Review of Literature
The relation between the levels of dietary and serum cholesterol has attracted the attention of the scientific community since Anichkov's cholesterol feeding experiments in rabbits at the beginning of this century.

Recent research has revealed a rather complex set of events that control plasma lipoprotein level. Specific proteins have been implicated in the regulation of lipoprotein synthesis. Besides this, many more factors like age, sex, smoking, obesity, hypertension, dietary habits and sedentary life style exert their influence on lipoprotein levels and development of atherosclerosis in their own way. Many of the risk factors are reversible but influences of age, sex and genetic factors are irreversible.

A single high cholesterol diet induced change in various subfractions of lipid lipoproteins and its relevance in predicting an individual's risk for future atherosclerosis still remains an unexplored field. The discovery of cell surface receptors for low density lipoprotein (LDL) by Dr. Joseph Goldstein, was fundamental to our understanding of how plasma cholesterol levels are controlled.

**Changes in Lipid Lipoprotein Levels After High Cholesterol Diet**

The effect of feeding cholesterol over a long term or short term period evokes variable responses and is subject to individual variation. Dietary fat and
cholesterol causes changes in specific lipoprotein in a variety of species (Mahley et al., 1978; Arora et al., 1987). Quantitatively, a change in specific lipoprotein may be dramatic in one species while insignificant in another.

**CHANGES IN TOTAL SERUM CHOLESTEROL**

In 1956 Ancelkeys and Anderson et al. concluded that serum cholesterol level is essentially independent of the cholesterol intake over the entire range of natural human diets. But later on it was proved beyond doubt that feeding cholesterol rich diet for 2–8 weeks raises total serum cholesterol in blood (Arora et al., 1987; Mesinger et al.; Conner et al., 1961; Deborah Applebaum et al., 1979).

In a recent study, (Martha and McMurry et al., 1991) a group of thirteen Tarahamurra Indians (a Mexican people known to consume a low fat high fibre diet and to have a very low incidence of risk for coronary heart disease) consumed their traditional diet (2700 Kcal per day) for one week, and were then fed a diet typical of affluent societies, which contained excessive calories (4700 Kcal per day), total fat, saturated fat, and cholesterol. After 5 weeks of consuming the affluent diet, the subjects' mean (±S.E.) plasma cholesterol level increased by 31 percent from 121±5 to 159±6 mg%. The increase in plasma cholesterol level was primarily in the low density lipoprotein fraction, which rose 39 percent from 72±3 to 100±4 mg%. HDL cholesterol, usually low in this population, increased by 31%.
An effect of dietary cholesterol in raising serum cholesterol levels has also been demonstrated in metabolic ward experiments and in some but not all studies of free living subjects. In studies by Connor and Connor (1989) serum cholesterol was increased by a dietary cholesterol intake up to approximately 400 to 500 mg/day; a higher intake had a minor additional effect. These findings imply that the efficiency of cholesterol absorption decreases as the amount of dietary cholesterol increases.

Recently, McNamara reviewed the data from 68 clinical studies published over a span of nearly 30 years. He found fairly consistent results, averaging an increase in serum cholesterol of 2-3 mg/dl (0.06 m mol/l) for each 100 mg increase in dietary cholesterol - quite different from the changes of 9.6 and 6.8 mg/dl (0.25 and 0.18 m mol/l) predicted by the classic Hegsted and Keys formulae, which describe linear and square root relations, respectively, between cholesterol consumption and serum cholesterol levels.

In an earlier report, Bruhn (1940) observed a 20% rise in mean cholesterol level after a fat load. Effect of high cholesterol fat load on post prandial cholesterol levels have also been studied in the past by several workers, but insignificant difference has been found between post prandial and 10 to 14 hours fasting value (Albrink and Man, 1956; Pomeranze et al, 1954, Schilling et al, 1964). All these workers observed plasma cholesterol values up to 24 hours after a test meal. On the other hand Nikkila and
Konttinem (1962) demonstrated a significant decrease in cholesterol level six hours after a fat diet in healthy soldiers.

Hanno Krauss, Picter Groot (1987) reported insignificant changes in total serum cholesterol fater feeding 0.5 gm/M^2 of cholesterol and taking readings at 2 hourly interval for 14 hours.

