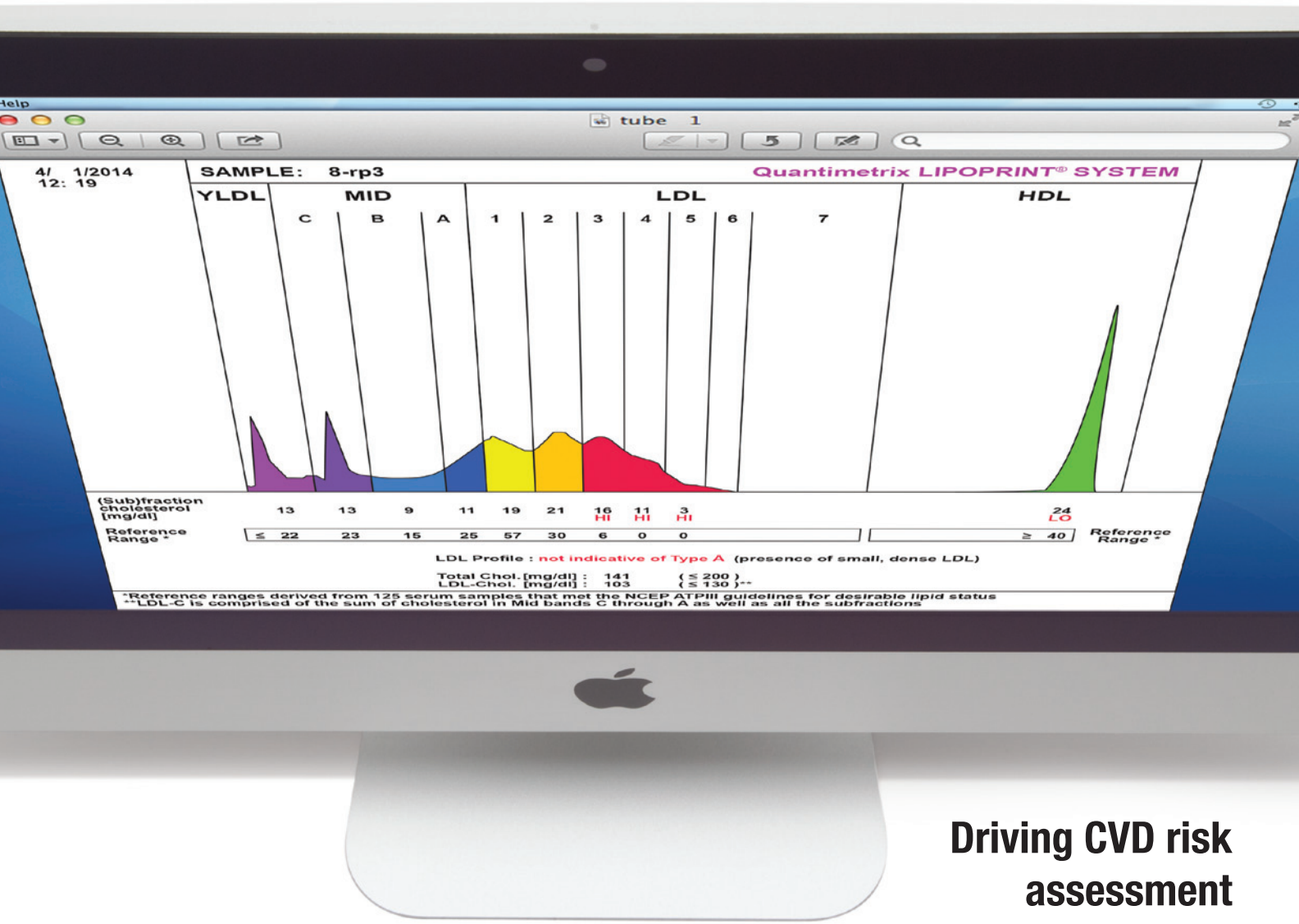
Lipoprint®

Personalized Diagnostics



Driving CVD risk assessment forward.

Quantimetrix

laboratory innovations for 40 years

It was with an unflinching entrepreneurial spirit and a distinguished PhD in biochemistry that, Dr. Robert Ban, my father, founded Quantimetrix. He pioneered a liquid control stabilization technology that simplified laboratory quality control methods and launched an industry that previously offered freeze-dried products.

He believed in the importance of applying the most rigorous standards in research, development and manufacturing, giving his customers confidence that their test results were accurate and clinically sound.

That was forty years ago, and today, Quantimetrix continues to develop next-generation laboratory technologies that will change how clinicians test, laboratories research, and patients interact with their physicians.

