Impaired Vascular Reactivity in Patients with Chronic Kidney Disease

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Abstract

Background: Patients with chronic kidney disease (CKD) show increased cardiovascular morbidity. We hypothesized that vascular properties which can be routinely evaluated noninvasively are related to different stages of CKD and their clinical and biochemical characteristics. Methods: Arterial vascular properties were quantified by the reflective index using digital photoplethysmography in 260 patients with CKD. Patients were grouped according to estimated glomerular filtration rate (eGFR). Additional measurements were performed in 50 healthy control subjects. Results: In patients with CKD stage 1 and 2 (n = 115; age 65 ± 1 year) the reflective index was 30 ± 1%, whereas in patients with CKD stage 3 and 4 (n = 60; age 72 ± 1 year) the reflective index was 36 ± 1%, and in patients with CKD stage 5 (n = 85; age 64 ± 1 year) the reflective index was 36 ± 1% (p < 0.01 by Kruskal-Wallis test) indicating increased arterial stiffness in advanced CKD. Arterial vascular reactivity was significantly impaired in patients with advanced stages of CKD (stage 1 and 2, 78 ± 12%; stage 3 and 4, 32 ± 12%; stage 5, 33 ± 12%; p < 0.01). Univariate analysis showed a significant correlation of the reflective index and eGFR (Pearson r = –0.24; p < 0.0001). Multivariate regression analysis showed an independent association of the reflective index and eGFR (adjusted correlation coefficient, –0.24; p < 0.001). Conclusion: The advanced stages of CKD are associated with increased vascular stiffness and impaired vascular reactivity and these changes are already present in CKD stage 3 and 4.

Introduction

Cardiovascular events are the leading cause of mortality in patients with chronic kidney disease (CKD). Several risk factors have been observed in patients with CKD, including diabetes mellitus, hypertension, dyslipidemia, anemia, increased calcium × phosphate product, endothelial dysfunction and inflammation which contribute to progressive atherosclerosis [1–5]. Early detection of impaired vascular properties by noninvasive methods can be an important diagnostic tool to take measurements for prevention of cardiovascular events. One accepted noninvasive method to determine vascular properties uses digital photoplethysmography, identifying changes of pulse-wave morphology, especially in the diastolic component, which is caused mainly by reflection from the lower limb [6, 7]. Digital photoplethysmography, a noninvasive and investigator-independent method, is a simple procedure for monitoring vascular proper-
ties [8, 9]. Using that method our group recently showed that impaired vascular reactivity was associated with progressive loss of renal allograft function in renal transplant recipients [10]. Now, we hypothesized that vascular properties which can be routinely evaluated noninvasively are related to different stages of CKD and their clinical and biochemical characteristics. We used noninvasive evaluation of vascular properties of patients with CKD at different stages.

Subjects and Methods

Patients

The study was conducted in 260 consecutive patients. The study was approved by the local ethics committee. Each patient gave written informed consent. Inclusion criteria were CKD defined by kidney damage as confirmed by kidney biopsy or markers of damage, or estimated glomerular filtration rate (eGFR) less than 60 ml/min/1.73 m² for more than 3 months. Markers of kidney damage included proteinuria, abnormalities on urine dipstick or sediment examination, or abnormalities on imaging studies of the kidneys. Patients aged less than 18 years, who were pregnant, or who refused to give their informed consent were excluded. Blood pressure was measured 3 times with a sphygmomanometer after 10 min of recumbency and averaged. Renal function was quantified by eGFR which was calculated according to the Modification of Diet in Renal Disease formula [11].