In adolescents with initial cholesterol levels greater than 200 mg/dl, a 50 percent decrease in cholesterol intake led to an appreciable drop (15.5%) in cholesterol levels, but the effect was much more modest (8.3%) in those with lower initial levels (Gandey et al, 1972).

In another large survey of school children, there was no positive correlation between the low (80-130 mg/dl) the intermediate (157 to 180 mg/dl) and the high (194 to 425 mg/dl) cholesterol levels with the mean daily intake of energy, sugar, fat, saturated fat and cholesterol (Weidman et al, 1978). However, in 7 different studies, summarised recently, significantly weak correlations were noted between serum lipids and dietary P/S ratio(Mellies and Glueck, 1983).

Textured vegetable proteins lowered total serum cholesterol in hypercholesterolemic subjects with no change or slight elevation in HDL cholesterol, no effect or only minor changes have been observed in normolipidaemic subjects (Sirtori et al, 1985).

The replacement of animal protein with vegetable protein in the diet has been suggested to reduce the diet
linked atherogenic risk (Carroll, 1982).

However, Sacks et al (1983) found no appreciable correlation between total intake of protein, when consumed above minimum requirement and serum cholesterol level.

Work in animals showing that sucrose and fructose are atherogenic prompted human studies, which have not shown consistent changes. In one study isocaloric replacement of starch with sucrose in mixed diet did not lead to changes in serum cholesterol (Mann and Truswell, 1972), changes that were documented in another study (Reider et al, 1978).

HIGH DENSITY LIPOPROTEINS (HDL)

High density lipoproteins are lipid lipoprotein complexes defined by flotation in the ultra centrifuge between density 1.063 and 1.21 g/ml, by the presence of major protein constituents, apolipoprotein A-I and A-II, and by alpha migration on electrophoresis. Three classes of HDL are separated on the basis of flotation rates on ultracentrifugation; HDL₃ have flotation rates between 0-3.5, HDL₂ have rates in excess of 3.5. The third and minor HDL₁ is sometimes found at d <1.063 and overlaps with the low density lipoprotein distribution. Recently Mahley and colleagues have identified a distinct sub type of HDL designated as HDLc or apo E-HDLc. This is found in the plasma of cholesterol fed animals and to a much smaller extent in human fed high cholesterol, high saturated fat diets. HDLc differs from other sub type by presence of
apolipoprotein E. This property confers an affinity for the low density lipoprotein receptor (Mahley and Weisgraber, 1978).

The lipid constituents of HDL exhibit variations. Cholesterol ester content may range from 10-20 percent, triglycerides are normally less than 4 percent. The ratio of cholesterol to triglycerides in HDL may show wide fluctuations with increase being observed after dietary cholesterol supplementation (Mistry et al, 1977) and decrease being found in patients with hypertriglyceridemia (Weisweiler et al, 1977), Uremia (Brunzell et al, 1977) and Ischaemic Heart disease (Carlson et al, 1975).

Conflicting reports have appeared on effect of dietary cholesterol on HDL levels:

Borden et al (1964) reported enhanced levels of HDL-c in rats fed cholesterol while Haf et al (1962) and Kritchevsky (1965) reported no change in HDL levels in cholesterol fed rats. Reiser et al (1966) and Howard et al (1968) reported decreased level of HDL cholesterol in rats fed with high cholesterol diet.

Narayan (1971) demonstrated that HDL₂ decreased drastically - about 50% in rats fed with high cholesterol diet. These results confirmed the earlier observations of Reiser et al (1966) that rat serum HDL level was decreased irrespective of whether a saturated or unsaturated fat was used in the diet supplemented with cholesterol. In short term feeding studies, marked reduction in dietary fat and isocaloric increase in carbohydrate resulted in decrease
in HDL cholesterol in conjugation with elevation of serum triglyceride and VLDL. Studies of HDL composition have shown a decrease in ratio of apolipoprotein A-I to A-II and a decrease in HDL cholesterol to protein ratio (Schonfeld et al, 1976) consistent with a selective decrease in HDL₂ species (Blum et al, 1977).

