Vascular Calcification in Patients with Chronic Kidney Disease

Santo Dellegrottaglie Javier Sanz Sanjay Rajagopalan

Zena and Michael A. Wiener Cardiovascular Institute and Marie-Josée and Henry R. Kravis Center for Cardiovascular Health, Mount Sinai Medical Center, New York, N.Y., USA

Key Words

Vascular calcification · Chronic kidney disease · End-stage renal disease · Cardiovascular disease · Atherosclerosis

Abstract

Chronic kidney disease (CKD) represents an extremely common condition, and cardiovascular diseases are frequently reported in this patient population. Traditional risk factors are not accurate prognostic predictors in CKD patients, and new potential markers to predict the cardiovascular involvement in uremic patients need to be identified. Vascular calcification (VC) represents a hallmark of the atherosclerotic process in CKD. This review summarizes the processes responsible for VC (particularly focusing on the mechanisms operative in the presence of renal dysfunction), discusses the utility of computer tomography modalities in the detection of VC in patients with CKD, and reports the potential role of VC as pathophysiological link between kidney disease and cardiovascular events.

Introduction

Chronic kidney disease (CKD) is globally emerging as an important risk factor for morbidity and mortality. The progressive aging of the global population and the escalating numbers of patients with type 2 diabetes mellitus are clearly major factors responsible for its growing significance [1]. As many as 6–11% of the adult population could have some degree of CKD, with a number of these patients progressing to end-stage renal disease (ESRD) [2]. The preponderance of subjects with CKD are more likely to die of cardiovascular disease than to go on to ESRD requiring renal replacement therapy [3]. Therefore, CKD is not surprisingly an independent risk factor for cardiovascular events, particularly in higher-risk populations [4–6]. Recently, an independent and graded association between reduced renal function and risk of cardiovascular events has been demonstrated in a single large cohort of subjects (approximately 1,000,000 individuals) [7]. A higher incidence of traditional cardiovascular risk factors, such as older age, hypertension, dyslipidemia and diabetes, in combination with risk factors specific to CKD (i.e. albuminuria, anemia, abnormal calcium/phosphate metabolism, extracellular fluid volume

Copyright © 2006 S. Karger AG, Basel

KARGER

Fax +41 61 306 12 34
E-Mail karger@karger.ch
www.karger.com

© 2006 S. Karger AG, Basel
0253–5068/06/0241–0056\$23.50/0

Accessible online at:
www.karger.com/bpu

Sanjay Rajagopalan, MD
Cardiovascular Institute, Mount Sinai Medical Center
Box 1030, One Gustave L. Levy Place
New York, NY 10029-6574 (USA)
Tel. +1 212 241 7911, Fax +1 212 423 9488, E-Mail sanjay.rajagopalan@mssm.edu

overload, electrolyte imbalance) may partly explain the very high prevalence of cardiovascular disease in these patients [8]. Additionally, it has been hypothesized that a number of pathophysiologic variables that are driven by the 'uremic milieu', such as oxidative stress, inflammation and endothelial dysfunction, may potentially promote atherosclerosis. A hallmark of the atherosclerotic process in CKD is vascular calcification (VC) [9, 10]. In view of the higher predilection of CKD patients for VC, measures of VC have been hypothesized to serve as non-invasive surrogates, especially in light of the fact that traditional risk factors are not good predictors in the CKD patient population.

This article briefly summarizes the state of the art of our knowledge regarding the mechanisms responsible for VC and its detection in patients with CKD and ESRD.

Mechanisms of VC in CKD

VC is observed either in the tunica intima (atherosclerosis) or in the tunica media. Medial calcification (Mönckeberg's sclerosis) is particularly common in patients with ESRD and CKD and may occur independent of atherosclerosis, implying different etiological mechanisms from intimal calcification [11]. Medial wall calcification increases vascular stiffness and reduces vascular compliance. Reduction in compliance (reflected as an increase in pulse wave velocity) may contribute to an increase in systolic blood pressure, loss of diastolic augmentation (fall in diastolic blood pressure) and rise in pulse pressure. Not surprisingly, the amount of coronary calcium correlates with arterial stiffness and the extent of calcification in the abdominal aorta in dialysis patients [12]. In atherosclerosis, VC probably occurs predominantly in the intima and through most stages of atherosclerosis, although it is lacking in the very early 'fatty streak' stage [13].

