Impact of Renal Functional/Morphological Dynamics on the Calcification of Coronary and Abdominal Arteries in Patients with Chronic Kidney Disease

Takeo Ichii1, Ryota Morimoto1,2, Takahiro Okumura1, Hideki Ishii1, Yosuke Tatami1, Dai Yamamoto1, Soichiro Aoki1, Hiroaki Hiraiwa1, Kenji Furusawa1, Toru Kondo1, Naoki Watanabe1, Naoaki Kano1, Kenji Fukaya1, Akinori Sawamura1, Susumu Suzuki1, Yoshinari Yasuda2 and Toyoaki Murohara1

1 Department of Cardiology, Nagoya University Graduate School of Medicine, Nagoya, Japan
2 Department of CKD Initiatives Internal Medicine, Nagoya University Graduate School of Medicine, Nagoya, Japan

Aim: Fast-progressing vascular calcification (VC) is accompanied by renal atrophy and functional deterioration along with atherosclerosis in patients with chronic kidney disease (CKD). However, the relationship between VC progression and renal functional and/or morphological changes remains unclear.

Methods: We included 70 asymptomatic patients with CKD without hemodialysis in our study. To identify temporal variations, the coronary artery calcification score (CACS), abdominal aortic calcification index (ACI), and renal parenchymal volume index (RPVI) were determined via spiral computed tomography scans taken during the study. We investigated significant factors related to annualized variations of CACS (ΔCACS/y) and ACI (ΔACI/y).

Results: During the follow-up period (4.6 years), median values of CACS [in Agatston units (AU)] and ACI increased from 40.2 to 113.3 AU (p=0.053) and from 13.2 to 21.7% (p=0.036), respectively. Multivariate analysis revealed that CACS at baseline (p<0.001) and diabetes mellitus (DM) status (p=0.037) for ΔCACS/y and ACI at baseline (p=0.017) and hypertension (HT) status (p=0.046) for ΔACI/y were significant independent predictors. Furthermore, annualized RPVI variation was significantly related to both ΔCACS/y and ΔACI/y (R=−0.565, p<0.001, and R=−0.289, p=0.015, respectively). On the other hand, independent contributions of the estimated glomerular filtration rate (eGFR) and annualized eGFR variation to VC progression were not confirmed.

Conclusion: The degree of VC at baseline, DM, HT, and changes in renal volume, but not eGFR, had a strong impact on VC progression in patients with CKD.

Key words: Coronary artery calcification, Abdominal aortic calcification index, Renal parenchymal volume, Chronic kidney disease

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Introduction

In patients with chronic kidney disease (CKD), cardiovascular disease (CVD) is a major cause of morbidity and mortality1-3. Vascular calcification (VC) is highly prevalent in patients with CKD undergoing dialysis4-6. Even in pre-dialysis patients with CKD, the high prevalence and rapid progression of VC is well recognized7-10. In addition, the presence and progression of VC is closely related to the increased morbidity and mortality of CVD11-15.

The progression of atherosclerosis, which results in CKD and CVD, is closely associated with lipid
abnormalities, nutrition status, and micro inflammation. VC is a key feature of advanced atherosclerosis and the extent of calcification is strongly correlated with the extent and severity of atherosclerotic lesions. Previous studies have shown that atherosclerotic risk factors, such as hypertension (HT), dyslipidemia (DL), and diabetes mellitus (DM), affect the prevalence and progression of VC. These factors affect renal functional deterioration and atrophy as well. Although both VC progression and renal functional/morphological changes are influenced by atherosclerosis, the relationship between VC and renal dynamics in patients with CKD remains unknown.

**Aim**

VC can be evaluated semi-quantitatively in coronary and abdominal arteries as the coronary artery calcification (CAC) score (CACS) and abdominal aortic calcification index (ACI), respectively. Therefore, this study aims to investigate the relationship between renal functional/morphological parameters and progression of CACS or ACI in patients with CKD without hemodialysis.