There is evidence that substitution of large quantities of poly-unsaturated fat for saturated fat in diet can result in lower levels of HDL lipids and proteins (Nichaman et al, 1967). An increase in the P/S fat ratio from 0.25 : 1 to 4 : 1 in food diet fed to four normal subjects for five weeks resulted in reduction of HDL cholesterol and apolipoproteins A-I concentration of 33 and 21 percent respectively, with an associated reduction in HDL₂ : HDL₃ ratio (Shepherd et al, 1978). Other studies have however, reported either no change (Lewis, 1978; Shore et al, 1981) or increase (Jackson and Glueck, 1980) in levels of HDL cholesterol with feeding of diets enriched in poly-unsaturated fats. High dietary intake of cholesterol, in the form of three to six egg yolks per day, has been reported to produce increase in apo-lipoprotein E containing HDL - sub species in human (Mahley et al, 1978). This effect was seen whether or not there was an increase in total plasma cholesterol. Despite the fact that HDL containing apolipoprotein E represented only a minor fraction to the total HDL its presence was shown to account for an increase of 2.6 to 4 times the
binding of HDL to LDL receptors of fibroblasts as compared to pretreatment HDL (Mahley et al, 1981). But this was not observed in another study (Applebaum et al, 1979). Recently it has been reported that level of HDL cholesterol and serum apolipoprotein A-I but not apolipoprotein E increased with the feeding of diets high in both cholesterol and saturated fat (Tan et al, 1974).

A final consideration in evaluating the effects of dietary variables on HDL is that, while levels of HDL cholesterol and plasma apolipoprotein A-I are similar after overnight fast and non fasting states (Henderson et al, 1980), Changes in levels and composition of HDL have been shown to occur acutely after meals containing fat. Cholesterol phospholipid and C-apolipoprotein levels in HDL₂ increases and cholesterol in HDL decreases (Havel, 1973; Saggio et al, 1980) in conjunction with transfer of chylomicron lipids to HDL during the course of their catabolism. Recently, it has been shown that HDL apolipoprotein A-I levels increased when fat was consumed in divided doses over a 10 hour period but not when the same amount of fat was ingested as a single load (Kay et al, 1980).

LOW DENSITY LIPOPROTEIN CHOLESTEROL (LDL-c)

More than 75 percent of the total cholesterol present in the plasma is in the form of LDL-c. LDL-c is generated by the degradation and removal of triglyceride from very low density lipoprotein (VLDL) in the plasma.
Their density is in the range of 1.019 to 1.063 and they contain apoprotein B 100.

In 1977, Goldstein hypothesized the concept of LDL receptor. The presence of these receptors have been confirmed by many laboratories. LDL receptors are present on the cell surface of liver, adrenal cortical cell, lymphocytes, muscle cells and renal cells. LDL that binds to this receptor is taken up by the receptor mediated endocytosis and digested by lysosome within the cells. The cholesterol esters of LDL are hydrolyzed by a lysosomal cholesteryl-esterase and the liberated cholesterol is used both for membrane synthesis and as a precursor for steroid hormone synthesis. Liver uses the LDL-c for synthesis of bile acids and for generation of free cholesterol which is secreted into the bile.

Diets high in fat and cholesterol cause an elevation in LDL in most animals (Mahley, 1978). The response in man varies, but in those subjects who have an elevation in plasma cholesterol, there is an elevation in plasma LDL levels. In 1979 Deborah-Applebaum et al demonstrated significant rise of LDL level in human volunteers after feeding 5000 mg of egg yolk cholesterol per day for 30 days.

Age related difference in rise of LDL was demonstrated by Arora and Gupta G (1987). They found that rise of total serum cholesterol after feeding high fat high cholesterol breakfast for one week was much more pronounced in young volunteers with major portion of rise being contri-
buted by increased HDL. In contrast, in the older age groups, the rise of total serum cholesterol was less marked with LDL-c contributing mainly towards the increased levels.

Baudet et al (1981) demonstrated that there was significant fall in level of LDL in five volunteers, three and five hour after taking a butter diet. They attributed this fall to a defect in VLDL hydrolysis by serum lipases and due to metabolic blocking in liver or adipose tissue.

