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Original Article

ORIGINAL ARTICLE

Prevalence, progression and implications of breast artery calcification in patients with chronic kidney disease

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ABSTRACT

Breast arterial calcification (BAC) is increasingly recognized as a specific marker of medial calcification. The present retrospective observational cohort study aimed to define the prevalence, progression rate, risk factors and clinical implications of BAC in chronic kidney disease (CKD) patients across stages of disease. The presence and extent of BAC were determined on mammograms in 310 females (58.7 ± 10.8 years, Caucasian) with CKD across various stages of disease [CKD G2–5D $n = 132$; transplant (Tx) recipients $n = 178$]. In a subset of 88 patients, repeat mammography was performed, allowing us to calculate the annualized BAC rate. Overall, BAC was observed in 34.7% of the patients. BAC prevalence ($P = 0.02$) and BAC score ($P = 0.05$) increased along the progression of CKD. In the overall cohort, patients with BAC were characterized by older age, more cardiovascular disease, more inflammation, higher pulse pressure and borderline higher prevalence of diabetes and were more often treated with a vitamin K antagonist (VKA). The BAC progression rate was significantly lower in Tx patients as compared with CKD G5D. Progressors were characterized by more inflammation, worse kidney function, higher BAC score and higher serum phosphate level (Tx only) at baseline and were more often treated with a VKA. Major adverse cardiovascular event-free survival was significantly worse in Tx patients with BAC. In conclusion, BAC is common among CKD patients, progresses at a slower pace in Tx patients as compared with CKD 5D and associates with dismal cardiovascular outcomes. BAC score, kidney function, serum phosphate at baseline and VKA usage seem to be important determinants of progression.

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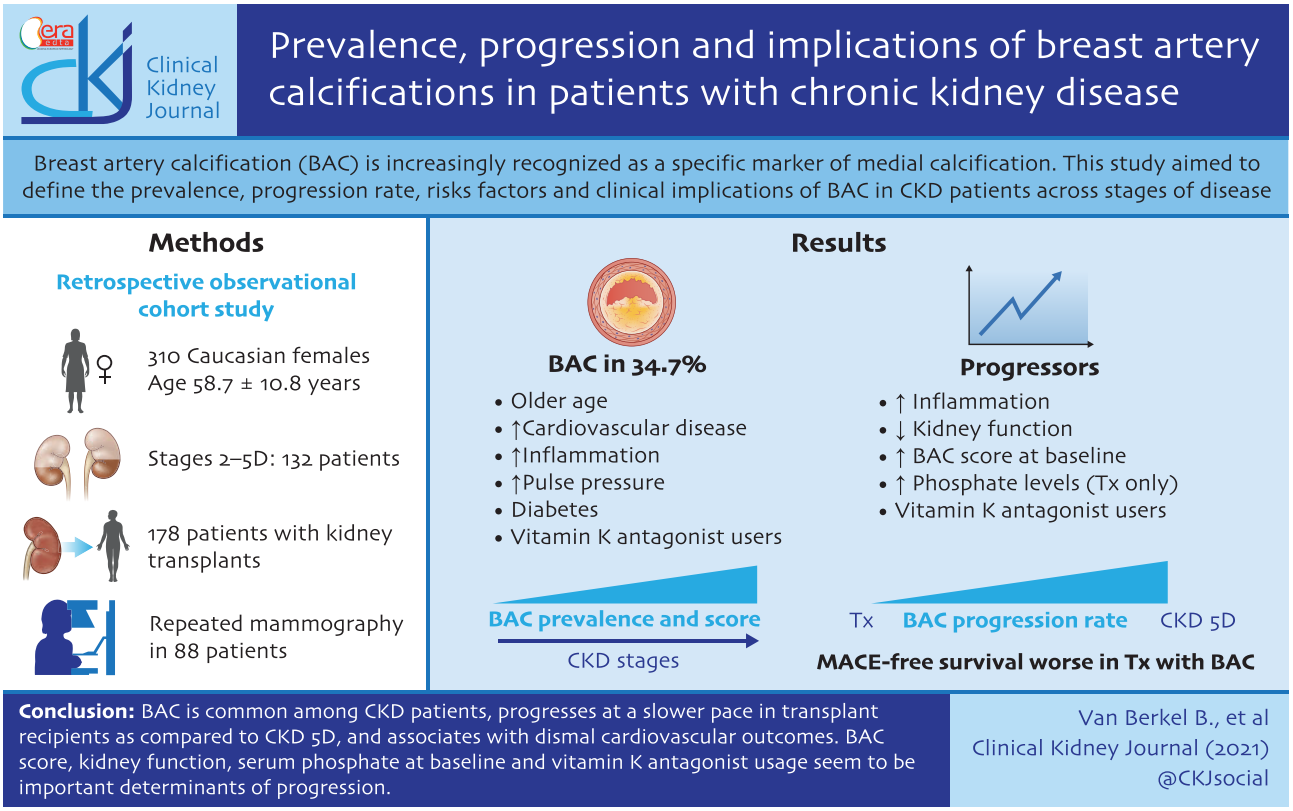
GRAPHICAL ABSTRACT



Keywords: chronic renal failure, kidney transplantation, mineral metabolism, phosphatemia, vascular calcification

INTRODUCTION

Vascular calcification (VC) is a common finding in patients with chronic kidney disease (CKD). Both prevalence and severity progress along with the decline of kidney function [1]. As in the general population, VC in CKD patients associates with dismal cardiovascular outcomes [2]. VC is an actively regulated biological process associated with crystallization of hydroxyapatite in the extracellular matrix and in cells of the media or intima of the arterial wall. Although intimal and medial calcification share many risk factors, pathophysiology and clinical implications differ substantially [3]. It appears that while deposition of hydroxyapatite represents the resulting commonality of VC, different initiating and propagating molecular mechanisms as well as diverse crystalline compositions of calcium apatite crystals may be present in various forms of VC [4]. Furthermore, the pathophysiology of medial calcification may vary across vascular beds, probably reflecting differences in the genetic origin of the vascular smooth muscle cells [5, 6]. In recent years, interest in breast arterial calcification (BAC) has increased. The reasons are 2-fold. First, on histologic examination, BAC was shown to be exclusively medial [7]. BAC may thus serve as a specific marker for medial calcification and its assessment may help identify specific risk factors [8, 9]. Second, measurement of BAC may offer a personalized, noninvasive approach to risk-stratify women for cardiovascular disease (CVD) at no additional cost or radiation since a majority of women >40 years of age undergo regular breast cancer screening with mammography.

