SERUM FATTY ACID CONCENTRATIONS IN SUBJECTS WHO HAVE RISK FACTORS OF CORONARY HEART DISEASE AND HEALTHY CONTROLS

KHAKHANANG RATANANIKOM

A THESIS SUBMITTED IN PARTIAL FULFILLMENT
OF THE REQUIREMENTS FOR
THE DEGREE OF MASTER OF SCIENCE (NUTRITION)
FACULTY OF GRADUATE STUDIES
MAHIDOL UNIVERSITY
2005

ISBN 974-04-6716-4

Thesis
Entitled

SERUM FATTY ACID CONCENTRATIONS IN SUBJECTS WHO HAVE RISK FACTORS OF CORONARY HEART DISEASE AND HEALTHY CONTROLS

Khakhanang Ratananikom

Miss Khakhanang Ratananikom
Candidate

Varapat Pakpeankitv

Asst. Prof. Varapat Pakpeankitvatana,
D.Sc.(Nutrition)
Major Advisor

Prof. Surat Komindr,
M.D.(Medicine)
Co-Advisor

Salee Kiewkarnka

Asst. Prof. Salee Kiewkarnka, Ph.D
Acting Dean
Faculty of Graduate Studies

Emorn Wasantwisut

Assoc. Prof. Emorn Wasantwisut
Ph.D.(Nutrition Biochemistry and Metabolism)
Chair
Master of Science Program in Nutrition,
Faculty of Medicine, Ramathibodi Hospital
and Institute of Nutrition

Thesis
Entitled

SERUM FATTY ACID CONCENTRATIONS IN SUBJECTS WHO HAVE RISK FACTORS OF CORONARY HEART DISEASE AND HEALTHY CONTROLS

was submitted to the Faculty of Graduate Studies, Mahidol University for the degree of Master of Science (Nutrition)

On
November 2, 2005

Khakhanang Ratananikom
Miss Khakhanang Ratananikom
Candidate

Varapat Pakpeankit
Asst. Prof. Varapat Pakpeankitvatana,
D.Sc.(Nutrition)
Chair

Surat Komindr, M.D.
Prof. Surat Komindr,
M.D.(Medicine)
Member

Paradee Temcharoen
Assoc. Prof. Paradee Temcharoen
Dr.P.H.
Member

Emorn Wasantwisut
Assoc. Prof. Emorn Wasantwisut,
Ph.D.(Nutrition Biochemistry and Metabolism)
Director
Master of Science Program in Nutrition,
Faculty of Medicine, Ramathibodi Hospital
and Institute of Nutrition

Salee Kiewkarnka
Asst. Prof. Salee Kiewkarnka, Ph.D
Acting Dean
Faculty of Graduate Studies
Mahidol University

Winit Phuapradit
Deputy Dean and Director
Acting Dean
Faculty of Medicine, Ramathibodi Hospital
Mahidol University

ACKNOWLEDGEMENT

I wish to express my sincere gratitude and deep appreciation to Assistant Professor Dr. Varapat Pakpeankitvatana, my advisor, for her supervision, constructive, valuable suggestion and consistent encouragement throughout the study.

Grateful acknowledgment is also extended to Professor Dr. Surat Kominnd, Associate Professor Dr. Paradee Temcharoen for their valuable comments and suggestion.

More sincerely, I wish to extend my gratitude to all of the staff of the research center, Ramathibodi Hospital, escecially Miss Nisakorn Tongmung for their kind assistances and suggestion.

Finally, my gratitude is express to my parents and my sister for their moral support throughout the period of my study.

Khakhanang Ratananikom

SERUM FATTY ACID CONCENTRATIONS IN SUBJECTS WHO HAVE RISK FACTORS OF CORONARY HEART DISEASE AND HEALTHY CONTROLS

KHAKHANANG RATANANIKOM 4536923 RANU/M

M.Sc.(NUTRITION)

THESIS ADVISORS : VARAPAT PAKPEANKITVATANA, D.Sc.(NUTRITION), SURAT KOMINDR, M.D. (MEDICINE)

ABSTRACT

Several studies have reported an association between dietary fat quality and risk of coronary heart disease, therefore the purpose of this study was to determine and examine the profile of serum total fatty acids concentration of n-3, n-6 and *trans* fatty acids between healthy control and subjects with various diseases. A cross-sectional study was performed to investigate 222 subjects from CEGAT study. They were divided into 38 apparently healthy groups, 11 subjects with hypertension, 25 subjects with hypercholesterol, 38 subjects with diabetes mellitus and hypertension and dyslipidemia, 78 subjects with hypertension and dyslipidemia and 37 subjects with coronary heart disease. Blood collection, lifestyle assessment, nutrition assessment and dietary assessment were investigated. Blood samples were assessed to serum lipid and serum fatty acids concentration and compare the serum lipid and serum fatty acids concentration between healthy control and subjects with various diseases.

Between healthy control and subjects with coronary heart disease, the clupadonic acuid (22:5, n-3) and DHA (22:6, n-3) in healthy control were significantly higher than subjects with coronary heart disease. The ALA (18:3, n-3) in subjects with hypertension was significantly higher than healthy control. The LA (18:2, n-6) in subjects with hypercholestserol was significantly higher than healthy control but the AA (20:5. n-3) was significantly lower than healthy control. Triglyceride in healthy control was significantly lower than subjects with diabetes mellitus and hypertension and dyslipidemia and subjects with hypertension and dyslipidemia. Cholesterol, HDL-C and LDL-C in subjects with hypercholerterol were significantly higher than healthy control. Lifestyle and dietary assessment were investigated from questionnaires and the results show that frequency of exercise influenced the level of serum HDL-C in subjects with CHD. Smoking status affected BMI in healthy controls and subjects with CHD. Alcohol intake affected BMI and WHR in subjects with HC.

KEY WORDS : CORONARY HEART DISEASE / N-3 FATTY ACIDS / N-6 FATTY ACIDS / *TRANS* FATTY ACID

84 P. ISBN 974-04-6716-4

ระดับกรดไขมันในซีรัมคนที่มีปัจจัยเสี่ยงของโรคหัวใจและหลอดเลือดและคนปกติ (SERUM FATTY ACID CONCENTRATIONS IN SUBJECTS WHO HAVE RISK FACTORS OF CORONARY HEART DISEASE AND HEALTHY CONTROLS)

คคนางค์ รัตนานิคม 4536923 RANU/M

วท.ม. (โภชนศาสตร์)

คณะกรรมการควบคุมวิทยานิพนธ์ : วราภัสร์ พากเพียรกิจวัฒนา, D.Sc. (Nutrition), สุรัตน์ โคมินทร์, M.D. (Medicine)

บทคัดย่อ

จากรายงานการศึกษาพบว่า การรับประทานไขมันที่เป็นองค์ประกอบในอาหารมีความสัมพันธ์เกี่ยวข้องกับการเกิดโรคหัวใจและหลอดเลือด ดังนั้นในการศึกษาครั้งนี้จึงมีจุดประสงค์เพื่อที่จะทำการศึกษาเปรียบเทียบปริมาณกรดไขมันชนิดกลุ่ม n-3, n-6 และ *trans* ในซีรัมของคนปกติเทียบกับกลุ่มคนที่เป็นโรคต่างๆ โดยได้ทำการศึกษาแบบตัดขวางในคน 222 คนจากกลุ่มที่ทำงานในการไฟฟ้าฝ่ายผลิต ซึ่งได้แบ่งกลุ่มตัวอย่างออกเป็นกลุ่มต่างๆ ดังนี้ คือ กลุ่มคนปกติ จำนวน 38 คน กลุ่มคนที่มีปัจจัยสี่ยงของโรคหัวใจและหลอดเลือด ที่ประกอบด้วย กลุ่มคนที่เป็นโรคความดันโลหิตสูง จำนวน 11 คน กลุ่มที่มีระดับโคเรสเตอรอลสูง จำนวน 25 คน กลุ่มที่เป็นโรคเบาหวานความดันโลหิตสูงและมีความผิดปกติของระดับไขมันในเลือด จำนวน 38 คน กลุ่มที่เป็นโรคความดันโลหิตสูงและมีความผิดปกติที่ระดับไขมันในเลือด จำนวน 78 คน และกลุ่มสุดท้าย คือ กลุ่มที่เป็นโรคหัวใจและหลอดเลือด จำนวน 38 คน หลังจากที่แบ่งกลุ่มตัวอย่างออกเป็นกลุ่มต่างๆแล้วได้ทำการศึกษาโดยเก็บตัวอย่างเลือดเพื่อวัดปริมาณไขมันและกรดไขมันที่เป็นองค์ประกอบในซีรัม ศึกษารูปแบบการใช้ชีวิต และศึกษาพฤติกรรมการรับประทานอาหาร เพื่อทำการวิเคราะห์เปรียบเทียบกลุ่มตัวอย่างที่เป็นโรคชนิดต่างๆกับคนปกติ