Digital Photoplethysmography

We noninvasively measured digital volume pulse using fingertip photoplethysmography. The basic principles of digital photoplethysmography and its applications have been described in detail by several authors including our group [6–10, 12–14]. The digital pulse waves were measured by the transmission of red and infrared light through the finger pulp using signal extraction technology (Masimo Corporation, Irvine, Calif., USA). Photoplethysmography was conducted using a Vitaguard VG3000 monitor (Getemed, Teltow, Germany) with the sensor (LNOP-Adt SpO2 sensor; Masimo Corporation) located at the third digit of the hand. In patients with arteriovenous shunt measurements were performed at the contralateral side. Photoplethysmographic measurements were performed at room temperature (21–23°C) in subjects lying in the supine position. Orthostatic changes were omitted during the study. Raw data were collected continuously at a rate of 32 per second and transferred to a personal computer. The first derivative of the digital pulse wave was calculated (GraphPad prism 3.0, Graph Pad Software, San Diego, Calif., USA). The local minimum of the first derivative of the digital pulse wave was determined, and the corresponding turning point (= inflection point) in the downslope of the pulse wave was thereby defined exactly. The reflective index was calculated as the mean of the 3rd to the 7th data point after this turning point (= inflection point) and represents the ‘shoulder region’ of the diastolic component of the digital volume pulse wave, which arises from arterial pressure waves reflected back along the aorta from small arteries. Each reflective index was normalized to the amplitude of the first peak of the digital volume pulse wave, which was set to 100. Data of the reflective index obtained were averaged every 2.5 min. As such, the reflective index represents the mean value from 150 to 250 digital volume pulse waves obtained during a period of 2.5 min. The analysis of measurements was performed using a computerized algorithm which was independent of the investigator. Furthermore, the investigator was unaware of the degree of renal impairment. Earlier experimental studies showed that an increased reflective index indicates increased reflection of arterial pressure waves from peripheral arteries, i.e. due to vasoconstriction or increased arterial stiffness.

To evaluate arterial vascular reactivity, the changes of the reflective index during reactive hyperemia evoked by the release of a cuff on the upper arm were investigated. The sphygmomanometric cuff was placed above the antecubital fossa and inflated to 240 mm Hg for 5 min. The reflective index from all pulse waves obtained during a period of 150 s before inflating the cuff and 150 s after release of the cuff were averaged, respectively. Bonetti et al. [15] showed that an attenuated digital response to reactive hyperemia measured by digital volume pulse changes identifies patients with impaired endothelial function. A strong correlation between forearm blood flow response to reactive hyperemia and intra-arterial infusion of acetylcholine has been observed, indicating that reactive hyperemia is at least in part related to endothelial function.

Statistics

Continuous data were given as mean ± SEM. Statistical analyses were performed using GraphPad Prism 3.0 (GraphPad Software, San Diego, Calif., USA) and SPSS (SPSS for Windows, version 11.5, SPSS, Chicago, Ill., USA). Differences between the groups were compared using the Kruskal-Wallis test with Dunn’s multiple comparison post-test and ANOVA post-test for linear trend. Relations between parameters were assessed using Pearson correlations. Multiple regression analysis was used to determine variables independently associated with reflective index. Variables tested were age, gender, body mass index (BMI), systolic and diastolic blood pressure, and pulse pressure. Two-sided p < 0.05 was considered statistically significant.

Results

The study was performed on 260 patients with CKD. 115 patients had CKD stage 1 and 2 (eGFR more than 60 ml/min/1.73 m²; 44%), 60 patients had CKD stage 3 and 4 (eGFR 15–60 ml/min/1.73 m²; 23%), and 85 patients had CKD stage 5 (eGFR less than 15 ml/min/1.73 m²; 33%). 88 patients (34%) underwent chronic hemodialysis. In the patients studied, the cause of CKD was diabetic nephropathy in 41 cases (16%), nephrosclerosis in 79 cases (30%), chronic glomerulonephritis in 18 cases (7%) and other/unknown in 122 cases (47%). In the subgroup of patients with CKD stage 5, the cause of CKD was diabetic nephropathy in 33%.
Clinical and biochemical characteristics of the groups are shown in Table 1. Patients with advanced stages of CKD were older, had higher systolic blood pressure, higher serum potassium, phosphate, and calcium \times phosphate product. Patients with CKD stage 5 had a total protein of 65 ± 1 g/l, albumin of 33 ± 1 g/l, parathyroid hormone of 156 ± 40 pg/ml. We observed a significant negative correlation between serum calcium and parathyroid hormone levels (r² = 0.244; p < 0.01).

