± 0.61  | 0.02 (0.04, 0.38) |
| Absolute fat mass (kg)   | 11.5 ± 5.0           | 18.4 ± 5.8            | 0.19 (0.01, 0.39) |
| Fat percentage (%)       | 16.2 ± 5.0           | 29.9 ± 6.4            | 0.44 (0.33, 0.55) |
| Insulin (mU/liter)       | 0.63 ± 0.16          | 0.95 ± 0.17           | 0.09 (0.16, 0.03) |
| C-peptide (nmol/liter)   | 0.68 ± 0.24          | 0.69 ± 0.21           | 0.06 (0.15, 0.04) |
| HOMA-IR                  | 0.30 ± 0.20          | 0.28 ± 0.18           | 0.09 (0.16, 0.01) |
| Total cholesterol (mmol/liter) | 3.98 ± 0.84     | 4.41 ± 0.87           | 0.04 (0.32, 0.42) |
| HDL (mmol/liter)         | 1.20 ± 0.24          | 1.44 ± 0.35           | 0.01 (0.14, 0.13) |
| LDL (mmol/liter)         | 2.36 ± 0.74          | 2.59 ± 0.76           | 0.06 (0.26, 0.38) |
| Systolic BP (mm Hg)      | 127 ± 12             | 121 ± 12              | 0.07 (0.05, 0.19) |
| Diastolic BP (mm Hg)     | 65 ± 5               | 68 ± 8                | 0.08 (0.02, 0.17) |
| Cortisol (nmol/liter)    | 2.59 ± 0.14          | 2.79 ± 0.24           | 0.02 (0.03, 0.46) |

CI, Confidence interval; WHR, waist-to-hip ratio; BP, blood pressure; HDL, high-density lipoprotein.

* For gender-genotype interaction, P < 0.05.
ies, showing decreased and increased gene expression in response to GR binding, respectively (35).

Because our participants were genotyped at 19 yr of age, a survivor effect of GR variation could not be excluded, considering the high neonatal mortality rate in the original cohort (16, 17). However, an argument against selective survival of a particular genotype is that the observed genotype frequencies of the studied polymorphisms did not deviate much from the genotype frequencies in the normal Dutch population, implying that gross selective survival of a particular genotype is not very likely to have occurred in our population.

Although sex specificity of the effects of the R23K polymorphism on linear growth and body composition have been reported by one study (2) and was attributed to a different regulation of HPA axis activity by androgens and estrogens, we did not find much evidence for sexually dimorphic effects of the R23K polymorphism, except for height at 5 yr of age and adult cholesterol levels. Clearly, any sex-specific observation must be balanced against the small numbers of subjects carrying the 23K variant (12 men and 12 women).

In conclusion, we found in 19-yr-old survivors of very preterm birth that the 23K variant was associated with lower fasting insulin levels and a lower HOMA-IR as well as with a taller stature departing from the age of 1 yr. Carriers of the 23K variant showed complete catch-up growth between the ages of 3 months and 1 yr and attained height was similar to the population reference mean. Therefore, carriers of the 23K variant are, at least in part, protected against postnatal growth failure and insulin resistance after preterm birth.

Acknowledgments

We thank Dennis Kremer for genotyping. Participants in the Dutch POPS-19 Collaborative Study Group are: TNO Quality of Life, Leiden (E. T. M. Hille, C. H. de Groot, H. Kloosterboer-Boerrigter, A. L. den Ouden, A. Rijpstra, S. P. Verloove-Vanhoorick, J. A. Vogelaar); Emma Children’s Hospital AMC, Amsterdam (J. H. Kok, A. Ilens, M. van der Lans, W. J. C. Boelen-van der Lou, T. Lundqvist, H. S. A. Heymans); University Hospital Groningen, Beatrix Children’s Hospital, Groningen (E. J. S. van der Worven, W. R. van den Bergh, L. G. Venken, E. J. L. E. Vrijlandt); University Hospital Maastricht, Maastricht (A. L. M. Mulder, A. Gerver); University Medical Center St. Radboud, Nijmegen (L. A. A. Kollée, L. Reijmers, R. Sonnemans); Leiden University Medical Center, Leiden (J. M. Wit, F. W. Dekker, M. J. J. Finken); Erasmus Medical Center-Sophia Children’s Hospital, University Medical Center Rotterdam (N. Weisglas-Kuperus, M. G. Keijzer-Veen, A. J. van der Heijden, J. B. van Goudoever); V. U. University Medical Center, Amsterdam (M. M. van Weissenbruch, A. Cranendonk, H. A. Delemarre-van der Waal, L. de Groot, J. F. Samsom); Wilhelmina Children’s Hospital, University Medical Center, Utrecht (L. S. de Vries, K. J. Rademaker, E. Moerman, M. Voogsgeerd); Máxima Medical Center, Veldhoven (M. J. K. de Kleine, P. Andriessen, C. C. M. Dillens-van Helvoort, I. Mohamed); Isala Clinics, Zwolle (H. L. M. van Straaten, W. Baerts, G. W. Veneklaas Slots-Kloosterboer, E. M. J. Tuller-Pikkemaat); Royal Effatha Guyot Group, Zoetermeer (M. H. Ens-Dokkum); and Association for Parents of Premature Babies (G. J. van Steenbrugge).