An additional alteration in LDL, induced by feeding high cholesterol diets involves the apoprotein constituents. In normal LDL the B-apoprotein is the major detectable apo-protein moiety. However, in several species, the LDL contains a variable amount of the E apoprotein following cholesterol feeding (Mahley et al, 1977; Rudel et al, 1979).

MECHANISMS FOR CONTROL OF PLASMA CHOLESTEROL LEVELS

Research till date has established a central role for lipoprotein receptors in regulating plasma cholesterol metabolism and transport. The homeostatic and regulatory mechanisms that maintain a relatively constant level of plasma cholesterol despite changes in dietary cholesterol intake include alterations in the efficiency of intestinal absorption and in the rates of cholesterol biosynthesis, LDL receptor activity, secretion of cholesterol into bile and hepatic conversion of cholesterol into bile acids, the chief metabolic product of cholesterol (Mistry and Miller et al, 1981; McNamara et al, 1987).
Cholesterol absorption is a complex process involving a number of steps. Although the rate limiting step is not known with certainty, it is currently believed to be the transport of micellar free cholesterol from the intestinal lumen through the unstirred water layer and, in molecular form across the cell membrane of the enterocyte. This had classically been thought to occur by passive diffusion, but several studies suggest a role for an yet unidentified brush border protein. Decreased absorption in the presence of increased dietary cholesterol serves as a major control mechanisms in cholesterol hemeostasis, but there is a great individual heterogeneity (Quintao et al, 1971). Miettinen and Kesaniemi et al (1988) cite the increased absorption of exogenous cholesterol by persons with apolipoprotein E4 as contributing to the heterogeneity in response to dietary cholesterol.

Another major adaptive mechanism is the rate of conversion of dietary cholesterol to bile acid and the loss of either cholesterol or bile acid in the stool. In 1972, Lofland et al found that hyporesponsive monkeys had increased biliary excretion of cholesterol. Recently Fred Ker et al (1991) studying a case of a normocholesterolemic 88-year old man who had been consuming 25 eggs a day for the last 15 years reported that the rate of bile acid synthesis in the patient was greater than in any of the 200 subjects studied by them for the last 13 years.
ROLE OF LDL RECEPTORS

Operationally, the LDL/LDL-receptor system can be considered the primary transport mechanism for endogenous cholesterol. LDL are generated in the plasma by the degradation of intermediate density lipoprotein (IDL). Generated LDL is removed relatively slowly from the plasma by binding to LDL receptors in the liver and extra hepatic tissues (Kita et al., 1982). In rabbits, rats and hamsters, more than half of the total LDL receptors are located in the liver. However, the precise distribution of these receptors in man is unknown.

REGULATION OF HEPATIC LDL RECEPTOR

Hepatic LDL receptors are suppressed whenever the liver's content of cholesterol increases or its demand for cholesterol is reduced. Thus receptor suppression occurs when a high cholesterol diet is consumed (Hui et al., 1981), or when bile acids are infused (Angelin et al., 1983). Conversely, LDL receptors increase when hepatic cholesterol synthesis is blocked by the drugs compactin or mevinolin (Goldstein et al., 1982; Belheimer et al., 1983), when bile acid binding resins are given (Shepherd et al., 1980), or when an ileal bypass is created (Spengel et al., 1982). Fasting has also been shown to suppress the LDL receptor in rabbit (Goldstein et al., 1982). LDL receptors can be stimulated by thyroxine (Thompson et al., 1981). Hepatic LDL receptors decline when rabbits are fed a diet composed only
of sucrose and casein (Chao et al, 1982). All these changes in receptor activity alter the rate of uptake of LDL by the liver and cause reciprocal changes in plasma LDL levels. Whenever hepatic LDL receptors are suppressed, the plasma LDL level rises, conversely, whenever, these receptors are induced, the plasma LDL level falls.

