Soluble (Pro)Renin Receptor and Blood Pressure During Pregnancy: A Prospective Cohort Study

Noriyoshi Watanabe, Kanako Bokuda, Takeo Fujiwara, Tomo Suzuki, Asako Mito, Satoshi Morimoto, Seung Chik Jwa, Makiko Egawa, Yoshie Arai, Fumiaki Suzuki, Haruhiko Sago and Atsuhiro Ichihara

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Abstract—The renin–angiotensin system is believed to influence blood pressure (BP) during pregnancy, but the associations between BP during pregnancy and the soluble form of the (pro)renin receptor (s[P]RR), a new component of the tissue renin–angiotensin system, remain undetermined. In this prospective cohort study of 437 pregnant women with normal BP (systolic <140 mm Hg and diastolic <90 mm Hg) during early pregnancy (<16 weeks of gestation) regression analysis was performed to examine the associations between plasma s(P)RR concentrations and BP in 3 gestational stages (20–24, 28–32, and 36–40 weeks of gestation) and logistic regression analysis to evaluate the incidence of preeclampsia. Plasma s(P)RR concentrations at early, middle (16–28 weeks), and late pregnancy (>28 weeks) and at delivery averaged 29.7±10.0, 31.3±12.0, 39.2±8.9, and 40.4±10.2 ng/mL (means±SD), respectively. A 1-ng/mL increase in plasma s(P)RR concentration in early pregnancy predicted systolic/diastolic BP elevation in the later 3 gestational stages: 0.11 (95% CI, 0.014–0.20)/0.093 (0.027–0.16) mm Hg for 20 to 24 weeks, 0.11 (0.029–0.19)/0.088 (0.027–0.15) mm Hg for 28 to 32 weeks, and 0.16 (0.058–0.26)/0.12 (0.043–0.19) mm Hg for 36 to 40 weeks, respectively. Plasma s(P)RR concentrations in middle and late pregnancy were not associated with BP. Adjusted models revealed that women with plasma s(P)RR concentrations above the 75th percentile at delivery had a significantly increased risk of preeclampsia (odds ratio, 22.5 [95% CI, 1.8–279.9]). In conclusion, high circulating levels of s(P)RR at early pregnancy predicted a subsequent elevation in BP, and high concentrations at delivery were significantly associated with preeclampsia. (Hypertension. 2012;60:1250-1256.)

Key Words: blood pressure ■ hypertension ■ pregnancy ■ receptors ■ women
invasion and migration. Clinically during pregnancy, it is well-known that even women with no complication exhibit BP changes with gestational age; the BP is typically at its lowest between 24 and 26 gestational weeks, increasing thereafter until the end of the pregnancy. In addition, the circulating RAS is involved in the development of hypertensive disorders, including preeclampsia, during pregnancy. However, no evidence suggests that the tissue RAS contributes to BP changes during pregnancy.

On the basis of these background findings, the present study was conducted to examine whether the tissue RAS contributed to BP changes during pregnancy and the incidence of preeclampsia. To this end, we assessed the relationship between plasma s(P)RR concentrations and BP levels during pregnancy in a prospective cohort study.

Methods

Study Participants

In this prospective cohort study, Japanese pregnant women whose first visit to the National Center for Child Health and Development hospital was at <16 weeks and 0 days of gestation were enrolled between January and December 2010. At recruitment, written informed consent was obtained from all participants. The study protocol was approved by the ethics committee of the National Center for Child Health and Development (Tokyo, Japan).

The expected due date was confirmed by ultrasound in all participants. Inclusion criteria were systolic BP <140 mm Hg and diastolic BP <90 mm Hg at the time of enrollment (<16 weeks of gestation) and the absence of preexisting hypertensive disorders and renal disease. The study initially enrolled 477 pregnant women who met the criteria; however, 40 pregnant women were excluded because of early abortion (n=8) or the inability to follow-up because of relocation (n=32), resulting in a total of 437 study participants.

