HYPERLIPIDEMIA POST HEART TRANSPLANTATION

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ABSTRACT

Hyperlipidemia Post Heart Transplantation

Hyperlipidemia is prevalent following heart transplantation, and may play a role in the development of late graft atherosclerosis. The charts of 35 heart transplant recipients (n=32 males and 3 females) were reviewed retrospectively up until three years post transplantation, to describe a time-course of hypercholesterolemia after transplantation, and to determine the mechanisms involved in its pathogenesis. All patients received prednisone, cyclosporine, and azathioprine for immunosuppression. A progressive rise in both serum cholesterol (2.4 ± 0.4 mmol/l, p < 0.01), and body weight (8.4 ± 1.6 kg, p < 0.01) were observed during the first 8 and 10 months respectively. Levels stabilized thereafter, remaining above pretransplant levels. Triglyceride, low-density lipoprotein cholesterol, and high-density lipoprotein cholesterol concentrations were all above normal limits following transplantation. Tapering of prednisone dose had a significant effect on serum cholesterol levels, whereas diet had a beneficial effect on body weight. A randomized, controlled, dietary intervention study then followed to further assess the effect of dietary intervention on minimizing or preventing post transplantation hyperlipidemia and weight gain. Five patients were counselled the Step One Lipid-Lowering diet, two patients were controls. All study patients demonstrated a lower overall
increase in serum cholesterol levels than other transplant recipients. Reported nutritional intakes were similar between both groups. Increases in body weight were related to increases in body fat. Patients in the diet group demonstrated improvements in their level of nutrition knowledge, which correlated with lower serum cholesterol levels. Changes in serum cholesterol were also associated with appetite, hunger, perceived interest, perceived benefits, perceived barriers, and attitudes toward food. Changes in body weight were associated with appetite, hunger, perceived barriers, and stress. As a result of small sample size, the effect of dietary intervention was difficult to assess, but results warrant the need for further intervention trials.
Hyperlipidémie Post-Transplantation Cardiaque

L'hyperlipidémie survient fréquemment après une transplantation cardiaque, et pourrait jouer un rôle dans l'apparition de l'athérosclérose tardive du greffon. Les dossiers de 35 receveurs de transplantation cardiaque (n=32 hommes et 3 femmes) furent analysés rétrospectivement jusqu'à trois ans post greffe, afin de décrire la chronologie de l'hypercholestérolémie post-transplantation cardiaque et d'étudier les mécanismes impliqués dans sa pathogénèse. Tous les patients reçurent de la prednisone, de la cyclosporine et de l'azathioprine comme immunosuppression. Une hausse progressive du cholestérol (2.4 ± 1.4 mmol/l, p < 0.01) et du poids corporel (8.4 ± 1.6 kg, p < 0.01) furent observés durant les premiers 8 et 10 mois respectivement. Les niveaux se stabilisèrent ensuite, demeurant en-dessus des niveaux pré-transplantation. Les concentrations de triglycérides et cholestérol LDL et HDL furent au-dessus des limites normales post-transplantation. Un sevrage de la prednisone diminua les niveaux de cholestérol de façon significative, alors que le régime eut un effet bénéfique sur le poids corporel. Une étude d'intervention diététique, randomisée et contrôlée suivit afin d'évaluer l'effet d'une intervention diététique pour prévenir ou diminuer l'hyperlipidémie et la prise de poids qui suivent la transplantation cardiaque. Le régime hypolipidémiant niveau un fut enseignée à cinq patients, deux autres patients étant témoins. Les patients
de l'étude eurent une hausse de cholestérol inférieure à celle des autres patients. Les apports nutritionnels furent similaires entre les deux groupes. L'augmentation du poids fut reliée à une augmentation du gras corporel. Les patients du groupe traité eurent une amélioration de leur niveau de connaissances en nutrition, qui fut en corrélation avec un niveau de cholestérol plus bas. Les changements du cholestérol sérique furent également associés avec l'appétit, la faim, les intérêts perçus, les bénéfices perçus, les barrières perçus, et les attitudes envers la nourriture. Les changements du poids corporel furent associés avec l'appétit, la faim, les barrières perçus et le stress. En raison du petit nombre de patients, l'effet de cette intervention diététique est difficile à évaluer, mais ces résultats justifient le besoin d'études d'intervention supplémentaires.
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UNIQUE CONTRIBUTIONS

To our knowledge, this is the first randomized, controlled, dietary intervention trial performed to determine the effect of diet on post transplantation hyperlipidemia and weight gain. It is also the first study to measure changes in body composition following transplantation, as well as to examine whether factors such as appetite, hunger, attitudes, beliefs, stress, and knowledge contribute to post transplantation hypercholesterolemia and weight gain.

This research demonstrates that: 1) dietary intervention may be beneficial in minimizing hypercholesterolemia post transplantation, 2) the increase in body weight post transplantation is related to an increase in body fat, and 3) contributing factors to post transplantation weight gain include appetite, hunger, perceived interest in changing eating habits, perceived barriers to changing eating habits, and stress, and to post transplantation hypercholesterolemia include appetite, hunger, perceived interest in changing eating habits, perceived barriers to changing eating habits, perceived benefits of changing eating habits, attitudes toward food, and nutrition knowledge.
CHAPTER ONE

LITERATURE REVIEW
INTRODUCTION

Cardiac transplantation was first performed in Cape Town, South Africa in the year 1967 by Christian Barnard (Barnard, 1967). Since then, there have been over 12,000 heart transplant procedures performed with nearly 7500 surviving recipients (Miller, 1991). A current average of 1600 transplants are performed per year in the United States and 2500 worldwide. Survival rates have improved dramatically from 20% at one year in 1968, to a current success rate of nearly 90% at one year and 70% at five years (Miller, 1991), no doubt a result of improvements in surgical procedures and knowledge with respect to post transplantation care and management.

Despite ameliorations in survival rates following heart transplantation, major complications which continue to be reported from nearly a decade ago remain a barrier to long-term patient survival. An intense amount of work has been performed to enhance our understanding of the mechanisms involved in each of these complications and the approaches needed for effective post transplant care and management (Miller, 1991).

Lipoprotein abnormalities which develop after heart transplantation have been reported as a major complication. Accelerated coronary atherosclerosis, present in as many as 40% of patients within three years post heart transplantation is a leading cause of mortality as a result of heart graft failure (Uretsky et al, 1987). Hyperlipidemia and obesity have been identified as risk factors for the development of coronary heart disease (CHD) (Winterfeldt, 1988), in the normal population, and therefore could pose a threat to post transplant longevity. Other risk factors associated with the incidence of CHD
include genetic predisposition, smoking, male sex, hypertension, diabetes, stress, and lack of exercise.

Time-related increases in total cholesterol (TC), low-density lipoprotein cholesterol (LDL-C), total to high-density lipoprotein cholesterol ratio (TC:HDL-C ratio), triglycerides (TG), apolipoprotein B (apoB), as well as weight gain following heart transplantation have been described, but the mechanisms underlying these changes remain to be elucidated (Stamler et al, 1991). Immunosuppressive therapy such as the administration of prednisone, cyclosporine (CsA), or both are associated with changes in serum lipid and lipoprotein levels and have been implicated as playing a role in the development of atherogenic lipid profiles following cardiac transplantation (Stamler et al, 1991). Body weight, preoperative diagnosis of ischemic cardiomyopathy, immunologic injury, ages of recipient and donor, pre-transplant lipid and lipoprotein levels, diet, antihypertensive agents, as well as other risk factors associated with hyperlipidemia in the general population are all possible contributors to post transplant atherogenic dyslipoproteinemias (Keogh et al, 1988; Becker et al, 1988; Ballantyne et al, 1989).

The relationship between diet, plasma lipid levels, and the incidence of coronary heart disease (CHD), has been well documented in the general population (Winterfeldt, 1988). A regime reduced in total fat (particularly saturated fat), and dietary cholesterol, and aimed at achieving a healthy body weight have been reported both epidemiologically and clinically to favorably influence plasma lipoprotein concentrations (Crimmins et al, 1985) and reduce the incidence of CHD (Winterfeldt, 1988). The 10 year Coronary Primary Prevention Trial demonstrated that a reduction in total serum cholesterol and
LDL-C resulted in a reduction in CHD incidence in men at high risk for CHD because of hypercholesterolemia. Serum cholesterol has been demonstrated to be an important determinant of atherosclerosis (National Institutes of Health Consensus, 1985; Lipid Research Clinics, 1984; Grundy, 1986). Both the type and amount of dietary fat affect plasma lipoprotein levels and changes in intake are reflected in changes in both total serum cholesterol and triglyceride levels. Plasma cholesterol levels, particularly the LDL-C component, are related to dietary cholesterol intake (Winterfeldt, 1988).

Dietary modification represents the first step in the phased approach to the nutritional management of hyperlipidemia (Crimmins et al, 1985). The American Heart Association (AHA) developed a two phase dietary treatment program, to improve lipid and lipoprotein abnormalities prior to the administration of medication. The AHA Step One Diet allows up to 30% of total daily energy as fat, 50-60% as carbohydrate, and 10-20% as protein. Total fat intake is further divided into 7-10% of energy as saturated, 10-15% as monounsaturated, and 10% as polyunsaturated. Daily cholesterol intake is limited to less than 300 mg/day (Moore et al, 1990). The Expert Panel has provided specific guidelines for the dietary treatment of hyperlipidemia which include the Step One Diet (The Expert Panel, 1988).

We therefore postulated that application of the same guidelines would control hyperlipidemia in heart transplant recipients. Therefore, this research tested whether the Step One lipid lowering diet commenced soon after transplantation could prevent or minimize hyperlipidemia and excessive weight gain following heart transplantation. The ultimate goal of this intervention is to prolong patient survival and improve quality of
life, by influencing the frequent early appearance of coronary heart disease. In addition, this research was carried out to gain insight into the possible mechanisms of post transplant hyperlipidemia and weight gain, with a view to understanding their role in the development of atherosclerosis in the transplanted heart.

Immunosuppression

The purpose of immunosuppressive medications is to help prevent rejection of the donor heart (Funk, 1986). Rejection is the process by which the immune system attacks the transplanted heart, when it recognizes it as a foreign organ. Rejection can be classified in three ways:

1) Hyperacute rejection: this type occurs immediately after placement of the new heart, and results from preformed antibodies in the recipient that act against donor antigens. Retransplantation is a necessity.

2) Acute rejection: this is the most common type and occurs with the greatest frequency and severity in the first 6-8 weeks after transplantation. It results from interstitial and perivascular mononuclear infiltration, and will lead to myocardial cell necrosis if not treated.

3) Chronic rejection: this is considered to be caused by immune mediated injury to the coronary arteries eventually resulting in atherosclerotic disease. The most effective treatment is retransplantation (Funk, 1986).
The three immunosuppressive drugs most commonly used are prednisone, CsA, and azathioprine (AZA). Corticosteroids such as prednisone and methylprednisolone act by suppressing T and B lymphocytes and inhibiting mobility of macrophages, migration of leukocytes, and vascular dilation (Funk, 1986). Cyclosporine A selectively inhibits T-cell function (Funk, 1986), probably by interfering with interleukin 2 driven proliferation (Kimball, 1986). Because of its mode of action, which allows humoral immunity to remain intact and does not suppress bone-marrow function, CsA has been successful in reducing morbidity and mortality from infection post transplantation. It therefore has contributed to the improved success rates of cardiac transplantation (Funk, 1986).

Azathioprine inhibits purine and DNA synthesis, thus acting as an antimitotic agent (Funk, 1986; Kimball, 1986). Hence, cell division is inhibited, including those cells involved in the immune response, and the immune system is suppressed. Side effects include infection, fluid retention, increased appetite, nephrotoxicity, hypertension, hepatic dysfunction, bone-marrow suppression and liver toxicity (Funk, 1986).

Time-courses

Various time-courses for the development of hyperlipidemia after heart transplantation have been described (Table 1). Stamler et al (1988) obtained sera before and at frequent intervals after transplantation from 20 heart transplant recipients, as well as through a retrospective chart review of 25 transplant patients. All patients received CsA and
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Table 1: Summary table of selected literature published on lipids in heart transplant patients. Includes the reference, sample size, study design, immunosuppressants used in study, lipid changes, and contributing factors attributed by the authors to lipid changes. Pred = prednisone; CsA = cyclosporine A; AZA = azathioprine; TC = total cholesterol; LDL-C = low-density lipoprotein cholesterol; HDL-C = high-density lipoprotein cholesterol; TG = triglyceride; ApoB = apolipoprotein B; BW = body weight; CAD = coronary artery disease.
corticosteroids. Azathioprine was added if resistant rejection occurred. They observed significant elevations in LDL-C and TC occurring within two weeks post transplantation, peaking at three months and remaining above desirable levels throughout the first year. Total cholesterol and LDL-C increased by approximately 2.6 and 1.9 mmol/l at one year respectively. They concluded that there is a time-dependent increase in TC levels in patients treated with CsA, predominantly a result of significant increases in LDL-C. However, other possible contributing factors to the elevations in both TC and LDL-C were not accounted for such as diet, medications, and pretransplant TC and LDL-C levels.

Keogh et al (1988) studied two groups of heart transplant patients, group one (n=10 males) with underlying ischemic heart disease, and group two (n=9 males, 3 females) with underlying idiopathic or valvular cardiomyopathy. They observed an increase in mean serum cholesterol concentrations in both groups (1.5 and 1.8 mmol/l respectively) at one year post transplantation. Serum cholesterol levels peaked at approximately three months as reported by Stamler et al. At 12 months, there was a significant degree of hypercholesterolemia in both groups. They also noted a gradual increase in body weight. Forty percent of group 1 and 42% of group 2 exceeded a body mass index (BMI) of 25 kg/m² during the first year. Body weight increased by 8% in group 1 and by 12% in group 2. One of the strengths of this study was the comparison with preoperative body weights and serum cholesterol levels. However, pretransplantation data must be interpreted with caution. Prior to transplantation, TC concentrations may be abnormally low as a result of congestive heart failure, with changes in hepatic function, and G.I.
absorption even among patients with known premorbid hypercholesterolemia and ischemic cardiomyopathy (Becker et al, 1988). Hence, preoperative TC levels may not be a true reflection of "usual" concentrations. Preoperative body weight may not be a true indicator of usual body weight, as a result of congestive heart failure and fluid retention, or as a result of weight loss due to pretransplantation morbidity. Thus, the time at which pretransplantation data are collected is very important when comparing changes in TC and body weight to preoperative values.

Further investigations have demonstrated progressive rises in TC (approx. 1.3 mmol/l), LDL-C (approx. 1.7 mmol/l), and TG (approx. 0.45 mmol/l) levels during the first 16 months post transplantation (Becker et al, 1988). HDL-C concentrations also augmented by 31% during the first 4-6 months following surgery, but then were restored to early post transplant levels thereafter. A criticism of this study is that at each timepoint, measurements were taken on a different group of patients. Hence, a different group of patients was compared over time, rather than the same group of patients. The actual changes reported therefore, cannot be justified.

Another 34 patients who had undergone heart transplantation and two of whom heart-lung transplantation was performed were also studied at an average time of 25.8 ± 14.0 months postoperatively (Bilodeau et al, 1989). All were administered steroids, AZA, and CsA for immunosuppression. ApoB and LDL-C were commonly elevated. They suggested that steroids may have stimulated hepatic ApoB production.

A more detailed time-course was described in 54 patients who were analysed retrospectively from before heart transplantation up until three years after transplantation
Both cholesterol and triglycerides became abnormally high postoperatively. Total cholesterol increased from $4.5 \pm 1.4$ to $6.5 \pm 1.7$ mmol/l (mean ± SD) during the first year, but then decreased to preoperative levels at three years. The latter was postulated to be a result of lower steroid doses, or of improved compliance with dietary and exercise instruction. Mean serum triglyceride concentrations increased significantly from $1.6 \pm 1.1$ mmol/l prior to transplantation, to $2.4 \pm 1.3$ mmol/l at three years following transplantation. There was however, no significant difference between triglyceride levels at one, two and three years post transplantation. LDL-C and HDL-C values were obtained at one, two, and three years postoperatively. LDL-C was highest at one year, reaching a concentration of $3.6 \pm 1.4$ mmol/l. HDL-C levels were desirable, with an overall mean of $1.1 \pm 0.4$ mmol/l. As mentioned previously, the interpretation of changes in comparison to preoperative levels is difficult as most patients suffer congestive heart failure prior to transplantation. Although serum cholesterol became abnormally high after transplantation, some patients may have actually been hypercholesterolemic prior to transplantation (prior to the severe stage of illness), and hence the increase would have a different implication.

Lipoprotein (a) [Lp(a)], is a lipoprotein highly correlated with coronary atherosclerosis. The Lp(a) structure consists of one LDL molecule joined by a disulfide bond to an ApoA molecule. It is synthesized in the liver, and although similar to LDL-C with regards to structure, circulating levels of Lp(a) do not appear to be regulated under the same metabolic control as LDL-C (Farmer et al, 1991). Lipid values for Lp(a), TC, HDL-C, LDL-C, and TG were obtained six weeks before transplantation and compared
with those measured three months after transplantation in the only report to date to examine changes in Lp(a) after heart transplantation (Farmer et al, 1991) (Table 1). Lp(a) surprisingly decreased after cardiac transplantation from 11.7 ± 1.7 to 6.8 ± 1.1 mg/dl (mean ± SEM, p<0.001). This decrement was accompanied by dramatic increases in TC, TG, LDL-C, ApoA1, ApoB, and HDL-C. The authors suggested that metabolism of Lp(a) is independent of LDL-C, and that the use of CsA and prednisone may affect its synthesis or catabolism.

**Association with immunosuppressants**

Corticosteroids:

The increased lipid levels just described have been attributed to steroid use (Table 1). Multivariate analyses identified cumulative prednisone dose as the strongest, significant predictor of post transplantation elevations in TC, LDL-C, and HDL-C, independent of age, gender, time since transplantation, and cumulative CsA dose (Becker et al, 1988). Two further studies have supported the view that prednisone is a major predictor of post transplantation hypercholesterolemia (Taylor et al, 1989; Renland et al, 1989). Patients discontinuing or not taking corticosteroids demonstrated lower serum cholesterol levels after transplantation (Taylor et al, 1989; Renlund et al, 1989). In the investigation of Taylor et al, three groups of heart transplant recipients with different
immunosuppressive regimens were compared at one year following surgery. Group one received AZA and prednisone, Group two received CsA and prednisone, and Group three received CsA and AZA. Group two had a significantly higher TC concentration (6.87 ± 0.31 mmol/l) than Group three (5.76 ± 0.21 mmol/l) but not significantly higher than Group one (6.10 ± 0.31 mmol/l). There was no significant difference between Groups one and three with respect to serum cholesterol. Group three who were receiving CsA without prednisone had a significantly higher total/HDL-C ratio which may have been related to the absence of prednisone which is known to increase HDL-C. Problems with this investigation include no description of the actual doses of immunosuppression administered as well as no baseline data with which to compare the levels at one year.