**TRIGLYCERIDES AND VERY LOW DENSITY LIPOPROTEIN (VLDL)**

The level of serum triglyceride (TG) rises considerably after fat ingestion. Rise in the triglyceride level after fat ingestion has been reported after giving different amounts of fat load and measuring the blood level at different intervals thereafter (Nikkila and Kontinen, 1962; Danborough et al, 1963). Angerwall (1963) has reported a significant correlation between fasting, 3½ hours and 7½ hours values of serum triglyceride postprandially.

Clefsky et al (1976) noted a biphasic plasma triglyceride curve with an initial peak occurring 1 to 3 hours after feeding and a second peak after 4 to 7 hours. The primary peak was accounted for by increase in chylomicron levels in more than 98% cases, whereas secondary peak represented a rise in VLDL level in 82% of the cases. Previously however, Havel et al (1957) had concluded that increment in the concentration of triglycerides in the serum following ingestion of fat is entirely the result of an increase in their concentration in VLDL.

Excess production of VLDL and triglycerides is more often due to secondary abnormalities than to primary
factors, perhaps the most common cause is high caloric intake associated with obesity, excess alcohol and excess carbohydrate. Increased levels are also found in diabetes mellitus, nephrotic syndrome and hypothyroidism with obesity. Delayed clearance of triglyceride from the serum is noted in cases of ischaemic heart disease after high fat diet (Arora et al, 1987, David et al, 1961).

**VLDL REMNANTS**

In addition to a report by Mistry et al (1976) that beta VLDL can be induced by cholesterol feeding in man, preliminary studies from the Gladstone Foundation Laboratories for Cardiovascular Disease indicate that certain individuals respond to high fat, high cholesterol diet by producing lipoproteins which are capable of delivering cholesterol to macrophages. The beta VLDL may occur transiently as minor components of the human plasma fractions after diets high in fat and cholesterol are consumed, and may cause repeated cholesterol deposition in cells in the arterial wall over the years. The beta VLDL, either chylomicron remnants or hepatic lipoprotein may represent the atherogenic particle postulated several years ago by Zilver- smit. This alteration in the lipoprotein fraction may represent the most significant diet induced changes in lipoprotein predisposing to accelerated atherosclerosis.

**CHYLMICRONS**

Chylomicrons are a means of lipid transport in the
exogenous pathway. They are large lipoprotein particles containing dietary triglyceride and cholesterol. Chylomicrons are rapidly cleared from the plasma and normally are not present after an overnight fast. The detection of these particles in fasting plasma samples, therefore, is always abnormal and may indicate presence of other hyperlipidaemias. Increased levels of chylomicrons in the plasma may be found in cases of genetic defects involving the enzyme lipoprotein lipase and in the familial form of hypertriglyceridemia.

ENDOCRINE & METABOLIC DISORDER AND LIPOPROTEINS

Changes in plasma lipoprotein levels in diabetes remain one of the most important risk factors in terms of accelerated atherosclerosis (Santen et al, 1972). In addition, diabetics may have altered lipoprotein structure and metabolism independent of increase in plasma lipid levels (Eckel et al, 1981; Howard et al, 1978 & Schonfeld et al, 1974), and these altered lipoprotein may be associated with accelerated atherosclerosis. It is generally appreciated that anatomic evidence of accelerated atherosclerosis frequently develops in insulin dependent diabetic patients, ten to fifteen years after onset of diabetes.

Hypertriglyceridemia is the most common lipid abnormality observed in diabetic patients. The main component of hypertriglyceridemia is VLDL and rarely chylomicrons (Nikkila et al, 1973 and 1974). VLDL can cause secondary hypercholesterolaemia. The hypercholesterolemia in diabetes
can occur because:

a. Increased plasma VLDL level causes a secondary increase in plasma cholesterol levels, since 20% of total lipid content of VLDL is cholesterol.

b. Diabetes affects the plasma LDL metabolism—the exact mechanism is not clear, but it has been proposed that:
   1. The increased synthesis of VLDL in diabetes causes increased LDL formation, since VLDL is a precursor for LDL
   2. Decreased catabolism of LDL in poorly controlled diabetes due to glycosylation of plasma LDL (Klitzman, 1982), which alters the configuration of LDL, so that less interaction occurs with specific receptors responsible for the majority of LDL catabolism in normal persons (Brown et al, 1981).