Plasma samples were obtained at 3 prenatal visits throughout the pregnancy and at time of delivery. The first sample was obtained before 16 weeks 0 days gestation (early pregnancy), the second between 16 weeks 0 days and 27 weeks 6 days gestation (mid-pregnancy), and the third after 28 weeks 0 days gestation (late pregnancy) at routine blood testing during prenatal visits. The fourth blood sample was obtained at time of delivery. We determined s(P)RR concentrations for first, second, and third trimester using the blood samples obtained in early, middle, and late pregnancy, respectively. We then analyzed s(P)RR concentrations in early pregnancy in association with BP values measured at 16 to 20, 20 to 24, 24 to 28, 28 to 32, 32 to 36, and 36 to 40 weeks of gestation (6 periods). Similarly, s(P)RR concentrations in mid-pregnancy were analyzed in association with BP levels measured at 28 to 32, 32 to 36, and 36 to 40 weeks gestation (3 periods) and s(P)RR concentrations in late pregnancy with BP levels measured at 36 to 40 weeks gestation (1 period). Finally, s(P)RR at delivery was used for analysis of the association between s(P)RR concentrations and the incidence of preeclampsia.

BP was measured in the sitting position with the right arm held at heart level after 5 minutes of rest using an automated sphygmomanometer (Omron BP-203RVIII Oscillometer; Nippon Colin, Tokyo, Japan) at enrollment and during every routine monthly prenatal check-up for study participants (a total of 7 measurements). Mean arterial pressure (MAP) was calculated by adding diastolic BP and one third of the pulse pressure. Plasma s(P)RR concentrations were determined for 437 participants during early pregnancy, 386 participants during mid-pregnancy, 364 participants during late pregnancy, and 384 participants at delivery. Clinical characteristics of study participants are shown in Table 1.

Procedure

Assays were performed by a person who was unaware of the outcome of the pregnancy. ELISAs for human s(P)RR were performed in duplicate, as described previously, with the use of commercial kits (Immuno-Biological Laboratories Co, Fujioka, Japan). The limit of detection was 24 pg/mL. Interassay and intrassay coefficients of variation were 7.5% and 5.5%, respectively.

Statistical Analysis

Paired t tests were used for comparisons of continuous variables (BP, plasma s(P)RR concentrations between each stage of pregnancy). To test the predictability of s(P)RR concentrations in early pregnancy for BP during pregnancy, the associations between s(P)RR in early pregnancy and BP at 20 to 24, 28 to 32, and 36 to 40 gestational weeks were determined using univariate and multivariate regression

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### Table 1. Baseline and Gestational Characteristics of Study Participants (n=437)

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>(No./total [%])</th>
<th>Mean (range)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, y</td>
<td>(No./total [%])</td>
<td>35.4 ± 4.2 (23–45)</td>
</tr>
<tr>
<td>Parity=0</td>
<td>(No./total [%])</td>
<td>235/437 (53.8)</td>
</tr>
<tr>
<td>Parity&gt;0</td>
<td>(No./total [%])</td>
<td>202/437 (46.2)</td>
</tr>
<tr>
<td>BMI before pregnancy</td>
<td>Mean (range)</td>
<td>20.5 ± 0.8 (15.2–38.9)</td>
</tr>
<tr>
<td>Smoking during pregnancy</td>
<td>(No./total [%])</td>
<td>10/437 (2.3)</td>
</tr>
<tr>
<td>Drinking during pregnancy</td>
<td>(No./total [%])</td>
<td>14/437 (3.2)</td>
</tr>
<tr>
<td>Twin pregnancy</td>
<td>(No./total [%])</td>
<td>18/437 (4.1)</td>
</tr>
<tr>
<td>Past history of pregnancy-induced hypertension</td>
<td>(No./total [%])</td>
<td>5/437 (1.1)</td>
</tr>
</tbody>
</table>

Family history

- Hypertension (No./total [%]) | 67/437 (15.3)
- Diabetes mellitus (No./total [%]) | 54/437 (12.4)
- Complicating disease
  - Diabetes mellitus (No./total [%]) | 1/437 (0.2)
  - Collagen disease (No./total [%]) | 7/437 (1.6)
  - Hyperthyroidism (No./total [%]) | 1/437 (0.2)
  - Hypothyroidism (No./total [%]) | 6/437 (1.4)
  - Asthma (No./total [%]) | 2/437 (0.5)
  - Antiphospholipid syndrome (No./total [%]) | 6/437 (1.4)
  - Epilepsy (No./total [%]) | 1/437 (0.2)
- Gestational age at early pregnancy blood sampling, wk | Mean (range) | 11.6 ± 0.6 (7.9–15.9)
- Gestational age at mid-pregnancy blood sampling, wk | Mean (range) | 24.7 ± 0.6 (19.9–27.7)
- Gestational age at late pregnancy blood sampling, wk | Mean (range) | 35.6 ± 0.8 (31.7–38.1)
- Gestational age at delivery, wk | Mean (range) | 39.0 ± 0.8 (26.4–41.9)