**Methods**

**Patients**

From September 2009 to January 2013, 261 asymptomatic, pre-dialysis patients with CKD were recruited to undergo non-contrast coronary and abdominal computed tomography (CT) scans simultaneously in the outpatient clinic at the Department of Nephrology in the Nagoya University Hospital. CKD was defined as an estimated glomerular filtration rate (eGFR) < 60 mL/min/1.73 m² or the presence of proteinuria and/or renal disease at study entry. Among these patients, 79 underwent follow-up non-contrast coronary and abdominal CT scans by August 2016 over a 1-year interval. Among the 79 patients, no patient had a history of coronary artery disease, abdominal aortic artery repair and stenting, active malignancy, and/or active autoimmune disease. After excluding patients with polycystic kidney disease (n=2), hemodialysis maintenance between follow-up periods (n=1), unclear CT images (n=1), and coronary intervention during the follow-up periods (n=5), we evaluated 70 patients in this study. Coronary CT scans were used to determine CACS in Agatston units (AU), and abdominal CT scans were used to determine ACI as the abdominal artery calcification (AAC) score and renal parenchymal volume (RPV). In addition, all patients underwent renal functional tests and comprehensive atherosclerotic screening. When we evaluated the progression of VC, we calculated the annualized variation (calculated as [second value – first value/days of follow-up] × 365) to determine the relationship between the annualized CACS variation (ΔCACS/y) or annualized ACI variation (ΔACI/y) and baseline variables or annualized variations with regard to renal function and morphology. The study protocol was approved by the Ethics Review Board of Nagoya University School of Medicine, and written informed consent was obtained from all patients prior to performing the first CT scan.

HT was defined as systolic blood pressure ≥ 140 mmHg, diastolic blood pressure ≥ 90 mmHg, and/or the present use of anti-hypertensive medications. DL was defined as low-density lipoprotein (LDL) cholesterol ≥ 140 mg/dL, high-density lipoprotein (HDL) cholesterol < 40 mg/dL, triglycerides ≥ 150 mg/dL, and/or the present use of anti-hyperlipidemic medications. DM was defined as fasting plasma glucose concentration > 126 mg/dL, glycosylated hemoglobin concentration ≥ 6.5% (National Glycohemoglobin Standardization Program), and/or the present use of anti-hyperglycemic medications. Those who reported as presently smoking or previously smoked at least one cigarette per day for more than 1 year were defined as patients with a history of smoking.

CKD stages were classified using eGFR and urinary protein excretion data. To estimate GFR, we used a modified equation for Japanese: eGFR (mL/min/1.73 m²) = 194 × serum creatinine – 1.094 × age – 0.287 × 0.739 (in females). Urinary protein excretion was determined via the urinary protein to creatinine ratio (PCR) in spot morning urine samples. We categorized eGFR levels into the following six groups according to the National Kidney Foundation Kidney Disease Outcomes Quality Initiative Guidelines: ≥ 90, 60–89, 45–49, 30–44, 15–29, and <15 mL/min/1.73 m², which were G1, G2, G3a, G3b, G4, and G5, respectively. We divided patients into the following three groups with regard to the urinary PCR: <0.15, 0.15–0.49, and ≥ 0.5 g/gCr, which were A1, A2, and A3, respectively.

**CACS Measurements**

All patients underwent CACS measurements using a multi-slice non-contrast CT scan (Siemens Medical Solutions, Forchheim, Germany) with a gantry rotation of 0.4 s, collimation of 2.5 mm (slice thickness), and reconstruction time of 6 frames/s. The images were scored by a single physician blinded to the clinical and biochemical aspects of the study patients. CACS was determined by multiplying the area of each calcified lesion by a weighting factor corresponding to the peak pixel intensity for each lesion.
and then summing the calcification scores of all coronary arteries. Lesions above a threshold of 130 Hounsfield units (HU) were defined as calcified.

**ACI Measurements**

ACI and RPV measurements for each patient were obtained in a manner similar to that for CACS measurements; that is, using multi-slice non-contrast CT scans. It is noteworthy that images were obtained with a 5-mm slice thickness. Calcification was considered to be present if an area of \( \geq 1 \text{ mm}^2 \) displayed a density of 130 HU. The AAC score was calculated from the site where the renal artery arises to the bifurcation into the common iliac arteries of the aorta. The cross-section of the abdominal aorta on each slice was radially divided into 12 segments. ACI was calculated as follows: 

\[
ACI = \frac{\text{total calcification score on all slices}}{12 \times 1/(\text{number of slices}) \times 100\%} \quad (6, 10, 15) 
\]

ACI was independently calculated by two physicians who were blinded to the clinical and biochemical aspects of each study patient.

