ALTERED FEEDING PATTERN: ITS EFFECT ON SELECTED BLOOD LIPIDS IN HUMANS

by

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(ABSTRACT)

Seventeen males participated in an investigation comparing the effects of a three meal per day feeding pattern versus a six meal per day feeding pattern on a serum lipid profile. During a two week acclimation period three meals per day were consumed by all subjects. The subjects were then divided into two groups. Group I consumed three meals per day for six weeks while group II consumed six meals per day for the same amount of time. The two groups then reversed feeding patterns for an additional six week period. Subjects completed five four-day food records which were analyzed qualitatively. Three of the four-day food records for each subject were hand coded and computer processed for kilocalorie, total fat, saturated fat, and cholesterol intake. Mean intake of cholesterol was found to be within a desirable range. Mean intake of total fat and saturated fat were noted as exceeding desirable values.

Blood samples were collected from each subject following a two week acclimation period and following each of the two six week feeding phases. The samples were analyzed for total cholesterol, triglyceride, LDL-cholesterol, and HDL-cholesterol levels. There was no statistically

significant difference found between the consumption of three meals per day and six meals per day for the lipid parameters of total cholesterol and LDL-cholesterol. There was a significant decrease in triglyceride level with the six meal per day feeding pattern with one group but not the other. For both groups there was a significant difference ($p \leq 0.05$) in HDL-cholesterol levels between the six meal per day and the three meal per day feeding patterns. However, the findings were dissimilar in that for Group I the HDL-cholesterol was higher after the six meal pattern as opposed to the three meal pattern, and for Group II, the opposite was true.

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TABLE OF CONTENTS

TABLE OF CONTENTS
(Contents)

LIST OF TABLES

LIST OF FIGURES

CHAPTER I

INTRODUCTION

Coronary heart disease remains the major cause of death and disability in the United States despite a recent decline in the incidence of mortality associated with it. It accounts for more deaths annually than any other disease, including the combination of all forms of cancer (1). Coronary heart disease ranks first as a cause for social security disabilities, and second only to arthritis for limitation of activity (2). Coronary heart disease cost the United States more than 60 billion dollars every year in direct health care costs as well as cost of compensatory wages and decreased productivity (2).

Because of the enormous burden placed on society by coronary heart disease, its possible prevention has been the subject of many studies. Multiple interrelated factors have been shown to play a role in the development of the disease. Of the identified risk factors, one of the strongest relationships is found between lipid levels and coronary heart disease (2).

The Nutrition Committee of the American Heart Association cautioned in 1978 (3),

> It appears prudent for the American people to follow a diet aimed at lowering serum lipid concentrations. There is substantial evidence that the diets recommended . . . will aid in the control of serum lipid levels in man. Diets similar to those recommended here have been consumed by many persons in the United States for periods of more than 15 years without any evidence of harmful effects.

Limited research is available regarding the effect of altered feeding frequency on selected serum lipids in humans in a free living situations who are following the dietary guidelines established by the American Heart Association. Thus additional data would be useful as a base for preventative counseling of those at risk for coronary heart disease. In light of this perceived need, this research project was undertaken. The specific purpose of this study was to investigate the effect of altered feeding patterns on selected serum lipids of humans who are at risk for coronary heart disease or have been diagnosed as having the disease.

A group of participants in the Cardiac and Intervention Program at Virginia Polytechnic Institute and State University were recruited and counseled extensively on the dietary guidelines of the American Heart Association and the feeding pattern protocol. Each subject participated in a two week acclimation phase, a six week three meal per day feeding phase, and a six week six meal per day feeding phase. Four-day food records were completed by each subject five times throughout the study phases. The food records were assessed qualitatively to give some indication of the degree of compliance with the established dietary guidelines and meal patterns and were used in ongoing counseling of the subjects. Three sets of the food records were analyzed by computer for kilocalorie, total fat, saturated fat, and cholesterol level. Correlations between meal pattern and the serum lipid parameters of total cholesterol triglyceride, LDL-cholesterol, and HDL-cholesterol as measured at the conclusion of each phase were examined.

CHAPTER II

REVIEW OF LITERATURE

Blood Lipid Levels Associated with Coronary Heart Disease

Cholesterol. For years, the relationship of serum cholesterol and its metabolism to cardiovascular disease has been an issue of concern (4-6). Cholesterol is necessary for normal cellular function; however, it also has been shown to be a major constituent of atherosclerotic plaques (7). The results of epidemiological studies confirm a positive relationship between elevated serum cholesterol levels and the risk of coronary heart disease (8-16). The association has been found to be stronger for men than for women (17). By applying discriminate function analysis to the Framingham study data, Cornfield (18) attempted to quantify the relationship of the serum cholesterol level to coronary heart disease. Although no critical values of serum cholesterol were found at which sharp increases in risk of coronary heart disease occur, it was determined that a one percent reduction of serum cholesterol should result in a 2.66 percent reduction in risk of coronary heart disease. Implications are that serum cholesterol levels do play an integral role in the determination of risk for coronary heart disease as well as of improvement of cardiac status.

Selected lipoproteins. The major lipoprotein secreted by the liver is very-low-density lipoprotein (VLDL). VLDLs are very rich in triglycerides with little if any cholesterol ester in their lipid core. The surface coat of VLDL contains unesterified cholesterol,

phospholipids, and apolipoprotein. As VLDLS circulate in the plasma they undergo numerous changes. Initially, they acquire cholesterol ester from high-density lipoprotein (HDL). At the same time they encounter lipoprotein lipase on the surface of systemic endothelial cells which degrades triglycerides with the release of free fatty acids and glycerol. In humans, the resultant VLDL-remnants are transformed into low-density lipoproteins (LDL). LDLs contain mostly cholesterol ester in their lipid core. Approximately two-thirds of the cholesterol in the blood is found in LDL (19).

The LDL cholesterol subfraction of total plasma cholesterol is the main contributor to the relationship between plasma cholesterol levels and coronary heart disease, while the HDL cholesterol portion is inversely related to the incidence of coronary heart disease (20-21). Schonfeld et al. (22) demonstrated dietary cholesterol causes a rise in LDL when consumed with a diet rich in saturated fatty acids. It is suggested that excess cholesterol in the diet might produce transitory increases in LDL-cholesterol that are not seen in the fasting state. Even if the consumption of cholesterol association with saturated fatty acids occurs only during a portion of the day, the resulting particles could contribute to atherosclerosis. It follows than that this rise would increase the risk for coronary heart disease.

Extrahepatic tissue concentration of cholesterol can be increased by at least two mechanisms: de novo synthesis or uptake of LDL (8). An increase by either means may result in an increase in plasma cholesterol

(8). Cholesterol must be returned to the liver for excretion or degradation. Although the mechanisms for such a process are still somewhat vague, they are thought to involve HDL. These mechanisms are often referred to as reverse cholesterol transport (8). HDLs contain mostly cholesterol ester in their neutral lipid core. Their lipoprotein coat consists of unesterified cholesterol, phospholipids, and apoproteins. The HDLs made by the liver act as carriers and reservoirs for lipids during the metabolism of lipoproteins. They are especially important in their role in the transfer of cholesterol. The HDLs accumulate cholesterol which is converted into cholesteryl esters of long-chain fatty acids via the enzyme cholesterol acyl transferase. The HDL can transfer some of its cholesteryl esters to VLDL in exchange for triglyceride. The cholestryl esters will then be returned to the liver for excretion or degradation (8). Removal of whole HDL particles for the extraction of cholesterol is also likely, but the mechanism is unknown. It is generally thought that a high concentration of HDL may protect against atherosclerosis (23-24). Both HDL- and LDL-cholesterol have potential for providing information predictive of coronary heart disease because of their roles in cholesterol metabolism. However, each cholesterol fraction is somewhat independent of the other as reflected in the small magnitude of correlation between the two (25).

Triglyceride. Whether triglyceride levels are independently related to coronary heart disease is still a matter of controversy (8,26). Chylomicrons and VLDLs are involved in the metabolism of triglycerides, but these are not generally considered atherogenic (27).

Confusion stems from the fact that a large proportion of patients with coronary heart disease have highly elevated triglyceride levels (27). Extensive research in this area is needed before further conclusions can be drawn.

Experimental Studies of Gorging Versus Nibbling in Experimental Animals

The inducement of a gorging versus a nibbling food consumption pattern has been used to study adaptive changes in experimental animals. Experimental studies with rats indicated that as food was given in larger amounts at longer intervals, the rats displayed such changes as alterations in overall energy metabolism, more rapid absorption of glucose and fat from the intestine, increased glycogen synthesis, and a marked increase in lipogenesis (28-33). Increased activity of malic enzyme, hexokinase, phosphoglycomutase, UDPG-pyrophosphorylase, glycogen synthetase, glucose-6-dehydrogenase, and 6-phosphogluconate dehydrogenase in adipose tissue of meal-fed as opposed to nibbling rats was also demonstrated (33-35). Wolffe (36) worked with the goose and reported that this animal, when force-fed, developed increased serum cholesterol levels and increased atherosclerosis, regardless of whether cholesterol was or was not present in the diet. The chicken, when allowed to eat for only two one-hour periods daily, exhibited highly elevated serum cholesterol levels and increased incidence of coronary atherosclerosis as compared to the nibbling control (37).

Cox and Taylor (38) investigated the induction of atherosclerosis in the monkey. Results again indicated an elevated serum cholesterol level and severity of atherosclerosis with a gorging type of feeding pattern. Similar results were found by Gopalan, et al. (39).

In summary, data from studies using experimental animals can be interpreted to indicate that gorging is associated with (a) a greater elevation of serum cholesterol levels when the diet contains modest levels of cholesterol, and (b) an enhancement in the production of and an inhibition in the regression of atherosclerosis.

Experimental Studies of Gorging Versus Nibbling in Humans

The reaction of humans to variations in eating habits related to selected blood lipids is controversial. Little is known about the effect of meal frequency in humans under normal living and working conditions. A large percentage of the United States population place a burden on their systems for disposing of nutrients by consuming 50 to 75 percent of their daily food intake at the evening meal (29).

Solid evidence of a positive or negative correlation between feeding frequency and serum cholesterol levels clearly is not present. Limitations of studies regarding this issue include the use of a formula type diet and hospitalization during the experimental period. Hashim et al. (40) noted that there is typically a decrease in serum cholesterol levels whenever individuals are changed from a regular to a formula type diet because of the type of fat utilized in the formula. Also, Hashim

et al. (40) postulated that the hospitalization of experimental participants contributes to a lowering of the cholesterol level; however, no rationale was given.

