RENIN ANGIOTENSIN ALDOSTERONE SYSTEM, URINARY PROSTAGLANDINS AND KALLIKREIN IN PREGNANCY INDUCED HYPERTENSION
EVIDENCE FOR A DISREGULATION OF THE RENIN-ANGIOTENSIN-PROSTACYCLIN LOOP

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Summary
Plasma renin activity and aldosterone concentrations were measured simultaneously with urinary excretion of kallikrein and four prostaglandins (PGE₂, PGF₂α, 6 keto PGF₁α and TXB₂) in 23 patients with pregnancy induced hypertension (17 with permanent PIH and six with labile PIH, since in these latter their hypertension was controlled only by home bed rest) and in 16 normotensive pregnant women at the same stage of gestation (31 ± 3 weeks). PRA was lower in permanent PIH than in controls and in labile PIH. No difference between the three groups was observed for plasma aldosterone and the urinary excretion of kallikrein and of the prostaglandins except that TXB₂ was higher in labile PIH than in permanent PIH. Correlation studies of kallikrein disclosed correlations with most prostaglandin excretions, explained by the physiological stimulation of phospholipase A₂ by kallidin. Correlation studies of PRA disclosed unexpected negative correlation with PGE₂ and 6 keto PGF₁α in the permanent PIH group.

In conclusion, labile PIH has a different biological profile than permanent PIH since they have higher PRA and higher TXB₂ excretion, an association which suggests a more pronounced ureteral compression by the gravid uterus in this group. Permanent PIH has a disregulation of the renin angiotensin-prostacyclin loop since PRA and 6 keto PGF₁ are negatively correlated. This suggests the role of an independent vasopressive substance which would stimulate PGI₂ and suppress renin secretion.

Introduction
In our previous studies, pregnancy induced hypertension not responding to bed rest (permanent PIH) was shown to be characterised by a hypovolaemic
state suggesting vasoconstriction, and an insufficient stimulation of the renin angiotensin aldosterone system contrasting with a more easily reactive adrenergic system. This finding could be compatible with a deficiency of production of the vasodilating prostaglandin (PGI₂) which has been shown to stimulate renin secretion and inhibit the release of noradrenaline [1] and which is more likely than PGE₂, a circulating hormone, since unlike PGE₂ it is not destroyed in the lungs. A decreased production of PGI₂ by the umbilical arteries of pre-eclamptic women has been reported by many authors and even correlated to decreased uteroplacental blood flow [2]. However these studies are in vitro studies performed after delivery and are therefore not relevant to the understanding of the mechanisms occurring at the onset of PIH. To grasp these mechanisms in vivo PGI₂ must be measured.

Methodological problems preclude adequate determination of decreased plasma PGI₂, therefore only the urinary excretion of its stable metabolites can be performed with reasonable assumption that they reflect plasma values [3]. Six women with PIH showed a 50 per cent decrease of 2–3 dinor 6 keto PGF₁α urinary excretion [4] without concomitant evaluation of the renin angiotensin aldosterone system, nor of the vasoconstrictive and pro-aggregative prostaglandin TXA₂. Evaluations of prostaglandin E₂, with natriuretic properties, have been performed more extensively in PIH by measurement of its urinary excretion, which reflects however only renal production. The results have been conflicting, with excretion in PIH being found to be either increased [5] or decreased [6].

Kallikrein excretion has been studied only once in PIH and found to be decreased compared to normal pregnant women [7]. However no simultaneous study has been performed of kallikrein and prostaglandin in spite of the known physiological link between them as kallidin (the product of the action of kallikrein on kininogen) is a physiological stimulus to phospholipase A₂ [3]. Because of these scarce and conflicting data, we have studied simultaneously in PIH the renin angiotensin system and the urinary excretion of kallikrein and of four prostaglandins (6 keto PGF₁α, PGE₂, PGF₂α and thromboxane B₂ the stable metabolite of thromboxane A₂).

Patients and methods

Patients

Twenty-three patients with PIH have been studied. PIH was defined by finding, after the twentieth week of pregnancy, a blood pressure higher than 140/90 mmHg at the obstetrical outpatient clinic, and the absence of hypertension before the twentieth week and three months after delivery. At the end of pregnancy two types of PIH have been distinguished according to their blood pressure measured at the nephrological clinic after lying supine for 30 minutes while home bed rest (4 hours lying in the left lateral position during the day time) has been prescribed throughout the pregnancy: labile hypertension (LH) when the patients were always found normotensive at the nephrological outpatient
clinical (6 patients): permanent hypertension (PH) when the patients were hypertensive at least once at the nephrological outpatient clinic (17 patients).

