Impact of the Preintervention Rate of Renal Function Decline on Outcome of Renoprotective Intervention

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Background and objectives: Randomized clinical trials on progression of renal diseases usually include patients according to criteria for BP, renal function, and proteinuria. There are no data showing that this provides groups with similar baseline rates of renal function loss. Accordingly, the impact of preintervention rate of renal function loss (slope) on outcome of studies has not been established.

Design, setting, participants, & measurements: Preintervention slope was established in 60 of 89 renal patients without diabetes in whom a 4-yr prospective, randomized intervention had been performed (enalapril versus atenolol), and whether (1) preintervention slope was distributed equally over the groups; (2) treatment benefit, defined as slope improvement, corresponded to study outcome; and (3) preintervention slope was a determinant of intervention slope were analyzed.

Results: The preintervention slope was different in the groups: $-3.7 \pm 3.2$ ml/min per yr in the group to receive enalapril versus $-2.2 \pm 3.3$ ml/min per yr in the group to receive atenolol. The intervention slopes were similar: $-1.9 \pm 0.8$ enalapril and $-1.8 \pm 0.7$ ml/min per yr atenolol. Accordingly, slope improved during enalapril only. When analyzed by angiotensin-converting enzyme (I/D) genotype, slope improvement was found only in DD genotype. On multivariate analysis, the preintervention slope was a main predictor of the intervention slope.

Conclusions: Differences in preintervention slope are relevant to outcome of trials and can induce bias. For future studies, allocation according to preintervention slope, although time-consuming, may be useful to allow conduction of more valid studies in a smaller number of patients.

In chronic renal disease, renal function usually deteriorates progressively toward end-stage renal failure. Major effort was made in the past decade to develop and evaluate renoprotective strategies by long-term intervention studies in humans (1–7). Whereas the rate of renal function loss is usually fairly constant in time for individual patients, the degree of loss varies greatly between patients (8). Theoretically, for studies on progressive renal function loss, it would be best to include or randomly assign patients according to their previous rate of renal function loss (preintervention slope). This could ensure an equal allocation of patients with high or low risk for progression to different treatment groups. Not surprising, such an approach has not been applied for comparative parallel trials, because of obvious practical problems in obtaining data on the rate of renal function loss before intervention; therefore, patients are usually included according to baseline cross-sectional parameters, such as renal function, BP, and proteinuria, as indirect indicators of renal prognosis.

We hypothesized that the previous rate of renal function loss might be the main determinant of progression during intervention and that, accordingly, neglect of this information could bias the interpretation of the results of intervention. To test this hypothesis, we collected data on preintervention rate of renal function loss for individuals who had participated in a 4-yr double-blind, randomized, intervention trial. Previously, two analyses were performed in this data set: (1) A double-blind, randomized comparison of enalapril versus atenolol, which revealed no differences between the regimens on i-ithalamate measured GFR slope (9); and (2) a post hoc analysis on the effect of angiotensin-converting enzyme (ACE) genotype that showed faster progression rate in the DD patients (10).

In this analysis, we evaluated, first, whether preintervention slope was distributed equally over the groups. Second, we reevaluated differences in treatment benefit, defined as slope improvement, in the comparison between enalapril and atenolol and the comparison between the genotypes, respectively. Finally, we evaluated whether preintervention slope was a determinant of the slope during intervention.
Concise Methods

Patients

In the previously published report, 89 patients with nondiabetic renal insufficiency were studied according to a prospective, parallel, randomized, double-blind design (10). In short, 4 wk after withdrawal of antihypertensive drugs, baseline measurements were obtained. Inclusion criteria were a creatinine clearance of 30 to 90 ml/min, 18 to 65 yr of age, and a diastolic BP of >80 mmHg. Patients were stratified according to baseline creatinine clearance (either 30 to 60 or 60 to 90 ml/min) and afterward randomly assigned to treatment with atenolol or enalapril (starting dosage 50 or 10 mg, respectively). Drug dosage was titrated on a goal diastolic BP of 10 mmHg below baseline and/or <95 mmHg. Patients were followed for 3 to 4 yr (mean 189 wk).