Despite the limitations involved, research has been done in an effort to explore the correlation between feeding frequency and serum cholesterol levels. Jagannathan et al. (41) studied the effect of gorging versus nibbling using two types of homogenized formula diets. They found a decrease in serum cholesterol levels of those subjects consuming eight meals per day as opposed to three meals per day. No change in triglyceride levels occurred. Cohen (3) reported on the unpublished data of Allweiss et al. which showed a change in serum cholesterol levels when the subjects consumed three meals per day initially, and subsequently, six meals per day. The changes appeared to be associated with differences in feeding frequency, since the quantitative and qualitative constituents of the diet remained constant. These observations were made on unhospitalized volunteers. Gwinup (42) reported on three hospitalized individuals whose serum cholesterol levels where elevated when changed when a nibbling to a gorging type of feeding pattern. Changes produced by gorging were readily reversed by a nibbling type of feeding pattern.

In summary, preliminary results are consistent that feeding frequency may contribute to abnormally elevated serum lipid concentrations.

Metabolic Adaptations Related to Frequency of Food Consumption

The activities of a number of enzymatic pathways have been shown to be influenced by the load of nutrients or substrates presented to them per unit of time (43). Thus, feeding frequency could be a significant factor in the regulation of intermediary metabolism. Furthermore, it follows that the adaptation in metabolism seen with various feeding patterns could result from the limited activities of some of the enzymatic pathways with consequent activation of alternative ones. For example, the rat maintained on a gorging type of feeding pattern exhibits such an adaptation to facilitate the conversion of glucose to fatty acids, alphaglycerophosphate, and glycogen as well as to promote a greater capacity for NADPH generation for the support of fatty acid biosynthesis (43).

A number of enzymes are responsible for the conversion of glucose to its storage forms of fatty acids and glycogen. For example, the conversion of glucose to pyruvate involves the enzymes hexokinase, phosphofructokinase, and pyruvate kinase. In the adipose tissue of rats maintained on a gorging type of feeding pattern, the activity of hexokinase and pyruvate kinase is significantly increased (43). The conversion of pyruvate to acetyl Coenzyme A requires the enzyme citrate lyase. The activity of this enzyme is markedly increased as a result of a gorging type of feeding pattern (43). The conversion of acetyl Coenzyme A to fatty acids involves the key enzymes acetyl Coenzyme carboxylase, fatty acid synthetase, and alphaglycerophosphate

dehydrogenase. The activity of these enzymes is also markedly increased in the adipose tissue of rats maintained on a gorging type of feeding pattern (43).

The pentose pathway is responsible for providing fifty percent of the reducing equivalents needed for lipogenesis. The two key enzymes involved are 6-phosphogluconate dehydrogenase and glucose-6-phosphate, the latter of which is significantly increased with gorging and determines the amount of substrate flowing through the pentose pathway. The remainder of the necessary reducing equivalents are thought to be derived from the malate transhydrogenase sequence (43). The initial substrate for this pathway is thought to be oxaloacetate derived from the cleavage of citrate. The oxaloacetate is converted to malate then oxidatively decarboxylated to pyruvate. A cycle can be formed by converting the pyruvate back to oxaloacetate. Two of the enzymes involved in the process, pyruvate carboxylase and malic enzyme, increase significantly with gorging.

During the relatively long periods of fasting accompanying a gorging type of feeding pattern, adaptation must occur such that a steady supply of blood glucose is provided for the metabolic processes requiring it. Hollifield et al. (26) showed that the liver glycogen is not decreased after a twenty-four hour fast in the rat that has gorged, as opposed to rats fasted after ad libitum feeding. Many enzymes involved in hepatic gluconeogenesis are increased during fasting (44). It appears then that there is also an increase in gluconeogenesis from amino acids transported from the protein stores of the body to the liver

during the relatively long fasting periods associated with the gorging type of feeding pattern. It is also possible that the net flux of free fatty acids from the fat depots is increased in those who gorge versus those who nibble as a result of prolonged periods of fasting (42). The result would be increased synthesis of serum lipids by the liver since the uptake of free fatty acids by the liver is largely determined by availability.

Exercise Effects on Serum Lipids

Results of investigations involving the effect of exercise on total and LDL-cholesterol concentration appear variable and inconsistent. Cooper et al. (45) found that total cholesterol levels were significantly lower in subjects considered to have excellent aerobic fitness when compared with those with very poor or poor fitness. However, adiposity may have accounted for some of the differences between the groups. Others, however, have failed to show a decrease in cholesterol concentration after training of weeks to months duration (46-48). No alteration in cholesterol levels were observed among middle aged men and patients enrolled in post coronary exercise training programs (46-47). Likewise, reduction of LDL-cholesterol has not been found after training despite improvement in maximal oxygen consumption (48).

Nakamura et al. (49) studied the effect of exercise on HDL-cholesterol levels. Twenty members of a joggers' club were matched

for sex, age, total cholesterol, serum triglyceride, and body weight index with healthy sedentary controls. Both HDL-cholesterol and HDL-cholesterol/total cholesterol concentrations were significantly higher in the joggers than in the matched controls. Sutherland and Woodhouse (50) found that changes in the HDL-cholesterol level were often dependent upon levels prior to onset of the exercise conditioning. Subjects who initially had a lower HDL-cholesterol concentration tended to show greater increases after training. Therefore, individuals with low HDL-cholesterol levels may appear to benefit more from exercise training and may show more consistent alterations in HDL-cholesterol than individuals who already have higher HDL-cholesterol levels.

Due to the known effect of diet on the triglyceride level of humans, Lipson et al. (51) studied men and women subjects maintained on a constant composition diet during a six week exercise protocol. Results indicated that there was no significant lowering of triglyceride levels. Streja et al. (49) also found no significant alteration in triglyceride levels among patients with coronary artery disease maintained on a traditional rehabilitation exercise conditioning program without dietary alteration.

The Use of Food Records in Research

The food record has been used for many years as a means of obtaining estimations of the nutrient intake of the free-living population. However, much variation is seen in the number of days used

for the recording process. Stuff (53) recommended the use of four day food records as opposed to three day food recores, using Friday through Monday. The rationale for the use of such a four day record stemmed from the concern for the variability reported in nutrient intake according to the day of the week. Four day food records which include weekends would cover the days of the week that have posed major concern regarding variability; i.e. weekend days versus weekdays.

There are limitations to the use of food records. Primarily, as accuracy is improved there is a rapid increase in the interference with the normal lifestyles and possibility with normal food intakes (53). Also, analysis of food records is dependent on food composition tables which, at their best, can only approximate the nutrient content of foods that are actually consumed (54).

CHAPTER III

METHODOLOGY

Selection of Subjects

Following the approval of the Institutional Review Board for Research Involving Human Subjects at Virginia Polytechnic Institute and State University, seventeen males were recruited from the Cardiac and Intervention Program conducted at the university. All subjects participated in either a swimming or walking/running routine sponsored by the Cardiac and Intervention Program and had been actively involved in this program for at least four months prior to initiation of the study. The subjects were classified by the Cardiac and Intervention Program as either cardiac participants or intervention participants. Such a classification was based primarily on history of cardiovascular illness, present cardiovascular condition, and the participants' physicians requests regarding degree of supervision while participating in the program (Table 1). The Cardiac and Intervention program is in operation on Monday, Wednesday, and Friday morning. Therefore, participants of that program as well as of this study routinely exercise three times per week in the structured program with the addition of any self-motivated, self-monitored exercising.

Recruitment was made from both the cardiac and intervention sections of the program based on the participant's most current total serum cholesterol and triglyceride levels (Appendix A). Criteria for recruitment were a serum cholesterol of greater than 240 milligrams per

TABLE 1

Distribution of subjects based on Cardiac and Intervention Program classification and activity type

	Walking/Running	Swumming
Cardiac	5	2
Intervention	6	4

deciliter and/or triglyceride of greater than 150 milligrams per deciliter since these values are indicated as in the upper range of normal, bordering on abnormal values (44). Food records on file with the Cardiac and Intervention Program were reviewed for each subject's past compliance with the dietary guidelines of the American Heart Association in an attempt to facilitate future compliance with the research protocol. Volunteers recruited from this target group signed a consent form prior to participation in the study (Appendix B).

Research Protocol

Initially all subjects completed a 24 hour food record. This food record was analyzed for approximate kilocalorie level and was used to assist in the counseling of the participants regarding adherence to the dietary guidelines of the American Heart Association. An individualized orientation session was then held for each subject. Individual counseling was provided to the subjects to facilitate integration of the research protocol (i.e., three meal plan and six meal plan) into their lifestyles. This session included an introduction to the purpose and procedures of the study as well as comprehensive instruction on the meal patterns to be used throughout the study. A booklet entitled Grocery Guide: Tips on Wise Food Selection (55) which delineates the dietary guidelines established by the American Heart Association was presented to and thoroughly reviewed with each subject. A kilocalorie level which promoted weight maintenance was determined, and meal patterns of three

meals per day and six meals per day were formulated and discussed with each subject. Finally, each subject was presented with a list of common foods in portions approximately 100 kilocalories to aid in food selection and adherence to the recommended daily kilocalorie level (Appendix C). Throughout the data collection period of the study, subjects were contacted individually at least every ten days to review dietary guidelines and discuss questions.

The data collection began with a two week acclimation period during which three meals per day were consumed by all subjects. The meals were such that approximately one quarter of the total daily kilocalorie intake for each subject was consumed at each of the morning and midday meals, and one half of the total daily kilocalorie intake was consumed at the evening meal. At the conclusion of the two week acclimation period, fifteen milliliters of blood were drawn via venipuncture from each subject. The blood was drawn following a twelve hour fast, and the data were used to establish baseline values. The subjects were then divided into two groups. Group I (eight subjects) consumed three meals per day for six weeks while group II (nine subjects) consumed six meals per day for the same amount of time. The caloric distribution for the three meals per day was in the same proportions as was consumed during the two week acclimation phase. The six meals consumed per day were of approximately equal caloric proportion divided throughout the waking hours. Upon conclusion of the six weeks, blood was drawn using the same methodology as described above. The two groups then reversed feeding patterns used in the previous six week period such that group I was

consuming six meals per day and group II was consuming three meals per day, again for six weeks. At the conclusion of the second six weeks, a third blood sample was drawn for analysis (Figure I).

During the entire experimental period the subjects were instructed to follow the dietary guidelines issued by the American Heart Association with emphasis placed on the limitation of foods high in cholesterol and saturated fat. Individual counseling was provided throughout the data collection period to facilitate compliance with the dietary guidelines. Four-day food records (Friday through Monday) were completed at the conclusion of the first, fourth, seven, tenth, and thirteenth weeks of the study. The subjects were provided with guidelines for reporting food intake, a completed sample of a food record, and food record collection sheets for their use (Appendix D). The four-day food records collected at the conclusion of weeks four and ten of the study were assessed qualitatively and used to give some indication of the degree of compliance with the established dietary guidelines and meal patterns. In addition, the food records collected at the conclusion of the first, seventh, and thirteen weeks of the study were analyzed by computer for kilocalorie, total fat, saturated fat, and cholesterol level. In all cases, indications of possible noncompliance were used in the ongoing counseling of the subjects.

