Effect of L-Thyroxine Therapy on Lipoprotein Fractions in Overt and Subclinical Hypothyroidism, With Special Reference to Lipoprotein(a)

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The effect of L-thyroxine therapy on lipoprotein fractions was assessed in 15 patients with overt hypothyroidism (14 women and one man aged 45 ± 3.9 years; thyrotropin [TSH]: mean ± SEM, 42 ± 6.5 mIU/L; range, 20.5 to 106.5) and 14 patients with subclinical hypothyroidism (13 women and one man aged 41 ± 4 years; TSH: mean ± SEM, 9.1 ± 1 mIU/L; range, 5.1 to 17.3). Fasting serum lipid levels were measured initially and 4 months after achievement of a euthyroid state with incremental L-thyroxine therapy (TSH: mean ± SEM, 1.8 ± 0.4 mIU/L; range, 0.3 to 4.9 for both groups). In the overtly hypothyroid group, restoration of a euthyroid state was associated with a significant reduction in total cholesterol, and apo B. In the subclinically hypothyroid group, there was a significant reduction of only total cholesterol (199.6 ± 13.2 *v* 183.4 ± 11.6 mg/dL) and LDL-C (131.6 ± 8.4 *v* 114 ± 9.25 mg/dL). In contrast, lipoprotein(a) [Lp(a)] was unaffected by the incremental adjustment of L-thyroxine therapy in both groups (overt, 34.3 ± 8.8 *v* 35.6 ± 6.7 mg/dL; subclinical, 23.0 ± 8.6 *v* 29.4 ± 9.5 mg/dL). We conclude that restoration of a euthyroid state in patients with overt hypothyroidism has no significant effect on Lp(a) levels, and confirm that subclinical hypothyroidism is associated with a significant increase in LDL-C, known to have an atherogenic effect.

THE MOST COMMON abnormalities of lipoprotein metabolism associated with overt hypothyroidism are hypercholesterolemia and elevated low-density lipoprotein cholesterol (LDL-C) and apolipoprotein (apo) B levels. These changes are at least in part accounted for by the effect of thyroid hormone on lipoprotein lipase activity, expression of LDL receptors, and subsequent cellular uptake of LDL and very–low-density lipoprotein particles,[1-4] and probably play an important role in atherogenesis in untreated overt hypothyroidism.

Although lipoprotein(a) [Lp(a)] has been shown to be an important pathogenic factor in atherosclerosis, its metabolism and the mechanisms involved in regulation of its synthesis and degradation are not fully elucidated. For instance, although it has been established that LDL receptors play a major role in the uptake and clearance of LDL, there is controversy as to whether the LDL receptor affects the clearance of a metabolically significant fraction of Lp(a).[5-7] In addition, although there is evidence that thyroid hormone plays an important regulatory role in LDL receptor number and LDL metabolism,[8-10] no clear-cut evidence exists that thyroid hormone plays a role in the regulation of Lp(a) metabolism. Thus, it has been questioned whether hypothyroidism could cause changes in Lp(a) concentration that would contribute to atherosclerotic cardiovascular disease, in conjunction with the known changes in other atherogenic fractions. Recently, four studies have reported Lp(a) levels in patients with overt hypothyroidism, with conflicting results.[11-14]

Numerous studies have also attempted to assess whether subclinical hypothyroidism causes significant changes in lipoprotein fractions and have provided different results.[1,2,15-22] However, most of these studies were cross-sectional and compared results in groups of patients with subclinical hypothyroidism with those of euthyroid controls. Recently, we have shown that restoration of a euthyroid state with incremental adjustment of L-thyroxine therapy causes a significant reduction in LDL-C and apo B in patients with subclinical hypothyroidism who have a mean basal thyrotropin (TSH) level of 16.6 ± 3.2 mIU/L.[15]

In this study, we have assessed lipoprotein fractions, including Lp(a), in patients with overt and subclinical hypothyroidism before and 4 months after achievement of a euthyroid state with L-thyroxine therapy to determine whether thyroid hormone deficiency affects Lp(a) in relationship to other lipoprotein fractions, and to reassess lipoprotein-fraction changes in subclinical hypothyroidism.