At each visit, BP was measured (during baseline and during the intervention period by automated device, Dinamap; GE Medical Systems, Milwaukee, WI), and 24-h urine was collected for determination of proteinuria (by pyrogallol red molybdate method) and creatinine. Blood was drawn for creatinine measurement (by standard autoanalyzer, SMA-C, Technicon, Tarrytown, NY) to calculate creatinine clearance. Values were corrected for body surface area (1.73 m²). ACE genotype was determined by PCR method using two different specific insertion primers to confirm putative DD genotypes and prevent mistyping, as described previously (11).

For this analysis, data on the period before study entry (during regular patient care) were retrospectively collected from patient records. Patients were included when at least 1 yr of follow-up of the preintervention period was available, with three or more data points for each parameter. ACE genotype was obtained for 81 patients. A total of 60 patients could be included in this analysis.

Statistical Analyses

Data on creatinine clearance and BP are given as means ± SD and for proteinuria as median and interquartile range during retrospective and prospective follow-up. The BP and antiproteinuric responses were analyzed as the change from baseline, that is, 4 wk after withdrawal of previous treatment. The baseline characteristics of the original and this study group were compared by χ² test (gender, age, ACE genotype, treatment, and type of disease) and nonparametric Mann-Whitney U test (continuous variables). Differences between atenolol/enalapril and ACE genotype groups were tested for slope, BP, and proteinuria by nonparametric ANOVA test. Mann-Whitney test was used to detect differences between the subgroups. Progression of renal function loss was estimated for each individual by calculation of the slope of creatinine clearance over time by the least squares regression method. The slope of renal function during the prospective study period is presented as the mean arterial pressure (MAP) and proteinuria. During intervention, in accordance with our previous report, MAP and proteinuria were similar during enalapril and atenolol (9). During the preintervention period, MAP and proteinuria were similar in the enalapril and atenolol groups. The rate of renal function loss (slope) was as well similar during intervention (Figure 1A); however, the intervention rate of renal function loss was higher in the enalapril group (P = 0.05). As a consequence, when the preintervention slopes were compared with the intervention slopes, the enalapril group had a significant improvement of slope (P = 0.018), which was absent in the atenolol group.

In the corresponding data for a break-up according to ACE genotype (Table 2), baseline creatinine clearance was higher in the II genotype, with similar MAP and proteinuria for the three genotypes. During intervention, in accordance with our previous report, MAP and proteinuria were similar for the genotypes, with a higher rate of renal function loss in DD homozygotes (Figure 1B) (9). Remarkably, preintervention rate of renal function loss was significantly higher in the DD genotype than in the other genotype groups. Consequently, in the DD genotype, a significant benefit of intervention was apparent from the improvement in rate of renal function loss during intervention (P = 0.0012; Figure 1). No such improvement was found in the other genotypes.

On univariate analysis, the preintervention slope was correlated to the intervention slope (R² = 0.13, P = 0.005). Baseline proteinuria was negatively correlated to the intervention slope (R² = −0.12, P = 0.008). Baseline creatinine clearance and MAP did not correlate to the intervention slope. On multivariate analysis, the preintervention slope and preintervention protein-
Table 1. Patient characteristics before start of the intervention study (after withdrawal of previous medication)a