Decreased conversion of VLDL to LDL and impaired LDL clearance are two opposing phenomena which may influence the LDL concentration of diabetics in either direction. Thus, despite minimal changes in LDL concentration, there are multiple defects in metabolism of LDL in non-insulin dependent diabetes, which may contribute to increased atherogenesis in this disorder (Howard and William et al, 1987).

In the Rockefeller monograph (Allen, 1919), "Lipaemia is largely associated with fat intake and other diabetic symptoms", according to Ervin (1919), the lipaemia in diabetics will disappear with the elimination of fat from diet. Joslin (1921) suggested a relation between high protein fat diet and a high degree of lipaemia, he stated
that with restricted diet, particularly of fat, the blood fat rapidly falls. Bloor (1921) stated that there was a deficiency of pancreatic hormone which is essential for the proper removal of fat from the blood.

Low fat and high carbohydrate in diet: The avoidance of low fat and high carbohydrate in diets of diabetic patients during treatment have been shown to result in lower serum cholesterol, lower insulin requirements, improved glucose tolerance and reduced severity of vascular complications (Ellis et al, 1934; Rabinowitch et al, 1935; Singh et al, 1955; Kampher et al, 1958; and Van Eck et al, 1959). On the other hand, hyperlipidemia has been noted in non diabetics during administration of low fat, high carbohydrate diets. The lipoaemic effect of such a diet may be a temporary one. In a study, the reduction of dietary fat over a long period showed that serum triglyceride returned to normal after several months (Autonis et al, 1961).

Recent studies in our department (Arora, Agarwal and Singh et al, 1989) on diabetic subjects given a high cholesterol test diet followed by evaluation of postprandial changes in lipid profile, revealed that one hour after such a meal serum triglyceride levels showed a fall in 40% cases, a rise in 50% cases, whereas remaining 10% cases showed no change. However, these levels were elevated in all cases 3 hours after the test meal. Changes in LDL levels exhibited a similar pattern, while HDL levels did not show any significant alteration, even though the basal HDL levels were higher.
CHRONIC LIVER DISEASE AND LIPOPROTEINS

Hypercholesterolemia and lipoprotein abnormalities have been reported in patients of primary biliary cirrhosis. There is an increased amount of unesterified cholesterol, along with increased levels of chylomicrons, and VLDL. These abnormalities are probably due to hepatic lipase inhibition as well as altered cholesterol esterification in patients of this disease (Jahn and Schaefer et al, 1985).

Manocha et al (1989) analysed fasting plasma samples from 29 patients of cirrhosis, for cholesterol, triglycerides and their lipoprotein fractions. The patients included 11 alcoholic cirrhotics consuming over 130 gm/day of absolute ethanol and 18 non-alcoholic cirrhotics. The difference in lipid values between the two patient groups was not significant except that VLDL cholesterol was raised in alcoholic cirrhotics. However, in comparison to normal healthy controls, the values were significantly altered. The dietary intake in the two groups showed no difference, except that the non alcoholic cirrhotics consumed more animal proteins. Low intake of exogenous fat and reduced synthesis of endogenous cholesterol in cirrhotic patients seemed to influence the total lipid values (Stigendahl and Olsson et al, 1984).

Alcohol ingestion per se has been reported to raise levels of HDL (Johansson et al, 1974; Belfrage et al, 1977). But the results of Glueck et al (1980) were contradictory to the above statement.

In a large epidemiological study, levels of HDL cholesterol and amount of habitual alcohol intake in
moderate range have been independently correlated (Castelli et al, 1977). A recent longitudinal study (Margerave et al, 1991) entailing follow up in 1983-89 of men investigated during a study in 1976 revealed that even though alcohol consumption decreased over the 12 years, there was no significant relation between the fall in HDL cholesterol concentration and fall in alcohol consumption.

**CHRONIC RENAL INSUFFICIENCY AND LIPOPROTEINS**

Abnormalities of lipoprotein metabolism leading to dyslipoproteinaemia are present already in the early stages of renal insufficiency, even though at this early stage, these abnormalities are not detected by measurements of plasma lipids (Attman and Alaupovic, 1990).