**RPV Measurements**

RPV was calculated based on a software that measures volume, which is available on ZioStation2 (Zio Software Inc., Tokyo, Japan). This is a three-dimensional (3D) volumetric method that is widely used to measure renal volume. The renal cortex was traced on each image showing the renal parenchyma, with the exclusion of sinus fat, vessels, and cysts. After complete tracing of each kidney, CT volume was automatically calculated and RPV was defined as the sum of renal volume on the left and right sides (Fig. 1). RPV measurement was conducted independently by two physicians who were blinded to the clinical and biochemical aspects of each study patient. Previous study showed that RPV can be corrected via the body surface area (BSA) (correction value = 0.68) \( ^{30} \); we applied this correction by dividing RPV with BSA and defined it as the RPV index (RPVI), which was used in downstream analyses.

**Statistical Analysis**

All variables were expressed as means ± standard deviations or as medians and interquartile ranges for non-parametric variables. Categorical data were expressed as a number (percentage). Between-group differences were tested using the Student’s \( t \)-test for
parametric variables and the Mann–Whitney U-test for non-parametric variables. Categorical variables were examined using the Pearson chi-squared test or Fisher’s exact test as appropriate. When the associations between ∆CACS/y or ∆ACI/y and various parameters were evaluated, baseline variables and annualized variations were independently calculated using correlation coefficients and a linear regression analysis. To reveal the independent factors associated with the progression of VC, significant variables identified by univariate analysis were entered in multivariable linear regression models. A significant difference is assumed with \( p < 0.05 \) for all analyses. All statistical analyses were performed using JMP statistical software version 11.0.0 (SAS Institute Inc., Cary, NC, USA).

**Results**

**Patient Characteristics and Variable Parameter Changes**

A total of 70 patients were evaluated in this study. The median duration between the first and repeated CT scans was 4.6 (3.2–5.4) years. The baseline clinical characteristics and changes of CACS, RPVI, and laboratory data are summarized in Table 1. The etiology of kidney disease was as follows: nephro sclerosis in 32 patients (45.7%), glomerulonephritis in 20 patients (28.6%), diabetic nephropathy in 7 patients (10.0%), interstitial nephritis in 1 patient (1.4%), and other diseases and unknown in 10 patients (14.3%). The percentage of medication usage that did not statistically increase during the study, with the exception of phosphate binders (from 5.7% at baseline to 17.1% at follow-up, \( p = 0.034 \)).

CACS (in AU) and ACI significantly increased from 40.2 (2.2–222.4) to 113.3 (11.0–576.0) AU (\( p = 0.053 \)) and from 13.2 (3.4–31.9) to 21.7 (8.3–47.1) % (\( p = 0.036 \)), respectively. The number of patients with CACS ≥ 100 or ≥ 400 increased from 26 patients (37.1%) to 35 patients (50.0%) and from 10 patients (14.3%) to 24 patients (34.3%), respectively, during the follow-up period. Although most patients had increased CAC and ACI progression, 14 patients (20.0%) out of 70 patients (17.1%) had no apparent ACI at baseline remained during the follow-up period. RPVI and eGFR significantly decreased from 131.8±43.2 to 115.5±39.3 mL/min/1.73 m² (\( p = 0.021 \)) and from 40.3±19.2 to 32.9±19.0 mL/min/1.73 m² (\( p = 0.022 \)), respectively. There was a positive correlation between RPVI and eGFR (R=0.634, \( p < 0.001 \)) (Supplemental Fig. 1). The baseline values of CACS, ACI, and RPVI in each CKD stages are summarized in Table 2. Although patients with CKD A3 exhibited a trend of higher CACS, there were no statistical differences between both CKD G and A stages at the baseline CACS or ACI. On the other hand, as the CKD G stage worsened, RPVI significantly decreased (\( p < 0.001 \)).