SUBJECTS AND METHODS

The subjects consisted of a group of 15 ambulatory patients (14 women and one man aged 17 to 75 years; mean ± SEM, 45 ± 3.9) with overt hypothyroidism (TSH > 20 mIU/L and low thyroid hormone levels, group I) and 14 patients (13 women and one man aged 22 to 66 years; mean ± SEM, 41 ± 4.0) with subclinical hypothyroidism (TSH = 5 to 20 mIU/L and normal thyroid hormone levels, group II). The patients' characteristics are shown in Table 1. The underlying etiology for hypothyroidism in group I was Hashimoto's thyroiditis in four patients, post–radioactive iodine ablation for Graves' disease in eight, and post–surgical ablation for a thyroid tumor in three. While six patients were receiving stable doses of L-thyroxine (50 to 150 μg daily), nine patients were newly diagnosed and were not receiving thyroid hormone therapy. The underlying cause of hypothyroidism in group II was Hashimoto's thyroiditis in six patients, post–surgical ablation for a thyroid tumor in three, atrophic hypothyroidism in three, and post–radioactive iodine ablation for Graves' disease in two. None of the study patients were receiving medications or had an associated illness known to alter TSH secretion and/or interfere with lipoprotein metabolism.

Thyroid-function tests including total thyroxine, triiodothyronine, free thyroxine index, and TSH were performed on serum samples obtained initially and 4 months after achievement of a euthyroid state with L-thyroxine therapy.

Morning blood samples were drawn after a 12-hour overnight fast into tubes containing EDTA, and plasma was stored at −70°C

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Table 1. Thyroid-Function Tests and L-Thyroxine Dose Before and After Achievement of a Euthyroid State

Parameter	Group I (n = 15)				Group II (n = 14)			
	Pretreatment		Posttreatment		Pretreatment		Posttreatment	
	Mean ± SE	Range	Mean ± SE	Range	Mean ± SE	Range	Mean ± SE	Range
FTI	3.17 ± .60	.5-7.2	8.75 ± .51	6.6-14.5	6.56 ± .61	5.4-9.7	8.7 ± .46	6.2-11.5
TSH (mIU/L)	42.9 ± 6.46	20.5-106.6	1.83 ± .43	0.3-4.9	9.1 ± 1.0	5.1-17.3	1.8 ± 0.42	0.3-4.9
Dose of L-thyroxine (μg)	31.66 ± 12.39	0-150	123.3 ± 8.5	75-175	55.42 ± 14.4	0-150	99.1 ± 11.8	50-175

Abbreviation: FTI, free thyroxine index.

for subsequent measurement of total cholesterol, triglyceride, LDL-C, high-density lipoprotein cholesterol (HDL-C), apo B-100, apo A-I, and Lp(a) levels. Four months after achievement of a euthyroid state, blood was drawn again for measurement of the same variables. Initial and repeat measurements were performed in the same assays.

Assays

Serum TSH level was measured in duplicate by a sensitive chemiluminescent assay (Immulite chemiluminescent; Diagnostic Products, Los Angeles, CA) with a detection limit of .005 mIU/L, a functional sensitivity of 0.01 mIU/L, and a normal range of 0.5 to 4.9 mIU/L. Serum total thyroxine level was measured by radioimmunoassay (Coat-a-Count; Diagnostic Products, Los Angeles, CA) (normal range, 5.5 to 11 μg/dL), and triiodothyronine resin uptake was measured using the same methodology (normal range, 25% to 35%). Concentrations of plasma cholesterol[23] and triglycerides[24] were measured by enzymatic procedures. These measurements were adapted for automatic analysis in a centrifugal analyzer (Cobas-Bio centrifugal analyzer; Roche, Basel, Switzerland). HDL-C level was determined by enzymatic measurement of the supernatant liquid after precipitation of the plasma with magnesium chloride and dextran sulfate.[25] LDL-C level was calculated subsequently using the Friedewald formula.[26] Apo A-I and apo B values were determined by rate immunonephelometry (Array Protein System; Beckman, Brea, CA). Plasma levels of Lp(a) were determined by a double-antibody, enzyme-linked immunosorbent assay[27] and multiplied by 3.3 to obtain plasma Lp(a) (total mass) levels, which are given in milligrams per deciliter.

Statistical analysis was performed using the Wilcoxon matched-pairs, signed-rank test and Student's *t* test. The *P* value was adjusted using the Bonferroni correction for comparison of the means.