จากการศึกษาพบว่าระดับกรดไขมันชนิด clupadonic acuid (22:5, n-3) และ DHA (22:6, n-3) ในซีรัมคนปกติมีระดับสูงกว่าอย่างมีนัยสำคัญเมื่อเทียบกับกลุ่มคนที่เป็นโรคหัวใจและหลอดเลือด ระดับกรดไขมันชนิด ALA (18:3, n-3) ในซีรัมคนที่เป็นโรคความดันโลหิตสูงมีระดับสูงกว่ากลุ่มคนปกติอย่างมีนัยสำคัญ ระดับกรดไขมันชนิด LA (18:2, n-6) ในซีรัมคนที่มีระดับโคเลสเตอรอลในเลือดสูงมีระดับสูงกว่าคนปกติอย่างมีนัยสำคัญ ขณะที่ระดับกรดไขมันชนิด AA (20:5. n-3) ของคนกลุ่มนี้มีระดับต่ำกว่าคนปกติอย่างมีนัยสำคัญ ระดับไตรกลีเซอไรด์ในเลือดของคนปกติมีระดับต่ำกว่ากลุ่มที่เป็นโรคเบาหวาน ความดันโลหิตสูงและมีความผิดปกติของระดับไขมันในเลือด และกลุ่มคนที่เป็นโรคความดันโลหิตสูงและมีความผิดปกติของระดับไขมันในเลือดอย่างมีนัยสำคัญ i ระดับโคเลสเตอรอล HDL-C และ LDL-C ของกลุ่มคนที่มีระดับโคเลสเตอรอลสูงจะมีระดับสูงกว่าคนปกติอย่างมีนัยสำคัญ รูปแบบการใช้ชีวิตและพฤติกรรมการรับแระทานอาหารสามารถศึกษาได้จากแบบสอบถาม พบว่าความถี่ของการออกกำลังกายจะมีผลต่อระดับ HDL-C ในคนที่เป็นโรคหัวใจและหลอดเลือด การสูบบุหรี่จะมีผลต่อ BMI ของคนปกติและคนที่เป็นโรคหัวใจและหลอดเลือด และการดื่มแอลกอฮอร์มีผลต่อ BMI และ WHR ของคนที่มีระดับโคเลสเตอรอลสูง

84 P. ISBN 974-04-6716-4

CONTENTS

CONTENTS (Continued)

LIST OF TABLES

LIST OF TABLES (Continued)

LIST OF TABLES (Continued)

LIST OF TABLES (Continued)

LIST OF FIGURES

LIST OF ABBREVIATIONS

AA	arachidonic acid
ALA	alpha linolenic acid
BMI	body mass index
BP	blood pressure
c	*cis* form
CHD	coronary heart disease
DHA	docosahexaenoic acid
DM	diabetes mellitus
DLp	dyslipidemia
EPA	eicosapentaenoic acid
HC	hypercholesterol
HDL-C	high-density lipoprotein cholesterol
HT	hypertension
LA	linoleic acid
LDL-C	very low-density lipoprotein cholesterol
Lp(a)	lipoprotein (a)
MUFA	monounsaturated fatty acid
PUFAs	polyunsaturated fatty acids
TC	total cholesterol
TG	triglyceride
18:1, *t*	elaidic acid
µl	microliter

CHAPTER I
INTRODUCTION

Coronary heart disease (CHD) is the main cause of death in United State (1), most Latin American countries, Canada (2), Western Europe (3) and several developing countries including Thailand (4). The number of death were 32231 persons in 2000 and 34903, 32896 persons in 2001 and 2002 respectively (5). Although the trend of the mortality rate from coronary heart disease was decreased from 2001 to 2002, however it is still leading cause of death in Thailand.

The Framingham heart study has contributed importantly to the understanding of the cause of coronary heart disease. They are called cardiovascular risk factors (6). The major risk factors included high blood pressure, abnormal blood lipid concentration, diabetes mellitus, smoking (1, 7) and factors other than those risk factors was abdominal obesity, inadequate levels of physical activity and family history of coronary heart disease (7-9). Numerous studies indicated that lifestyle change and nutritional habit are believed to be the major factors to reduce mobid from coronary heart disease (10). It was found that Japanese risk factors of coronary heart disease, other than dietary intake, were the same as for Europeans and Americans which had the high death rate by coronary heart disease, resulting in a lower death rate of coronary heart disease (11). But now, the Japanese lifestyle especially eating habit is changing to western style diet, resulting in a higher death rate due to coronary heart disease (12). There were data to support dietary fat quality and development and prevention of coronary heart disease. They found the protective effects of long chain n-3, n-6 fatty acids on coronary heart disease (13-15) whereas a diet high in saturated fats and *trans* fats was associated with increase risk of coronary heart disease (16-18).

Numerous studies reported that populations who consume large amounts of marine foods have a low prevalence of arterial thrombosis and cardiovascular disease (19-21). The data from epidemiological studies showed an inverse relation between fish consumption and death from coronary heart disease (22-

24). These beneficial effect are attributed to the n-3 fatty acids especially docosahexaenoic acid (DHA) and eicosapentaenoic acid (EPA), the main dietary sources are fishes and marine mammals. The positive effect of n-3 fatty acids are implicated in the pathogenesis of artherosclerosis, thrombotic disease, including act on lowering triglycerides and very low-density lipoprotein (VLDL) cholesterol (19). In addition, the study from Schmidt and Dyerberd,1994 (25) indicated that n-6 fatty acids played important roles in a wide variety of biological function because they are incorporated into cell membrane where they affect membrane characteristics and give to biological active substance. The Nurses Health Study (NHS) found that n-3 and n-6 fatty acids were strongly protective against coronary heart disease. They explained the possible mechanisms of both types of polyunsaturated fatty acids which included the reduction in LDL-C, total cholesterol to HDL-C ratio and the suppression of cardiac ventricular arrhythmias (26).

Trans fatty acids intake has been associated with an increased risk of coronary heart disease (27-28). Several studies found that the *trans* fatty acids affected on plasma lipoproteins and promoted coronary heart disease by raisesing low-density lipoprotein cholesterol (LDL-C), triglycerides (329-30) and lipoprotein (a) [Lp (a)] plasma concentrations (31) and decreasing high-density lipoprotein cholesterol (HDL-C) (29-30).

CHAPTER II
LITERATURE REVIEWS

Coronary Heart Disease (CHD)

Cardiovascular disease (CVD) is the general term for all disease of the heart and blood vessels. Coronary heart disease (CHD) is the blood vessel disease in the heart results in coronary event which commonly called acute coronary syndrome, acute myocardial infarction. Coronary artery disease (CAD) and ischemic heart disease are other names for coronary heart disease.

Coronary heart disease (CHD) accounts for about one-quarter of all deaths worldwide, with is the highest proportion of all causes of death. They are sometimes referred to as “killer number one”. In the developing countries, about one-half of all deaths are caused by CHD. In Thailand, an agricultural and developing country. The occurrence of CHD among urban population seems to be progressively increased each year. This is, at least, due to the change of life style towards westernized-society pattern (32-34).

Coronary heart disease (CHD) is closely associated with advanced atherosclerosis, with reflects several deteriorative phenomena (involving interaction between plasma lipids, lipoproteins, monocytes, platelets and endothelium and smooth muscle of arterial wall) that gradually result in narrowing of coronary arteries, terminating in thrombosis and coronary infarction. (35-37)

Atherosclerosis

Coronary atherosclerosis, the major cause of CHD, is the characterized histologically by the accumulation of lipid, predominantly cholesterol, in the arterial wall together with a local connective tissue reaction. Four points related to atherosclerotic lesion are brought up here.

Both the early and advanced lesion of atherosclerosis consists of smooth muscle cells and macrophages. Macrophages predominate in fatty streaks,

which may represent a form of inflammatory response, whereas advanced lesions contain a fibrous cup of smooth muscle that covers proliferated smooth muscle and macrophages and variable numbers of other types of leukocytes, including lymphocytes.

Diet-induced hypercholesterolemia leads to changes in endothelium and monocytes that result in increased monocyte adherence, subendothelial migration, accumulation of lipid to form foam cells, and gradual accumulation of smooth muscle to form fatty streaks (38-39).

All four principle cells are involved in atherosclerosis, i.e., endothelial cells, smooth muscle cells, platelets and monocyte/macrophages. For instance, platelets contain or can synthesizes and release chemoattractents and growth factors, at least one of which is identical or very closed related to platelet-derived growth factor. It is thus possible that injury or other stimulus may induce growth-factor release and autocrine or paracrine stimulation of cell in the artery.

A modified response to injury hypothesis of atherosclerosis suggests that at least two pathways may lead to formation of intimal smooth muscle proliferation lesions. The first pathway, demonstrated in hypercholesterolemia, involves monocyte and possibly platelet interactions, which may stimulate fibrous plaque formation by growth-factor release from the different cells. The second pathway involves direct stimulation of endothelium, which may release growth factors that can induce smooth muscle migration and proliferation. This pathway may be important in diabetes mellitus, hypertension, cigarette smoking or other circumstance associated with an increased incidence of atherosclerosis (40-41).

Scientific studies in laboratory, clinic and population have pointed to the contributing roles of dyslipidemia including high serum total cholesterol (TC), low serum high density lipoprotein-cholesterol (HDL-C) and high serum triglyceride (TG) level, hypertension, cigarette smoking, overweight, DM and impaired glucose tolerance, physical inactively in the genesis of CHD. The risk factors represent statistical diagnosis which are accurate for large populations but which may have less predictive value for individuals (7-9).

Fatty acid

The diet generally includes at least 30 percent of calories as fat, mostly from triglycerides in oils, margarine, dietary products and meats. Triglycerides are the most abundant in foods and they are esters of fatty acids and glycerol. One, two, or three fatty acids may be attached to the glycerol backbone, resulting in mono-, di- or triglycerides respectively. Because more than 90 percent of triglyceride molecule is accounted for its 3 fatty acids, so fatty acids are the major dietary lipid.

Chemical structures of fatty acids share some common characteristic. Fatty acids are composed of a chain carbon atoms, with a methyl (CH_3) group at one end and a carboxyl (COOH) group at the other. The number of carbon atoms in common fatty acids ranges from 4 to 22, and they can be linked by single of double bond, as can be seen in the following representation of butyric acid (Figure 1), a short-chain fatty acid found in butter (42).

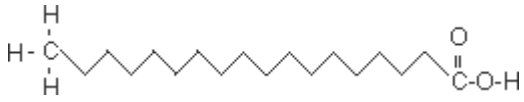
Figure 1 Butyric acid

Fatty acids differ from each other most significantly in chain length, position and configuration of the double bond. Fatty acids with twelve or more carbon atoms are referred to as long-chain fatty acids and are typical of fats. The predominance of short to medium chain fatty acids (4-6 C or 8-12 C) contributes lower melting points and accounts for relative softness of butterfat and coconut oil. The attached fatty acids give lipid their special characteristics (43).