Renlund et al (1989), tested the hypothesis that corticosteroids contribute to post transplant hypercholesterolemia by reviewing 117 consecutive heart transplant recipients, 51 of whom required maintenance corticosteroids (mean dose of 12.4 ± 1.2 mg/day), and 66 of whom did not, and who were maintained with CsA and AZA only, once corticosteroids were discontinued. From 3-18 months after heart transplantation, serum cholesterol levels were 21% to 26% lower in the corticosteroid-free group. However, possible confounders did exist. More than 90% of the patients in this study required antihypertensive medications which have been reported to be a potential contributor to post transplant elevations in serum cholesterol (Becker, 1988). The number of antihypertensive agents was less in the corticosteroid-free group at 6 and 12 months, and significantly less at 18 months after transplantation. Secondly, one patient in the corticosteroid free group was taking gemfibrozil, a lipid-lowering drug, and another in
the corticosteroid maintenance group was taking oral contraceptives which are known to raise serum cholesterol levels (Wynn, 1991). Hence other factors may have been contributing to the differences between the two groups.

Further substantiations of an association between prednisone therapy and TC elevations after transplantation come from the investigation of Livi et al (1989) (Table 1). They found a significant increase vs pretransplantation, in TC levels up to 21 months post transplantation, in 31 patients receiving CsA, AZA, and prednisone, and no significant difference in another group (n=28) receiving only CsA and AZA. Rudas et al (1990) demonstrated a positive association between total serum cholesterol and actual prednisone dose at one year, as well as with the cumulative one year steroid dose. The strength of this relationship is questionable since multiple regression analysis failed to demonstrate an association between these variables as discussed by Stamler et al (1991). Possible variables which were acting as confounders include diuretic therapy which was received by 39% of the study sample at one year after transplantation, lipid-lowering therapy (cholestyramine) which was initiated in one patient, and the effect of diet. Although all patients received detailed and individual instruction regarding the maintenance of a low cholesterol diet, the actual nutrient breakdown of the diet is not described, nor was dietary compliance measured to verify whether patients were adhering to these recommendations. Hence, the exclusion of diet as a contributing factor cannot be justified.

The relationship between prednisone treatment and lipoproteins is not limited to the heart transplant population. Although corticosteroid therapy is a primary treatment for
patients following cardiac transplantation, it is widely used for many diseases (Zimmerman et al, 1984). In a prospective study of the effect of prednisone therapy on plasma lipoproteins and apolipoproteins, 12 patients were treated for diseases that required the administration of corticosteroids (Zimmerman et al, 1984). Diagnoses included uveitis, sarcoidosis, endocrine exophthalmos, eosinophilic pneumonia, allergic reaction, hypereosinophilic syndrome, Bell’s palsy, idiopathic pericarditis, pemphigus vulgaris, and optic neuritis. Initial prednisone doses ranged from 30-80 mg/day depending on the diagnosis. These diseases are not known to have a direct effect on plasma lipid and lipoprotein levels, unless associated with marked nutritional changes, or in the case of exophthalmos, with hyperthyroidism. One of the patients had non-insulin dependent diabetes mellitus, which can have such an effect. Plasma cholesterol concentrations increased, mostly as a result of a dramatic increase in HDL-C, and a moderate rise in LDL-C. These changes were statistically significant after 48 hours of prednisone treatment. The increase in HDL-C was independent of prednisone dosage. No change in plasma apolipoproteins was observed. The authors suggest that the marked increase in HDL-C level without a concomitant increase in HDL apolipoproteins indicates an altered protein-lipid ratio for HDL. This suggests a change in HDL subfraction distribution, with an increase in HDL2, an HDL subfraction with a higher lipid-protein ratio than HDL3 (Zimmerman et al, 1984). The HDL2 subfraction has been shown to be the subfraction predominantly elevated following cardiac transplantation (Atger et al, 1990).

Importantly though, patients from the study above comprised a heterogeneous group in a good general condition, with no major metabolic abnormality. Other patient
populations receiving long-term corticosteroid therapy including patients with asthma, systemic lupus erythematosus, and other rheumatic conditions, have demonstrated elevations in TC, TG, and LDL-C when compared with sex- and age- matched control subjects. Patients with untreated Cushing’s syndrome have also shown similar lipid elevations (Becker et al, 1988).

As well, renal transplant patients have demonstrated a significant degree of hyperlipidemia associated with corticosteroid use (Stamler, 1991). A positive correlation between total steroid dose and serum TG has been observed (Ibels et al, 1978). Alternate-day steroid use has been suggested to minimize lipid elevations (Miller, 1991). In contrast to cardiac transplant recipients, the lipid fractions most commonly elevated following renal transplantation are triglycerides, and HDL-C (Miller, 1991). Various mechanisms have been proposed to explain the association between hyperlipidemia and prednisone: 1) impaired receptor uptake of normal LDL-C (Allegra, et al, 1989; Becker et al, 1988), 2) hyperinsulinemia caused by exogenous corticosteroids, stimulates hepatic synthesis of very-low-density lipoprotein cholesterol (VLDL-C) inducing augmentations of both TGs and LDL-C (Becker et al, 1988), 3) an impaired immune system due to rejection may have an effect on lipoprotein metabolism (Becker et al, 1988). It is plausible that this process is a response to the high doses of prednisone administered when episodes of rejection occur, suggesting that patients with more rejection episodes would have greater lipid elevations. Studies have also demonstrated increases in the hepatic production of HDL-C with corticosteroid use (Becker et al, 1988; Ballantyne, 1989).
Cyclosporine A:

Cyclosporine A has also been implicated in post transplantation hyperlipidemia. (Stamler et al, 1988; Superko et al, 1990) (Table 1). Stamler et al (1988) suggested that CsA induced hepatotoxicity impairs cellular clearance of LDL-C, and adversely affects TG levels through lower lipoprotein lipase activity (LPL). Work performed by Superko et al (1990), investigated LPL and hepatic lipase (HL) activity at one year after cardiac transplantation. Seventy-two cardiac transplant patients (62 men, 10 women) and 51 healthy male volunteers from a coffee-drinking and lipoprotein metabolism trial who were used as controls participated in the study. There were significant differences in TG (2.0 ± 1.5 mmol/l), LDL-C (4.2 ± 1.4 mmol/l), TC (6.3 ± 1.7 mmol/l), LPL activity (-0.4 ± 2.0 μmol/FFA/ml/hr), and HL activity (11.8 ± 3.5 μmol/FFA/ml/hr) between heart transplant recipients and healthy control subjects (1.1 ± 0.46 mmol/l, 3.6 ± 0.72 mmol/l, 5.4 ± 0.88 mmol/l, 1.7 ± 1.1 μmol/FFA/ml/hr, and 5.9 ± 1.9 μmol/FFA/ml/hr respectively). HDL-C concentrations were not significantly different between the two groups. In this study, LPL activity was lower and HL activity was higher within the transplant group, and both were correlated to TGs and LDL-C. LPL activity correlated negatively with TG levels in the control and treatment group, and HL levels approached a significant inverse correlation with LDL-C concentrations. Furthermore, CsA dose was the only immunosuppressant to correlate positively with HL activity, and inversely with LPL activity. These results suggest that CsA dose affects lipoprotein metabolism by contributing to changes in enzymatic activity and consequently
the lipid profile. Furthermore, evidence from both animal and human studies suggest that immunologically mediated arterial (endothelial) injury is aggravated by hypercholesterolemia (Stamler et al, 1991). The resulting endotheliopathy may further impair peripheral cholesterol metabolism due to alterations in endothelial-bound lipase activity (Stamler et al, 1991).

Both lipoprotein lipase and hepatic lipase are involved in lipoprotein metabolism. Hepatic lipase aids in processing VLDL lipoprotein particles to LDL particles and helps identify LDL characteristics (Kinnuren et al, 1983). Lipoprotein lipase facilitates the uptake of unesterified cholesterol, and the transfer of cholesterol ester from lipoproteins to cells (Eckel, 1989; Fielding, 1978). It is also involved in the breakdown of TG in the liver to free fatty acids and glycerol, which are then transported to tissues and resynthesized into TG (Stryer, 1988).

To further examine the effects of CsA on lipoprotein levels, a prospective, double-blind, randomized, placebo-controlled, two month study was conducted with 39 men diagnosed with amyotrophic lateral sclerosis, (a progressive, degenerative, neurological disease) treated with CsA (Ballantyne et al, 1989). By two weeks of CsA treatment, there was an 18% increase in total cholesterol from 5.22 ± 0.34 mmol/l to 6.21 ± 0.39 mmol/l, and no significant change was observed in the placebo group. At two months, a 21% rise in TC was still observed from baseline in the CsA group, and still no significant change with the placebo group. The increase in TC was primarily attributed to an increase in LDL-C of 0.91 ± 0.23 mmol/l. The placebo group showed a non-significant change of 0.10 ± 0.18 mmol/l. Apo B levels augmented by 11 ± 3
mg/dl in the CsA group compared to 3 ± 3 mg/dl in the placebo group (p<0.05). No significant changes in TG, HDL-C, or weight were noted in either group. Interestingly, no association was observed between dosage (6 mg/kg/day) or plasma levels of the drug and the degree of hyperlipidemia.

Similar results have been observed among renal transplant patients. After conversion from CsA to AZA following 4 months of CsA therapy, 13 renal transplant recipients had a 19% fall in TC (7.9 ± 0.4 to 6.5 ± 0.4 mmol/l) (Harris et al, 1986). The same magnitude of decrease in TC was reported in 23 cadaveric renal transplant patients in whom CsA was converted to AZA one year post transplantation (Versluis et al, 1987). A subsequent decline in both TG and TC was found when CsA was converted to AZA in another study involving renal transplant recipients (Markwell et al, 1989). Cyclosporine A administration was significantly associated with hypertriglyceridemia in bone marrow transplant recipients as well (Carreras et al, 1989). It has been suggested that because CsA is transported bound to cholesterol-rich lipoproteins, and because of its inherent hydrophobicity, which makes it soluble in lipid membranes, it could alter membrane and receptor-mediated clearance of LDL-C (Stamler et al, 1991). CsA also has a major binding affinity to HDL which could possibly alter its metabolism (Mraz et al, 1986).

It thus remains uncertain what the relative roles of prednisone and CsA are in the causation of the plasma lipid changes following transplantation. In a cohort study of 566 patients who had undergone renal transplantation, there was an equal incidence of hypercholesterolemia in CsA-Prednisone (Pred) vs AZA-Pred treated patients (Vathsala
et al., 1989). Several of their observations can be interpreted to suggest that CsA therapy per se contributes less to post transplantation hypercholesterolemia. First, there was a strong correlation between prednisone doses and serum cholesterol concentrations in both the CsA-Pred and AZA-Pred groups as well as an improvement in cholesterol levels in 10 patients who discontinued the use of prednisone. Second, there was an inverse correlation between CsA dose and cholesterol levels. Third, mean CsA doses were significantly lower in hyperlipidemic than nonhyperlipidemic CsA-Pred treated patients until two years post transplantation, whereas prednisone doses were significantly higher in hypercholesterolemic than nonhypercholesterolemic patients at 6 and 36 months post transplantation. These results suggest that prednisone rather than CsA is the primary cause of post transplantation hypercholesterolemia, at least in this patient population. The data also showed that CsA-Pred therapy produces an elevation in the LDL-C fraction rather than the HDL-C fraction as reported for AZA-Pred patients. This is consistent with the proposed mechanism that CsA affects LDL clearance resulting in elevated LDL-C concentrations.

Controlled randomized trials comparing the various immunosuppressant regimens would be difficult to justify with lipid endpoints being primary, as the priority must remain on optimizing graft acceptance and function. Likewise, all heart transplant recipients receive a form of combined immunosuppressant therapy that currently includes both CsA and prednisone. The types and doses of drugs administered are furthermore dependent on the patient's response to transplantation, making randomization with different immunosuppressants virtually impossible, and indeed unethical. This accounts
for the absence of controlled randomized trials reported to date.

Other study design variables render interpretation of lipid outcomes, and especially determination of mechanisms, uncertain and complex. These include differences in sample sizes, time intervals at which data were collected, duration of data collection after surgery, type of data collection (retrospective vs prospective), differences in follow-up treatment procedures among centres, the various immunosuppressant regimens administered and the time at which they were initiated, as well as the characteristics of the population being studied (i.e. age, etiology of heart failure, existence of pretransplantation hyperlipidemia and obesity, which are difficult to interpret because of preoperative cachexia, the use of antihypertensive medication, various dietary patterns etc.). The lack of control of variables such as exercise, diet, and weight gain can also bias results (Miller, 1991).

The lipid elevations that follow cardiac transplantation clearly have a multifactorial causation, and it appears justified to implicate steroids in combination with CsA as primary. The literature has provided data and potential mechanisms implicating both steroids and CsA as strong contributing factors, but it is difficult to assign priority to one or the other. In my view, the strongest lines of evidence are the significant improvements in serum cholesterol levels with prednisone withdrawal, and the correlations between steroid doses and lipid levels. The absence of a correlation between CsA doses and lipid levels can be interpreted to suggest that CsA is not an independent causal factor. However, a threshold effect, or a possible dose-response at lower than therapeutic doses of CsA cannot be excluded. Furthermore, Ost (1984) suggested that CsA may potentiate
the effects of prednisone therapy by reducing the clearance of the drug, suggesting that
the combination of both prednisone and CsA administration may have the greatest effects
on lipoprotein elevations.

Association with body weight

Significant elevations in body weight have been observed after transplantation (Keogh et al, 1988) (Table 1). Patients with pretransplant diagnoses of both underlying ischemic heart disease (IHD), and underlying idiopathic dilated or valvular cardiomyopathy (CM) showed a steady and progressive increase in body weight throughout the first year after transplantation. Patients with IHD showed an increase from 68.0 ± 9.2 kg preoperatively to 73.1 ± 8.7 kg (n=10, mean ± SD). Patients with CM showed an increase of 7.1 ± 1.7 kg (n=12, mean ± SD), from preoperative to one year postoperative. The authors do not report whether these changes are significant. However, it is self-evident that these patients were seriously ill preoperatively. Therefore body weights could have been lower than usual, premorbid levels, or higher as a result of congestive heart failure, and body composition likewise altered, making the comparison to preoperative levels difficult to interpret.

In another study by Grady et al (1991), patient weight increased significantly from 89% of usual body weight and 100% of ideal body weight preoperatively to 106% of usual body weight and 117% of ideal body weight one year postoperatively. Body weight was not significantly lower at three years after surgery. Actual body weights are not
provided, and the changes in body weight are reported according to changes in %ideal body weight, and %usual body weight. Preillness usual body weight was determined through patient inquiry. Ideal body weights were determined using the following formula: men: 106 lbs for the first 5 feet, then 6 lbs for each additional inch; women: 100 lbs for the first 5 feet, then 5 lbs for each additional inch.

The authors reported that patients were obese prior to cardiac illness. A criticism of this method of comparing body weights, is that ideal body weight is no longer used as a reference for desirable, healthy body weights. Desirable body weights determined from reference tables according to patients' height, age, and sex, or BMI would have provided a better estimation of patients' weight status both prior to and following surgery. Hence, the designation of premorbid "obesity" is not justified. These subjects not only regained but also exceeded their usual body weight. Most of the increase in body weight however (11 %), was a return to preillness levels. These changes in body weight were significant.

To our knowledge, the evolution of body composition following heart transplantation has not been assessed in a detailed manner in the literature. The increase in body weight may very well be a result of an increase in body fat, or may be a normal post surgical response occurring as a result of 1) increased appetite both relative to preoperative state, and on a absolute scale, 2) improved GI absorption with reversal of pretransplant cachexia (Keogh et al, 1988), as well as 3) a return to preillness dietary habits (Grady et al, 1991). Side effects of corticosteroids include fluid retention and increased appetite which may also contribute to weight gain (Funk, 1886). Finally, CsA therapy may potentiate the effect of prednisone therapy by reducing the clearance of the drug (Ost,
1984), and hence influence weight by exacerbating its usual effects.

It is plausible that the elevations in lipoproteins following transplantation are related to the weight gained post transplantation. Many population studies have reported the relationship between body weight, body fat, and the distribution of body fat and serum lipid and lipoprotein levels (Albrink et al, 1980; Avogaro et al, 1978; Freedman et al, 1985; Wattigney et al, 1991; Weinsier et al, 1976; Freedman et al, 1989; Haffner et al, 1987; Coon et al, 1989; Micozzi et al, 1989; Glueck et al, 1980). Models developed from the Framingham study showed that for each 10% increase in weight in 35-54 year old men, a 30% increase in coronary heart disease incidence would be expected due to simultaneous changes in levels of TC and systolic blood pressure (Freedman et al, 1985).

Weight changes in males aged 24-48 years has been positively associated with LDL-C and VLDL-C levels (Harlan et al, 1987). Longitudinal changes in weight and triglycerides in men have also been positively associated (Larsson, 1978). Weight gain has also been related to elevations in TC and TG in middle-aged females (Noppa, 1980). During a 14 year period, the Framingham study reported that changes in body weight were associated with changes in TC levels (Ashley and Kannel, 1974). Weight gain between the ages of 18 and 30-55 years has also been demonstrated to be related to TG and LDL-C levels and inversely related to changes in HDL-C levels (Khoury et al, 1983).

Measurements of body fat such as changes in triceps-skinfold-thickness (TRSF) and estimated percent body fat have also been correlated with LDL-C and TC, and could be used as predictors for follow-up serum lipid and lipoprotein levels (Freedman et al, 1985). Percent body fat showed evidence of being an important determinant of lipoprotein
profiles in healthy, nonsmoking males aged 46-73 years (Coon et al, 1989), and was strongly associated with TC concentrations in U.S. men and women in the First National Health and Nutrition Examination Survey (NHANESI) (Micozzi et al, 1989).

Many studies support the view that central adiposity (storage of fat located predominantly in the abdominal region) rather than peripheral adiposity (storage of fat located at the periphery i.e. limbs, arms, skin) is associated with more adverse lipid and lipoprotein profiles (Albrink et al, 1980; Wattigney et al, 1991; Freedman, 1989; Haffner et al, 1987; Coon et al, 1989; Micozzi et al, 1989). The Bogalusa study on children and young adults reported on the prediction of lipids and lipoproteins using subscapular skinfold thickness (Wattigney et al, 1991). Predicted levels of LDL-C and VLDL-C increased, whereas predicted levels of HDL-C decreased with increasing levels of subscapular skinfold measurements. In another report, which compared the ability of the ratio of subscapular to triceps skinfold (TSF) and the waist:hip ratio (WHR) to predict lipids and lipoproteins in adults aged 25-64 years, both indices were predictive of low HDL-C and high TG levels, though, the WHR was a better predictor (Haffner et al, 1987). Truncal fat was associated with decreased concentrations of HDL-C and APOA1 in children aged 6-18 years. Children with higher levels of VLDL-C and LDL-C had more truncal fat and less peripheral fat independently of the overall level of obesity (Wattigney et al, 1991).