Recent estimates are that 12.7% of women undergoing breast cancer screening have some degree of BAC. Factors associated with a higher prevalence include older age, diabetes mellitus, hypertension and CKD [10–12]. A significant association between BAC and coronary artery calcification has been reported in many, but not all studies [10, 13].

Studies exploring the prevalence and progression of BAC in CKD across stages of disease are scanty. Furthermore, only a few studies have explored the association between BAC and mineral metabolism disturbances and inflammation, i.e. wel- established risk factors of medial calcification. The present retrospective cross-sectional observational study aimed to fill these knowledge gaps. More specifically, we aimed to define the prevalence of BAC in CKD patients across stages of disease and to assess the association with parameters of mineral metabolism and inflammation. Furthermore, we explored the association between BAC and cardiovascular morbidity and mortality.

MATERIALS AND METHODS

Patient selection and data collection

Computer files of all women followed at the Division of Nephrology, University Hospitals Leuven, were screened for availability of mammography performed between 1 January 2006 and 1 January 2017 (Supplementary data, Figure S1). Since this cohort study aimed to study the association with laboratory parameters of mineral metabolism, only patients with parathyroid hormone (PTH) measurement available within 1 year of the date of the

mammography were eligible for inclusion. The date of the most recent PTH measurement within this time frame was defined as the index date. Patients were categorized according to transplant status [CKD vs. transplant (Tx) patients]. CKD patients were further categorized according to CKD stage (CKD G1–3, CKD G4–5 and CKD G5D). Relevant clinical [age, body mass index (BMI), smoking habits, comorbidity etc.] and laboratory data were retrieved from an automated (computerized) database. CVD was defined by a history of myocardial infarction or ischemia, coronary intervention, ischemic stroke or peripheral vascular disease. Patients with more than one data set available, separated by a time interval exceeding 6 months and without a change in transplant status within this period were enrolled in a longitudinal substudy that aimed at defining the natural history of BAC and its correlates. The clinical and research activities being reported are consistent with the principles of the Declaration of Istanbul as outlined in the Declaration of Istanbul on Organ Trafficking and Transplant Tourism. The study was performed in adherence with the Declaration of Helsinki and approved by the Ethics Committee Research UZ/KU Leuven.

Biochemistry

Serum creatinine, calcium (Ca), phosphate (Phos), alkaline phosphatases, C-reactive protein, total cholesterol, high-density lipoprotein (HDL) cholesterol and other parameters were determined by standard laboratory techniques. In dialysis patients, only predialysis determinations were recorded. Serum concentrations of PTH were determined by a 1–84 PTH assay [14]. The estimated glomerular filtration rate (eGFR) was calculated using the Chronic Kidney Disease Epidemiology Collaboration formula.

Mammography

Left and right digital mammograms (mediolateral oblique views) were screened for BAC by visual inspection on standard 5 megapixel mammography monitors. BAC appears as thin, parallel, linear densities along the margins of blood vessels and is easily distinguished from other ductal or parenchymal calcifications, which appear as tiny solitary densities (size varying between 150 and 30 μm), most often organized in clusters (Figure 1). Assessment was performed by two physicians (B.V.B. and C.V.O.), blinded to patient characteristics. Inter reader correlation, assessed on a random sample of 14 mammograms, was 0.91. The extent of BAC (i.e. the BAC score) was determined by summation of the lengths of BAC (mm) in both left and right mammograms, according to Trimboli *et al.* [15]. The change in BAC score over time (i.e. the BAC rate) was expressed as millimeters per year per breast. Patients were categorized according to presence of BAC (BAC^+ vs. BAC^-).

Outcomes

Kidney transplant recipients (KTRs) were followed from the date of the first mammography after transplantation until the first cardiovascular event, death or end of follow-up (defined as the date of the last visit prior to 25 March 2018). The primary endpoint was a major adverse cardiac event (MACE; coronary artery bypass surgery, percutaneous intervention or myocardial infarction, and/or electrocardiogram changes), cerebrovascular accidents or peripheral arterial disease (PAD; claudication with proven stenosis, vascular intervention) or cardiovascular death. The time to the first event was analyzed.

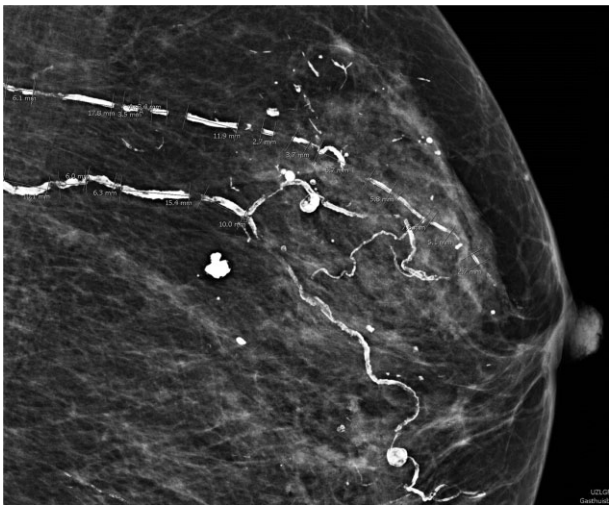


FIGURE 1: Oblique view mammogram (with demarcated/measured calcified vessels).

