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Reduced Fetuin A Serum Level is Associated with Faster Stenosis Progression and Increased Valvular Calcification in Elderly Patients with Aortic Stenosis

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Abstract

Background: Fetuin A is a circulating calcium-regulatory glycoprotein that inhibits ectopic and vascular calcification. Aortic stenosis (AS) is a disease process involving an active calcification of the aortic valve (AV). Its prevalence increases markedly with aging. We examined the associations between serum level of Fetuin A with AV calcification, and disease progression rate of AS, in function of age.

Methods: 226 patients operated for AS, were divided into 2 groups according to the median value of age: the younger group (<70 years) and the elderly group (≥70 years). Serum fetuin A levels and calcium content of AV were determined respectively by Elisa method and *0*-cresolphtalein complexone method. The annualized progression rate of AS was calculated for the subset of patients (n=113) in whom at least 2 transthoracic Doppler-echocardiographic exams separated by at least 6 months were available pre-operatively.

Results: There was no correlation between fetuin A level and AV calcium content or AS progression rate in the subset of younger patients. On the other hand, in the elderly group, the preoperative progression rate of the peak transvalvular gradient was 2-fold faster in patients with serum fetuin A level <0.36 g/L (median value) when compared to those with higher level of fetuin A (9±1 mmHg/year vs. 5±1 mmHg/year, p=0.02). Moreover, there was a negative correlation between calcium content of the AV explanted at the time of surgery and fetuin A serum level (r=-0.22, p=0.05). After adjusting for age, male gender, triglycerides and the morphology of the AV (bicuspid vs. tricuspid), fetuin A remained significantly and inversely associated with AV calcification (r^2=0.09, β=-67.5, p= 0.04) and AS progression rate (r^2=0.30, β=-10.4, p=0.02).

Conclusion: In the elderly patients, reduced level of Fetuin A is associated with enhanced valvular calcification and faster stenosis progression rate. These findings also support that the determinants and mechanisms of the progression of AS may be different in younger patients.

Keywords: Aging; Aortic stenosis; Fetuin A; Calcific aortic valve disease

Introduction

Studies have shown that calcific aortic stenosis (AS) is not a passive degenerative disease, but rather an active cellular process which shares clinical and histological similarities with atherosclerosis including: lipoprotein deposition, chronic inflammation, and fibro-calcific tissue remodelling [1]. Calcification of the aortic valve (AV), a predominant feature of AS, is a highly active and regulated process of biomineralization, which shares similarities with bone formation. In fact, previous studies demonstrated that bone associated proteins such as osteopontin, osteonectin, osteocalcin and tenascin C are expressed within AS valves [2].

Powerful inhibitors of ectopic mineralization are required to prevent spontaneous calcification of the AV. Fetuin A is a circulating calcium-regulatory glycoprotein secreted by the liver, which prevents soft tissue calcification [3]. It inhibits vascular and ectopic mineralization by preventing nucleation of hydroxyapatite of calcium. In patients with chronic kidney diseases, fetuin A levels were reported to be reduced as compared to control healthy subjects [4]. Studies also suggest that low fetuin A level is associated with increased calcification of the mitral and aortic valves in patients with coronary artery disease [5]. Kadden et al. showed that patients with AS had lower fetuin A levels as compared to control patients [6].

Recent reports also indicate that the pathobiology of AS may vary in function of age [7]. To this effect, even after correction for covariates, when compared to younger patients, elderly subjects have a less altered lipid profile and a lower prevalence of the metabolic syndrome. Hence, it is suspected that whereas in the younger patients mineralization of the AV is, to a large extent, driven by lipid-derived products, in the elderly calcification of valvular tissue may be less driven by lipids and more influenced by mineralization-controlling factors [8]. In this work,

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we hypothesized that fetuin A has an age-dependent relationship with mineralization of the AV and the progression rate of AS.

Methods

Patients and tissue collection

226 patients with a moderate to severe AS and referred for an AV surgery were recruited in this study. Blood sample were taken and kept for further analyses. At the time of surgery the AV was retrieved, immediately immersed in liquid nitrogen and kept for the analysis of calcium content. The protocol was approved by local ethical committee and informed consent has been obtained from the subjects. All patients had moderate to severe AS and an aortic regurgitation grade ≤2+. Patients with a history of rheumatic disease, endocarditis or an inflammatory disease were excluded from the study.