Representative pulse waves obtained from digital photoplethysmography are shown for patients with CKD stage 1 and 2 (fig. 1a), patients with CKD stage 3 and 4 (fig. 1b), and patients with CKD stage 5 (fig. 1c), respectively. Advanced CKD was associated with a rise of the ‘shoulder’ region of the diastolic component of the pulse wave, indicating increased arterial stiffness. Increased arterial stiffness was quantified by an increased reflective index in patients with advanced CKD. The mean of reflective index was 30 ± 1% (n = 115) in patients with CKD stage 1 and 2, and 36 ± 1% in patients with CKD stage 3 and 4 (n = 60). The mean reflective index was 36 ± 1% in patients with CKD stage 5 (n = 85). Compared to patients with CKD stage 1 and 2 the reflective index was significantly increased in patients with CKD stage 3 and 4 or in patients with CKD stage 5 (p < 0.01 by Kruskal-Wallis test and Dunn’s nonparametric multiple comparison post-test; fig. 2).

For comparison, the reflective index was also measured in 50 healthy control subjects (mean age, 65 ± 1 years; gender, 29 male, 21 female; body weight, 78 ± 3 kg; height, 1.69 ± 0.01 m; body mass index, 26.4 ± 0.8; systolic blood pressure, 119 ± 1 mm Hg; diastolic blood pressure, 66 ± 1 mm Hg; pulse pressure, 53 ± 1 mm Hg; heart rate, 72 ± 2 per minute; serum creatinine, 81 ± 2 μmol/l; blood urea nitrogen, 7 ± 1 mmol/l; potassium, 4.1 ± 0.1; calcium, 2.30 ± 0.02 mmol/l; phosphate, 1.1 ± 0.0 mmol/l). In these healthy control subjects, the reflective index was 29 ± 1% (n = 50).

Arterial vascular reactivity, i.e. the changes of the reflective index during reactive hyperemia, has been shown to characterize endothelial function. Arterial vascular reactivity was 78 ± 12% in patients with CKD stage 1 and 2 (n = 115) and 32 ± 12% in patients with CKD stage 3 and 4 (n = 60). In patients with CKD stage 5 arterial vascular reactivity was 33 ± 12% (n = 85), indicating impaired endothelial function in patients with advanced kidney disease. Compared to patients with CKD stage 1 and 2 the arterial vascular reactivity was significantly lower in patients with CKD stage 3 and 4 or in patients with CKD stage 5 (p < 0.05 by Kruskal-Wallis test and Dunn’s nonparametric multiple comparison post-test; fig. 3).

Univariate analysis demonstrated a significant correlation between the reflective index and eGFR (Pearson