Lipoprint® Lipoprotein Subfractions Testing System™ is our comittment to these core principles. Bringing Lipoprint to the marketplace has been a personal achievement for the Quantimetrix family. Many of us have known people who may have benefitted from more advanced clinical information about their cardiac health. Take a moment to review our Lipoprint brochure and see how our test could benefit you.

Monty Ban
President, Quantimetrix Corporation

Lipoprint®
Lipoprotein Subfractions Testing System

The Lipoprint Lipoprotein Subfractions Testing System analyzes all lipoprotein fractions and subfractions in fasting serum or plasma. Lipoprint uses high resolution polyacrylamide gel electrophoresis that separates and measures the amount of cholesterol in each LDL and HDL subfraction. The Lipoprint LDL Kit is FDA cleared for *in vitro* diagnostic use. The Lipoprint HDL Kit is intended for research use only. Not for use in diagnostic procedures.

PERSONALIZED CVD RISK ASSESSMENT BY LIPOPRINT

There has been much confusion lately about the new cholesterol treatment recommendations for primary and secondary ASCVD risk management. Lipoprint Lipoprotein Subfractions Testing System creates clarity by providing more comprehensive analysis for more specific treatment. The new cholesterol treatment guidelines weigh heavily on non-lipid risk factors, raising concerns of under-treatment of some individuals that may be at risk and overtreatment of low risk individuals based on well established risk factors such as LDL-C, Apo B and new emerging lipid and lipoprotein risk factors such as Non-HDL cholesterol or the atherogenic small dense LDL subfractions.

Coronary artery disease (CAD) continues to be the leading cause of death in most of the developed countries and much of the developing world in spite of more risks awareness and increased treatment of individuals at risk. The role of lipids and lipoproteins in atherogenesis has been demonstrated by numerous studies but the methods for assessing CAD risk have continued to be the subject of much debate. Until recently, LDL cholesterol (LDL-C) was accepted as the primary target for CAD treatment.

The ACC/AHA Task Force released new guidelines in November 2013 along with a new CVD risk calculator based on data from randomized control trials. The risk calculator weighs heavily upon non-lipid risk factors reducing the role of LDL-C and other dyslipidemias as risk factors.

A recent study[1] suggested that blood cholesterol and LDL-C levels as currently measured may not reflect the actual risk of CAD since many individuals that develop CAD have the same levels as those that do not develop CAD. These findings suggest that dyslipidemia, as currently defined, may not be a good indicator of CAD risk. The most common lipid disorder associated with CAD is a pro-atherogenic dyslipidemia characterized by the presence of highly atherogenic small dense LDL particles, intermediate density lipoproteins and VLDL remnants particles that may not be reflected by measuring total cholesterol, LDL-C or any other single metric. Recent studies suggest that pro-atherogenic dyslipidemias can be best identified by measuring the individual atherogenic lipoprotein such as VLDL remnants, IDL and the small dense LDL subfractions.

The study randomly selected 22 individuals who were tested for CVD risk using the new ASCVD risk calculator. In addition, the samples were tested by other traditional and non-traditional test methods such as LDL-C, NMR LDL particle number, total cholesterol, triglycerides, non-HDL cholesterol, total cholesterol to HDL-C ratio and LDL-C to HDL-C ratio. The samples were also analyzed using the Lipoprint Lipoprotein Subfractions Testing System that can discriminate atherogenic from non-atherogenic LDL subfractions.

The results of the study showed significant discordance between the various test methods, (See Table 1 on the next page). Based on the ASCVD risk calculator, 7 (31.8%) of the 22 individuals were at high risk of CVD. Five of the seven samples were classified high risk based on non-lipid risk factors such as hypertension, diabetes and advanced age and only 2 were classified high risk due to dyslipidemias. The Lipoprint LDL test measures VLDL remnants, IDL and small dense LDL subfractions in addition to the non-atherogenic large buoyant LDL. Based on the Lipoprint LDL subfractions test, 15 (68.2%) of the individuals were classified intermediate or high risk.

The study suggests that the ASCVD risk calculator appears to underestimate the risk associated with dyslipidemias when compared to other test methods while the Lipoprint® LDL subfractions test captures all the high-risk individuals identified by the other methods in the study. The Lipoprint LDL subfractions test provides a more individualized assessment of CVD risk than the generalized approach of the ASCVD risk calculator. Considering the fact that CVD is a life long progressive disease, early recognition is very important in primary prevention and treatment of individuals at risk.