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Original Study (n = 81)</th>
<th>This Analysis (n = 60)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male/female</td>
<td>49/32</td>
<td>35/25</td>
</tr>
<tr>
<td>Age (yr)</td>
<td>49 ± 14</td>
<td>49 ± 12</td>
</tr>
<tr>
<td>Creatinine clearance (ml/min; mean ± SD)</td>
<td>55.1 ± 23.4</td>
<td>56.3 ± 23.7</td>
</tr>
<tr>
<td>MAP (mmHg; mean ± SD)</td>
<td>110 ± 14</td>
<td>108 ± 9</td>
</tr>
<tr>
<td>Proteinuria (g/d; median [IQR])</td>
<td>0.6 (0.0 to 2.5)</td>
<td>0.5 (0.0 to 2.4)</td>
</tr>
<tr>
<td>Sodium excretion (mmol/d; mean ± SD)</td>
<td>126 ± 17</td>
<td>127 ± 13</td>
</tr>
<tr>
<td>DD/ID/II (n)</td>
<td>17/37/27</td>
<td>15/24/21</td>
</tr>
</tbody>
</table>

Previously taking (n)
- antihypertensives
- diuretics
- β blockers
- ACE inhibitors
- miscellaneous

Comorbid condition (n)
- glomerulosclerosis/hypertension
- IgA nephropathy
- urolithiasis/reflux
- PKD
- miscellaneous

aData are given for the original study and for the patients in the present analysis. ACE, angiotensin-converting enzyme; IQR, interquartile range; MAP, mean arterial pressure; PKD, polycystic kidney disease.

Table 2. BP and proteinuria during the preintervention and intervention periodsa

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Enalapril (n = 31)</th>
<th>Atenolol (n = 29)</th>
<th>DD (n = 15)</th>
<th>ID (n = 24)</th>
<th>II (n = 21)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (yr; mean ± SD)</td>
<td>50 ± 14</td>
<td>48 ± 11</td>
<td>49 ± 15</td>
<td>49 ± 12</td>
<td>49 ± 12</td>
</tr>
<tr>
<td>Gender (male/female)</td>
<td>21/10</td>
<td>15/14</td>
<td>9/6</td>
<td>13/11</td>
<td>11/10</td>
</tr>
<tr>
<td>Genotype (DD/ID/II)</td>
<td>9/11/11</td>
<td>6/13/10</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>MAP (mmHg; mean ± SD)</td>
<td>108 ± 8</td>
<td>107 ± 10</td>
<td>110 ± 11</td>
<td>106 ± 9</td>
<td>108 ± 7</td>
</tr>
<tr>
<td>proteinuria (g/d; median [IQR])</td>
<td>0.2 (0.0 to 1.6)</td>
<td>0.1 (0.0 to 1.1)</td>
<td>0.2 (0.0 to 2.3)</td>
<td>0.1 (0.0 to 1.8)</td>
<td>0.0 (0.0 to 1.1)</td>
</tr>
<tr>
<td>creatinine clearance (ml/min; mean ± SD)</td>
<td>57.8 ± 28.6</td>
<td>56.9 ± 21.4</td>
<td>45.9 ± 23.5</td>
<td>50.1 ± 15.7</td>
<td>71.8 ± 24.3b</td>
</tr>
<tr>
<td>MAP (mmHg; mean ± SD)</td>
<td>108 ± 8</td>
<td>111 ± 16</td>
<td>113 ± 10</td>
<td>106 ± 13</td>
<td>112 ± 12</td>
</tr>
<tr>
<td>proteinuria (g/d; median [IQR])</td>
<td>0.4 (0.0 to 2.2)</td>
<td>0.7 (0.0 to 3.8)</td>
<td>1.1 (0.0 to 3.6)</td>
<td>0.1 (0.0 to 2.3)</td>
<td>0.3 (0.0 to 2.2)</td>
</tr>
<tr>
<td>proteinuria (g/d; median [IQR])</td>
<td>94 ± 3c</td>
<td>96 ± 5c</td>
<td>98 ± 5c</td>
<td>94 ± 5c</td>
<td>95 ± 4c</td>
</tr>
</tbody>
</table>

aData are given for the original study and for the patients in the present analysis. ACE, angiotensin-converting enzyme; IQR, interquartile range; MAP, mean arterial pressure; PKD, polycystic kidney disease.

uria both were independent predictors of intervention slope (R² of model = 0.24, P < 0.001; Table 3). ACE genotype contributed significantly to a model including proteinuria, with faster progression rate in DD genotype (R² model = 0.21; standardized B: genotype −1.342 and proteinuria −0.510; both P < 0.05); however, when preintervention slope was included, the contribution of ACE genotype was no longer significant.