Received June 11, 2007. Accepted September 4, 2007.

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This specific part of the POPS-19 study was supported by grants from The Netherlands Organisation for Scientific Research (NWO) and the Center of Medical System Biology. The POPS-19 study was supported by grants from The Netherlands Organisation for Health Research and Development (ZonMw), Edgar Doncker Foundation, Foundation for Public Health Fundraising Campaigns, Phelps Foundation, Svartz-van Essen Foundation, Foundation for Children’s Welfare Stamps, TNO Quality of Life, Netherlands Organisation for Scientific Research (NWO), Dutch Kidney Foundation, Sophia Foundation for Medical Research, Stichting Astmabestrijding, Royal Effatha Guyot group.

Disclosure Summary: M. J. J. Finken has received consulting fees and lecture fees from Ferrin, Novo Nordisk, and Ipsen.

References

1. Van Rossum EF, Koper JW, Huizenga NA, Uitterlinden AG, Janssen JA, Brinkmann AO, Grootbee DE, de Jongh FH, van Duyn CM, Pols HA, Lamber
ts SW 2002 A polymorphism in the glucocorticoid receptor gene, which decreases sensitivity to glucocorticoids in vivo, is associated with low insulin sensitivity and hypertension in healthy young men. J Clin Endocrinol Metab 87:2227–2231

2. Van Rossum EF, Voorhoeve PG, te Velde SJ, Koper JW, Delemarre-van de Waal HA, Kemper HC, Lamberts SW 2004 The ER22/23EK polymorphism in the glucocorticoid receptor gene is associated with a beneficial body composition and muscle strength in young adults. J Clin Endocrinol Metab 90:4004–4010

3. Huizenga NA, Koper JW, de Lange P, Pols HA, Stolk RP, Burger H, Grootbee DE, Brinkmann AO, de Jongh FH, Lamberts SW 1998 A polymorphism in the glucocorticoid receptor gene may be associated with increased sensitivity to glucocorticoids in vivo. J Clin Endocrinol Metab 83:144–151

4. Dobson MG, Redfern CP, Unwin N, Weaver JU 2001 The N363S polymorphism of the glucocorticoid receptor: potential contribution to central obesity in men and lack of association with other risk factors for coronary heart disease and diabetes mellitus. J Clin Endocrinol Metab 86:2220–2224

5. Lin RC, Wang XL, Dalziel B, Caterson ID, Morris BJ 2003 Association of obesity, but not diabetes or hypertension, with glucocorticoid receptor N363S variant. Obes Res 11:640–643

6. Euser AM, Finken MJ, Keijzer-Veen MG, Hille ET, Wit JM, Dekker FW, Dutch POPS-19 Collaborative Study Group 2005 Associations between prenatal and infant weight gain and BMI, fat mass, and fat distribution in young adulthood: a prospective cohort study in males and females born very preterm. Am J Clin Nutr 81:480–487

7. Hofman PL, Regan F, Jackson WE, Jefferies C, Knight DB, Robinson EM, Cutfield WS 2004 Premature birth and later insulin resistance. N Engl J Med 351:2179–2186

8. Finken MJ, Keijzer-Veen MG, Dekker FW, Frölich M, Hille ET, Romijn JA, Wit JM, Dutch POPS-19 Collaborative Study Group 2006 Preterm birth and later insulin resistance: effects of birth weight and postnatal growth in a population-based longitudinal study from birth into adult life. Diabetologia 49:1641–1649

9. Hovi P, Andersson S, Eriksson JS, Jarvenpaa AL, Strang-Karlsson S, Makitie O, Kajantie E 2007 Glucose regulation in young adults with very low birth weight. N Engl J Med 358:2053–2063

10. Keijzer-Veen MG, Finken MJ, Nauta J, Dekker FW, Hille ET, Frölich M, Wit JM, van der Heijden AJ, Dutch POPS-19 Collaborative Study Group 2005 Is blood pressure increased 19 years after intrauterine growth restriction and preterm birth? A prospective follow-up study in the Netherlands. Pediatrics 116:725–732

11. Kist-van Holthe JE, van Zwieten PH, Schell-Feith EA, Zonderland HM, Holscher HC, Wolterbeek R, Veen S, Frolich M, van der Heijden BJ 2007 Is nephralocinosis in preterm neonates harmful for long-term blood pressure and renal function? Pediatrics 119:468–475

12. Szathmari M, Vasarhelyi B, Tulassay T 2001 Effect of low birth weight on adrenal steroids and carbohydrate metabolism in early adulthood. Horm Res 55:172–178