Statistics

Continuous parameters are expressed as mean ± standard deviation (SD) (in case of normality) or as median [interquartile range (IQR)] (for nonnormal data). eGFR in CKD 5D (dialysis) patients was arbitrarily set at 5 mL/min/1.73 m^2. Differences between two independent groups (BAC^+ vs. BAC^-) were analyzed using the two-sample *t*-test (when test assumptions are fulfilled) or Mann–Whitney U-test (nonparametric equivalent). To compare three or more groups (CKD categories), analysis of variance (ANOVA) (when test assumptions are fulfilled) or its nonparametric equivalent Kruskal–Wallis was used. Fisher's exact test was used to compare two (independent) proportions; the exact Pearson chi-squared test was used to test the association between categorical variables in case of more than two proportions. Linear regression analyzes were performed to identify independent determinants of BAC score (changes) in CKD and Tx patients. Variables presenting a skewed distribution were log-transformed to test the selected prognostic factors of overall survival. Overall survival analysis was performed using the time from the first mammography to the primary endpoint. Survival curves were computed by the Kaplan–Meier method and compared by the logrank test. Cox regression models were used for multivariable survival analyses. SAS version 9.4 (SAS Institute, Cary, NC, USA) was used for the statistical analysis. Two-sided P-values <0.05 were considered statistically significant.

RESULTS

Determinants of the BAC score

The final study cohort consisted of 310 females; 178 Tx recipients and 132 CKD patients. Relevant clinical and laboratory parameters are summarized in Table 1. The mean age was 58.8 ± 10.9 years (range 24–89). Twenty-four patients (7.7%) were treated with a vitamin K antagonist (VKA).

With 108 patients having BAC, the overall prevalence of BAC was 34.7%. BAC^+ patients were characterized by significantly older age, more CVD, higher pulse pressure and borderline higher inflammation and prevalence of diabetes (Table 1). The prevalence of BAC, moreover, was twice as high in VKA users compared with nonusers (66.7 vs. 32.2%; P = 0.0007).

Table 1. Cohort characteristics

Characteristics	Total ($N = 310$)	BAC$^-$ ($N = 202$)	BAC$^+$ ($N = 108$)	P-value[a]
Age (years), mean ± SD	58.8 ± 10.9	56.6 ± 10.9	62.7 ± 9.6	<0.0001
BMI (kg/m²), mean ± SD	25.1 ± 6.2 ($n = 307$)	24.9 ± 5.6	25.5 ± 7.0	0.44
CKD/Tx (%)	43/57	39/61	49/51	0.09
(Ever) smoked (%)	19.4	19.9	18.4	0.87
DM (%)	26.8	23.3	33.3	0.06
CVD (%)	10.0	5.9	17.6	0.0023
VKA (%)	7.7	4.0	14.8	0.0007
CRP (mg/dL), median (IQR)	2.3 (1.0–8.0) ($n = 259$)	2.1 (1.0–6.3)	3.0 (1.1–11.6)	0.051
Alkaline phosphotase (U/L), median (IQR)	123 (71–203) ($n = 284$)	57 (45–70)	147 (73–230)	0.031
SBP (mmHg), mean ± SD	136.1 ± 21.5 ($n = 264$)	135.2 ± 19.6	137.8 ± 24.6	0.36
DBP (mmHg), mean ± SD	76.3 ± 13.4 ($n = 264$)	77.2 ± 12.1	74.7 ± 15.4	0.17
MAP (mmHg), mean ± SD	96.3 ± 14.3 ($n = 264$)	96.5 ± 12.9	95.7 ± 16.4	0.68
PP (mmHg), mean ± SD	59.8 ± 17.9 ($n = 264$)	57.9 ± 16.3	63.1 ± 20.1	0.032
BAC score (mm), median (IQR)		0	76.4 (18.1–231.3)	NA

CRP: C-reactive protein; SBP: systolic blood pressure; DBP: diastolic blood pressure; MAP: mean arterial pressure; PP: pulse pressure.
[a]Two-sample *t*-test or Wilcoxon rank sum test for continuous data or Fisher's exact test for categorical data.

