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Characterization of coronary atherosclerosis in familial hypercholesterolemia and familial combined hypercholesterolemia patients utilizing intravascular ultrasound, coronary angiography and serum lipid profiles

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Characterization of Coronary Atherosclerosis in Familial Hypercholesterolemia and Familial Combined Hypercholesterolemia Patients Utilizing Intravascular Ultrasound, Coronary Angiography and Serum Lipid Profiles.

by

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ABSTRACT

Objectives: This study sought to compare the extent of atherosclerosis in coronary arteries of patients with heterozygous Familial Hypercholesterolemia (FH) and Familial Combined Hypercholesterolemia (FCH) using intravascular ultrasound, coronary angiography and serum lipid profiles.

Methods: Ninety-nine asymptomatic patients with heterozygous FH or FCH underwent angiography and intravascular ultrasound imaging of the Left Anterior Descending and/or Left Main Coronary Arteries. Two angiographic scores of diameter stenosis (Gensini and summation score) were derived for each patient. Intravascular ultrasound images obtained during catheter pullback underwent morphometric analysis. Plaque burden was expressed as mean and maximal intimal areas. Risk factors for coronary artery disease and serum lipid profiles were obtained for every patient.

Results: FH patients had significantly higher summation scores than FCH patients (FH=2.48±2.1, FCH=1.65±1.3, $p=0.012$). There was no difference in plaque burden as measured by intravascular ultrasound in the two patient populations ($p>0.3$). By stepwise multiple regression analysis, lipoprotein (a) level was the only significant predictor of angiographic scores in FH patients. In the FCH patients, predictors of angiographic measures were female gender and alcohol consumption, both negative. Female gender remained a significant, negative predictor of all intravascular ultrasound measurements in the FH cohort. In the FCH group, alcohol

consumption and high density lipoprotein cholesterol level were the only two predictors (negative) of ultrasound measurements.

Conclusions: Patients with heterozygous FH and FCH have the same degree of atherosclerosis early in the coronary disease process. In advanced disease, FH patients develop more severe atherosclerosis than their FCH counterparts. Lipoprotein (a) is a risk factor for advanced coronary artery disease in both populations. Women with FH have markedly less atherosclerosis than males with FH, perhaps mediated by a high-estrogen state. Alcohol has an anti-atherosclerotic effect in FCH but not FH patients. Ethanol's protective effect on the coronary arteries is possibly blunted in FH patients by high levels of low density lipoprotein cholesterol.

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INTRODUCTION

Familial hypercholesterolemia (FH) and familial combined hypercholesterolemia (FCH) are genetic disorders of lipoprotein metabolism which lead to accelerated atherosclerosis and premature cardiovascular events. The genetic defects underlying FH, one of the most common inborn errors of metabolism, are now well characterized as mutations affecting the structure of the LDL receptor, its synthesis and/or processing. Different and as yet unknown genetic defect(s) govern the FCH phenotype. Both diseases lead to significant elevations in LDL cholesterol levels and a predilection for early plaque accumulation, particularly in the coronary, carotid and iliac vessels [1]. Unique to FCH is a marked elevation in total triglyceride level. Recent studies indicate that in addition to LDL and HDL, TG levels independently predict coronary risk [2]. Thus, although patients with FH and FCH ultimately have similar clinical symptoms of coronary artery disease (CAD), they may have significantly different presentations of early atheroma formation and disease progression.

Intravascular ultrasound (IVUS) is a novel imaging technique which provides detailed cross-sectional images of the epicardial coronary arterial wall in vivo and allows quantitation of atherosclerosis and plaque burden [3, 4, 5, 6]. Few studies have utilized IVUS to examine the coronary disease of patients with genetic hyperlipidemias. IVUS analysis has confirmed angiographic findings of an increased prevalence of coronary ectasia in FH patients [7]. The efficacy of cholesterol lowering agents has been monitored with IVUS in a limited number of patients [8]. In a recent study of asymptomatic patients with FH and FCH, IVUS detected disease in a significant portion of patients who had minimal or no angiographic changes [9]. However, no studies to date have directly compared the morphology of the coronary arteries of FH and FCH patients utilizing either IVUS or fiberoptic angioscopy. In this study, we detail the severity of atherosclerosis and its relationship to coronary risk factors using IVUS and quantitative angiographic imaging techniques in 99 asymptomatic patients with heterozygous FH and FCH.

PATIENTS AND METHODS

1) Patients. Ninety-nine patients with heterozygous FH or FCH were recruited from the Lipid Clinic of the University of California, San Francisco. Patients diagnosed with FH had to have tendon xanthomas on physical examination or had a first degree relative with tendon xanthomas. In addition, FH was diagnosed in patients with lipoprotein profiles of LDL cholesterol greater than 200 mg/dL (5.17 mmol/L) and total triglyceride level less than 275 mg/dL (3.1 mmol/L). Criteria for diagnosis of heterozygous FCH included serum LDL levels less than 250 mg/dL, total TG levels greater than 200 mg/dL (3.1 mmol/L) and absence of tendon xanthomas on examination. Patients homozygous for apolipoprotein E-2 and patients who had known diseases producing secondary hyperlipidemia were excluded from this study. Any patient with symptoms of coronary artery disease or a history of systemic disease other than hypertension was excluded as well.

A detailed medical history was obtained, including a history of smoking, alcohol consumption, diabetes, hypertension and the use of prescription and/or over-the-counter medications. Weight and blood pressure measurements from three consecutive clinic visits were

averaged for each patient. Following the approval of the study by the University of California, San Francisco, Committee on Human Research, all patients were given a written consent prior to participation.

2) Angiography. Coronary angiography was performed via the femoral artery approach utilizing 7F catheters. Cineangiograms were filmed at 60 frames per second through a 135mm focal length lens, with an X-ray field of 15cm. Multiple frames of orthogonal views of the left and right coronary arteries were obtained. All films were viewed at x5 magnification (Vanguard Instruments) by two angiographers (KJH and T-ML) without knowledge of the patient's clinical history or IVUS data. Images of lesions identified on angiogram were captured for display using a high resolution video card (Scion, Inc.) onto a video monitor and Macintosh PowerPC computer (Apple, Inc., Cupertino, CA). Images were captured during end-systole, defined as the frame with the heart occupying the smallest area. Only lesions present throughout at least one cardiac cycle were captured. The location and percent diameter stenosis (diameter of stenotic segment/diameter of adjacent reference

segment) of each lesion was recorded. Vessel measurements were made utilizing public domain NIH image analysis software, NIH Image Version 1.55 (National Institutes of Health, Bethesta, MD) modified for this specialized purpose [10]. Using the modified Gensini Score [11], lesions were scored and summed for each patient. A second within-patient score, the summation score of the sum percent of lesions, was derived for each patient as well [9].