RESULTS

Before correction of the hypothyroid state, mean TSH was 42.9 ± 6.46 (mean ± SE) mIU/L in group I and 9.1 ± 1.0 mIU/L in group II. After incremental adjustment of L-thyroxine therapy, TSH became normal in all patients (Table 1). In group I, restoration of a euthyroid state was associated with a significant reduction in total cholesterol (from 276.7 ± 26.5 to 200.9 ± 13.3 mg/dL), LDL-C (from 207.3 ± 22 to 137.3 ± 12.2 mg/dL), apo A-I (from 178.5 ± 13 to 134.3 ± 9.1 mg/dL), and apo B-100 (from 180.5 ± 18.8 to 127.1 ± 11.2 mg/dL). However, Lp(a) levels were not different in the hypothyroid state (34.3 ± 8.8 mg/dL; range, 5 to 120) and euthyroid state (35.6 ± 7.7 mg/dL; range, 4 to 136). Figure 1 shows individual Lp(a) values before and after L-thyroxine therapy in patients with overt hypothyroidism. Thus, almost all patients who had initial Lp(a) values higher than the accepted cutoff level for increased risk of atherosclerosis (30 mg/dL) continued to have elevated values after achievement of a euthyroid state. Table 2 shows lipoprotein concentrations and ratios before and after achievement of a euthyroid state in patients with subclinical hypothyroidism. There was a statistically significant reduction in total cholesterol and LDL-C and a small but not statistically significant reduction in mean levels of apo B-100. Lp(a) was also unaffected by incremental adjustment of L-thyroxine therapy in patients with subclinical hypothyroidism. Comparison of lipid results in the two groups before L-thyroxine therapy is shown in Fig 2. Thus, in patients with overt hypothyroidism, total cholesterol, LDL-C, and apo B-100, but not Lp(a), are significantly higher than in patients with subclinical hypothyroidism.

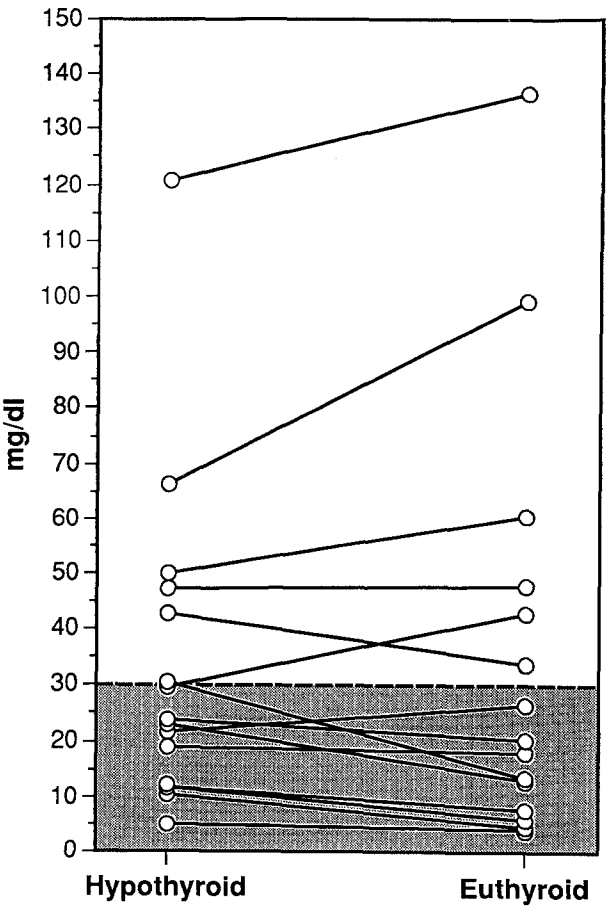

Fig 1. Individual Lp(a) values in patients with overt hypothyroidism before and after restoration of a euthyroid state with L-thyroxine therapy. (———) Accepted cutoff level (30 mg/dL) above which there is an increased risk for cardiovascular disease.

Table 2. Lipoprotein Fractions and Ratios in Patients With Subclinical Hypothyrodism Before and After Treatment

	Before Treatment		After Treatment	
Lipoprotein	Mean ± SE	Range	Mean ± SE	Range
Total cholesterol (mg/dL)	199.6 ± 13.20	138-324	183.4 ± 11.62*	118-281
Triglycerides (mg/dL)	112.6 ± 15.24	46-238	116.4 ± 16.90	50-272
HDL-C (mg/dL)	45.4 ± 4.77	30-95	45.4 ± 4.23	21-74
LDL-C (mg/dL)	131.6 ± 8.43	78-196	114.8 ± 9.25*	64-195
Apo A-I (mg/dL)	171.8 ± 12.17	110-266	177 ± 14.59	103-297
Apo B (mg/dL)	112.1 ± 9.86	59-207	105.7 ± 9.39	54-197
Lp(a) (mg/dL)	22.99 ± 8.6	.99-101.64	29.40 ± 9.47	1.98-100.32
Apo A-I/ HDL-C	4.2 ± .34	2.81-7.03	4.2 ± .45	2.62-8
LDL-C/apo B	1.2 ± .04	.99-1.47	1.1 ± .04*	.84-1.44

*$P < .01$.