1. AACC 2014 Scientific Poster Session, *LDL Subfractions Analysis in Pro-atherogenic Dyslipidemia*; N. Muñiz, E. Nuñez, M. Rivera, M. Ban – Quantimetrix, Redondo Beach, CA

THE LIPOPRINT LDL SYSTEM

The Lipoprint LDL System from Quantimetrix is a device for *in vitro* diagnostic use only, intended to measure cholesterol levels in all lipoprotein fractions and LDL subfractions in fasting serum or plasma. The Lipoprint system uses polyacrylamide gel electrophoresis to separate the various lipoprotein subfractions on the basis of size (See Figure 1). The electrophoresed gels are analyzed with Lipoware, a configured software that calculates the levels of cholesterol in each subfraction. A color coded profile is generated for ease of interpretation.

PERSONALIZED ATTRIBUTES OF THE LIPOPRINT LDL TEST

To determine the clinical significance of an emerging risk factor, the ATP III requires, the following criteria: "Laboratory or clinical measurements must be widely available, well standardized, inexpensive, have accepted population-reference values, and be relatively stable biologically."

Table 1: CVD Risk Data

Sample ID	/AHA	Lipoprint						
	10-Year ASCVD Risk	VLDL	IDL-C	IDL-B	IDL-A	LDL-1	LDL-2	LDL-3+
	%	[mg/dL]	[mg/dL]	[mg/dL]	[mg/dL]	[mg/dL]	[mg/dL]	[mg/dL]
002	8.6	49	26	23	14	23	31	20
005	17.7	28	24	20	25	52	40	6
006	4.8	33	19	18	22	37	49	21
008	1.8	43	23	17	12	18	25	12
009	8.6	43	29	20	15	23	26	17
010	3.6	42	22	19	13	20	25	22
011	4.4	22	23	15	18	46	40	7
012	7.1	35	21	23	27	43	32	6
016	1.7	28	20	15	16	45	46	9
017	1.6	31	20	14	20	54	31	2
022	1.7	23	21	16	23	58	43	5
023	5	36	20	12	14	37	18	0
026	11.9	26	17	14	20	41	30	4
028	5.7	43	21	13	9	22	33	14
029	21.8	36	22	16	15	30	32	11
030	0.6	33	21	14	20	55	27	2
031	4.2	17	14	13	28	42	11	0
034	9.7	22	14	13	16	32	26	7
036	0.5	21	13	15	27	51	8	0
037	4.5	44	22	13	11	27	30	7
039	1.9	22	15	16	21	49	45	8
040	18.8	42	22	13	11	30	25	5
	< 7.4 % Desirable	≤ 22	≤ 23	≤ 15	≤ 25	≤ 57	≤ 30	≤ 6
		23-30	24-26	16-18	26-28	58-60	31-33	7-9
	≥ 7.5 % High	> 30	> 26	> 18	> 28	> 60	> 33	> 9

The Lipoprint LDL subfraction test is an *in vitro* test device for new emerging risk factors with the following characteristics:

- The Lipoprint test is available for use in any clinical or research laboratory.
- The Lipoprint test reveals heart disease risk factors not indicated by conventional lipoprotein cholesterol measurements. It is accurate, easy to perform, and relatively inexpensive.
- The Lipoprint test was compared to established methods such as direct LDL and HDL cholesterol test methods. It was also compared to the CDC beta quantification and ultracentrifugation methods.
- Normal reference ranges for all lipoprotein fractions and LDL subfractions were derived from a normal population as defined by NCEP ATP III guidelines for desirable lipid levels.
- The test is performed on fasting serum or EDTA plasma. Samples are stable refrigerated for up to seven days. For prolonged storage, cryogenic freezing of the samples is recommended.