The impact of VC on plaque vulnerability is controversial, with some studies demonstrating an increase in stability and some noting increased vulnerability. To some extent, this may relate to the stage of atherosclerosis and the extent of calcification, with the larger more heavily calcified lesions reflecting lesions at an advanced state lacking the extent of inflammation and lipid core seen in earlier plaques [14, 15]. An interesting hypothesis that may potentially explain the somewhat paradoxical increased and decreased association of VC with vulnerability is the possibility that VC may confer a biphasic risk depending on the stage of atherosclerosis and the extent

of VC. Model calculations support an increased wall stress in the transition areas between the calcified plaques and the adjacent non-involved areas [16]. As the degree of calcification increases, the extension of the transition area between rigid and distensible plaque would initially increase until the point at which the calcified plaques coalesce. Calcification beyond this point may theoretically reduce transition zones, result in lower wall stress and may be associated with a lower risk of plaque rupture. Thus, it could be hypothesized that the most relevant prognostic parameter may not be the extent of VC but rather the 'total transition area' [17]. The ability to arrive at such a metric in the clinical realm remains unrealistic at this point.

Molecular Potentiators of VC

VC has been traditionally considered a passive process, associated with atherosclerosis or aging. However, recent evidence refutes this notion and supports the concept that VC is a dynamically regulated process [18–20]. A plethora of genes and proteins that normally function as key modulators of bone and mineral metabolism are involved, either directly or indirectly in the process of VC [18, 21]. Bone-related proteins, such as osteonectin, parathyroid hormone, parathyroid hormone-related peptide and bone morphogenetic protein 7, are expressed in the atherosclerotic plaques as well as at sites of medial arterial calcification [20–24]. Osteoprotegerin (OPG), a member of the tumor necrosis factor- α receptor family with inhibitory effects on osteoclastogenesis, introduces another link between bone and vascular metabolism [25]. OPG is a soluble molecule that binds and inhibits the ligand for receptor activator of nuclear factor- κ B, a tumor necrosis factor- α superfamily member demonstrated in the vasculature and essential for the maturation of osteoclast progenitors [26]. In agreement with the permissive role of OPG in VC, serum levels of OPG are elevated in patients with ESRD [27]. Vascular smooth muscle cells (VSMCs) and possibly vascular endothelial cells can be induced to transform into an osteoblast-like phenotype in vitro [28]. VSMCs and osteoblasts derive from a common mesenchymal precursor cell, and core binding factor (Cbfa)-1 is a transcription factor thought to be responsible for this switch that turns this precursor to the osteoblast phenotype [29].

Phosphate levels were long thought to influence mineralization only through physicochemical means. However, new evidence indicates that phosphate regulates and coordinates cell signaling and gene expression by dynamic transport processes. Primary cultures of human VSMCs

express bone proteins and mineralize when treated with β -glycerophosphate, which serves as an inorganic phosphate donor in the presence of alkaline phosphatase [28]. High intracellular phosphate downregulates typical VSMC genes and induces osteoblastic-like phenotypic changes of VSMCs. In particular, phosphate stimulates the expression of alkaline phosphatase on their surface and the production of Cbfa-1 and calcium-binding proteins, such as osteocalcin and osteopontin (OPN) [30, 31]. Recent studies demonstrate that Cbfa-1, alkaline phosphatase and OPN are present in calcified arteries, but are absent from the vessel wall of non-calcified arteries [19, 20].

Molecular Inhibitors of Calcification

Plasma components (such as citrate and magnesium) play a key physiological role by maintaining mineral in solution. Specific proteins may serve an inhibitory function and prevent ectopic calcification. For instance, fetuin-A, matrix Gla protein (MGP) and OPN are important inhibitors of calcification *in vivo*. Serum levels of fetuin-A are significantly reduced in subjects with renal failure [32, 33]. Circulating levels of fetuin-A decline with increasing inflammation in patients on dialysis and may additionally provide a link between inflammation and propensity for VC [32]. MGP is a small-size protein and is found in normal arterial wall and appears to be upregulated in atherosclerotic plaques [21]. The MGP knockout mouse has extensive aortic calcification, and recent evidence indicates that MGP inhibits mesenchymal cell differentiation to osteogenic phenotype [34, 35]. OPN is another important negative regulator of calcification and is expressed by macrophages, smooth muscle cells and endothelial cells [36–38], and is found to colocalize with these cell types in calcified atherosclerotic plaques. Mice deficient in both OPN and MGP have accelerated aortic calcification compared with mice deficient in only MGP, consistent with the concept that OPN inhibits mineralization [39].

The high prevalence of traditional risk factors for atherosclerosis combined with uremia-specific factors may be responsible for the increased VC observed in these patients. Inflammation in CKD may play a central role in the predisposition of patients with renal failure to VC [22]. Inflammation is a fundamental component of the atherosclerotic process and high level of serum inflammatory markers (i.e. C-reactive protein) have been reported in patients with ESRD [40]. The role of inflammation in VC is further strengthened by recent observations linking levels of circulating inhibitors of VC, such as fetuin-A and OPN, with increase inflammatory markers [32].