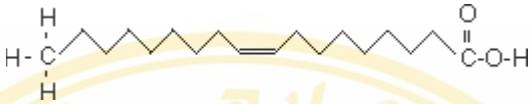
.

Type of fatty acids

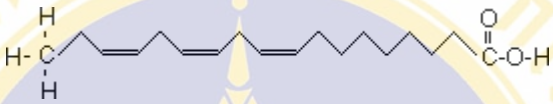
Fatty acids can be grouped in families according to their number of double bonds, they can also be classified into three main group: saturated fatty acids (SFAs), monounsaturated fatty acids (MUFAs) and polyunsaturated fatty acids (PUFAs). (Figure 2)



saturated fatty acids (SFAs)



monounsaturated fatty acids (MUFAs)



polyunsaturated fatty acids (PUFAs)

Figure 2 three type of fatty acids

Saturated fatty acids (SFAs) (44)

Saturated fatty acids (SFAs) contain carbon atoms only by single bonds and are usually solid at room temperature. They are principally obtained from animal fats and animal products (eg. Meat fat, lard dripping, milk, butter, cheese and cream). Food of plant origin generally have a much lower content of SFAs, although there are some exception, such as coconut and palm oil. Margarines and fat spreads made from vegetable oils also contain significant amounts of SFAs.

Monounsaturated fatty acids (MUFAs) (44)

Monounsaturated fatty acids (MUFAs) contain only one double bond and are usually liquid at room temperature. The most concentrated dietary sources of MUFAs are olive oil and rapeseed oil. However, MUFAs also comprise about one third of fatty acids content in meat fat and most of fat present in nuts and seeds.

Polyunsaturated fatty acids (PUFAs) (43-46)

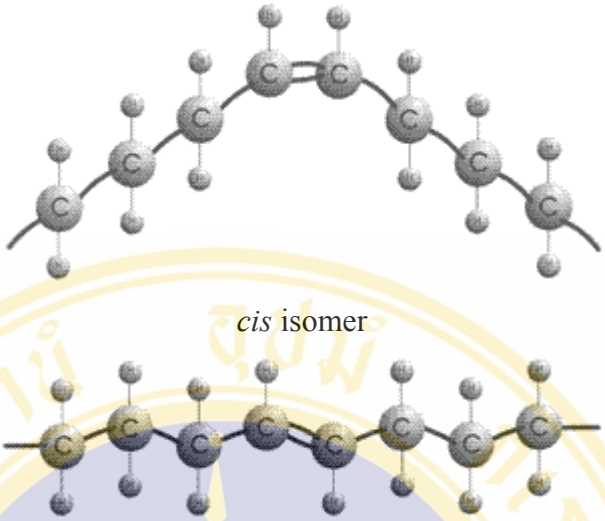
Polyunsaturated fatty acids (PUFAs) contain two or more double bonds and are liquid at room temperature. They can be classified by the omega classification into four families. The omega group is determined by the number of carbons between

the methyl end of the molecule and the nearest double bond. For example, if the double bond nearest the methyl end of the molecule is 3 carbons from the methyl end, the fatty acids is known as an omega-3 or ω-3 (n-3) fatty acids. The four families of polyunsaturated fatty acids are omega-3 or ω-3 (n-3) fatty acids, omega-6 or ω-6 (n-6) fatty acids, omega-7 or ω-7 (n-7) fatty acids and omega-9 or ω-9 (n-9) fatty acids.

There are four parent fatty acids of this four families, α-linolenic acid (C18:3, ω-3), linoleic acid (C18:2, ω-6), palmitoleic acid (C16:1, ω-7) and oleic acid (C18:1, ω-9). Interconversion among there four families of unsaturated fatty acids does not occur in humans. However, the enzymes for the desaturation and elongation reactions, by which the parent fatty acids are converted via CoA esters, to their derivative fatty acids are shared by all four families.

The parent fatty acids in the ω-3and ω-6 groups; α-linolenic acid (C18:3), and linoleic acid (C18:2) are also called “essential fatty acids” (EFAs) because we lack the enzymes to make them and must, therefore, get them form our diet. EFAs are principle derived from vegetable oils nuts and seeds.

In addition, the double bond can be located anywhere along the length of the molecule becomes more rigid at this point, and has two types of isomers, positional and geometric; *cis trans*. The cis from; the hydrogen atoms on either side can point in the same direction or *trans* from; the hydrogen atoms on either side can point in the opposite direction. The present of double bond and its spatial configuration has a remarable influence on the melting point. In nature, most fatty acids are *cis* isomer. (Figure 3 *cis* and *trans* configulation)



cis isomer

trans isomer

Figure 3 *cis* and *trans* configulation

***Trans* fatty acids** (45)

Trans fatty acids are unsaturated fatty acids with at least a double bond in the *trans* configuration, resulting in a more rigid molecule close to a saturated fatty acids. The most common trans fatty acids are monounsaturated, but various diunsaturated *cis*, *trans* and *trans*, *cis* isomer also occur. *Trans* fatty acids appear in dairy fat because of ruminal activity, and in hydrogenated oils; margarines, shortening and baked goods contain relatively high levels of trans fatty acids.

Trans fatty acids produce a straight fatty acids chain and more closely packed configuration of the resulting straight-chained triglycerides. As a consequently of these changes, the 18 carbon oleic acids (contain 1 cis bond) has a melting point of 13 °C whereas the 18 carbon elidic acids (containing 1 trans bond) has a melting point of 44 °C. By comparion, the melting point of the carbon fully saturated strearic acids 70 °C.

Dietary fatty acids (44, 46)

The fatty acid composition of various substances in listed in Table 1. Most food fats consist predominantly of long-chain fatty acids (16 or more carbons), with butter, milk (including human milk) and milk products, and , particularly,

coconut oil, containing significant amounts of short- and medium-chain fatty acids as well. Animal products generally contain significant amounts of saturated and monounsaturated fatty acids, with lesser amounts of PUFA. Vegetable products, on the other hand, except for coconut oil, contain higher proportions of the long-chain unsaturated oleic and linoleic acids and lessee amounts of saturated fatty acids. Although coconut oil is largely saturated, it consists for the most part of medium or short-chain fatty acids containing 12 or fewer carbon atoms, and is therefore liquid.

Table 1 Dietary fatty acids

Type of fatty acids	Dietary source
Saturated fatty acids	
Butyric acid (4:0)	Dietary produce, coconut oil
Caproic acid (6:0)	Dietary produce, coconut oil
Caprylic acid (8:0)	Dietary produce, coconut oil
Capric acid (10:0)	Dietary produce, coconut oil
Lauric acid (12:0)	Dietary produce, coconut oil, palm kernel
Myristic acid (14:0)	Dietary produce, coconut oil, palm kernel
Palmitic acid (16:0)	Palm oil, cottonseed oil, butter, meat fat
Stearic acid (18:0)	Butter, meat fat, lard, chocolate
Arachidic acid (20:0)	Nut and seed oils
Behenic acid (22:0)	Peanuts, peanut oil
Lignoceric acid (24:0)	Peanut
Monounsaturated fatty acids	
Oleic acid (18:1,n-9)	Olive oil, nut and seed oils, meat fat, butter, eggs, avocados
Elaidic acid (*t*18:1, n-9)	Hydrogenated fats
Transvaccenic acid (*t*18:1, n-7)	Hydrogenated fats, butter
Erucic acid (22:1, n-9)	Rapeseed oil

Table 1 Dietary fatty acids (continues)

Type of fatty acids	Dietary source
Polyunsaturated fatty acids	
Linoleic acid (18:2, n-6)	Nut, seeds vegetable oils (Sunflower, safflower, corn, soyabean) lean meat, eggs
Alpha-linolenic acid (18:3, n-3)	Nuts, seeds vegetable oils (flaxseed, hemp, pumpkin, walnut, rapeseed, soybean)
Gamma-linoleic acid (18:3, n-6)	Evening primrose oil, borage oil, blackcurrant oil
Dihomo-γ-linoleic acid (20:3, n-6)	Shark liver
Arachidonic acid (20:4, n-6)	Lean meat, game, eggs
Eicosapentaenoic acid (20:5, n-3)	Oily fish (herring, mackerel, tuna, salmon, sardines)
Clupadonic acid (22:5, n-3)	Seafood
Docosahexaenoic acid (22:6, n-6)	Fish, liver, egg yolk

Metabolic effect of dietary fatty acids on coronary heart disease

Dietary fat plays important role in heart disease by affecting atherogenesis, thrombosis and coronary infraction (47) because, many researches found that dietary fat quality has been shown to be associated with development and prevention of coronary heart disease (15). They found that a diet high in saturated fats and trans fats also have been associated with increase risk of coronary heart disease but n-6 fatty acids and n-3 fatty acids collectively protect against coronary heart disease (48).

Saturated fatty acids

The most common dietary saturated fatty acids are palmitic and stearic acid. These are important for energy metabolism, cell membrane structure and normal growth. Saturated with 20 to 24 carbon atoms are important constituents of myelin. However, the requirement for saturated fatty acids can be met by endogenouse synthesis and it seems unlikely that a dietary source is necessary. Furthermore, chronic excessive intake or synthesis, or both, of palmitic and stearic acids tends to raise the level of LDL cholesterol in blood (44). Nicolosi et al., 1990 reposted that the increase in total cholesterol concentration in plasma induced by saturated fatty acids occurs primary in the low-density lipoprotein cholesterol (LDL-C) fraction, and it appears to act by suppressing the expression of LDL recepters. Therefore, a high intake of saturated fatty acids could enhance the process of atherogenesis and increase the risk of cardiovascular disease (49).