Table 2. Transplant patient characteristics

Characteristics	Tx ($N = 178$)	BAC$^-$ ($N = 123$)	BAC$^+$ ($N = 55$)	P-value[a]
Age (years)	57.1 ± 9.0	56.3 ± 9.0	59.9 ± 8.3	0.014
BMI (kg/m²)	25.0 ± 5.7	24.9 ± 5.4	25.2 ± 6.3	0.85
DM (%)	32.0	28.7	40	0.1
CVD (%)	10.7	7.4	18.2	0.03
VKA (%)	6.2	4.9	9.1	0.3
SBP (mmHg)	138.2 ± 19.4 ($n = 156$)	136.2 ± 19.4	142.6 ± 18.9	0.054
DBP (mmHg)	77.9 ± 12.7 ($n = 156$)	77.5 ± 11.5	78.6 ± 14.9	0.64
MAP (mmHg)	98.0 ± 12.9 ($n = 156$)	97.1 ± 12.5	99.9 ± 13.6	0.193
PP (mmHg)	60.3 ± 17.4 ($n = 156$)	58.6 ± 16.2	63.9 ± 19.3	0.075
eGFR (mL/min/1.73 m²), median (IQR)	38.7 (26.2–51.2)	47.7 ± 18.1	46.4 ± 20.8	0.687
Ca^{++} (mg/dL)	9.7 ± 0.9 ($n = 159$)	9.8 ± 0.7	9.7 ± 0.5	0.981
Phosphorus (mg/dL)	3.2 ± 0.7 ($n = 166$)	3.1 ± 0.6	3.5 ± 0.7	0.0017
Alkaline phosphotase (IU/L), median (IQR)	110.0 (70.0–167.0) ($n = 161$)	105.0 (70.0–148.0)	120.5 (68.5–208.5)	0.191
PTH (ng/L), median (IQR)	47.5 (26.1–87.4)	48.2 (26.2–83.9)	46.9 (24.0–106.0)	0.857
CRP (mg/dL)	8.6 ± 22.6 ($n = 140$)	1.8 (1.0–5.3)	2.5 (1.0–5.8)	0.378
Total cholesterol (mg/dL)	184.5 ± 39.8 ($n = 130$)	184.8 ± 37.8	184.1 ± 43.6	0.920
HDL cholesterol (mg/dL)	64.6 ± 17.4 ($n = 120$)	64.2 ± 17.4	65.3 ± 17.7	0.721
Triglycerides (mg/dL), median (IQR)	122 (91–164) ($n = 129$)	130 (91–167)	119 (96–159)	0.578
BAC score (mm), median (IQR)		0	73.1 ([18.8–248.6)	NA

Values are presented as mean ± SD unless stated otherwise. SBP: systolic blood pressure; DBP: diastolic blood pressure; MAP: mean arterial pressure; PP: pulse pressure; CRP: C-reactive protein; HDL: high-density lipoprotein.
[a]Two-sample *t*-test, Wilcoxon rank sum test or Fisher's exact test.

In the Tx subcohort, BAC$^+$ patients (30.9%) were characterized by significantly older age, more CVD, higher serum phosphate and borderline higher pulse pressure (Table 2). In linear regression analysis, older age and higher phosphate were identified as independent determinants of BAC score (Supplementary data, Table S1). Mammograms were obtained on average 5.6 years after transplantation.

In the CKD subcohort, BAC$^+$ patients (47.7%) were characterized by significantly older age, higher blood pressure and more CVD and were more often treated with a VKA (Table 3). In linear regression analysis, older age, history of CVD and treatment with VKA were identified as independent determinants of BAC score (Supplementary data, Table S2). The prevalence of BAC increases with the progression of CKD was CKD G1–3, 27.0%; CKD G4–5, 38.3%; CKD G5D, 52.1% ($P = 0.02$) by the Cochran–Armitage trend test (Figure 2). Median BAC scores (for BAC$^+$ patients) in

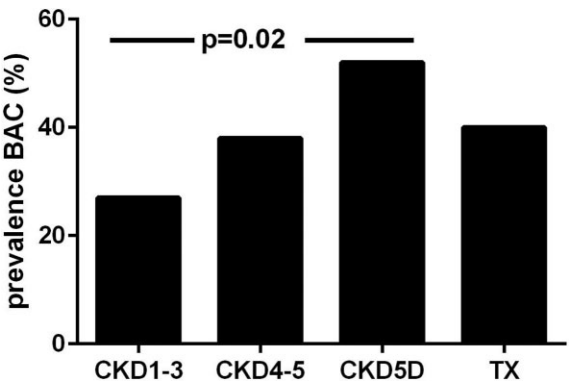


FIGURE 2: BAC prevalence according to CKD stage (Cochran–Armitage test for trend).

Table 3. CKD patients characteristics

Characteristics	CKD (N = 132)	BAC$^-$ (N = 79)	BAC$^+$ (N = 53)	P-value[a]
Age (years)	60.5 ± 12.8	57.1 ± 13.3	65.6 ± 10.1	<0.0001
BMI (kg/m²)	25.3 ± 6.7	24.8 ± 6.0	25.9 ± 7.7	0.39
DM (%)	19.7	15.2	26.4	0.12
CVD (%)	9.1	3.8	17.0	0.013
VKA (%)	9.9	2.5	20.8	0.0006
SBP (mmHg)	133.1 ± 24.0 (*n* = 108)	133.5 ± 20.0	132.6 ± 29.0	0.86
DBP (mmHg)	74.1 ± 14.2 (*n* = 108)	76.8 ± 13.1	70.3 ± 14.8	0.02
MAP (mmHg)	93.8 ± 15.7 (*n* = 108)	95.7 ± 13.7	91.1 ± 18.1	0.15
PP (mmHg)	59.0 ± 18.8 (*n* = 108)	56.7 ± 16.6	62.3 ± 21.1	0.13
eGFR[b](mL/min/1.73 m²), median (IQR)	16.5 (9.3–39.0)	17.1 (9.9–47.7)	15.1 (8.3–26.0)	0.32
Ca^{++} (mg/dL)	9.3 ± 0.8 (*n* = 128)	9.3 ± 0.6	9.4 ± 1.1	0.68
Phosphorus (mg/dL)	3.6 ± 1.0 (*n* = 128)	3.6 ± 1.0	3.6 ± 1.1	0.85
Alkaline phosphotase (IU/L), median (IQR)	145.0 (74.0–236.0) (*n* = 123)	123.0 (71.0–226.5)	168.0 (91.0–263.0)	0.12
PTH (ng/L), median (IQR)	68.8 (25.8–138.8)	59.8 (27.9–123.8)	85.8 (25.0–158.8)	0.30
CRP (mg/dL), median (IQR)	3.2 (1.0–11.4) (*n* = 119)	2.3 (1.0–7.6)	5.0 (1.5–12.4)	0.16
Total cholesterol (mg/dL)	174.5 ± 42.3 (*n* = 102)	178.3 ± 37.0	168.3 ± 49.6	0.28
HDL cholesterol (mg/dL)	61.0 ± 24.2 (*n* = 96)	64.8 ± 24.8	55.6 ± 22.6	0.068
Triglycerides (mg/dL), median (IQR)	118.0 (84.5–167.5) (*n* = 96)	108 (77–167)	124 (92–168)	0.30
BAC score (mm) (positive only)		0	81.2 (17.4–191.8)	NA