Following the drawing of the blood samples, the blood was centrifuged and the serum drawn off. The blood serum samples were sent to Roche Biomedical Laboratory for analysis of total serum cholesterol,

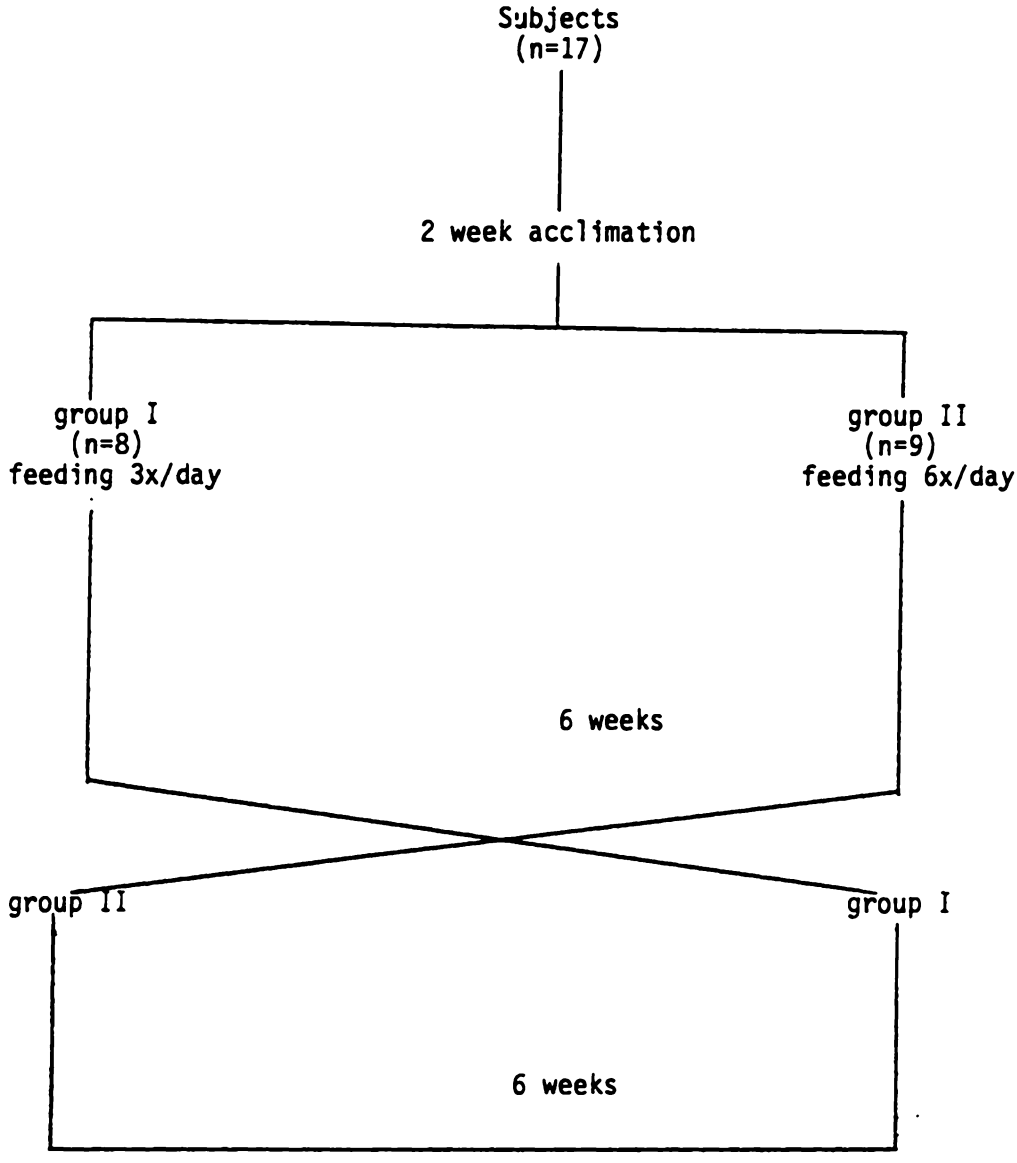

Figure 1. Experimental Design

low-density lipoprotein cholesterol, high-density lipoprotein cholesterol, and triglyceride levels.

Statistical Analysis

Mean values with standard deviations of the means were calculated for kilocalorie, total fat, saturated fat, and cholesterol intake as well as for weight. Paired t-tests were used to evaluate the significant of changes in each of the blood lipid parameters during the three meal per day feeding pattern and the six meal per day feeding patterns (56).

CHAPTER IV

RESULTS AND DISCUSSIONS

Seventeen males were included in the study which compared the effect of the consumption of three meals per day versus six meals per day on total cholesterol, HDL-cholesterol, LDL-cholesterol, and triglyceride levels. The degree of compliance with the research protocol was good as indicated by the following: completion of food records, adherence to the meal patterns, food selection as recommended by the American Heart Association, and cooperation in the procedures of drawing blood.

Subject Description

Age range of the subjects was 43 to 68 years with a mean age of 54.5 years. Although one of the goals of the American Heart Association is to obtain and maintain ideal body weight, the focus of this study was to promote weight maintenance at current weight because of the possible effect that weight reduction itself might have on the serum lipid profile (19,57). Therefore, weight change was monitored. The subjects were advised to monitor their own weight weekly, and the kilocalorie levels of the diets were adjusted to promote weight maintenance. The subjects' heights and weights as measured at intervals throughout the study are displayed in Appendix E. Of those consuming three meals per day during phase II of the study, four subjects lost weight, three

subjects gained weight, and one remained at a constant weight. Of those consuming six meals per day, five subjects gained weight and four subjects lost weight. During phase III of the study, three subjects who consumed six meals per day gained weight, four subjects lost weight, and one remained at a constant weight. And, of those consuming three meals per day, six and three subjects gained and lost weight, respectively. The mean changes in weight and standard deviations for each group at each phase of the study are presented in Table 2. The overall mean change in weight for both groups I and II cumulatively from initiation to the end of the study was a loss of 0.30 ± 2.648 percent of original body weight. Some subjects initially sought to use the diet patterns involved in the study protocol as an opportunity to lose weight. The purpose of the study and rationale for weight maintenance were reviewed to reinforce compliance with weight maintenance.

Dietary Analysis

The five sets of food records were assessed qualitatively and used to give some indication of the degree of compliance with the established dietary guidelines and meal patterns. Records from the first, seventh, and thirteenth weeks were further analyzed by computer. Each subject's mean intakes of kilocalories, total fat, percent of kilocalories as fat, saturated fat, percent saturated fat, and cholesterol as indicated by the four day food records from these weeks are presented in Appendix F. Mean intake and standard deviations of kilocalorie total fat, percent of

TABLE 2

Group mean and standard deviation of weight at the conclusion of each phase of the study and overall group mean and standard deviation of percent weight change.

	weight (kg) end phase I	weight (kg) end phase II	weight (kg) end phase III	weight change %
Group I	79.81	79.38	79.50	- 0.713
	± 8.379	± 8.895	± 9.040	± 2.777
Group II	75.69	75.41	75.61	+ 0.067
	122.056	±11.149	±10.892	± 2.638

kilocalories at fat, saturated fat, percent saturated fat, and cholesterol at the first, seventh, and thirteenth weeks for Group I and Group II are presented in Table 3.

Assessment of the total set of food records indicated compliance with dietary guidelines and meal patterns. Generally, a wide variety of foods was consumed. Many subjects were routinely consuming the same foods at the second, fourth, and sixth feeding during the six meal per day feeding pattern. When confronted with this, the subjects commented on the fact that the snacks being consumed routinely were easy to prepare and convenient to carry and consume and were therefore easier to work into their daily routine.

The largest discrepancy found when comparing the food records to the dietary guidelines and patterns was an over consumption of kilocalories at the feeding time most closely associated with the evening meal. This high kilocalorie intake most generally was associated with large meat or meat substitute servings.

When discussing with the subjects what their greatest difficulty with the study protocol was, it was found that adherence to the specific meal patterns, and especially six meals per day, proved to be most difficult. Since most of the participants were employed, the consumption of feedings in mid-morning and mid-afternoon proved to require a major adjustment in their daily routine.

The American Heart Association does not quantify the dietary goals which they offer as suggestions for the United States population. However, the Senate Select Committee on Nutrition and Human Needs also

TABLE 3

Mean intakes and standard deviations of kilocalories, total fat, percent kilocalories as fat, saturated fat, percent kilocalories as fat, saturated fta, percent saturated fat, and cholesterol for groups I and II at weeks one, seven and thirteen.

	week number	kilo-calories	total fat (g)	% kilocalories as fat	saturated as fat (g)	% fat as saturated fat (g)	cholesterol (mg)
Group I	1	2241 ±651.930	96.01 ±54.084	36.70 ±12.410	29.85 ±16.164	32.113 ±6.856	280.43 ±121.548
	7	2180 ±651.729	83.10 ±54.342	32.71 ±13.115	27.58 ±11.178	36.780 ±11.456	219.755 ±107.143
	13	1982 ±202.779	73.21 ±28.171	33.05 ±11.741	31.03 ±16.497	41.040 ±7.761	206.06 ±87.790
Group II	1	1957 ±508.640	66.72 ±17.295	31.356 ±6.4168	22.47 ±6.168	33.900 ±4.560	225.71 ±78.483
	7	2048 ±602.687	77.21 ±28.812	33.540 ±4.942	28.76 ±12.556	36.456 ±3.493	237.36 ±82.532
	13	1656 ±522.346	66.70 ±31.170	33.22 ±7.170	24.28 ±11.548	37.440 ±7.505	252.49 ±135.023

published a set of dietary goals which do suggest specific levels of fat and cholesterol intake (58). Included in those goals are the reduction of overall fat consumption to thirty percent of the energy intake, reduction of saturated fat consumption to account for about ten percent of the total energy intake or approximately thirty-three percent of the total fat intake, and reduction of cholesterol intake to about 300 milligrams per day. Brown and co-workers (59) have shown that such a diet is effective in lowering serum cholesterol. Computer analysis of the four-day food records for the first, seventh and thirteenth weeks indicated that neither group I or II met the recommendation of percent total fat for any of the weeks analyzed. Qualitative analysis indicates that this was generally due to a high intake of meats, cheeses, and margarine.

The criteria of thirty-three percent of fat consumed as saturated fat was met only by Group I and only in phase I of the study. This again is attributable to a high intake of meat and cheese.

The intake of cholesterol by both groups I and II met the criteria of less than 300 milligrams per day for all three phases of the study as indicated by the four-day food records. A high emphasis on the elimination of foods high in cholesterol is present in on-going counseling by the supervisors of the Cardiac and Intervention Program as well as throughout the public media. In counseling the subjects prior to the initiation of the data collection as well as throughout the study, it was apparent that the level of knowledge and concern regarding

dietary cholesterol content of food was very high which aided in their elimination of those foods from the diet.