Role of Calcium and Phosphate Flux in VC

Recent evidence suggests that abnormalities in calcium and phosphorus metabolism, including therapeutic interventions affecting total body calcium balance, may also influence the development and progression of VC. Thus, it has been postulated that accelerated VC at the medial level may be favored (especially in ESRD patients) by the putative systemic calcium load arising from the use of calcium-containing phosphate binders and the use of high concentrations of calcium salts in the dialysate [41]. This may be distinct from the concomitant intimal VC that may occur in the same patients through disparate mechanisms. In direct agreement with these hypotheses is the correlation between VC and the calcium-phosphate product observed in studies involving young patients with ESRD (who have predominantly medial VC) but not in studies including older ESRD subjects who may have concomitant intimal VC [12, 42–44].

Detection and Significance of VC in CKD and ESRD

Although various methods have been proposed, electron beam computed tomography (EBCT) and multi-detector computer tomography (MDCT) represent the most accurate techniques available for calcium quantification. EBCT has been applied for more than a decade in the detection and quantification of cardiac and VC with good results and is currently considered the gold standard [45]. However, most medical centers do not have access to EBCT scanners due to relatively high costs and limited applications other than the calcium scoring. MDCT scanners which are more widely available and can be employed for a number of cardiac and non-cardiac applications are therefore more practical for the assessment of VC [46]. A number of studies have directly compared EBCT and MDCT (4- and 16-slice), demonstrating excellent correlation between these techniques for calcium quantification [47–49]. There are three different methods of calcium quantification and scoring: the Agatston method, the volumetric method and the mass method [50–52]. The Agatston score, although the most commonly used index to date, has the worst reproducibility (inter-scan). Reproducibility appears to be intermediate for the volumetric approach and best for the mass method [53].

The presence of coronary calcium, although a sensitive test for the presence of coronary artery disease, is not specific for prediction of ‘significant’ coronary lesions [54, 55]. Coronary calcification closely correlates with the

Table 1. Studies using EBCT or MDCT to assess coronary calcium score in patients with renal failure

Reference	Study population	Imaging modality	Correlates for coronary calcium score				
			mineral metabolism				others
			Ca	P	Ca-P product	PTH	
Braun et al. [66], 1996	49 HD	EBCT	–	–	NA	–	age, hypertension
Goodmann et al. [42], 2000	39 HD	EBCT	–	–	+	–	age, body mass index, cholesterolemia, dialysis vintage
Oh et al. [43], 2002	39 ESRD (13 HD + 26 RT)	EBCT	–	–	+	+	C-reactive protein, homocysteine
Raggi et al. [67], 2002	205 HD	EBCT	+	+	–	–	age, DM
Moe et al. [44], 2003	55 ESRD (33 HD + 38 RT)	MDCT	–	–	–	–	age, longer dialysis
Haydar et al. [78], 2004	46 HD	EBCT	NA	NA	NA	NA	angiographic coronary artery disease
Mehrotra et al. [69], 2004	60 CKD + DM	EBCD	–	–	–	–	age, creatinine, GFR
Nitta et al. [12], 2004	53 HD	MDCT	–	–	–	–	age, C-reactive protein

PTH = Parathyroid hormone; HD = hemodialysis; NA = not assessed; RT = renal transplantation; DM = diabetes mellitus; GFR = glomerular filtration rate.

presence of calcium in the aorta (at thoracic and abdominal levels) and in the aortic valve, suggesting common pathophysiologic mechanisms for this process, be it in the valve or in conduit arteries [56, 57]. According to published guidelines, the coronary calcium score may help in identifying asymptomatic patients at low-to-intermediate risk, who may benefit from more aggressive risk factor modification [45]. In recent years, several studies have investigated the prognostic value of calcium scoring [58]. As expected, coronary VC detected by EBCT was strongly associated with all-cause mortality [59, 60]. VC also predicts cardiovascular death in different patient populations, although the predictive value ranges quite widely depending on the patient population and the underlying mix of risk factors [61–63].

VC has been recognized for many years as a common complication in patients with ESRD [64, 65]. Table 1 reports a list of the available studies that have evaluated the presence of coronary VC in patients with renal failure. Braun et al. [66] first reported the use of EBCT in 49 hemodialysis patients, showing coronary calcium score values 2.5- to 5-fold higher than non-hemodialysis patients. Other studies have revealed pronounced VC even in young adults with ESRD, who are otherwise not at risk for VC [42, 43]. In another report, Raggi et al. [67] studied a population of 205 patients on maintenance hemodialysis therapy observing high values of calcium score at the level of coronary arteries, thoracic aorta and cardiac valves. Furthermore, the coronary calcium score was significantly related to previous history

of myocardial infarction ($p < 0.0001$) and angina ($p < 0.0001$).