N-3 fatty acids

Alpha-linolenic acid (ALA, 18:3 n-3) is the primary essential fatty acid that can be elongation and desaturated to form highly specialized 22-carbon fatty acids with the n-3 family. Adequate intake of ALA and long-chain n-3 fattty acids is especially important for infants, young children and patients requiring parenteral and enteral nutrition. In animals and humans, ALA can be metabolized to long-chain polyunsaturated n-3 fatty acids, including eicosapentaenioc (EPA) and docosahexaenoic acid (DHA) through the desaturation-chain elongation pathway. Common available dietary sources of EPA and DHA are fish and seafood. Recent

human studies suggest that dietary ALA and its subsequent metabolic and products may protect against coronary heart disease (48).

A low rate of cardiovascular disease in population with very high intake of fish, such as Alaskan Native Americana, Greenland Eskimos and Japanese living fishing villages suggest that fish oil may be protective against atherosclerosis. Subsequent prospective cohort studies have found an inverse association between fish consumption and risk of cardiovascular mortality in diverse population. The potential benefits of marine n-3 fatty acids include a reduction triglyceride level, reducing platelet aggregation and anti-arrhythmic effects. However, this can be accompanied by increase in total and LDL cholesterol in patients with low LDL levels and hypertriglyridemia (50).

In addition, Bruce J. Holub, 2002 reported that the beneficial effects of n-3 fatty acids on cardiovascular disease are mediated by both eicosanoid-depent and eicosanoid-independent processes. For example, the reduced blood platelet reactivity (antitrombotic effect) observed with increase EPA and DHA intake involves the reduced formation of proaggregatory eicosanoid known as thromboxane A_2 (TxA_2). The replacement of AA (the n-6 fatty acid and TxA_2 precursor) in blood platlet membrane phospholipids by EPA and DHA yield less TxA_2 upon platelet stimulation; furthermore, EPA has an inhibitory effect on the cyclo-oxygenase enzyme that connects AA to TxA_2 thus leading to a lessened thrombogenic state (51).

They summarized the report that, the various mechanisms by witch increased dietary consumption of omega-3 fatty acids from fish and fish oil are considered to favorably modify cardiovascular disease and associated disorders as in Table 2 (51).

Table 2 Mechanisms for the cardioprotective effects of EPA and DHA

Mechanisms for the cardioprotective effects of n-3 fatty acids
• Reduction in malignant ventricular arrhythmias (via enrichment of cardiac lipid in EPA and DHA)
• Increase in heart rate variability (possibly via increased parasypmpathetic tone, altered cytokine levels, other factors)
• Antithrombotic effects and other effects on the hemostatic system (e.g., reduced blood platalat reactivity, moderately longer bleeding times, reduced plasma viscosity)
• Lipid lowering (reduction in fasting triglyceride and VLDL levels often accompanied by a moderate raise in HDL cholesterol, attenuation of postpranial triglyceride response)
• Improved endothelial relaxation (via enhancement of nitric oxide-dependent and nitric oxide-independent vasodilation)
• Inhibitory effect on atherosclerosis and inflammation (via inhibition of smooth-muscle cell proliferation, altered eicosanoid synthesis, reduced expression of cell adhesion molecules)
• Suppressed production of inflammatory cytokines (interleukins, tumour necrosis factor) and mitogens

In addition, Weber and Leaf, 1991 reported the hypolipidemic, antithrombotic and anti-inflammatory effects of n-3 fatty acids that have been studied extensively in animal models, tissue cultures and cell in table 3 (52).

Table 3 Effects of n-3 fatty acids on factors involved in the pathophysiology of atherosclerosis and inflammation

Factor	Function	Effect of n-3 fatty acid on factor concentration
Arachidonic acid	Eicosanoid precursor , aggregateds plateleta and stimulates white blood cells	increased
Thromboxane A_2	Platalet aggregation, vasoconstriction, increase intracellular Ca^{2+}	decreased
Prostaacyclin	Prevents platelet aggregation, vasodilator, increase cyclic AMP	increased
Leukotriene B_4	Neutrophil chemoattractant increases intracellular Ca^{2+}	decreased
Tissue plasminogen activator	Increase endogenous fibrinolysis	increased
Fibrinogen	Blood clotting factor	decreased
Red blood cell deformability	Decrease tendency to thrombosis and improves oxygen delivery to tissues	increased
Platelet activating factor	Activates platelets and white blood cells	decreased
Platelet-derived growth factor	Chemoattractant and mitogen for smooth muscles and macrophages	decreased
Oxygen free radicals	Causes cellular damage, enhances LDL uptake via the scavenger pathway, stimulates arachidonic acid metabolism	decreased
Lipid hydroperoxides	Stimulates eicosanoid formation	decreased
Interleukin I and tumor necrosis factor	Stimulate neutrophil oxygen free radical formation, lymphocyte proliferation and platelet activating factor; expresses intercellular adhesion molecule I on endothelial cells; and inhibits plasmiogen activator and thus is procoagulant	decreased
Endothelial-derived relaxation factor	Reduces arterial vasoconstrictor response	increased
VLDL	Related to LDL and HDL concentrations	decreased
HDL	Decreases the risk of coronary heart disease	increased
Lipoprotein (a)	Atherogenic and trombogenic	decreased
Triglyceride and chylomicrons	Contribute to postprandial lipemia	decreased

N-6 fatty acids

Linolaic acid (LA, 18:2 n-6) is the primary essential fatty acid and represents the basis of the n-6 family. It is the major polyunsaturated fat in the diet and comes from vegetable oils, including corn, soybean, safflower and sunflower. It can be elongated and desaturated form 20 carbon fatty acids, such ad arachidonic acids (AA, 20:4 n-6), which together function via structural lipid and cell signals to control numerous cell activities responsible for daily living. In fact, epidemiological evidence supports a role for dietary n-6 fatty acids in reducing the risk of coronary heart disease and a cross-population study in healthy men from European populations found that higher adipose tissue LA, a marker of dietary LA intake, was associated with lower coronary heart disease mortality. Further, serum linoleic acid was negatively related to cardiovascular death in postinfarction and middle-aged men, while low dietary LA intake predisposed to myocardial infarction. Therefore, n-6 fatty acids appear to be strongly protective against coronary heart disease (48-53).

N-6 polyunsaturated fat may have beneficial effects on cardiovascular disease besides improving lipid profile. In animals and metabolic studies, an increased intake of n-6 fatty acids improve insulin sensitivity (54-55). Possible mechanisms of n-6 fatty acids include reduction plasma total cholesterol (TC) and low-density lipoprotein cholesterol (LDL-C), each of which is an established coronary heart disease risk factor. N-6 fatty acids might lower LDL levels by inbibition of hepatic synthesis of apoB-containing lipoproteins. Decease synthesis of VLDL-apoB, for example, should ultimately reduce formation of LDL apoB (56-58).

Following, Simopoulos report in 1999 that, a diet rich in n-6 fatty acids shifts the physiological state to one that is prothrombotic and proaggregatory, with increases in blood viscosity, vasospasm and vasoconstriction and decreases in bleeding time. Because, bleeding time is shorter in groups of patients with hypercholesterolemia, hypoproteinemia, myocardial infarction, other forms of atheroasclerotic disease, type 2 diabetes, obesity and hypertriglyceridemia (59).

In addition, Vasuki Wijendran and K.C. Hayes summaried the effect of n-3 and n-6 fatty acids on cardiovascular risk factors as seen in Table 4 (48).

Table 4 Biological effects of n-3 and n-6 fatty acids on cardiovascular risk factors

Fatty acids	Parameters	Effects
Linoleic acid	Plasma lipids	
	TC	Decreased
	LDL-C	Decreased
	HDL-C	Increased
	LDL-C:HDL-C	Decreased
	Hepatic LDL-C clearance	Increased
	Hepatic LDL-C production	Decreased
Linolenic acid	Plasma lipids	
	LDL-C	Decreased
	HDL-C	Decreased
	LDL-C:HDL-C	Increased
	Arrhythmia	Decreased
	Thrombosis	Decreased
EPA+DHA	Plasma lipids	
	TC	Decreased
	LDL-C	Increased
	HDL-C	Increased
	LDL-C:HDL-C	Decreased
	Hepatic TG and apo B recretion	Decreased
	Arrhythmia	Improved
	Vascular endothelial function	
	Blood pressure	Decreased
	Proinflammatory factors	Decreased

Trans fatty acids

Trans fatty acids are unsaturated fatty acids with at least a double bond in trans configuration. The physical properties of *trans* fatty acids are intermediate between *cis* and saturated fatty acids, but a *trans* double bond is chemically less reactive than a *cis* double bond. Biochemical data indicate that *trans* fatty acids are subject to the same metabolic control mechanisms that regulate the metabolism of saturated and *cis*-isomeric fatty acids.

Concerns have been raised for several decades that consumption of *trans* fatty acids might have contributed to the 20th century epidemic of coronary heart disease and many studies such as epidemiologic, metabolic and clinical studies found that *trans* fats have also been associated with an increase risk of coronary heart disease. For example, based on the available metabolic studies, we estimated in a 1994 report that approximately 30,000 premature coronary heart disease deaths annually could be attributable to consumption of *trans* fatty acids and both adipose and plasma *trans* fatty acids level reflect dietary intake of them (60).

A higher intake of *trans* fat can contribute to increase risk of coronary heart disease through multiple mechanisms. First, *trans* fatty acids raise LDL cholesterol similarly to saturated fat, but unlike saturated fat, they also decrease HDL cholesterol. As a result, the net effect of *trans* fat on the LDL/HDL cholesterol ratio is approximately double that of saturated fat. Second, trans fat increase lipoprotein (a) [Lp(a)] level, which are positively associated with risk of coronary heart disease. Third, *trans* fat raises plasma triglyceride level and increase triglycerides are independently associated with increased risk of coronary heart disease. Fourth, *trans* fatty acids can adversely affect essential fatty acid metabolism and prostaglandin balance by inhibiting the enzyme delta-6-desaturase and, as a result, may promote thrombogenesis. Finally, recent data have suggested that high intake of *trans* fat may promote insulin resistance in human. In addition, data from rats have suggested that this may be a variable with respect to *trans* fatty acid intake and some measures of immune function (31, 50, 61).