**∆CACS/y or ∆ACI/y profiles in patients with CKD**

The comparisons of ∆CACS/y and ∆ACI/y with clinical characteristics are presented in Fig. 2. Progressive CAC was significantly associated with risk factors, such as HT, DL, DM, and CKD stage A3 (Fig. 2-A), and progressive ACI was associated with HT (Fig. 2-B). At baseline, CACS, RPVI, serum albumin, alkaline phosphatase, glycated hemoglobin (HbA1c), and urinary PCR for ∆CACS/y and ACI, serum albumin, calcium, brain natriuretic peptide (BNP), and hemoglobin for ∆ACI/y were significantly related variables (Supplemental Table 1). Multivariable linear regression analysis revealed that DM status (\( \beta = 0.161, p = 0.037 \)) and CACS at baseline (\( \beta = 0.735, p < 0.001 \)) for ∆CACS/y and HT status (\( \beta = 0.238, p = 0.046 \)) and ACI at baseline (\( \beta = 0.285, p = 0.017 \)) for ∆ACI/y were significantly independent predictors (Table 3-A). In the annualized variation, there was a positive correlation between ∆CACS/y and ∆ACI/y (R=0.351, \( p = 0.003 \)) (Fig. 3-A). ∆CACS/y significantly correlated with the annualized variations of BSA (∆BSA/y) (R=−0.249, \( p = 0.038 \)) and RPVI (∆RPVI/y) (R=−0.565, \( p < 0.001 \)) (Supplemental Table 2, Fig. 3-B). Moreover, multiple linear regression analysis revealed that ∆RPVI/y (\( \beta = -0.537, p < 0.001 \)) was an independent factor for ∆CACS/y (Table 3-B). A similar trend was observed in the analysis for ∆ACI/y. Although many annualized variations, such as body weight (BW), RPVI, eGFR, calcium, intact parathyroid hormone, BNP, and HbA1c were corrected with ∆ACI/y (Supplemental Table 2, Fig. 3-C), multiple linear regression analysis revealed that ∆BW/y (\( \beta = -0.446, p = 0.001 \)), ∆RPVI/y (\( \beta = -0.371, p = 0.015 \)), and ∆HbA1c/y (\( \beta = -0.264, p = 0.041 \)) were independent factors for ∆ACI/y (Table 3-B). On the other hand, eGFR at baseline and annualized eGFR variation (∆eGFR/y) did not significantly correlate with ∆CACS/y and ∆ACI/y in the multivariable analysis.

**Discussion**

There is a high prevalence of rapid VC progression in patients with CKD7-10, which is prognostically related to the increased morbidity and mortality of CVD11-15. In this study, we discerned the relationship between CAC or AAC progression and renal functional/morphological changes and revealed the following results:
Table 1. Clinical characteristics at baseline and follow-up period (n=70)

|                              | Baseline          | Follow-up         | p     |
|------------------------------|-------------------|-------------------|-------|
| Age, years                   | 68.9 ± 9.9        | 73.2 ± 9.9        | -     |
| Female, n (%)                | 24 (34.3)         |                   |       |
| BW, kg                       | 62.2 ± 12.0       | 61.0 ± 12.7       | 0.57  |
| BMI, kg/m²                   | 24.0 ± 3.5        | 23.7 ± 3.5        | 0.61  |
| BSA, m²                      | 1.65 ± 0.18       | 1.63 ± 0.20       | 0.56  |
| Hypertension, n (%)          | 56 (80.0)         | 59 (84.3)         | 0.51  |
| SBP, mmHg                    | 135.1 ± 18.4      | 130.0 ± 14.5      | 0.072 |
| DBP, mmHg                    | 77.7 ± 12.1       | 73.0 ± 11.4       | 0.02  |
| Dyslipidemia, n (%)          | 48 (68.6)         | 50 (71.4)         | 0.71  |
| Diabetes mellitus, n (%)     | 27 (38.6)         | 26 (37.1)         | 0.86  |
| Smoking history, n (%)       | 31 (44.3)         | 31 (44.3)         | 1.00  |
| CKD G stage                  |                   |                   |       |
| G1, n (%)                    | 1 (1.4)           | 1 (1.4)           | 0.091 |
| G2, n (%)                    | 7 (10.0)          | 5 (7.1)           |       |
| G3a, n (%)                   | 20 (28.6)         | 10 (14.3)         |       |
| G3b, n (%)                   | 17 (24.3)         | 20 (28.6)         |       |
| G4, n (%)                    | 21 (30.0)         | 20 (28.6)         |       |
| G5, n (%)                    | 4 (5.7)           | 14 (20.0)         |       |
| CKD A stage                  |                   |                   | 0.1   |
| A1, n (%)                    | 21 (30.0)         | 20 (28.6)         |       |
| A2, n (%)                    | 26 (37.1)         | 16 (22.9)         |       |
| A3, n (%)                    | 23 (32.9)         | 34 (48.6)         |       |
| Medications, n (%)           |                   |                   |       |
| Anti-hypertension drugs, n (%) | 55 (78.6)     | 58 (82.9)         | 0.52  |
| ACE-I/ARBs, n (%)            | 49 (70.0)         | 51 (72.3)         | 0.71  |
| Warfarin, n (%)              | 2 (2.9)           | 4 (5.7)           | 0.4   |
| Statins, n (%)               | 36 (51.4)         | 39 (55.7)         | 0.72  |
| Anti-diabetes drugs, n (%)   | 18 (25.7)         | 18 (25.7)         | 1.00  |
| Vitamin-D, n (%)             | 5 (7.1)           | 8 (11.4)          | 0.38  |
| Phosphate binders, n (%)     | 4 (5.7)           | 12 (17.1)         | 0.034 |
| CACS, AU                     | 40.2 (2.2-222.4)  | 113.3 (11.0-576.0)| 0.053 |
| ACL, %                       | 13.2 (3.4-31.9)   | 21.7 (8.3-47.1)   | 0.036 |
| RPVI, mL/m²                   | 131.8 ± 43.2      | 115.5 ± 39.3      | 0.021 |
| Laboratory data              |                   |                   |       |
| Serum albumin, g/dL          | 3.79 ± 0.45       | 3.91 ± 0.35       | 0.099 |
| Creatinine, mg/dL            | 1.38 (1.06-1.90)  | 1.60 (1.11-2.74)  | 0.036 |
| eGFR, mL/min/1.73m²          | 40.3 ± 19.2       | 32.9 ± 19.0       | 0.022 |
| Calcium, mg/dL               | 4.74 ± 0.18       | 4.68 ± 0.20       | 0.061 |
| Phosphorus, mg/dL            | 3.37 ± 0.51       | 3.56 ± 0.71       | 0.078 |
| Alkaline phosphatase, IU/L    | 237.6 ± 81.1      | 244.3 ± 92.6      | 0.65  |
| Intact-PTH, pg/mL            | 62.1 (38.1-102.8) | 76.8 (52.6-127.0)| 0.006 |
| BNP, pg/mL                   | 24.1 (10.4-61.1)  | 25.4 (12.4-75.6)  | 0.42  |
| Hemoglobin, g/dL             | 12.3 ± 1.7        | 12.4 ± 1.8        | 0.79  |
| HbA1c, %                     | 6.0 ± 0.5         | 6.2 ± 0.7         | 0.22  |
| LDL-cholesterol, mg/dL       | 97.9 ± 25.6       | 96.9 ± 29.6       | 0.82  |
| Urinary PCR, g/gCr           | 0.35 (0.12-0.74)  | 0.51 (0.11-1.37)  | 0.14  |