3) IVUS. Patients with Left Main or Left Anterior Descending artery luminal narrowing estimated to be less than 40% during angiography underwent 2-dimensional IVUS analysis utilizing commercially available, 4.3F, 3.2F and 2.9F, 30-MHz. ultrasound catheters (Boston Scientific Corp., Sunnyvale, CA). The theoretical axial and lateral resolution of the transducer was 80μm and 150μm, respectively. After treatment with 200mcg intracoronary nitroglycerin and 5000 units intravenous heparin, an 8F guiding catheter was placed at the left main coronary osteum. The IVUS catheter was advanced over a 0.014 inch guide-wire into the mid-portion of the left anterior descending artery under fluoroscopic guidance. The ultrasound transducer was then withdrawn at a

constant rate of 1mm/sec using a motorized, timed, automated pullback device. A series of cross sectional images of the Left Main and/or Left Anterior Descending arteries were displayed and ultrasound contrast, gain and reject settings were adjusted to produce well-balanced, gray scale images. The real-time ultrasound images and a concurrent standard electrocardiogram (lead II) were recorded on high quality, 0.5-in sVHS video tape for subsequent off-line analysis.

Image analysis was described elsewhere in detail [12]. In brief, the videotape was replayed on a video monitor to identify segments of interest. Septal, diagonal and the circumflex artery branch points were utilized as reference points for identifying the Left Main and Left Anterior Descending artery segments on ultrasound. Images obtained during the first 5 seconds of pullback were discarded due to possible variations in the speed of the catheter at the beginning of pullback. Videotape segments meeting these criteria were digitized into a 600 x 480-pixel matrix image with 8 bit depth (256 gray levels, Raster Ops) and stored on a Macintosh PowerPC personal computer (Apple, Inc., Cupertino, CA).

Frames were retrieved for qualitative (plaque composition and morphology) and quantitative (cross-sectional) analysis by an ultrasonographer not familiar with the patient's clinical history or angiographic findings. The in-vitro validation of IVUS analysis was reported elsewhere [4, 13, 14]. Morphometric analysis using NIH Image was performed. After calibration of the planimeter to cross-hair markers on the imaging screen, the vessel wall was examined for presence of plaque. A normal wall was defined as one with no three-layered appearance or one which had three layers but an intima of less than 200μm [15]. The lumen border, identified as the inner border of echo-dense plaque, and the internal elastic lamina (IEL), identified as the area between the echo-dense intima and the relatively echo-lucent media, were traced with the planimeter. IEL and lumen areas were then measured, and plaque area was calculated as the difference between IEL and lumen. Using the geometric lumen center as the reference, 360 equally spaced radial measurements were made to calculate maximal and mean intimal thickness.

4) Lipid quantitation. Within one month of angiography and IVUS catheterization, blood was obtained from every patient following a

10 to 16-hour fast. After separating the serum at room temperature, total cholesterol and triglyceride contents were measured and lipoprotein concentrations were determined by sequential preparative ultracentrifugation [16]. Concentrations were measured for the cholesterol and triglyceride content of low density lipoproteins (LDL), very low density lipoproteins (VLDL) and high density lipoproteins (HDL). The plasma content of Lp(a) was measured by an ELISA technique in which the apo B-100 moiety of the complex was quantified in molar terms. Analyses were standardized against reference material provided by the Standardization of the National Center for Disease Control.

5) Statistical analysis. Statistical analysis was performed with SAS Version 6.04. Qualitative data are presented as frequencies. Quantitative data are presented as mean ± 1SD. To test for differences of mean values of continuous variables between subgroups, paired and unpaired Student's t-tests were utilized. Univariate and multivariate regression analysis was used to find the best predictors of angiographic and IVUS measurements of atherosclerotic disease. The following risk factors were considered

for predictor variables: age, gender, smoking history (never, past, present), mean arterial pressure, body mass index, history of hypertension and alcohol consumption. For "Stage 1" analysis on candidate sets of predictor variables, the subset regression model having the minimum value of Akaike Information Criteria Corrected (AICC) was selected [17]. The model with the minimal AICC may be interpreted as the most likely model given the data and candidate predictors examined. Once the preliminary first order model was selected, product terms were added to examine interaction effects, after which a final minimum AIC model was determined. Residual plots, partial regression plots and Cook's distance were examined to check analysis assumptions and detect outliers. Square root transforms of variables were examined because of poor fitting of the larger values on the original scale. However, transformation yielded similar results as the original data. For simplicity, we present raw data rather than the square root transformed variables.

For "Stage 2" multiple regression models, first order predictors and lipid measurements (total cholesterol, HDL cholesterol, LDL cholesterol, VLDL cholesterol, non-HDL cholesterol (total minus HDL cholesterol), Lp(a) and total triglycerides) were

considered. Predictors were selected in a step-wise procedure. Univariate variables with a p-value < 0.15 were entered into and remained in the multivariate model. A value of $p<0.05$ was considered statistically significant.

RESULTS

Patients. Ninety nine patients (48 men and 51 women; mean age ±SD: 49±10 years, range 28 to 71) with FH or FCH participated in this study. The baseline clinical characteristics of the patients are presented in Table I. Baseline characteristics were not statistically significant between the two diagnostic groups by Student's two sample T-test except for mean age (FCH=52±9, FH=47±11; p=0.016). In the FH patient population (n=55), the mean serum level of fasting LDL cholesterol was 284 mg/dL and the mean total TG level was 155 mg/dL (Table II). FCH patients (n=44) had a mean serum LDL level of 218 mg/dL and mean total TG level of 212 mg/dL. None of the 99 patients had any systemic illness including diabetes mellitus or any other known disease leading to secondary hyperlipidemia. No patient smoked at the time of the study. 20 patients carried the diagnosis of hypertension.

