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Impact of Plasma Lp-PLA2 Activity on the Progression of Aortic Stenosis

The PROGRESSA Study

Romain Capoulade, PhD, Ablajan Mahmut, MD, MSc, Lionel Tastet, BSc, Marie Arsenault, MD, Élisabeth Bédard, MD, Jean G. Dumesnil, MD, Jean-Pierre Després, PhD, Éric Larose, DVM, MD, Benoit J. Arsenault, PhD, Yohan Bossé, PhD, Patrick Mathieu, MD, Philippe Pibarot, DVM, PhD

ABSTRACT

OBJECTIVES The purpose of this prospective study was to examine the relationship between plasma lipoprotein-associated phospholipase A2 (Lp-PLA2) activity and the progression rate of aortic stenosis (AS).

BACKGROUND We recently reported that Lp-PLA2 is highly expressed in stenotic aortic valves where it may contribute to the mineralization of valvular interstitial cells.

METHODS Patients with AS were prospectively recruited in the PROGRESSA (Metabolic Determinants of the Progression of Aortic Stenosis) study. AS progression rate was assessed by annualized increase in peak aortic jet velocity (V_{peak}), mean gradient (MG), and aortic valve area index (AVAi). Circulating Lp-PLA2 activity was measured and dichotomized based on the median value.

RESULTS Of 183 patients included in this subanalysis of the PROGRESSA study, 70% were men and the mean age was 66 ± 13 years. Over the 2.5 ± 1.4 years of follow up, the AS progression rate tended to be higher in patients with high versus low Lp-PLA2 activity (annualized V_{peak} = 0.17 ± 0.23 m/s vs. 0.12 ± 0.18 m/s; p = 0.14). There was a significant interaction (p < 0.05) between baseline AS severity and Lp-PLA2 activity with respect to impact on AS progression rate. In patients with mild AS (i.e., V_{peak} <3 m/s; n = 123), increased Lp-PLA2 activity was associated with a significantly faster AS progression rate (V_{peak} 0.16 ± 0.18 m/s vs. 0.09 ± 0.14 m/s; p = 0.01) but not in patients with moderate or severe AS (p = 0.99). After adjustment for other risk factors, increased Lp-PLA2 activity remained independently associated with faster AS progression rate (p = 0.005) in the former subset.

CONCLUSIONS There was no significant association between plasma Lp-PLA2 activity or mass and stenosis progression in the whole cohort. However, increased Lp-PLA2 activity was associated with a faster stenosis progression rate in the subset of patients with mild AS. These findings provide an impetus for the elaboration of a randomized trial targeting Lp-PLA2 activity in patients with early stages of calcific aortic valve disease. (J Am Coll Cardiol Img 2015;8:26–33)
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Calcific aortic stenosis (AS) is a chronic and multifactorial disorder characterized by an abnormal mineralization of aortic valve (AV) cusps (1). The pathological processes that lead to ectopic mineralization of the valvular tissue may involve lipid-derived factors (1). Studies conducted with human AS valve specimens demonstrated that oxidized low-density lipoprotein (LDL) accumulates in areas of inflammation and mineralization (2,3). Circulating levels of oxidized LDL are positively associated with several indexes of aortic valve remodeling, suggesting that oxidized-derived lipids may play a significant role in the development of AS (4).

Lipoprotein-associated phospholipase A2 (Lp-PLA2) uses oxidized LDL as substrate and produces free fatty acids and lysophosphatidylcholine (LPC), a powerful proinflammatory and procalcifying factor (5). We recently reported that Lp-PLA2 is highly expressed in stenotic AVs and that LPC induces mineralization of valvular interstitial cells in vitro (6). In another recent cross-sectional study, plasma levels of Lp-PLA2 was increased in patients with AS (7).

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These findings support the hypothesis that Lp-PLA2 may play a role in AV mineralization. The objective of this prospective study was thus to examine the relationship between circulating Lp-PLA2 activity and the progression rate of AS.