DISCUSSION

Our results agree with findings reported in previous studies with respect to changes in total cholesterol, LDL-C, apo B, and apo A-I in overt hypothyroidism before and after achievement of a euthyroid state with L-thyroxine.[1-4,28] These results are also consistent with previous observations that the magnitude of changes in lipoprotein fractions in hypothyroid patients correlates with the severity of thyroid hormone deficiency.[2] Achievement of euthyroidism also results in a significant reduction in LDL-C in patients with subclinical hypothyroidism, consistent with results of our previous study.[15] However, we have not found a significant reduction in apo B levels in patients with subclinical hypothyroidism, as we previously reported,[15] although we have noted a trend for reduction in apo B-100 levels with L-thyroxine therapy. This difference may be explained by the fact that patients recruited in the current study had lower TSH values (TSH, 9.1 ± 1 mIU/L) than those previously studied (TSH, 16.6 ± 2.2 mIU/L).[15] Thus, apparent changes in LDL-C may precede those of apo B-100 with gradually worsening thyroid hormone deficiency. Higher LDL-C levels in patients with slightly elevated TSH as compared with euthyroid controls have also been shown by Althaus et al[21] and Staub et al.[2] However, in the latter study, only patients with TSH greater than 12 mIU/L had an elevation in LDL-C and apo B. In contrast, other studies did not show a change in LDL-C in patients with subclinical hypothyroidism.[1,20]

The elucidation of whether thyroid hormone deficiency is associated with an increase in Lp(a) is important for several reasons. Lp(a) may be an independent risk factor for atherosclerosis, as suggested by both cross-sectional and prospective studies.[29-39] In addition, establishing whether Lp(a) changes parallel those of LDL-C and apo B may provide further insight into whether Lp(a) and LDL-C are subject to similar regulatory mechanisms.

Whereas one cross-sectional study from Denmark showed no difference in Lp(a) levels in hypothyroid patients as compared with euthyroid controls,[11] another study in which serial measurements were performed suggested that L-thyroxine therapy caused an initial reduction followed by a secondary increase in Lp(a) concentration.[12] Spandrio et al[13] showed that L-thyroxine therapy did not significantly affect Lp(a) levels in a group of patients with postsurgical hypothyroidism, but Engler and Riesen[14] did demonstrate a reduction of Lp(a) levels during L-thyroxine treatment, attributed to a direct effect of thyroid hormone on Lp(a) synthesis rather than LDL receptor effects. Even though we have not sequentially measured Lp(a) levels during L-thyroxine therapy, our data do not provide evidence that hypothyroidism is associated with a significant change in Lp(a). This is further supported by the observation that despite significant differences in total cholesterol, LDL-C, and apo B-100 levels between patients with overt and subclinical hypothyroidism, Lp(a) levels were not different in the two groups in the untreated state. Our data also

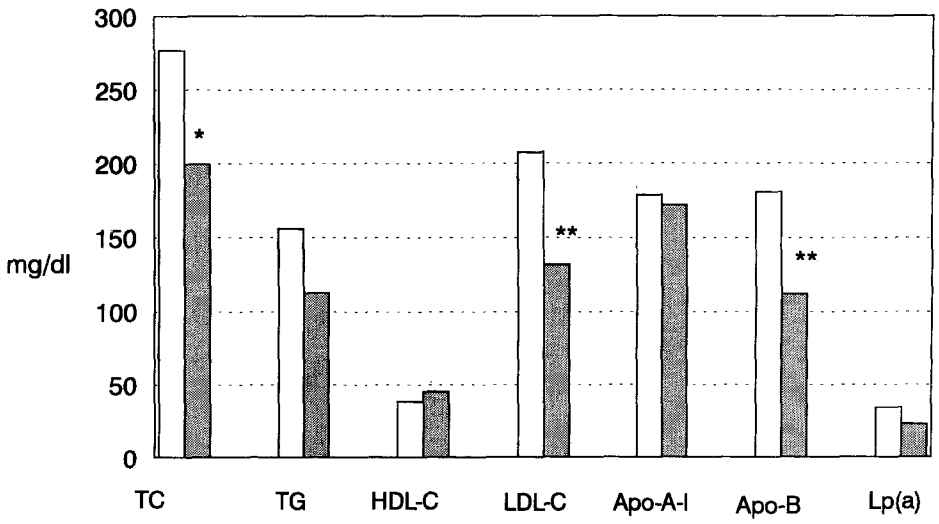

Fig 2. Mean lipoprotein fractions in patients with (□) overt and (■) subclinical hypothyroidism before L-thyroxine therapy. Lp(a) is expressed as total Lp(a) mass. TC, total cholesterol; TG, triglycerides. *$P < .001$, **$P < .005$.