CT follow-up period: 4.6 (3.2-5.4) years

Data are expressed as means ± SD, or numbers (percentages), or median (interquartile range).

BW, Body weight; BMI, body mass index; BSA, body surface area; SBP, systolic blood pressure; DBP, diastolic blood pressure; CKD, chronic kidney disease; ACE-I, angiotensin converting enzyme inhibitor; ARBs, angiotensin receptor blockers; CACS, coronary artery calcification score; AU, Agatston unit; ACI, abdominal aortic calcification index; RPVI, renal parenchymal volume index; eGFR, estimated glomerular filtration rate; PTH, parathyroid hormone; BNP, brain natriuretic peptide; HbA1c, glycated hemoglobin; LDL, low-density lipoprotein; PCR, protein to creatinine ratio.
Table 2. The baseline values of CACS, ACI, and RPVI in each CKD stages

|          | G1-2          | G3          | G4-5         | p    |
|----------|---------------|-------------|--------------|------|
| CACS     | 133.1 (38.3-582.1) | 27.2 (0.6-146.6) | 30.3 (1.7-350.0) | 0.21 |
| ACI      | 20.6 (4.6-41.2) | 11.9 (3.2-30.3) | 17.2 (3.3-32.0) | 0.71 |
| RPVI     | 172.6 ± 32.4 | 144.2 ± 38.2 | 100.5 ± 32.4 | <0.001 |

|          | A1            | A2            | A3            |
|----------|---------------|---------------|---------------|
| CACS     | 20.0 (0-209.5) | 27.3 (2.4-91.8) | 153.8 (9.6-338.2) | 0.11 |
| ACI      | 12.1 (3.0-26.8) | 9.3 (2.7-31.1) | 22.7 (7.0-37.0) | 0.31 |
| RPVI     | 137.7 ± 39.0 | 118.7 ± 33.2 | 141.3 ± 53.8 | 0.14 |

Data are expressed as means ± SD, or median (interquartile range). Abbreviations as in Table 1.

Fig. 2.