METHODS

PATIENT POPULATION. Patients with AS recruited in the PROGRESSA (Metabolic Determinants of the Progression of Aortic Stenosis) study underwent Doppler echocardiography annually. Inclusion criteria were 21 years of age and older and peak aortic jet velocity (V_{peak}) >2.0 m/s. Patients were excluded if they had symptomatic AS, moderate to severe aortic regurgitation or mitral valve disease (mitral stenosis or regurgitation), and left ventricular ejection fraction (LVEF) <50%, and if they were pregnant or lactating. The study was approved by the Ethics Committee of the Quebec Heart and Lung Institute, and patients signed a written informed consent at the time of inclusion in the study. Among the 216 patients recruited until March 31, 2014, 183 patients with at least 1 year of follow-up were included in the present subanalysis of the PROGRESSA study.

CLINICAL DATA. Clinical data included age, sex, height, weight, body surface area (BSA), body mass index, systolic and diastolic blood pressures, documented diagnoses of hypertension (patients taking antihypertensive medications or having known but untreated hypertension [blood pressure ≥140/90 mm Hg]), diabetes (patients taking antidiabetic medication or, in the absence of such medication, having a fasting glucose ≥7 mmol/l), dyslipidemia (patients taking cholesterol-lowering medication or, in the absence of such medication, having a total plasma cholesterol level >6.2 mmol/l), and coronary artery disease (history of myocardial infarction or coronary artery stenosis on coronary angiography).

LABORATORY DATA. Fasting plasma samples were collected to measure levels of glucose and creatinine as well as a complete lipid profile, which included total cholesterol, LDL cholesterol, high-density lipoprotein (HDL) cholesterol, and triglycerides using automated techniques standardized with the Canadian reference laboratory.

After centrifugation, plasma samples were stored at −80°C for future measurements of Lp-PLA2 activity and mass. Briefly, circulating Lp-PLA2 activity was measured by a commercial colorimetric activity method (Alanine Transaminase Colorimetric Activity Assay Kit, Cayman Chemical, Ann Arbor, Michigan). We also measured the Lp-PLA2 mass using a commercial ELISA kit (R&D Systems, Minneapolis, Minnesota). Results were expressed as nanomole per minute per milliliter for Lp-PLA2 activity and nanogram per milliliter for Lp-PLA2 mass.

DOPPLER ECHOCARDIOGRAPHIC DATA. All Doppler echocardiographic examinations were performed and analyzed in the same laboratory by the same team of sonographers and cardiologists. The investigators who did the acquisition and analysis of the Doppler echocardiographic images were blinded to the results of all blood analyses including those of Lp-PLA2.

Aortic valve morphology and function. The AV phenotype (i.e., bicuspid vs. tricuspid AV) was recorded. The Doppler echocardiographic indexes of AS severity included V_{peak}, peak and mean transvalvular pressure gradients (MG) obtained with the modified Bernoulli equation, and the AV area index (AVAi) calculated by the standard continuity equation and indexed to the BSA. The degree of AV calcification was scored according to the criteria proposed by Rosenhek et al. (8).

Left ventricular hemodynamic load and function. As a measure of global left ventricular hemodynamic load, we calculated the valvuloarterial impedance: $Z_{va} = (SBP + \Delta P_{mean})/SVi$, where SBP is the systolic blood pressure, ΔP_{mean} is the mean transvalvular gradient, and SVi is the stroke volume indexed to the BSA (9). LVEF was measured with the biplane Simpson method.

STUDY ENDPOINTS. The primary endpoint for this study was the progression rate of valve stenosis

ABBREVIATIONS AND ACRONYMS

AS = aortic stenosis
AV = aortic valve
AVAi = aortic valve area index
BSA = body surface area
HDL = high-density lipoprotein
LDL = low-density lipoprotein
Lp-PLA2 = lipoprotein-associated phospholipase A2
LPC = lysophosphatidylcholine
LVEF = left ventricular ejection fraction
MG = mean transvalvular pressure gradient
V_{peak} = peak aortic jet velocity

TABLE 1 Baseline Characteristics of the Study Population According to Circulating Lp-PLA2 Activity