13. Verloove-Vanhorick SP, Verwey RA, Brand R, Gravenhorst JB, Verloove-Vanhorick SP, Wit JM 2005 Are age references for waist circumference, hip circumference and waist–hip ratio useful in Dutch children useful in clinical practice? Eur J Pediatr 164:216–222

14. Durnin JV, Rahaman MM 1967 The assessment of the amount of fat in the human body from measurements of skinfold thickness. Br J Nutr 21:681–689
20. Matthews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF, Turner RC 1985 Homeostasis model assessment: insulin resistance and β-cell function from fasting plasma glucose and insulin concentrations in man. Diabetologia 28:412–419

21. Gungor N, Saad R, Janosky J, Arslanian S 2004 Validation of surrogate estimates of insulin sensitivity and insulin secretion in children and adolescents. J Pediatr 144:47–55

22. Conwell LS, Trost SG, Brown WJ, Batch JA 2004 Indexes of insulin resistance and secretion in obese children and adolescents: a validation study. Diabetes Care 27:314–319

23. Van den Akker E, Nouwen JL, Melles DC, van Rossum EF, Koper JW, Uitterlinden AG, Hofman A, Verbrugh HA, Pols HA, Lamberts SW, van Belkum A 2006 Staphylococcus aureus nasal carriage is associated with glucocorticoid receptor polymorphisms. J Infect Dis 194:814–818

24. Kuningas M, Mooijaart SP, Slagboom PE, Westendorp RG, van Heemst D 2006 Genetic variants in the glucocorticoid receptor gene (NR3C1) and cardiovascular disease risk. The Leiden 85-plus Study. Biogerontology 7:231–238

25. Niklasson A, Ericson A, Fryer JG, Karlberg J, Lawrence C, Karlberg P 1991 An update of the Swedish reference standards for weight, length and head circumference at birth for given gestational age (1977–1981). Acta Paediatr Scand 80:756–762

26. Fredriks AM, van Buuren S, Burgmeijer RJ, Meulmeester JF, Breuker RJ, Brugman E, Roede MJ, Verloove-Vanhorick SP, Wit JM 2000 Continuing positive secular growth change in The Netherlands 1955–1997. Pediatr Res 47:316–323

27. Fredriks AM, van Buuren S, Wit JM, Verloove-Vanhorick SP 2000 Body index measurements in 1996–7 compared with 1980. Arch Dis Child 82:107–112

28. Perola M, Sammalisto S, Hiekkaninna T, Martin NG, Visscher PM, Montgomery GW, Benyamin B, Harris JR, Boomsma D, Willemsen G, Hottenga JJ, Christensen K, Kylvik KO, Sorensen TI, Pedersen NL, Magnusson PK, Spector TD, Widen E, Silventoinen K, Kaprio J, Palotie A, Pedersen L 2007 Combined genome scans for body stature in 6,602 European twins: evidence for common Caucasian loci. PLoS Genet 3:e97

29. Deng HW, Xu FH, Liu YZ, Shen H, Deng H, Huang QY, Liu YJ, Conway T, Li JL, Davies KM, Becker RR 2002 A whole-genome linkage scan suggests several genomic regions potentially containing QTLs underlying the variation of stature. Am J Med Genet 113:29–39

30. Wu X, Cooper RS, Boerwinkle E, Turner ST, Hunt S, Myers R, Olshen RA, Curf D, Zhu X, Kan D, Luke A 2003 Combined analysis of genomewide scans for adult height: results from the NHLBI Family Blood Pressure Program. Eur J Hum Genet 11:271–274

31. Meaney MJ, Aitken DH, Vieu V, Sharma S, Sarrieau A 1989 Neonatal handling alters adrenocortical negative feedback sensitivity and hippocampal type II glucocorticoid receptor binding in the rat. Neuroendocrinology 50:597–604

32. Liu D, Diorio J, Tannenbaum B, Caldji C, Francis D, Freedman A, Sharma S, Pearson D, Plotsky PM, Meaney MJ 1997 Maternal care, hippocampal glucocorticoid receptors, and hypothalamic-pituitary-adrenal responses to stress. Science 277:1659–1662

33. Weaver IC, Cervoni N, Champagne FA, D’Alessio AC, Sharma S, Seckl JR, Dymov S, Szyf M, Meaney MJ 2004 Epigenetic programming by maternal behavior. Nat Neurosci 7:847–854

34. Heijmans BT, Kremer D, Tobi EW, Boomsma DI, Slagboom PE 2007 Heritable rather than age-related environmental and stochastic factors dominate variation in DNA methylation of the human IGF2/H19 locus. Hum Mol Genet 647–654

35. Russcher H, Smit P, van den Akker EL, van Rossum EF, Brinkmann AO, de Jongh FH, Lamberts SW, Koper JW 2005 Two polymorphisms in the glucocorticoid receptor gene directly affect glucocorticoid-regulated gene expression. J Clin Endocrinol Metab 90:5804–5810

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