Values are presented as mean ± SD unless stated otherwise. DM: diabetes mellitus; SBP: systolic blood pressure; DBP: diastolic blood pressure; MAP: mean arterial pressure; PP: pulse pressure; CRP: C-reactive protein; HDL: high-density lipoprotein.
[a]Two-sample *t*-test or Wilcoxon rank sum test for continuous data and Fisher's exact test for categorical data. [b]bCKD-5D: eGFR arbitrarily set at 5 mL/min/1.73 m^2.

Table 4. Longitudinal substudy characteristics (*n* = 88)

Characteristics	All (N = 88)	Nonprogressors (N = 60)	Progressors (N = 28)	P-value[a]
Age (years)	57.4 ± 8.5	56.0 ± 7.4	60.6 ± 10.0	0.017
BMI (kg/m²)	24.6 ± 5.8	24.1 ± 5.1	25.5 ± 7.0	0.35
DM (%)	27.3	23.3	35.7	0.30
CVD (%)	10.2	8.3	14.3	0.46
VKA (%)	10.2	5.0	21.4	0.02
SBP (mmHg)	134.3 ± 19.0 (*n* = 72)	135.2 ± 19.0	132.4 ± 19.4	0.57
DBP (mmHg)	75.9 ± 12.4 (*n* = 72)	76.8 ± 10.9	74.1 ± 15.1	0.45
eGFR (mL/min/1.73 m²)	44.3 (25.8–54.8)	48.2 (32.5–58.2)	34.1 (10.1–45.5)	0.0019
Ca^{++} (mg/dL)	9.6 ± 0.8 (*n* = 80)	9.7 ± 0.7	9.4 ± 0.8	0.15
Phosphorus (mg/dL)	3.3 ± 0.8 (*n* = 84)	3.2 ± 0.7	3.4 ± 1.0	0.33
PTH (ng/L)	40.5 (23.6–71.5)	36.6 (18.9–67.4)	43.4 (26.2–132.7)	0.31
CRP (mg/dL)	2.6 (1.0–11.5) (*n* = 68)	2.1 (0.9–6.5)	5.8 (2.1–23.1)	0.021
Total cholesterol (mg/dL)	180.4 ± 41.2 (*n* = 61)	188.4 ± 38.3	167.1 ± 43.1	0.049
HDL cholesterol (mg/dL)	64.9 ± 20.0 (*n* = 53)	65.6 ± 20.2	63.9 ± 20.1	0.77
Triglycerides (mg/dL)	137.0 (103.0–167.0) (*n* = 53)	150.0 (103.0–173.0)	123.5 (88.0–150.0)	0.29
BAC score (mm) (gain only)		0	63.2 (15.1–227.3)	<0.001

Values are presented as mean ± SD unless stated otherwise. DM: diabetes mellitus; SBP: systolic blood pressure; DBP: diastolic blood pressure; CRP: C-reactive protein; HDL: high-density lipoprotein.
[a]Two-sample *t*-test or Wilcoxon rank sum test for continuous variables and Fisher's exact test for categorical variables.

these groups were 37.1 mm (IQR 10.8–81.2), 56.2 (IQR 4.9–131.6) and 166.1 (IQR 24.0–327.1), respectively (P = 0.05).

Determinants of BAC progression

In a subset of 88 patients (Tx, *n* = 74; CKD G5D, *n* = 14), a repeat mammography was performed as part of routine practice after a mean interval of 3.5 ± 2.2 years (Table 4). The mean annualized BAC rate was 1.2 ± 0.4 mm in the overall cohort (*n* = 88) and 6.6 ± 2.2 mm after exclusion of 54 patients free of BAC both at baseline and follow-up. The mean annualized BAC rate was significantly lower in Tx patients as compared with CKD G5D patients (1.0 ± 0.4 vs. 2.2 ± 1.2 mm; P = 0.02, overall cohort). This significance was lost when patients free of BAC at baseline and follow-up were excluded (6.6 vs. 7.8 mm/year/breast, mean TX, vs. CKD G5D). Lacking data on the least significant change, patients with increasing BAC scores were arbitrarily defined as 'progressors'. A total of 25% and 36% of Tx and CKD patients, respectively, were classified as progressors (P = 0.005). In the overall cohort (*n* = 88), progressors were characterized by older age, more inflammation, worse kidney function and higher baseline BAC score and were more often treated with a VKA. In the Tx subcohort (*n* = 74), progressors were characterized by lower eGFR, higher serum phosphate, higher baseline BAC score and more diabetes.

BAC and major cardiovascular outcome after kidney transplantation

The association between BAC and outcomes was investigated in the subset of KTRs. Thirty-five KTRs (18.5%) died during follow-up (median 5 years); causes of death were as follows:

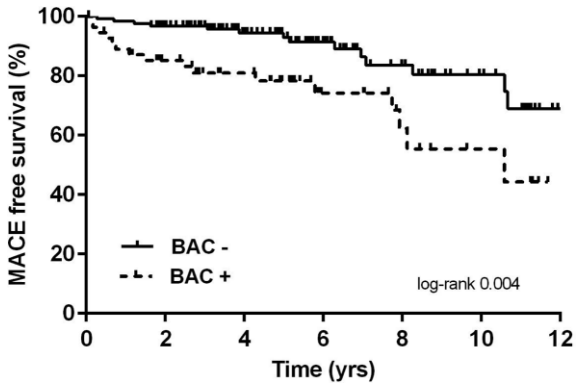


FIGURE 3: Kaplan–Meier survival curve in kidney transplant recipients with MACE as the composite endpoint.