	All Patients (N = 183)	Lp-PLA2 Activity <11.4 (n = 96) (52%)	Lp-PLA2 Activity >11.4 (n = 87) (48%)	p Value
Clinical				
Age, yrs	66.00 ± 13.00	67.00 ± 13.00	65.00 ± 13.00	NS
Male	70	66	77	0.07
Height, cm	166.00 ± 9.00	165.00 ± 8.00	168.00 ± 9.00	0.03
Weight, kg	79.00 ± 14.00	77.00 ± 15.00	80.00 ± 14.00	NS
Body surface area, m^2	1.87 ± 0.20	1.84 ± 0.20	1.90 ± 0.19	0.05
Body mass index, kg/m^2	28.30 ± 4.10	28.10 ± 4.30	28.40 ± 4.00	NS
History of hypertension	74	77	70	NS
Systolic blood pressure, mm Hg	134.00 ± 20.00	135.00 ± 20.00	133.00 ± 19.00	NS
Diastolic blood pressure, mm Hg	74.00 ± 10.00	73.00 ± 11.00	75.00 ± 10.00	NS
History of dyslipidemia	67	75	57	0.02
History of diabetes	22	28	16	0.05
History of coronary artery disease	40	48	31	0.02
Medication				
Beta-blockers	33	41	25	0.03
ACE inhibitors	32	32	31	NS
ARBs	30	35	23	0.07
Statin	67	76	57	0.007
Antidiabetes	22	28	15	0.03
Laboratory data				
LDL cholesterol, mmol/l	2.35 ± 0.82	2.07 ± 0.63	2.65 ± 0.91	<0.0001
HDL cholesterol, mmol/l	1.43 ± 0.40	1.48 ± 0.44	1.37 ± 0.33	0.05
Triglycerides, mmol/l	1.44 ± 0.77	1.26 ± 0.57	1.63 ± 0.10	0.001
Fasting glucose, mmol/l	5.70 ± 1.40	5.80 ± 1.60	5.20 ± 1.30	NS
Creatinine, µmol/l	86.00 ± 25.00	87.00 ± 26.00	85.00 ± 25.00	NS
Lp-PLA2 activity, nmol/min/ml	11.90 ± 2.90	9.70 ± 1.20	14.20 ± 2.30	<0.0001
Lp-PLA2 mass, ng/ml	122.00 ± 43.00	97.00 ± 29.00	149.00 ± 39.00	<0.0001
Doppler echocardiographic data				
Bicuspid aortic valve	18	15	21	NS
Aortic valve calcification score	2.50 ± 0.60	2.50 ± 0.60	2.60 ± 0.50	NS
Peak aortic jet velocity, m/s	2.80 ± 0.60	2.80 ± 0.60	2.80 ± 0.60	NS
Peak transvalvular gradient, mm Hg	34.00 ± 14.00	34.00 ± 14.00	34.00 ± 14.00	NS
Mean transvalvular gradient, mm Hg	19.00 ± 9.00	19.00 ± 9.00	19.00 ± 9.00	NS
Aortic valve area, cm^2	1.26 ± 0.29	1.22 ± 0.29	1.30 ± 0.29	NS
Indexed aortic valve area, cm^2/m^2	0.68 ± 0.16	0.67 ± 0.17	0.69 ± 0.15	NS
Valvuloarterial impedance, mm $Hg/ml/m^2$	3.60 ± 0.70	3.60 ± 0.70	3.50 ± 0.80	NS
LV ejection fraction, %	65.00 ± 6.00	66.00 ± 6.00	65.00 ± 6.00	NS

Values are mean ± SD or %.

ACE = angiotensin-converting enzyme; ARBs = angiotensin receptor blockers; HDL high-density lipoprotein; LDL = low-density lipoprotein; Lp-PLA2 = lipoprotein-associated phospholipase A2; LV = left ventricular.

measured by Doppler echocardiography. To account for different follow-up lengths, annualized changes in V_{peak}, MG, and AVAi were calculated by dividing the difference between last follow-up and baseline values by the time duration of follow-up. The secondary endpoint was the change in AS severity class (i.e., transition from mild to moderate or severe AS) during follow-up.