Serum Lipid Analysis

Results related to changes in serum lipid parameters for subjects are presented in Appendix G. Mean and standard deviations of serum lipid parameters for each group at each phase are presented in Table 4. The paired t-test analysis results are found in Table 5. Normal ranges for total cholesterol triglyceride and LDL-cholesterol as indicated by Roche Biomedical Laboratory are as follows:

total cholesterol	115-295 milligrams per deciliter
triglyceride	10-190 milligrams per deciliter
LDL-cholesterol	80-210 milligrams per deciliter

There was no comparable normal range for HDL-cholesterol presented by the laboratory. Krupp and Chatton (4) indicated that a HDL-cholesterol value of greater than 40 is desirable.

Mean total cholesterol, triglyceride, and LDL-cholesterol levels for both groups I and II were within the specified normal ranges for all three phases of the study. The HDL-cholesterol level was above 40 for both groups at phases I and III, but below 40 at phase II.

For Group I, the mean total cholesterol value after the six meal per day feeding pattern was lower than the mean value after the three meal per day feeding pattern; however, the difference was not statistically significant. This pattern did not hold true for Group II.

TABLE 4

Means and standard deviations of serum total cholesterol, triglyceride, LDL-cholesterol, and HDL-cholesterol levels for groups I and II at phases I, II, and III of the study.

	phase number	total cholesterol (mg/dl)	tri-glyceride (mg/dl)	LDL-cholesterol (mg/dl)	HDL-cholesterol (mg/dl)
Group I	I	236.250 ±36.189	150.13 ±78.547	164.13 ±33.267	41.63 ±9.61
	II	236.500 ±43.579	180.50 ±68.521	164.88 ±36.117	35.13 ±7.08
	III	230.500 ±28.132	141.63 ±48.071	161.13 ±19.142	40.75 ±6.11
Group II	I	224.440 ±31.516	146.89 ±72.951	151.89 ±31.970	42.89 ±5.99
	II	230.778 ±40.521	149.44 ±57.378	164.56 ±34.854	35.89 ±8.81
	III	217.556 ±27.835	124.11 ±45.947	147.89 ±22.839	44.44 ±8.02

TABLE 5

Paired t-test analysis of effects of feeding frequency on lipid parameters for groups I and II

	Group I (n = 8)	Critical Value	Group II (n = 9)	Critical Value
total cholesterol	- 0.56	2.3646	- 1.24	2.3060
triglyceride	- 2.78	2.3646	- 1.44	2.3060
LDL-cholesterol	- 0.30	2.3646	- 1.57	2.3060
HDL-cholesterol	+ 2.61	2.3646	+ 3.44	2.3060

The lack of significant change in total cholesterol level when changing from a gorging to a nibbling type of feeding pattern is inconsistent with the results of Jagannathan (41) and Gwinup (42). However, this inconsistency may be attributable to the differences found in the research protocol of these studies. The individual subjects within each group differed in their response to the two meal patterns with some subjects showing a lowering of cholesterol level. Therefore, high individual variation may account for a lack of statistical significance. Also, it has been shown that a adinine nucleotide diphosphate (NADP) generating system is necessary for cholesterol formation (60). That NADP formation has been shown to be accelerated by full spaced meals also has been established (61-62). It is possible that the spacing of the meals with the six meal per day feeding pattern remained large enough such that the TPN generating system was accelerated promoting cholesterol formation.

No significant difference in the triglyceride level of Group II between the nibbling and the gorging feeding patterns is consistent with the findings of Jagannathan (41). However, a significant difference ($p \leq 0.05$) in triglyceride level between the two feeding patterns was found with Group I. Figures 2 and 3 reflect the plot of the median values of triglyceride for group I and II, respectively. In plotting, median values rather than mean values were used to compensate for outliers within each group. Both plots show a trend of decreasing triglyceride levels with the six meal per day feeding pattern. With Group II this decreasing trend is continued when the subjects returned to the three

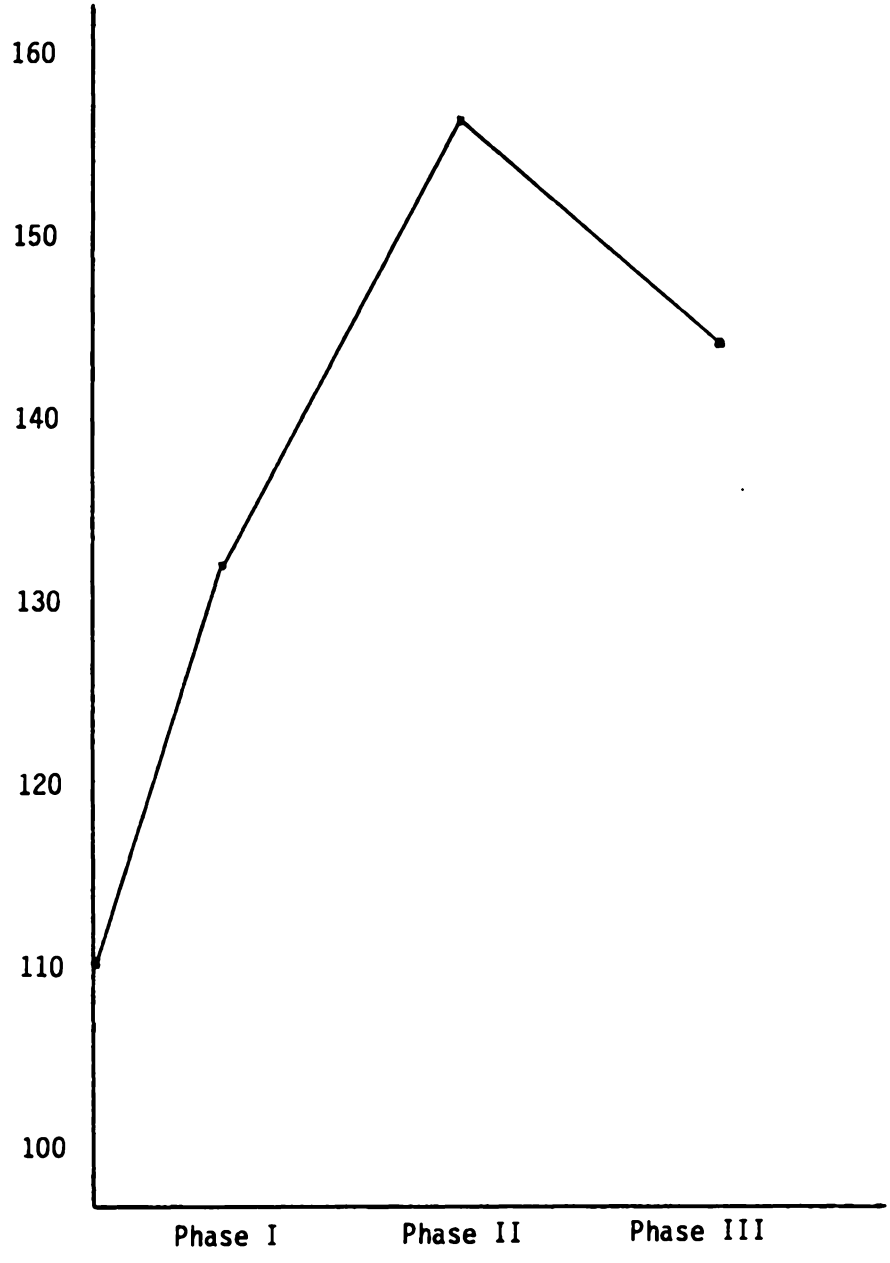

Figure 2. Median serum triglyceride values for group I upon recruitment (y axis) and phases I (3 meals per day), II (3 meals per day), and III (6 meals per day).

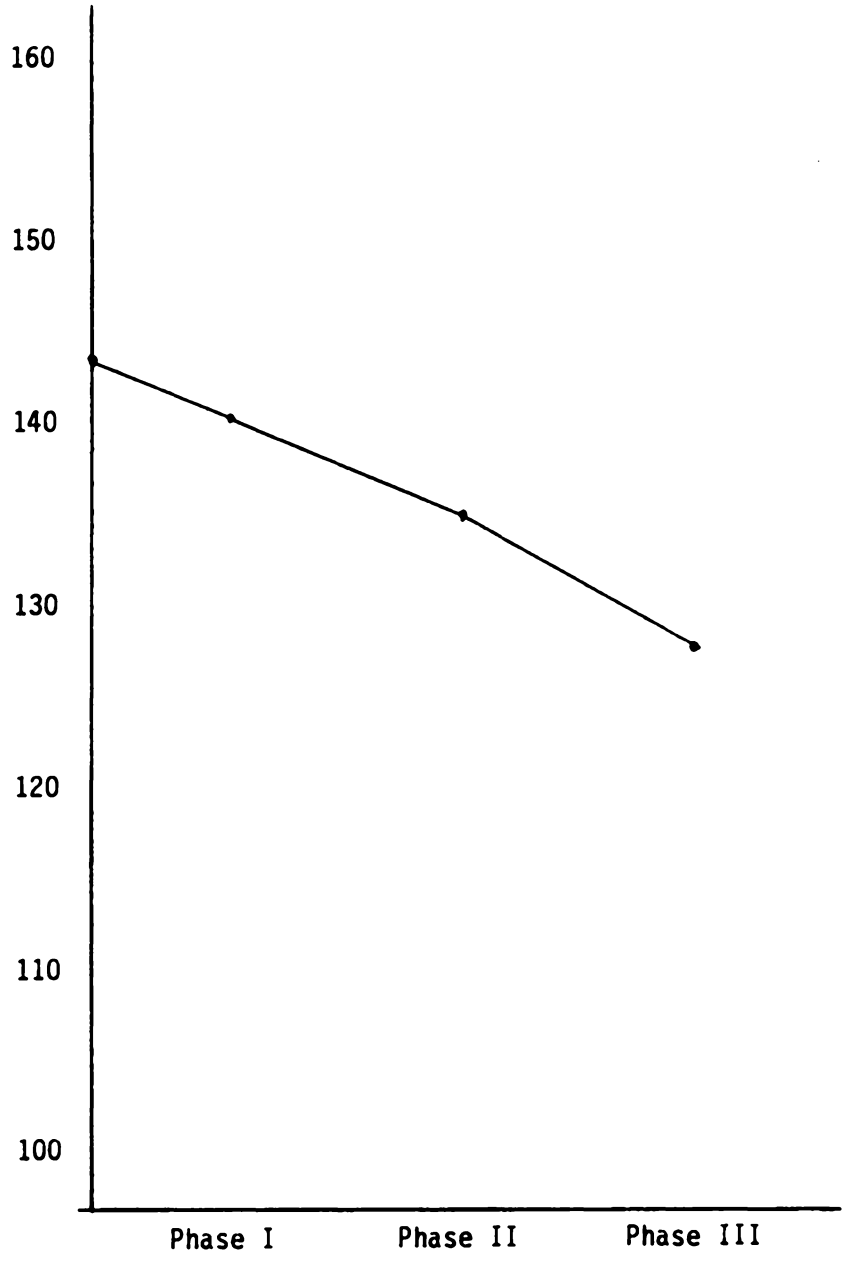

Figure 3. Median serum triglyceride values for group II upon recruitment (y axis) and phases I (3 meals per day), II (3 meals per day), and III (6 meals per day).

meal per day feeding pattern. The synthesis of triglyceride during a six meal per day feeding pattern is likely to be lower with the oxidation of fatty acids for energy being favored over their incorporation into triglycerides. It could be hypothesized that these patterns of triglyceride levels are attributable to changes in types of food consumed; however, there is no data to support such a theory.