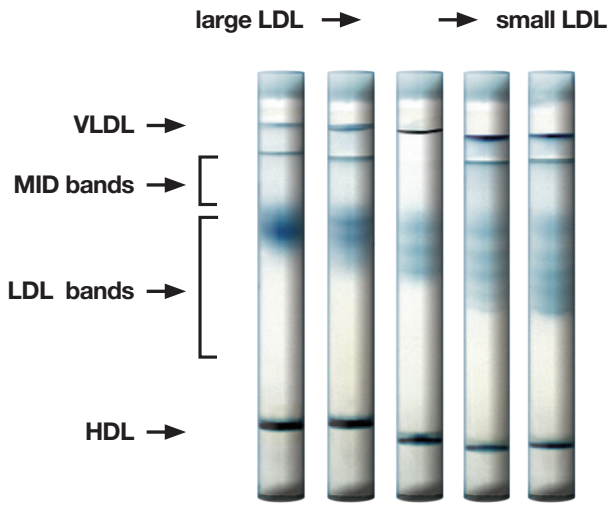
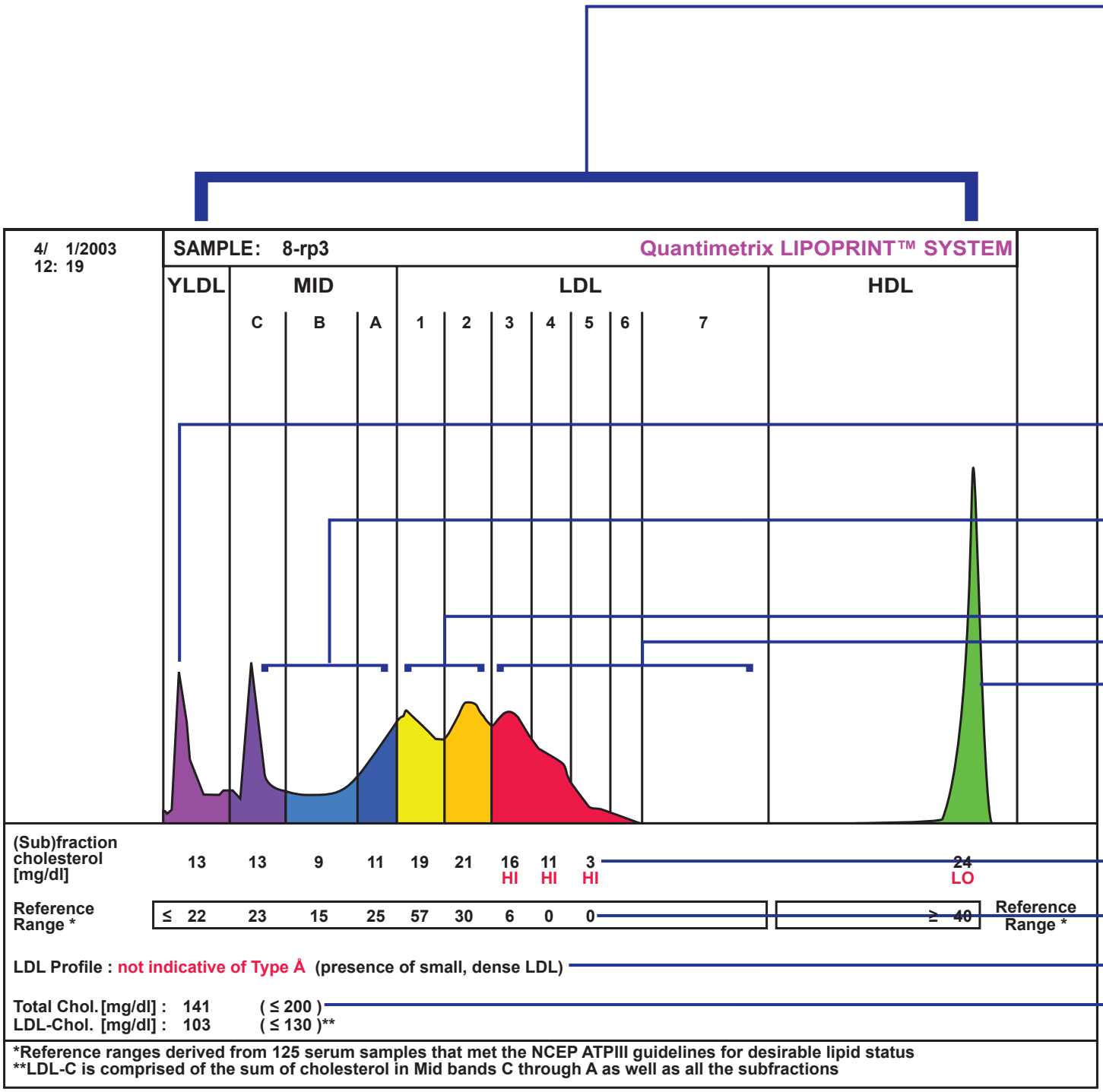