STATISTICAL ANALYSIS. Continuous data were expressed as mean ± SD and compared with the Student *t* test according to Lp-PLA2 activity dichotomized at the median value (i.e., 11.4 nmol/min/ml). The continuous variables were tested for normality of distribution and homogeneity of variances with the Shapiro-Wilk and Levene tests, respectively. Categorical data were expressed as percentages and compared with the chi-square or Fisher exact test where appropriate. Univariable linear regression analyses were performed to identify the impact of plasma Lp-PLA2 activity and mass on AS progression rate (i.e., annualized progression rates of V_{peak}, MG, and AVAi) and the interaction between Lp-PLA2 activity or mass and baseline AS severity. To determine whether Lp-PLA2 activity is independently associated with the stenosis progression rate, we built a multivariable linear regression model that included

variables with a p value <0.10 on individual analysis as well as traditional cardiovascular risk factors. Variables entered in the model, other than Lp-PLA2 activity, were age, sex, hypertension, diabetes, LDL cholesterol, statin therapy, creatinine level, bicuspid AV phenotype, degree of AV calcification, baseline AS severity (i.e., V_{peak}, MG, or AVAi). The independent correlates of Lp-PLA2 activity were assessed by multivariable linear regression. Standardized regression coefficients were presented as mean ± SE (beta coefficient ± SE). The Kaplan-Meier curve and log-rank test of the time-to-event data were used to assess the transition from mild to moderate or severe AS according to median value of plasma Lp-PLA2 activity. Multivariable Cox proportional hazard analysis adjusted for age, sex, degree of AV calcification and baseline AS severity, was performed to determine the independent association between Lp-PLA2 activity and the worsening of AS severity class. Multivariate models were inspected for multicollinearity by calculating the variance inflation factor. A p value <0.05 was considered statistically significant.

RESULTS

POPULATION CHARACTERISTICS. Table 1 shows the baseline characteristics of the 183 patients included in this study. The mean age was 66 ± 13 years, 70% were men, 74% had systemic arterial hypertension, 67% had dyslipidemia, 22% had diabetes, and 40% had coronary artery disease. Eighteen percent of patients presented a bicuspid AV phenotype, 67% had mild (V_{peak} <3.0 m/s), 29% moderate (V_{peak} 3.0 to 3.9 m/s), and 4% severe (V_{peak} >4.0 m/s) AS at baseline.

The baseline Lp-PLA2 activity was 11.9 ± 2.9 nmol/min/ml, and the plasma Lp-PLA2 mass was 122 ± 43 ng/ml (Table 1). Lp-PLA2 activity correlated with Lp-PLA2 mass ($r = 0.78$, $p < 0.001$). Patients with increased Lp-PLA2 activity (i.e., median value, >11.4 nmol/min/ml) had lower prevalence of dyslipidemia ($p = 0.02$), diabetes ($p = 0.05$), and coronary artery disease ($p = 0.02$), and they were less often treated with beta-blockers, statins, antidiabetes drugs compared with patients with low Lp-PLA2 activity (Table 1). They also had higher LDL cholesterol ($p < 0.0001$) and triglycerides ($p = 0.001$) and lower HDL cholesterol ($p = 0.05$) (Table 1). Regarding Doppler echocardiographic data, patients with high versus low plasma Lp-PLA2 activity had similar baseline AS severity, Z_{va}, and LVEF (Table 1).

IMPACT OF PLASMA LP-PLA2 ON AS PROGRESSION RATE AND INTERACTION WITH BASELINE AS SEVERITY. During the mean follow-up of 2.5 ± 1.4 years, patients with increased Lp-PLA2 activity had a nonsignificant trend for faster AS progression rate (annualized V_{peak} 0.17 ± 0.23 m/s/year vs. 0.12 ± 0.18 m/s/year, $p = 0.14$) (Figure 1A). There was no significant association between increased plasma Lp-PLA2 mass (i.e., median value >119.3 ng/ml) and AS progression rate (annualized V_{peak} 0.15 ± 0.23 m/s/year vs. 0.14 ± 0.19 m/s/year, $p = 0.88$).

There was a significant interaction between Lp-PLA2 activity and baseline AS severity on AS progression rate (i.e., all interaction p values <0.05 for V_{peak}, MG, and AVAi), suggesting that the impact of circulating plasma Lp-PLA2 activity on AS progression rate differed depending on patients' baseline AS severity. Interaction between circulating plasma Lp-PLA2 mass and baseline AS severity on AS progression did not reach statistical significance (i.e., all interaction p values >0.20). We thus performed separated analyses in the subset of patients with mild AS and moderate or severe AS. In patients with mild AS (i.e., V_{peak} <3.0 m/s, $n = 123$) (Online Table 1), stenosis progression rate was 2-fold faster in the subset of patients with increased Lp-PLA2 activity (annualized progression of V_{peak} 0.16 ± 0.18 m/s/year vs. 0.09 ± 0.14 m/s/year, $p = 0.01$) (Figure 1B). On the other hand, there was no association between Lp-PLA2 activity and stenosis progression rate in the patients with moderate or severe AS (annualized V_{peak} 0.20 ± 0.33 m/s/year vs. 0.19 ± 0.23 m/s/year, $p = 0.99$) (Figure 1C). Similar results were obtained with annualized progression of MG and AVAi (Online Figure 1 and Online Table 1).