Cholesterol ester is the preferred form for cholesterol storage in the liver, since an increase in total hepatic cholesterol is reflected more as esterified than free cholesterol. A possible relationship between cholesterol ester turnover and lipoprotein transport has emerged from the study of Nestel and Associates (1968; 1970). Cholesterol ester turnover has been found to be raised together with triglyceride turnover in subjects with the nephrotic syndrome (McKenzie and Nestel, 1968).

Low plasma HDL cholesterol concentrations in patients with CRF are related to decreases in the synthetic rate of apo A-I/HDL (Martin and Lee et al, 1990). Whereas overproduction of lipoproteins containing apoprotein B is the principal cause of hyperlipidaemia in patients with
the nephrotic syndrome (Joven and Villabona et al, 1990). Karadi and Romics et al (1989) found that serum lipoprotein (a) levels may be increased in patients of nephrotic syndrome, particularly those with membranous or membranoproliferative glomerulonephritis or primary amyloidosis. But measurements of HDL in patients with the nephrotic syndrome have yielded contradictory results, because lipoprotein (a) floats in the same density range as HDL₂, hence serum HDL concentrations determined solely by ultracentrifugation may be falsely elevated by contamination with substantial amounts of lipoprotein-(a) (Kostner et al, 1983). However, the composition of plasma cholesterol esters is only minimally affected, by a single meal of a specific fat (Kayden et al, 1963), but is readily influenced thereafter.

Patients with chronic renal failure (CRF) tend to have lower than normal plasma HDL cholesterol concentrations (Lewis et al, 1966; Rapoport and Aviram et al, 1978; Goldberg and Harter et al, 1983), although the mechanism responsible for this defect has not been defined. Decreases in HDL cholesterol are often associated with increases in plasma VLDL-TG concentration (Schaefer et al, 1978; Fuller and Pinney et al, 1978) and in two of these instances - endogenous hypertriglyceridermia and NIDDM - it has been shown that the HDL catabolic rate is faster than normal (Saku et al, 1985; Golay and Zech et al, 1987). Hypertriglyceridermia also occurs in patients with CRF, but whereas
patients with endogenous hypertriglyceridemia and NIDDM have an increase in VLDL-TG synthetic rate, a decrease in VLDL-TG synthetic rate is seen in patients with chronic renal failure (Reaven & Swenson et al, 1980). Thus it seems possible that the abnormality in HDL kinetics might also be different in patients with chronic renal failure. Recent studies (Martin and Lee et al, 1990) provided support for the view that low plasma HDL cholesterol concentrations in patients with CRF are related to decreases in synthetic rate of apo A-I/HDL.

There is also evidence that patients with CRF have a factor in their plasma which inhibits lipoprotein lipase (LPL) activity (Murase et al, 1975) and it has also been shown that the lower the LPL activity, the lower the plasma HDL cholesterol concentration (Nikkila and Taskinen et al, 1978).

Patients with CRF are at an increased risk for coronary artery disease (Lindner and Charra et al, 1974) as are patients with NIDDM and endogenous hypertriglyceridemia. None of these clinical syndromes are characterized by an increase in plasma LDL cholesterol concentrations, suggesting that some other mechanism must account for the increased prevalence of coronary artery disease. An obvious contender for this role is the decrease in plasma HDL cholesterol concentration seen in patients with CRF, NIDDM or endogenous hypertriglyceridemia. In this context, it seems worth emphasizing that patients with CRF have a low plasma
HDL cholesterol concentration and a slower than normal fractional catabolic rate (FCR) of apo A-I/HDL. This combination of defects is in marked contrast to the situation in patients with NIDDM and endogenous hypertriglyceridemia, in whom a faster than normal FCR of apo A-I/HDL is associated with a low plasma HDL cholesterol concentration (Fidge and Nestel et al., 1980; Saku and Gartside et al., 1985). Thus, it is not possible to predict the change in HDL kinetics that will be present in patients with a low plasma HDL cholesterol concentration. These observations focus attention on the relationship between the FCR of apo A-I/HDL, plasma HDL cholesterol concentration, and coronary artery disease. It is generally assumed that it is a defect in reverse cholesterol transport that explains why patients with a low plasma HDL cholesterol are at increased risk for coronary artery disease (Miller and Miller, 1975).