Figure 1: Lipoprint Subfractions

		NMR	ATP III	ATP III	ATP III	ATP III	ATP III	ATP III
HDL-C	Mean LDL Particle Size	LDL-P	TC	TRIGS	LDL-C	Non-HDL	TC:HDL-C	LDL-C: HDL-C
[mg/dL]	Å	(nmol/L)	[mg/dL]	[mg/dL]	[mg/dL]	[mg/dL]	Ratio	Ratio
38	258	1281	235	471	147	197	6.2	3.9
43	269	1761	238	103	166	195	5.5	3.9
26	263	2425	228	186	169	202	8.8	6.5
33	262	1152	185	321	108	152	5.6	3.3
35	260	1376	215	245	136	180	6.1	3.9
27	254	1772	208	284	139	181	7.7	5.1
50	268	1936	222	115	149	172	4.4	3.0
37	268	1843	223	139	150	186	6.0	4.1
37	266	2038	217	153	152	180	5.9	4.1
48	270	1641	220	116	141	172	4.6	2.9
48	269	1980	237	101	165	189	4.9	3.4
31	272	1236	169	173	101	138	5.5	3.3
45	269	1690	196	105	125	151	4.4	2.8
30	262	1723	186	219	114	156	6.2	3.8
33	265	1445	196	196	127	163	5.9	3.8
54	271	1656	226	169	139	172	4.2	2.6
54	274	1299	180	55	108	126	3.3	2.0
51	266	1283	183	109	109	132	3.6	2.1
60	275	1484	196	71	115	136	3.3	1.9
32	266	1375	186	233	111	154	5.8	3.5
40	267	1998	216	83	153	176	5.4	3.8
51	267	939	199	287	107	148	3.9	2.1
≥ 50 High	≥ 269 Type A	< 1000 Low	< 200 Desirable	< 150 Desirable	< 100 Desirable	< 130 Desirable	< 4.0 Desirable	< 3.0 Desirable
40-49					100 - 129 Average	130 - 159 Average	4.0 - 5.0 Average	3.0 - 4.0 Average
	> 265 ≤ 268 Intermediate	1000-1299 Moderate	200 - 239 Borderline high	> 150 ≤ 199 Borderline high	130 - 159 Borderline high	160 - 189 Borderline high	> 5.0 - 6.0 Borderline high	> 4.0 ≤ 6.0 Bordeline High
< 40 Low	≤ 265 Type B	> 1300 High	≥ 240 High	≥ 200 High	≥ 160 High	≥ 190 High	> 6.0 High	> 6.0 High

Understanding the Lipoprint® Profile



Measurement of up to twelve lipoprotein fractions and subfractions. Results for every lipoprotein subfraction reported as mg/dL cholesterol which should be the basis for patient treatment

VLDL increased levels - associated with hypertriglyceridemia, one of the components of the lipid triad of atherogenic dyslipidemia

Midbands A, B and C (include VLDL remnants and IDL) - associated with atherogenic Type III dyslipidemia and combined hyperlipoproteinemia

LDL 1 and 2 (large buoyant LDL) - not associated with CVD risk

LDL 3 through 7 (small dense LDL) - component of the lipid triad associated with 3 fold increase of CVD, metabolic syndrome and diabetes

HDL cholesterol (good cholesterol) - Low HDL is a component of the lipid triad associated with increased CHD risk

Cholesterol values outside the normal reference range are flagged in red for ease of interpretation. Elevated Mid B, Mid C (IDL and VLDL remnants) and small-dense LDL 3 through LDL 7 pose the highest risk for CVD

Normal Reference Range for each subfraction based on NCEP ATP III guidelines for desirable lipid levels

Lipoprotein Profile Classification - predominance of large LDL is classified as Type A and predominance of small dense LDL is classified as Not Indicative of Type A

TRADITIONAL RISK FACTORS

Total cholesterol - CHD risk factor

COLOR-CODED PROFILE

The Lipoprint Profile is color-coded making it easy for doctor and patient to understand the results of the test at a glance. Good HDL is shown in green; large low risk LDL 1 and 2 in yellow; for caution, and the atherogenic; small dense LDL 3 through 7 in red; for high risk.

Add Lipoprint® Testing to Your Laboratory

Lipoprint
Lipoprotein Subfractions Testing System Components

TEST CHARACTERISTICS

- Easy to interpret color coded profile
- Results available in less than 3 hours
- Fasting serum or EDTA plasma
- LDL subfractions cholesterol derived from total cholesterol assay
- Normal reference ranges derived from NCEP ATPIII guidelines
- Costs are reimbursable (CPT Code: 83701)*

LIPOPRINT CONSUMABLES INFORMATION

LDL Subfractions	48-7002	100 Tests	100 quantity
HDL Subfractions**	48-9002	100 Tests	100 quantity
Liposure Serum Lipoprotein	48-7060	Level 1	4 x 0.5 mL

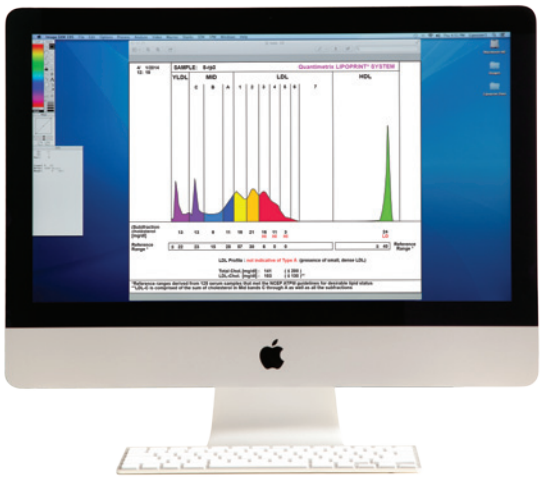
* Reimbursable costs may vary. Please consult with the insurance provider for their specific guidelines.