cardiovascular, $n = 7$; infectious, $n = 9$; malignant disease, $n = 7$; miscellaneous or unknown, $n = 12$. MACEs occurred in 30 patients (fatal cardiovascular event, $n = 6$; nonfatal cardiac event, $n = 15$; nonfatal cerebrovascular event, $n = 9$, 1 prior to a fatal cardiovascular event). Patients who reached the primary endpoint during follow-up were significantly older, had a more frequent history of CVD, had higher levels of phosphate and had a higher BAC score (Supplementary data, Table S2). MACE-free survival was significantly worse in Tx patients with BAC [log rank 0.0036, HR 2.9 (IQR 1.42–5.96)] (Figure 3). In a backward or forward selection procedure, with age, blood pressure, history of CVD, diabetes, alkaline phosphatase, eGFR and BAC, the final Cox regression model retained BAC as the only significant predictor of MACEs.

In univariate logistic regression (such a model does not take into account the time to event), the traditional risk factors (age, diabetes, CVD, blood pressure and eGFR) failed to associate with MACEs, contrary to BAC ($P = 0.0047$) and alkaline phosphatase ($P = 0.036$). A (backward/forward) logistic regression model for MACEs using the same covariates resulted in a model with BAC [odds ratio (OR 2.50 (IQR 1.07–5.84); $P = 0.034$] and alkaline phosphatase [OR 1.035 (IQR 1.003–1.068) for a 10-unit change; $P = 0.034$] as significant predictors for MACEs.

DISCUSSION

The main findings of the present observational study are as follows: BAC is common in CKD patients and KTRs, hyperphosphatemia is a (nontraditional) risk factor for BAC in KTRs and BAC associates with dismal cardiovascular outcomes in KTRs.

BAC prevalence is well known to vary widely depending on the population's age and comorbidities. Literature data show BAC prevalence ranging from 10–12% in healthier population-based cohort studies [11, 16, 17] to 60–70% among women >70 years of age [18] or with CKD [9, 19]. In the present cohort of CKD patients, the overall prevalence of BAC was 34.7%. This prevalence is twice as high as reported in age-matched healthy controls [18]. Patients with BAC were characterized by significantly older age, more CVD, higher pulse pressure and borderline more inflammation and higher prevalence of diabetes. These findings are sound from a pathophysiological perspective and align with data from the general population [10, 16, 18, 20]. The higher pulse pressure in patients with BAC most probably is a reflection of increased arterial stiffness, commonly observed in patients with medial arterial calcification [3]. In line with previous reports [21], we also identified VKA usage as an independent determinant of BAC. This association of VKA usage with medial arterial calcification is consistent with inhibition of matrix Gla protein (MGP), a vitamin K–dependent inhibitor of VC that is synthesized by vascular smooth muscle cells [22, 23].

In the subcohort of patients with CKD G2–5D, BAC prevalence increased parallel to the severity of kidney dysfunction (from 27 to 52%). This finding aligns with literature data [9, 19] and confirms that medial calcification at least partly accounts for the increased VC burden in CKD [1]. The BAC score was >3-fold higher in dialysis patients as compared with patients with earlier-stage CKD, consistent with the notion that advanced CKD, and especially dialysis, is a catalyzer of medial calcification [24–26]. The prevalence of BAC in CKD G5D patients (52%) was slightly lower than previously reported in similar cohorts [8, 9, 27, 28]. Most probably, differences in measurement techniques, with digital mammography being more accurate than plain mammography, and case mix account for the variability.

BAC prevalence in KTRs was 30.9%, which is substantially lower than the prevalence observed in patients with CKD G5D. This should not come as a surprise, as kidney Tx patients are younger and among the fittest of patients with end-stage kidney disease. KTRs with BAC were characterized by older age, more CVD and higher serum phosphate as compared with counterparts without BAC. In linear regression analysis, older age and higher phosphate were identified as independent determinants of BAC score.

In a subset of patients we were able to assess the natural history of BAC. The median annualized change of BAC amounted to 2.2 mm per breast in CKD 5D patients, which is significantly higher than 1.0 mm per breast in KTRs. A higher proportion of CKD 5D patients as compared with KTRs showed progression. This finding aligns with recent data by Alappan *et al.* [29], showing a slowing of the progression of BAC in KTRs. Of note, progression rates differ among cohorts, most probably reflecting case mix and analytical issues. Since the assumption of linearity of progression of BAC is unproven, we preferred, as opposed to Alappan *et al.*, not to exclude patients free of calcification on the first mammogram.

Among KTRs, progressors were characterized by poorer graft function, higher serum phosphate, higher baseline BAC score at baseline and more diabetes. This finding confirms and extends data from the Brussels Renal Transplant Cohort study [30]. In the latter study, the natural history and determinants of aorta and coronary artery calcifications were studied in prevalent KTRs by means of multislice spiral computed tomography. Baseline VC and serum phosphate were identified as independent determinants of VC progression. Individuals without VC at baseline rarely showed progression. The decisive impact of baseline VC on progression in present and previous studies supports the thesis that VC might itself induce further inflammation and calcification in a positive feedback loop [31, 32]. The observation of an independent association between serum phosphate levels and progression of VC is consistent with current notions of the biological roles of a disturbed phosphate metabolism in the pathogenesis of VC [33].