Lipid profile and CRP measurements

Overnight fasting plasma was collected and immediately processed by the laboratory for the measurement of glucose, total cholesterol, low density-cholesterol (LDL), high density-cholesterol (HDL), and triglyceride levels. After centrifugation, plasma and serum samples were kept and stored at -80°C until measurement of C-reactive protein (CRP) and fetuin A levels. Methods used to measure CRP, LDL and HDL have been detailed previously [9].

Determination of serum fetuin A concentration

Serum fetuin A was measured with a commercially available enzyme-linked immunosorbent assay (ELISA) according to the manufacturer instructions (B-Bridge International, Sunnyvale, CA, USA).

Determination of valvular calcium content

Leaflets were homogenized and treated with HCl 6N at 95°C during 24 hours. Treated tissues were then centrifuged at 4400 RPM during 30 min and supernatants were collected. Calcium content was determined by the *0*-cresolphtalein complexone method. Results were expressed as mg of calcium per wet weight of tissue (Ca mg/g ww).

Preoperative doppler-echocardiographic data

For all patients included in this study, a transthoracic Doppler-echocardiography was performed pre-operatively. The hemodynamic severity of the stenosis was assessed by the measurement of the peak transvalvular gradient and aortic valve area (AVA) calculated by the standard continuity equation. When patients had ≥2 serial echocardiograms separated by at least 6 months, hemodynamic progression between the first and last echocardiographic study was calculated. To determine the hemodynamic progression rate of AS, annualized changes in the peak transvalvular gradients (mmHg/year) were calculated by dividing the difference between the first and last measurements by the time between examinations.

Statistical analysis

Results are expressed as means ±SEM. Values were compared between groups with a Student t-test for continuous data, whereas categorical data were compared using a χ^2 test. Correlations between variables were determined using Spearman's coefficients. Multiple

Variables	Younger <70 years (n=115)	Elderly ≥70 years (n=111)	p value
Age, years	59.7 ± 0.88	77.1 ± 0.4	<0.0001
Male, %	71.3 %	63.8 %	0.31
Smoking status, %	17.1 %	3.0 %	0.0009
BMI, Kg/m^2	29.6 ± 0.59	27.9 ± 0.5	0.02
Waist circumference, cm	104.2 ± 1.59	101.4 ± 1.55	0.21
Hypertension, %	56.4 %	68.9 %	0.04
Diabetes, %	23.3 %	34.3 %	0.09
Glycemia, mmol/L	5.87 ± 0.13	5.80 ± 0.15	0.88
Metabolic syndrome, %	28.1 %	23.0 %	0.43
CAD, %	66.4 %	50.0 %	0.02
Lipids			
LDL-cholesterol, mmol/L	2.5 ± 0.08	2.1 ± 0.08	0.002
HDL-cholesterol, mmol/L	1.3 ± 0.03	1.3 ± 0.04	0.85
Triglycerides, mmol/L	1.58 ± 0.08	1.32 ± 0.59	0.01
Statines, %	97.1 %	96.1 %	1
ACE inhibitors/ARBs, %	40.7 %	47.5 %	0.33
Bicuspid valve, %	49.5 %	7.7 %	<0.0001
Creatinine, mmol/L	86.9 ± 1.7	97.1 ± 2.4	0.0007
CRP, mg/L	2.45 ± 0.36	2.52 ± 0.31	0.89
Fetuin A, g/L	0.46 ± 0.02	0.36 ± 0.02	0.0006
Preoperative peak transvalvular gradient, mmHg	71 ± 3	71 ± 2	0.5
Preoperative aortic valve area, cm^2	0.78 ± 0.2	0.73 ± 0.18	0.1
Annualized peak transvalvular gradient, mmHg/year	7.99 ± 0.9	9.10 ± 0.8	0.38
Annualized aortic valve area, cm^2/year	-0.12 ± 0.1	-0.12 ± 0.1	0.1
Aortic valve calcium content, mg/ g ww tissue	56.9 ± 3.86	58.7 ± 3.26	0.72

Legend: BMI: body mass index; CAD: coronary artery disease; LDL: low-density lipoprotein; HDL: high-density lipoprotein; ACE: angiotensin-converting enzyme; ARB: angiotensin receptor blockers; CRP: C-reactive protein. Values are mean ± SEM

Table 1: Baseline characteristics of patients in younger and elderly group.

Variables	β coefficient	p value
Age	-0.004	0.001
Male gender	0.01	0.47
Diabetes	-0.04	0.02
TGs	0.06	0.004
CRP	-0.009	0.1

r^2 adjusted=0.23; p<0.0001
Legend: TGs: triglycerides; CRP: C-reactive protein

Table 2: Multivariate linear analysis of fetuin A in the whole cohort.