Lp-PLA2 activity or mass expressed in continuous variables was not associated with AS progression in the whole cohort (all $p > 0.20$) (Table 2). On univariable analysis, Lp-PLA2 activity was significantly associated with faster AS progression rate (annualized V_{peak} $p = 0.02$; annualized MG: $p = 0.05$; annualized AVAi: $p = 0.03$) in patients with mild AS but not patients with moderate or severe AS (all $p > 0.20$) (Table 2). On multivariable analysis adjusted for age, sex, hypertension, diabetes, LDL cholesterol, statin therapy, creatinine level, bicuspid AV phenotype, degree of AV calcification, and AS severity, Lp-PLA2 activity was independently associated with a faster annualized progression rate of V_{peak} ($p = 0.005$), MG ($p = 0.02$), or AVAi ($p = 0.02$) in the subset of patients with mild AS (Table 2, multivariable models). The variance inflation factor was <5, thus confirming that the level of multicollinearity in these multivariable models is acceptable.

AS SEVERITY CLASS. During follow-up, 46 patients with mild AS progressed from mild to moderate or severe AS. Increased Lp-PLA2 activity was associated with a higher incidence of progression to more severe

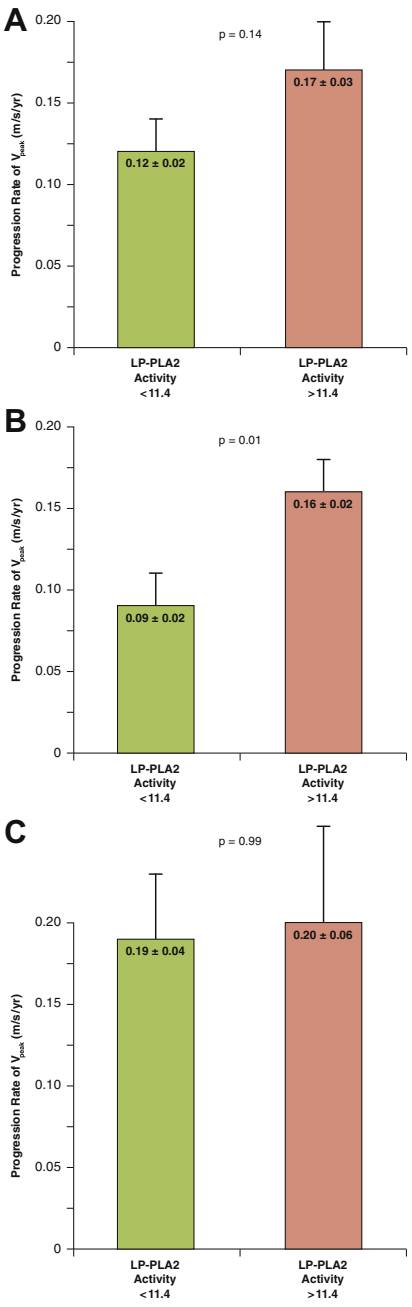

FIGURE 1 Progression Rate of AS According to the Median Value of Plasma Lp-PLA2 Activity

Comparison of annualized progression of V_{peak} in the whole cohort (N = 183) **(A)**, in patients with mild AS **(B)** (V_{peak} <3.0 m/s; n = 123), and in patients with moderate/severe AS **(C)** (V_{peak} ≥3.0 m/s; n = 60) according to the median value of Lp-PLA2 activity (i.e., 11.4 nmol/min/ml). The numbers on top of the bars are mean annualized V_{peak} ± SEM. Error bars represent the SEM. AS = aortic stenosis; Lp-PLA2 = lipoprotein-associated phospholipase A2; V_{peak} = peak aortic jet velocity.