Angiography. Angiograms of the left and right coronary arteries were available for 96 patients. A total of 736 lesions were identified and measured. Patients with FH had significantly higher summation scores than FCH patients (2.48±2.1 in FH, 1.65±1.3 in FCH, p=0.012; Table III). Gensini scores for the two patient populations approached statistical significance (p=0.087). Table IV details the angiographic findings. 69 (9%) lesions had lumen diameter stenoses of >50%. Of the 69 patients; 77% had the diagnosis of FH. 16 lesions had >70% stenosis; 74% of these patients were FH patients. No patient had Left Main artery stenosis of >50%. On univariate regression analysis, the predictors of Gensini Scores and summation scores were gender (Table V) and alcohol consumption (figures 5 & 6), both negative. In multivariate analysis, the relationship between risk factors and angiographic measures of atherosclerosis were examined separately for the two diagnostic groups.

For the FCH patient population, univariate predictors of alcohol and gender remained significant in multivariate analysis (Table VI). Gender effect, in particular, was quite large for both angiographic measures.

Table VII shows the predictors of Gensini Score and summation score in the FH patient population. For the FH patients, mean arterial pressure, non-HDL cholesterol and Lp(a) remained significant on multivariate analysis ($p<0.05$). In contrast to the FCH population, gender and alcohol were not significant predictors of angiographic scores in the FH patients.

Intravascular Ultrasound. Predictors for the following two IVUS variables were examined with univariate and multivariate regression analysis: 1) maximal intimal area, 2) average intimal area. These two IVUS parameters were examined in the Left Anterior Descending (n=84) and/or Left Main (n=82) coronary arteries of study patients.

Left Main Artery

In the Left Main artery, plaque burden as measured by IVUS was not significantly different between FH and FCH patients ($p > 0.3$). Statistically significant predictors of maximum intimal area and average intimal area for the Left Main artery on univariate analysis

were: age, age-by-gender interaction effect, diagnostic group-by-gender interaction effect and gender (Figures 1 & 2, Table IX).

In multiple regression analysis within the FCH patient population, maximal and mean intimal area measurements were correlated inversely with alcohol intake (Table VI). In addition, there was significant correlation of maximum intimal area with non-HDL cholesterol and VLDL triglyceride. Serum HDL levels were inversely correlated with maximum intimal area but not average intimal area measurements.

In the FH patients, larger maximum intimal area measurements were associated with male gender and mean arterial pressure. The average intimal area measure was predicted by male gender, mean arterial pressure and serum Lp(a) levels (Table VII).

Left Anterior Descending Artery

As in the Left Main artery, IVUS measurements of plaque burden in the Left Anterior Descending artery were not significantly different between the two diagnostic groups ($p > 0.3$). In univariate analysis, gender was the only predictor of the IVUS parameter, maximum intimal area, for the Left Anterior Descending artery

(Table IX). Predictors for the average intimal area parameter in the Left Anterior Descending artery were gender and diagnostic group-by-alcohol consumption interaction effect.

In multivariate analysis, significant predictors of maximum intimal area and average intimal area within the FCH patient group were female gender, alcohol consumption and HDL cholesterol (Figures 3 & 4), all negative (Table VI). In the FH patient population, female gender inversely related to both IVUS measurements (Figures 1 & 2). In addition, in the FH group, alcohol consumption was negatively correlated with maximum intimal area measurements and mean arterial pressure was positively correlated with average intimal area values (Table VII).

DISCUSSION

In this study, we examined the coronary arteries of asymptomatic patients with heterozygous FH and FCH utilizing traditional angiographic measurements and the more detailed imaging provided by IVUS. The results of this study indicate that: 1) by angiography, FH patients have more severe coronary atherosclerosis than FCH patients; however, by IVUS measurements, these is no difference in

plaque burden between the two patient populations 2) elevated Lp(a) level is associated with more severe disease as measured by cineangiography but not IVUS in the FH patient population 3) the female gender offers significant protection from CAD in the FH patient population; in contrast, the women in the FCH group experience significantly less protection 4) FCH patients who consume alcohol have markedly less atherosclerosis by all angiographic and IVUS measurements than those who abstain from alcohol. This difference is not seen in the FH cohort.

Atherosclerosis in FH and FCH patients

Our results indicate that patients with heterozygous FH have significantly more atherosclerosis by angiographic summation score than patients with FCH. In addition, correlation of CAD severity by Gensini score in FH patients approached significance. This is not surprising, as FH patients have markedly higher plasma total- and LDL-cholesterol levels than their FCH counterparts, and both total- and LDL-cholesterol are established risk factors for advanced CAD [18, 19]. However, our study failed to demonstrate a difference in plaque burden by IVUS between the two patient populations. This

suggests that early in the disease process, patients with both hyperlipidemias have similar manifestations of coronary atherosclerosis. Later, when the lesions are detectable by angiogram, atherosclerosis becomes significantly more severe in the FH population. Therefore, early atheroma formation may be independent of serum total cholesterol, LDL cholesterol or triglyceride levels, which are the distinguishing features of the two diagnostic groups.

An additional risk factor, Lp(a), was an independent predictor of advanced disease in our FH patient population on multiple regression analysis. Lp(a) is a recently identified risk factor that has been associated with coronary atherosclerosis in many but not all studies [20, 2, 21, 22, 23, 24]. In the only study to date of FH patients examining the relationship between angiographic disease and Lp(a) levels, there was no difference in serum Lp(a) levels between FH patients with and without CAD [25]. Moreover, cardiovascular risk in the FH patients was not related to Lp(a) levels. However, in the present study, we use a new, highly reproducible and accurate ELISA technique to quantify serum Lp(a) levels. The increased accuracy of Lp(a) measurements may account

for the significant association between Lp(a) and angiographically detected atherosclerosis in our FH and FCH patients.

Our finding that FH patients have significantly more severe atherosclerosis by angiography than their FCH counterparts may be explained by the interaction of high circulating serum LDL cholesterol with Lp(a). LDL and Lp(a) have been found in high concentrations in atheromas; both are implicated in plaque formation via increased influx [26] and decreased fractional loss of LDL from the intima [27]. Lp(a) may also accelerate the delivery of cholesterol to the injured blood vessel [28]. One prospective study of FH patients suggested that Lp(a) pathogenesis may be dependent on elevated LDL levels [29]. Treatment of 20 hyper-LDL patients with apharesis resulted in a concomitant decrease of Lp(a). Thus, although Lp(a) was associated with angiographically detectable atherosclerosis in both FH and FCH patients in the present study, the interaction may be less dramatic due to lower levels of LDL cholesterol in the FCH phenotype. This may account for the lower Gensini and summation scores found in the FCH cohort.