Literature reviews of n-3, n-6 and *trans* fatty acids on coronary heart disease

Dietary fat quality has been shown to be associated with develop and against coronary heart disease because, fatty acids composition of dietary fat markedly influences the fatty acids composition of serum lipid (62). In addition, Arja T Erkklä et al, 2003 report that we can use the content of fatty acid in plasma or adipose tissue as the biomarkers of intake of dietary fat.

N-3 fatty acids

There is some evidence from epidemiological data that intake of n-3 fatty acids is associated with a reduced risk of coronary heart disease. This was originally found in Greenland Eskimos and later a low occurrence of coronary heart disease was also reported in other populations with diets in rich in seafood in Alaska, Japan and China. In Western populations, with an average intake of n-3 fatty acids well below 1 g/day, an inverse correlation between coronary heart disease and fish consumption was reported (63).

In 1971, Bang and Dyerberg reportedthat in Eskimos, serum total cholesterol, triglycerides, low-density lipoprotein (LDL), very low-density lipoprotein (VLDL) were lower, but increased levels of high-density lipoprotein (HDL) when compared with those in the Danes. Fewer Eskimos had high plasma levels of lipoprotein (a) [Lp(a)] above 30 mg/dl, while their median Lp(a) did not differ from Danes (64-65)

Kromhout et al, 1985 retrospectively reviewed the Zutphen dietary study in which 852 middle-age men who did not have coronary artery disease on enrollment were followed for 20 year. An inverse relation was found between the amount of fish eaten weekly and the mortality from coronary artery disease. As little as 35 g of dietary fish per day resulted in about a 50~ reduction in mortality from coronary artery disease (23). A reexamination of the Western Electric Study revealed a similar inverse relation between the ingestion of fish and mortality from coronary artery disease (25). A large Swedish study also found a reduction of coronary artery disease among the subjects consuming a large amount of fish (66)

Many studies investigated the correlation of n-3 fatty acids and coronary heart disease. Dewailly et al, 2002 (67) examined the profile of plasma

phospholipids concentration of n-3 fatty acids, eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) among 917 subjects in James Bay Cree and verify the relation between these concentration and cardiovascular disease risk factors. In plasma n-3 fatty acids were higher among coastal resident than among inland residents. A positive association was observed between plasma high-density lipoprotein (HDL) and n-3 fatty acids. Eicosahexaenoic acid (EPA) and eicosahexaenoic acid (EPA) + docosahexaenoic acid (DHA) were inversely association with triglycerides. From the results, they concluded that n-3 fatty acids may favorably influence some cardiovascular risk factors. This research has conclusion similar to another study from Dewailly in 2001(68). They concluded that concentrations of n-3 fatty acids were positive association with fish intake and they found positive associations between eicosahexaenoic acid (EPA) and total cholesterol, HDL cholesterol, plasma glucose and systolic and diastolic blood pressure.

In 2001, Eric Dewaily et. al, verify the relation between plasma phospholipids concentrations if n-3 fatty acids eicosapentaenoic acid (EPA) and docosahexsaenoic acid (DHA) and various cardiovascular diseases risk factors among the 426 Inuit of Nunavik, Canada. The results expressed as the percentage of total fatty acids, geo metric mean concentrations of EPA, DHA and their combination in plasma phospholipids were 1.99% and 6.38% respectively. N-3 fatty acids were positively associated with HDL-cholesterol concentrations and inversely associated with triglyceride concentration and ratio of total to HDL cholesterol. In contrast, concentrations of total cholesterol, LDL cholesterol and plasma glucose increase as n-3 fatty acid concentrations increased. There were no significant associations between n-3 fatty acids and diastolic and systolic blood pressure and plasma insulin. And they concluded the report that traditional Inuit diet, which is rich in n-3 fatty acids, is probably responsible for the low mortality rate from ischemic heart disease in this population (69).

Erkkila et al, 2003 (15) tried to test hypothesis that a high proportion of n-3 fatty acids in serum lipid would be associated with reduced risk of death and coronary events in patients with established cardiovascular disease. After Follow-up 285 men and 130 women with cardiovascular disease, they found a high proportion of eicosahexsaenoic acid (EPA) was associated with a lower risk of death. They

concluded that high proportion of n-3 fatty acids in serum lipids are associated with a substantially reduced risk of death. And the study from Yamori Y. et al, 1985 (11) report that the percentage of n-3 fatty acids, i.e., eicosahexaenoic acid (EPA) and docosahexaenoic acid (DHA) was significantly low in United State inlanders with high coronary heart disease morbidity compared with fishing and farming Japanese with low morbidity.

P. Yli-JAMA et al., 2002 (70) investigated the associated between composition of serum free fatty acids fraction and risk of first myocardial infarction (MI) in 103 patients with first MI and 104 population control from Ullevål Hospital in Oslo and Østfold Central Hospital in Fredrikstad and Sarpsborg, Norway. As the result, they found that the mean percentage content of docosahexaenoic (DHA), eicosapentaenoic (EPA), stearic and myristic acid in serum free fatty acid fraction was significantly lower in cases than in controls, whereas that of oleic and linoleic acid was higher in cases. Increased percentage content of total n-3 fatty acids in serum free fatty acid fraction was associated with decreased risk of MI.

Following Hongtong K. et al, 2003 (71), they studied platelet fatty acids in coronary heart disease and cases. The cross sectional study was performed to investigate 250 volunteers from Pramongkutklao Hospital, Samphanthawong district, Wat Chaiyapreukmala and at pradoo in Taling Chan district. They found Platelet fatty acids levels were found to have no significant difference between the different male groups. In female groups, the alpha-linoleic (ALA) in hypertension was significantly higher than in coronary heart disease ($p<0.05$), whereas the arachidonic acid (AA) level in hypertension was significantly higher than in the healthy females ($p<0.05$). Positive correlations were shown between alpha-linoleic acid (ALA) and eicosapentaenoic acid (EPA), arachidonic acid (AA) and docosahexsaenoic acid (DHA), alpha-linoleic acid (ALA) and the diastolic blood pressure, docosahexsaenoic acid (DHA) andtotal cholesterol (TC), and between low-density lipoprotein cholesterol (LDL-C) and plasma glucose.

In addition, Hojo N. et al., 1998 (72) compared the serum lipid profiles of patients with or without significant coronary stenosis and control subjects. They founded that level of high-density lipoprotein cholesterol and eicosapentaenoic acid were significantly lower in patients with significant coronary stenosis than in the

control subjects. It suggests that high-density lipoprotein cholesterol and eicosapentaenoic acid may have a protective effect on the pagress of coronary atherosclerosis.

N-6 fatty acids

Following the retrospective study, Lewis, 1958 (73) and Kingsbory et al, 1962 (74) reported the fatty acids composition of blood lipids in 12 and 9 coronary heart disease patients respectively and found significantly lower linoleic acid in cases when compared with healthy volunteers. And Schrade et al, 1960 (75) reported that linoleic acid in serum, cholesterol esters and phospholipids was significantly lower in atherosclerotic patients when compared with healthy controls.

In 1997, Brouwer DA et al. (76) invertigated the dietary linoleic acid intake of 57 patients with coronary artery disease (47 males, 10 females; 61+/-10 years) in Curacao is higher as compared with 77 controls (51 males, 26 females; 56+/-7 years). For this, we measured plasma cholesterol ester fatty acids, which reflect the dietary fatty acid composition of the preceding weeks. Patients with coronary artery disease and controls had differences in cholesterol ester fatty acids. This cholesterol eater linoleic content suggest that the dietary polyunsaturated/saturated fatty acid ratio is far below 1. Comparison with data reported for The Netherlands, Greenland and Crete showed that the dietary fatty acid composition in Curacao is typically Western with a high intake of saturated fatty acids, a low intake of monounsaturated fatty acids and the consumption of linoleic acid as the predominant polyunsaturated fatty acid. And they concluded that reduction of dietary saturated fatty acids and increase of the alpha-linolenic/linoleic acid ratio are likely to be benefit to both primary and secondary prevention from coronary artery disease in Curaco.

In addition, Miettinen TA et al., 2003 (77) examined the fatty acids composition of serum lipids predicts myocardial infarction, a follow-up of five to seven years 33 out of 1222 middle-aged men initially free of coronary heart disease sustained fatal or non-fatal myocardial infarction or died suddenly. The fatty acid composition of serum triglycerides, phospholipids and cholesterol esters had been measured at the start of the surveillance in these men and in a control group of 64 men match foe age, serum cholesterol and triglyceride concentration, blood pressure,

obesity, smoking and 0ne-hour glucose tolerance. Palmitic and stearic acids of phospholipids were significantly higher and linoleic and most polyunsaturated fatty acids, including arachidonic acid and eicosapentaenoic acid of phospholipids were lower in the subjects who sustained coronary events compared with the controls. Linoleic acid tended to correlate negatively with blood pressure while other polyunsaturated fatty acids, especially eicosapentaenoic acid, exhibited a negative correlation with blood pressure and relative body weight in the controls but not in the subjects who sustained coronary events. These findings suggest that the fatty-acid pattern of serum phospholipids is an independent risk factor coronary heart disease.