The studies described above all used anthropometric measurements to measure the distribution of body fat. However, intra-abdominal fat has also been suggested to be an important determinant of lipoprotein abnormalities (Després et al, 1990). Subcutaneous
fat as well as visceral fat can be measured with considerable accuracy by using either computed tomography (CT) or magnetic resonance imaging (MRI) (Bouchard et al., 1990). Increased subcutaneous truncal-abdominal fat is associated with increased insulin resistance and a decrease in HDL-C. High levels of visceral fat enhances insulin resistance, and further decreases HDL-C (Bouchard et al., 1990). Després et al (1990), studied the associations between body fat distribution and plasma lipid levels in a sample of 52 premenopausal obese women. Computed tomography was used to measure the amount of intra-abdominal fat. They demonstrated a high proportion of intra-abdominal fat to be associated with a reduction in HDL-C concentration, and with significant reductions in lipoprotein ratios (HDL-C/LDL-C; ApoA1/ApoB; HDL2-C/HDL3-C). The latter associations were independent from the effect of obesity. Abdominal obesity has also been shown to be associated with high plasma TG concentrations (Kissebah et al, 1982; Krotkiewski et al, 1983; Evans et al, 1984).

It has been suggested previously that 81% of the variation in serum cholesterol and 72% of the variation in TG levels following heart transplantation could be explained by body weight (Keogh et al, 1988), suggesting that weight gain is a contributing factor to post transplantation hyperlipidemia. Transplant patients who were overweight (BMI > 25 kg/m²) at one year postoperatively, demonstrated significantly higher cholesterol and triglyceride levels than those who were not overweight (Rudas et al, 1990). In contrast to these findings, no correlation was found between postoperative body weight and serum lipid levels in the Grady report (Grady et al, 1991). Thus, the composition of weight gain post transplantation, and where any excess fat is deposited, and their relationship to
lipoprotein abnormalities requires further investigation.

**Association with pre-transplant ischemic heart disease**

Cardiac diagnosis prior to surgery may influence susceptibility to hyperlipidemia following transplantation. Correlations have been demonstrated between a preoperative diagnosis of coronary artery disease (CAD) and post transplant lipid abnormalities (Taylor et al, 1989; Keogh et al, 1988; Rudas et al, 1990; Grady et al, 1991). In the Rudas series, patients diagnosed with CAD prior to transplantation had a significantly higher pretransplant TC level than patients with a diagnosis other than CAD (Rudas et al, 1990), although levels for both groups were within the normal range. Thus, greater serum lipid abnormalities following heart transplantation in patients with a preoperative diagnosis of CAD, may be influenced by higher pretransplant serum lipid concentrations. Importantly however, the pattern of increase in serum cholesterol in patients with and without CAD is similar, implicating other external and common factors such as weight gain and immunosuppressive drugs (Keogh et al, 1988).

**Association with diet**

Diet has demonstrable effects on both body weight and serum cholesterol. Although cardiac transplant recipients are usually requested to follow a low fat, low cholesterol, no added salt diet following transplantation, to our knowledge no study has addressed the
effects of such a dietary intervention on hyperlipidemia and weight gain following heart transplantation, using a randomized, controlled, clinical intervention trial. Significant improvements in serum cholesterol, body weight, and triglycerides following a lipid-lowering diet have been noted with renal transplant patients (Nelson et al, 1988; Moore et al, 1990). Nine renal transplant recipients who were hospitalized for a three-week period, were given a calculated diet providing $7259 \pm 79$ KJ (mean for three days $\pm$ SEM), consisting of 21% protein, 50% carbohydrate (CHO), 29% total fat, (7% saturated, 8% polyunsaturated-PUFA), less than 300mg cholesterol/day, a P/S ratio $> 1$, and dietary fiber intake of 29g/1000 kcal (Nelson et al, 1988). Patients' usual diet provided $9171 \pm 590$ KJ (mean for six days $\pm$ SEM), consisting of 18% protein, 52% CHO, 30% total fat (11% saturated, 4% PUFA), greater than 400mg cholesterol/day, a P/S ratio $< 1$, and a lower fiber intake. After two weeks of following the diet, mean body weight significantly decreased from $79 \pm 4$ kg to $76 \pm 4$ kg, and mean body mass index (BMI) was significantly reduced from $31 \pm 2$ kg/m$^2$ to $30 \pm 2$ kg/m$^2$. The abnormal lipid profiles observed before the initiation of the diet were significantly ameliorated after two weeks of the diet. Total cholesterol concentrations fell from $8.2 \pm 0.93$ mmol/l to $6.9 \pm 0.78$ mmol/l. Triglyceride levels improved from $4.8 \pm 1.1$ mmol/l to $3.3 \pm 0.68$ mmol/l. No significant change in apolipoprotein levels was shown. A significant correlation was found between the quantity of dietary cholesterol consumed and serum cholesterol and serum TG before administration of the experimental diet. A significant correlation between the degree of reduction of dietary cholesterol and the degree of lowering serum lipids was also found. No correlation between changes in serum
lipids and changes in energy, total fat, saturated fat, P/S ratio, or dietary fiber was reported indicating dietary cholesterol to be the most influential factor in improving serum lipid levels. The absence of a correlation between changes in total fat and changes in serum lipid levels may be related to the fact that although total fat intake prior to the experimental diet was significantly different from total fat intake during the experimental diet, usual fat intake was not unreasonably high. The absence of a correlation with saturated fat coupled with a correlation with dietary cholesterol is ironic since higher serum cholesterol concentrations and dietary cholesterol intakes are usually a reflection of high saturated fat intakes (Winterfeldt, 1988). Perhaps the amount of days for which dietary data were collected (i.e. six day food diary for usual intake, and three day food diary during the experimental diet) was not sufficient to provide adequate estimations of saturated fat intake (Sempos et al, 1985). Furthermore it’s been shown that food records of at least nine days are required to adequately estimate dietary cholesterol intakes (White et al, 1981).

In another dietary intervention study, 12 out of 17 renal transplant patients who followed the Step One lipid-lowering diet for a period of eight weeks, lost weight (mean weight loss of 0.9 kg) and had a significant reduction in TC (6.8 to 6.2 mmol/l). (Moore et al, 1990). There was no control group. Becker et al (1988) reported no correlation between dietary fat and cholesterol intakes and concentrations of TC, LDL-C, or triglycerides in their retrospective analysis of 92 heart transplant patients. However, no information pertaining to the type of diet followed by these patients, nor to their actual total fat and dietary cholesterol intakes were provided.
Nutritional programs specific for heart transplant recipients have reported improved outcomes with dietary intervention (Ragsdale, 1987). Although an actual study was not performed, changes in dietary recommendations resulted in less obesity, and less death due to atherosclerosis. Although other factors may be responsible for the improvements seen i.e. changes in immunosuppressive therapies, incidence of hyperlipidemia and obesity prior to surgery, preoperative cardiac diagnosis, and exercise programs, this report describes the possible benefit of a nutritional program in reducing post transplantation weight gain and increasing patient survival by reducing the incidence of graft atherosclerosis.
PURPOSE OF RESEARCH

The literature has generally described a potentially atherogenic effect of heart transplantation on serum lipid levels, and has examined some possible contributors associated with this undesirable effect. Although associations between hyperlipidemia after cardiac transplantation and prednisone, cyclosporine, body weight, pre-transplant cardiac diagnosis, and diet have been observed, researchers in the field agree that more research is required, as none of these contributors have been fully defined as to their role in causation, and the associations remain controversial. Different time-courses have been observed, probably as a result of the different time intervals (i.e. weekly, monthly, or yearly measurements) being studied.

To our knowledge, a randomized, controlled dietary intervention study to assess the role of diet on hyperlipidemia post heart transplantation has not been conducted. This Masters thesis was therefore divided into two parts. The first was a retrospective chart review of 35 heart transplant patients, to define as many potential variables as possible involved with hypercholesterolemia post transplantation in our population, in comparison to the literature, and to provide us with a base for conducting our intervention study. Our intent was to describe a more detailed time-course of serum cholesterol changes post heart transplantation than in the literature, and to further identify factors associated with these changes, specifically body weight, doses of medication, and pre-transplant cardiac diagnosis. The effect of diet on the reversal of hypercholesterolemia and weight gain was also reviewed retrospectively in patients who had been referred to a dietitian. Changes
in body weight and serum lipid levels following counselling by a dietitian were recorded. This was not an intervention trial. The results of this study were then used as a prelude to the second part of our proposal: the dietary intervention study.

Our primary hypothesis of the intervention study is that the Step One Lipid-Lowering diet, with an energy intake appropriate to achieve a healthy body weight, commenced immediately after transplantation, would minimize or prevent elevations in lipids, lipoproteins, and body weight which occur after transplantation.
CHAPTER 2

TIME-COURSE OF HYPERCHOLESTEROLEMIA POST HEART TRANSPLANTATION
MATERIALS AND METHODS

Patient Population

The medical charts of 35 consecutive heart transplant recipients (32 males, 3 females) in whom the surgical procedure was successful enough to allow a return home, were reviewed retrospectively. Patient demographics are illustrated in Table 2. The transplantations were performed between July 1982 and May 1987 at the Royal Victoria and Notre-Dame Hospitals in Montreal, Quebec. All patients received prednisone, CsA, AZA, and antithymocyte globulin for immunosuppression. Methylprednisolone was initiated at a dose of 500 mg/day i.v., then reduced to 100 mg/day i.v., one day after surgery to three days postoperatively. Prednisone was then initiated at 30 mg/day and tapered according to each individual's tolerance. Cyclosporine A was started when hemodynamic parameters were stable, around two days postoperatively, at a dose of 100 mg/day, then increased such that CsA levels were 350 ng/ml. It was then adjusted according to creatinine levels and results of heart biopsy. Azathioprine started at 1.5 mg/kg body weight i.v., then by mouth as soon as tolerated usually by four days postoperatively, then adjusted according to white blood cell count. Maintenance dose was 1-1.2 mg/kg body weight. Antithymocyte globulin was initiated at 2.5 mg/kg body weight, then reduced to 1.5 mg/kg body weight, and given after administration of prednisone. Doses would vary depending on patients' response to transplantation and incidence and severity of rejection. Rejection was treated with 500 mg
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Table 2: Patient Demographics. F = female; M = male; BMI = Body Mass Index; IHD = Ischemic Heart Disease; CHF = Congestive cardiomyopathy; IDIO. = Idiopathic cardiomyopathy; Hyper = Hypertrophic cardiomyopathy.
methylprednisolone i.v., and optimal use of CsA, AZA, and prednisone on day one. Methylprednisolone was then reduced to 250-500 mg i.v. on days two and three. Prednisone was continued at 25 mg/day on day 4, and the biopsy was repeated.

**Data Collection**

Information was gathered from hospital records of patients before (if available) and after surgery. Data were obtained at monthly intervals up until three years after transplantation, and included patients’ age, body weight, etiology of heart failure resulting in need for transplantation, lipid profile (TC, HDL-C, LDL-C, VLDL-C, TG, and APOB when available), and medications. All data were entered into files using the Dbase III computer program.

The body weight and TC level presented in our data at the time closest to the date of surgery were used as the weight and cholesterol value at the time of transplantation (time 0). There were limited data available prior to transplantation with regards to body weight and serum cholesterol levels. Only a small number of patients had pretransplantation data available in their medical charts. Thirteen of the 35 patients had body weight data at an average time of 100 days before surgery. Eleven out of the 35 patients had pretransplantation TC levels indicated at 100 days in their chart. An average time of 100 days was used, as this was the timepoint at which the majority of patients had data available. Monthly complete lipid profile data were not available for each patient, however, complete lipid profile data were collected at 12, 18, and 24 months post
transplantation for some patients.

Lipid Determinations

All lipid and lipoprotein concentrations were determined after an overnight fast, with the Technicon RA-1000 Analyser system by the technical staff at the Biochemistry Laboratory of the Royal Victoria Hospital.

Total Cholesterol:

Total cholesterol was determined using the cholesterol esterase (CE) / cholesterol oxidase (CO) / peroxidase method. The CE hydrolysed cholesterol esters to free cholesterol and fatty acids. The free cholesterol was then oxidized to cholesten-3-one and hydrogen peroxide by CO. Peroxidase catalysed the reaction of hydrogen peroxide with 4- aminoantipyrine (4-AAP) and phenol which produced a colored quinoneimine product. The Technicon Analyser then proportioned the appropriate sample and reagent volumes. The system monitored the change in absorbance at 520 nanometers at a fixed-time interval. This change in absorbance was directly proportional to the concentration of cholesterol in the sample and was used by the system to calculate the cholesterol concentration.

HDL-C:

High density lipoprotein cholesterol was first separated from serum LDL and VLDL
cholesterol by precipitation with phosphotungstate and magnesium. The LDL and VLDL portions were then removed by centrifugation. The cholesterol in the HDL fraction which remained in the supernatant was assayed with an enzymatic cholesterol reagent, as described above for total cholesterol.

Triglycerides:

Triglycerides (TG) in the sample were hydrolysed to glycerol and free fatty acids by the action of triacylglycerol acyl-hydrolase EC 3.1.1.3. Three enzymatic reactions involving glycerol kinase (GK), glycerol-1-phosphate-dehydrogenase (G1PDH), and diaphorase reduced the end product 2-(p-iodophenyl)-3-p-nitrophenyl-5-phenyltetrazolium chloride (INT) to a colored formazan product. The change in absorbance was used to calculate and express the triglyceride concentration.

LDL-C:

Low density lipoprotein cholesterol was calculated using the Friedewald formula: 

\[
\text{LDL-C (mmol/l)} = \frac{\text{Total cholesterol} - \frac{\text{Serum Triglyceride}}{2.2} - \text{HDL-C}}{2.2}
\]

Note that this method will not accurately estimate plasma LDL-C levels when the TG concentration exceeds 4.5 mmol/l (Friedewald et al, 1972). None of the patients participating in this study for which LDL-C was determined, had TG levels exceeding this level.
ApoB:

Apolipoprotein B was measured by Radial Immuno Diffusion (Sniderman et al, 1975) by the technical staff at the Endocrinology Laboratory at the Royal Victoria Hospital.

Effect of Weight

To determine an association between weight gain and serum cholesterol elevations after heart transplantation, patients were divided into two groups according to their maximum weight gain after transplantation. The names of all patients in the study, together with their maximum weight gain after transplantation were listed in descending order according to their magnitude of weight gain. The mean of the first 18 patients was determined to give Group A’s mean maximum weight gain (9 kg), and the mean of the next 17 patients was determined to give Group B’s mean maximum weight gain (4 kg). The cut-off point was 5 kg. Serum cholesterol levels were compared between groups.

Effect of diet

Data were collected for 17 of the 35 patients who had been referred to the Lipid Clinic of the Royal Victoria Hospital, and who had seen a dietitian for hypercholesterolemia. Body weight and serum cholesterol data were entered into dbase files for this subset, and analysed from one year prior to one year post the initial lipid
clinic visit, to determine the effect of diet on post transplantation serum cholesterol elevations and weight gain. Furthermore, this subgroup was also divided into two groups according to their weight response to the diet, to further evaluate an association between body weight and serum cholesterol. Group A \( (n=8) \) lost 6 kg, and group B \( (n=9) \) lost 2 kg following dietary instruction. The groups were divided according to the same method described above. Changes in serum cholesterol levels were compared between groups.

**Effect of prednisone**

Nine patients discontinued the use of prednisone. A separate database file was created for this group. Monthly changes in body weight and serum cholesterol were analysed from one year prior to one year after prednisone discontinuation. Prednisone dose was slowly tapered from 15 mg/day to 2.5 mg/day, and then was completely withdrawn from the immunosuppressive regimen.

**Effect of pre-transplantation cardiac diagnosis**

Seventeen patients were diagnosed with ischemic heart disease before surgery, eight with congestive cardiomyopathy, seven with idiopathic cardiomyopathy, one with post-infection cardiomyopathy, one with hypertrophic cardiomyopathy, and one with sarcoid cardiomyopathy. These patients were classified as patients with non-ischemic...
cardiomyopathy. Changes in body weight and serum cholesterol levels were analysed between groups.

**Statistical Analyses**

Changes in body weight and serum cholesterol over time, and comparisons of groups were analysed using repeated measures analysis of variance. Paired t-test was used to compare differences between pre and post transplant data using the Primer of Biostatistics software package. Multivariate analysis was performed to determine the variables associated with the change in serum cholesterol at six months post transplantation. The six month timepoint was used as data were available for the majority of patients at this timepoint. The forward selection and stepwise procedures were used. All repeated measures analyses and multivariate analysis were performed using the SAS/STAT software (SAS Institute Inc., Cary, NC, USA) on a Hewlett-Packard Vectra computer. Results were considered significant at $p < .05$. Points presented at each time interval represent the mean monthly value ± standard error, and were determined using SAS. Changes in cholesterol levels and body weight were also analysed with respect to three different time periods: 1) the time of transplantation, 2) the time of prednisone discontinuation (for those patients who discontinued prednisone), and 3) the time of initial lipid clinic visit (for those patients referred to the lipid clinic). The data were expressed as the change from that particular reference timepoint (the actual time at which the above occurred [time 0]).
Body weight and serum cholesterol data were not always available at each monthly time interval for each patient up until three years after transplantation due to the lack of such information in the medical records. When possible, we calculated the mean between available data points to estimate a value for the missing data. Analyses were then performed on subsets of patients for which there were no missing data, and for those for which data were interpolated. Mean values at each timepoint therefore represent the same group of patients.
RESULTS

**Time-course of body weight and serum cholesterol changes after transplantation**

There were 15 patients with complete data on body weight, and 16 patients with complete data on serum cholesterol concentrations. During the first 30 days following transplantation, patients (n = 15) lost an average of 1.5 ± 1.7 kg (mean ± SEM). This was followed by a progressive, significant increase in body weight of approximately 8.4 ± 1.6 kg from time 0 during the next 10 months (p < 0.01)(Fig.1). Body weight stabilized thereafter, and did not return to pretransplantation levels (Table 3).

A progressive rise in serum cholesterol (2.4 ± 0.4 mmol/l) (p < 0.01) was also observed during the first 8 months (n = 16), then levels stabilized up until 2.5 years following surgery, remaining above normal levels (Fig.2). A normal serum cholesterol level was defined as < 5.2 mmol/l. The mean serum cholesterol level prior to transplantation for 11 patients for whom available data existed at 100 days before surgery was 4.9 ± 0.3 mmol/l, and hence was within the normal range (Table 3). Complete lipid profile data were available for some patients at 12, 18, and 24 months following transplantation (Table 4). Mean TC, LDL-C, and TG concentrations were all above desirable levels defined as 5.2, 3.4, (The Expert Panel, 1988) and 2.3 mmol/l (Canadian Consensus Conference, 1988) respectively. Out of these patients (Table 4), 71-86% had TC and 50-62% had LDL-C levels above the 75th percentile for age and sex at all three month time periods. HDL-C levels were within desirable limits, although at 12 and 18
Figure 1: Changes in body weight from time of transplantation (time 0) during the first 2.5 years. Each point represents the mean change from time 0 ± SEM, (n=15).