Gender-protection in FH

Our study also found a strong negative association between female gender and atherosclerosis as measured by both angiography and IVUS. Interestingly, this "gender protective" effect was far better defined in the FH diagnostic group than the FCH group. Although the relatively high estrogen state of the female gender has been shown to exert substantial protection from symptomatic CAD in many large, prospective studies [30], this is the first study to highlight the difference between males and females in a genetically hyperlipidemic cohort.

We found a disproportionately greater effect of estrogen on atherosclerosis in the FH women as compared to the FCH women. Perhaps the hyperlipidemia associated with FH, that of marked LDL-cholesterol elevation, may play a role in mediating the effects of estrogen on CAD progression in this population. Interestingly, in non-genetically hypercholesterolemic women, the protective effects of estrogens were found to be more pronounced [31]. The age-adjusted mortality in estrogen users in one study was 13.1 (per 10,000) in normolipidemic women and 8.2 in hyperlipidemic women. In non-estrogen users, the adjusted mortality rate was 27.1 and 38.7

in normolipidemic and hyperlipidemic women, respectively. Thus, the elevated serum LDL levels that characterize the FH phenotype may account for the marked estrogen-mediated protection found in the FH but not FCH women. On the other hand, one may also speculate that elevated triglycerides in FCH women suppress levels of HDL-cholesterol and therefore counter the beneficial effects of estrogen on lipids. However, after controlling for lipid levels by multiple regression analysis, the FCH females in our study continued to have higher angiographic and IVUS scores in the Left Anterior Descending artery artery as compared to FH females.

Another explanation for this difference in protection may be related to the hypertriglyceridemia that is the defining phenotype of FCH. In the Framingham study, elevated triglyceride level, although only a weak risk factor in men, was a strong, independent risk factor for coronary artery disease in women [32]. Triglycerides may have direct, not yet elucidated atherosclerotic properties, which counteract the beneficial effects of estrogen in the FCH females. Finally, the difference in gender-protection between FH and FCH women may be explained by the lack of compensatory dilation in the FCH population. Endothelium-dependent vasodilatation improves

with the administration of physiological levels of 17-beta estradiol in peri- and post-menopausal women [33, 34]. Further, there is now considerable evidence that estrogens effect the vascular smooth muscles and endothelium through nitric oxide (NO) via an estrogen receptor [35]. Nitric oxide is a potent endothelium-derived vasodilator. This augmentation of vasodilatation by estrogen via NO is seen in healthy females as well as females with angiographic evidence of atherosclerosis [34]. Perhaps the genetic defect underlying the FCH genotype interferes with the function of the endothelium, including compensatory dilatation, thereby negating estrogen's beneficial, vasodilitory effect on the coronary arteries. Future studies comparing vasoactivity in FCH and FH women may help shed light on the coronary disease process in the female gender.

The mechanism of estrogen's protection is unclear. Numerous studies have examined the effect of endogenous and exogenous estrogen on CAD. One prospective, case controlled study showed a direct effect of female sex hormones on Lp(a) levels [36]. Pre-menopausal or post-menopausal women with estrogen replacement therapy (ERT) had decreased concentrations of Lp(a) as compared to post-menopausal women not on estrogen therapy. In addition, Lp(a)

levels increased after bilateral oophorectomy and returned to baseline after the initiation of ERT. ERT also decreases mortality and morbidity from CAD in women by 25% to 50%, likely from reversing the increase in atherogenic lipids after the onset of menopause [37,38,39,40]. The menopausal status of our FH and FCH patients was unfortunately not available at the time of this study. However, more detailed information about the age of menarche and/or menopause and any hormonal therapy in our cohort will help clarify the role of estrogens in genetically hyperlipidemic women.

Of note, the mean age of the FCH women in our study was almost a decade more than their FH counterparts. Although multiple regression analysis controlled for this age difference, more FCH women are expected to be post-menopausal, losing the protective effects of endogenous estrogen. If this is the case, one might hypothesize that circulating estrogens are necessary for its anti-atherosclerotic effect at all times, because even relatively brief periods without estrogen (a few years) cause significant acceleration of atherosclerosis.

Cardio-protection by alcohol

Alcohol consumption is inversely and significantly associated with atherosclerosis in the FCH cohort in multivariate analysis of all angiographic and most IVUS measurements in this study. This finding supports epidemiologic [41,42,43,44,45,46], histopathologic [47] and autopsy [48] studies of the cardio-protective effects of low to moderate alcohol consumption. However, no study to date has examined this inverse relationship between alcohol and coronary artery disease in genetically hyperlipidemic patients.

The mechanism of alcohol's anti-atherogenic action is unclear and controversial but is believed to be mediated through elevated HDL-C levels induced by ethanol [49,50,51]. The inhibition of platelet aggregation may be a second mechanism [52,53,54]. Furthermore, ethanol decreases serum levels of Lp(a), factor VIII and vonWillebrand factor [55, 56, 57], which are considered possible risk factors for atherosclerosis. Alcohol also increases apolipoprotein A-I levels in human fibroblasts *in vitro* [58, 56]. Apolipoprotein A-I, the major protein of HDL, is believed to provide cardio-protection through modulation of HDL-mediated efflux of cholesterol from human fibroblasts [58]. Finally, Bello *et al* showed that the B1-

isoenzyme of alcohol dehydrogenase is expressed in blood vessels [59]. The authors hypothesize that alcohol dehydrogenase, which produces reductive NADH molecules when stimulated by alcohol, may counteract lipoprotein oxidation. This intriguing finding may provide a direct link between alcohol and its anti-atherogenic properties.

Interestingly, alcohol did not remain a significant predictor of atherosclerosis in the FH population on multivariate analysis in the present study. Perhaps the severe hyperlipidemia associated with the FH phenotype may offset the beneficial effects of alcohol in this population. Or, because the female gender offers such a large magnitude of protection in the FH population, the influence of alcohol remains hidden in statistical analysis. Future studies which employ detailed alcohol histories, including length of alcohol consumption, initial age of alcohol consumption, type of alcohol consumed and binge versus consistent drinking will help elucidate the role of ethanol in cardiovascular disease progression.

