



by Harold M. Bates, Ph.D.

HDL-Cholesterol And Coronary Heart Disease

In 1951, Dr. David P. Barr and his associates¹ at New York Hospital-Cornell Medical Center examined the cholesterol content of the serum high-density lipoprotein (HDL) fraction in normal individuals and in patients with conditions in which there is a variable disposition to atherosclerosis. Based on their results (Figure 1), they concluded: "The outstanding fact in our observations is the relative and absolute reduction in alphasipoprotein (HDL) in atherosclerosis."

Two years later, Dr. Barr² told the American Heart Association that the high proportion of HDL-cholesterol in human babies was similar to that found in dogs and rabbits who appear to have a high resistance to atherosclerosis. It is only in later life as HDL-cholesterol levels decrease that human beings become susceptible, and when atherosclerosis is induced in dogs and rabbits the HDL-cholesterol concentration is also decreased. The evidence supporting this view is shown in Figure 2.

Except for a prospective study in 1966 by Gofman et al.,³ who reported that HDL-cholesterol levels were lower than normal in young men who developed coronary heart disease, the findings presented by Barr's group were largely ignored until 1975. In that year, Miller and Miller⁴ presented evidence that HDL-cholesterol is inversely related to total body cholesterol, but

is unrelated to the plasma concentrations of total cholesterol and other lipoproteins.

Based on these studies and epidemiologic data, they postulated that low plasma concentrations of HDL-cholesterol may accelerate the development of coronary atherosclerosis by impairing the normal clearance of cholesterol from the arterial wall and its transport to the liver for catabolism and excretion.

The liver has been shown to be the only organ that can catabolize and excrete quantitatively important amounts of cholesterol.⁵ Viewing HDL as a "scavenger" lipoprotein, Miller and Miller⁴ fur-

ther proposed that the development of atherosclerosis might be more successfully prevented by increasing plasma HDL-cholesterol, and hence the clearance of cholesterol from the arterial wall, than by conventional attempts to reduce the plasma cholesterol and other lipoproteins.

Mechanism of HDL action

It is difficult to explain how lower-than-average levels of HDL-cholesterol make an individual more susceptible to coronary artery disease, whereas higher-than-average levels of HDL-cholesterol appear to delay the development of atherosclerosis and, in fact, seems to exert a protective effect. Most laboratory data dealing with the mechanism of HDL action have supported the view that HDL serves a carrier function, clearing cholesterol from the arterial tissues.

For example, one of the earliest studies on the mechanism of HDL action, reported by Glomset,⁶ showed that HDL enhances the enzymatic conversion of free cholesterol to esterified cholesterol by facilitating the action of lecithin/cholesterol acyltransferase (LCAT). Since esterified cholesterol molecules are larger than free cholesterol molecules and move

Figure 1.

	Mean % of Total Cholesterol as HDL	Mean Cholesterol (mg/dl)
Normal men and women (age 18-35)	29.3	192
Normal men and women (age 45-65)	23.2	245
Survivors of Myocardial Infarction	14.1	258
Familial Xanthomatosis	7.1	459
Nephrotics	5.1	577

Figure 2.

	Mean % of Total Cholesterol as HDL	Mean Cholesterol (mg/dl)
Human Infants (cord blood)	43.3	65
Dogs		
Normal	82.9	167
Atherosclerotic	5.8	2698
Rabbits		
Normal	52.7	41
Atherosclerotic	9.6	364

High risk for coronary heart disease

Low plasma levels of HDL
High plasma levels of LDL

Protection from coronary heart disease

High plasma levels of HDL
Low plasma levels of LDL

Figure 3

Lipoprotein cholesterol

At the present time, the "normal" level of plasma LDL in Western man is thought to be unphysiologically high, whereas the "normal" level of plasma HDL is considered to be unphysiologically low. The plasma concentrations of these lipoproteins are now thought to be related to the propensity of Western man to develop coronary artery disease for the following reason: LDL is supposed to be atherogenic by carrying cholesterol into the inner layer (the intima) of the arterial wall, and HDL is believed to protect persons from atherosclerosis by transporting cholesterol out of the vessel wall. To summarize, see Figure 3.