Figure 2: Changes in serum cholesterol concentration from time of transplantation (time 0) during the first 2.5 years. Each point represents mean change from time 0 ± SEM, (n = 16).
### Table 3: Changes in total cholesterol (TC, mmol/l), body weight (BW, kg), and prednisone dose (pred, mg/day) over time in patients with available pre-transplant (pretr) data. Each point represents the mean absolute value, or mean change from time of transplantation (time 0) ± SEM. Data shown at 100 days pre-transplantion (100d pretr), time of transplantation, and at 90, 180, 360, and 720 days post transplantation (posttr). Var. = variable, n=11 for TC and pred, n=13 for BW.

<table>
<thead>
<tr>
<th>Var.</th>
<th>100d pretr</th>
<th>time 0</th>
<th>90d posttr</th>
<th>180d posttr</th>
<th>360d posttr</th>
<th>720d posttr</th>
</tr>
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<tr>
<td>TC</td>
<td>4.9 ± 0.3</td>
<td>4.6 ± 0.3</td>
<td>6.2 ± 0.4</td>
<td>6.6 ± 0.3</td>
<td>6.6 ± 0.3</td>
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<tr>
<td>Δ TC</td>
<td>0.3 ± 0.3</td>
<td>0</td>
<td>1.6 ± 0.5</td>
<td>1.9 ± 0.4</td>
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<tr>
<td>BW</td>
<td>71.2 ± 4.3</td>
<td>72.3 ± 4.5</td>
<td>75.7 ± 4.1</td>
<td>79.9 ± 4.3</td>
<td>81.7 ± 4.6</td>
<td>83.5 ± 4.8</td>
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<td>Δ BW</td>
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<td>0</td>
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<td>7.6 ± 1.2</td>
<td>9.4 ± 1.4</td>
<td>9.6 ± 1.6</td>
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<td>0</td>
<td>20.6 ± 2.8</td>
<td>13.3 ± 0.6</td>
<td>11.0 ± 0.6</td>
<td>8.4 ± 0.4</td>
</tr>
<tr>
<td>VARIABLE</td>
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<td>18 MONTHS</td>
<td>24 MONTHS</td>
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<tr>
<td>TC</td>
<td>6.5 ± 0.5</td>
<td>6.7 ± 0.4</td>
<td>6.6 ± 0.3</td>
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<td>(12)</td>
<td>(10)</td>
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<tr>
<td>HDL-C</td>
<td>1.1 ± 0.1</td>
<td>1.3 ± 0.1</td>
<td>1.2 ± 0.1</td>
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<td></td>
<td>(8)</td>
<td>(12)</td>
<td>(10)</td>
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<tr>
<td>LDL-C</td>
<td>4.2 ± 0.4</td>
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<td>(8)</td>
<td>(12)</td>
<td>(10)</td>
<td></td>
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<tr>
<td>TG</td>
<td>2.5 ± 0.4</td>
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<td>(8)</td>
<td>(12)</td>
<td>(10)</td>
<td></td>
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<tr>
<td>APOB</td>
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</table>

Table 4: Complete lipid profile data of some patients after transplantation, n is indicated in parentheses. Each value represents mean ± SEM. TC = total cholesterol; HDL-C = high-density lipoprotein cholesterol; LDL-C = low-density lipoprotein cholesterol; TG = triglyceride; APOB = apolipoprotein B.
months, 71% and 57% were still below the 50th percentile respectively. At 24 months, 50% of patients were above the 50th percentile which is desirable.

**Effect of weight**

A negative association was observed between weight gain and elevations in serum cholesterol. Those patients who gained significantly more weight following transplantation (Group A, n=8, 15.2 ± 1.8 kg at 930 days, p < 0.01) had significantly lower increases in serum cholesterol levels (p < 0.05) than those patients who gained less weight (Group B, n=7, 4.2 ± 1.0 kg at 930 days) (Figs.3 and 4). Repeated measures analysis of variance demonstrated a significant change over time in serum cholesterol in both groups (p < 0.01). No significant difference in prednisone dose was observed (Fig.5).

**Effect of Diet**

There were nine patients seen by a dietitian at the lipid clinic for whom data were available. There was an average time of 1.8 years (range: 1-3 years) after transplantation, at which patients were seen at the clinic. Prior to the initial lipid clinic visit, patients gained weight (4.6 ± 0.7 kg), (p < 0.01) (Fig.6). During the first 6.5 months following dietary instruction, patients lost weight (2.6 ± 1.1 kg), (p < 0.01) which was then followed by a weight gain of 0.9 ± 0.1 kg at one year. There was a significant increase over time in serum cholesterol prior to dietary instruction (p < 0.01), however, levels
Figure 3: Changes in body weight from time of transplantation (time 0) during the first 2.5 years in 2 groups divided according to their mean maximum weight gain postoperatively (Group A in closed squares, n=8 and Group B in open circles, n=7). Each point represents the mean change from time 0 ± SEM.

Figure 4: Changes in serum cholesterol concentration from time of transplantation (time 0) during the first 2.5 years in the same 2 groups as in Figure 3. Each point represents the mean change from time 0 ± SEM.
Figure 5: Time-course of prednisone dose from time of transplantation (time 0) during the first 2 years, for group A (closed squares, n=8) and for group B (open circles, n=7). Each point represents the mean prednisone dose ± SEM.
DAYS OF FOLLOW-UP VISITS (DAY 0 = FIRST VISIT)

Figure 6: Weight response of 9 patients who received dietary instruction at the Lipid Clinic. Each point represents the mean change in body weight in relation to the start of dietary intervention (Day 0). Data shown from 11.5 months prior to 10.5 months post the initial Lipid Clinic visit ± SEM.

DAYS OF FOLLOW-UP VISITS (DAY 0 = FIRST VISIT)

Figure 7: Serum cholesterol response of the same group of patients to diet. Data shown as in Figure 6.
showed no decrease after dietary counselling (Fig. 7).

This group was further divided into two groups (Group A, n=4; Group B n=5) according to their weight response to the diet (Fig. 8). Group A had a mean weight loss of 4.4 ± 1.3 kg, whereas Group B tended to gain weight (0.3 ± 0.4 kg) at 315 days following dietary instruction. Changes in body weight over time were significant for both groups (p < 0.01), and there was a significant difference in weight change over time between groups (p < 0.01). There was a significant difference between groups in their weight response to the diet (p < 0.01), which was expected since it was the manner by which they were classified. Prior to dietary instruction, both groups gained weight, and these changes in body weight over time were significantly different between groups (p < 0.05). Thus, although both groups gained weight (Group A: 7.1 ± 0.4 kg; Group B: 3.8 ± 1.4 kg) prior to dietary instruction, Group A who had a better response to the diet, was comprised of those who had gained more weight prior to diet treatment. Group B who gained less weight, tended to maintain the increased body weight, or slightly gain weight following diet treatment.

For both groups there was a significant effect of time on serum cholesterol before dietary counselling (p < 0.01), but not after. There were no differences between groups in terms of their cholesterol response to the diet (Fig. 9).

**Effect of prednisone**

The effect of prednisone discontinuation on body weight and serum cholesterol was
Figure 8: Weight response of the same group as in Figure 6 divided in 2 according to their weight response to the diet (Group A in closed squares, n=4 and Group B in open circles, n=5). Each point represents the mean change in body weight from the start of dietary intervention (Day 0), from 11.5 months prior to 10.5 months post the initial Lipid Clinic visit ± SEM.

Figure 9: Serum cholesterol response of the same 2 groups to diet. Data shown as in Figure 8.
examined in 7 and 8 patients respectively whose prednisone dose was slowly tapered, then completely withdrawn (Figs. 10 and 11). Prednisone dose was gradually tapered from 15 to 2.5 mg/day, then completely withdrawn.

There was a significant increase in body weight as prednisone dose decreased from 15 mg/day ($p < 0.01$), and from 12.5 mg/day ($p < 0.01$) to 0 mg/day. (Fig. 10). However, mean body weight did tend to decrease when prednisone was tapered from 7.5 to 2.5 mg/day.

There was a significant decrease of $0.8 \pm 0.3$ mmol/l ($p < 0.01$), and $0.6 \pm 0.1$ mmol/l ($p < 0.05$) in serum cholesterol concentrations as prednisone was tapered from 10 to 0 mg/day and 5 to 0 mg/day respectively (Fig. 11). Following total withdrawal however, no significant change in serum cholesterol was observed (data not shown).

By multivariate analysis, prednisone dose was the only variable to be significantly associated with the change in serum cholesterol at six months post transplantation ($p < 0.05, n=25$).

It was interesting to note, that the most dramatic increases in serum cholesterol levels occurred when the prednisone dose was at its highest levels. A prednisone dose of $12.6 \pm 0.9$ mg/day was the cut-off point, after which higher doses resulted in serum cholesterol elevations (Figs. 2 and 5).
Figure 10: Body weight response to prednisone withdrawal in 7 patients who discontinued the use of prednisone. Each point represents the mean change in body weight (± SEM), in relation to the decreasing prednisone dosages.

Figure 11: Serum cholesterol response to prednisone withdrawal in 8 patients who discontinued the use of prednisone. Data shown as in Figure 10.
Effect of pretransplantation cardiac diagnosis

There was a significant change over time in body weight and serum cholesterol for both the ischemic and non-ischemic groups ($p < 0.01$), (Figs. 12 and 13). There was no significant difference in changes in body weight and serum cholesterol levels between groups. There was no significant difference in prednisone dose between the groups.
Figure 12: Effect of pretransplant diagnosis on the mean change in body weight from the time of transplantation (Day 0) ± SEM. Ischemic group: closed squares, n=4; Non-ischemic group: open circles, n=11.

Figure 13: Effect of pretransplant diagnosis on the mean change in body weight from the time of transplantation (Day 0) ± SEM. Ischemic group: closed squares, n=4; Non-ischemic group: open circles, n=11.
DISCUSSION

This retrospective study demonstrated a time-dependent and sustained increase in both total cholesterol levels and body weight following transplantation. Progressive augmentations during the first 8 and 10 months, respectively, were observed, followed by a stabilization which remained above pretransplant levels during the first 2.5 years after surgery.

Although the literature has been consistent in reporting a significant degree of hyperlipidemia following transplantation, there remains a disparity regarding the incidence and type of hyperlipidemia described. This has probably resulted from differences in sample sizes, time intervals at which data were collected, duration of data collection after surgery, type of data collection (retrospective vs prospective), as well as differences in follow-up treatment procedures, and characteristics of the population being studied (i.e. age, immunosuppressant regimens, etiology of heart failure, existence of pretransplantation hyperlipidemia and obesity, use of antihypertensive medication, and dietary and exercise patterns). The lack of control of variables such as exercise, diet, and weight gain within the various studies can also influence results (Miller, 1991).

The mechanisms responsible for post cardiac transplantation hyperlipidemia have yet to be elucidated, and researchers in the field agree that future research is required to identify its predictors more fully, in an effort to target effective intervention programs to reduce or prevent excessive elevations in body weight and lipid levels (Grady, 1991).

This study collected body weight and serum cholesterol data at monthly time intervals
prior to transplantation and throughout the first three years following transplantation, and therefore represents a more detailed time-course than previously reported. Grady et al (1991) also collected data both preoperatively and postoperatively up until three years, but collection was yearly rather than monthly. Others have observed changes every 3 and 6 months over a period of one year (Keogh et al, 1988; Stamler et al, 1988; Taylor et al, 1989). Still others have examined the effects of transplantation at an average time of approximately two years postoperatively (Bilodeau et al, 1989). Although variable, the literature reports lipid elevations from as early as two weeks, which are maximum between 3 and 6 months after transplantation. Total cholesterol is the lipid fraction most commonly elevated, predominantly due to elevations in the LDL fraction (Miller, 1991).

In our study, serum cholesterol peaked at about eight months, also in conjunction with elevated LDL-C and TG levels.

In contrast to our results, Grady et al (1991) observed a significant decrease in serum cholesterol levels from the first to the third postoperative year. They attributed this finding to the lower corticosteroid doses by three years post transplantation, and to perhaps improved adherence to diet and exercise plans with their study sample. By 2.5 years, our patients still exhibited a significant degree of hypercholesterolemia, perhaps related to poor compliance with diet and exercise plans in our sample, or to a greater incidence of rejection among our sample, resulting in greater prednisone doses. The smaller sample size used in our study may have accounted for the difference as well (16 patients as compared to 54 in the Grady et al study).

As with the results of Keogh et al (1988) and Grady et al (1991), we also observed
a significant increase in body weight. Body weight remained above pretransplant levels at 2.5 years after surgery both in the Grady et al series and our study. In the former however, premorbid body weight data were collected, and results indicated a weight gain in excess of preillness levels. Although our study collected body weight data at 100 days prior to transplantation from patients for which this data were available, such body weights may not have been a true reflection of premorbid body weights. It is therefore difficult to determine what percentage of body weight (if any) was actually a result of transplantation, or was a return to premorbid levels.

In our study, patients tended to lose weight during the first postoperative month. Although not significant, this could have been due to the recovery of surgery and hospitalization which can cause a transient decrease in appetite and food intake. The medication regimen may have affected appetite as well. Weight loss may have also been related to water losses, as a result of improved cardiac function, and reversal of congestive heart failure.

Possible contributors to post transplantation weight gain include increased appetite as a result of corticosteroid therapy, a sense of well-being with improved heart function, improved nutrient absorption, return to premorbid body weight, as well as lack of compliance with prescribed diets, and exercise schedules (Grady et al, 1991; Keogh et al, 1988). Although corticosteroid therapy is a potential contributor to weight gain, note that we did not observe a beneficial effect of prednisone discontinuation on body weight. We are not aware of another study that has examined the effects of prednisone withdrawal on body weight.
Tapering of prednisone did have a significant effect on serum cholesterol. However as with body weight, no further improvement was observed when prednisone was totally discontinued. Serum cholesterol actually reached a minimum value of 5.3 ± 1.1 mmol/l when prednisone was completely discontinued. Levels then tended to increase, not significantly though and not returning to pre-prednisone withdrawal levels (data not shown). These results suggest an association between prednisone dose and post transplantation elevations in total cholesterol. Interestingly, there seems to be a threshold effect, whereby lower prednisone doses will decrease serum cholesterol to a certain level after which no further improvement occurs.

Much of the literature has identified corticosteroid use as a major contributor to posttransplant hypercholesterolemia (Becker et al, 1988; Taylor et al, 1989; Renland et al, 1989). Our results support this relationship, as multivariate analysis demonstrated prednisone dose to be the only variable significantly associated with changes in serum cholesterol. However, there is controversy as to whether prednisone, cyclosporine, or both are the primary predictors (Miller, 1991). Both have been reported to be associated with changes in lipoprotein concentrations, and have been implicated in the abnormal lipoprotein profile that develops after cardiac transplantation. We did not evaluate the contribution of cyclosporine to post transplant hypercholesterolemia since none of our patients discontinued its use, and all followed the same immunosuppressive regimen. Stamler et al (1991) conclude that in clinical practice, tapering of prednisone is commenced soon after transplantation, and therefore is unlikely to be the primary cause of the concomitant significant elevations in serum cholesterol that evolve. However, our
results suggest that although prednisone dose decreased as serum cholesterol levels increased, the dramatic increases occurred initially after transplantation when prednisone doses were highest. A mean prednisone dose of $12.6 \pm 0.9$ mg/day was the level above which elevations in mean serum cholesterol were observed. Cholesterol levels stabilized thereafter with lower doses.

Our results suggest no statistically significant association between weight gain and elevations in serum cholesterol, which contrast with the results of Keogh et al (1988). This disparity perhaps lies within the different time intervals at which data were collected, as well as our small sample size. The lack of an association between body weight and serum cholesterol was further observed when dietary counselling showed a beneficial effect on body weight, but not on serum cholesterol. Even those patients who had a better weight response to the diet showed no improvement in serum cholesterol levels. However, our sample size may have been too small to show any beneficial effect of diet on reducing serum cholesterol. In addition, ameliorations in serum cholesterol associated with lower prednisone doses did not accompany decreases in body weight. Although, the literature has reported strong associations between body weight and serum cholesterol levels in the normal population, it is important to note that these studies usually are comprised of heterogeneous persons in fairly good health with no metabolic disturbances, and taking no immunosuppressive agents.

Improvements in body weight but not in serum cholesterol following dietary intervention (data collected retrospectively) may be a reflection of corticosteroid use, since post transplantation hypercholesterolemia not weight gain, was associated with
prednisone. Hence, diet may be a better predictor of post transplantation weight gain, whereas corticosteroid use may be a better predictor of post transplantation hypercholesterolemia. The poor relationship between body weight and prednisone use was further substantiated by the lack of difference in prednisone dose between groups A and B who were divided according to weight gain. It's important to note however, that although prednisone dose was not significantly different between the two groups, it did tend to be higher in the group with significantly higher serum cholesterol elevations.

It was interesting to observe that the group of patients who had a better weight response to the diet gained more weight prior to dietary instruction. Possible reasons for the appearance of two distinct groups are the following: 1) it may have been easier for some patients in the group who gained more weight to lose weight after dietary instruction; 2) some patients in the "non-responsive" group may have actually reached their normal body weight post transplantation and, therefore, it was more difficult for them to lose additional weight; 3) some patients may have already been consuming a diet similar to the prescribed Step One diet, hence minor dietary modifications may not have resulted in any further weight changes; 4) regression towards the mean; some patients may have been given a weight loss diet because they had reached a high body weight at one time, and this level then came down naturally; 4) differences in fluid retention and water losses.