(A) Comparison of annualized coronary artery calcification score variation (ΔCACS/y) values with clinical characteristics. (B) Comparison of annualized abdominal aortic calcification index variation (ΔACI/y) values with clinical characteristics. ΔCACS/y was higher in patients with hypertension (35.7 vs. 0.6; p < 0.001), dyslipidemia (28.9 vs. 4.2; p = 0.023), diabetes mellitus (42.0 vs. 11.9; p = 0.006), and chronic kidney disease (CKD) stage A3 (A1: A2: A3 = 15.8: 11.6: 54.5; vs. A1, p = 0.027; vs. A2, p = 0.066). ΔACI/y was higher in patients with hypertension (1.8 vs. 0.9; p = 0.004). All values represented are median.
1) CACS at baseline and DM were independent predictors for CAC progression.
2) ACI at baseline and HT were independent predictors for AAC progression.
3) In annualized variations, a decrease in RPV, but not in eGFR, were significantly related to both CAC and AAC progression.

To our knowledge, this is the first study dissecting and clarifying the intimate relationships between RPV and VC progression.

Several studies have reported that the presence of calcification, DM, and HT are strong indicators for progressive calcification, while VC absence in patients with CKD strongly implies a lack of calcification during disease progression. Stavroulopoulos et al. have reported that patients with CKD with DM have a higher prevalence and greater increase in CACS than patients with CKD with non-DM. Hyperglycemia

| Table 3-A. Linear regression analysis in baseline variable |
|---------------------------------------------------------------|
| For \( \Delta \text{CACS}/y \)                                  |
| Univariate analysis                                           | Multivariate analysis |
| \( R \)            | \( p \) | \( \beta \) | \( p \) |
| Hypertension       | 0.281  | <0.001 | 0.032  | 0.68  |
| Dyslipidemia       | 0.203  | 0.023  | -0.026 | 0.74  |
| Diabetes mellitus  | 0.38   | 0.006  | 0.161  | 0.037 |
| CACS               | 0.825  | <0.001 | 0.735  | <0.001|
| RPVI               | 0.257  | 0.032  | 0.066  | 0.37  |
| Serum albumin      | -0.273 | 0.022  | -0.077 | 0.35  |
| Alkaline phosphatase| 0.274  | 0.022  | 0.128  | 0.079 |
| Urinary PCR        | 0.305  | 0.01   | -0.026 | 0.77  |

\( \Delta (\text{variable})/y \) means annualized variation of the variable. Abbreviations as in Table 1.

| Table 3-B. Linear regression analysis in annualized variation |
|---------------------------------------------------------------|
| For \( \Delta \text{CACS}/y \)                                  |
| Univariate analysis                                           | Multivariate analysis |
| \( R \)            | \( p \) | \( \beta \) | \( p \) |
| \( \Delta \text{BSA}/y \) | -0.249 | 0.38   | -0.156 | 0.12  |
| \( \Delta \text{RPVI}/y \) | -0.565 | <0.001 | -0.537 | <0.001|

| For \( \Delta \text{ACI}/y \)                                  |
| Univariate analysis                                           | Multivariate analysis |
| \( R \)            | \( p \) | \( \beta \) | \( p \) |
| \( \Delta \text{BW}/y \) | -0.378 | 0.001  | -0.446 | 0.001 |
| \( \Delta \text{RPVI}/y \) | -0.289 | 0.015  | -0.371 | 0.015 |
| \( \Delta \text{eGFR}/y \) | -0.323 | 0.006  | 0.191  | 0.27  |
| \( \Delta \text{Calcium}/y \) | -0.27  | 0.006  | -0.019 | 0.91  |
| \( \Delta \text{Intact-PTH}/y \) | 0.321  | 0.012  | 0.227  | 0.22  |
| \( \Delta \text{BNP}/y \) | 0.369  | 0.005  | 0.116  | 0.49  |
| \( \Delta \text{HbA1c}/y \) | -0.341 | 0.012  | -0.264 | 0.041 |

\( \Delta (\text{variable})/y \) means annualized variation of the variable. Abbreviations as in Table 1.
and insulin resistance lead to osteogenic differentiation of vascular smooth muscle cells, and an accumulation of advanced glycation end products resulting from long-term hyperglycemia have been reported to be associated with CAC. HT is a well-established risk factor for arterial stiffness and medial calcification, and some previous studies have revealed that pulse blood pressure levels promote AAC progression. VC occurs in the intima in association with atherosclerosis, which is characterized by an increase in the deposition of plaques, and in the media in association with arteriosclerosis, which is characterized by vascular stiffening due to loss of elasticity of the arterial musculature. A previous study has revealed that there is a significant relationship between CAC and AAC progression, and our study revealed that both the annualized variations had a significant correlation in the same manner. Although a combination of intimal and medial calcification may occur in patients with CKD, CAC is considered to be mainly due to intimal calcification and AAC to be due to medial calcification. This is why there is a difference in the predict factors between CAC and AAC progression.