Limitations of coronary angiography

Although angiography has long been considered the "gold standard" in coronary vessel visualization, recent studies with in vivo intracoronary ultrasound devices indicate that angiography significantly underestimates the severity of atherosclerosis, particularly in early disease [60, 61, 9]. Haussman *et al* showed, in a subset patient population of this study, that angiograms detect luminal narrowing in only 30% of Left Anterior Descending artery vessel segments, while IVUS detects atherosclerotic plaque in 89% of the same segments [9]. The authors hypothesize that compensatory vessel enlargement early in the disease process may account for these strikingly different results. Nishioka reports compensatory enlargement of the coronaries in 54% of patients studied with IVUS [62]. Coronary dilation in response to plaque deposition helps preserve coronary flow and maintain low coronary pressures [63, 64]. However, it may eventually lead to structural weakening of the vascular connective tissues. In fact, pathologic thinning of the vessel media and marked coronary ectasia have been documented in advanced atherosclerosis [65, 7]. Thus, dilation of the coronary arteries to preserve lumen area, a relatively common phenomenon in

hyperlipidemic patients [7], is likely to result in false negative cineangiography results.

Studies of plaque eccentricity further highlight the limitations of angiography and the strengths of IVUS in characterizing coronary artery disease [6]. One study of 1446 lesions indicated that angiography was particularly poor in detecting plaque eccentricity as compared to IVUS, with a concordance rate of only 50% between these two imaging modalities [66]. Because an eccentric lesion preserves the lumenal area more so than a concentric lesion, angiography is less likely to detect eccentric lesions. However, eccentric lesions may represent early atherosclerosis that later develop into clinically symptomatic stenoses. Thus, the utilization of IVUS is particularly important in assessing the coronary vasculature of asymptomatic patients such as those of this study, where such eccentric, "young" lesions are likely to be prevalent.

In this study, we utilize IVUS to measure mean and maximal intimal areas in the left anterior descending and left main coronary arteries in genetically hyperlipidemic patients without symptoms of coronary artery disease. We expect our patient population to have

more severe atherosclerosis than asymptomatic, normolipidemic patients. However, in the continuum of atherosclerotic disease, these patients are likely to have "early" atherosclerotic disease as compared to genetically hyperlipidemic patients *with* symptoms of CAD [8]. IVUS provides accurate, anatomical, cross-sectional images of early lesions; thus, it is the ideal tool for examining in detail the coronary vasculature of our patients.

CONCLUSIONS AND FUTURE DIRECTIONS

In this study, we found profound differences in atherosclerotic disease between FH and FCH patients undergoing coronary angiography and IVUS. FH patients had significantly more advanced disease than FCH patients by angiography, but had no difference in plaque burden measured by IVUS. Lp(a) was an independent risk factor for atherosclerotic disease measured by angiography in FH patients. On the other hand, the female gender offered significant cardio-protection for the FH women compared to the FH men. This protection was not seen to the same degree in the FCH women. Finally, alcohol had a strong, negative association with angiographic

and IVUS measurements of CAD in the FCH but not the FH patient population.

Larger, prospective studies of genetically hyperlipidemic patients examining the effects of estrogen and alcohol are needed to confirm the above findings. Angiographic and IVUS comparisons of patients treated with lipid lowering agents will help elucidate the role of Lp(a) in atherogenesis. Long-term IVUS follow-up studies of FH and FCH patients will allow investigators to track the development of atherosclerosis, from the early, angiographically "silent" to advanced, clinically symptomatic stages of coronary artery disease. Ultimately, one hopes to gain better understanding of the mechanisms underlying atherosclerosis in order to develop more effective treatment modalities, and ultimately, a potential cure for these unfortunate patiens with genetic hyperlipidemia.

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Table I

Baseline Characteristics of the 99 Study Patients*

	# of patients	*FH*	*FCH*
All patients		55 (56%)	44 (44%)
Men	48 (48%)	24 (44%)	23 (52%)
Age (yr)	49±10	47±11	52±9
Hypertention diagnosis	20 (20%)	9 (16%)	11 (25%)
Tobacco history			
current	0	0	0
past	32 (32%)	16 (29%)	16 (29%)
pack year history	6±11	5.8±11	6.3±12
Alcohol consumed (oz/week)	4±6	4±6	4±6

* Two-sample T-test showed no statistical significance in baseline characteristics between diagnostic groups except in age ($p<0.05$). FH: familial hypercholesterolemia, FCH: familial combined hypercholesterolemia.

Table II

Baseline Levels of Serum Lipids in the 99 Study Patients

*Cholesterol**	*Heterozygous FH (mean±SD)*	*FCH (mean±SD)*	*p-value*
Total	363±64	308±45	0.000
HDL	52±17	53±17	0.87
VLDL	25±24	38±27	0.018
LDL	284±62	218±33	0.000
Triglycerides			
Total	156±88	212±93	0.003
HDL	18±8	28±39	0.12
VLDL	85±68	140±69	0.002
LDL	49±20	49±17	0.93

* all values are in mg/dL

HDL: high density lipoprotein; LDL: low density lipoprotein, VLDL: very low density lipoprotein; FH: Familial Hypercholesterolemia; FCH: Familial Combined Hypercholesterolemia

Table III

Angiographic findings, statistics

	FH (n=53)	*FCH (n=43)*	*p*
Gensini score	22±20	16±16	0.087
Summation score	2.48±2.1	1.65±1.3	0.012

Table IV

Angiographic findings in 736 lesions

Number of Lesions >70% (n=16)	13 (n=8)	3 (n=2)
Number of Lesions >50% (n=69)	53 (77%)	16 (23%)
Number of Patients with >50% lesions	20 (74%)	7 (26%)
Number of LM lesions >50%	0	0
Number of LAD lesions >50%	17 (n=10)	3 (n=3)
Number of LCx lesions >50%	6 (n=6)	3 (n=2)
Number of OM lesions >50%	10 (n=8)	3 (n=3)
Number of RCA lesions >50%	15 (n=11)	5 (n=3)
Other lesions*	5	2

* Diagonal, Septal, Posterior Descending, Postero-lateral Arteries.
Abbreviations: LM: Left Main Artery, LAD: Left Anterior Descending Artery, Lcx: Left Circumflex Artery, OM: Obtuse Marginal Artery, RCA: Right Coronary Artery.