Variables	β coefficient	p value
Age	-0.37	0.7
Male gender	-1.99	0.62
TGs	-0.5	0.95
Bicuspid valve	-16.6	0.01
Fetuin A	-67.5	0.02

r^2 adjusted=0.09; p=0.04;
Legend : TGs : triglycerides

Table 3: Multivariate linear analysis of aortic valve calcium content in elderly patients.

Variables	β coefficient	P-value
Age	-0.15	0.51
Male gender	1.14	0.22
Bicuspid valve	0.58	0.71
Creatinine	-0.08	0.01
TGs	-0.1	0.95
Glycemia	0.55	0.28
Fetuin A	-10.4	0.04

r^2 adjusted=0.30; p=0.02
Legend : TGs : triglycerides

Table 4: Multivariate linear analysis of annualized peak transvalvular gradient in elderly patients.

linear regression analysis was used to identify the factors that are independently associated with the amount of valvular calcium as well as with the serum level of fetuin A. Variables with a p value ≤0.1 were entered in the multivariate models. A p value <0.05 was considered as significant. Statistical analysis was performed with a commercially available software package JMP IN 5.1.

Results

Interaction between age and fetuin A with regard to impact on AS progression

In Table 1, clinical characteristics as well as metabolic data are presented according to age (median of 70 years). In the elderly group (≥70 years), fetuin A level was significantly reduced compared to younger patients (<70 years) (0.46 g/L ± 0.02 vs. 0.36 g/L ± 0.02; p=0.0006). With regard to cardiovascular risk factors, there was a higher prevalence of hypertension (56.4 % vs. 68.9 %; p=0.04) and a lower prevalence of smoking (17.1 % vs. 3.0 %; p=0.0009) and bicuspid valves (49.5 % vs. 7.7 %; p<0.0001) in the elderly group. Also, patients in the elderly group had a significantly lower plasma level of LDL (2.5 mmol/L ± 0.08 vs. 2.1 mmol/L ± 0.08; p=0.002) and higher creatinine level (86.9 mmol/L ± 1.7 vs. 97.1 mmol/L ± 2.4; p=0.0007). However, elderly patients had similar preoperative transvalvular peak gradient (71 mmHg ± 3 vs. 71 mmHg ± 2; p=0.5), preoperative AVA (0.78 cm^2 ± 0.2 vs. 0.73 cm^2 ± 0.18; p=0.1), AS progression rate (peak gradient: 7.99 mmHg/year ± 0.9 vs. 9.10 mmHg/year ± 0.8; p=0.38, AVA: -0.12 cm2/year ± 0.1 vs. -0.12 cm^2/year ± 0.1; p=0.1), and valvular amount of calcium (56.9 mg/g ww tissue ± 3.86 vs. 58.7 mg/g ww tissue ± 3.26; p=0.72) compared to the younger group.

In the whole cohort of patients, serum levels of fetuin A were inversely related to age (r=-0.33; p<0.0001) and directly related to plasma triglycerides (r=0.17; p=0.03). Also, there was a non-significant trend for negative correlation between fetuin A and CRP levels (r=-0.17; p=0.07). Among the clinical risk factors, patients with diabetes had a tendency of having lower fetuin A levels. The mean serum levels of fetuin A were 0.37 ± 0.02 g/L and 0.41 ± 0.01 g/L respectively in patients with and without diabetes (p=0.09). On multivariate analysis, age (p=0.001), diabetes (p=0.02), and triglycerides (p=0.004) were independently related to serum level of fetuin A (r^2=0.23; β=-0.004; p<0.0001) (Table 2).

Relationships between fetuin A levels and valve calcification in function of age

In the whole series of patients with AS, fetuin A had a tendency of being inversely related with the valvular amount of calcium (-0.15; p=0.07). When stratified according to age, we found that serum level of fetuin A was not associated with valvular amount of calcium (r=-0.05; p=0.57) in the middle aged-group, whereas it was inversely correlated (r=-0.22; p=0.05) with valvular calcium in the elderly group.

In the latter group, after correction for age, gender, triglycerides, and bicuspid valves, fetuin A level (p=0.02) was significantly and inversely associated with valvular calcium content (r^2=0.09; β=-67.5, p=0.04) (Table 3). In the elderly group, when stratified to the median value of fetuin A (median value 0.36 g/L), the amount of valvular calcium were 70±5 mg/g ww and 56±5 mg/g ww respectively in patients with low and high values of fetuin A (p=0.03) (Figure 1).