No association was observed between body weight and serum cholesterol elevations and a pretransplant diagnosis of ischemic heart disease. Both the ischemic and non-ischemic group exhibited a significant degree of hypercholesterolemia and body weight
gain after transplantation. The disparity within the literature of associations between cardiac diagnoses and post transplant hypercholesterolemia may reflect different pretransplant serum cholesterol levels among ischemics and non-ischemics, the persistence of pretransplant hypercholesterolemia after transplantation, and differences in time at which pretransplant data were collected (i.e. one year prior to surgery, three months, etc.). The subset of patients for which pretransplant data were available demonstrated no hypercholesterolemia prior to transplantation (Table 2). However, the existence of premorbid hypercholesterolemia was not ascertained in this study. Comparisons between ischemics and non-ischemics prior to transplantation were difficult as a result of small sample size, and inequalities in the number of subjects between groups for whom pretransplant data were available. Furthermore, the three females of this study were in the nonischemic group which lowered the group’s mean body weight.
CONCLUSIONS

Substantial elevations in both body weight and serum cholesterol were observed during the first 10 and 8 months respectively following transplantation. Levels stabilized thereafter. No association was observed between serum cholesterol levels, body weight, and a pretransplant cardiac diagnosis of ischemic heart disease, which may be a reflection of small sample size. Prednisone withdrawal had a significant effect on serum cholesterol levels. A significant decrease of $0.8 \pm 0.3$ mmol/l and $0.6 \pm 0.1$ mmol/l was observed as the prednisone dosage was slowly tapered from 10 to 0 mg/day, and from 5 to 0 mg/day respectively. Although higher prednisone doses are associated with weight gain via increased appetite, we did not observe a benefit of decreasing prednisone dose on body weight. Dietary intervention had a beneficial effect on body weight, but was not associated with decreased serum cholesterol levels. No association between changes in body weight and changes in serum cholesterol levels was observed, which may be a reflection of small sample size, or the effect of prednisone on serum cholesterol.
STUDY LIMITATIONS

Limitations of this study included: 1) small sample sizes on which the various analyses were performed as a result of missing data 2) the lack of complete, monthly, lipid profile data, 3) uncontrolled confounders such as diabetes mellitus, smoking, sex, age, exercise, and antihypertensive medications which may have all influenced post transplant lipid levels, 4) disadvantages of a retrospective study including: inaccurate recording of data in medical records, missing data in medical records, and a non-randomized, non-controlled study design, and 5) unavailable data regarding premorbid, usual body weights and serum lipid levels, to make meaningful comparisons with post transplant data.
PURPOSE OF INTERVENTION STUDY

As shown through this study and others, obesity and hyperlipidemia are serious problems following cardiac transplantation, and may act synergistically with immune factors to accelerate the development of CAD (Farmer et al, 1991; Eich et al, 1991; Carrier et al, 1991), and threaten patient survival. They therefore require further exploration and intervention.

The existence of a group of patients who responded well to diet revealed the effectiveness of diet in reducing excessive weight gain after transplantation. Furthermore, nutritional programs for heart transplant recipients have improved patient status (Ragsdale, 1987). Thus, we conducted a controlled dietary intervention study, as a means of identifying an effective intervention program targeted at preventing elevations in body weight and serum lipid levels after cardiac transplantation.
CHAPTER 3
DIETARY INTERVENTION OF HYPERLIPIDEMIA AND WEIGHT GAIN POST CARDIAC TRANSPLANTATION
HYPOTHESES

We hypothesized that the Step One lipid lowering diet, with an energy intake appropriate to achieve a healthy body weight, commenced immediately after transplantation would minimize or prevent elevations in lipids, lipoproteins, and body weight which occur after cardiac transplantation.

To our knowledge, the evolution of whole body composition following heart transplantation has not been assessed in a detailed manner in the literature. Although post transplant elevations in body weight have been observed, the composition of this excess weight gain has not been studied. As part of our intervention study, we set out to answer the following: are changes in body weight following transplantation a function of changes in body fat, and body fat distribution, changes in body water balance which may be a reflection of immunosuppressant therapy (Funk, 1986), or changes in lean body mass (LBM) as a reflection of improved nutritional status (Keogh et al, 1988)? We hypothesized that changes in body weight are principally a result of changes in body fat due to the dramatic increases above pretransplant levels observed during our retrospective study.

From clinical observations, in following such patients individually, we noted that heart transplant patients seem to possess great enthusiasm about food, and are very descriptive when explaining what they consume. The study was therefore also designed to assess appetite, hunger, attitudes toward food, beliefs toward changing eating habits,
stress from following a diet, and nutrition knowledge after transplantation, to gain further insight about the potential factors associated with weight gain after heart transplantation. We hypothesized further, that patients with a greater appetite after transplantation, who possess a less favorable attitude towards food, who are less interested in changing their eating habits, who perceive fewer benefits in changing eating habits, who perceive more difficulties in changing eating habits, who have more stress, and who have less nutrition knowledge will consume a diet higher in energy intake following transplantation, and will be more likely to have greater weight gains following transplantation.
MATERIALS AND METHODS

Patient Population

All patients on the active list for cardiac transplantation at the Royal Victoria Hospital between April 1991 and March 1992 were requested to participate in the study. The protocol was approved by the Department of Medicine Human Ethics Committee at the Royal Victoria Hospital. Written consent was obtained from all participating subjects following a complete explanation of the protocol. Any patient requiring lipid lowering medication following transplantation was excluded from the study.

Data Collection

Data were collected prior to transplantation (when possible), and then monthly for a period of seven months after transplantation. Table 5 describes the protocol. Data collection included the following: height, body weight, total cholesterol, complete lipid profile (LDL-C, HDL-C, TG, ApoA1, ApoB, Lp(a)), fibrinogen, body composition measurements (body fat, LBM, body water), waist:hip circumference ratio, activity level, and nutritional information from food records.

Prior to surgery, subjects were also requested to respond to a questionnaire (see Appendix) to gather information on appetite, attitudes, beliefs, hunger, stress, and nutrition knowledge. The questionnaire was completed again at 3 and 6 months after
Table 5: Summary table illustrating times at which data were collected. Pretx = pre-transplantation, 0 = time of transplantation (or prior to hospital discharge for questionnaire completion), 1-7 = 1-7 months after transplantation. Fib. = fibrinogen; Body Comp. = body composition measurements; Body Fat Dis. = body fat distribution measurements; Quest = questionnaire completion. Each X represents the time at which each variable was measured.

<table>
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<th>DATA</th>
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<td>X</td>
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</table>

- Pretx = pre-transplantation
- 0 = time of transplantation (or prior to hospital discharge for questionnaire completion)
- 1-7 = 1-7 months after transplantation
- Fib. = fibrinogen
- Body Comp. = body composition measurements
- Body Fat Dis. = body fat distribution measurements
- Quest = questionnaire completion
- Each X represents the time at which each variable was measured.
transplantation. In addition, a complete diet history and assessment of usual intake were performed by the investigator to obtain information concerning the subjects' eating habits and dietary patterns prior to surgery.

Following transplantation, all subjects received a copy of the Heart Smart: Good Food Choices pamphlet published by the Heart and Stroke Foundation of Canada, and were given guidelines for a prudent diet. Patients were then randomized (by Dr. Katherine Gray-Donald, Department of Dietetics and Human Nutrition, and Epidemiology, McGill University) into either a dietary treatment group or control group. Randomization was stratified according to the following body weight criteria: overweight patients (BMI > 25 kg/m²) or normal weight patients (BMI between 20-25 kg/m²). Two sets of envelopes were developed: one set for the overweight group, and one set for the normal weight group. Each envelope contained an index card which indicated either "diet" or "no diet" which determined to which group each patient would be allocated.

Follow-up visits were monthly. Patients were seen by nurses and doctors of the transplant clinic, and if necessary by a dietitian if hypercholesterolemia developed, or diabetes worsened (one patient had Type 2 diabetes). Monthly follow-up visits included a physical examination by the attending physician, blood tests, body weight measurements, bioelectrical-impedance analysis (BIA) for body composition estimation, waist and hip measurements for an indication of body fat distribution, and a review of food records for a determination of the nutrient composition of diets. Completion of the questionnaire occurred at months 3 and 6. The treatment group were instructed the Step One Lipid Lowering diet by the investigator, and were followed monthly on an individual
basis. Problems concerning the diet, and food diaries were discussed at each visit and
adjustments to their diet were made accordingly (if necessary).

**Step One Diet**

Diets for the treatment group were calculated according to individual needs, and
individual eating patterns as revealed through the diet history and usual intake. Energy
requirements were determined using the Harris-Benedict equation and were adjusted
according to activity level. They were then readjusted (if necessary), according to the
needs of the patient, (weight gain or weight loss), to produce a weight change of 0.45 -
0.91 kg a week until the desired weight was achieved. Nutrient distribution was then
calculated according to the following scheme (all percentages were a function of total
energy) : total fat < 30%; saturated fat (SFA) < 10%; polyunsaturated fat (PUFA) 7-
10%; monounsaturated fat (MUFA) 10-15%; carbohydrate (CHO) 50-60%; protein 10-
20%; cholesterol (CHOL) < 300 mg/day.

Diets were based on the exchange system using the Good Health Eating Guide
published by the Canadian Diabetes Association. A commercial booklet, commonly used
in clinical practice for teaching low cholesterol diets, developed by Becel, was used as
a teaching guide for information concerning foods allowed and foods to avoid on the diet.
Dietary instruction with regards to fiber consumption was also provided.
Body Weight

Body weight was recorded to the nearest 0.1 kg, and was usually measured following an overnight fast. Patients removed their shoes, jackets, belts, and wallets from pants before the measurement.

Blood Values

Total cholesterol, HDL-C, TG, and LDL-C levels were determined as described in Chapter one, but using the Beckman C7 system. ApoA1, and ApoB levels were determined using the Beckman Array Nephelometer (Sternberg, 1977).

Lipoprotein (a):

Lipoprotein (a) was determined by the technical staff at the Endocrinology Laboratory of the Royal Victoria Hospital also using the Rate Nephelometer Approach. This system permits the quantitative determination of Lp(a) by immunoprecipitin analysis. Standards, controls, and samples are pipetted undiluted into sample cups. Microvolumes of these samples and polymeric enhancer are automatically pipetted into individual cuvettes. Undiluted antiserum is then added. The sample (antigen) solution and antiserum are then mixed. Insoluble antigen-antibody complexes begin to form immediately. Following an incubation period of 10 minutes, the absorbance of the solution is measured at the
analytical wavelength. Concentrations of samples and controls are interpolated from the calibration curve.

**Fibrinogen:**

Fibrinogen concentrations were determined using a fibrometer at the Hematology Laboratory at the Royal Victoria Hospital. Thrombin which was warmed to room temperature was added to the samples of plasma containing fibrinogen which were then converted enzymatically to fibrin. Fibrin then underwent polymerization to form a fibrin network. Factor XII, activated by thrombin, catalysed the formation of stabilizing cross-links to produce a visible clot. The elapsed time, from the addition of thrombin to the formation of a clot was inversely proportional to fibrinogen concentrations. Time for clot was recorded to the 0.1 second. Fibrinogen concentrations were interpolated from the calibration curve.

**Bio-Impedance Analysis**

Bio-impedance analysis is a rapid and painless procedure used to evaluate body composition (% LBM, % body fat, % body water). A four-terminal bioimpedance analyzer was used. Measurements were taken while patients lay in a supine position, in the fasting state. Two electrodes were cut in half lengthwise. The cut edge of the electrodes placed on the ankle and wrist, face toward the shoulder and thigh respectively.
One half was placed on an imaginary longitudinal line on the dorsal surface of the right hand proximal to the metacarpal-phalangeal joint, another half was placed at the right pisiform prominence of the wrist, the proximal edge dissecting the ulnar tubercle, a third half was placed anteriorly on an imaginary longitudinal line between the medial and lateral malleoli, the proximal edge bisecting the medial malleolus, the last half was placed dorsal on the right foot proximal to the metatarsal-phalangeal joint. A small electrical current was introduced into the body, and the impedance to the current flow resulting from resistance and reactance was measured (Brylowski, 1992, Ross et al, 1989). Two readings for both resistance and reactance were taken. The resistance and reactance values, as well as values for body weight, age, sex, height, plasma sodium and potassium were entered into a bio-impedance program developed by Dr. Harry M. Shizgal of the Royal Victoria Hospital. This provided a set of values for LBM, body fat, total body water, body cell mass, and extracellular mass for each patient, along with reference ranges.

**Body Fat Distribution**

Body fat distribution was estimated using the waist:hip ratio procedure (Després et al, 1990). Waist measurements were taken across the umbilicus. Hip measurements were taken across the largest girth protruding between waist and thigh. Both were measured using a tape measure, and were recorded to the nearest inch, using one decimal place.
Food Diary

Food diaries were collected every month, and included time of food consumption, description of the food consumed, method of preparation, and the quantity consumed. Prior to transplantation, and at 3 and 6 months following, patients were requested to complete the food diary for a period of two consecutive weeks for an adequate estimation of dietary fat and cholesterol intake (Sembros et al, 1985). Food diaries completed for the other months were recorded for three consecutive days including a weekend day, and were used to review food intake with patients on follow-up visits. The actual days used for food recording varied among patients. Two week food diaries were analysed using the CBORD diet analyser program, 1991 Canadian Nutrient File.

Activity

The activity level was determined for each patient at each follow-up visit and classified according to the following activity factors: sedentary: 20-25%; mobile, but not involved in an exercise program: 30%; exercising three times or more a week (walking, bicycle, etc.: 35-40%).
The questionnaire was developed in collaboration with Dr. Irene Strychar from the Nutrition Department of the Université de Montréal (Appendix 1). It was divided into six sections: appetite, hunger, attitudes, beliefs, stress, and knowledge.

Appetite was measured using continuous scores ranging from 1-5 (very poor to very good) and 1-7 (decreased a great deal to increased a great deal). Patients were asked to rate their appetite at three different times: within the past month, today, and since heart transplantation. Overall changes in appetite, as well as changes in appetite during specific eating times (breakfast, lunch, supper, and snacks), during specific situations (social gatherings, anxiousness, loneliness, boredom, and when with family or friends), and for specific food items (desserts, snack foods, fried foods, and the four food groups) were asked. Patients were also asked to describe why they felt their appetite had changed, if applicable. Questions from this section were based on the type of assessment we wanted to make, and were not adapted from another scale.

Seven questions comprised the hunger section which also used continuous scores ranging from 1-7 (strongly agree to strongly disagree). Questions from this section were developed to identify the level of hunger after cardiac transplantation, and to differentiate the concept of hunger from that of a greater appetite. Questions were adapted from the Three-Factor Eating Questionnaire developed by Albert Stunkard, which measures dietary restraint, disinhibition, and hunger among chronic dieters (Stunkard, 1985).

The attitudes section included 10 questions describing attitudes toward eating in
general, toward eating nutritious foods, toward eating a lot of food, toward eating fattening foods, and toward eating foods high in cholesterol. The scores were also continuous (strongly agree to strongly disagree). Questions were also adapted from parts of the Stunkard questionnaire.

Beliefs were operationalized according to the Health Belief Model which is applied to explain and predict what influences behavior (Rosenstock, 1990). Three components of this model were used in this questionnaire: perceived interest concern, perceived benefits, and perceived barriers. Perceived interest concern refers to the level of motivation needed to make health issues important (i.e. how interested are you in improving your eating habits?). Perceived benefits refer to the belief that following a particular health recommendation would be beneficial in reducing susceptibility to a serious health problem (i.e. how useful do you think improving your eating habits would be in maintaining your weight?) Perceived barriers refer to the barriers that must be overcome to adhere to that recommendation (i.e. how difficult would it be for you to change your eating habits to control your weight?) All questions were based on continuous scores.

Overall level of stress, its effect on eating habits, and the effect of diet on stress level were also assessed using continuous and dichotomous scores to determine whether a stress effect on appetite or from diet existed and to evaluate its relationship to body weight. Questions from this section were not adapted from another measure.

To evaluate nutrition knowledge with respect to fat, and cholesterol, and its relationship with changes in body weight and serum cholesterol levels, 11 questions were
developed asking to identify sources of foods high in fat and cholesterol, the differences between saturated, monounsaturated, and polyunsaturated fats with respect to food sources and effects on blood cholesterol levels, and effective measures of controlling body weight and cholesterol concentrations. All questions were scored dichotomously. The knowledge test questions were based upon the content of my dietary intervention.

Pre-pilot study

Once the first draft of the questionnaire was completed, a pre-pilot study was performed. The questionnaire was given to four heart transplant recipients, one of whom had been recently transplanted. One female and three males participated. They were asked whether the questionnaire was easy to understand, whether questions were repetitive, what should be changed, as well as to provide any other additional comments they may have had. The necessary revisions were then made, and a pilot study was performed to determine the questionnaire’s reliability.

Pilot study

The pilot study involved two sets of heart transplant patients; those who had been transplanted from 8 months to 5 years prior to the pilot study, who were not participating in the dietary intervention study, and those who had been recently transplanted (except for one), who were subjects in the intervention study at the time the pilot study was
taking place. One patient from this group was on the active list for transplantation, and had completed the questionnaire for a pretransplantation assessment, and therefore was included in the pilot study. The number of patients in each group was 8 and 10 respectively, for a total of 18 patients.

Subjects were asked to complete the revised questionnaire. Once completed, all answers were entered into files using the Dbase 3 computer program. We then computed reliability coefficients for hunger, attitudes, beliefs, stress, and knowledge to determine their internal consistency. For continuous variables, the Cronbach’s coefficient alpha (CCA) was used, and for dichotomous variables, the Kuder-Richardson method was used. If a patient did not answer a particular question, it was coded as a missing value, and not included in the analysis. Only patients with complete answers were included. Reliability coefficients were computed for each group separately, and for both groups combined as a whole.

Intervention Study

For the intervention study, analyses were based on three groups: the diet group, the control group, and the pilot group (those in the pilot study but who were not part of the intervention study). The mean of all responses for each patient was computed for each section and classified according to group and time. Time one referred to patients who completed the questionnaire at, or before one month after surgery, time three referred to patients who completed the questionnaire 2-5 months after surgery, and time six referred
to patients who completed the questionnaire six months after surgery.

For the knowledge section, when a numbered answer was given, that number was entered into the file. When more than one answer existed for a question, a 0 or 1 code was used. Each patient was required to circle the answer that they thought was correct, hence if a letter was not circled, the code was 0, and if a letter was circled, the code was 1. A Dbase program was then developed to score the answers. For each correct answer, a score of 1 was given, and for each incorrect answer, a score of 0 was given. Total scores for each patient were computed, and the mean total score per group was determined. The total score was a weighted score out of 100 points. The weight of each question was determined according to the amount of items asked per question and according to how much it was emphasized during dietary instruction. The number of points allocated to each question are as follows: Question 36: 11 points; questions 37-40: 15 points; question 41: 10 points; questions 42 and 43: 2 points; question 44: 4 points; question 45: 6 points; question 46: 5 points.

Statistical Analyses

Changes over time in the aforementioned variables in the treatment and control groups, were graphed using a graphics program designed by Dr. Jean-Francois Yale, Royal Victoria Hospital, Montreal. Comparisons between groups were difficult in light of the small sample size, and the uneven distribution of patients between the treatment
and control groups. Hence statistical analyses between groups were not performed. It was therefore decided to describe our results as a whole rather than assess any differences between groups as a result of dietary treatment.

To determine any relationship between body weight and serum cholesterol levels with questionnaire items (appetite, attitudes toward food, hunger, beliefs, stress, and knowledge), Pearson correlation coefficients were computed using SAS.
RESULTS

Sample size

Fifteen patients were transplanted during the study period. One patient completed only three months of the study then dropped out as he did not want to continue following the diet. Another patient was excluded from the study, since he was administered cholesterol lowering medication which would have been a potential confounder. Another patient died following surgery. Five patients refused to participate. Hence, our final sample size consisted of the remaining seven patients who agreed to participate in the study.

From randomization, five patients were allocated to the treatment (diet) group, and two patients were allocated to the control group. Both patients from the control group were classified as overweight at the start of the study. From the treatment group, three patients were overweight, and two were of normal weight. Demographic characteristics of the patients are described in Table 6.