The predictive value of baseline proteinuria for an intervention slope in the highest quartile was significant, shown by an area under the ROC of 0.65 ± 0.08 (P = 0.04 versus reference line; Figure 2). The predictive value of the preintervention slope was somewhat better (0.73 ± 0.07; P = 0.008 versus reference line). When the preintervention slope and proteinuria were combined, the predictive value for rapid progression was the highest, as shown by an area of 0.75 ± 0.07 (P = 0.005 versus reference line).

The Bland-Altman plot comparing the preintervention slope with the intervention slope showed clearly a horizontal scatter.
and not correlation between the initial preintervention value and the subsequent change ($R^2/\text{H11005} = 0.09$, $P/\text{H11021} = 0.15$; Figure 3), indicating that the slowing of GFR decline in the enalapril group cannot be explained by regression to the mean. This analysis was performed separately for the atenolol group and genotype groups and showed no correlation either (data not shown).

**Discussion**

This study shows that stratification over different therapy arms according to baseline creatinine does not guarantee equal distribution of previous rate of renal function loss and that this may be relevant to study outcome. First, we found a difference in preintervention slope between the randomly assigned groups. Second, the therapeutic benefit in terms of slope improvement did not correspond on a one-to-one basis with the therapeutic outcome based on between-group comparison during treatment only. Finally, the preintervention slope was an independent determinant of intervention slope, and consideration of preintervention slope improved the prediction of progression during the study.

We retrospectively investigated whether the stratification according to baseline renal function resulted in an equal distribution of renal function loss before intervention. The preintervention slope was drawn by taking baseline renal function as reference and calculating renal function loss backward. The intervention slope is calculated from 3 mo on treatment to end of follow-up. A significant improvement in slope was present in the enalapril group, which was absent in the atenolol group. A significant benefit of intervention was apparent from the improvement in rate of renal function loss only in the DD genotype. No such improvement was found in the other genotypes.

**Table 3. Regression analysis of relation between proteinuria and preintervention slope and intervention slope**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Standardized $\beta$</th>
<th>$P$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Proteinuria</td>
<td>-0.356</td>
<td>0.004</td>
</tr>
<tr>
<td>Preintervention slope</td>
<td>0.355</td>
<td>0.004</td>
</tr>
</tbody>
</table>

$^aR^2$ of model = 0.24, $P < 0.001$. 

and not correlation between the initial preintervention value and the subsequent change ($R^2 = 0.09$, $P = 0.15$; Figure 3), indicating that the slowing of GFR decline in the enalapril group cannot be explained by regression to the mean. This analysis was performed separately for the atenolol group and genotype groups and showed no correlation either (data not shown).

**Figure 1.** Creatinine clearance slopes (ml/min per 1.73 m$^2$/yr) for atenolol and enalapril (A; $^aP < 0.05$ versus atenolol; $^bP = 0.018$ versus preintervention period) and for the genotypes (B; $^aP < 0.05$ versus other genotypes; $^bP = 0.001$ versus preintervention period) separately during the preintervention and intervention periods. The preintervention slope is drawn by taking baseline renal function as reference and calculating renal function loss backward. The intervention slope is calculated from 3 mo on treatment to end of follow-up. A significant improvement in slope was present in the enalapril group, which was absent in the atenolol group. A significant benefit of intervention was apparent from the improvement in rate of renal function loss only in the DD genotype.