Relationship between serum fetuin A and preoperative stenosis progression rate

In this study, 50% (n=113) of the 226 patients had ≥2 serial echocardiograms separated by at least 6 months (mean follow up: 4±3 years) in the preoperative period. Fetuin A level was not associated with the progression rate of AS (r=0.22; p=0.18) in the younger group, whereas it was inversely related to the preoperative stenosis progression rate evaluated with the annualized peak transvalvular gradient (r=-

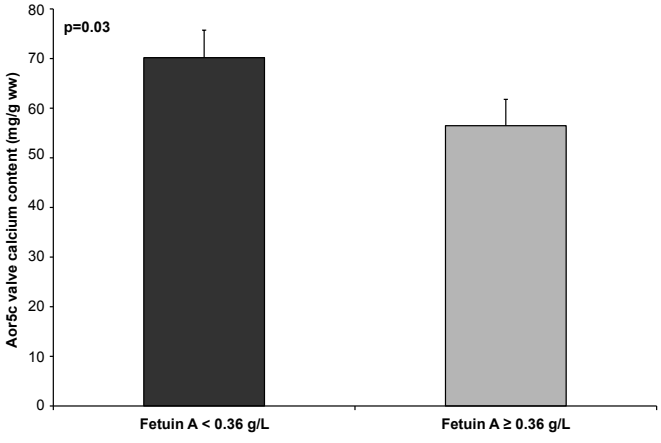

Figure 1: Aortic valve calcium content in relation with fetuin A level in the elderly population.

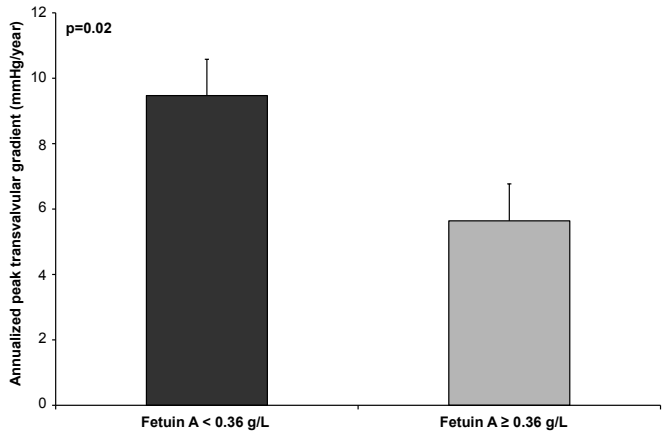

Figure 2: Annualized peak transvalvular gradient in relation with fetuin A level in the elderly patients.

0.37; p=0.02) in the elderly group. Although the peak transvalvular gradient was not significantly different at baseline, AS progression rate was nearly 2-fold faster in patients with decreased serum fetuin A level (median value <0.36 g/L) when compared to those with higher level of fetuin A (9±1 mmHg/year vs. 5±1 mmHg/year; p=0.02) (Figure 2). After adjustment for age, gender, bicuspid valve, creatinine, triglyceride level and glycemia, fetuin A (p=0.04) remained independently and inversely associated with a faster hemodynamic progression rate as measured with the annualized transaortic peak gradient (r^2=0.30; β=-10.4; p=0.02) (Table 4).

Discussion

The most important contribution of this study was to demonstrate for the first time, that reduced circulating level of fetuin A is associated with a faster progression of AS and enhanced valvular calcification in elderly patients, whereas there was no such association in the younger group of patients. Furthermore, we found that aging is associated with lower serum levels of fetuin A.

Pathobiology of aortic stenosis: an age-dependent process?

Recent findings indicate that risk factors/biomarkers that are associated with aortic valve calcification are, at least in part, age-dependent [7]. To this effect, it was found that lipid variables, including plasma levels of LDL, were different in younger vs. elderly patients with AS. Of note, LDL levels were higher and the size of LDL particles was smaller in younger patients with AS when compared to elderly subjects. It is worth to underline that small, dense LDL particles have been associated with AV inflammation and a faster progression of AS [8]. Also, one study reported in a large cohort of subjects that plasma level of LDL is a risk factor for AV calcification, only in the population younger than 65 years-old [10]. In the same line, a recent study has found that pathological mineralization of atherosclerotic plaque was closely associated with plasma LDL levels particularly in younger patients, whereas this association was less important with aging [11]. When taken together, these findings indicate that mineralization of the AV may be more dependent on lipid variables in younger patients, whereas in elderly other factors may contribute to this process. In fact, survival bias is likely one explanation behind the observation that elderly patients with AS have a less deteriorated blood lipid profile. However, there are biological modifications with the aging process that may explain the high incidence of AV calcification that is encountered in the elderly subjects [12,13]. In this regard, in the present study we documented that serum level of fetuin A was independently and inversely related with age. Hence, lower levels of serum fetuin A may be one factor explaining the frequent association of aging with AS.