Body Weight, Lipids and Lipoproteins

Patients in the diet group initially tended to gain weight (0.8 ± 1.3 kg) (Fig.14), then tended to lose weight, during the first month post transplantation. This initial change in body weight was probably related to immediate postoperative edema followed by a
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<th>GROUP</th>
<th>N</th>
<th>MEAN AGE</th>
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<th># NON-ISCH</th>
<th># OVERWT</th>
<th># AVE. WT</th>
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<td>DIET</td>
<td>5</td>
<td>55.4 ± 2.3</td>
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<td>1</td>
<td>3</td>
<td>2</td>
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<td>CONTROL</td>
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<td>2</td>
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</table>

Table 6: Patient Demographics. N = sample size; mean age = age at time of transplantation; # NON-ISCH = # of patients with pre-transplant diagnosis other than ischemic heart disease; # overwt/# ave. wt = # of patients in each group who were over weight, and of average weight. All subjects were male.
MONTHS POST TRANSPLANTATION

Figure 14: Mean changes in body weight from the value closest to (but prior to) transplantation (time -1) for the diet group (closed squares, ± SEM, n = 5) and controls (open squares, n=2).

MONTHS POST TRANSPLANTATION

Figure 15: Mean changes in serum cholesterol from the value closest to (but prior to) transplantation (time -1) for the diet group (closed squares, ± SEM, n = 5) and controls (open squares, n=2).
return to a normal hydration status. Time -1 is the time closest to transplantation whereas time 0 is post transplantation during recovery period, hence most of the weight gain shown is postoperative. An overall increase in body weight of 4.2 ± 2.3 kg by the end of the study was observed. The control group followed a similar pattern, although their initial weight gain was approximately 8.0 kg. They demonstrated an overall mean weight gain of 5.7 kg by the end of the study. The results suggest an overall mean increase in serum cholesterol concentrations in both the diet and control groups, of 0.7 ± 0.4 mmol/l and 1.2 mmol/l respectively (Fig. 15). Immediately after transplantation, serum cholesterol levels tended to decrease.

At seven months post transplantation, the diet group tended to demonstrate increases in HDL-C, TG, and ApoA1, of 0.7 ± 0.1 mmol/l, 0.2 ± 0.1 mmol/l, and 34.2 ± 8.9 mg/dl respectively (Table 7). The change in LDL-C and ApoB at seven months was fairly variable among patients in this group. The mean change in LDL-C and ApoB was -0.05 ± 0.37 mmol/l and 2.8 ± 13.0 mg/dl respectively. Mean Lp(a) concentrations tended to decrease by 14.7 ± 3.4 mg/dl. Fibrinogen was also fairly variable, and had a slight overall increase of 0.09 ± 0.28 mg/dl. One patient from the control group showed an overall decrease in LDL-C (-1.7 mmol/l), and a final increase in HDL-C (0.4 mmol/l). This data was missing for the other control. Mean triglyceride levels tended to increase by 1.06 mmol/l for this group. We were not able to obtain baseline data on ApoA1, ApoB, Lp(a), and fibrinogen levels for the control group as they were emergency transplants, and these variables are not routinely measured. However, throughout the study, the trend was for ApoA1, ApoB, and Lp(a) to increase.
<table>
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<td>4.2 ± 2.3</td>
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<td>6.0 ± 1.1</td>
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<td>Δ TC (mmol/l)</td>
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<td>2.1 ± 1.3</td>
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<td>Δ LDL-C (mmol/l)</td>
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<td>Δ HDL-C (mmol/l)</td>
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<td>Δ TG (mmol/l)</td>
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<tr>
<td>Δ ApoB (mg/dl)</td>
<td>2.8 ± 13.0</td>
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<td>-</td>
</tr>
<tr>
<td>Δ ApoA1 (mg/dl)</td>
<td>34.2 ± 8.9</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Δ Lp(a) (mg/dl)</td>
<td>-14.7 ± 3.4</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Δ Fib. (mg/dl)</td>
<td>.09 ± 0.3</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
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Table 7: Comparison of the change in study variables from the time of transplantation (time 0) to the end of the study (time 7) between groups. Each value represents the mean change at time 7 from time 0 ± SEM. Retro = patients from the retrospective study (chapter 2) (n=35). Diet (n=5), control (n=2 for TG, and n=1 for LDL and HDL), BW = body weight; TC = total cholesterol; LDL-C = low-density lipoprotein cholesterol; HDL-C = high-density lipoprotein cholesterol; TG = triglyceride; ApoB = apolipoprotein B; ApoA1 = apolipoprotein A1; Lp(a) = lipoprotein (a); Fib. = fibrinogen.
Activity Level:

Both the diet and control group had increases in their level of activity. However, the control group participated in some form of physical activity program (cycling, cross-country skiing, or walking), whereas the diet group maintained an average level of activity, although they were encouraged to do more (Fig. 16).

Body Composition:

Percent body fat was the only indicator of body composition to increase after transplantation among both the control and diet group (Table 8). The percentage of body fat increased by 27.1 ± 8.6% in the diet group, and by 18.1% in the control group. Levels of lean body mass and body water tended to decrease.

Body Fat Distribution

Waist:Hip Ratio:

No apparent change was observed in body fat distribution among both the control and diet groups. Subjects from both groups tended to maintain healthy waist:hip ratios (< 1.0).
MONTHS POST TRANSPLANTATION
Figure 16: Activity level in relation to time after transplantation (Day 0, ±SEM), for the diet group (closed squares, n=5) and the controls (open squares, n=2).

MONTHS POST TRANSPLANTATION
Figure 17: Level of hunger in relation to time after transplantation (Day 0, ±SEM), for the diet group (closed squares, n=5), the controls (open circles, n=2) and the pilot subjects (open squares, n=8).
Table 8: Changes in levels of body composition related to transplantation. Each point represents the mean change at the end of the study from time of transplantation ± SEM. LBM = lean body mass; C = control group; D = diet group.

<table>
<thead>
<tr>
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<th>C (n=2)</th>
<th>D (n=5)</th>
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<tr>
<td>Δ % fat</td>
<td>18.1 ± 21.3</td>
<td>27.1 ± 8.6</td>
</tr>
<tr>
<td>Δ % LBM</td>
<td>-9.7 ± 3.7</td>
<td>-0.3 ± 4.4</td>
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<tr>
<td>Δ % body water</td>
<td>-9.7 ± 3.7</td>
<td>-0.3 ± 4.4</td>
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Table 9 illustrates changes in nutrient intakes throughout the study period. Reported energy intake tended to decrease throughout the study. Prior to transplantation, patients reported a greater energy consumption. Patients in the diet group reported energy intakes between the range of 68.8-101% of estimated energy requirements. Patients of the control group reported energy intakes between the range of 74.5-85% of estimated energy requirements.

There was a trend for a decrease in total fat consumption among all subjects. The recommended level of 30% or less was achieved at three months for the diet group (26.8 ± 4.1%, n=5), and at six months for the control group (28.6%, n=2), from pretransplant levels of 32.4 ± 1.8%, n=6 and 33.2%, n=2 respectively. Percent fat intakes at the time of transplantation were 31.1%, n=2, and 30.2 ± 2.9%, n=5 for the control and diet group respectively.

The decrease in total fat intake tended to reflect reductions in saturated and polyunsaturated fat intake. Percent saturated fat was between 7-10% for the diet group at three months (8.5 ± 0.9%, n=5), and at six months for the control group (8.5%, n=2). Throughout the study, subjects maintained PUFA levels below 10%, except for the control group prior to transplantation, who had a mean intake of 11.6% (n=2). Monounsaturated fat intake tended to decline during the first three months after surgery, then increased during the latter three months. At the end of the study, the control group tended to surpass their pretransplant mean intake, whereas the diet group tended not to
<table>
<thead>
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<td>C (n=2)</td>
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<tr>
<td>Energy (kcal)</td>
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<td>1858</td>
<td>1528 ± 179</td>
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<td>297</td>
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<td>179</td>
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<td>Energy (% needs)</td>
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<td>101</td>
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<td></td>
<td>74.5</td>
<td>68.8</td>
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<tr>
<td>Total Fat (%)</td>
<td>33.2</td>
<td>32.4 ± 1.8</td>
<td>31.1</td>
<td>30.2 ± 2.9</td>
</tr>
<tr>
<td></td>
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<td></td>
<td>(n=2)</td>
<td>(n=5)</td>
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<tr>
<td></td>
<td>28.6</td>
<td>26.8 ± 2.8</td>
<td></td>
<td></td>
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<tr>
<td>Sat. Fat (%)</td>
<td>10.1</td>
<td>12.9 ± 1.6</td>
<td>9.8</td>
<td>11.3 ± 1.3</td>
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<tr>
<td></td>
<td>8.5</td>
<td>9.4 ± 1.4</td>
<td></td>
<td></td>
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<tr>
<td>MUFA (%)</td>
<td>11.5</td>
<td>13.4 ± 1.3</td>
<td>11.6</td>
<td>11.9 ± 1.2</td>
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<tr>
<td></td>
<td>12.2</td>
<td>12.1 ± 1.4</td>
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<td></td>
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<tr>
<td>PUFA (%)</td>
<td>11.6</td>
<td>6.1 ± 1.1</td>
<td>9.7</td>
<td>7.0 ± 1.0</td>
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<td>(n=2)</td>
<td>(n=5)</td>
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<tr>
<td></td>
<td>8.0</td>
<td>5.3 ± 0.3</td>
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<tr>
<td>Protein (%)</td>
<td>16.0</td>
<td>19.9 ± 0.9</td>
<td>15.7</td>
<td>22.9 ± 0.8</td>
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<td>(n=2)</td>
<td>(n=5)</td>
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<td></td>
<td>18.4</td>
<td>22.0 ± 2.1</td>
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<tr>
<td>Carbohydrate (%)</td>
<td>50.8</td>
<td>47.7 ± 2.1</td>
<td>53.2</td>
<td>46.9 ± 2.7</td>
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<td>(n=5)</td>
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<td></td>
<td>53.0</td>
<td>51.2 ± 3.5</td>
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<tr>
<td>Cholesterol (mg)</td>
<td>217.5</td>
<td>258.7 ± 61.7</td>
<td>201.5</td>
<td>216.6 ± 36.5</td>
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<td>(n=2)</td>
<td>(n=5)</td>
</tr>
<tr>
<td></td>
<td>210.0</td>
<td>195.8 ± 18.5</td>
<td></td>
<td></td>
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<tr>
<td>Fiber (g)</td>
<td>21</td>
<td>19.2 ± 3.6</td>
<td>19.5</td>
<td>19.2 ± 8.2</td>
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</tr>
<tr>
<td></td>
<td>21.0</td>
<td>14.9 ± 2.6</td>
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</table>

Table 9: Nutritional intakes of patients during study period. Intakes were collected prior to transplantation (pre-transplant) by diet history, and then prior to hospital discharge (time 0), 3 months post transplantation (time 3), and 6 months post transplantation (time 6), using two week food records. Each value represents the mean ± SEM. C = control group, D = diet group, sat = saturated, MUFA = monounsaturated fat, PUFA = polyunsaturated fat, % = percent.
consume as much MUFA as they did prior to transplantation. All subjects consumed mean intakes between 10-15% during the study, which represented a healthy intake.

The recommended protein intake of 15-20% of energy was maintained throughout the study within the control group, although a slight increase in protein intake was observed. The diet group showed a slight decrease in protein intake from time 0, but a slight increase from pretransplant intakes. At six months post transplantation, the diet group's mean protein intake was $22 \pm 2.1\%$ of total energy, $n=4$. The control group had a mean protein intake of $18.4\%$ of total energy.

Subjects tended to demonstrate an overall increase in CHO consumption. The control group maintained intakes above 50% of energy consumption. The diet group tended to increase their intakes from $47.7 \pm 2.1\%$ ($n=6$) prior to surgery to $51.2 \pm 3.5\%$ ($n=4$) at six months following surgery. The recommended intake was defined as greater than 50% of energy intake.

Reported dietary cholesterol intake was below 300 mg/day throughout the study for both groups which was the instructed intake. The diet group tended to lower even further their cholesterol intake throughout the study.

Mean fiber intakes were below 25 g/day among all patients throughout the study. No apparent change in fiber consumption was observed.

**Questionnaire Results**

Following the pre-pilot study, the format and wording of the questions were
subsequently modified in order to improve readability and comprehension of the questionnaire.

Pilot study-Reliability Results (n = 18):

Hunger

To determine the internal consistency of the various measures used in our questionnaire, the Cronbach’s coefficient alpha (CCA) was used. Like correlation coefficients, reliability coefficients fall within a range of 0.00 to 1.00, where 1.00 indicates perfect reliability, and 0.00 indicates no reliability. The CCA determined for hunger was 0.78 for all the variables in this measure (10A-10G). When calculating the CCA, SAS also identified the value of the CCA when each of the items is individually deleted from the analysis. Hence, item #10D was deleted since its absence increased the reliability to 0.83. Question #10F was poorly correlated with the other variables (0.29), and therefore was also deleted from the analysis, raising the reliability of this section to 0.86. Possible explanations include: question 10D was more concerned with eating rather than with overall hunger, and eating more than three times a day did not necessarily signify hunger as suggested by question 10F. Therefore, the final hunger measure consisted of questions #10A, 10B, 10C, 10E, and 10G with a CCA of 0.86.
Attitudes

The reliability coefficient for questions 11-20 was not very high, a value of only 0.60, probably due to the fact that not all the questions in this section were measuring the same thing. Therefore, a factor analysis was performed using SAS to determine which questions were common among one another. The principle factor analysis was used. The analysis demonstrated questions 11-17 to be related in nature, as they dealt with the attitude of eating as an enjoyable, nutritious, and healthy experience (when eating the right foods). However, question #17 was deleted as each respondent had the same response. Questions 19-20 were related, as they identified an attitude of guilt after the consumption of foods high in fat and cholesterol. Question #18 did not fit into either group, as it referred to the quantity (i.e. a lot of food), rather than the quality of food (i.e. high in fat and cholesterol), and therefore was indicative of a different measurement of attitude. The final reliability coefficients for questions 11-16 (attitude towards food), and questions 19-20 (attitude towards guilt) were 0.61 and 0.90, respectively.
The internal consistency coefficient of reliability was determined for each component of the health belief model:

<table>
<thead>
<tr>
<th>Question</th>
<th>CCA</th>
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<tr>
<td>perceived interest concern (#21-22)</td>
<td>0.88</td>
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<tr>
<td>perceived benefits (#23-26)</td>
<td>0.72</td>
</tr>
<tr>
<td>perceived barriers (#27-28)</td>
<td>0.18</td>
</tr>
</tbody>
</table>

Although questions #27 and #28 identified perceived barriers, i.e. difficulty in changing eating habits to control weight and blood cholesterol levels, they may not have correlated well with each other because the perception of how to control body weight and blood cholesterol levels may have differed among these patients.

**Stress and Knowledge**

The CCA determined for the stress and knowledge measures were 0.89 and 0.72 respectively. The final knowledge measure consisted of questions #36 (excluding 36A), #37 (excluding 37j and r), #39, #40, #41, #42, #43, #44, #45, and #46.

When deleting question 36A, reliability increased, as all respondents answered this
question correctly. Question 37j was deleted as there was a translational error in skim milk cheese on the French version of the questionnaire. Reliability increased with the omission of 37r (ricotta), perhaps because of its possible unfamiliarity to this patient population. Finally question 38 which asked patients to identify foods high in cholesterol showed poor reliability, probably because patients may have had difficulty differentiating between those foods containing cholesterol per se, and those that should be avoided when following a low cholesterol diet because of saturated fat content. Patients tended to circle those items that should not be consumed on a low cholesterol diet, rather than those items that are high in cholesterol per se.

Changes in Study Variables

There were no apparent changes in appetite, and attitudes toward food or guilt following transplantation. Patients in our study tended to have a good attitude toward food. Patients in all groups tended to feel guilty about the type and quantity of foods consumed following transplantation, and there was no apparent trend for this to change with time.

Figure 17 demonstrates level of hunger following transplantation in our subjects. Both control subjects, as well as one diet subject became more hungry. Three diet subjects became less hungry, and one showed no overall change in his level of hunger. There was, however, considerable interindividual variability between patients, in terms of each one's recorded rate of hunger at each time point. Patients from the pilot group (those not in the
study) tended to feel more hungry than less hungry following transplantation.

Most patients in all groups were interested in improving their eating habits, and remained interested throughout the study (Fig. 18). However, the general trend among the diet group was to become less interested. One control patient became more interested, but showed less interest than the rest. The other control patient remained very interested during the entire course of the study. One patient from the diet group who had dropped out of the study had no interest in changing his eating habits. The pilot group was similar to the study group. Changes in perceived benefit demonstrated similarity to changes in perceived interest (Fig. 19). As with the perceived interest curve, most patients in all three groups understood the usefulness and influence of improving eating habits on serum cholesterol levels and body weight. Perceived benefit for two patients of the diet group did decrease during the study, and increased for one patient in both the diet and control group. Three patients reported that changing eating habits were very beneficial throughout the entire study period. Two patients (drop-out, and pilot patient) saw no benefit in changing dietary patterns. There was variability among patients in how difficult it would be for them to change their eating habits to control serum cholesterol and body weight (Fig. 20). The majority of patients did perceive some difficulty in modifying eating patterns. The pilot group fell within the same range as the study group.

All three groups exhibited the same level of stress throughout the study. Patients rated their level of stress as moderate. Those patients who were instructed a diet did not have more stress than those not given a diet. Patients from both the diet and control group found that stress had some influence on what they ate, and that diet did have some
Figure 18: Level of perceived interest in relation to time after transplantation (Day 0, ±SEM), for the diet group (closed squares, n=5), the controls (open circles, n=2) and the pilot subjects (open squares, n=8).

MONTHS POST TRANSPLANTATION

Figure 19: Level of perceived benefit in relation to time after transplantation (Day 0, ±SEM), for the diet group (closed squares, n=5), the controls (open circles, n=2) and the pilot subjects (open squares, n=8).
MONTHS POST TRANSPLANTATION

Figure 20: Level of perceived barrier in relation to time after transplantation (Day 0, ±SEM), for the diet group (closed squares, n=5), the controls (open circles, n=2) and the pilot subjects (n=8).

MONTHS POST TRANSPLANTATION

Figure 21: Knowledge score in relation to time after transplantation (Day 0, ±SEM), for the diet group (closed squares, n=5), the controls (open circles, n=2) and the pilot subjects (open squares, n=8).
influence on their level of stress, whereas the pilot group reported little influence for these questions.

Only one patient, who was from the pilot group, reported not receiving any dietary advice from a physician, dietitian, nurse, or someone else. For the others who had received some dietary advice, two patients from the diet group felt little stress to follow the dietary advice, one felt somewhat stressed, and two felt moderately stressed by the end of the study. Their stress level did tend to increase as the study progressed. Control patients felt little or no stress following dietary advice. From the pilot group, there were mixed stress levels including moderate (2 patients), little (1 patient), or no stress (4 patients).