AS severity class (p = 0.005) (Figure 2). For each unit of Lp-PLA2 activity (1 nmol/min/ml), patients had 19% increased risk of progressing to a more severe AS class (hazard ratio: 1.19, 95% confidence interval: 1.08 to 1.32, p = 0.001) (Figure 2). After adjustment for age, sex, degree of AV calcification, and baseline AS severity, Lp-PLA2 activity remained significantly associated with a higher risk of progression to a more severe class (hazard ratio: 1.16, 95% confidence interval: 1.02 to 1.31, p = 0.02) (Figure 2).

CORRELATES OF PLASMA LP-PLA2 ACTIVITY. Lp-PLA2 activity was associated with male sex (p = 0.04), larger BSA (p = 0.006), higher LDL cholesterol and triglycerides (both p < 0.001), as well as lower HDL cholesterol (p = 0.001) and fasting glucose (p = 0.04) (Table 3). There was a trend for an association between Lp-PLA2 activity level and younger age (p = 0.09). After adjustment for these variables, higher LDL cholesterol (p < 0.001) as well as lower HDL cholesterol (p = 0.001) and lower fasting glucose (p = 0.02) remained independently associated with higher circulating Lp-PLA2 activity (Table 3). There was no significant interaction (p > 0.20) between LDL cholesterol and Lp-PLA2 activity with regard to AS progression rate.

DISCUSSION

The main finding of this study is that increased plasma Lp-PLA2 activity is associated with a faster progression rate of AS in earlier stages of the disease. Lp-PLA2 mass was not associated with AS progression. This finding could be explained by the fact that there is an important interindividual variation in the Lp-PLA2 activity for a given Lp-PLA2 mass, and a previous study reported that Lp-PLA2 activity is, at least in part, genetically determined (10). It is thus possible that the enzyme activity, which takes into account both the mass and the genotype, provides a better marker of the impact of Lp-PLA2 on AS development and progression.

LP-PLA2 AND AS. We recently reported that Lp-PLA2 is highly expressed in AS valves and promotes ectopic valve mineralization (6). Lp-PLA2 uses oxidized phospholipids to produce LPC (11–13). We showed that Lp-PLA2 is transported in the AV by LDL particles and also secreted locally by macrophages and that the product of Lp-PLA2 activity, LPC, is present in AS valves (6). Tissue Lp-PLA2 activity also correlated with the mineral content of the AV. In vitro, we documented that LPC is a strong promoter of AV mineralization through both an osteogenic transition of cells and apoptosis. In the present study, we found

TABLE 2 Impact of Lp-PLA2 Activity and Mass on AS Progression Rate

	Whole Cohort (N = 183)		Mild AS Patients (n = 123)				Moderate/Severe AS Patients (n = 60)	
	Univariable Analysis		Univariable Analysis		Multivariable Analysis		Univariable Analysis	
	Beta-Coefficient ± SE	p Value	Beta-Coefficient ± SE	p Value	Beta-Coefficient ± SE	p Value	Beta-Coefficient ± SE	p Value
Annualized progression rate of peak aortic jet velocity, m/s/yr								
Lp-PLA2 activity, nmol/min/ml	0.08 ± 0.54	0.27	**0.21 ± 0.50**	**0.02**	**0.33 ± 0.63**	**0.005**	−0.05 ± 1.34	0.72
Lp-PLA2 mass, ng/ml	0.03 ± 0.04	0.71	0.11 ± 0.03	0.25		NA	−0.03 ± 0.09	0.81
Annualized progression rate of mean gradient, mm Hg/yr								
Lp-PLA2 activity, nmol/min/ml	0.02 ± 0.09	0.75	**0.17 ± 0.07**	**0.05**	**0.27 ± 0.09**	**0.02**	−0.10 ± 0.23	0.45
Lp-PLA2 mass, ng/ml	−0.02 ± 0.006	0.82	0.11 ± 0.005	0.19		NA	−0.10 ± 0.01	0.44
Annualized progression rate of aortic valve area index, $cm^2/m^2/yr$								
Lp-PLA2 activity, nmol/min/ml	−0.05 ± 0.001	0.48	**−0.19 ± 0.002**	**0.03**	**−0.30 ± 0.002**	**0.02**	0.05 ± 0.003	0.70
Lp-PLA2 mass, ng/ml	0.01 ± 0.001	0.85	0.05 ± 0.001	0.56		NA	−0.06 ± 0.001	0.66

Beta-coefficient is the standardized regression coefficient ± SE. The multivariate model is adjusted for age, sex, hypertension, diabetes, low-density lipoprotein cholesterol, statin therapy, creatinine level, bicuspid aortic valve phenotype, degree of aortic valve calcification, and aortic stenosis severity at baseline (i.e., peak aortic jet velocity, mean gradient, or aortic valve area index, respectively). **Bold** values indicate statistical significance.