suggest that even if minimal changes in Lp(a) do occur as a result of thyroid hormone deficiency, these changes may be difficult to assess due to the wide range of Lp(a) values found in both normal euthyroid and hypothyroid individuals.[6,7]

In conclusion, our study indicates that both overt and subclinical hypothyroidism cause an elevation in LDL-C commensurate with the severity of thyroid hormone deficiency, but these analytes are not associated with significant changes in Lp(a). These results suggest that Lp(a) and LDL metabolism are not subject to the same regulatory mechanisms.

REFERENCES

1. Lithell H, Boberg J, Hellsing K, et al: Serum lipoprotein and apolipoprotein concentrations and tissue lipoprotein-lipase activity in overt and subclinical hypothyroidism: The effect of substitution therapy. Eur J Clin Invest 11:3-10, 1981
2. Staub J-J, Althaus BU, Engler H, et al: Spectrum of subclinical and overt hypothyroidism: Effect on thyrotropin, prolactin, and thyroid reserve, and metabolic impact on peripheral target tissues. Am J Med 92:631-642, 1992
3. Mishkel MA, Crowther SM: Hypothyroidism: An important cause of reversible hyperlipidemia. Clin Chim Acta 74:139-144, 1977
4. Muls E, Rosseneau M, Blanton V, et al: Serum lipids and apoproteins A-I, A-II, and B in primary hypothyroidism before and during treatment. Eur J Clin Invest 14:12-15, 1984
5. Scanu AM, Scandiani L: Lipoprotein(a): Structure, biology, and clinical relevance. Adv Intern Med, 36:249-270, 1991
6. Scanu AM, Fless GM: Lipoprotein(a): Heterogeneity and biological relevance. J Clin Invest 85:1709-1715, 1990
7. Mims MP, Paubatz JW, Ghazzaly KK, et al: Interaction of LDL and Lp(a) with human fibroblasts. Chem Phys Lipids 67/68: 145-152, 1994
8. Thompson GR, Soutar AK, Spengel FA, et al: Defects of receptor-mediated low density lipoprotein catabolism in homozygous familial hypercholesterolemia and hypothyroidism. Proc Natl Acad Sci USA 78:2591-2595, 1981
9. Chait A, Bierman EL, Albers JJ: Regulatory role of triiodothyronine in the degradation of low density lipoprotein by cultured human skin fibroblasts. J Clin Endocrinol Metab 48:887-889, 1979
10. Dory L, Roheim PS: Rat plasma lipoproteins and apolipoproteins in experimental hypothyroidism. J Lipid Res 22:287-296, 1981
11. Klausen IC, Nielsen FE, Hegedus L, et al: Treatment of hypothyroidism reduces low-density lipoproteins but not lipoprotein(a). Metabolism 41:911-914, 1992
12. De Bruin TWA, Van Barlingen H, Trip MV-L, et al: Lipoprotein(a) and apolipoprotein B plasma concentrations in hypothyroid, euthyroid, and hyperthyroid subjects. J Clin Endocrinol Metab 76:121-126, 1993
13. Spandrio S, Sleiman I, Scalvini T, et al: Lipoprotein(a) in thyroid dysfunction before and after treatment. Horm Metab Res 25:586-589, 1993
14. Engler E, Riesen WF: Effect of thyroid function on concentrations of lipoprotein(a). Clin Chem 39:2466-2469, 1993
15. Arem R, Patsch W: Lipoprotein and apolipoprotein levels in subclinical hypothyroidism: Effect of levothyroxine therapy. Arch Intern Med 150:2097-2100, 1990
16. Nilsson G, Nordlander S, Levin K: Studies on subclinical hypothyroidism with special reference to the serum lipid pattern. Acta Med Scand 200:63-67, 1976
17. Kutty KM, Bryant DG, Farid NR: Serum lipids in hypothyroidism—A re-evaluation. J Clin Endocrinol Metab 46:55-60, 1978
18. Feely J, Iqgal SJ, Isles TE, et al: Total cholesterol/HDL cholesterol ratio in hyperthyroidism, hypothyroidism, and subclinical hypothyroidism. Horm Metab Res 12:560-561, 1980
19. Raziel A, Rosenzweig B, Botvinic V, et al: The influence of thyroid function on serum lipid profile. Atherosclerosis 41:321-326, 1982