Table V

Effect of gender on angiographic measures of atherosclerosis

	female	male	p-value
Gensini score	16.4	22.3	0.02
Summation score	1.88	2.36	0.03

Table VI

Selected Predictors of Atherosclerosis in FCH patients: Multivariate Analysis

	FH	Female	Alcohol	HDL-C
Gensini score	* *	-12.424	-0.966	*
Summation score	* *	-1.099	-0.070	*
MIA in LM	#	* *	-0.245	-0.081
AIA in LM	#	* *	-0.164	*
MIA in LAD	#	-2.315	-0.246	-0.089
AIA in LAD	#	-1.759	-0.178	-0.045

**= significant on univariate analysis but not significant on multivariate analysis. #= p value > 0.3. * = not significant (p>0.05); MIA: maximum intimal area, AIA: average intimal area, LM: left main coronary artery, LAD: left anterior descending coronary artery, FH: familial hypercholesterolemia, HDL-CL high density lipoprotein cholesterol.

Table VII

Selected Predictors of Atherosclerosis in FH patients: Multivariate Analysis

	Lp(a)	Female	Alcohol	HDL-C
Gensini score	0.043	*	*	*
Summation score	0.003	*	*	*
MIA in LM	*	-3.150	*	*
AIA in LM	0.007	-3.058	*	*
MIA in LAD	*	-3.691	0.183	*
AIA in LAD	*	-3.178	*	*

Abbreviations as in Table V.

Table VIII

Effect of gender on IVUS indices of plaque burden:

Left Main Coronary Artery

	female	male	p-value
Maximum intimal area*	3.43	5.67	0.006
Average intimal area*	2.21	4.24	0.002

*= cm^2

Left Anterior Descending Artery

	female	male	p-value
Maximum intimal area*	3.97	6.78	0.0003
Average intimal area*	2.31	4.43	0.0001

*= cm^2

Table IX

Selected Predictors of Atherosclerosis on Univariate Analysis

	FH	Female	Alcohol	Age
Gensini score	3.107	-4.196	-0.581	*
Summation score	0.453	-0.394	-0.070	*
MIA in LM	*	-1.331	*	*
AIA in LM	*	-1.243	*	0.080
MIA in LAD	*	-1.651	*	*
AIA in LAD	*	-1.247	*	*

* = not significant ($p>0.05$); MIA: maximum intimal area, AIA: average intimal area, LM: left main coronary artery, LAD: left anterior descending coronary artery, FH: familial hypercholesterolemia.

Figure 1

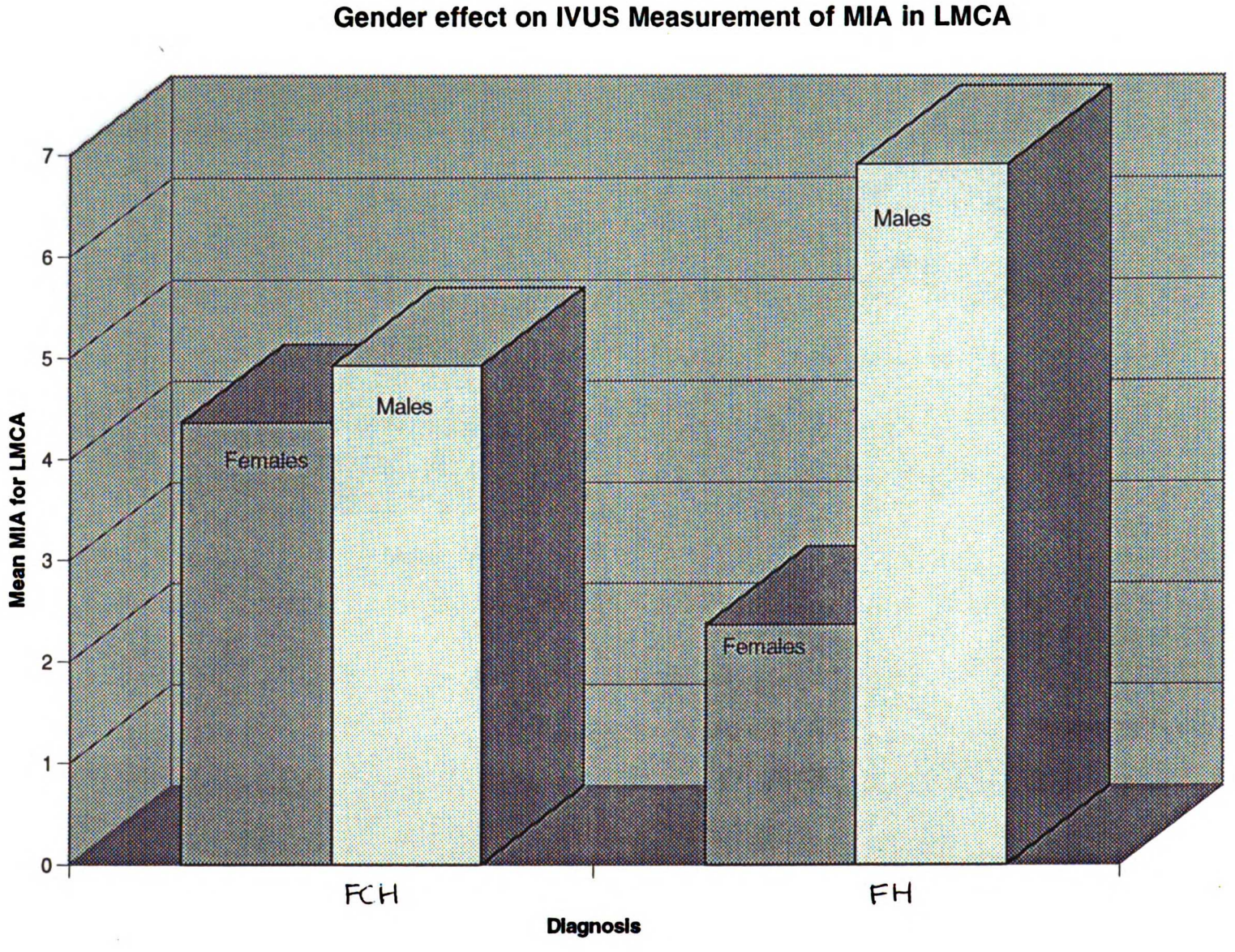
Gender effect on IVUS Measurement of MIA in LMCA
Mean MIA for LMCA
0
1
2
3
4
5
6
7
Females
Males
Females
Males
FCH
FH
Diagnosis