Fetuin A, mineralization and aortic stenosis

Fetuin A is synthesized by the liver and reaches high serum concentrations [14]. It prevents hydroxyapatite formation by reducing crystal formation [15] and it can also assemble a high molecular mass complex with calcium phosphate mineral and Matrix Gla protein (MGP), a key regulator of tissue calcification [16]. Also, fetuin A binds to bone morphogenetic proteins (BMPs) and transforming growth factor beta1 (TGF-β1) and, in doing so, may prevent osteogenic signals [17]. Mice that lack fetuin A exhibit extensive soft tissue calcification which is amplified on a mineral/vitamin D-rich diet, which suggests that fetuin A acts to inhibit calcification systemically [18]. Hence, fetuin A appears as a critical regulator of ectopic mineralization. At this point, it is worth to highlight that the development and progression of AS is largely dependent on the mineralization of the AV [19,20]. Therefore, factors that may have a control on ectopic mineralization of the AV may participate to disease initiation/progression. In the present study, we documented in elderly patients that serum levels of fetuin A were independently and inversely related with calcium content of the explanted stenotic AV. These findings are in line with reports indicating that fetuin A is inversely related with AV calcification [5,6]. However, more important perhaps, in the present work we have also documented that lower level of fetuin A was independently associated with a faster hemodynamic progression of AS in elderly patients.

Clinical implications

Increasing evidence suggest that the relative importance of lipid factors may be more relevant with the mineralization of the AV in younger patients, whereas in elderly its impact may be, at most, modest. Hence, it is possible that other factors may participate to the frequent association of AS with aging. In this regard, we found that fetuin A may well be one such factor participating to the calcification of the aortic valve in the elderly population. Progression of AS is variable on an individual basis, and it is likely that genetic, biological and environmental factors explain that some patients have a faster disease course than others [21]. It then follows that efforts should be dedicated to identify patients at high risk for rapid progression of AS. In this regard, a low serum level of fetuin A in a patient ≥70 y.o. indicates a higher risk for a rapid AS progression. These patients should have a closer Doppler-echocardiographic follow-up.

Limitations

Insofar the present study included patients with severe AS, the present conclusion cannot be transposed to the population with early mineralization of the AV. The present independent associations derived from this study do not necessarily imply cause-and-effect relationship between fetuin A and AS. Nonetheless, the present study has contributed to identify fetuin A as a potential marker of disease activity that is age-dependent.

Conclusions

Fetuin A is a critical regulator of ectopic mineralization. In the present work, we found that the serum level of fetuin A decreases with aging in the population with AS. In addition, we documented in the elderly patients that serum level of fetuin A was inversely and

independently related with the AV content in calcium as well as with the hemodynamic progression of stenosis. Further studies aimed at identifying the role of fetuin A, particularly in the elderly population with AS, are required. These studies may help eventually to design novel medical treatments for patients with AS, a condition that is affecting our aging societies.