Table 1. Clinical and biochemical characteristics of 260 patients with CKD

<table>
<thead>
<tr>
<th>CKD</th>
<th>Stage 1 and 2 GFR &gt;60 (n = 115)</th>
<th>Stage 3 and 4 GFR 15–60 (n = 60)</th>
<th>Stage 5 GFR &lt;15 (n = 85)</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, years</td>
<td>65 ± 1</td>
<td>72 ± 1</td>
<td>64 ± 1</td>
<td>0.0008</td>
</tr>
<tr>
<td>Gender, male/female</td>
<td>59/56</td>
<td>35/25</td>
<td>53/32</td>
<td></td>
</tr>
<tr>
<td>Body weight, kg</td>
<td>75 ± 2</td>
<td>77 ± 2</td>
<td>70 ± 2</td>
<td>0.1206</td>
</tr>
<tr>
<td>Height, m</td>
<td>1.69 ± 0.01</td>
<td>1.71 ± 0.01</td>
<td>1.69 ± 0.01</td>
<td>0.1327</td>
</tr>
<tr>
<td>BMI</td>
<td>26.3 ± 0.7</td>
<td>26.3 ± 0.6</td>
<td>24.5 ± 0.6</td>
<td>0.1721</td>
</tr>
<tr>
<td>Systolic blood pressure, mm Hg</td>
<td>133 ± 2</td>
<td>142 ± 4</td>
<td>141 ± 3</td>
<td>0.0316</td>
</tr>
<tr>
<td>Diastolic blood pressure, mm Hg</td>
<td>73 ± 1</td>
<td>74 ± 2</td>
<td>74 ± 2</td>
<td>0.7474</td>
</tr>
<tr>
<td>Pulse pressure, mm Hg</td>
<td>60 ± 1</td>
<td>68 ± 2</td>
<td>67 ± 2</td>
<td>0.0029</td>
</tr>
<tr>
<td>Heart rate, /min</td>
<td>73 ± 1</td>
<td>74 ± 2</td>
<td>76 ± 2</td>
<td>0.2521</td>
</tr>
<tr>
<td>Serum creatinine, μmol/l</td>
<td>74 ± 2</td>
<td>181 ± 11</td>
<td>721 ± 32</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Blood urea nitrogen, mmol/l</td>
<td>6 ± 1</td>
<td>16 ± 1</td>
<td>26 ± 2</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Potassium, mmol/l</td>
<td>4.1 ± 0.0</td>
<td>4.2 ± 0.1</td>
<td>5.0 ± 0.1</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Calcium, mmol/l</td>
<td>2.33 ± 0.02</td>
<td>2.37 ± 0.06</td>
<td>2.33 ± 0.04</td>
<td>0.8275</td>
</tr>
<tr>
<td>Phosphate, mmol/l</td>
<td>1.1 ± 0.0</td>
<td>1.3 ± 0.0</td>
<td>1.9 ± 0.1</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Calcium \times phosphate product, mmol²/l²</td>
<td>2.6 ± 0.1</td>
<td>3.0 ± 0.1</td>
<td>4.3 ± 0.2</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>
Fig. 1. Typical pulse waves obtained from digital photoplethysmography from patients with CKD stage 1 and 2 (a), stage 3 and 4 (b), and stage 5 (c). The shaded area indicates the 'shoulder region' of the diastolic component of the pulse wave, which covers arterial pressure waves reflected back along the aorta from distal reflecting sites. These examples show that the reflective index was 24% in a patient with CKD stage 2, whereas it was 38% in a patient with CKD stage 4, and 33% in a patient with CKD stage 5.

Fig. 2. Bar graph showing mean ± SEM of the reflective index in 115 patients with CKD stage 1 and 2, 60 patients with CKD stage 3 and 4, and 85 patients with CKD stage 5. ** p < 0.01 compared to CKD stage 1 and 2 by the nonparametric Kruskal-Wallis test and Dunn’s multiple comparison post-test.

Fig. 3. Bar graph showing mean ± SEM of arterial vascular reactivity in 115 patients with CKD stage 1 and 2, 60 patients with CKD stage 3 and 4, and 85 patients with CKD stage 5. * p < 0.05 compared to CKD stage 1 and 2 by the nonparametric Kruskal-Wallis test and Dunn’s multiple comparison post-test.

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r = −0.2399; p < 0.0001), body mass index (Pearson r = −0.1420; p < 0.05), serum creatinine (Pearson r = 0.1218; p < 0.05), and urea (Pearson r = 0.1409; p < 0.05).

Multivariate regression analysis confirmed that the reflective index was independently associated with eGFR (adjusted correlation coefficient, −0.24; p < 0.001) whereas age or blood pressure were not significantly associated with the reflective index.