These two groups were compared with a control group (C) of 16 pregnant women normotensive before, during and after pregnancy.

Methods

Protocol  Studies have been performed as soon as possible after the discovery of hypertension in ambulatory patients taking no drugs and on a normal salt diet (confirmed by 24 hr sodium excretion). Blood pressure was measured after thirty minutes of supine rest and they were weighed. Blood samples were taken through a catheter inserted 30 minutes previously. Samples for PRA and aldosterone were put into ice, centrifuged immediately at 4°C, then stored at -30°C. All measurements were made within three months.

Analytical methods  Plasma renin activity (PRA) and plasma aldosterone (PA) were estimated by radioimmunoassay technique [1]. Urinary prostaglandins, PGE₂, PGF₂α, 6 keto PGF₁α, TXB₂ were measured by radioimmunoassay after prior extraction and separation by chromatography on silicic acid [8]. Urinary kallikrein was measured by esterolytic activity on the synthetic substrate BAEE [9].

Statistical methods  Comparison of the groups were made using the Wilcoxon's test for non paired data. Link between two parameters was evaluated first by linear regression analysis and then by covariance analysis.

Results

Groups comparison

There was no difference between the three groups for age, parity ratio, duration of pregnancy (30.4, 31.5 and 31.2 weeks in C, PH and LH) and weight gain during pregnancy at the discovery of hypertension and for sodium excretion (respectively 12.2, 12.2 and 10.2mmol/mmol creatinine). The weight was higher in PH than in C (respectively 73 ± 3.8, 54.1 ± 1.1kg, p<0.01) but not different in LH (66.7 ± 6.8kg). At the time of first attendance at the nephrological clinic the mean arterial pressure (MP) was higher in permanent PIH than in C (99.5 ± 3.8 versus 82.4 ± 1.4mmHg, p<0.01) but was comparable in labile PIH and in C (89 ± 3.1mmHg). However at the discovery of hypertension at the obstetrical clinic MAP was higher in permanent and labile PIH than in C (respectively 116.1 ± 2.2, 111.6 ± 1.8 and 82.4 ± 1.4mmHg, p<0.01).

The mean (±SEM) values of the various biological parameters observed in the three groups at the first exploration after discovery of hypertension are summarised in Table I. PRA was significantly lower in PH than in C and LH. PRA was significantly higher in LH than in C and PH. Plasma aldosterone levels were not significantly different in the three groups. Urinary kallikrein was not significantly different in the two hypertensive groups and the control group. Urinary
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<th>Plasma</th>
<th>Urinary excretion/mmol creatinine</th>
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<td>Renin activity ng/ml/hr</td>
<td>Aldosterone pg/ml</td>
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<td>Controls (C) n = 16</td>
<td>6.9 ± 0.6</td>
<td>246.6 ± 34.1</td>
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<td>Permanent hypertension (PH) n = 17</td>
<td>4.9* ± 0.3††</td>
<td>218 ± 53.7</td>
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<td>Labile hypertension n = 6</td>
<td>12.2† ± 0.9</td>
<td>271 ± 63.2</td>
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Significance of comparisons: permanent hypertension versus control: *p<0.01
labile hypertension versus control: †p<0.01
permanent versus labile hypertension **p<0.05
††p<0.01
excretion of prostaglandins were not significantly different in the two hypertensive groups except for \( \text{TXB}_2 \) which is significantly greater in labile than in permanent PIH. No difference between the hypertensive groups and the controls was observed. The ratio \( \text{PGE}_2 : \text{PGF}_{2\alpha} \) was no different in the three groups being respectively 1.47, 1.6 and 1.5 in control, permanent and labile PIH.

**Correlation studies**

*PRA and urinary prostaglandins*  In the control group, the only significant linear correlation found was between PRA and \( \text{PGF}_{2\alpha} \) \( (r = 0.85, n = 15, p<0.001) \). In labile hypertension group, no correlation was found. In the permanent hypertensive group, \( (n = 17) \) a negative linear correlation was found with \( \text{PGE}_2 \) \( (r = -0.51, p<0.05) \) and 6 keto \( \text{PGF}_{1\alpha} \) \( (r = -0.62, p<0.01) \). Two way covariance analysis confirmed these negative correlations with a \( D_c \) of respectively 0.72 \( (f<0.01) \) and 0.83 \( (f<0.002) \). (Figure 1).