In the overall cohort, progressors were more often treated with a VKA and were characterized by more inflammation. The latter finding confirms the previously described relationship between VC and markers of inflammation in CKD patients [34, 35] as well as in the general population [36] and in diabetics [37]. Underlying pathophysiological mechanisms are complex and multiple. Inflammatory cytokines can promote vascular smooth muscle cell calcification [38], in part through activation of Msx2-Wnt/β-catenin signalling [39]. Moreover,

inflammatory cytokines may promote the process of endothelial–mesenchymal transition, leading to osteogenic gene expression and cytokine production by endothelial cells [40]. Finally, inflammation may repress the important calcification inhibitor fetuin-A [41].

In KTRs, BAC was identified as a predictor of MACEs, defined as the composite of nonfatal cardiovascular, cerebrovascular and fatal cardiovascular events. Of note, traditional risk factors failed to associate with MACEs, even in univariate logistic regression analysis. BAC most probably is a more robust risk marker, as it integrates/recapitulates various traditional and nontraditional cardiovascular risk factors such as older age, higher blood pressure and inflammation. The identification of VC as an independent predictor of dismal cardiovascular outcome in the present study aligns with data from a comparable study in 281 prevalent KTRs showing a strong association between coronary artery calcification and cardiovascular outcomes [42]. Also in the general population, BAC predicts CV outcomes better than traditional risk factors.

Besides BAC, alkaline phosphatase was identified as an independent predictor of MACEs in logistic regression analysis. The latter observation confirms and extends data from previous cohort studies linking elevated alkaline phosphatase levels to increased residual cardiovascular risk [43] and mortality [44].

This study presents significant strengths. These include a relatively large sample size, availability of laboratory parameters of mineral metabolism and inflammation, and serial mammograms and outcomes in a substantial subset of patients. Finally, we used full-field digital mammography to evaluate the calcifications. This technique is superior to screen-film mammography because of its better resolution and possibility to adjust contrast.

In conclusion, BAC is common among CKD patients, progresses at a slower pace in KTRs as compared with CKD G5D and associates with dismal (cardiovascular) outcomes. BAC score, kidney function and serum phosphate at baseline seem to be important determinants of progression. Measurement of BAC may offer a personalized, noninvasive approach to risk-stratify CKD patients for CVD at no additional cost or radiation since a majority of women >40 years of age undergo regular breast cancer screening.

SUPPLEMENTARY DATA

Supplementary data are available at *ckj* online.

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CONFLICT OF INTEREST STATEMENT

None declared.

DATA AVAILABILITY STATEMENT

The data underlying this article cannot be shared publicly to protect the privacy of individuals who participated in the study. The data will be shared on reasonable request to the corresponding author.

REFERENCES

1. Budoff MJ, Rader DJ, Reilly MP *et al*. Relationship of estimated GFR and coronary artery calcification in the CRIC (Chronic renal insufficiency cohort) study. *Am J Kidney Dis* 2011; 58: 519–526
2. Chen J, Budoff MJ, Reilly MP *et al*. Coronary artery calcification and risk of cardiovascular disease and death among patients with chronic kidney disease. *JAMA Cardiol* 2017; 2: 635–643
3. Vervloet M, Cozzolino M. Vascular calcification in chronic kidney disease: different bricks in the wall? *Kidney Int* 2017; 91: 808–817
4. Lanzer P, Boehm M, Sorribas V *et al*. Medial vascular calcification revisited: review and perspectives. *Eur Heart J* 2014; 35: 1515–1525
5. Kirsch AH, Kirsch A, Artinger K *et al*. Heterogeneous susceptibility for uraemic media calcification and concomitant inflammation within the arterial tree. *Nephrol Dial Transplant* 2015; 30: 1995–2005
6. Schlieper G. Vascular calcification in chronic kidney disease: not all arteries are created equal. *Kidney Int* 2014; 85: 501–503
7. O'Neill WC, Adams AL. Breast arterial calcification in chronic kidney disease: absence of smooth muscle apoptosis and osteogenic transdifferentiation. *Kidney Int* 2014; 85: 668–676
8. Abou-Hassan N, Tantisattamo E, D'Orsi ET *et al*. The clinical significance of medial arterial calcification in end-stage renal disease in women. *Kidney Int* 2015; 87: 195–199
9. Duhn V, D'Orsi ET, Johnson S *et al*. Breast arterial calcification: a marker of medial vascular calcification in chronic kidney disease. *Clin J Am Soc Nephrol* 2011; 6: 377–382
10. Margolies L, Salvatore M, Hecht HS *et al*. Digital mammography and screening for coronary artery disease. *JACC Cardiovasc Imaging* 2016; 9: 350–360
11. Hendriks EJ, de Jong PA, van der Graaf Y *et al*. Breast arterial calcifications: a systematic review and meta-analysis of their determinants and their association with cardiovascular events. *Atherosclerosis* 2015; 239: 11–20
12. Chadashvili T, Litmanovich D, Hall F *et al*. Do breast arterial calcifications on mammography predict elevated risk of coronary artery disease? *Eur J Radiol* 2016; 85: 1121–1124
13. Mostafavi L, Marfori W, Arellano C *et al*. Prevalence of coronary artery disease evaluated by coronary CT angiography in women with mammographically detected breast arterial calcifications. *PLoS One* 2015; 10: e0122289
14. Bouillon R, Coopmans W, Degroote DE *et al*. Immunoradiometric assay of parathyrin with polyclonal and monoclonal region-specific antibodies. *Clin Chem* 1990; 36: 271–276
15. Trimboli RM, Codari M, Bert A *et al*. Breast arterial calcifications on mammography: intra- and inter-observer reproducibility of a semi-automatic quantification tool. *Radiol Med* 2018; 123: 168–173
16. van Noord PA, Beijerinck D, Kemmeren JM *et al*. Mammograms may convey more than breast cancer risk: breast arterial calcification and arterio-sclerotic related diseases in women of the DOM cohort. *Eur J Cancer Prev* 1996; 5: 483–487
17. Kataoka M, Warren R, Luben R *et al*. How predictive is breast arterial calcification of cardiovascular disease and risk factors when found at screening mammography? *AJR Am J Roentgenol* 2006; 187: 73–80
18. Iribarren C, Sanchez G, Husson G *et al*. MultIethNic study of BrEast ARterial calcium gradation and cardiovascular disease: cohort recruitment and baseline characteristics. *Ann Epidemiol* 2018; 28: 41–47