Although not statistically significant, the trend in changes of LDL-cholesterol for groups I and II for all three phases is similar to the changes seen with total cholesterol. Figures 4 and 5 reflect the similarity in the changes of total cholesterol and LDL-cholesterol for groups I and II, respectively. Because of the close association between the serum levels of total cholesterol and LDL-cholesterol (8), one would expect to see similar changes between the two lipid fractions. Shonefield's report (22) claimed that dietary cholesterol causes a rise in LDL-cholesterol when given concurrently with a diet rich in saturated fatty acids. It was found in this study that the subjects moderate consumption of cholesterol was accompanied by a relatively higher intake of saturated fat. This may account for the elevated LDL-cholesterol findings.

For both groups there was a significant difference ($p \leq 0.05$) in HDL-cholesterol levels between the six meal per day and the three meal per day feeding patterns. However, the findings are dissimilar in that for Group I the HDL-cholesterol was higher after the six meal pattern as opposed to the three meal pattern, and for Group II, the opposite was true. It has been reported that fluctuations in HDL-cholesterol level

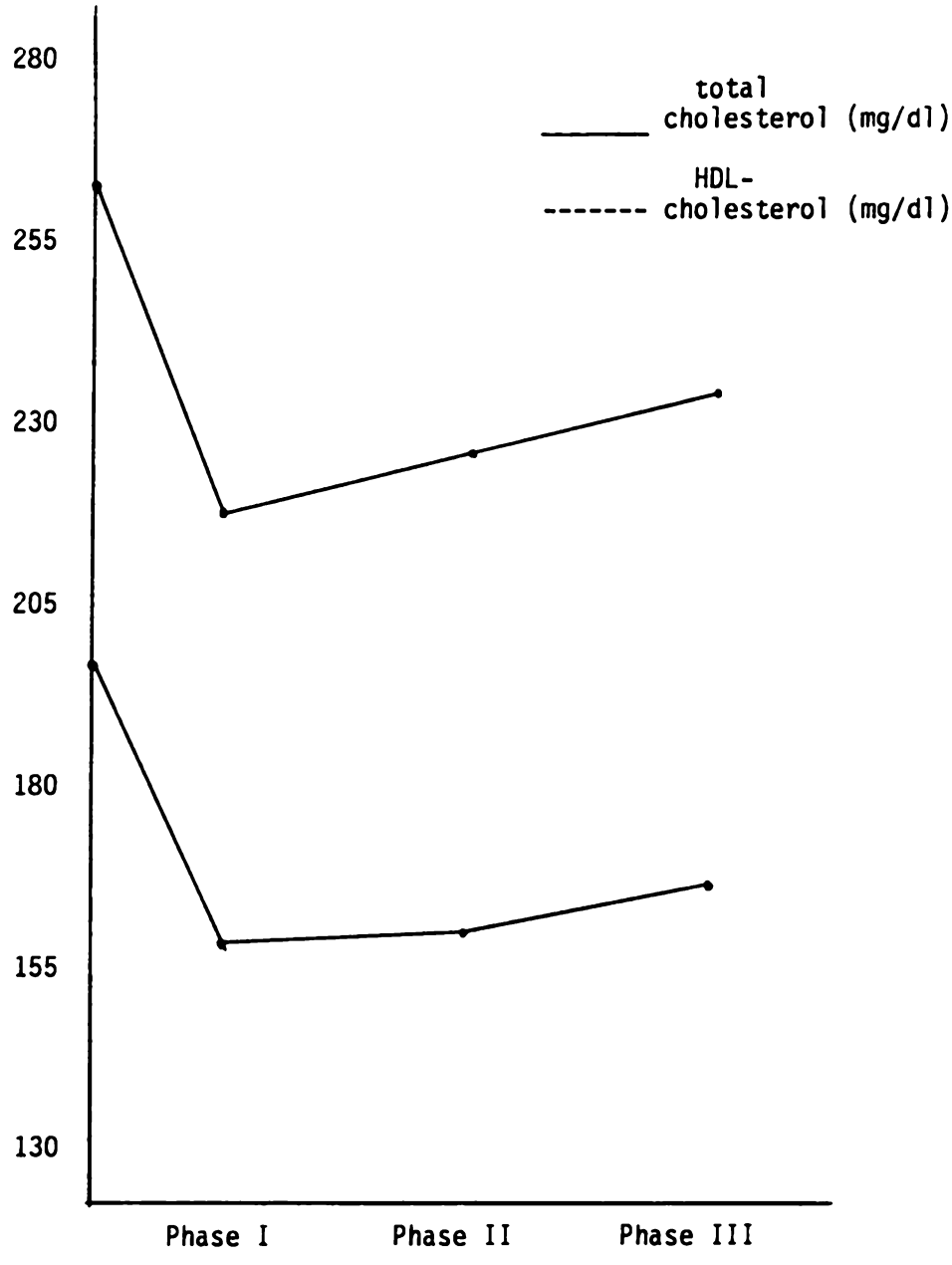

Figure 4. Median total cholesterol and LDL-cholesterol values for group I upon recruitment (y axis) at phase I (3 meals per day), II (3 meals per day), and III (6 meals per day).

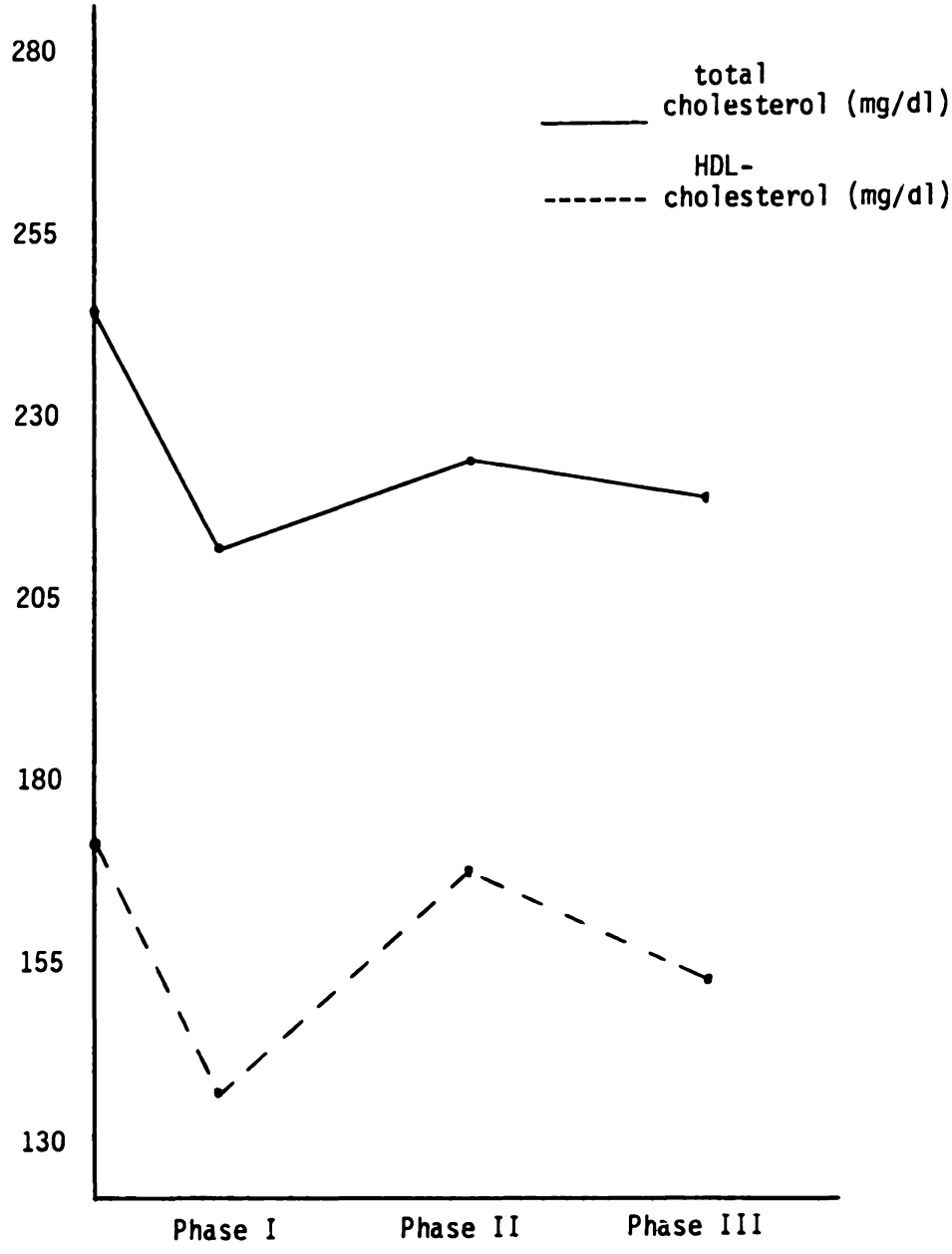

Figure 5. Median total cholesterol and LDL-cholesterol values for group II upon recruitment (y axis) at phase I (3 meals per day), II (3 meals per day), and III (6 meals per day).

with time appear related to variations in serum triglyceride concentrations (63). This may account for the variability in Group I. (Figure 6) The variance in expected HDL-cholesterol levels for Group II may be based on high individual variation or possibly on the activity of one or several enzymes known to be major determinants of HDL-cholesterol concentrations such as lipoprotein lipase (64-65) or lecithin: cholesterol acyltransferase (66). It is beyond the scope of this study to make any suggestions as to which if any of the enzymatic activities may in fact have been responsible for the alteration of the HDL-cholesterol concentration of Group II.