Figure 2

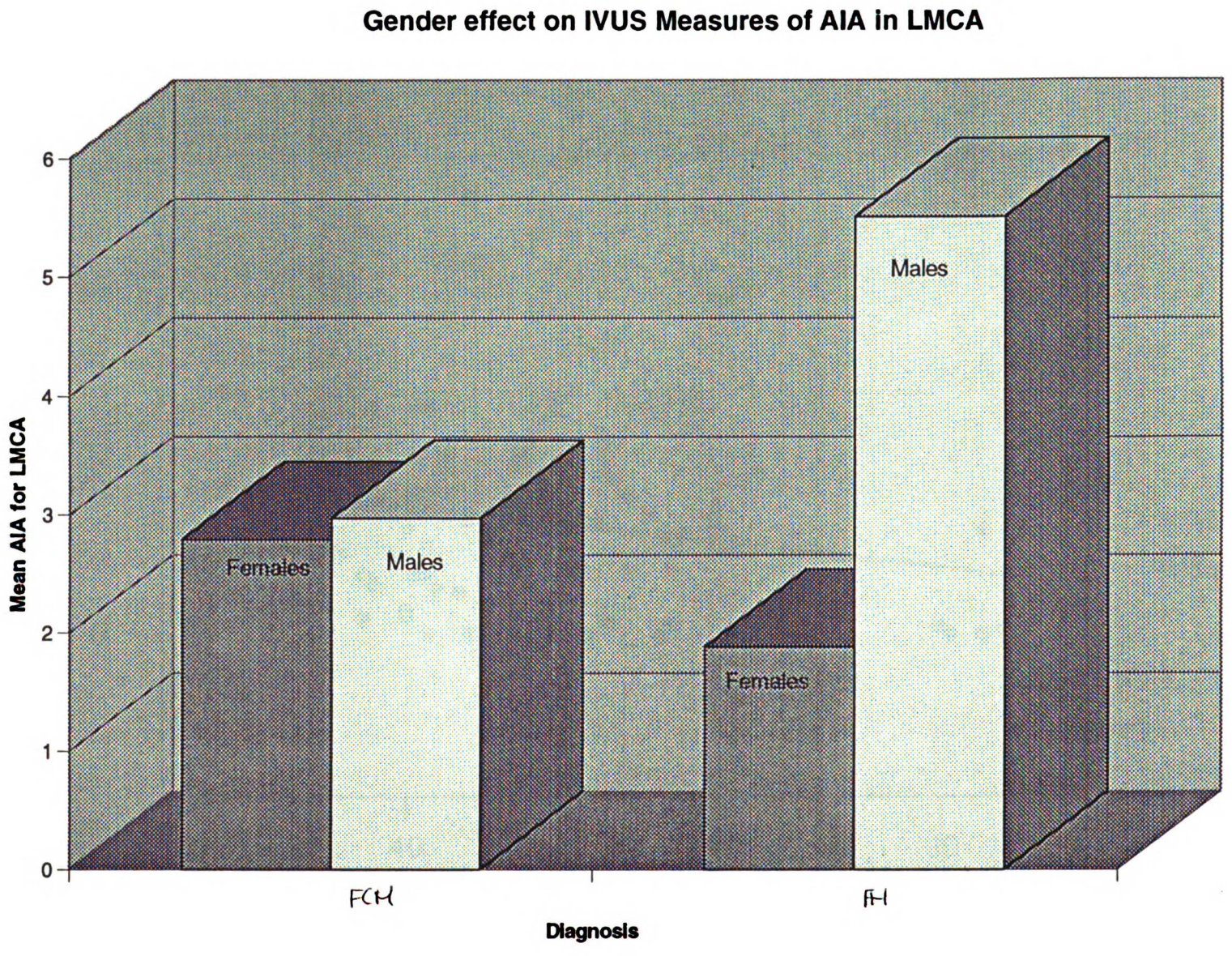
Gender effect on IVUS Measures of AIA in LMCA
Mean AIA for LMCA
0
1
2
3
4
5
6
Females
Males
Females
Males
FCH
FH
Diagnosis

Figure 3

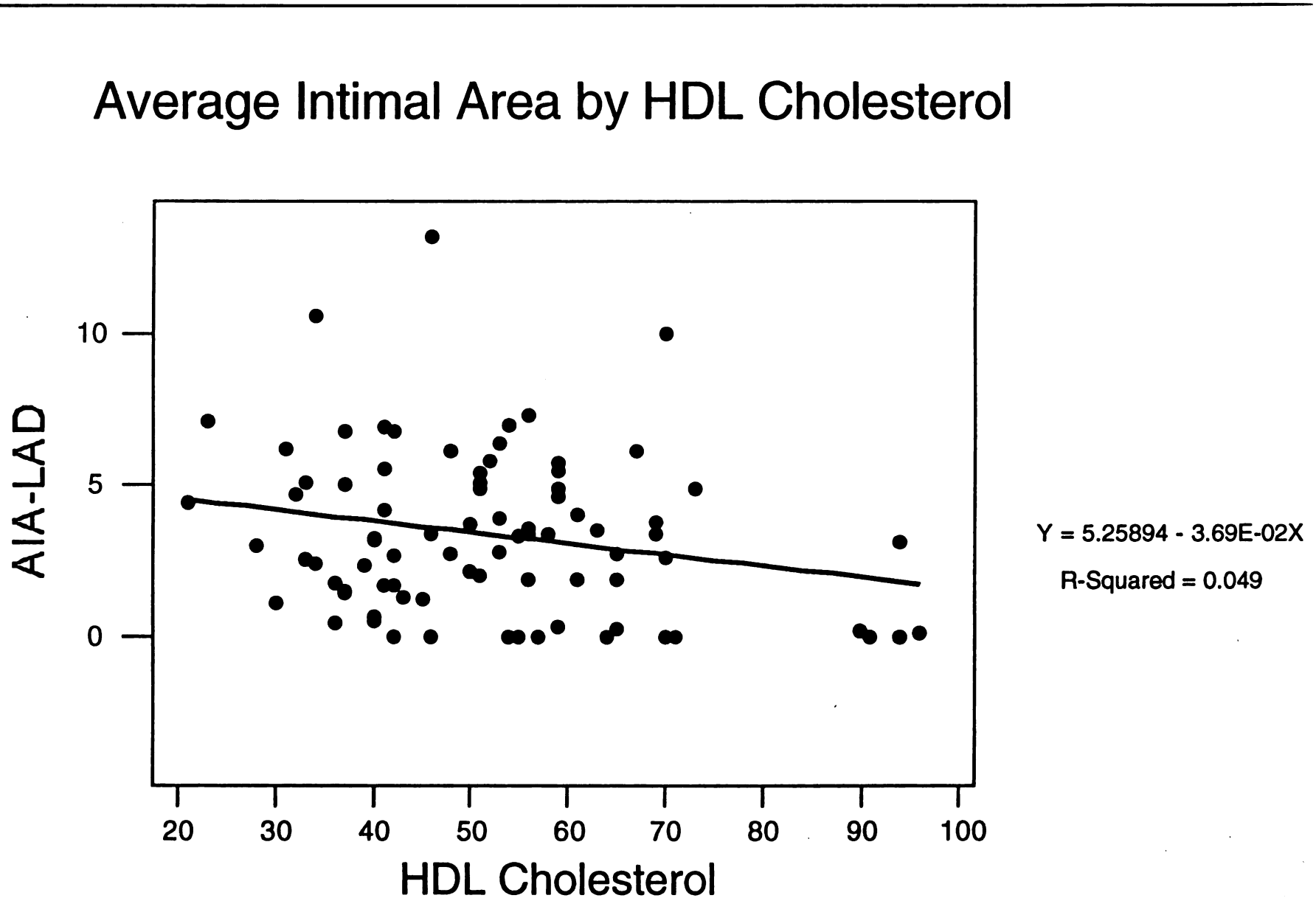
Average Intimal Area by HDL Cholesterol
AIA-LAD
10
5
0
20
30
40
50
60
70
80
90
100
HDL Cholesterol
Y = 5.25894 - 3.69E-02X
R-Squared = 0.049