***Trans* fatty acids**

Several case-control or cross-sectional studies have also been conducted. In a case-control study of subjects in the Boston area, the researches found a strong and significant positive association between the intake of *trans* fatty acids, assessed with the use of dietary questionnaires, and risk of acute myocardial infraction. The relative risk of acute myocardial infarction for the quintile with the highest intake of *trans* fatty acids as compared with the quintile with the lowest intake was 2.4 (P for trend<0.001); this association was entirely explained by the intake of these fats from hydrogenated vegetable oil. Bolton-Smith et al. performed a cross-sectional analysis of the association between the intake of *trans* fatty acids and the presence of previously undiagnosed coronary heart disease among participants in the Scttish Heart Study. The intake of *trans* fatty acids was positively correlated with the ratio of LDL plus very-low-density lipoprotein cholesterol to HDL cholesterol. The odds ratio for coronary heart disease in the quintile with the highest intake as compared with the quintile with the lowest intake were elevated but not significantly so (1.26 in women and 1.08 in men)

In addition, there is the report about the association between dietary intake, including *trans* fatty acids and LDL size, in 414 radomly selected subjects living in Purisca, Costa Rica. They found that women had larger LDL sixe compares with men (263 v 261), and large LDL particle were coorlated with increased intake of *trans* fatty acids (p<0.001). The correlation between *trans* fatty acids intake and large LDL was significant in multivariate model that included dietary and nondietary factor;

a 1% difference in *trans* fatty acids was associated with 2.44 Å increase in LDL size (p=0.004) and they summarized that the effects of dietary factors such as intake of *trans* fatty acids on coronary heart disease are mediated throught their effects on LDL size (79).

Baylin et al, 2003 (80) concluded that total adipose tissue *trans*-fat was positively associated with risk of myocardial infarction. From this research, they examined the correlation of individual *trans* fatty acids from partially hydrogenated oil (18:1 and 18:2) or meat and dairy products (16:1 and 18:1) with myocardial infarction (MI). They found that 16:1 and 18:2 *trans* fatty acid in adipose tissue are also associated with increase risk of MI but an association with 18:1 *trans* fatty acids is not detected.

In 1996, Lucy P.L. (81) performed a study in coronay heart disease patients, and measured the *trans* fatty acids concentration of the plasma phospholipids fraction. Comparison was made between a case group with angiographically documentae severs coronary heart disease ans a control group of patients who had just minor stenosis on the coronary angiograpy. They found that, controls had higher plasma HDL levels and the difference in total *trans* faty acids content between cases and control (0.32±0.02% versus 0.35±0.02%)

Objective

General objective :

To examine the profile of serum total fatty acids concentration of n-3, n-6 and *trans* fatty acids among subjects who have risk factors of CHD: hypertension, hypercholesterolemia, dyslipidemia, diabetes mellitus and healthy subjects.

Specific objectives :

This study was aimed to:

(1). To determine the profile of serum total fatty acids concentration of n-3, n-6 and *trans* fatty acids among the risk group of coronary heart disease, subjects with coronary heart disease and healthy subjects.

(2). To compare the concentration of these polyunsaturated fatty acids among the risk group of coronary heart disease, subjects with coronary heart disease and healthy subjects.

(3). To compare the serum lipid concentration and lifestyle (eg, exercise, smoking status) among the risk group of coronary heart disease, subjects with coronary heart disease and healthy subjects.

(4). To compare the fatty acids concentration and dietary habit among the risk group of coronary heart disease, subjects with coronary heart disease and healthy subjects.

CHAPTER III
MATERIALS AND METHODS

3.1 Study populations

Cross sectional study was investigated in Electricity Generating Authority of Thailand (EGAT) officials who had already participated in the Cardiovascular Electricity Generating Authority of Thailand Study (CEGAT study) to assess the prevalence of CVD and risk factors of CVD in 2002. The sample size was calculated by 10% of total subjects of CEGAT study and 222 subjects with age range of 53- 73 years were participated in this study. The study was approved by Ramathibodi Hospital Research Ethics Committee and written informed consent was obtained from all participants.

Risk factors of CHD were defined as the followings: hypertension (HT; BP ≥140/90 mmHg), hypercholesterol (HC; TC ≥ 200 mg/dL), diabetes mellitus (DM; FBG ≥126 mg/dL), dyslipidemia (DLp; TG ≥ 150 mg/dL or TC ≥ 200 mg/dL or LDL-C ≥130 mg/dL or HDL-C ≤ 35 mg/dL). Healthy control subjects were those who were apparently healthy and did not have any risk factors of CHD.

3.2 Lifestyle assessment and dietary assessment

Questionnaire covering frequency of exercise, smoking habit, alcohol consumption, previous medical history and socioeconomic characteristics and dietary habit including food frequency were sent to the participants.

All participants were requested to reported their frequency of exercise as never performed, sometime (<1 time/wk), often (2-3 times/wk), usually (4-5 times/ wk) or always performed (>5 times/wk).

All participants were requested to specify their cigarette smoking behavior as never, quit or current drinkers. Never smokers were those who never smoked cigarette in their lives, quit were those who smoked previously but had

already ceased smoking for a certain period of time prior to the study. Current smokers were those who had smoked up to the time of the study.

All participants were requested to report their alcohol consumption behavior as never, quit or current drinkers. Never drinkers were those who never drank alcohol beverages in their lives, quit were those who drank alcoholic beverages previously and had ceased alcohol consumption for a certain period prior to the study. Current drinkers were those who drank alcoholic beverage regularly, regardless of social activities.

All participants were requested to specify their habits of taking breakfast, lunch, supper, and snacks as well as their frequency of high fat intake. Their frequencies of high fat intake were specified as sometime (<1 time/wk), often (2-3 times/wk), or usually (>3 times /wk).

3.3 Nutrition status assessment

Anthropometric measurement such as body weight, height, waist and hip circumferences in each subjects were measured by the standard techniques. Body mass index (BMI) was calculated from body weight in kg/ (height in m)^2. Waist-over hip circumference ratio (WHR) was computed.

3.4 Blood collection

Subjects were asked to fast for 12 hours before giving blood samples. Venous blood was obtained for determinations of fasting blood glucose (FBG), serum TC, HDL-C, and TG by enzymatic-colorimetric methods. Serum LDL-C was calculated by Friedewald's formula. Serum n-3, n-6 and *trans* fatty acids profile were analyzed by Gas chromatography.

3.4.1 Determination of serum n-3, n-6 and *trans* fatty acids profile by Gas chromatography

Lipids in serum were extracted by methanol:hexane (4:1) and transesterified by acetylchloride by modified method of Lepage G and Roy CC (82) as described in the following procedure:

1. Added 100 µl of C15:0 (1 mg/ml), an internal standard, to the screw cap test tube containing 200 µl serum sample

2. Added 2.0 ml of methanol : hexane (4:1) , screw on cap and stirred

3. Slowly added 200 µl acetyl chloride to crude extract while stirring

4. Tubes were tightly closed with Teflon-lined caps and subjected to methanolysis at 100°c for 1 hour

5. After tubes had been cooled, 5 ml of potassium carbonate solution was slowly added to stop the reaction and neutralize the mixture

6. The tubes were then shaken and centrifuged, and an aliquot of the hexane upper phase was injected into the chromatograph

The fatty acid methyl esters were separated and identified by the capillary gas liquid chromatography, Hewlette Packard model 2890 series II with a 50 m x 0.2 mm x 0.33 µm film thickness of SP 2330 fused-silica capillary column, using He as carrier gas at flow rate 1 ml/min and flame ionization detector

Identify of individual fatty acid methyl esters in the sample by comparison of their retention times with fatty acid methyl ester of known standards. The peak areas were measured by an electronic integration. The individual fatty acids were reported as percentages of total fatty acids.

3.5 Statistics

Statistical analysis was performed using SPSS for Windows version 11.0. Results were expressed as means ± SEM. Comparison were made among group of subjects who have various risk factors of coronary heart disease and healthy control subjects; and among the frequency of exercise, the frequency of high fat intake, smoking habit and alcohol intake by using analysis of variances at p-value <0.05 level of significance. Linear regression between various parameters was also computed.

CHAPTER IV
RESULTS

Two hundred and twenty two subjects were divided into 6 groups according to their risk factors of CHD and their previous medical history, ie. 38 healthy control, 11 subjects who had BP ≥140/90 mmHg only (HT group), 25 subjects who had TC ≥200 mg/dL only (HC group), 79 subjects who had BP ≥140/90 mmHg and TG ≥150 mg/dL or TC ≥200 mg/dL or LDL-C ≥130 mg/dL or HDL-C ≤35mg/dL(HT+DLp group), 32 subjects who had FBG ≥126 mg/dL together with BP ≥140/90 mmHg and TG ≥150 mg/dL or TC ≥200 mg/dL or LDL-C ≥130 mg/dL or HDL-C ≤35 mg/dL(DM+HT+DLp group) and 38 subjects who had CHD risk factors and suffered from CHD (CHD group) (Table 5).

Table 5 Number of healthy control and subjects with various diseases according to criteria

Groups	Number
Control group	38
HT	11
HC	25
HT+DLp	79
DM+HT+DLp	32
CHD	37
Total	222

Means ± SEM of fatty acids composition in total serum fatty acids in healthy controls and subjects with various diseases were shown in Table 6. The myristic acid (14:0) in subjects with DM and HT and DLp was significantly lower than healthy controls and subjects with CHD but the palmitic acid (16:0) was significantly higher than healthy controls and subjects with various diseases. The stearic acid (18:0) in subjects with DM and HT and DLp was significantly higher than healthy controls and subjects with HC, subjects with HT and DLp and subjects with CHD. The stearic acid (18:0) in subjects with HT and DLp was also higher than healthy controls. The oleic acid (18:1, n-9) in subjects with DM and HT and DLp was significantly higher than healthy controls and subjects with HT, subjects with HC and subjects with HT and DLp but only the *trans* fatty acid; elaidic acid (18:1, *t*) in subjects with DM and HT and DLp was lower than healthy controls and subjects with HT and DLp. The 5, 8, 11-eicosatrienoic acid (20:3, n-9) in subjects with DM and HT and DLp was significantly lower than subjects with HT and DLp and subjects with CHD. The palmitoleic acid (16:1, n-7) content in subjects with DM and HT and DLp was significantly lower than subjects with HC. The linoleic acid (18:2, n-6) in subjects with HC was significantly higher than healthy controls and subjects with HT and subjects with HT and DLp. The 11, 14-eicosadienoic acid (20:2, n-6) in subjects with HT was significantly lower than subjects with CHD. The dihomo gramma linolenic acid (20:3, n-6) in subjects with HT was significantly higher than healthy controls but the 20:3, n-6 in DM and HT and DLp was significantly lower than subjects with HT. The arachidonic acid (20:4, n-6) in subjects with DM and HT and DLp was significantly lower than healthy controls and subjects with HT, subjects with HC, subjects with HT and DLp and subjects with CHD. The linolenic acid (18:3, n-3) in subjects with HT was significantly higher than healthy controls and subjects with HC, subjects with HT and DLp and subjects with CHD. EPA content in subjects with DM and HT and DLp was significantly lower than subjects with HC, subjects with HT and DLp and subjects with CHD and EPA (20:5, n-3) in subjects with HC and subjects with HT and DLp was significantly lower than healthy controls. The clupanodonic acid (22:5, n-3) in subjects with DM and HT and DLp and subjects with CHD was significantly lower than healthy controls but the 22:5, n-3 in subjects with HT and DLp was significantly higher than subjects with HC and subjects with DM and HT and

DLp. The DHA (22:6, n-3) in subjects with DM and HT and DLp was significantly lower than healthy controls and subjects with HT, subjects with HC and subjects with HT and DLp. The DHA (22:6, n-3) in subjects with CHD was significantly lower than healthy controls and DHA (22:6, n-3) in subjects with HT and DLp was significantly higher than subjects with HC.