Gestational characteristics

- Fetal sex (male/female) (No.%)/No.% | 203 (46.5)/234 (53.5)
- Small for gestational age* (No./total [%]) | 37/437 (8.5)
- Gestational diabetes mellitus (No./total [%]) | 18/437 (4.1)
- Placenta previa (No./total [%]) | 6/437 (1.4)
- Preeclampsia† (No./total [%]) | 8/437 (1.8)

BMI indicates body mass index. Means are expressed as mean±SD.

* This was defined as an infant whose birth weight was below the 10th percentile.

† Preeclampsia was diagnosed by elevation of blood pressure >140/90 mm Hg and proteinuria (>300 mg/24 h or >1+ dipstick).
analysis. To avoid multiple comparisons of repeated measurements, we limited the 6 gestational stages of BP measurements into these 3 stages to analyze the associations between s(P)RR and BP. Similarly, the associations between s(P)RR in mid-pregnancy and BP at 28 to 32 and 36 to 40 gestational weeks and that between s(P)RR in late pregnancy and BP at 36 to 40 gestational weeks were also investigated. All multivariate models were adjusted for age, parity, body mass index before pregnancy, smoking, past history (diabetes mellitus), complicating diseases (collagen disease, hyperthyroidism, hypothyroidism, asthma, antiphospholipid syndrome, or epilepsy), family history (hypertension or diabetes mellitus), twin pregnancy, and each BP parameter at the time of s(P)RR measurement as confounders. For logistic regression analysis of the association between plasma s(P)RR concentration and the incidence of preeclampsia, s(P)RR concentration quartiles during each stage of pregnancy were used as cutoffs to define groups. All multivariate models were adjusted for age, parity, body mass index before pregnancy, family history (hypertension or diabetes mellitus), twin pregnancy, and MAP at enrollment. Sensitivity analysis was conducted by characterizing the association between s(P)RR and possible mediators, such as gestational characteristics (presence of gestational diabetes mellitus, fetal sex, small for gestational age, or placenta previa), and adding this to the above model. All analyses were conducted using STATA software (version 11.0; Stata Corporation, College Station, TX).

Results

Blood Pressure

Figure 1 shows a time course of systolic BP, diastolic BP, and MAP for each stage of pregnancy. Systolic BP at <16 weeks of gestation and every 4 weeks between 16 and 40 weeks of gestation (7 stages total) was 109.2 ± 10.2, 110.6 ± 10.9, and 113.4 ± 11.3 mm Hg (mean ± SD), respectively. There was a significant difference in MAP between before 16 weeks (P < 0.001), between 16 to 20 weeks of gestation (P = 0.009), between 28 to 32 and 36 to 40 weeks of gestation (P = 0.001), and between 32 to 36 and 40 weeks of gestation (P < 0.001).

Plasma s(P)RR Levels

All s(P)RR concentrations were above the level required for detection (ie, >24 pg/mL). Figure 2 shows the frequency histogram for plasma s(P)RR concentrations, which revealed a normal distribution for all 4 pregnancy periods. Plasma s(P)RR concentrations increased significantly stepwise from early pregnancy to delivery.

Association Between Plasma s(P)RR Level and BP

Table 2 shows the results of univariate and multivariate analyses for the association between plasma s(P)RR concentration and BP. The coefficient estimate shows BP elevation (in millimeters of mercury) per (in nanograms per milliliter) higher plasma s(P)RR concentration.