Interestingly, we found that renal morphological change was associated more strongly with VC variability over time than eGFR in our patient cohort despite there being a correlation between RPVI and eGFR. This means that the more advanced renal atrophy becomes, the more severe VC is formed. Although GFR is an indicator of the filtration ability of kidneys and eGFR is an excellent marker that can be estimated based on serum creatinine levels, serum creatinine is affected by muscle mass and nutritional status. In addition, serum creatinine levels may be underestimated in the elderly. On the other hand, RPV, excluding tissues that do not contribute to renal function, can be quantitatively and accurately measured based on CT images and more reflective of renal function, including renal endothelial vascular function in addition to filtration ability. Renal atrophy results from global glomerulosclerosis, tubular atrophy, and interstitial fibrosis due to macro- and microvascular damage. α-Klotho is one of the most important regulatory factors influenced by vascular calcification and facilitates normal phosphaturic function of FGF23 in the kidneys. α-Klotho is expressed mostly in renal tubular cells and parathyroid glands, and production and serum levels decline upon decreased renal function. Moreover, tubular atrophy downregulates the expression of various proteins associated with vascular calcification. Thus, changes in RPV, but renal function estimated by eGFR, may strongly synchronize with VC progression.

The decrease of BW was another independent associated factor for ΔACI/y in our study. This is considered to occur because of malnutrition in patients with CKD. Recent data have suggested that malnutrition can trigger CKD–mineral bone disorder and accelerate VC progression. Previous studies have revealed that phosphate overload leads to mineral bone abnormalities and directly promotes systemic malnutrition and VC in CKD, and this is one of the reasons why lowering the phosphate level is effective in reducing calcification. In our study, HbA1c

Fig. 3. (A) Correlation between annualized coronary artery calcification score variation (ΔCACS/y) and annualized abdominal aortic calcification index variation (ΔACI/y). (B) Correlation between ΔCACS/y and annualized renal parenchymal volume index variation (ΔRPVI/y). (C) Correlation between ΔACI/y and ΔRPVI/y. ΔCACS/y revealed a positive correlation to ΔACI/y (R=0.351, p=0.003). Furthermore, ΔCACS/y revealed a negative correlation to ΔRPVI/y (R=−0.565, p<0.001). In a similar manner, ΔACI/y revealed a weak negative correlation to ΔRPVI/y (R=−0.289, p=0.015).
changes are also negatively associated with ΔACI/y. This mechanism is not clear; however, because HbA1c improves according to the deterioration of renal function and malnutrition, these changes are considered to have secondarily affected ΔACI/y.

There are several limitations to our study. First, this study was conducted in a single center and included few patients. Second, 7 patients with diabetic nephropathy were enrolled in our study. Although, in some cases, renal volume is expanded in the early phase of diabetic nephropathy, there was no impact on the relationship between ΔCACS/y or ΔACI/y and ΔRPVI/y when we eliminated these patients from our study. Furthermore, the prevalence of diabetic nephropathy in this study was much less than that in the general patient population. There are two possible reasons for this. The first reason is that this study was conducted at a single university hospital and had an etiologic bias. The second reason is that some patients with diabetic nephropathy were excluded because they had undergone coronary intervention in the past. At last, when we performed the measurement of renal volume by CT scan, radiation exposure was increased in comparison with non-invasive sonography. However, we adopted this method because estimating CT-based renal parenchymal volume was more accurate than sonographic measurement.

**Conclusion**

VC progression was strongly associated with changes in renal volume and predicted by a high baseline VC as well as the presence of atherosclerotic diseases, such as DM and HT, in patients with CKD who were not on dialysis. With regard to VC progression, changes in renal volume provided more information than conventional indicators of kidney function, such as eGFR and urinary protein. When treating patients with CKD, clinicians should bear in mind that baseline VC presence, DM, HT, and renal volumes are crucial factors that may enhance VC progression and thus cardiovascular disease.