Predialysis CKD patients constitute a very large population with a documented high risk for cardiovascular events [7]. VC in predialysis patients with CKD, in contrast to hemodialysis patients, is much less intensively studied. Recent data revealed that advanced atherosclerosis (as demonstrated by the thickening of the arterial wall) is already present in patients with CKD before starting hemodialysis treatment [68]. Mehrotra et al. [69] studied a group of 60 patients with diabetic nephropathy with EBCT, revealing a higher prevalence and severity of coronary VC compared with diabetic controls with normal renal function. Interestingly, in this population, the high degree of VC was not related to measures of disordered mineral metabolism (table 1).

The Renal Research Institute (RRI)-CKD study is currently evaluating the severity of VC at the level of the coronary artery, thoracic aorta and cardiac valves in a subgroup of approximately 100 patients studied by 16-slice MDCT [70]. This is the first prospective study that will be specifically addressed to study prevalence and correlates of VC in an unselected population of predialysis CKD patients. In this population, the preliminary results seem to show the presence of high prevalence of VC, whereas no clear correlation with an index of abnormal mineral metabolism has been noted. The National Institutes of Health-sponsored Chronic Renal Insufficiency Cohort study will also address some of the same issues.

Table 2. Studies using EBCT or MDCT to assess the progression in coronary calcium score in patients with renal failure

Reference	Study population	Duration of follow-up	Imaging modality	Findings
Tamashiro et al. [71], 2001	24 HD	17 months	EBCT	progression correlates with higher triglycerides and lower high-density lipoprotein cholesterol
Chertow et al. [76], 2002	132 HD	11 months	EBCT	progression reduced by sevelamer
Bursztyn et al. [73], 2003	53 CKD + HTN	3 years	MDCT	progression faster in CKD compared with non-CKD
Stompor et al. [75], 2004	47 PD	1 year	MDCT	progression correlates with higher C-reactive protein, serum phosphate and calcium-phosphate product

HD = Hemodialysis; HTN = hypertension; PD = peritoneal dialysis.

Progression in VC in ESRD and CKD

VCs progress more rapidly in patients treated with dialysis than in the general population (table 2) [42, 71]. In asymptomatic subjects with normal renal function, the Agatston score progresses at an average rate of 33% per year [72]. Coronary calcium score progression (measured by MDCT) was 2-fold greater in hypertensive patients with CKD compared with hypertensives with normal renal function [73]. It is possible (although still a hypothesis) that medial wall calcification plays a dominant role in the progression of VC in patients with ESRD, while VC progression in patients with normal renal function mainly reflects the evolution of atherosclerotic calcification [74]. In a group of peritoneal dialysis patients, studied by MDCT at baseline and after 1 year of follow-up, Stompor et al. [75] reported a significant correlation between the change in coronary calcium score and the mean values of phosphate and calcium-phosphate product. Corroborating the association of VC progression with abnormal mineral metabolism, greater rates of VC progression have been demonstrated in ESRD patients treated with large oral doses of calcium-containing compounds, in comparison with patients treated with the calcium-free, phosphate-binding agent sevelamer [76].

There are limited data regarding the progression of VC in predialysis CKD patients [73]. A subgroup of approximately 50 patients enrolled in the RRI-CKD study have been studied with 16-slice MDCT at baseline and after about 1 year of follow-up. Our preliminary data reveal a high rate of VC progression in this unselected population of CKD patients (unpublished observations).

The utility of VC measurements (by EBCT or MDCT) as a predictor of adverse cardiovascular outcomes is yet to be demonstrated in patients with renal failure. A num-

ber of issues relevant to the renal patients need to be carefully studied. These include the evolution of VC and its relationship to decline in renal function. Although it is likely that medial versus intimal VC may have a differential impact on prognosis in CKD and for that matter dialysis patients, the image resolution of EBCT and MDCT is currently insufficient to distinguish between these two processes [77].

Conclusions

VC is ubiquitous and progresses rapidly in patients with advanced kidney disease. The paracrine and molecular determinants of VC in renal failure are continuously evolving. Modalities such as EBCT and MDCT allow non-invasive detection and quantification of VC and may provide useful prognostic information in this patient population. The ongoing Chronic Renal Insufficiency Cohort study and the RRI-CKD study should help provide information on the role for calcium scoring in cohorts of patients with predialysis CKD.