** The Lipoprint HDL Kit is intended for research use only. Not for use in diagnostic procedures.

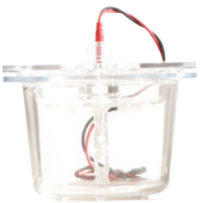
THE LIPOPRINT SYSTEM COMES FULLY LOADED!

CAT# 48-9150 (120V)

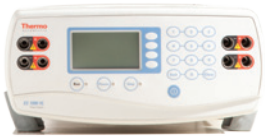
A. iMac Computer and Licensed Lipoprint Software
B. Electrophoresis Chamber
C. Electrophoresis Power Supply
D. Lipoprint LDL Reagent Kit
E. Preparation Light
F. Digital Scanner Densitometer
G. Color Printer



A



B



C

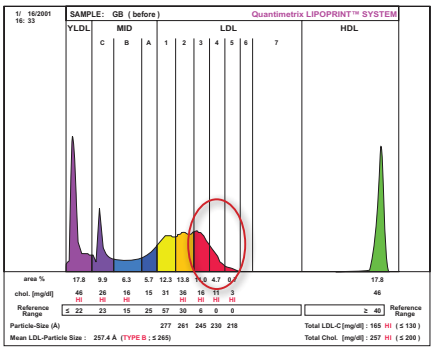
D

E

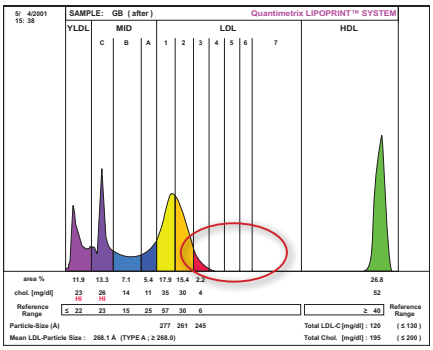
F

G

Case Study 1



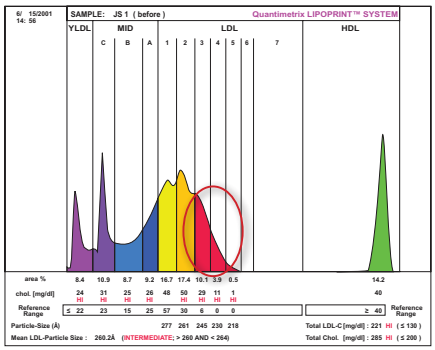
Before Treatment



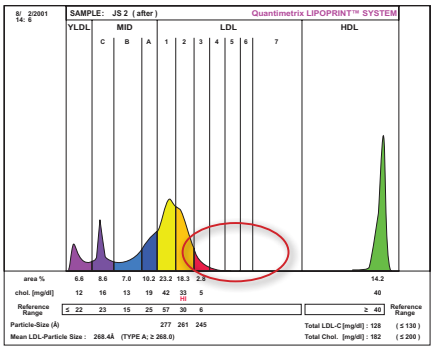
After Treatment

George, a 65 year-old Caucasian male had a Lipoprint LDL Profile showing predominance of small dense LDL 3, 4 and 5 which has been associated with increase risk of cardiovascular disease (before treatment). After 3 months of niacin and dietary therapy, his profile became normal (after treatment).

Case Study 2



Before Treatment



After Treatment

James, a 42 year-old male on a diet rich in carbohydrates and fats had a Lipoprint LDL Profile with high cholesterol levels in most subfractions (before Treatment). After 2 months on a vegetarian diet with emphasis on soy protein and low carbohydrate and exercise, his profile became essentially normal (after treatment).

Lipoproteins and Longevity

LIPOPRINT®—IN RESEARCH CLINICAL STUDIES

Recent studies suggest that larger lipoprotein particle sizes of both high-density lipoprotein (HDL) and low-density lipoprotein (LDL) are characteristic of long and healthy lives. A large number of clinical studies have demonstrated that the size of the LDL particles present is a better predictor of cardiovascular and other health risks than the total LDL concentration. It appears that it is not only the amount of cholesterol that is important but how the cholesterol is distributed among the various LDL and HDL subfractions. Measurements of the cholesterol levels in these lipoprotein subfractions provide a more personalized assessment of the Cardio Vascular disease (CVD) status of the individual than traditional lipid measurements. The Quantimetrix Lipoprint Lipoprotein Subfractions Testing System, is now being utilized routinely in the clinical laboratory for assessment of cardiac risk based on the lipoprotein subfractions distribution.