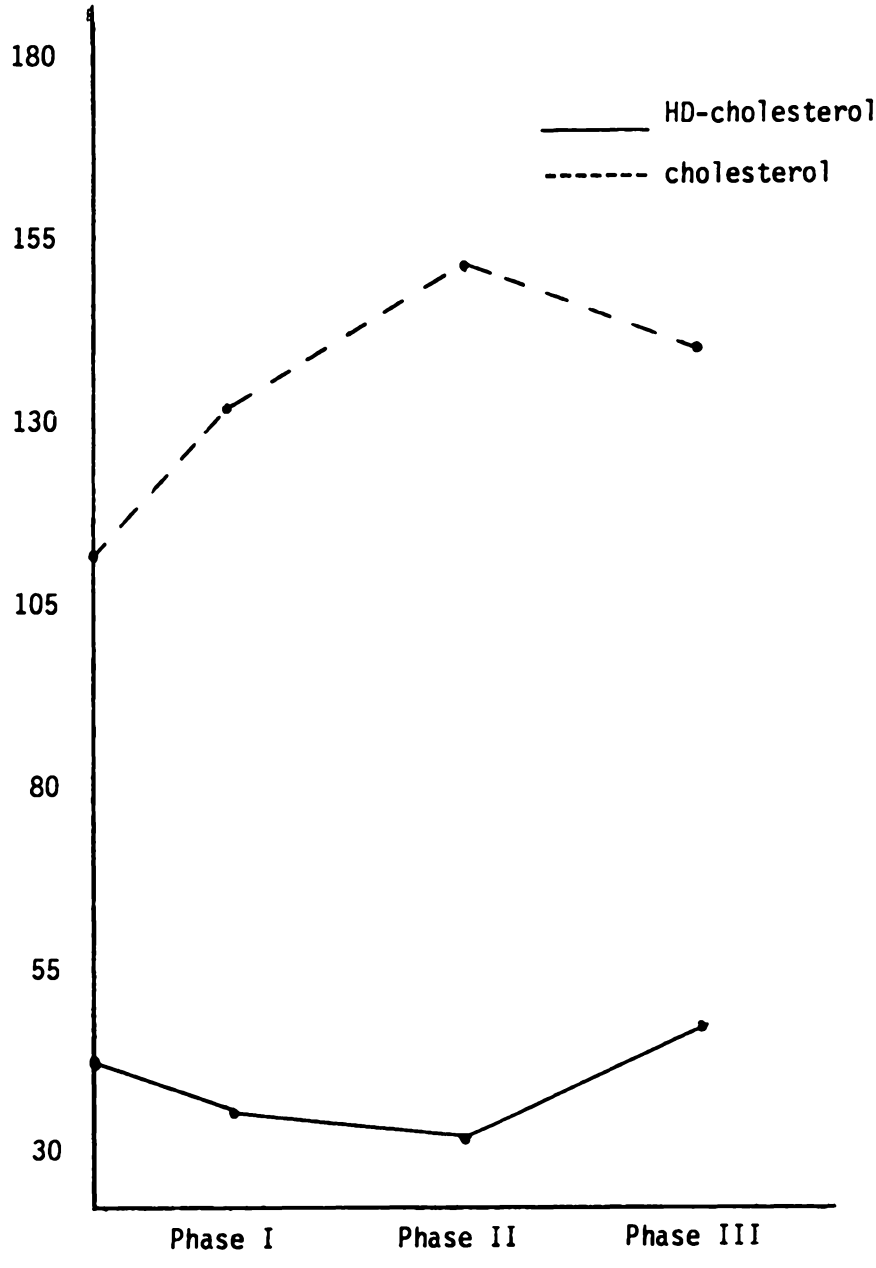

Figure 6. Median serum HDL-cholesterol and triglyceride values for group I upon recruitment (y axis) at phase I (3 meals per day), II (3 meals per day), and III (6 meals per day).

CHAPTER V

Summary and Conclusions

The effect of the consumption of three meals per day versus six meals per day on selected serum lipids was assessed. Seventeen males diagnosed as having cardiovascular disease or at risk for the disease participated in the study by following the research protocol of three meals per day and six meals per day, maintaining dietary food records, and donating blood for serum lipid analysis. The food records were assessed qualitatively and quantitatively to provide information on the degree of compliance with the dietary guidelines of the American Heart Association and the established meal patterns as well as to provide information to be used in the ongoing counseling of the subjects.

Blood samples were obtained from each subject at the conclusion of each phase of the study, and the following serum lipid parameters were determined: total cholesterol, triglyceride, LDL-cholesterol, and HDL-cholesterol.

Computer analysis of the food records for each group of subjects indicated that, generally, the intake of total fat and saturated fat was relatively high while the intake of cholesterol was within an acceptable range.

When considering mean lipid parameters for each group of subjects, total cholesterol, triglyceride and LDL-cholesterol levels were within ranges considered acceptable for each phase of the study. The

HDL-cholesterol level fell below a desirable level for both groups during phase II of the study.

There was no statistically significant difference found between the consumption of three meals per day and six meals per day regarding the lipid parameters of total cholesterol and LDL-cholesterol for both Groups I and II. A significant decrease in triglyceride with the six meal per day feeding pattern occurred in Group I but not Group II. HDL-cholesterol increased significantly for Group I when those subjects changed from three meals per day to six meals per day. The HDL-cholesterol level for Group II increased significantly when those subjects changed from a six meal per day pattern to a three meal per day pattern.

The high level of deviation in results between the two groups indicate the widely divergent response of each of the individual subjects. The variability illustrates the hazard that would be involved in attempting to draw valid conclusions from the results on a small number of subjects involved in a study of limited duration. It is possible that a feeding trial of longer duration with a higher level of subject control, if achievable, might reveal a greater effect of feeding pattern on serum lipids. Finally, more meaningful information might have been obtained by performing serial lipid analyses on samples of blood taken at intervals throughout the various phases of the study as opposed to simply at the end of each phase.

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APPENDIX A

Most current total serum cholesterol and triglyceride levels of subjects prior to initiation of data collection

Subject code	Date	Serum Cholesterol (mg/dl)	Triglyceride (mg/dl)
01	5/21/85	239	149
02	7/29/85	260	99
03	10/17/85	273	160
04	10/09/85	244	156
05	8/03/85	257	80
06	1/28/85	278	87
07	11/13/85	355	297
08	10/12/85	264	172
09	8/23/85	276	137
10	12/05/85	225	116
11	11/26/85	261	103
12	11/11/85	246	138
13	8/05/85	294	142
14	3/15/85	297	139
15	9/27/85	246	52
16	12/12/85	238	143
17	5/15/85	232	219

APPENDIX B

Consent for Participation

I have received written and oral explanation of the study and understand the following:

Data on subjects presently on file with the Cardiac and Intervention Program will be reviewed. Records will be reviewed for laboratory values including total cholesterol and triglyceride. Past food records also will be reviewed by the investigator. Subjects will participate in an orientation program in which the dietary guidelines of the American Heart Association will be reviewed. Subjects will comply with a three meal per day feeding pattern during the acclimation phase of the study. The subjects will also comply with a three meal per day feeding pattern for six weeks and a six meal per day feeding pattern for another six week period. Approximately 15 milliliters of blood will be taken a total of three times throughout the study by a Registered Nurse. The subjects will complete five four-day food records during the course of the study.

The identity of the subjects will be held confidential in all reports of this research. Subjects are at essentially no risk by participating in the project. No compensation is available if injury is suffered as a result of this research; however, subjects can gain valuable information about their nutritional health. There will be no monetary cost for participation in the study and neither will there be any monetary reward. Consent for participation may be withdrawn at any time. Questions regarding this project will be answered by the investigator.

I understand the above, and agree to participate in the nutrition study at the times designated between December 1, 1985, and May 1, 1986.

_____

Subject

APPENDIX C

The following is a list of foods to help you in approximating your calorie intake and portioning your food intake for the three and six meal per day feeding patterns. The amounts of food listed below contain approximately 100 Calories for each food.

Beverages

- apple juice - 7 oz (scant 1 cup)
- carrot juice - 1 cup (8 oz)
- cranberry juice cocktail = 2/3 cup
- low-cal cranberry juice cocktail - 18 oz
- sweetened grape juice - 6 oz
- canned grapefruit juice - 1 cup
- orange juice from concentrate - 7 oz
- V-8 juice cocktail - 2 cups
- canned tomato juice - 2 1/2 cups
- regular soda - 8 oz (1 cup)
- skim milk - 9 oz
- 1% fat chocolate milk - 2/3 cup
- 2% milk - 3/4 cup

Eggs/Cheese/Oils

- egg beaters - 3/4 cup
- Morning Star Farms scramblers - 1/2 cup
- 2% fat cottage cheese - 1/2 cup
- 1% fat cottage cheese - 2 oz
- Low-cal American cheese - 2 oz
- part-skim ricotta cheese - 1/3 cup
- margarine - 2 tsp.
- safflower, sunflower, corn, wheat germ, or soybean oil - 3/4 cup
- mayonnaise - 2 tbsp.
- French dressing - 1 1/2 tbsp.
- thousand island dressing - 2 tbsp.

APPENDIX C (cont.)

Grains

biscuits - 1
bagel - 1/2
plain bread - 1 1/2 slice
French bread - 1 1/2 slice
hot dog/hamburger bun - 3/4 bun
English muffin - 2/3
bread sticks - 4
croutons - 1 oz.
enriched/quick grits - 2/3 cup
cream of wheat - 3/4 cup
oatmeal - 3/4 cup
bran cereal - 2/3 cup
cheerios - 1 cup
puffed rice, puffed wheat - 1 oz
shredded wheat - 1 oz or 1 biscuit
graham crackers - 3 squares
melba toast - 3 pieces
rye krisp - 4 triple crackers
pretzels - 1 oz
popcorn - 2 cups
cooked macaroni, noodles - 3/5 cup
cooked spaghetti - 1/2 cup
cooked rice - 1/2 cup

Fruit

apple - 1 large
sweetened applesauce - 1/2 cup
banana - 1 medium
cantaloupe pieces - 2 cups
sweet cherries - 20
dried dates - 6
dried figs - 2
juice packed fruit cocktail - 1 cup
grapefruit - 1
seedless grapes - 2 cups
orange - 2 small
peaches - 2 medium
pear - 1 medium
raw pineapple pieces - 1 1/3 cup
raisins - 1/4 cup
tangerines - 2 medium

APPENDIX C (cont.)

Vegetables

- cooked asparagus, broccoli, or green beans - 3 cups
- pork and beans or baked beans - 1/3 cup
- cooked beets or carrots - 2 cups
- carrots, raw - 2 large
- cooked yellow corn, mixed vegetables, or peas - 1 cup
- cooked lima beans - 1/2 cup
- baked potato - 1 medium
- mashed potato (with milk and margarine) - 1/2 cup
- baked acorn squash - 1/2 squash
- baked butternut squash - 2/3 cup
- baked sweet potato - 2/3 small

Meat/Fish/Poultry

- broiled bass - 1 1/2 oz
- baked flounder or sole - 1 3/4 oz
- broiled cod - 2 oz
- baked or broiled salmon - 2 oz
- cooked brook trout - 1 3/4 oz
- clams, meat only - 5 large
- crab cake - 2 oz
- raw shrimp or lobster - 3 1/2 oz
- raw scallops - 4 oz
- chicken, roasted without skin - 2 oz
- lean beef - 1 oz
- pork - 1 oz

Soups

- chicken noodle soup - 1 1/3 cup
- beef noodle soup - 1 1/4 cup
- manhattan clam chowder - 1 1/4 cup
- minestrone soup - 1 1/4 cup
- vegetable soup - 1 1/4 cup

Nuts

- black walnut halves - 10
- pine nuts - 2 tbsp.
- pecan halves - 12
- almonds - 17

APPENDIX C (cont.)