References

- Meguid EI, Nahas A, Bello AK: Chronic kidney disease: the global challenge. *Lancet* 2005;365:331–340.
- Coresh J, Astor BC, Greene T, Eknoyan G, Levey AS: Prevalence of chronic kidney disease and decreased kidney function in the adult US population: Third National Health and Nutrition Examination Survey. *Am J Kidney Dis* 2003;41:1–12.
- Shulman NB, Ford CE, Hall WD, Blaufox MD, Simon D, Langford HG, et al: Prognostic value of serum creatinine and effect of treatment of hypertension on renal function. Results from the hypertension detection and follow-up program. The Hypertension Detection and Follow-up Program Cooperative Group. *Hypertension* 1989;13(suppl 5):180–193.
- Manjunath G, Tighiouart H, Ibrahim H, MacLeod B, Salem DN, Griffith JL, et al: Level of kidney function as a risk factor for atherosclerotic cardiovascular outcomes in the community. *J Am Coll Cardiol* 2003;41:47–55.
- Fried LF, Shlipak MG, Crump C, Bleyer AJ, Gottdiener JS, Kronmal RA, et al: Renal insufficiency as a predictor of cardiovascular outcomes and mortality in elderly individuals. *J Am Coll Cardiol* 2003;41:1364–1372.
- Shlipak MG, Heidenreich PA, Noguchi H, Chertow GM, Browner WS, McClellan MB: Association of renal insufficiency with treatment and outcomes after myocardial infarction in elderly patients. *Ann Intern Med* 2002;137:555–562.
- Go AS, Chertow GM, Fan D, McCulloch CE, Hsu CY: Chronic kidney disease and the risks of death, cardiovascular events, and hospitalization. *N Engl J Med* 2004;351:1296–1305.
- Sarnak MJ, Levey AS, Schoolwerth AC, Coresh J, Culleton B, Hamm LL, et al: Kidney disease as a risk factor for development of cardiovascular disease: a statement from the American Heart Association Councils on Kidney in Cardiovascular Disease, High Blood Pressure Research, Clinical Cardiology, and Epidemiology and Prevention. *Circulation* 2003;108:2154–2169.
- Schwarz U, Buzello M, Ritz E, Stein G, Raabe G, Wiest G, et al: Morphology of coronary atherosclerotic lesions in patients with end-stage renal failure. *Nephrol Dial Transplant* 2000;15:218–223.
- Jungers P, Massy ZA, Khoa TN, Fumeron C, Labrunie M, Lacour B, et al: Incidence and risk factors of atherosclerotic cardiovascular accidents in predialysis chronic renal failure patients: a prospective study. *Nephrol Dial Transplant* 1997;12:2597–2602.
- Moe SM, Chen NX: Calciphylaxis and vascular calcification: a continuum of extra-skeletal osteogenesis. *Pediatr Nephrol* 2003;18:969–975.
- Nitta K, Akiba T, Suzuki K, Uchida K, Ogawa T, Majima K, et al: Assessment of coronary artery calcification in hemodialysis patients using multi-detector spiral CT scan. *Hypertens Res* 2004;27:527–533.
- Stary HC, Chandler AB, Dinsmore RE, Fuster V, Glagov S, Insull W Jr, et al: A definition of advanced types of atherosclerotic lesions and a histological classification of atherosclerosis. A report from the Committee on Vascular Lesions of the Council on Arteriosclerosis, American Heart Association. *Circulation* 1995;92:1355–1374.
- Beckman JA, Ganz J, Creager MA, Ganz P, Kinlay S: Relationship of clinical presentation and calcification of culprit coronary artery stenoses. *Arterioscler Thromb Vasc Biol* 2001;21:1618–1622.
- Huang H, Virmani R, Younis H, Burke AP, Kamm RD, Lee RT: The impact of calcification on the biomechanical stability of atherosclerotic plaques. *Circulation* 2001;103:1051–1056.
- Loree HM, Kamm RD, Stringfellow RG, Lee RT: Effects of fibrous cap thickness on peak circumferential stress in model atherosclerotic vessels. *Circ Res* 1992;71:850–858.
- Abedin M, Tintut Y, Demer LL: Vascular calcification: mechanisms and clinical ramifications. *Arterioscler Thromb Vasc Biol* 2004;24:1161–1170.
- Bostrom K, Watson KE, Horn S, Wortham C, Herman IM, Demer LL: Bone morphogenetic protein expression in human atherosclerotic lesions. *J Clin Invest* 1993;91:1800–1809.