Unadjusted models showed significant associations between higher plasma s(P)RR concentrations in early pregnancy and

![Figure 1](http://hyper.ahajournals.org/)

**Figure 1.** Systolic and diastolic blood pressure (BP) and mean arterial pressure (MAP) levels during pregnancy. The diagram shows the distribution of systolic BP, diastolic BP, and MAP in each stage of pregnancy. **Closed circles** and **vertical bars** denote mean BP values and SDs, respectively; *P < 0.049, †P < 0.001, ‡P < 0.009, §P < 0.008.
increased plasma s(P)RR concentrations in early pregnancy, systolic and diastolic BPs, as well as MAP, were predicted to increase in the later gestational stages as follows: 0.11 (95% CI, 0.014–0.20), 0.093 (95% CI, 0.027–0.16), and 0.095 mm Hg (95% CI, 0.028–0.16 mm Hg) (s(P)RR) for 20 to 24 weeks; 0.11 (95% CI, 0.029–0.19), 0.088 (95% CI, 0.027–0.15), and 0.094 mm Hg (95% CI, 0.034–0.15 mm Hg) for 28 to 32 weeks; and 0.16 (95% CI, 0.058–0.26), 0.12 (95% CI, 0.043–0.19), and 0.13 mm Hg (95% CI, 0.052 to 0.21 mm Hg) for 36 to 40 weeks, respectively. However, mid-pregnancy and late pregnancy plasma s(P)RR concentrations were not predictive of elevated BP values in later stages of pregnancy.

Association Between Plasma s(P)RR Concentration and Preeclampsia

We computed odds ratios and 95% CIs for preeclampsia in the highest quartile of s(P)RR concentrations in each stage of pregnancy with respect to the lower 3 quartiles (Table 3). Overall incidence of preeclampsia was 1.8% (8 of 437) as shown in Table 1. There was a significant association between plasma s(P)RR levels at delivery and the incidence of preeclampsia (unadjusted odds ratio, 18.9 [95% CI, 2.2–158.7]; P=0.007). This association was also significant in multivariate models adjusted for age, parity, body mass index before pregnancy, family history (hypertension or diabetes mellitus), twin pregnancy, and MAP at enrollment (adjusted odds ratio, 22.5; [95% CI, 1.8–279.9]; P=0.016). The present study demonstrated 3 major findings regarding the association between s(P)RR and pregnancy. The first is the periodic stepwise increase of s(P)RR levels during pregnancy. The second is that increased plasma s(P)RR levels, especially in early pregnancy, were significantly predictive of elevated BP in later pregnancy. Finally, high s(P)RR levels at delivery were strongly associated with incidence of preeclampsia.

Although the present study found a stepwise elevation of s(P)RR levels during pregnancy, the source of increased plasma s(P)RR during pregnancy remains unclear. Nguyen et al.21 reported that (P)RR mRNA was strongly expressed in the brain, heart, and placenta. In the present study, the plasma s(P)RR concentrations were significantly higher in twin pregnancies compared with singleton pregnancies during late pregnancy and at delivery (50.9 ± 14.4 versus 38.7 ± 8.3 ng/mL, P<0.001; 54.7 ± 18.1 versus 39.8 ± 9.4 ng/mL [mean±SD], P<0.001, respectively), although there was no significant difference at early and mid-pregnancy. In addition, a previous study demonstrated that plasma s(P)RR concentrations of nonpregnant women averaged 22.8 ± 5.3 ng/mL (mean±SD), lower than shown in pregnant women in this study.13 These findings suggest that fetal and utero-placental components, including the placenta, may be major sources of s(P)RR in pregnant women and that plasma s(P)RR concentrations may increase with fetal and placental growth. Furthermore, although the components of the RAS in intrauterine tissues are reportedly altered by fetal sex during pregnancy,20 plasma s(P)RR concentrations at each gestational period did not vary significantly between women carrying a male versus a female...
The second finding, which is the most important part of the study, is that plasma s(P)RR levels were significantly associated with BP during pregnancy. In particular, increased plasma s(P)RR levels in early pregnancy (before 16 weeks and 0 days of gestation) predicted elevated BP values at all later stages of pregnancy. However, plasma s(P)RR levels at mid- and late pregnancy were not predictive of BP values in late pregnancy. Thus, a high plasma s(P)RR concentration in early pregnancy serves as a marker for elevated BP in later pregnancy.

The causal relationship between s(P)RR and BP could not be determined in the present study, but previous animal and human studies implied that (P)RR contributed to the development of hypertension. Overexpression of the human (P)RR gene in vascular smooth-muscle cells elevated BP and heart rate in rats. Human population studies showed that (P)RR gene polymorphism was associated with BP levels.