Desserts/Snacks

gum drops - 28
jelly beans - 15
marshmallows - 4 large
hard candy - 6 pieces
angel food cake - 1/14 of cake
animal crackers - 13
fig bars - 2
ginger snaps - 6 small
molasses cookies - 1 1/2
popsicle - 1 twin pop
fruit ice - 1/3 cup
fudgesicle - 1
sherbet - 1/3 to 1/2 cup
ice milk - 1/2 cup
gelatin desserts - 1/2 cup
honey - 1 1/2 tbsp.
jam or jelly - 2 tbsp.
low-cal jam or jelly - 4 tbsp.
maple syrup - 2 tbsp.

Other

cooked lentils - 2/3 cup
peanut butter - 1 tbsp.

APPENDIX D

GUIDELINES FOR RECORDING FOOD EATEN

An important consideration in this study is the composition of your diet. You will be asked to complete a total of 5 four-day food records throughout the course of this study. It is very important that we get accurate information regarding the kind and amount of food you eat.

Here are some hints for recording information:

1. Do not eat any differently than normal on the days you are recording. It is important that you consistently eat the number of meals requested throughout the study.

2. Be accurate! Write down what you have eaten immediately after it has been consumed. Do not wait until the end of the day to try to remember everything you have eaten. We do not care about spilled coffee or smeared mayonnaise on the form, but we do care about accuracy of information.

3. Start the record with the first thing you eat or drink after you get up in the morning and end with the last thing you eat or drink before bed at night.

4. Be sure to record any extra fat, flour, sugar, etc. that is added to vegetables, spread on sandwiches, or added to desserts. Whenever possible, give preparation of eggs, meat, vegetables, i.e., scrambled eggs, batter fried fish, barbequed chicken.

5. Amounts are very important!! Follow these guidelines when recording amounts eaten:

   Milk, fruit juice, and other liquids: record in terms of measuring cup or ounces (1 measuring cup = 8 ounces).

   Butter: record as level teaspoon (1 average pat = 1 level teaspoon).

   Sugar: record as level teaspoon (1 packet = 1 teaspoon).

   Egg: note whether whole egg, white or yolk only, size of egg and preparation method.

APPENDIX D (cont.)

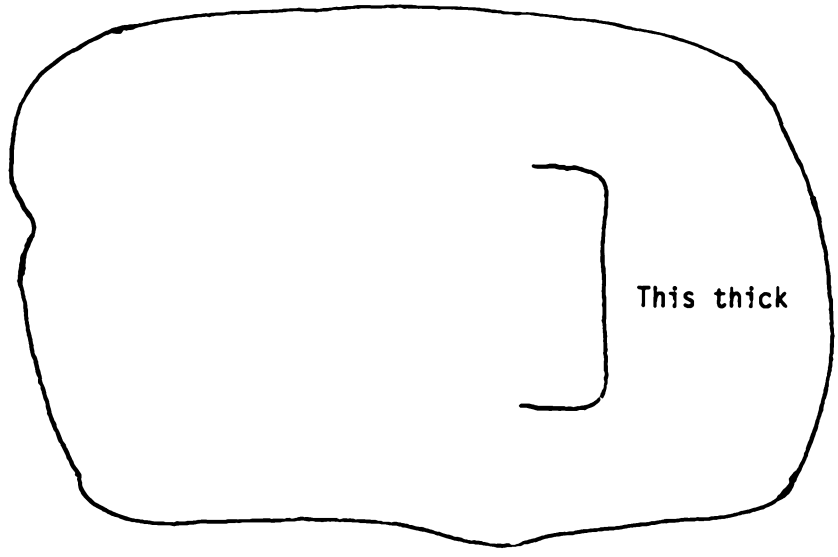
Cereal and Vegetables: record in terms of measuring cup or tablespoons (1/2 measuring cup = 8 level tablespoons). Use level tablespoons when measuring food.

Fruit: if whole, give size (small, medium, large). If sliced or canned, express in terms of measuring cup or tablespoon as above.

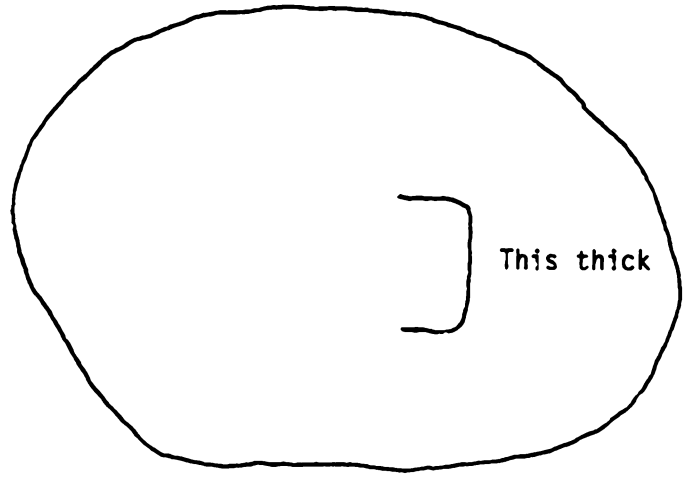
Meat: record as ounces, if possible. If not possible, record size clearly. Example: one 2-inch diameter hamburger patty.

Other foods: record as accurately as possible in terms of ordinary household measurements.

APPENDIX D (cont.)

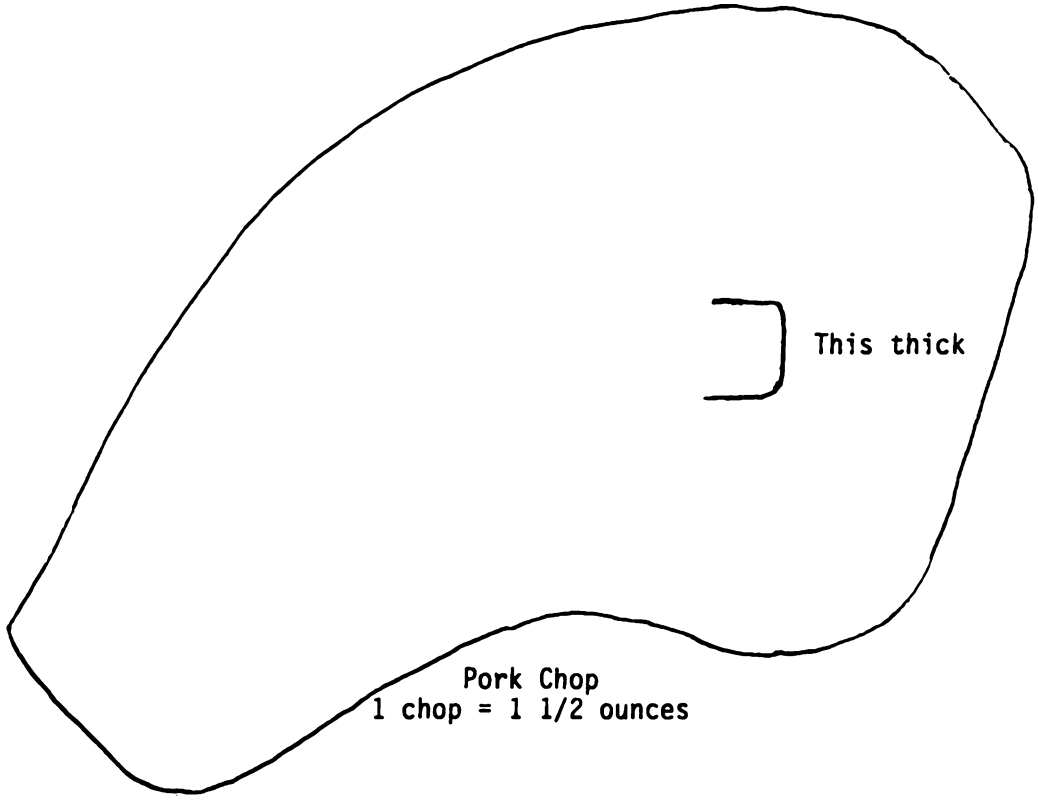


Baked Perch
1 piece = 3 ounces

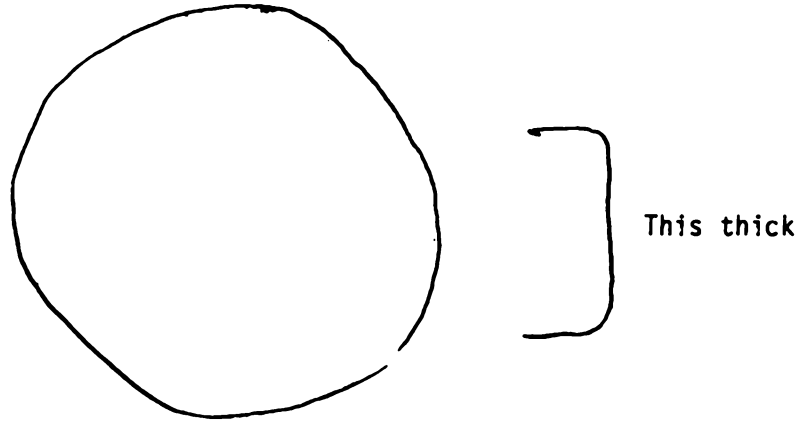


Sliced Turkey
1 slice = 2 ounces

APPENDIX D (cont.)