- Fitzpatrick LA, Severson A, Edwards WD, Ingram RT: Diffuse calcification in human coronary arteries. Association of osteopontin with atherosclerosis. *J Clin Invest* 1994;94:1597–1604.
- Moe SM, O'Neill KD, Duan D, Ahmed S, Chen NX, Leapman SB, et al: Medial artery calcification in ESRD patients is associated with deposition of bone matrix proteins. *Kidney Int* 2002;61:638–647.
- Shanahan CM, Cary NR, Metcalfe JC, Weissberg PL: High expression of genes for calcification-regulating proteins in human atherosclerotic plaques. *J Clin Invest* 1994;93:2393–2402.
- Hujairi NM, Afzali B, Goldsmith DJ: Cardiac calcification in renal patients: what we do and don't know. *Am J Kidney Dis* 2004;43:234–243.
- Shanahan CM, Cary NR, Salisbury JR, Proudfoot D, Weissberg PL, Edmonds ME: Medial localization of mineralization-regulating proteins in association with Monckeberg's sclerosis: evidence for smooth muscle cell-mediated vascular calcification. *Circulation* 1999;100:2168–2176.
- Jono S, Nishizawa Y, Shioi A, Morii H: Parathyroid hormone-related peptide as a local regulator of vascular calcification. Its inhibitory action on *in vitro* calcification by bovine vascular smooth muscle cells. *Arterioscler Thromb Vasc Biol* 1997;17:1135–1142.
- Simonet WS, Lacey DL, Dunstan CR, Kelley M, Chang MS, Luthy R, et al: Osteoprotegerin: a novel secreted protein involved in the regulation of bone density. *Cell* 1997;89:309–319.
- Kaden JJ, Bickelhaupt S, Grobholz R, Haase KK, Sarikoc A, Kilic R, et al: Receptor activator of nuclear factor kappaB ligand and osteoprotegerin regulate aortic valve calcification. *J Mol Cell Cardiol* 2004;36:57–66.
- Avbersek-Luznik I, Malesic I, Rus I, Marc J: Increased levels of osteoprotegerin in hemodialysis patients. *Clin Chem Lab Med* 2002;40:1019–1023.
- Jono S, McKee MD, Murry CE, Shioi A, Nishizawa Y, Mori K, et al: Phosphate regulation of vascular smooth muscle cell calcification. *Circ Res* 2000;87:E10–E17.
- Ducy P, Zhang R, Geoffroy V, Ridall AL, Karsenty G: *Osf2/Cbfa1*: a transcriptional activator of osteoblast differentiation. *Cell* 1997;89:747–754.
- Chen NX, O'Neill KD, Duan D, Moe SM: Phosphorus and uremic serum up-regulate osteopontin expression in vascular smooth muscle cells. *Kidney Int* 2002;62:1724–1731.
- Giachelli CM: Vascular calcification mechanisms. *J Am Soc Nephrol* 2004;15:2959–2964.
- Ketteler M, Bongartz P, Westenfeld R, Wildberger JE, Mahnen AH, Bohm 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.
- Mehrotra R, Westenfeld R, Christenson P, Budoff M, Ipp E, Takasu J, et al: Serum fetuin-A in nondialyzed patients with diabetic nephropathy: relationship with coronary artery calcification. *Kidney Int* 2005;67:1070–1077.
- Farzaneh-Far A, Proudfoot D, Weissberg PL, Shanahan CM: Matrix Gla protein is regulated by a mechanism functionally related to the calcium-sensing receptor. *Biochem Biophys Res Commun* 2000;277:736–740.
- Luo G, Ducy P, McKee MD, Pinero GJ, Loyer E, Behringer RR, et al: Spontaneous calcification of arteries and cartilage in mice lacking matrix Gla protein. *Nature* 1997;386:78–81.
- Wada T, McKee MD, Steitz S, Giachelli CM: Calcification of vascular smooth muscle cell cultures: inhibition by osteopontin. *Circ Res* 1999;84:166–178.
- Steitz SA, Speer MY, McKee MD, Liaw L, Almeida M, Yang H, et al: Osteopontin inhibits mineral deposition and promotes regression of ectopic calcification. *Am J Pathol* 2002;161:2035–2046.
- Giachelli CM, Bac N, Almeida M, Denhardt DT, Alpers CE, Schwartz SM: Osteopontin is elevated during neointima formation in rat arteries and is a novel component of human atherosclerotic plaques. *J Clin Invest* 1993;92:1686–1696.
- Speer MY, McKee MD, Goldberg RE, Liaw L, Yang HY, Tung E, et al: Inactivation of the osteopontin gene enhances vascular calcification of matrix Gla protein-deficient mice: evidence for osteopontin as an inducible inhibitor of vascular calcification *in vivo*. *J Exp Med* 2002;196:1047–1055.