Four out of five patients from the diet group had improvements in their level of nutrition knowledge concerning dietary fat and cholesterol. One patient from this group remained fairly stable in terms of his knowledge base. Both patients from the control group demonstrated a decline in their level of nutrition knowledge from the start to the end of the study. By the end of the study period, the average score for the diet group was 86.7%. The control group showed a final average score of 72.2%. The pilot group exhibited the lowest mean nutrition knowledge score with respect to fat and cholesterol, with the exception of one patient (score of 86.2%). Their mean score was 60.7% (Fig.21).
Associations

Pearson correlation coefficients were computed using SAS, to determine which variables were associated with the change in body weight and serum cholesterol levels at the end of our study (Table 10). Responses from the first (time 1) and final (time 7) times at which patients completed the questionnaire were used for the analysis. We also subdivided our questionnaire into those questions (for perceived interest, perceived benefits, perceived barriers, and knowledge) pertaining to weight, and those pertaining to cholesterol. We then correlated the change in body weight at the end of our study with the mean score of questions pertaining to body weight, and the change in serum cholesterol with the mean score of questions pertaining to cholesterol (Table 10).

Despite our small sample size, certain associations were significant. There was a significant positive association between the change in body weight and perceived barriers at time 1 (0.86, \(p=0.01\)), and a significant inverse association between the change in serum cholesterol and perceived interest at time 7 (-0.79, \(p=0.04\)).

At time 0 and 7, the trend, though not significant, indicates a positive correlation, between final change in body weight and hunger (0.69, \(p=0.08\); 0.75, \(p=0.05\) respectively), a negative correlation with perceived interest (-0.44, \(p=0.32\); -0.61, \(p=0.15\) respectively), and a negative correlation with stress (-0.45, \(p=0.32\) respectively). At time 7, the change in body weight was associated with appetite (0.49, \(p=0.26\)).

At time 1 and 7 the change in serum cholesterol was positively related to hunger (0.50, \(p=0.25\); 0.75, \(p=0.05\) respectively). At time 1, the change in serum cholesterol
Table 10: Correlation coefficients between the final change in body weight (BW) and total cholesterol (TC) and questionnaire study variables. Time 0 = completion of questionnaire prior to hospital discharge; time 6 = final completion of questionnaire at 6 months after transplantation. APP. = appetite; ATT. FOOD = attitudes toward food; PER. INT. = perceived interest in changing eating habits; PER. BEN. = perceived benefits of changing eating habits; PER. BAR. = perceived barriers toward changing eating habits; KNOW. = nutrition knowledge. Just BW refers to the correlation between the final change in body weight and those questions pertaining only to body weight; Just TC refer to the correlation between the final change in total cholesterol and those questions pertaining only to total cholesterol.
was inversely related to perceived interest (-0.75, p=0.05), and tended to correlate with perceived benefits (-0.44, p=0.33), and perceived barriers (0.60, p=0.16) toward changing eating habits. At time 7, the trend suggests a direct correlation between the change in serum cholesterol and appetite (0.62, p=0.13) and an inverse association with attitudes toward food (-0.59, p=0.17), and knowledge (-0.37, p=0.41).

When questions were subdivided according to weight and cholesterol, and correlated with final change in body weight and serum cholesterol levels, perceived barriers were correlated with the change in body weight (0.42, p=0.35) as shown above, and knowledge was inversely correlated with the change in serum cholesterol (-0.63, p=0.13) as shown above, but positively associated with the change in body weight (0.61, p=0.15).

Patients were also divided according to weight gain (> or = to 5 kg or less than 5 kg) and TC change (> or = to 2.0 mmol/l or less than 2.0 mmol/l), and questionnaire items were compared between groups (Table 11). As confirmed by the correlations, those who gained more weight and who had greater elevations in serum cholesterol tended to have a greater appetite, and to be more hungry. Those who had a more favourable attitude towards food and who felt more guilty about eating foods rich in cholesterol and fat tended to gain less weight and have a lower increase in serum cholesterol levels. However only an association between the change in serum cholesterol and attitudes toward food was found. Those slightly less interested in changing eating habits, and who perceived more difficulties in changing eating habits tended to gain more weight and show greater elevations in serum cholesterol. Nutrition knowledge tended to be similar
Table 11: Mean score for each of the questionnaire items (see Appendix), divided according to weight and total cholesterol changes after transplantation (n=7). Weight change is divided according to those who gained more than 5 kg (n=2), and those who gained less than 5 kg (n=5) after transplantation. Total cholesterol change is divided according to those who had an increase of greater than 2.0 mmol/l (n=4), and those with an increase less than 2.0 mmol/l after transplantation (n=3). A value close to 1 indicates a low value, and a value close to 5 or 7 depending on scale (see Appendix) indicates a high value.

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<th>CHOLESTEROL CHANGE</th>
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<td>&lt; 5.0 kg</td>
<td>&gt; 2.0 mmol/l</td>
<td>&lt; 2.0 mmol/l</td>
</tr>
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<td>3.5</td>
<td>2.3</td>
</tr>
<tr>
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<td>1.7</td>
<td>3.5</td>
<td>1.5</td>
</tr>
<tr>
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<td>5.1</td>
<td>4.7</td>
<td>5.4</td>
</tr>
<tr>
<td>Attitude (guilt)</td>
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<td>5.7</td>
<td>4.7</td>
<td>5.5</td>
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<td>3.8</td>
<td>4.2</td>
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<td>4.7</td>
<td>4.6</td>
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<td>Knowledge</td>
<td>84.4%</td>
<td>81.8%</td>
<td>82.3%</td>
<td>82.9%</td>
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</table>
among all groups.
DISCUSSION

This study was a clinical intervention study designed to determine whether the Step One lipid lowering diet is an effective method to prevent or minimize post heart transplantation hyperlipidemia and weight gain. Seven patients completed the study. Five were randomly assigned to the diet group, in which they received individual nutritional instruction and education pertaining to the Step One diet, and two were assigned to the control group whereby no dietary instruction was given.

As a result of small sample size, and uneven distribution of patients within each group, the use of statistical analyses to compare the two groups was limited, and conclusions on the efficacy of the diet are difficult. It was therefore decided to evaluate this study as a descriptive one rather than a comparison between treatment and control groups. Any conclusions made from the results are suggestive.

To our knowledge, this was the first randomized study to examine the effects of diet on serum lipid levels and body weight in this patient population. One hospital reported that a nutritional program was effective in reducing weight and arteriosclerotic complications following cardiac transplantation, however an actual study was not performed (Ragsdale, 1987). Two other retrospective studies found no correlation between dietary fat and cholesterol intake and levels of TC, LDL-C, or TG, and only a marginal significant positive association between weight change and fat intake at the time of study, but not at 1, 2, and 3 years post transplantation (Becker et al, 1988; Rubin et al, 1991 respectively). These studies were performed retrospectively. Both groups in our
study had an overall increase in serum cholesterol concentrations. The diet group did tend to have the lowest mean overall increase by the end of the study (0.7 ± 0.4 mmol/l), compared to the control group (1.2 mmol/l), and both groups demonstrated a lower overall increase in serum cholesterol concentrations than that observed in our retrospective study (2.1 ± 1.3 mmol/l). These results suggest that dietary intervention may be somewhat beneficial in minimizing serum cholesterol concentrations after heart transplantation. The similar order of increase observed between the diet and control group may be associated with the fact that both groups reported dietary intakes in accordance with the Step One Lipid Lowering diet. Despite limited nutritional counselling among the control group, they managed to reduce total fat and saturated fat intakes, and consumed desirable amounts of complex carbohydrates and dietary cholesterol. In addition, the questionnaire did show evidence that the control group (those in the study) had greater nutrition knowledge than the pilot group (those not in the study) expressing differences between those in the study and those not in the study. Furthermore, they were instructed to complete food diaries like the diet group, and their body weights, waist-hip ratios, and body composition were measured monthly like the diet group, potentially making them aware of their eating habits. One control patient received instruction from his physician to lose weight, and hence the patient on his own was watching the quantity and quality of foods he consumed regardless of the limited dietary teaching given. Similarly, patients in both groups also gained weight, although the diet group tended to show less of a weight gain. The diet group had a mean overall weight gain of 4.2 ± 2.3 kg at seven months post transplantation, as compared to the control group who had a mean overall
gain of 5.7 kg. These weight changes were similar to what we observed in our retrospective study. Patients from the retrospective study demonstrated a significant increase in body weight of 6.0 ± 1.1 kg during the first 7 months post transplantation (Table 7). The large increase in body weight, followed by a large decrease in body weight soon after transplantation, observed among the control group may be a reflection of changes in hydration status postoperatively.

These results suggest that dietary intervention does not minimize post transplantation weight gain. Ironically, weight gain occurred despite a reported decrease in energy intake, an intake below estimated energy requirements, and an increase in activity level among both groups. However, prior to transplantation, patients were too sick to complete two consecutive weeks of food diaries. Dietary information was therefore obtained using two different methods: 1) diet history and 2) usual intake, and hence may explain the large decrease in energy intake observed between pre and post transplant timepoints. Another potential explanation is inaccurate recording of food items consumed in food records. Food diary information confirmed similarities between the diet and control group with respect to dietary intake, which may explain why little difference was observed between groups. Patients of the control group were seen regularly by their physicians where weight loss was discussed and patients were conscious of their intakes and weight from follow-up visits with the dietitian (investigator). Patients from both groups showed an overall decrease in both total fat and saturated fat consumption. The recommended level of consumption of less than 30% of energy from total fat and between 7-10% from saturated fat was achieved for both groups at the end of the study.
Despite nutritional education to maintain PUFA intakes at 10% of energy, the diet group consumed less PUFA than their recommended diets allowed. This was probably a response to their efforts to lower total fat intakes. The control group showed a similar response, and consumed similar amounts during the study. No differences between groups with regard to MUFA intake were observed and intakes were within the recommended range. Dietary recommendations for protein and carbohydrate intakes were achieved for the control group despite only general nutritional counselling, and no teaching with regards to a special diet. This however was expected since intakes prior to transplantation were already within the Step One dietary guidelines. The diet group improved their overall intake of complex carbohydrate to greater than 50% of overall intake. Overall protein intake was above the recommended level. Dietary cholesterol intakes were below 300 mg/day for both groups possibly a reflection of recommended dietary fat and complex carbohydrate intakes. No improvements were shown with fiber consumption as nutritional intervention did not primarily focus on this area.

Both the diet and control group tended to demonstrate increases in HDL-C concentrations. LDL-C tended to decrease. Triglycerides, ApoA1, and ApoB tended to increase. Lp(a) tended to decrease among the diet group, and increase among the control group. Significant elevations in LDL-C, and APOB levels have been reported (Stamler et al, 1988; Becker et al, 1988; Bilodeau et al, 1989; Grady et al 1991; Farmer et al, 1991). Increases in TC levels have been demonstrated to be predominantly a result of significant increases in LDL-C (approximately 150%) (Stamler et al, 1988; Becker et al, 1988), which contrasts our results. This is probably a result of small sample size.
Increases in HDL-C and APOA1 are consistent with the literature (Atger et al, 1990). Elevations in HDL-C following transplantation have been associated with CsA use (Mraz et al, 1986), and corticosteroid therapy (Becker et al, 1988). Transplanted patients were found to have a HDL profile different from that of matched controls, with a predominance of HDL2, which has been demonstrated to be negatively correlated with coronary artery disease (Atger et al, 1990). There were no apparent differences between the diet and control group with respect to changes in HDL-C profile.

Elevated levels of fibrinogen and Lp(a) are correlated with coronary atherosclerosis (Hunt et al, 1991; Farmer et al, 1991). The present investigation demonstrated great variability among fibrinogen and Lp(a) concentrations, and trends could not be established as a result of small sample size.

We hypothesized that any increase in body weight is actually a result of an increase in body fat. Weight gain post transplantation has been represented as a return to premorbid levels (Rubin et al, 1991). Through bio-impedence analysis, we recorded changes in LBM, body water, and body fat monthly for seven months. Levels of both LBM and body water tended to decline following transplantation despite weight gain, confirming our hypothesis that weight gain post transplantation is related to an increase in body fat. One explanation for the decrease in LBM and concomitant increase in body fat may be related to prednisone therapy. Corticosteroids reduce the utilization of amino acids for protein synthesis everywhere except in the liver (Griffin and Ojeda, 1992). Extrahepatic protein stores are reduced, and blood amino acid levels increase. Amino acids are not transported into muscle cells, decreasing protein synthesis. With respect to
fat metabolism, corticosteroids stimulate appetite and the laying down of additional fat in the central or truncal areas (Griffin and Ojeda, 1992).

Resistance is affected by agents or actions that change total body water, such as diuretics, alcohol consumption, the presence of edema, ascites, congestive heart failure, or exercise that causes sweating or dehydration. Patients with a higher proportion of body water will show a lower proportion of body fat (Brylowski, 1992). In normal, healthy individuals, BIA has been well correlated with total body water, LBM, and total body potassium. However, body composition, total body water, and intracellular/extracellular fluid distribution change with disease and nutritional status. We realize that the use of BIA in the heart transplant population may have been somewhat inaccurate as a result of prednisone and diuretic therapies, as well as the presence of congestive heart failure prior to transplantation. We hoped to overcome such discrepancies with the frequency at which measurements were taken, to obtain accurate, overall, mean values. Patients however, may have had varying degrees of hydration which could have affected our data. BIA has been demonstrated to estimate abnormal fluid retention and total body water changes, and therefore has been applied to measure the hydration status of burn, dialysis, cardiac, and critical ill patients (Brylowski, 1992). Further studies using the BIA system among the heart transplant population is required.

One patient from the control group, and two from the diet group developed waist:hip ratios above 1.0 (below which is considered healthy for males) during the study period. All patients were male and gained most of their weight in the truncal area. The WHR has been reported to be a good determinant of lipid profiles, and links between truncal fat and

A secondary hypothesis of our study was to examine whether patients with a greater appetite following transplantation, patients with a less favorable attitude towards food, patients with a lower degree of perceived interest concern and perceived benefits, patients with a greater degree of perceived barriers, patients with more stress, and patients with less nutrition knowledge would be more likely to have a higher weight gain and higher serum cholesterol levels following cardiac transplantation.

Patients participating in the study shared similar responses to those not in the study and hence were representative of other heart transplant populations. Most patients exhibited no dramatic change in appetite during the study period. This was somewhat unexpected considering an improved health status from pretransplant condition, and the effect of prednisone on increasing appetite (Funk, 1986). However as has been reported by other heart transplant recipients, improvement is contingent upon the degree of treatment-induced symptoms and overall life change (Hook et al, 1990). Both the social and psychosocial consequences of cardiac transplantation were not addressed in this study but could have played a significant role in influencing changes in appetite. A greater appetite was correlated with a higher weight gain and greater serum cholesterol elevations at the end of the study. Although the correlation coefficients were not statistically significant, likely due to the small sample size, the results suggest that appetite is associated with post transplant elevations in weight and serum cholesterol. There was no
specific trend in hunger among the patients in our study. Some showed an increased level whereas others showed a decreased level of hunger. There was considerable variability in the level of hunger reported among patients. Non-participants of the study tended to be more hungry following transplantation. An increased hunger following transplantation could be the result of an improved cardiac and nutritional status, as well as an increase in activity level. A decrease in hunger could be related to psychosocial factors not investigated in this study or to the incidence of rejection (not investigated in this study). Hunger was also found to be associated with greater weight gain and greater serum cholesterol concentrations at the end of the study, confirming our hypothesis.

Most patients had a favorable attitude toward food after surgery, suggesting a joy for eating, and an interest in eating nutritious foods for good health. Most also indicated a feeling of guilt after the consumption of foods high in fat and cholesterol also suggesting an awareness for a healthier lifestyle. A greater change in serum cholesterol levels was associated with a less favourable attitude towards food at the end of the study, which confirmed, in part, our hypothesis. Attitude towards food however, was not associated with changes in body weight. Furthermore, patients who felt a greater sense of guilt when eating fattening foods, tended to gain less weight and have lower elevations in serum cholesterol post transplantation. Perhaps individuals who felt a greater sense of guilt when eating fattening foods were more conscious about their eating habits.

Questions based on the health-belief model demonstrated that heart transplant patients do recognize the benefits of healthy eating habits on serum cholesterol levels and maintaining healthy body weights, and most demonstrated an interest in wanting to
improve eating habits. Perceived interest in changing dietary behaviours was negatively correlated with changes in body weight (non-significantly) and serum cholesterol (significantly), suggesting that those patients interested in changing their eating habits tended to gain less weight (or lose weight), and have lower elevations in serum cholesterol compared to those not interested, supporting in part the study hypothesis. These results are similar to Strychar's et al (1993) findings whereby perceived interest was associated with decreased frequency in consumption of high fat meat products, sauces, gravies, and snack foods such as potato chips, all food items which can potentially increase serum cholesterol levels.

Patients who perceived benefits in changing in eating habits (time 0) tended to have lower elevations in serum cholesterol levels, suggesting that the understanding of the benefits associated with changing behavior can succeed in positive outcomes. This, in part, supported our hypothesis. However, this association occurred only at the beginning of our study suggesting that perceived benefits may not always be predictive of changes in serum cholesterol or perhaps the sample size was too small. It’s important to note however, that perceived benefits of changing eating habits to influence serum cholesterol levels only (not body weight) were not correlated with the change in serum cholesterol, possibly as a result of small sample size.

Most patients perceived some difficulty in making the appropriate dietary modifications. Those who perceived more difficulty in changing eating habits to control weight tended to have greater elevations in weight and those who perceived more difficulty in changing eating habits to control serum cholesterol tended to have greater
elevations in serum cholesterol, which again confirmed our hypothesis.

The health-belief model is used as a psychosocial approach to explain health-related behavior (Rosenstock, 1990). The above associations suggest that health beliefs are associated with behaviour change among patients following cardiac transplant surgery, and should be considered if nutritional counselling with regards to body weight and cholesterol is to be effective. Further research is required with a larger sample size as many of the correlation coefficients are not statistically significant.

Patients of the diet group responded as having a similar level of stress to patients of the control group. Contributing factors to stress level may have included frequency and severity of rejection episodes, home situation, lifestyle modification, and the emotional, and physical side effects of immunosuppressive therapy (Hook et al., 1990). Patients of the diet group who were feeling stressed as a result of trying to comply to dietary recommendations tended to display more stress as the study progressed suggesting that long-term changes of familiar and customary eating patterns is difficult. Stress tended to be inversely related to body weight, suggesting that a higher stress level produced less weight gain.