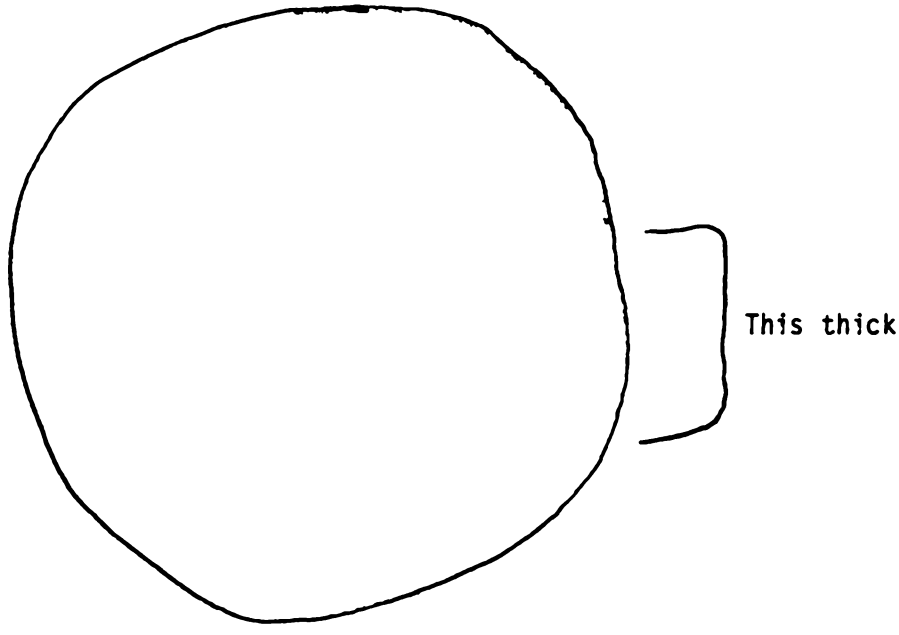


Pork Chop
1 chop = 1 1/2 ounces

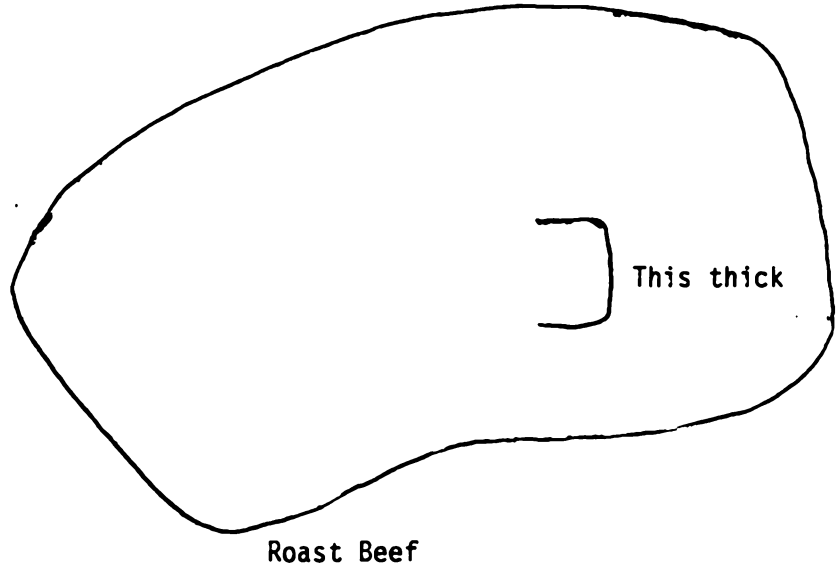


Meat Balls
2 = 3 ounces

APPENDIX D (cont.)



Hamburger Patty
1 = 3 ounces



Roast Beef
2 slices this size = 3 ounces

APPENDIX D (cont.)

SAMPLE 4-DAY FOOD RECORD

Name: _____

Date: _____

Please use a separate sheet of paper for each day. Consult the GUIDELINES FOR RECORDING FOOD EATEN when completing this record.

	Time of day	Description of food consumed	Amount of food consumed
1	8:00 a.m.	cornflakes, Kelloggs	1 cup
		1% milk	1/2 cup
		wheat toast	2 slices
		margarine	2 tsp.
		frozen concentrated orange juice	8 oz.
		banana	1/2 small
2	12:00 p.m.	turkey sandwich (2 slices bread, 2 oz. white meat turkey, 2 tsp. mayonnaise)	2
		carrot sticks	2 med.
		apple	1 med.
3	7:00 p.m.	baked chicken (white meat, no skin)	5 oz.
		1% milk	8 oz.
		green beans (steamed)	1 cup
		all vegetable tossed salad (lettuce, carrot, tomato)	1 cup
		1000 island dressing	2 tbsp.

APPENDIX D (cont.)

4-DAY FOOD RECORD

Name: _____

Date: _____

Please use a separate sheet of paper for each day. Consult the GUIDELINES FOR RECORDING FOOD EATEN when completing this record.

Time of day	Description of food consumed	Amount of food consumed
1		
2		
3		

APPENDIX D (cont.)

4-DAY FOOD RECORD

Name: _____

Date: _____

Please use a separate sheet of paper for each day. Consult the GUIDELINES FOR RECORDING FOOD EATEN when completing this record.

Time of day	Description of food consumed	Amount of food consumed
1		
2		
3		
4		
5		
6		

APPENDIX E

Subjects initial height and weights as measured at the end of phases I, II, and III of the study.

number	subject number	initial ht. (kg)	phase I wt. (kg)	phase II wt. (kg)	phase III wt. (kg).
Group 1	1	180.5	81.6	81.4	81.4
	2	174.6	63.1	59.1	59.0
	3	172.7	79.1	79.1	80.9
	4	177.8	81.0	81.4	81.5
	5	177.8	87.3	87.6	87.1
	6	172.7	76.5	76.0	75.6
	7	188.0	87.2	85.0	84.0
	8	174.0	84.5	85.4	86.5
Group II	9	175.3	77.0	78.1	79.1
	10	175.3	99.8	100.2	99.7
	11	174.0	82.2	80.2	78.1
	12	175.3	77.7	79.5	80.2
	13	180.3	74.1	72.0	72.1
	14	178.5	75.5	73.2	75.1
	15	170.2	65.6	64.6	66.0
	16	172.7	65.0	66.2	66.4
	17	177.0	64.3	64.7	63.8

APPENDIX F

Four day mean intakes of kilocalories, total fat, percent kilocalories as fat, saturated fat, percent saturated fat, and cholesterol for weeks one, seven and thirteen.

subject number	week number	kcals	total fat (g)	%kcals as fat	saturated fat (g)	% unsaturated fat	cholesterol (mg)
1	1	3220	181.2	50.6	33.3	18.4	304.9
	7	3250	204.0	57.3	36.4	17.8	293.7
	13	2275	101.8	40.3	46.3	46.5	207.9
2	1	1876	57.3	27.5	20.7	33.9	171.5
	7	1894	45.0	21.4	17.3	38.4	112.7
	13	1794	29.0	14.5	13.5	46.6	99.3
3	1	2583	168.1	58.6	58.3	35.3	171.5
	7	2286	59.4	21.6	24.4	44.4	191.7
	13	2301	85.4	33.4	36.8	43.1	330.0
4	1	1557	44.0	25.4	13.3	30.2	183.9
	7	1519	77.5	45.9	23.6	30.7	418.8
	13	1847	74.5	36.3	32.3	43.4	172.6
5	1	1308	47.4	32.6	13.4	28.3	131.0
	7	1190	28.7	21.7	8.0	27.9	159.5
	13	1441	50.1	31.3	15.0	29.9	106.2
6	1	2263	82.7	32.9	27.2	32.9	222.7
	7	2573	104.4	36.5	36.3	34.8	262.8
	13	2076	58.2	25.2	19.1	32.8	215.9

APPENDIX F (cont.)

subject number	week number	kcals	total fat (g)	%kcals as fat	saturated fat (g)	% unsaturated fat	cholesterol (mg)
7	1	2057	67.9	29.7	27.0	39.8	222.7
	7	2142	64.3	27.0	34.4	53.5	87.8
	13	2211	70.3	28.6	24.1	34.3	185.7
8	1	2523	118.6	42.3	46.5	39.2	431.3
	7	2583	86.1	30.3	40.2	46.7	231.0
	13	1912	116.4	54.8	60.2	51.7	330.9
9	1	2184	74.1	30.5	27.4	37.0	267.7
	7	2355	97.7	37.3	38.6	39.5	312.0
	13	1506	59.7	35.7	21.3	35.7	236.5
10	1	1640	58.4	32.0	20.7	35.4	184.9
	7	1294	54.2	37.7	17.8	32.8	133.4
	13	996	43.8	39.6	13.6	31.1	112.8
11	1	1384	43.2	28.1	14.3	33.1	187.6
	7	1324	46.3	31.5	15.6	33.7	124.8
	13	994	32.3	29.2	17.0	52.9	298.2
12	1	2802	83.6	26.9	26.6	31.8	348.7
	7	2985	116.8	35.2	47.7	40.8	497.5
	13	2374	121.8	46.2	38.3	31.4	559.0

APPENDIX F (cont.)

subject number	week number	kcals	total fat (g)	%kcals as fat	saturated fat (g)	% unsaturated fat	cholesterol (mg)
13	1	2033	73.0	32.3	18.4	25.2	166.6
	7	2005	57.4	25.8	23.3	40.6	183.5
	13	1704	44.0	23.2	16.4	37.3	182.5
14	1	2755	91.7	30.0	33.2	36.2	367.6
	7	2625	113.5	38.9	41.9	36.9	328.5
	13	2463	111.2	40.6	48.4	43.5	340.7
15	1	1073	52.3	43.9	16.4	31.4	115.0
	7	1405	49.0	31.4	15.9	32.4	113.7
	13	1589	60.5	34.3	19.8	32.7	162.2
16	1	2124	79.2	33.6	35.4	32.1	232.4
	7	2349	98.0	37.5	37.5	38.3	249.4
	13	1877	82.3	39.5	24.9	30.3	163.2
17	1	2162	45.9	19.1	19.2	41.8	196.6
	7	2094	62.0	26.6	20.5	33.1	193.4
	13	1403	44.7	28.7	18.8	42.1	217.3

APPENDIX G

Results of total cholesterol, triglyceride, LDL-cholesterol, and HDL-cholesterol analysis for each subject at the conclusion of phases I, II, and III.

subject number	phase	T.chol. (mg/dl)	trigly. (mg/dl)	LDL (mg/dl)	HDL (mg/dl)
1	I	207	81	153	34
	II	210	146	154	26
	III	206	133	144	35
2	I	265	125	193	47
	II	256	159	181	43
	III	237	91	170	48
3	I	274	135	190	47
	II	297	140	224	45
	III	259	169	178	47
4	I	215	175	146	34
	II	190	222	111	34
	III	229	149	160	39
5	I	202	127	135	41
	II	223	140	164	31
	III	199	107	135	42
6	I	217	84	167	33
	II	194	121	141	28
	III	194	90	143	33
7	I	296	329	177	53
	II	301	331	201	33
	III	263	234	169	47
8	I	214	145	152	33
	II	221	185	143	41
	III	257	160	190	34
9	I	201	108	131	48
	II	275	236	182	45
	III	220	128	151	43
10	I	225	120	166	35
	II	203	133	153	23
	III	186	113	128	35

APPENDIX G (cont.)

subject number	phase	T.chol. (mg/dl)	trigly. (mg/dl)	LDL (mg/dl)	HDL (mg/dl)
11	I	196	78	131	49
	II	173	63	117	43
	III	218	60	152	54
12	I	199	140	134	37
	II	220	125	167	28
	III	181	157	113	37
13	I	286	169	213	39
	II	223	152	153	39
	III	242	159	170	40
14	I	255	144	177	49
	II	260	126	198	36
	III	245	132	162	56
15	I	212	313	109	40
	II	240	211	165	32
	III	237	91	169	49
16	I	200	68	136	50
	II	188	94	120	49
	III	182	73	117	50
17	I	246	182	170	39
	II	295	205	226	28
	III	247	206	169	36

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