<table>
<thead>
<tr>
<th>Preintervention</th>
<th>Intervention</th>
</tr>
</thead>
<tbody>
<tr>
<td>Slope</td>
<td>Slope</td>
</tr>
<tr>
<td>Atenolol</td>
<td>$-2.2 \pm 3.3$</td>
</tr>
<tr>
<td>Enalapril</td>
<td>$-3.7 \pm 3.2^a$</td>
</tr>
</tbody>
</table>

$^aP < 0.05$ compared to atenolol
$^bP = 0.018$ compared to pre-intervention period

<table>
<thead>
<tr>
<th>Pre-intervention</th>
<th>Intervention</th>
</tr>
</thead>
<tbody>
<tr>
<td>Slope</td>
<td>Slope</td>
</tr>
<tr>
<td>II</td>
<td>$-2.1 \pm 2.6$</td>
</tr>
<tr>
<td>ID</td>
<td>$-1.8 \pm 2.5$</td>
</tr>
<tr>
<td>DD</td>
<td>$-6.1 \pm 3.6^a$</td>
</tr>
</tbody>
</table>

$^aP < 0.05$ compared to other genotypes
$^bP = 0.001$ compared to pre-intervention period
proteinuria that was calculated to affect its outcome (12). Our data suggest that inclusion of data on preintervention rate of renal function decline may serve to obtain groups with a comparable renal risk during intervention, thereby increasing the validity of the study.

The impact of previous rate of renal function decline was even more remarkable for the analysis of the effect of ACE genotype on study outcome (9). Assessed from parallel intervention data only, the steeper slope in the DD genotype during intervention suggests lack of renoprotective benefit in these patients. The preintervention data revealed, remarkably, that slope improvement was most pronounced in DD homozygotes, which sheds a different light on the nature of their responsiveness to therapy. This treatment benefit in DD patients is in accordance with other studies (13–15) and, according to our data, may be explained by their steeper preintervention slope rather than by ACE genotype as such, because slope improvement closely correlated with the preintervention rate of renal function decline independent of genotype. Several studies have reported on the slope in DD genotype, with conflicting results (9,13–19). Recent data from the Reduction of Endpoints in NIDDM with the Angiotensin II Antagonist Losartan (RENAAL) study, published in abstract form, show that patients with type 2 diabetes, proteinuric, and the D allele of the ACE gene have an unfavorable renal prognosis that can be mitigated by losartan treatment (20). The RENAAL data are in line with our results that show a steeper preintervention slope in patients with the DD genotype along with better risk reduction during intervention. The parallel with our data is remarkable, and, taken together, these data suggest that optimal treatment is even more important in patients with rapid progression associated with the D allele.

When designing intervention studies on renoprotection, a solid estimate of progression rate during the study is of major importance. Proteinuria is a consistent predictor of progression rate. In concordance, in this analysis, baseline proteinuria predicted 12% of the variability in progression rate during the study. It is interesting that by including the preintervention slope, the predictive power of the model doubled to 24%. Thus, knowledge of the preintervention slope allowed a better prediction of the slope during intervention. This suggests that it would be worthwhile to document previous rate of renal function loss of patients who are considered for intervention studies. In our own study, proteinuria and preintervention slope contributed equally to the model, but it should be mentioned that baseline proteinuria was fairly low in our study and that the relative contributions of proteinuria and preintervention slope might have been different in populations with higher proteinuria.

Our data strongly suggest that taking into account preintervention slope during stratification allows a more valid comparison of treatment effects in the study arms. In our own study, a possible difference between the two agents could have been missed by neglecting preintervention slope in the stratification. The predictive value of preintervention slope for the slope during intervention, a frequently used surrogate end point, is of clear relevance for future studies on renoprotective intervention. This holds particularly true for trials with relatively small numbers of patients, in which inclusion of nonprogressors may have a relatively large impact. Our data suggest that it would be fruitful to consider previous rate of renal function loss, if available, because stratification on cross-sectional parameters may not warrant a sufficient match for the risk for renal func-

**Figure 2.** Receiver operating characteristic (ROC) curve showing the sensitivity and 1 − specificity of baseline proteinuria and the combination with preintervention for an intervention slope of >3.8 ml/min per yr (highest quartile). The area under the proteinuria curve is 0.65 ± 0.08. The area under the preintervention curve is higher, 0.73 ± 0.07. The area under the curve is slightly higher when the preintervention slope and baseline proteinuria are combined (0.75 ± 0.07).