Means ± SEM of triglyceride, cholesterol, HDL-C, LDL-C, fasting blood sugar, weight, height, BMI, waist, hip and WHR of healthy controls and subjects with various diseases were shown in Table 7. Triglyceride of subjects with DM and HT and DLp was significantly higher than healthy controls and subjects with various diseases except subjects with HT and triglyceride in subjects with HT and DLp was significantly higher than healthy controls. Cholesterol in healthy controls and subjects with HT were significantly lower than subjects with HC, subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD and cholesterol in subjects with HC was significantly higher than subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD. HDL-C in subjects with HC was significantly higher than healthy controls and subjects with various diseases. LDL-C in healthy controls and subjects with HT was significantly lower than subjects with HC, subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD and LDL-C in subjects with HC was significantly higher than subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD. Fasting blood sugar in DM and HT and DLp was significantly higher than healthy controls and subjects with various disease and fasting blood sugar in subjects with CHD was significantly higher than healthy controls and subjects with HC, subjects with DM and HT and DLp. Weight in healthy controls and subjects with HC were significantly lower than subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD. BMI in healthy controls and subjects with HC were significantly lower than subjects with HT, subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD. Waist girth in healthy controls and subjects with HC were significantly lower than subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD and waist girth in subjects with HT was significantly higher than healthy controls. Hip circumference in healthy controls was significantly lower than subjects with various diseases except subjects with HC. WHR in healthy

controls and subjects with HC were significantly lower than subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD.

Table 8 showed no significant different from frequency of exercise pattern of healthy controls and subjects with various diseases by chi-square test. Table 9-14 showed Means ± SEM triglyceride, serum total cholesterol, LDL-C, HDL-C, BMI and WHR in healthy controls and subjects with various diseases according to frequency of exercise, respectively. Table 9, triglyceride level in subjects with CHD who frequent had exercise was significantly higher than control groups. Serum triglyceride in subjects with DM and HT and DLp who usual had exercise was significantly higher than healthy controls and subjects with various diseases. Table 10, subjects with HC, subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD who frequent, usual and always had exercise had significantly higher than healthy controls and subjects with HT. LDL-C in subjects with HC, subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD who frequent, usual and always had exercise were significantly higher than healthy controls and subjects with HT (Table 11). Table 12 showed that serum HDL-C level in subjects with HT and DLp and subjects with CHD who occasional had exercise was significantly lower than healthy controls and subjects with HC. Serum HDL-C level in subjects with HC who frequent had exercise was significantly higher than subjects with HT and DLp and HDL-C level in subjects with HC who usual had exercise was significantly higher than subjects with DM and HT and DLp and subjects with HT and DLp. Subjects with CHD who usual and always had exercise had serum HDL-C level significantly higher than subjects with CHD who occasional and frequent had exercise. Table 13, BMI in subjects with HT and DLp who occasional had exercise and BMI in subjects with DM and HT and DLp who frequent had exercise was significantly higher than healthy controls and subjects with HC. BMI in subjects with HC who frequent had exercise was significantly lower than subjects with HT. Healthy controls and subjects with HC who usual had exercise had BMI significantly lower than subjects with HT and DLp and subjects with CHD. Table 14 showed that WHR in subjects with HC who frequent had exercise was significantly lower than healthy controls and subjects with various diseases, healthy controls and subjects with HC who usual had exercise had WHR significantly lower than subjects with various diseases except

subjects with HT and subjects with HC who usual had exercise had WHR significantly lower than subjects with HC who always had exercise.

No significant different from smoking status in healthy controls and subjects with various diseases by chi-square test (Table 15). Means ± SEM of triglyceride, serum total cholesterol, serum HDL-C level, serum LDL-C level, BMI and WHR in healthy controls and subjects with various diseases according to smoking status showed in Table 16, 17, 18, 19, 20 and 21, respectively. Table 16, triglyceride level in subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD who newer smoking were significant different from healthy control and triglyceride level in subjects with DM and HT and DLp who newer smoking were also significant different from subjects with HT. Table 17, serum total cholesterol level in healthy control and subjects with HT were significantly different from subjects with various diseases. Serum LDL-C level in healthy controls and subjects with HT who never smoking and quit smoking were significantly lower than subjects with various diseases and subjects with CHD who never smoking had serum LDL-C level significantly lower than subjects with HC and subjects with HT and DLp (Table 18). Serum HDL-C level in subjects with HC who never smoking was significantly higher than healthy controls, subjects with HT and DLp and subjects with CHD, serum HDL-C level in subjects with HC who quit smoking was significantly higher than subjects with HT, subjects with HT and DLp and subjects with CHD and subjects with CHD who quit smoking had serum HDL-C level significantly lower than subjects with HT and DLp (Table 19). BMI in healthy controls and subjects with HC who never had smoking intake was significantly lower than subjects with various except subjects with HT, BMI in healthy controls who quit smoking was significantly lower than subjects with various diseases except subjects with HC and healthy controls and subjects with CHD who never smoking had BMI significantly higher than healthy controls and subjects with CHD who current smoking, respectively (Table 20). Table 21 showed that WHR in subjects with HC who never smoking and healthy controls who quit smoking was significantly lower than subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD and WHR in subjects with HC who quit smoking was significantly lower than subjects with DM and HT and DLp.

No significant different from frequency of high fat intake pattern of healthy controls and subjects with various diseases by chi-square test. Table 23, 24, 25, 26, 27 and 28 showed means ± SEM of triglyceride, serum total cholesterol, serum LDL-C, serum HDL-C level, BMI and WHR in healthy controls and subjects with various diseases according to frequency of high fat intake. Table 23, triglyceride level in healthy control who frequent had high fat intake was significantly different from subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD. Serum cholesterol level in healthy controls who occasional and frequent had high fat intake was significantly lower than subjects with various diseases except subjects with HT, subjects with HT who frequent had high fat intake had serum cholesterol level significantly lower than subjects with various disease and subjects with HC who frequent had high fat intake had serum cholesterol level significantly higher than subjects with DM and HT and CHD, subjects with HT and DLp and subjects with CHD (Table 24). Table 25, serum LDL-C level in subjects with various diseases who frequent had high fat intake were significantly different from healthy controls and subjects with HT. No significantly different from serum HDL-C in healthy controls and subjects with various diseases according to frequency of high fat intake (Table 26). BMI in healthy controls and subjects with HC who frequent had high fat intake was significantly lower than subjects with various except subjects with HT (Table 27). WHR in subjects with DM and HT and DLp who occasional had high fat intake was significantly higher than healthy controls and subjects with HC. The healthy controls and subjects with HC who frequent high fat intake had WHR significantly lower than subjects with various diseases except subjects with HT (Table 28). Serum fatty acid concentrations in healthy controls and subjects with various diseases according to frequency of high fat intake were showed in Tables 29-37. Table 29 showed the palmitic acid (16:0) level, subjects with DM and HT and DLp who frequent had high fat intake was significantly higher than healthy controls and subjects with various diseases. The palmitoleic acid (16:1, n-7) concentration in subjects with HC who frequent had high fat intake was significantly higher than subjects with DM and HT and DLp and subjects with CHD who frequent had high fat intake was significantly high than subjects with HT, subjects with DM and HT and DLp and subjects with HT and DLp (Table 30). The serum elaidic acid (18:1, *t*) concentration in

subjects with HC who frequent had high fat intake was significantly higher than subjects with various diseases (Table 31). The LA (18:2, n-6) level in subjects with HT and DLp who frequent had high fat intake was significantly lower than subjects with CHD and the AA (20:4, n-6) level in subjects with DM and HT and DLp who frequent had high fat intake was significantly lower than healthy controls and subjects with various diseases (Table 32-33). Table 33, ALA (18:3, n-3) concentration in subjects with HT who frequent had high fat intake was significantly higher than healthy controls and subjects with various diseases. Table 34, EPA (22:5, n-3) content in subjects with DM and HT and DLp who occasional had high fat intake was significantly lower than healthy controls and EPA in subjects with CHD who frequent had high fat intake was significantly higher than subjects with DM and HT and DLp and subjects with HT and DLp. Clupadonic acid (22:5, n-3) in healthy controls who occasional had high fat intake was significantly lower than and subjects with DM and HT and CHD, subjects with HT and DLp and subjects with CHD (Table 36). DHA (22:6, n-3) content in healthy controls who occasional and frequent had high fat intake were significantly higher than subjects with DM and HT and DLp and subjects with HC, respectively. DHA in healthy controls and subjects with HT and DLp who frequent had high fat intake were significantly high than subjects with DM and HT and DLp and subjects with CHD (Table 37).