As the world's population grows older, medicine has increasingly turned to questions of longevity and healthy aging. Researchers have reported correlation between a pattern of large lipoprotein particle size and human longevity. They studied a group of Ashkenazi Jews with a mean age of 98.2 years as well as the children of these long lived individuals.

The researchers found that these individuals and their children had HDL and LDL populations of significantly larger diameter than that of the study control groups. In contrast to the particle size, results were found to be quite similar to the control groups with regard to their concentrations of total cholesterol, LDL cholesterol, HDL cholesterol, and triglycerides. Individuals with large buoyant populations of LDL and HDL cholesterol were also found to have a lower prevalence of hypertension, metabolic syndrome, and cardiovascular disease when compared to those of a similar age with small dense particles. A study conducted in the Netherlands reached a similar conclusion with regard to the link between inherited longevity and LDL particle size. This study looked at sibling pairs with an age of 89 or older for men and 91 or older for women.

The children of these individuals were also studied. Both the long lived siblings and their children were found to have significantly larger LDL populations than the control group and suggests that inherited longevity may either be due to or identified by a pattern of large LDL particles. The significant linkage between LDL particle size and longevity was found to be independent of total cholesterol, HDL cholesterol, and triglycerides.

The study then took the question of LDL particle size and longevity a step further. They wanted to determine whether non-inherited longevity could be predicted by a person's mean LDL diameter. The study attempted to determine the LDL particle size of all the residents of the city of Leiden who had reached an age of ninety or greater regardless of family history. The elderly population of Leiden was found to have a pattern of large LDL particles similar to the long lived siblings in their study. In this study larger LDL size was again linked to better overall health regardless of family history.

What do the results of these studies mean for people who do not have LDL cholesterol loaded with large particles? Statin drugs commonly taken for lowering cholesterol have been shown to alter the LDL subfraction pattern favorably but the results are inconsistent. Also, Fibrate drugs have consistently improved the LDL subfraction pattern based on a number of studies. In fact, the advice that physicians have been giving their patients for years may be the best approach; maintain a healthy life style, follow a diet rich in fruits and vegetables and do some moderate aerobic exercise. This not only reduces the total LDL cholesterol but actually produces a significant increase in the mean size of the LDL particle population that may be conducive of a longer healthier life.

LIPOPRINT HDL SUBFRACTIONS* TESTING

The total cholesterol to HDL cholesterol ratio is very commonly used to assess CVD risk. Lipoprint measures the mg/dL of cholesterol in each lipoprotein fraction and subfraction from VLDL to HDL. (10 parameters)

*Lipoprint HDL is for Research use only. Not for diagnostic use.

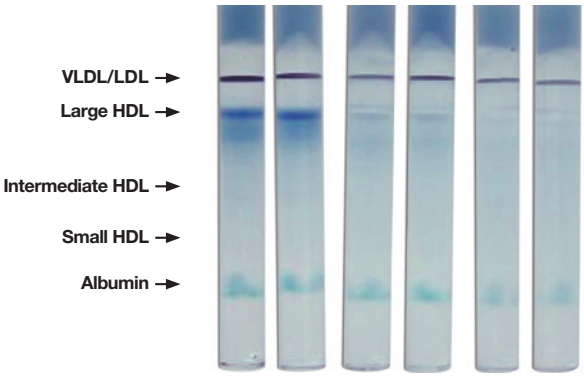


Figure 2: Lipoprint HDL Subfractions

BIBLIOGRAPHY

Barzilai N, Atzmon G, Schechter C, Schaefer E, Cupples A, Lipton R, Cheng, Shuldiner A. Unique Lipoprotein Phenotype and Genotype Associated With Exceptional Longevity. JAMA. 2003; 290(15):2030-2040.

Heijmans B, Beekman M, Houwing-Duistermaat, Cobain M, Powell J, Blauw G, van der Ouderaa F, Westendorp R, Slagboom P. Lipoprotein Particle Profiles mark Familial and Sporadic Human Longevity. PLOS Medicine. 2006; 3(12): 2317-2323.