- 40 Zimmermann J, Herrlinger S, Pruy A, Metzger T, Wanner C: Inflammation enhances cardiovascular risk and mortality in hemodialysis patients. *Kidney Int* 1999;55:648–658.
- 41 Raggi P: Effects of excess calcium load on the cardiovascular system measured with electron beam tomography in end-stage renal disease. *Nephrol Dial Transplant* 2002;17:332–335.
- 42 Goodman WG, Goldin J, Kuizon BD, Yoon C, Gales B, Sider D, et al: Coronary-artery calcification in young adults with end-stage renal disease who are undergoing dialysis. *N Engl J Med* 2000;342:1478–1483.
- 43 Oh J, Wunsch R, Turzer M, Bahner M, Raggi P, Querfeld U, et al: Advanced coronary and carotid arteriopathy in young adults with childhood-onset chronic renal failure. *Circulation* 2002;106:100–105.
- 44 Moe SM, O'Neill KD, Fineberg N, Persohn S, Ahmed S, Garrett P, et al: Assessment of vascular calcification in ESRD patients using spiral CT. *Nephrol Dial Transplant* 2003;18:1152–1158.
- 45 O'Rourke RA, Brundage BH, Froelicher VF, Greenland P, Grundy SM, Hachamovitch R, et al: American College of Cardiology/American Heart Association Expert Consensus document on electron-beam computed tomography for the diagnosis and prognosis of coronary artery disease. *Circulation* 2000;102:126–140.
- 46 Moselewski F, Ropers D, Pohle K, Hoffmann U, Ferencik M, Chan RC, et al: Comparison of measurement of cross-sectional coronary atherosclerotic plaque and vessel areas by 16-slice multidetector computed tomography versus intravascular ultrasound. *Am J Cardiol* 2004;94:1294–1297.
- 47 Becker CR, Kleffel T, Crispin A, Knez A, Young J, Schoepf UJ, et al: Coronary artery calcium measurement: agreement of multirow detector and electron beam CT. *AJR Am J Roentgenol* 2001;176:1295–1298.
- 48 Stanford W, Thompson BH, Burns TL, Heery SD, Burr MC: Coronary artery calcium quantification at multi-detector row helical CT versus electron-beam CT. *Radiology* 2004;230:397–402.
- 49 Horiguchi J, Yamamoto H, Akiyama Y, Marukawa K, Hirai N, Ito K: Coronary artery calcium scoring using 16-MDCT and a retrospective ECG-gating reconstruction algorithm. *AJR Am J Roentgenol* 2004;183:103–108.
- 50 Agatston AS, Janowitz WR, Hildner FJ, Zusmer NR, Viamonte M Jr, Detrano R: Quantification of coronary artery calcium using ultrafast computed tomography. *J Am Coll Cardiol* 1990;15:827–832.
- 51 Callister TQ, Cooil B, Raya SP, Lippolis NJ, Russo DJ, Raggi P: Coronary artery disease: improved reproducibility of calcium scoring with an electron-beam CT volumetric method. *Radiology* 1998;208:807–814.
- 52 Ohnesorge B, Flohr T, Fischbach R, Kopp AF, Knez A, Schroder S, et al: Reproducibility of coronary calcium quantification in repeat examinations with retrospectively ECG-gated multisection spiral CT. *Eur Radiol* 2002;12:1532–1540.
- 53 Rumberger JA, Kaufman L: A rosetta stone for coronary calcium risk stratification: agatston, volume, and mass scores in 11,490 individuals. *AJR Am J Roentgenol* 2003;181:743–748.
- 54 Sangiorgi G, Rumberger JA, Severson A, Edwards WD, Gregoire J, Fitzpatrick LA, et al: Arterial calcification and not lumen stenosis is highly correlated with atherosclerotic plaque burden in humans: a histologic study of 723 coronary artery segments using noncalcifying methodology. *J Am Coll Cardiol* 1998;31:126–133.
- 55 Nallamothu BK, Saint S, Bielak LF, Sonnada SS, Peyser PA, Rubenfire M, et al: Electron-beam computed tomography in the diagnosis of coronary artery disease: a meta-analysis. *Arch Intern Med* 2001;161:833–838.
- 56 Yamamoto H, Shavelle D, Takasu J, Lu B, Mao SS, Fischer H, et al: Valvular and thoracic aortic calcium as a marker of the extent and severity of angiographic coronary artery disease. *Am Heart J* 2003;146:153–159.
- 57 Cury RC, Ferencik M, Hoffmann U, Ferullo A, Moselewski F, Abbata S, et al: Epidemiology and association of vascular and valvular calcium quantified by multidetector computed tomography in elderly asymptomatic subjects. *Am J Cardiol* 2004;94:348–351.
- 58 Raggi P, Callister TQ, Cooil B, He ZX, Lippolis NJ, Russo DJ, et al: Identification of patients at increased risk of first unheralded acute myocardial infarction by electron-beam computed tomography. *Circulation* 2000;101:850–855.