Nutritional counselling and education regarding fat and cholesterol had a beneficial effect in improving the knowledge base of patients in the diet group. There was an obvious difference between diet and control subjects in terms of nutrition knowledge. Patients of the control group demonstrated a decrease in their degree of knowledge most probably related to the fact that some initial nutritional guidelines were given but these were not followed-up. Patients of the diet group were counselled and educated monthly.
Patients receiving no nutrition education (pilot group) scored the lowest. By the end of the study, greater nutrition knowledge was related, although not significantly, to smaller elevations in serum cholesterol, suggesting that nutrition education and follow-up may be beneficial in minimizing post transplant hypercholesterolemia. This supported our hypothesis. The effectiveness of cholesterol education programs in improving nutrition knowledge and dietary behavior has been demonstrated in other investigations related to cardiovascular disease (Greene and Strychar, 1992). No association was observed between knowledge and changes in body weight, likely due to the fact that the majority of questions were based on dietary cholesterol and fat knowledge rather than knowledge required to change body weight.

The aforementioned associations, and the obvious improvement in nutrition knowledge among the diet group reveal the need for more clinical trials to explore the effect of diet on post transplantation hyperlipidemia and weight gain.
Conclusions

As a result of small sample size, it is difficult to make meaningful conclusions as to the effect of our dietary intervention on post transplant hyperlipidemia and weight gain. Upon comparison of our study group to the group of patients from our retrospective study, there is evidence to suggest that a study effect may have existed. The smaller increase in serum cholesterol observed among our intervention study group as compared to patients from our retrospective study suggests that dietary intervention, may have had a beneficial effect in minimizing post transplant hypercholesterolemia. Patients of the control group did show similar dietary intakes to patients of the diet group, and this contributed to the similar outcomes.

The overall increases in HDL-C, TG, APOB, and APOA1 concentrations confirm what has been observed in the literature. No specific trends with respect to diet can be made.

Weight gain post transplantation was observed to be due to an increase in body fat, confirming our hypothesis. However, as a result of varying degrees of hydration status among this population, results may not be completely accurate. Patients in the diet group clearly had an improvement with respect to their nutrition knowledge base, and demonstrated the highest scores on the nutrition knowledge questions. Greater nutrition knowledge, a favourable attitude towards food, a greater degree of perceived interest in wanting to improve eating habits, a greater degree of perceived benefits in changing eating habits, and fewer perceived barriers to changing eating habits tended to be related
to lower serum cholesterol elevations. However, knowledge, attitude towards food, perceived interest, and perceived benefits were not found to be associated with lower weight gain following transplant surgery. Individuals who perceived less barriers to changing their eating habits to control their weight, actually tended to gain less weight following transplant surgery, as postulated according to the Health Belief Model. Changes in body weight and serum cholesterol concentrations were also associated with appetite and hunger. Stress was also related to changes in body weight.
FINAL CONCLUSIONS

This thesis describes a time-course of hypercholesterolemia and weight gain post heart transplantation, identifies potential factors involved in its pathogenesis, and attempts to identify an effective intervention program to minimize or prevent the observed changes in body weight and lipid levels. Our results from the retrospective study confirmed that following cardiac transplantation, serum cholesterol concentrations become abnormally elevated, accompanied by a progressive increase in body weight. Serum cholesterol levels respond well to decreases in prednisone dose, whereas body weight responds to diet.

It is difficult to draw firm conclusions on the effectiveness of our intervention study which attempted to evaluate whether a lipid-lowering diet would prevent elevations in body weight and lipid levels post transplantation. The principle limitation was our smaller than anticipated sample size. As was reported in the retrospective study, body weight responds to the prescription and monitoring of a specific diet. This however was not observed in our intervention study which may have been a result of small sample size.

According to food records, patients in both the diet and control group were following the Step One Lipid-Lowering diet, and tended to show less dramatic elevations in serum cholesterol concentrations post transplantation. This suggests a potentially beneficial effect of diet in minimizing post transplantation hypercholesterolemia. Lipid levels as well as body weight may not respond very well to diet post transplantation because of the use of immunosuppressants.

Increases in body weight were related to increases in body fat. Levels of LBM and
Total body water tended to decrease post transplantation.

Changes in body weight and serum cholesterol levels were correlated with appetite, hunger, and perceived barriers toward changing eating habits. Perceived interest in changing eating habits and perceived benefits of changing eating habits, attitudes toward food and nutrition knowledge were related to changes in serum cholesterol. Increased stress was associated with less weight gain. Patients in the diet group demonstrated improvements in their level of nutrition knowledge, which correlated with a more favourable attitude towards food.

This study therefore demonstrated a possible beneficial effect of diet in reducing hypercholesterolemia post transplantation. It further demonstrated that nutrition counselling and education will improve nutrition knowledge, which tends to correlate with a smaller increase in serum cholesterol levels. Further clinical, randomized, controlled, dietary intervention trials are therefore warranted to evaluate the effect of diet and improved nutrition knowledge on post transplant hyperlipidemia and weight gain, in an effort to help reduce the incidence of graft atherosclerosis, and thereby hopefully increase patient survival.
REFERENCES


Canadian Consensus Conference on Cholesterol. Meeting Report (July 1988) Rapport. 3(3). Suppl to the CMAJ.


APPENDIX
APPETITE

The following will ask you questions about your appetite DURING THE LAST MONTH.

1. Overall, within the past month, how would you rate your appetite (desire for food)? Please circle your answer.

<table>
<thead>
<tr>
<th></th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Very Poor</td>
<td>Poor</td>
<td>Fair</td>
<td>Good</td>
<td>Very Good</td>
</tr>
</tbody>
</table>

2. Within the past month, how would you rate your appetite during the following eating times? Please circle your answer.

<table>
<thead>
<tr>
<th></th>
<th>Very Poor</th>
<th>Poor</th>
<th>Fair</th>
<th>Good</th>
<th>Very Good</th>
</tr>
</thead>
<tbody>
<tr>
<td>a)</td>
<td>breakfast</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>b)</td>
<td>morning snack</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>c)</td>
<td>lunch</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>d)</td>
<td>afternoon snack</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>e)</td>
<td>supper</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>f)</td>
<td>evening snack</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
</tbody>
</table>

3. Within the past month, how would you rate your appetite during the following situations? Please circle your answer.

<table>
<thead>
<tr>
<th></th>
<th>Very Poor</th>
<th>Poor</th>
<th>Fair</th>
<th>Good</th>
<th>Very Good</th>
</tr>
</thead>
<tbody>
<tr>
<td>a)</td>
<td>when you are anxious</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>b)</td>
<td>when you are bored</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>c)</td>
<td>when you are alone</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>d)</td>
<td>when you are with friends</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>e)</td>
<td>when you are with family members</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>f)</td>
<td>when you are at a social occasion (i.e. party)</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
</tbody>
</table>
4. Within the past month, how would you rate your appetite for the following food items? Please circle your answer.

<table>
<thead>
<tr>
<th></th>
<th>Very Poor</th>
<th>Poor</th>
<th>Fair</th>
<th>Good</th>
<th>Very Good</th>
</tr>
</thead>
<tbody>
<tr>
<td>a) Desserts</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>pastries, pies, cookies, cake</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>chocolates, candies</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>ice-cream</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>b) Snack Foods</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>chips, pretzels, cheese snacks</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>c) Fried foods</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>d) Starches</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>bread, cereals, pasta</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>e) Milk Products</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>f) Meat, fish, poultry</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>g) Fruits</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>h) Vegetables</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
</tbody>
</table>

The following asks you to rate your appetite TODAY.

5. FOR TODAY, rate your appetite for the following food items. Please circle your answer.

<table>
<thead>
<tr>
<th></th>
<th>Very Poor</th>
<th>Poor</th>
<th>Fair</th>
<th>Good</th>
<th>Very Good</th>
</tr>
</thead>
<tbody>
<tr>
<td>a) Desserts</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>pastries, pies, cookies, cake</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>chocolates, candies</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>ice-cream</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>b) Snack Foods</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>chips, pretzels, cheese snacks</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>c) Fried foods</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>d) Starches</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>bread, cereals, pasta</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>e) Milk Products</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>f) Meat, fish, poultry</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>g) Fruits</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>h) Vegetables</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
</tr>
</tbody>
</table>
The following will ask you questions about your appetite SINCE YOUR HEART TRANSPLANT.

6. Overall, has your appetite changed since your heart transplant? Please circle your answer.

a) ________________
   
   1  2  3  4  5  6  7
   decreased decreased decreased neither increased increased increased
decreased moderately a little decreased moderately increased
decreased a great deal

b) Why has your appetite changed? (If applicable). Place an X next to your answer.

   _______ more hungry
   _______ food tastes better
   _______ increased physical activity
   _______ feel healthier
   _______ eating is more pleasurable

   _______ less hungry
   _______ food tastes worse
   _______ decreased physical activity
   _______ feel less healthy
   _______ eating is less pleasurable

   Other reasons: ________________________________
7. For the following eating times, indicate whether your appetite has changed since heart transplant. Please circle your answer.

<table>
<thead>
<tr>
<th></th>
<th>decreased a great deal</th>
<th>decreased moderately</th>
<th>decreased a little</th>
<th>neither decreased nor increased</th>
<th>increased a little</th>
<th>increased moderately</th>
<th>increased a great deal</th>
</tr>
</thead>
<tbody>
<tr>
<td>a)</td>
<td>breakfast</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>b)</td>
<td>morning snack</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>c)</td>
<td>lunch</td>
<td>1</td>
<td>2</td>
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<td>4</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>d)</td>
<td>afternoon snack</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>e)</td>
<td>supper</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>f)</td>
<td>evening snack</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
<td>6</td>
</tr>
</tbody>
</table>

8. For the following situations, indicate whether your appetite has changed since your heart transplant. Please circle your answer.

<table>
<thead>
<tr>
<th></th>
<th>decreased a great deal</th>
<th>decreased moderately</th>
<th>decreased a little</th>
<th>neither decreased nor increased</th>
<th>increased a little</th>
<th>increased moderately</th>
<th>increased a great deal</th>
</tr>
</thead>
<tbody>
<tr>
<td>a)</td>
<td>when you are anxious</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>b)</td>
<td>when you are bored</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>c)</td>
<td>when you are alone</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>d)</td>
<td>when you are with friends</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>e)</td>
<td>when you are with family members</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>f)</td>
<td>when you are at a social occasion (e.g. party)</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
<td>6</td>
</tr>
</tbody>
</table>
9. For the following foods, indicate whether your appetite has changed since your heart transplant. Please circle your answer.

<table>
<thead>
<tr>
<th></th>
<th>decreased a great deal</th>
<th>decreased moderately</th>
<th>decreased a little</th>
<th>neither decreased nor increased</th>
<th>increased a little</th>
<th>increased moderately</th>
<th>increased a great deal</th>
</tr>
</thead>
<tbody>
<tr>
<td>Desserts</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>a) Pastries, pies, cookies, cakes</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>b) Snack Foods</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>c) Fried Foods</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>d) Starches</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>e) Dairy</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>f) Meats, fish, poultry</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>g) Fruits</td>
<td></td>
<td></td>
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<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>h) Vegetables</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

HUNGER

10. Please indicate how you feel about the following statements. Please circle your answer.

<table>
<thead>
<tr>
<th></th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
</tr>
</thead>
<tbody>
<tr>
<td>a) Sometimes things just taste so good that I keep on eating even when I am no longer hungry</td>
<td>strongly agree</td>
<td>moderately agree</td>
<td>slightly agree</td>
<td>neither agree nor disagree</td>
<td>slightly disagree</td>
<td>moderately disagree</td>
<td>strongly disagree</td>
</tr>
<tr>
<td>b) When I see a real delicacy I often get so hungry that I have to eat it right away</td>
<td>strongly agree</td>
<td>moderately agree</td>
<td>slightly agree</td>
<td>neither agree nor disagree</td>
<td>slightly disagree</td>
<td>moderately disagree</td>
<td>strongly disagree</td>
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</tbody>
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I am always hungry enough to eat at any time.

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d) I eat anything I want at anytime I want.

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e) I am always so hungry that it is difficult for me to stop eating before I finish the food on my plate.

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f) I am usually so hungry that I eat more than 3 times a day.

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g) Following a diet is so difficult because I just get too hungry.

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ATTITUDES

PLEASE CIRCLE YOUR RESPONSE FOR THE FOLLOWING STATEMENTS:

11 Eating is an Important activity in my life.

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12 Eating is an enjoyable experience.

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13 Eating the foods I like is an enjoyable experience.

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Eating nutritious foods is something I am very conscious of.

15. Eating nutritious foods makes me feel good.

16. Eating nutritious foods is a regular part of my diet.

17. Eating nutritious foods is good for my health.

18. Eating a lot of food makes me feel guilty.

19. Eating fattening foods makes me feel guilty.

20. Eating foods high in cholesterol makes me feel guilty.

BELIEFS

21. How interested are you in improving your eating habits?

22. How interested are you in learning ways to improve your eating habits?
23. How useful do you think improving your eating habits would be in maintaining your weight?

1. not useful 2. a little useful 3. somewhat useful 4. moderately useful 5. very useful

24. How useful do you think improving your eating habits would be in maintaining a normal blood cholesterol level?

1. not useful 2. a little useful 3. somewhat useful 4. moderately useful 5. very useful

25. How much do you think your eating habits influence your weight?

1. no influence 2. little influence 3. some influence 4. moderate influence 5. great influence

26. How much do you think your eating habits influence your blood cholesterol level?

1. no influence 2. little influence 3. some influence 4. moderate influence 5. great influence

27. How difficult would it be for you to change your eating habits to control your weight?

1. not difficult 2. little difficult 3. somewhat difficult 4. moderately difficult 5. very difficult

28. How difficult would it be for you to change your eating habits to control your blood cholesterol level?

1. not difficult 2. little difficult 3. somewhat difficult 4. moderately difficult 5. very difficult

STRESS

29. Overall, within the past month, how would you rate your level of stress? Please circle your answer.

1. not stressed 2. a little stressed 3. somewhat stressed 4. moderately stressed 5. very stressed

30. When you feel stressed, does it influence what you eat?

1. no influence 2. a little influence 3. some influence 4. moderate influence 5. great influence
31. How much does your diet influence your level of stress?

1  2  3  4  5
no a little some moderate great
Influence Influence Influence Influence Influence

32. Did you receive dietary advice from your doctor? Please place an X next to your answer.

a) No _______
    Yes _______

b) If yes, what kind of advice?
   ____________________________________________________________
   ____________________________________________________________
   ____________________________________________________________
   ____________________________________________________________
   ____________________________________________________________

33. Did you receive dietary advice from a nurse?

a) No _______
    Yes _______

b) If yes, what kind of advice?
   ____________________________________________________________
   ____________________________________________________________
   ____________________________________________________________
   ____________________________________________________________
   ____________________________________________________________

34. If yes, were you given a diet?

a) No _______
    Yes _______

b) If yes, what kind?
   ____________________________________________________________
   ____________________________________________________________
   ____________________________________________________________
   ____________________________________________________________
   ____________________________________________________________

35. If yes, how stressful was it for you to follow the dietary advice given by your doctor? Please circle your answer.

1  2  3  4  5
not somewhat moderately very
stressful stressful stressful stressful stressful

36. How many calories?
   ____________________________________________________________
   ____________________________________________________________
   ____________________________________________________________
   ____________________________________________________________
   ____________________________________________________________
34. a) Did you receive dietary advice from a dietician? 
   No ________
   Yes ________

b) If yes, what kind of advice? ____________________________

35. a) Did you receive dietary advice from anyone else? 
   No ________
   Yes ________

b) If yes, who gave you this advice? _________________________

c) If yes, what kind of advice? ____________________________

34. c) If yes, how stressful was it for you to follow the dietary advice given by the dietician? 
   1 not stressful  2 a little stressful  3 somewhat stressful  4 moderately stressful  5 very stressful

35. c) If yes, were you given a diet? ____________________________
   If so, what kind? ____________________________
   How many calories? ____________________________
e) If yes, how stressful was it for you to follow this dietary advice?

<table>
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<tr>
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<th>2 a little stressful</th>
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KNOWLEDGE

For each statement below, circle True, False or Don't Know.

36.

a) The most effective way to lower blood cholesterol levels is to eat less foods which contain a lot of fat and a lot of cholesterol.

<table>
<thead>
<tr>
<th></th>
<th>True</th>
<th>False</th>
<th>Don't Know</th>
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<tbody>
<tr>
<td>1</td>
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b) Eating fried foods is a good way to lower blood cholesterol levels.

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c) Polyunsaturated fats help to lower blood cholesterol levels.

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d) Monounsaturated fats help to lower blood cholesterol levels.

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e) Saturated fats help to lower blood cholesterol levels.

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37.

Which of the following foods contain a lot of fat? Circle all that apply.

a) whole milk
b) margarine
c) sausages
d) ice-cream
e) cheddar cheese
f) lean ground beef
g) butter
h) white chicken meat
i) chocolate
j) skin milk cheese
k) cream
l) shortening
m) fish
n) pasta
o) bread
p) jelly
q) honey
r) ricotta
38 Which of the following foods contain a lot of cholesterol? Circle all that apply.

a) egg yolk
b) butter
c) skim milk cheese
d) cream cheese
e) liver
f) margarine
g) shrimps
h) shortening
i) shrimp
j) mayonnaise
k) nuts
l) vegetable oil
m) egg whites
n) butter

39 Have you heard about saturated fat?

a) No ________
   Yes ________

b) If yes, which of the following foods contain a lot of saturated fat?
   Please circle your answers.

a) lard
b) corn oil
c) butter
d) cream
e) shortening
f) olive oil
g) canola oil
h) soft margarine
i) fish
j) peanut oil
k) avocados
l) whole milk
m) sunflower oil
n) soybean oil
o) olives
p) red meat
q) cheese

40 Have you heard about polyunsaturated fat?

a) No ________
   Yes ________

b) If yes, which of the following foods contain a lot of polyunsaturated fat?
   Please circle your answers.

a) lard
b) corn oil
c) butter
d) cream
e) shortening
f) olive oil
g) canola oil
h) soft margarine
i) fish
j) peanut oil
k) avocados
l) whole milk
m) sunflower oil
n) soybean oil
o) olives
p) red meat
41. Have you heard about monounsaturated fat?
   a) Yes ____
   b) No ____

41. b) If yes, which of the following foods contain alot of monounsaturated fat? Please circle your answers.
   a) lard  b) corn oil  c) butter  d) cream
   e) shortening  f) olive oil  g) canola oil  h) soft margarine
   i) fish  j) peanut oil  k) avocados  l) whole milk
   m) sunflower oil  n) soybean oil  o) olives  p) red meat
   q) cheese

42. What is a low fat yogurt? Indicate the percentage of milk fat.

43. What is a low fat cheese? Indicate the percentage of milk fat.

44. On a low cholesterol diet, how often can you have:
   a) shrimps? ________
   b) egg yolk? ________

45. Which of the following ways can help you to control your weight? Circle all that apply.
   a) kind of foods consumed
   b) amount of food consumed
   c) exercise
   d) cooking methods
   e) skipping meals
   f) eating during other activities, i.e., television, reading, etc
46. Which of the following ways can help you to control your blood cholesterol levels? Circle all that apply.

a) kind of foods consumed
b) amount of food consumed
c) exercise
d) cooking methods
e) skipping meals