AS = aortic stenosis; Lp-PLA2 = lipoprotein-associated phospholipase A2; NA = not applicable.

an independent association between Lp-PLA2 activity and faster AS progression in patients with mild AS but not in patients with more advanced AS. These findings support the hypothesis that lipid-mediated mineralizing processes may be predominantly involved in earlier stages of disease, as suggested by previous studies (14–20). In more advanced disease stages, further mineralization of the AV may be, in large part, determined by phosphocalcic factors (21,22).

In the present work, we found that higher levels of LDL cholesterol as well as lower levels of HDL cholesterol were independently associated with higher activity of circulating Lp-PLA2. These metabolic features have previously been associated with enhanced production, accumulation, and oxidation of lipids within the AV (2,4,18,19), which may in turn increase the production of substrate for Lp-PLA2 and thus the production of the pro-mineralizing LPC. The oxidized lipids-Lp-PLA2-LPC pathway may thus explain, at least in part, the previously reported associations between dyslipidemia, metabolic syndrome, diabetes, and AV calcification (17,23–28). However, although Lp-PLA2 activity was correlated with LDL cholesterol levels, the results of the multivariable analyses performed in this study suggest that the effect of Lp-PLA2 activity on AS progression is independent of LDL.

We previously reported that the proportion of circulating small, dense LDLs was associated with the accumulation of oxidized LDL within the AV (2). Small dense LDLs are prone to oxidation and carry oxidized phospholipids, which is the main substrate for Lp-PLA2 (29–31). Moreover, oxidized phospholipids are also transported and sequestered in the blood by lipoprotein(a) (32,33) and it has been shown in recent studies that plasma levels of lipoprotein(a) and corresponding genotypes were

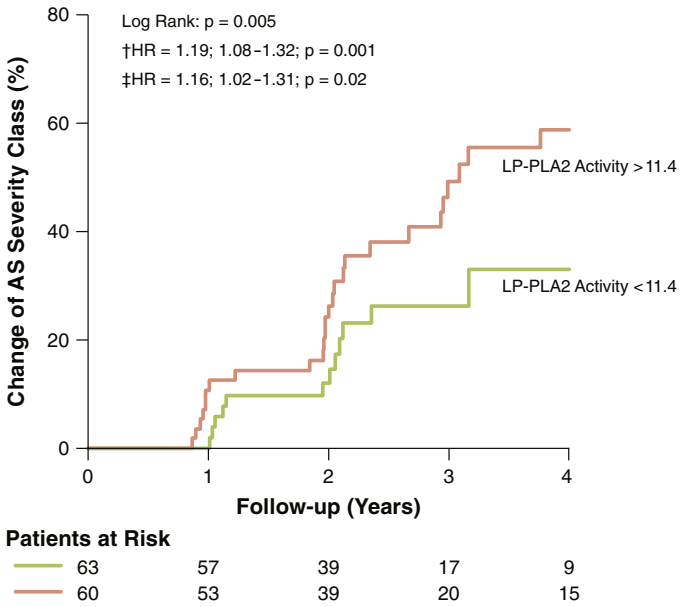

FIGURE 2 Comparison of the Time-to-Event Curve for the Change from Mild to Moderate or Severe AS According to a Median Value of Plasma Lp-PLA2 Activity

The Kaplan-Meier curves for the change in AS severity class (i.e., change from mild to moderate or severe AS defined as V_{peak} >3.0 m/s). †Univariable hazard ratio (HR) for increase in 1 nmol/min/ml. ‡Multivariate HR adjusted for age, sex, degree of aortic valve calcification, and baseline AS severity for increase in 1 nmol/min/ml. Abbreviations as in Figure 1.