**CHRONIC OBSTRUCTIVE PULMONARY DISEASE AND LIPOPROTEINS**

Specific non-cancer respiratory causes of mortality have been infrequently studied in relation to cholesterol concentration. One of the few ongoing long-term cohort studies, that has been the source of most such data, is the Whitehall study, in which 18,403 men aged 40 to 64 years were examined between 1967 and 1969. Plasma cholesterol concentrations were available in 17,718 of these men.
Subjects with respiratory symptoms had lower cholesterol levels than those without respiratory symptoms, and this was reflected in the relationship between cholesterol concentration and quartile of $FEV_1$ : respiratory symptoms and low $FEV_1$ are associated with increased overall mortality as well as mortality from respiratory conditions (Ebi-Kryston, 1988). Subjects who reported that they had experienced unexplained weight loss over the year preceding examination had a mean plasma cholesterol concentration of 0.31 mmol/1 lower than those who had not experienced weight loss, and their relative rate of mortality was 1.64. Mortality rates were similarly elevated for lung cancer (1.75) and non cancer respiratory disease (2.6).

These findings are in agreement with those from similar other studies (Kozarevic and McGee et al, 1981; Kagan and McGee et al, 1981; Neaton and Blackburn et al, In press), viz. that respiratory disease mortality has shown inverse associations with cholesterol levels. In the present study also (Smith and Shipley et al, 1992), mortality rates from all respiratory disease, plus the subcategories of bronchitis, pneumonia and other respiratory disease displayed consistent inverse associations with cholesterol level - a trend which remained unaltered even after 15 years of follow up. On the other hand, cholesterol concentration showed a continuous positive relationship with coronary heart disease, which is in agreement with results
from other large cohorts (Tornberg and Holm et al, 1989; Neaton and Kuller et al, 1984). Cancer mortality showed no trend with plasma cholesterol concentration.

However, of the individual cancer sites, only lung cancer demonstrated a consistent inverse trend with cholesterol concentration, but pancreas and liver cancer rates were highest in the lowest cholesterol group. Smoking would be the obvious confounder in the relationship between cholesterol level and lung cancer and respiratory disease mortality. There was however, no suggestion of any association between smoking and cholesterol concentration in this cohort. When cancers were divided into those that have consistently been related to smoking (Doll and Peto et al, 1975; Hammond et al, 1966), and those that have not, the negative relationship with cholesterol was seen only for the former. For the smoking related cancers, the mortality rate was highest in the lowest cholesterol group than in the rest of the population.

It could be argued that if low cholesterol levels predispose persons to respiratory mortality, low cholesterol levels will also predispose persons to respiratory disease before death occurs. If this were the case, then adjusting for the presence of respiratory symptoms would be inappropriate. The present study does not allow examination of the temporal relationship between the development of respiratory morbidity and plasma cholesterol level. Studies in
humans and other primates have, however, demonstrated that respiratory infections lead to lowered plasma cholesterol levels (Kerttula and Weber et al, 1988; Fiser and Denniston et al, 1972). There are two implications of this. First, repeated respiratory infections are a feature of some forms of chronic respiratory morbidity, which could be responsible for the lower plasma cholesterol levels in patients with chronic obstructive pulmonary disease. Second, patients suffering from respiratory infections at the time of examination would have lowered cholesterol levels and also would be at higher risk for further respiratory infections and death attributed to respiratory disease. This could produce the inverse association between plasma cholesterol level at examination and future respiratory disease mortality. Respiratory disease does, therefore, appear to lead to lowered cholesterol levels, whereas there is no substantive evidence to suggest that the converse occurs.

In the Framingham study, subjects whose serum cholesterol concentrations fell between examinations had elevated mortality rates (Anderson and Castelli et al, 1987), which could have been due to illness causing cholesterol levels to decrease. Falling cholesterol levels, however, could have different biological effects than consistently low levels, and the means of lowering cholesterol levels used in the intervention studies could, by themselves, lead to increased mortality (Davey Smit & Pekkanen et al, in Press).