Recently, well-documented epidemiologic data from NIH-sponsored coronary heart disease studies in Framingham (Mass.), Albany (N.Y.), San Francisco, Honolulu, Evans County (Georgia), and Puerto Rico⁹⁻¹¹ show that a strong inverse relationship exists between HDL levels and coronary heart disease. Based on the published epidemiologic data, our laboratory provides physicians who order HDL tests the information shown in Figure 4 to help them predict the risk of coronary heart disease.

Measurement of HDL Levels

Our laboratory performs more than 10,000 HDL determinations each month. We follow the HDL procedure of the Lipid Research Clinic Program of NIH.¹² Briefly, a solution of heparin-manganese chloride is added to serum which is kept cold in an ice bath. The LDL and VLDL is precipitated from the serum by the heparin-manganese. The HDL, which remains in solution after centrifugation (1500 x g, 30 min, 4°C), is analyzed for cholesterol. We have found this method to be accurate, reproducible, and reliable, and, moreover, we find that it is particularly suitable for the large clinical laboratory.

A number of laboratorians prefer the sodium phosphotungstate-magnesium chloride precipitation

method of Burnstein et al.¹³ as described by Lopes-Virella,¹⁴ who reported that the procedure is more appropriate for routine clinical laboratory use than the heparin-manganese procedure. She claims that this method is "simple, reliable, precise, and inexpensive."

There are no diagnostic kits presently available that perform HDL assays. To perform HDL determinations, technicians must prepare their own reagents and standardize and quality control their test method. The HDL assay has been promoted as a "simple-as-ABC" type test by some medical reporters.

For example, in the February 23, 1977 issue of *Medical Tribune*, an article entitled "HDL a Simple Test" stated, "Testing for HDL is simplicity itself, says Dr. Castelli. The lab technician adds heparin and manganese chloride to a sample of plasma, and it precipitates all lipoproteins except HDL. The precipitates are then spun away on an ordinary lab centrifuge. An aliquot of supernatant material, containing only the HDL cholesterol, is then quantified in the usual lab procedure for total cholesterol."

In contrast, in the May 1977 issue of the *American Journal of Medicine*, Dr. Castelli's group¹⁰ states: "If HDL cholesterol is to become part of a standard risk profile for coronary heart disease, great care must be taken with laboratory precision. A good laboratory can achieve a technical error of 5 mg/dl in measuring this lipid. When it is remembered that an average HDL cholesterol for men is around 45 mg/dl whereas a significantly high risk of coronary heart disease is evident at 35 mg/dl, it is clear that a technical error of 5 mg/dl is by no means a comfortable one. More precise methods would be helpful." LM

A complete reference list is available from: The Editor, Laboratory Management, 750 Third Ave., New York, NY 10017.

more slowly, they have trouble passing back and forth across cell membranes. Thus, if cholesterol at the peripheral cellular level is held out of the cell for a time by LCAT action, there is a greater chance that it will be picked up by an HDL molecule and carried back to the liver for elimination.

In another study, Bondjers and Bjorkerud⁷ demonstrated that the in vitro efflux of cholesterol from atheromatous tissue is promoted by the addition of HDL to the incubation medium. And more recently, in a study of the metabolism of lipoproteins by arterial smooth muscle cells, Carew and his associates⁸ demonstrated that HDL competitively inhibits the uptake and degradation of low-density lipoproteins (LDL) by the cells. This ensures that the arterial smooth muscle cells accumulate less cholesterol. It appears that HDL binds to the surface of the muscle cells as effectively as LDL, but is internalized and degraded more slowly, thus significantly modifying the metabolism of LDL by these cells. According to these investigators, "The demonstrated interaction of HDL and LDL could be a second mechanism contributing to the protective effect of high plasma HDL concentrations in relation to atherogenesis."⁸

Figure 4

Serum HDL (mg/dl)	Risk of Coronary Heart Disease
25 or less	Risk at dangerous level
26 to 35	High risk
36 to 44	Moderate risk
45 to 59	Average risk
60 to 74	Below average risk
75 or more	Protection probable, associated with longevity.