![Figure 1](image-url)
**Kallikrein and urinary prostaglandins** In the labile group, no correlation was found. In the permanent hypertensive group, a positive linear correlation was found with PGF$_{2\alpha}$ ($r = 0.55$, $p<0.05$), 6 keto PGF$_{1\alpha}$ ($r = 0.76$, $p<0.001$) and thromboxane B$_2$ ($r = 0.66$, $p<0.001$). These correlations were confirmed by covariance analysis for PGF$_{2\alpha}$ ($Dc = 0.39$, $f<0.03$) and 6 keto PGF$_{1\alpha}$ ($Dc = 0.72$, $f<0.001$) but not for thromboxane B$_2$. Covariance analysis unmasked a positive correlation between kallikrein and PGE$_2$ ($Dc = 0.47$, $f<0.01$).

**Discussion**

Our data in women with PIH soon after the discovery of hypertension at 25–35 weeks gestation, do not show significant differences with normtensive pregnant women as regards the urinary excretion of kallikrein and of the four prostaglandins PGE$_2$, PGF$_{2\alpha}$, 6 keto PGF$_{1\alpha}$ and TXB$_2$. As regards PGE$_2$, the main difference with the population of Pedersen, who found a decreased excretion of PGE$_2$, was the fact that most of his patients had proteinuria and a sodium retaining state since urinary sodium excretion was lower in his pre-eclamptic women than in the controls [6]. As regards 6 keto PGF$_{1\alpha}$, our results differ from those of Goodman who measured another metabolite, the 2–3 dinor 6 keto PGF$_{1\alpha}$, by gas chromatography-mass spectrometry, which is considered as a better index of circulating PG1$_2$ than the 6 keto PGF$_{1\alpha}$ [3]. Furthermore, his women were studied later in pregnancy (36 weeks) and no indication is given on their sodium diet compared to controls.

Since we had previously found that pregnancy induced hypertension could be differentiated into permanent hypertension with lower PRA and a more easily reactive adrenergic system, and labile hypertension with stimulated PRA and normally reactive adrenergic system, we differentiated our patients with PIH according to the response of their blood pressure to home bed rest in order to interpret their urinary excretion of kallikrein and prostaglandins. No difference was found between the two hypertensive groups except for the excretion of TXB$_2$ which has been found paradoxically higher in the labile hypertensive group. Since one cause of increased TXB$_2$ and renin secretion is ureteral obstruction [3], we plan to look for more severe ureteral compression by the gravid uterus in this group of patients by ultrasonography.

Correlation studies of kallikrein disclosed physiological correlations with the urinary excretion of most prostaglandins which is in accordance with the known fact that kallidin is a normal stimulus of phospholipase A$_2$ which liberates arachidonic acid from the membrane phospholipids. Correlation studies of PRA disclosed an unexpected negative correlation in the permanent hypertension group with urinary PGE$_2$ and 6 keto PGF$_{1\alpha}$ whereas a physiological correlation was found in the control group between PRA and urinary PGF$_{2\alpha}$.

This negative correlation between PRA and 6 keto PGF$_{1\alpha}$ in the permanent hypertension group is quite surprising since the physiological modulation of the vascular tone (at least in the kidney) is the result of the balance between the angiotensin II induced vasoconstriction and the prostacyclin induced vasodilatation and that this balance is achieved by the following loop [3]:

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This loop implies a positive correlation between PRA and prostacyclin metabolites.

The negative correlation between PRA and 6 keto PGF_1α shows that there is a disregulation of this loop. This could be explained by the independent production of a vasoconstrictive factor which like angiotensin II would suppress renin secretion and stimulate prostacyclin synthesis. Since this latter is actually normal in our patients an inappropriate synthesis of prostacyclin in response to this stimulus is also implied. The negative correlation found by Symmonds et al [9] between renin concentration and plasma angiotensin II in PIH is compatible with this hypothesis.

In conclusion our study shows that the hypovolaemic vasoconstrictive hypertensive state of PIH is not associated with abnormal excretion of kallikrein and prostaglandins but is associated with a disregulation of the renin-angiotensin-prostacyclin loop implying an inappropriate synthesis of prostacyclin which is not stimulated in response to a hypothetical vasoconstrictive factor with renin suppressive effect.

References

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