Hoefner D, Hodel S, O'Brien J, Branum E, Sun D, Meissner I, McConnell J, Development of a Rapid, Quantitative Method for LDL Subfractionation with Use of the Quantimetrix lipoprint LDL System. Clinical Chemistry. 2001; 47(2): 266-274

Rizzo M, Berneis K. Low Density Lipoprotein Size and Cardiovascular Risk Assessment. QJ Medicine. 2006; 99: 1-14

Berneis K, Krauss R. Metabolic Origins and Clinical Significance of LDL Heterogeneity. Journal of Lipid Research. 2002; 43: 1363-1379

Rizzo M, Berneis K, Corrado E, Novo S. The Significance of Low-Density Lipoproteins Size in Vascular Disease. International Angiology. 2005; 25(1): 4-9

Beard C, Barnard R, Robbins D, Ordovas J, Schaefer E. Effects of Diet and Exercise on Qualitative and Quantitative Measures of LDL and Its Susceptibility to Oxidation. Ateriosclerosis, Thrombosis, and Vascular Biology. 1996; 16: 201-207

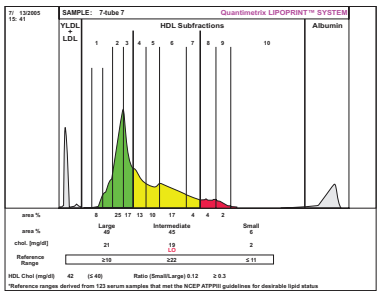


Figure 3. Large HDL
(subfractions 1 to 3)

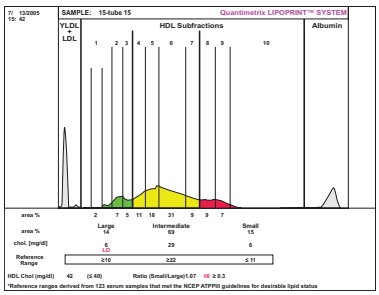


Figure 4. Intermediate HDL
(subfractions 4 to 7)

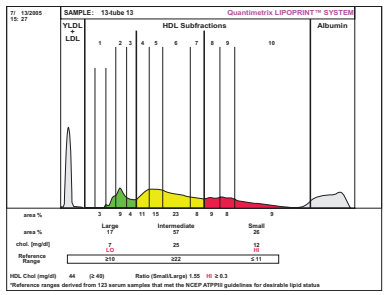
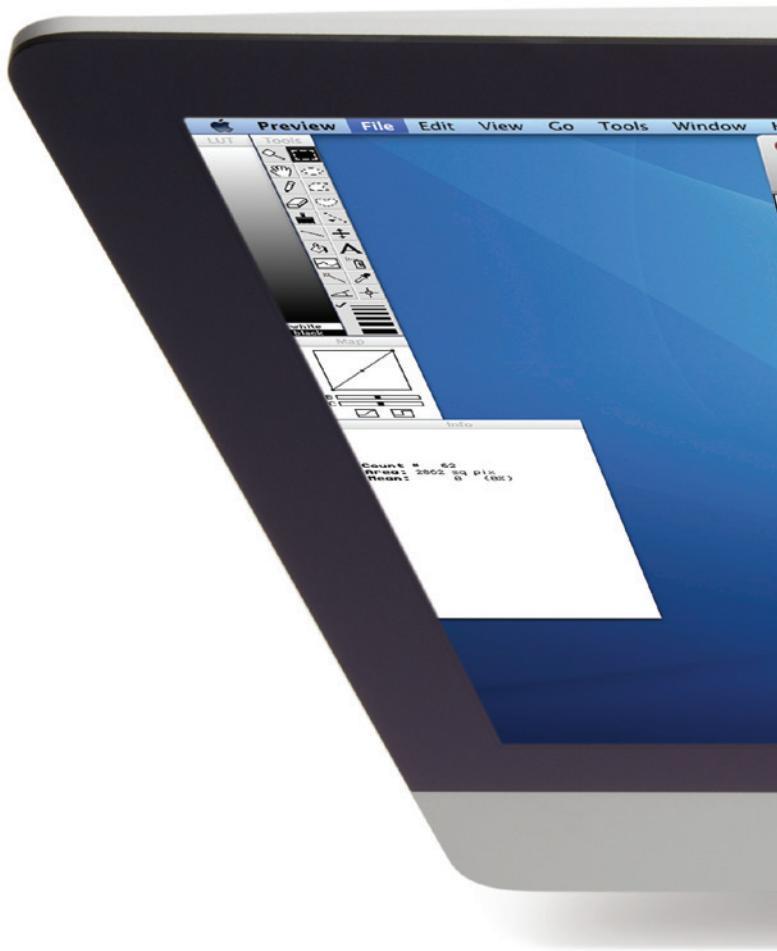


Figure 5. Small HDL
(subfractions 8 to 10)



Quantimetrix™
laboratory innovations for 40 years

2005 Manhattan Beach Blvd.
Redondo Beach CA 90278-1205 USA
(310) 536.0006
www.quantimetrix.com

10-0013 100114