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References

1. Cote C, Pibarot P, Despres JP, Mohty D, Cartier A, et al. (2008) Association between circulating oxidised low-density lipoprotein and fibrocalcific remodelling of the aortic valve in aortic stenosis. Heart 94: 1175-1180.
2. Charest A, Pepin A, Shetty R, Côté C, Voisine P, et al. (2006) Distribution of SPARC during neovascularisation of degenerative aortic stenosis. Heart 92: 1844-1849.
3. Schafer C, Heiss A, Schwarz A, Westenfeld R, Ketteler M, et al. (2003) The serum protein alpha 2-Heremans-Schmid glycoprotein/fetuin-A is a systemically acting inhibitor of ectopic calcification. J Clin Invest 112: 357-366.
4. Ketteler M, Wanner C, Metzger T, Bongartz P, Westenfeld R, et al. (2003) Deficiencies of calcium-regulatory proteins in dialysis patients: a novel concept of cardiovascular calcification in uremia. Kidney Int Suppl S84-S87.
5. Ix JH, Chertow GM, Shlipak MG, Brandenburg VM, Ketteler M, et al. (2007) Association of fetuin-A with mitral annular calcification and aortic stenosis among persons with coronary heart disease: data from the Heart and Soul Study. Circulation 115: 2533-2539.
6. Kaden JJ, Reinohl JO, Blesch B, Brueckmann M, Haghi D, et al. (2007) Systemic and local levels of fetuin-A in calcific aortic valve stenosis. Int. J Mol Med 20: 193-197.
7. Mohty D, Pibarot P, Despres JP, Cartier A, Arsenault B, et al. (2010) Age-related differences in the pathogenesis of calcific aortic stenosis: the potential role of resistin. Int J Cardiol 142: 126-132.
8. Mohty D, Pibarot P, Despres JP, Côté C, Arsenault B, et al. (2008) Association between plasma LDL particle size, valvular accumulation of oxidized LDL, and inflammation in patients with aortic stenosis. Arterioscler Thromb Vasc Biol 28: 187-193.
9. Lamarche B, Despres JP, Moorjani S, Cantin B, Dagenais GR, et al. (1995) Prevalence of dyslipidemic phenotypes in ischemic heart disease (prospective results from the Quebec Cardiovascular Study). Am J Cardiol 75: 1189-1195.
10. Owens DS, Katz R, Johnson E, Shavelle DM, Probstfield JL, et al. (2008) Interaction of age with lipoproteins as predictors of aortic valve calcification in the multi-ethnic study of atherosclerosis. Arch Intern Med 168: 1200-1207.
11. Paramsothy P, Katz R, Owens DS, Burke GL, Probstfield JL, et al. (2010) Age-modification of lipoprotein, lipid, and lipoprotein ratio-associated risk for coronary artery calcium (from the Multi-Ethnic Study of Atherosclerosis [MESA]). Am.J Cardiol 105: 352-358.
12. Stewart BF, Siscovick D, Lind BK, Gardin JM, Gottdiener JS, et al. (1997) Clinical factors associated with calcific aortic valve disease. Cardiovascular Health Study. J Am Coll Cardiol 29: 630-634.
13. Lindroos M, Kupari M, Heikkila J, Tilvis R (1993) Prevalence of aortic valve abnormalities in the elderly: an echocardiographic study of a random population sample. J Am Coll Cardiol 21: 1220-1225.
14. Heiss A, Eckert T, Aretz A, Richtering W, van Dorp W, et al. (2008) Hierarchical role of fetuin-A and acidic serum proteins in the formation and stabilization of calcium phosphate particles. J Biol Chem 283: 14815-14825.
15. Heiss A, DuChesne A, Denecke B, Grötzinger J, Yamamoto K, et al. (2003) Structural basis of calcification inhibition by alpha 2-HS glycoprotein/fetuin-A. Formation of colloidal calciprotein particles. J Biol Chem 278: 13333-13341.
16. Price PA, Caputo JM, Williamson MK (2002) Bone origin of the serum complex of calcium, phosphate, fetuin, and matrix Gla protein: biochemical evidence for the cancellous bone-remodeling compartment. J Bone Miner Res 17: 1171-1179.
17. Binkert C, Demetriou M, Sukhu B, Szweras M, Tenenbaum HC, et al. (1999) Regulation of osteogenesis by fetuin. J Biol Chem 274: 28514-28520.
18. Schafer C, Heiss A, Schwarz A, Westenfeld R, Ketteler M, et al. (2003) The serum protein alpha 2-Heremans-Schmid glycoprotein/fetuin-A is a systemically acting inhibitor of ectopic calcification. J Clin Invest 112: 357-366.
19. Messika-Zeitoun D, Aubry MC, Detaint D, Bielak LF, Peyser PA, et al. (2004) Evaluation and clinical implications of aortic valve calcification measured by electron-beam computed tomography. Circulation 110: 356-362.
20. Bosse Y, Miqdad A, Fournier D, Pepin A, Pibarot P, et al. (2009) Refining molecular pathways leading to calcific aortic valve stenosis by studying gene expression profile of normal and calcified stenotic human aortic valves. Circ Cardiovasc Genet 2: 489-498.
21. Bosse Y, Mathieu P, Pibarot P (2008) Genomics: the next step to elucidate the etiology of calcific aortic valve stenosis. J Am Coll Cardiol 51: 1327-1336.