Discussion

The main finding of the present study in a large cohort of patients with CKD is that vascular properties which can be routinely evaluated noninvasively using digital photoplethysmography are related to different stages of CKD. Advanced stages of CKD are associated with increased vascular stiffness and impaired vascular reactivity in CKD stage 5 as well as in CKD stage 3 and 4. Differences between the groups were compared using the *Kruskal-Wallis* test with Dunn’s multiple comparison post-test. Hence, these tests showed significant differences firstly between CKD stage 1 and 2 and CKD stage 3 and 4, and secondly between CKD stage 1 and 2 and CKD stage 5. The results were similar for the baseline reflective index and for arterial vascular reactivity.

Using noninvasive digital photoplethysmography and quantification by the reflective index, we showed that impaired renal function directly affects vascular properties. Other studies used different methods to evaluate arterial properties in patients with CKD. Several studies indicated increased arterial stiffness in patients with CKD [16 – 18]. Increased arterial stiffness was associated with elevated cardiovascular mortality [18, 19]. Changes of reflective index during reactive hyperemia, i.e. arterial vascular reactivity, were significantly lower in patients with advanced CKD. These changes, caused by flow-mediated dilation, were attributed to endothelial function [20, 21]. This is in agreement with other observations of an impaired endothelial function in patients with advanced CKD [5, 22].

The advantage of the described method is that the reflective index and vascular reactivity can be measured routinely in patients with CKD. In contrast to other measurements of endothelial function, the present method is simple, noninvasive, cheap, quick, and reproducible. The method quantifies vascular reactivity during reactive hyperemia, which is related to endothelial function. However, until now no outcome data are available; therefore, one can only speculate about the threshold values. On the other hand, comparisons with healthy subjects and future studies using repeated measurements for longer periods may help clarify the impact of this new method.

Several methods for quantification of vascular function have been introduced; however, no method beyond blood pressure measurements had been introduced in clinical practice. A reliable method for quantification of vascular properties could be desirable before major vascular events including myocardial infarction or stroke occur in particular because of putative prophylaxis and treatment. However, due to limited data the question whether measurements of the reflective index or vascular reactivity are promising tools cannot be answered yet.

For the first time, we clearly indicated by multivariate analysis that increased arterial stiffness was independently associated with reduced eGFR in a large cohort of patients with CKD. On the other hand, age or blood pressure did not predict arterial stiffness. Reduced eGFR is often associated with impaired calcium and phosphate metabolism. Previous findings by Block et al. [23] showed an association between calcium \( \times \) phosphate product and mortality in hemodialysis patients. This finding was confirmed in a study by Kalantar-Zadeh et al. [24] showing that hypercalcemia and hyperphosphatemia predicted a higher death risk in a cohort of 58,058 maintenance hemodialysis patients. Our present results indicate that well-known disturbances in calcium metabolism directly affect vascular properties in patients with CKD which are not on renal replacement therapy.

Furthermore, we could show that increased arterial stiffness as well as reduced vascular reactivity is already present in CKD stage 3 and 4 and that intervention is already required in these stages of CKD. This may play a role in the rate of long-term success of earlier interventions in patients with earlier stages of CKD, which should be examined in follow-up studies.

A limitation of the present study was that patients were stratified according to glomerular filtration rate to CKD stage 1 through 5 as recommended. However, according to this definition we cannot exclude that patients at stage 1 and 2 are clinically stable without any progression to major renal insufficiency. Therefore, measurements of the reflective index and vascular reactivity in these patients may not predict further renal outcome. Another limitation of the present study may be that age could have contributed to some of the differences observed between the different stages of CKD.
However, it should be taken into consideration that multivariate regression analysis confirmed that the reflective index was independently associated with eGFR but not with age.

In summary, the present data from a large cohort of patients with CKD indicate that advanced stages of CKD are associated with increased vascular stiffness and impaired vascular reactivity and that these alterations are already present in CKD stage 3 and 4.

References