20. Parle JV, Franklyn JA, Cross KW, et al: Circulating lipids and minor abnormalities of thyroid function. Clin Endocrinol (Oxf) 37:411-414, 1992
21. Althaus BU, Staub JJ, Ryff-De Leche A, et al: LDL/HDL changes in subclinical hypothyroidism: Possible risk factors for coronary heart disease. Clin Endocrinol (Oxf) 28:157-163, 1988
22. Caron P, Calazel C, Parra HJ, et al: Decreased HDL cholesterol in subclinical hypothyroidism: The effect of L-thyroxine therapy. Clin Endocrinol (Oxf) 33:519-523, 1990
23. Siedel J, Hagele E, Ziegenhorne J, et al: Reagent for enzymatic determination of serum cholesterol with improved lipolytic efficiency. Clin Chem 29:1075-1080, 1983
24. Nagele U, Hagele E, Sauer G, et al: Reagent for enzymatic determination of serum total triglyceride with improved lipolytic efficiency. J Clin Chem Clin Biochem 22:165-174, 1984
25. Warnick GR, Benderson J, Abers JJ: Dextran sulfate–Mg^{2+} precipitation procedure for quantification of high-density-lipoprotein cholesterol. Clin Chem 28:1379-1388, 1982
26. Friedewald WT, Levy RI, Fredrickson DS: Estimation of the concentration of low-density lipoprotein in the plasma without the preparative ultracentrifuge. Clin Chem 18:499-502, 1972
27. Gaubatz JW, Cushing GL, Morrisett JD: Quantification, isolation, and characterization of human lipoprotein [a]. Methods Enzymol 129:167-186, 1986
28. Ballantyne FC, Epenetos AA, Caslake M, et al: The composition of low-density lipoprotein and very–low-density lipoprotein subfractions in primary hypothyroidism and the effect of thyroid hormone replacement therapy. Clin Sci 57:83-88, 1979
29. Schreiner PJ, Morrisett JD, Sharrett AR, et al: Lipoprotein(a) as a risk factor for preclinical atherosclerosis. Arterioscler Thromb 13:826-833, 1993
30. Dahlen GH: Lipoprotein(a) in relation to atherosclerotic diseases. Prog Clin Biol Res 255:27-36, 1988
31. Hoefler G, Harnoncourt F, Paschke E, et al: Lipoprotein Lp(a): A risk factor for myocardial infarction. Arteriosclerosis 8:398-401, 1988
32. Durrington PN, Ishola M, Hunt L, et al: Apolipoproteins (a), AI, and B and parental history in men with early onset ischaemic heart disease. Lancet 1:1070-1073, 1988
33. Dahlen GH, Guyton JR, Attar M, et al: Association of levels of lipoprotein Lp(a), plasma lipids, and other lipoproteins with coronary artery disease documented by angiography. Circulation 74:758-765, 1986
34. Hoff HF, Beck GJ, Skibinski CI, et al: Serum Lp(a) level as a predictor of vein graft stenosis after coronary artery bypass surgery in patients. Circulation 77:1238-1244, 1988
35. Dahlen G, Berg K, Gillnas T, et al: Lp(a) lipoprotein/prebeta 1-lipoprotein in Swedish middle-aged males and in patients with coronary heart disease. Clin Genet 7:334-341, 1975
36. Brown SA, Morrisett JD, Boerwinkle E, et al: The relation of lipoprotein(a) concentrations and apolipoprotein(a) phenotypes with asymptomatic atherosclerosis in subjects of the Atherosclero-

sis Risk in Communities (ARIC) study. Arterioscler Thromb 13:1558-1566, 1993

37. Berg K, Dahlen G, Borresen A-L: Lp(a) phenotypes, other lipoprotein parameters and family history of heart disease in middle-aged males. Clin Genet 16:347-352, 1979

38. Kostner GM, Avogaro P, Cozzolato G, et al: Lipoprotein Lp(a) and the risk for myocardial infarction. Atherosclerosis 38:51-61, 1981

39. Rosengren A, Wilhelmsen L, Erikson E, et al: Lipoprotein(a) and coronary heart disease: A prospective case-control study in a general population sample of middle aged men. Br Med J 301:1248-1251, 1990