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**Disclosures**

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Supplemental Fig. 1. Correlation between estimated glomerular filtration rate (eGFR) and renal parenchymal volume index (RPVI) (open symbols: at baseline, solid symbols: at follow-up period)

RPVI revealed a positive correlation to eGFR ($R=0.634$, $p<0.001$).
Supplemental Table 1. Correlations between baseline values and $\Delta$CACS/y or $\Delta$ACI/y

|                | $\Delta$CACS/y R | $\Delta$CACS/y p | $\Delta$ACI/y R | $\Delta$ACI/y p |
|----------------|-------------------|------------------|-----------------|-----------------|
| Age            | 0.199             | 0.099            | 0.203           | 0.092           |
| BW             | -0.055            | 0.65             | 0.065           | 0.6             |
| BMI            | -0.044            | 0.72             | 0.124           | 0.31            |
| BSA            | -0.046            | 0.71             | 0.06            | 0.62            |
| SBP            | 0.072             | 0.56             | 0.105           | 0.39            |
| DBP            | -0.222            | 0.065            | -0.009          | 0.94            |
| CACS           | 0.825             | <0.001           | 0.255           | 0.033           |
| ACI            | 0.452             | <0.001           | 0.44            | <0.001          |
| RPVI           | 0.257             | 0.032            | 0.067           | 0.58            |
| Serum albumin  | -0.273            | 0.022            | -0.300          | 0.012           |
| Creatinine     | -0.023            | 0.85             | 0.024           | 0.84            |
| eGFR           | -0.031            | 0.8              | 0.002           | 0.98            |
| Calcium        | 0.11              | 0.37             | 0.278           | 0.02            |
| Phosphorus     | 0.149             | 0.22             | 0.067           | 0.58            |
| Alkaline phospatase | 0.274     | 0.022            | 0.179           | 0.14            |
| Intact-PTH     | -0.146            | 0.26             | 0.079           | 0.54            |
| BNP            | 0.112             | 0.39             | 0.253           | 0.049           |
| Hemoglobin     | -0.23             | 0.055            | -0.291          | 0.015           |
| HbA1c          | 0.256             | 0.034            | 0.066           | 0.59            |
| LDL-cholesterol| -0.025            | 0.84             | -0.114          | 0.35            |
| Urinary PCR    | 0.305             | 0.01             | 0.228           | 0.057           |

$\Delta$(variable)/y means annualized variation of the variable. Abbreviations as in Table 1.

Supplemental Table 2. Correlations between annualized variations and $\Delta$CACS/y or $\Delta$ACI/y

|                | $\Delta$CACS/y R | $\Delta$CACS/y p | $\Delta$ACI/y R | $\Delta$ACI/y p |
|----------------|-------------------|------------------|-----------------|-----------------|
| $\Delta$BW/y   | -0.223            | 0.064            | -0.378          | 0.001           |
| $\Delta$BMI/y  | -0.193            | 0.11             | -0.346          | 0.003           |
| $\Delta$BSA/y  | -0.249            | 0.038            | -0.358          | 0.002           |
| $\Delta$SBP/y  | -0.149            | 0.22             | -0.156          | 0.2             |
| $\Delta$DBP/y  | -0.105            | 0.39             | -0.198          | 0.1             |
| $\Delta$CACS/y | -0.565            | <0.001           | -0.289          | 0.015           |
| $\Delta$ACI/y  | 0.351             | 0.003            | -              | -               |
| $\Delta$RPVI/y | -0.013            | 0.91             | 0.113           | 0.35            |
| $\Delta$Serum albumin/y | 0.042 | 0.73             | 0.281           | 0.019           |
| $\Delta$Creatinine/y | -0.184     | 0.13             | -0.323          | 0.006           |
| $\Delta$eGFR/y | -0.088            | 0.47             | -0.27           | 0.024           |
| $\Delta$Calcium/y | -0.033    | 0.79             | 0.151           | 0.21            |
| $\Delta$Phosphorus/y | -0.191   | 0.11             | -0.134          | 0.27            |
| $\Delta$Alkaline phospatase/y | 0.205 | 0.11             | 0.321           | 0.012           |
| $\Delta$Intact-PTH/y | 0.087    | 0.52             | 0.369           | 0.005           |
| $\Delta$BNP/y  | -0.033            | 0.79             | 0.162           | 0.18            |
| $\Delta$Hemoglobin/y | 0.077         | 0.58             | -0.341          | 0.012           |
| $\Delta$HbA1c/y | -0.162            | 0.18             | 0.13            | 0.29            |
| $\Delta$LDL-cholesterol/y | 0.037     | 0.76             | 0.171           | 0.16            |

$\Delta$(variable)/y means annualized variation of the variable. Abbreviations as in Table 1.