TABLE 3 Correlates of Plasma Lp-PLA2 Activity

	Univariable Analysis		Multivariable Analysis	
Lp-PLA2 activity	**Beta-Coefficient ± SE**	**p Value**	**Beta-Coefficient ± SE**	**p Value**
Age, yrs	−0.13 ± 0.02	0.09	−0.01 ± 0.01	0.87
Male	0.16 ± 0.46	0.04	0.15 ± 0.50	0.07
Body surface area, m^2	0.20 ± 1.07	0.006	0.10 ± 1.17	0.19
Fasting glucose	−0.16 ± 0.15	0.04	**−0.15 ± 0.12**	**0.02**
LDL cholesterol	0.46 ± 0.23	<0.001	**0.51 ± 0.22**	**<0.001**
HDL cholesterol	−0.25 ± 0.53	0.001	**−0.24 ± 0.51**	**0.001**
Triglycerides	0.28 ± 0.27	<0.001	0.11 ± 0.24	0.10

Beta-coefficient is the standardized regression coefficient ± SE. **Bold** values indicate statistical significance.

HDL = high-density lipoprotein; LDL = low-density lipoprotein; Lp-PLA2 = lipoprotein-associated phospholipase A2.

associated with AS (34-36). Hence, several mechanisms, which are independent of the effect of statins, may promote the entry of lipids within the AV.

CLINICAL IMPLICATIONS. Statins failed to slow the progression of AS, even in early stages of the disease (37-40). This may be explained by the fact that several mechanisms, which are independent of the effect of statins or circulating LDL cholesterol levels, may promote the entry, retention, and oxidation of lipids within the AV (2,4,28,41). Although statins are very efficient in lowering the total LDL levels, they do not appreciably modify the proportion of small, dense LDLs or the lipoprotein(a) levels (24,42,43).

Our results, combined with those of recent studies lend support to the hypothesis that reducing Lp-PLA2 activity may reduce the lipid-mediated mineralization of AVs and thereby contribute to slowing the stenosis progression rate, particularly in patients with mild AS. Lifestyle modification aimed at increased physical activity and dietary changes may help to reduce visceral obesity and associated metabolic abnormalities including Lp-PLA2 activation (44). Direct inhibition of Lp-PLA2 activity with pharmacological agents such as darapladib may also be considered (36). However, recent randomized, controlled trials failed to demonstrate any significant benefit of darapladib in patients with acute coronary syndrome or stable ischemic heart disease (45,46). However, the cellular structure and biology are different in the AV versus the vasculature, and the mechanisms determining disease progression and clinical events also differ substantially between AS and atherosclerosis. In AS, the calcification of the AV is the predominant mechanism responsible for disease progression and occurrence of events, whereas in coronary artery disease, lipid deposition/modification and inflammation are the main causative mechanisms. The results of the present study as well as those of our previous experimental study (6) suggest that increased Lp-PLA2 activity may enhance the production of LPC within the valve and thereby induce mineralization of valvular tissues. Further trials are needed to assess the efficacy and safety of Lp-PLA2 inhibitors in patients with aortic sclerosis and/or mild AS.

STUDY LIMITATIONS. This study is subject to the inherent limitation of a prospective observational study. In particular, this design does not allow us to establish a causal relationship between Lp-PLA2 and pathogenesis of AS.

CONCLUSIONS

In this study, there was no significant association between plasma Lp-PLA2 activity or mass and stenosis progression in the whole cohort. However, increased Lp-PLA2 activity was a powerful independent predictor of a faster AS progression rate in patients with mild AS. These findings suggest that inhibiting Lp-PLA2 activity may be a valuable approach to slow AS progression and provide an impetus for the elaboration of a randomized trial to assess the effect of such intervention in patients with early stages of calcific AV disease.

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REPRINT REQUESTS AND CORRESPONDENCE: Dr. Philippe Pibarot, Institut Universitaire de Cardiologie et de Pneumologie de Québec, 2725 Chemin Sainte-Foy, Québec City, Québec G1V-4G5, Canada. E-mail: philippe.pibarot@med.ulaval.ca.

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KEY WORDS aortic stenosis, calcific aortic valve disease, Doppler echocardiography, lipoprotein-associated phospholipase A2

APPENDIX For a supplemental table and figure, please see the online version of this article.