Table 2. Regression Analysis of the Associations Between Plasma s(P)RR Concentrations and Blood Pressure

<table>
<thead>
<tr>
<th>s(P)RR in early pregnancy</th>
<th>20–24 wk (n=410)</th>
<th>28–32 wk (n=426)</th>
<th>36–40 wk (n=401)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>CE</td>
<td>95% CI</td>
<td>P Value</td>
</tr>
<tr>
<td>Systolic BP</td>
<td>0.15</td>
<td>0.043 to 0.25</td>
<td>0.006</td>
</tr>
<tr>
<td>Diastolic BP</td>
<td>0.11</td>
<td>0.039 to 0.19</td>
<td>0.003</td>
</tr>
<tr>
<td>MAP</td>
<td>0.12</td>
<td>0.046 to 0.20</td>
<td>0.002</td>
</tr>
<tr>
<td>Adjusted*</td>
<td>Systolic BP</td>
<td>0.11</td>
<td>0.014 to 0.20</td>
</tr>
<tr>
<td>Diastolic BP</td>
<td>0.093</td>
<td>0.027 to 0.16</td>
<td>0.006</td>
</tr>
<tr>
<td>MAP</td>
<td>0.095</td>
<td>0.028 to 0.16</td>
<td>0.005</td>
</tr>
</tbody>
</table>

s(P)RR indicates soluble (pro)renin receptor concentration; CE, coefficient estimate; BP, blood pressure; MAP, mean arterial pressure. Early pregnancy, mid-pregnancy, and late pregnancy are defined as the periods before 16 wk 0 d of gestation, between 16 wk 0 d and 27 wk 6 d of gestation, and after 28 wk 0 d of gestation, respectively.

*All multivariate models were adjusted for age, parity, body mass index before pregnancy, smoking, past history (diabetes mellitus), complicating diseases (diabetes mellitus, collagen disease, hyperthyroidism, hypothyroidism, asthma, antiphospholipid syndrome, or epilepsy), family history (hypertension or diabetes mellitus), twin pregnancy, and each blood pressure parameter at the time of prorenin measurement.
increased plasma s(P)RR concentrations in early pregnancy was unclear in the present study, we speculate that increased levels of both prorenin and (P)RR in early pregnancy may contribute to BP elevation in later stages of pregnancy. (P)RR is essential for embryonic and fetal development, and plasma prorenin concentrations are increased during pregnancy, especially in early pregnancy. Taken together, the interaction between (P)RR and prorenin may be necessary for embryonic and fetal development. Prorenin may trigger an elevation of (P)RR levels given that the amount of (P)RR is for embryonic and fetal development. Prorenin may trigger an elevation in later pregnancy. We speculate that increased s(P)RR levels during middle and late pregnancy predicted BP elevation by s(P)RR in middle and late pregnancy. In conclusion, increased circulating levels of s(P)RR were associated with elevated BP during pregnancy, suggesting that plasma s(P)RR concentration in early pregnancy is a useful marker for predicting BP elevation during late pregnancy. However, although increased s(P)RR levels at delivery were strongly associated with the incidence of preeclampsia, whether plasma s(P)RR levels predict preeclampsia remains undetermined. Future studies, including a larger sample size, are needed to clarify the association between plasma s(P)RR and preeclampsia.

**Perspectives**

Increased circulating levels of s(P)RR were associated with elevated BP during pregnancy, suggesting that plasma s(P)RR concentration in early pregnancy is a useful marker for predicting BP elevation during late pregnancy. Because s(P)RR serves as a biomarker reflecting the tissue RAS status, our result indicates that the tissue RAS may be significantly associated with BP changes during pregnancy. However, although increased s(P)RR levels at delivery were strongly associated with the incidence of preeclampsia, whether plasma s(P)RR levels predict preeclampsia remains undetermined. Future studies, including a larger sample size, are needed to clarify the association between plasma s(P)RR and preeclampsia.

**Acknowledgment**

We thank Dr Nobuhiro Maruyama for his technical support of the s(P)RR assay.
The renin–angiotensin system is believed to influence blood pressure during pregnancy, but the associations between blood pressure during pregnancy and soluble (pro)renin receptor (s(P)RR), a new component of the tissue renin–angiotensin system, remain undetermined.

The present study showed that s(P)RR in early pregnancy was predictive of elevated blood pressure during late pregnancy and that increased s(P)RR levels at delivery were strongly associated with the incidence of preeclampsia.

References