Figure 4

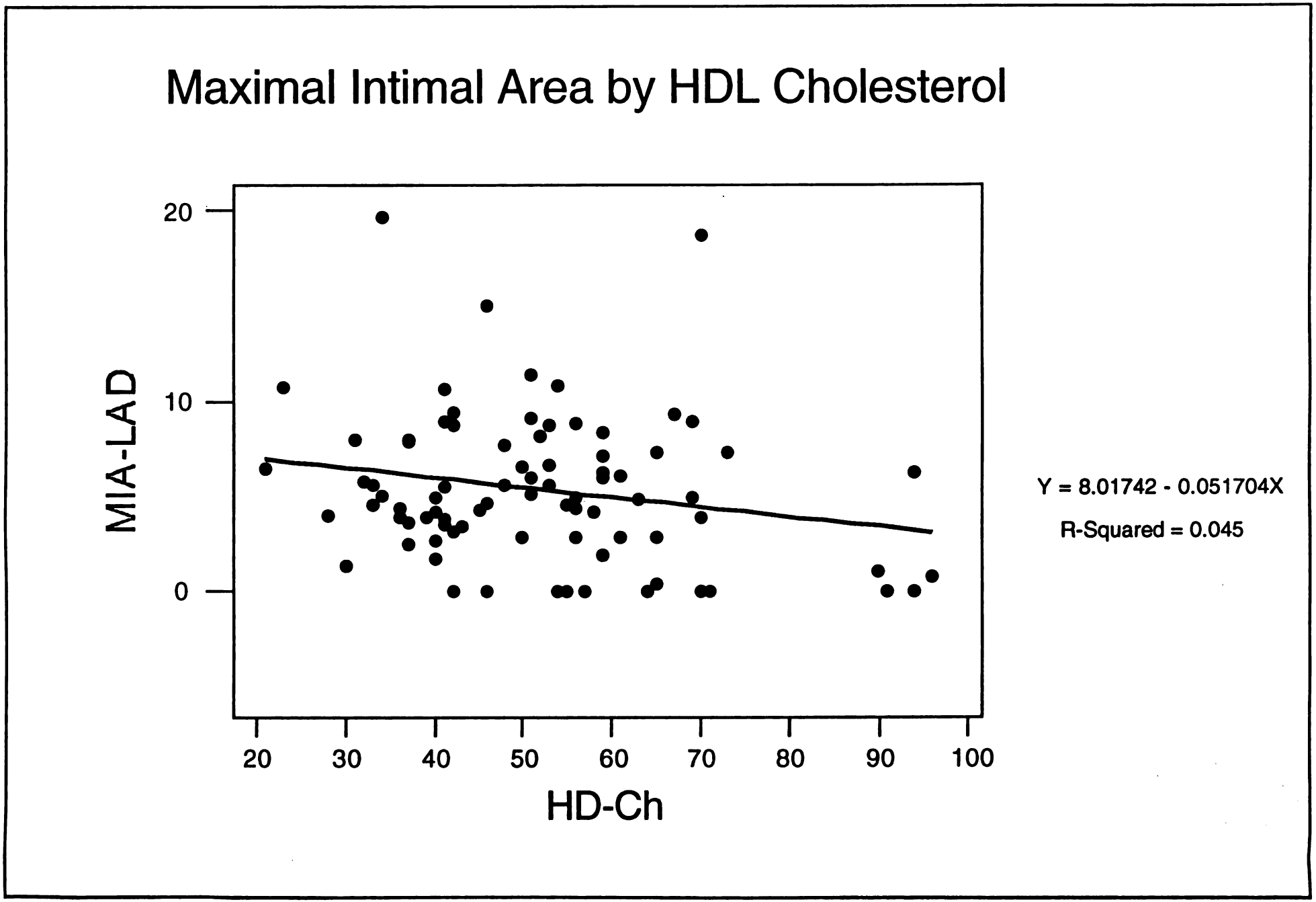
Maximal Intimal Area by HDL Cholesterol
MIA-LAD
20
10
0
20
30
40
50
60
70
80
90
100
HD-Ch
Y = 8.01742 - 0.051704X
R-Squared = 0.045

Figure 5


Alcohol consumption by Gensini Score
Alcohol (oz/wk)
20
10
0
-10
0
10
20
30
40
50
60
70
80
Gensini Score
Y = 5.20772 - 5.83E-02X
R-Squared = 0.029

Alcohol consumption by Summation Score