19. Hassan NA, D'Orsi ET, D'Orsi CJ *et al*. The risk for medial arterial calcification in CKD. *Clin J Am Soc Nephrol* 2012; 7: 275–279
20. Ferreira JA, Pompei LM, Fernandes CE *et al*. Breast arterial calcification is a predictive factor of cardiovascular disease in Brazilian postmenopausal women. *Climacteric* 2009; 12: 439–444
21. Tantisattamo E, Han KH, O'Neill WC. Increased vascular calcification in patients receiving warfarin. *Arterioscler Thromb Vasc Biol* 2015; 35: 237–242
22. Lomashvili KA, Wang X, Wallin R *et al*. Matrix Gla protein metabolism in vascular smooth muscle and role in uremic vascular calcification. *J Biol Chem* 2011; 286: 28715–28722
23. Luo G, Ducy P, McKee MD *et al*. Spontaneous calcification of arteries and cartilage in mice lacking matrix GLA protein. *Nature* 1997; 386: 78–81
24. Shroff RC, McNair R, Figg N *et al*. Dialysis accelerates medial vascular calcification in part by triggering smooth muscle cell apoptosis. *Circulation* 2008; 118: 1748–1757
25. Parikh RV, Iribarren C, Lee C *et al*. Kidney function, proteinuria and breast arterial calcification in women without clinical cardiovascular disease: the MINERVA study. *PLoS One* 2019; 14: e0210973
26. Manzoor S, Ahmed S, Ali A *et al*. Progression of medial arterial calcification in CKD. *Kidney Int Rep* 2018; 3: 1328–1335
27. Castellanos M, Varma S, Ahern K *et al*. Increased breast calcifications in women with ESRD on dialysis: implications for breast cancer screening. *Am J Kidney Dis* 2006; 48: 301–306
28. Sommer G, Kopsa H, Zazgornik J *et al*. Breast calcifications in renal hyperparathyroidism. *AJR Am J Roentgenol* 1987; 148: 855–857
29. Alappan HR, Vasanth P, Manzoor S *et al*. Vascular calcification slows but does not regress after kidney transplantation. *Kidney Int Rep* 2020; 5: 2212–2217
30. Evenepoel P, Goffin E, Meijers B *et al*. Sclerostin serum levels and vascular calcification progression in prevalent renal transplant recipients. *J Clin Endocrinol Metab* 2015; 100: 4669–4676
31. Yoon HC, Emerick AM, Hill JA *et al*. Calcium begets calcium: progression of coronary artery calcification in asymptomatic subjects. *Radiology* 2002; 224: 236–241
32. Nadra I, Mason JC, Philippidis P *et al*. Proinflammatory activation of macrophages by basic calcium phosphate crystals via protein kinase c and MAP kinase pathways: a vicious cycle of inflammation and arterial calcification? *Circ Res* 2005; 96: 1248–1256
33. Shanahan CM, Crouthamel MH, Kapustin A *et al*. Arterial calcification in chronic kidney disease: key roles for calcium and phosphate. *Circ Res* 2011; 109: 697–711
34. Guerin AP, London GM, Marchais SJ *et al*. Arterial stiffening and vascular calcifications in end-stage renal disease. *Nephrol Dial Transplant* 2000; 15: 1014–1021
35. Viaene L, Behets GJ, Heye S *et al*. Inflammation and the bone-vascular axis in end-stage renal disease. *Osteoporos Int* 2016; 27: 489–497
36. Jenny NS, Brown ER, Detrano R *et al*. Associations of inflammatory markers with coronary artery calcification: results from the multi-ethnic study of atherosclerosis. *Atherosclerosis* 2010; 209: 226–229
37. Alman AC, Kinney GL, Tracy RP *et al*. Prospective association between inflammatory markers and progression of coronary artery calcification in adults with and without type 1 diabetes. *Diabetes Care* 2013; 36: 1967–1973
38. Tintut Y, Patel J, Territo M *et al*. Monocyte/macrophage regulation of vascular calcification in vitro. *Circulation* 2002; 105: 650–655
39. Shao JS, Aly ZA, Lai CF *et al*. Vascular Bmp Msx2 Wnt signaling and oxidative stress in arterial calcification. *Ann N Y Acad Sci* 2007; 1117: 40–50
40. Khosla S. The bone and beyond: a shift in calcium. *Nat Med* 2011; 17: 430–431
41. Ketteler M, Bongartz P, Westenfeld R *et al*. Association of low fetuin-A (AHSG) concentrations in serum with cardiovascular mortality in patients on dialysis: a cross-sectional study. *Lancet* 2003; 361: 827–833
42. Nguyen PT, Henrard S, Coche E *et al*. Coronary artery calcification: a strong predictor of cardiovascular events in renal transplant recipients. *Nephrol Dial Transplant* 2010; 25: 3773–3778
43. Haarhaus M, Ray KK, Nicholls SJ *et al*. Apabetalone lowers serum alkaline phosphatase and improves cardiovascular risk in patients with cardiovascular disease. *Atherosclerosis* 2019; 290: 59–65
44. Drechsler C, Verduijn M, Pilz S *et al*. Bone alkaline phosphatase and mortality in dialysis patients. *Clin J Am Soc Nephrol* 2011; 6: 1752–1759