**Figure 3.** Bland-Altman plot enalapril group showing the average pre- and postintervention values of the creatinine clearance slope against the change variable. This analysis shows a horizontal scatter without significant correlation ($R^2 = 0.09, P = 0.15$), indicating that the slowing of GFR decline in the enalapril group cannot be explained by regression of the mean.
tion loss. This may substantially enhance the power to detect differences between treatment arms and thus reduce the required number of patients.

Our study has several limitations. Most important, the pre-intervention data in this study were obtained retrospectively; therefore, the potential flaws of a post hoc analysis and notably selection bias should be considered. From the original 81 patients, 21 could not be included; however, the only reason for noninclusion of these patients was that the preintervention period was <1 yr, precluding accurate assessment of the pre-intervention slope. No patients were lost to follow-up, and the intention-to-treat principle was not violated. Moreover, there were no significant differences between patients who were and were not included in this analysis. Thus, by lack of knowledge on preintervention slope in these 21 patients, we cannot exclude selection bias completely, but we consider it unlikely that it plays a major role in these results.

Regression to the mean did not seem to influence our results. Especially for the enalapril group, showed by the horizontal scatter in the Bland-Altman plot. Regression to the mean and mathematical coupling are important to consider when analyzing the relationship between a baseline value and its subsequent change; therefore, it is important when using parameters such as a renal function slope to take this into account. However, regression to the mean is more likely to occur when comparing two single observations (e.g., baseline and change) per individual than in the analysis of serial observations, as was the case in our slope estimations. Of note, a single intervention slope value reflects multiple measurements (mean number of data points 12), which renders regression to the mean less likely; therefore, for future studies, when using slope analysis, it is important to calculate slopes with as many measurements as possible. Another issue when selecting patients according to slope is that fast progressors are more likely to become slow progressors and vice versa. When slope is calculated with multiple measurements, this is less likely to occur. As pointed out, our Bland-Altman plot showed that this issue was not present in our study; however, the possibility of regression of slope has to be taken into account when using slope as a selection parameter in future studies.

The implication of our study is that renal intervention studies should include an assessment of preintervention data in the inclusion criteria. This will pose considerable practical challenges, but it is not entirely at variance with current practice. In several large trials, the inclusion criteria included data on the course of preintervention renal function, usually to exclude patients with a very rapid rate of renal function loss (2,4,21). The Angiotensin-Converting-Enzyme Inhibition in Progressive Renal Insufficiency (AIPRI) trial, for instance, included only patients with <30% variation in 24-h estimated creatinine clearance in a 3-mo screening period (2). To assess the slope, preintervention data on a period of >3 mo (at least 1 yr with a minimum of three data points) would be necessary for all patients. This would increase the total observation period and, in newly identified patients, postpone the start of the intervention. Whereas this can be considered a disadvantage in view of the usual time pressure, it might be balanced by a greater power to detect intervention-associated differences between the groups. If no creatinine clearance data are available, then a slope derived from the reciprocal of serum creatinine might serve the purpose. It should be emphasized that, if adopting this policy, preintervention data should be available for all patients so as not to introduce a selection bias.

Conclusion

Preintervention rate of renal function loss is a main determinant of future rate of renal function loss. Neglect of the previous rate of renal function loss may result in differences in risk for progression between treatment arms in randomized trials, thereby confounding the outcome of studies on long-term renoprotection. Considering previous rate of renal function loss as a randomization parameter, albeit cumbersome, may enhance study power and allow conduction of valid studies in smaller numbers of patients.

Disclosures

None.

References

9. van Essen GG, Rensma PL, de Zeeuw D, Sluiter WJ, Schef-fer H, Apperloo AJ, de Jong PE: Association between an-