- 59 Shaw LJ, Raggi P, Schisterman E, Berman DS, Callister TQ: Prognostic value of cardiac risk factors and coronary artery calcium screening for all-cause mortality. *Radiology* 2003;228:826–833.
- 60 Raggi P, Shaw LJ, Berman DS, Callister TQ: Prognostic value of coronary artery calcium screening in subjects with and without diabetes. *J Am Coll Cardiol* 2004;43:1663–1669.
- 61 Detrano RC, Wong ND, Doherty TM, Shavelle RM, Tang W, Ginzton LE, et al: Coronary calcium does not accurately predict near-term future coronary events in high-risk adults. *Circulation* 1999;99:2633–2638.
- 62 Arad Y, Spadaro LA, Goodman W, Newstein D, Guerci AD: Prediction of coronary events with electron beam computed tomography. *J Am Coll Cardiol* 2000;36:1253–1260.
- 63 Greenland P, LaBree L, Azen SP, Doherty TM, Detrano RC: Coronary artery calcium score combined with Framingham score for risk prediction in asymptomatic individuals. *JAMA* 2004;291:210–215.
- 64 Salusky IB, Goodman WG: Cardiovascular calcification in end-stage renal disease. *Nephrol Dial Transplant* 2002;17:336–339.
- 65 Davies MR, Hruska KA: Pathophysiological mechanisms of vascular calcification in end-stage renal disease. *Kidney Int* 2001;60:472–479.
- 66 Braun J, Oldendorf M, Moshage W, Heidler R, Zeitler E, Luft FC: Electron beam computed tomography in the evaluation of cardiac calcification in chronic dialysis patients. *Am J Kidney Dis* 1996;27:394–401.
- 67 Raggi P, Boulay A, Chasan-Taber S, Amin N, Dillon M, Burke SK, et al: Cardiac calcification in adult hemodialysis patients. A link between end-stage renal disease and cardiovascular disease? *J Am Coll Cardiol* 2002;39:695–701.
- 68 Shoji T, Emoto M, Tabata T, Kimoto E, Shinohara K, Maekawa K, et al: Advanced atherosclerosis in predialysis patients with chronic renal failure. *Kidney Int* 2002;61:2187–2192.
- 69 Mehrotra R, Budoff M, Christenson P, Ipp E, Takasu J, Gupta A, et al: Determinants of coronary artery calcification in diabetics with and without nephropathy. *Kidney Int* 2004;66:2022–2031.
- 70 Perlman RL, Kiser M, Finkelstein F, Eisele G, Roys E, Liu L, et al: The longitudinal chronic kidney disease study: a prospective cohort study of predialysis renal failure. *Semin Dial* 2003;16:418–423.
- 71 Tamashiro M, Iseki K, Sunagawa O, Inoue T, Higa S, Afuso H, et al: Significant association between the progression of coronary artery calcification and dyslipidemia in patients on chronic hemodialysis. *Am J Kidney Dis* 2001;38:64–69.
- 72 Budoff MJ, Lane KL, Bakhsheshi H, Mao S, Grassmann BO, Friedman BC, et al: Rates of progression of coronary calcium by electron beam tomography. *Am J Cardiol* 2000;86:8–11.
- 73 Bursztyjn M, Motro M, Grossman E, Shemesh J: Accelerated coronary artery calcification in mildly reduced renal function of high-risk hypertensives: a 3-year prospective observation. *J Hypertens* 2003;21:1953–1959.
- 74 Callister TQ, Raggi P, Cooil B, Lippolis NJ, Russo DJ: Effect of HMG-CoA reductase inhibitors on coronary artery disease as assessed by electron-beam computed tomography. *N Engl J Med* 1998;339:1972–1978.
- 75 Stompou TP, Pasowicz M, Sulowicz W, Dembinska-Kiec A, Janda K, Wojcik K, et al: Trends and dynamics of changes in calcification score over the 1-year observation period in patients on peritoneal dialysis. *Am J Kidney Dis* 2004;44:517–528.
- 76 Chertow GM, Burke SK, Raggi P: Sevelamer attenuates the progression of coronary and aortic calcification in hemodialysis patients. *Kidney Int* 2002;62:245–252.
- 77 London GM, Guerin AP, Marchais SJ, Metivier F, Pannier B, Adda H: Arterial media calcification in end-stage renal disease: impact on all-cause and cardiovascular mortality. *Nephrol Dial Transplant* 2003;18:1731–1740.
- 78 Haydar AA, Hujairi NM, Covic AA, Pereira D, Rubens M, Goldsmith DJ: Coronary artery calcification is related to coronary atherosclerosis in chronic renal disease patients: a study comparing EBCT-generated coronary artery calcium scores and coronary angiography. *Nephrol Dial Transplant* 2004;19:2307–2312.