Table 38, no significant different from alcohol intake of healthy controls and subjects with various diseases by chi-square test. Means ± SEM of triglyceride, serum total cholesterol, serum LDL-C level, serum HDL-C level serum, , fasting blood sugar level, BMI, WHR and various serum fatty acid concentrations in healthy controls and subjects with various diseases according to alcohol intake showed in Table 39-54. Table 39, serum triglyceride level in healthy controls was significantly lower than subjects with DM and HT and DLp, subjects with HT and Dlp and subjects with CHD. Serum triglyceride level in subjects with DM and HT and CHD who current had alcohol intake was significantly higher than healthy controls, subjects with HC and subjects with HT and DLp. Serum total cholesterol in subjects with various disease who quit and current had alcohol intake were significantly different from healthy controls and subjects with HT (Table 40). Healthy controls and and subjects with HT who never, quit and current had alcohol intake had serum LDL-C level

significantly lower than subjects with various diseases (Table 41). Table 42 showed that serum HDL-C level in subjects with HC who never had alcohol intake was significantly higher than healthy controls and subjects with various diseases except subjects with HT but serum HDL-C level in subjects with HC who quit had alcohol intake was significantly higher than subjects with HT. Serum fasting blood sugar in subjects with DM and HT and DLp who never and current had alcohol intake was significantly higher than healthy controls and subjects with various diseases. Healthy controls and subjects with HT and DLp who quit had alcohol intake had serum fasting blood sugar significantly lower than subjects with DM and HT and DLp and subjects with CHD (Table 43). BMI in healthy controls who never and quit had alcohol intake was significantly lower than subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD but BMI in healthy controls who current had alcohol intake was significantly lower than subjects with various diseases. Subjects with HC who never had alcohol intake had BMI significantly lower than subjects with HT, subjects with DM and HT and DLp, subjects with HT and DL p and subjects with CHD. BMI in healthy controls who never drink was significantly higher than healthy controls who quit and current had alcohol intake and BMI in subjects with HC who never had alcohol intake was significantly lower than subjects with HC who current had alcohol intake (Table 44). WHR in healthy controls who never had alcohol intake was significantly lower than subjects with DM and HT and DLp and subjects with CHD, WHR in healthy controls who quit had alcohol intake was significantly lower than subjects with various diseases except subjects with HC and WHR in healthy controls who current had alcohol intake was significantly lower than subjects with various diseases except subjects with HT. WHR in subjects with HC who never had alcohol intake was significantly lower than subjects with various diseases except healthy controls and subjects with HT. Subjects with HC who never had alcohol intake had WHR significantly lower than subjects with HC who current had alcohol intake (Table 45). The palmitic acid (16:0) in subjects with DM and HT and DLp who never had alcohol intake were higher than healthy controls and subjects with HT and DLp, palmitic acid in subjects with DM and HT and DLp who current had alcohol intake was significantly higher than healthy controls, subjects with various diseases (Table 46). The palmitic acid in subjects with DM and HT and DLp who current had alcohol

intake was significantly higher than healthy controls and subjects with various diseases. The palmitoleic acid (16:1, n-7) level in healthy controls, subjects with HT and DLp who never had alcohol intake and subjects with DM and HT and DLp who current had alcohol intake were significantly lower than subjects with CHD (Table 47). The elaidic acid (18:1, *t*) in subjects with DM and HT and DLp who never had alcohol intake was significantly lower than healthy controls and subjects with HC and elaidic acid in subjects with HC who current had alcohol intake was significantly higher than healthy controls, subjects with HT and DLp and subjects with CHD (Table 48). Table 49 showed the LA (18:2, n-6) concentration in healthy controls and subjects with various diseases. LA in subjects with HC who never had alcohol intake was significantly higher than healthy controls, subjects with DM and HT and DLp and subjects with HT and DLp and LA in subjects with HC who current had alcohol intake was significantly higher than subjects with HT and subjects with HT and DLp. Table 50, AA level in subjects with DM and HT and DLp who current had alcohol intake was significantly lower than healthy controls and subjects with various diseases except subjects with CHD. The ALA (18:3, n3) in subjects with HT who never and current had alcohol intake were significantly higher than healthy controls and subjects with various diseases (Table 51). Table 52, EPA in healthy controls who never and quit had alcohol intake were significantly higher than subjects with DM and HT and DLp. Table 53, clupadonuic acid (22:5, n-3) in subjects with CHD who never had alcohol intake was significantly lower than healthy controls and subjects with HT and DLp, clupadonic acid in subjects with HC and subjects with CHD who current had alcohol intake were significantly lower than healthy controls, subjects with HT and subjects with HT and DLp. Table 54, the DHA (22:6,n-3) level in healthy controls, subjects with HT and subjects with HT and DLp who current had alcohol intake were significantly higher than subjects with HC and subjects with DM and HT and DLp.

Regression equations of total subjects showed in Table 55 (N=222). Alcohol intake and smoking status were as the independent variables and WHR, HDL-C and BMI were as the dependent variables.

Table 6 Means ± SEM of fatty acids composition in total serum fatty acids in healthy controls and subjects with various diseases

Fatty acids	Control (N=38)	HT (N=11)	HC (N=25)	DM+HT+DLp (N=32)	HT+DLp (N=79)	CHD (N=37)
	← percent of total fatty acids →					
14:0	1.46±0.11	1.26±0.17	1.40±0.25	0.93±0.21 $^{a1, f1}$	1.27±0.11	1.66±0.21
16:0	26.44±1.13	23.34±3.21	26.63±1.92	31.93±1.57 $^{a2, b1, c1, e2, f1}$	27.21±0.93	27.11±1.68
18:0	15.73±0.65	18.72±3.02	16.39±1.56	20.93±0.94 $^{a3, c1, e1, f2}$	18.03±0.67 ^{a1}	16.36±1.30
20:0	0.54±0.61	0.47±0.18	1.05±0.50	0.43±0.09	0.61±0.09	0.80±0.15
18:1, n-9	12.57±0.75	11.81±0.94	12.38±0.97	16.44±0.83 $^{a3, b3, c1, e3}$	13.01±0.50	13.94±0.17
18:1, *t*	0.37±0.53	0.31±0.09	0.61±0.20	0.23±0.05 $^{a2, e1}$	0.32±0.25	0.29±0.04
20:1, n-9	0.53±0.11	0.49±0.21	0.33±0.08	0.34±0.13	0.45±0.07	0.55±0.08
20:3, n-9	1.34±0.22	1.12±0.32	1.42±0.32	0.84±0.18 $^{e1, f1}$	1.50±0.17	1.51±0.26
16:1, n-7	1.25±0.13	1.16±0.33	1.59±0.22	0.90±0.16 ^{c1}	1.18±0.12	2.60±0.82
18:2, n-6	9.81±1.07	9.27±2.09	14.62±2.02 $^{a1, b1, e2}$	9.59±0.85	8.71±0.58	11.25±1.20
20:2, n-6	1.15±0.28	0.49±0.17 ^{f1}	1.13±0.23	1.23±0.33	1.10±0.17	1.42±0.36
20:3, n-6	0.71±0.14	1.70±0.41 ^{a2}	0.99±0.21	0.81±0.18 ^{b1}	0.85±0.16	0.92±0.31
20:4, n-6	6.82±0.65	8.55±1.48	7.64±0.87	4.77±0.58 $^{a1, b1, c1, e3, f2}$	7.47±0.50	7.63±0.76
18:3, n-3	3.65±0.60	9.41±2.00 $^{a1, c1, e1, f1}$	3.87±0.56	2.50±0.33 $^{b2, c1, e1, f1}$	3.96±0.32	4.08±0.64
20:5, n-3	1.80±0.41	0.86±0.29	0.57±0.76 ^{a2}	0.31±0.05 $^{a3, c2, e3, f1}$	0.88±0.14 ^{a1}	1.31±0.47
22:5, n-3	3.71±0.37	2.50±0.52	2.25±0.44	2.37±0.35 ^{a1}	3.38±0.26 $^{c1, d1}$	1.53±0.27 ^{a3}
22:6, n-6	8.19±0.79	6.65±1.17	5.69±0.85	3.54±0.53 $^{a3, b2, c1, e3}$	7.74±0.50 ^{c1}	5.04±0.77 ^{a2}

Significant different from Control group: $^{a1}p<0.05$, $^{a2}p<0.01$, $^{a3}p<0.005$

Significant different from HT: $^{b1}p<0.05$, $^{b2}p<0.01$, $^{b3}p<0.005$

Significant different from HC: $^{c1}p<0.05$, $^{c2}p<0.01$

Significant different from DM+HT+DLp: $^{d1}p<0.05$

Significant different from HT+DLp: $^{e1}p<0.05$, $^{e2}p<0.01$, $^{e3}p<0.005$

Significant different from CHD: $^{f1}p<0.05$, $^{f2}p<0.01$, $^{f3}p<0.005$

Table 7 Means ± SEM of some parameters of healthy controls and subjects with various diseases

Parameters	Cases					
	Control (N=38)	HT (N=11)	HC (N=25)	DM+HT+DLp (N=32)	HT+DLp (N=79)	CHD (N=37)
Triglyceride	91.8±4.49	135.1±22.83	119.1±6.37	211.8±61.14 $^{a2, c1, e1, f1}$	149.5±7.70 ^{a1}	142.9±9.34
Cholesterol	177.9±3.20 $^{c2, d2, e2, f3}$	181.6±4.55 $^{c2, d2, e3, f3}$	275.0±5.85 $^{d1, e2, f3}$	250.7±6.35	254.3±3.86	242.1±6.98
HDL-C	54.1±1.59	51.4±4.58	62.2±1.82 $^{a1, b1, d2, e2, f3}$	51.7±2.21	52.5±1.39	50.6±2.47
LDL-C	105.4±3.26 $^{c2, d2, e3, f3}$	103.2±6.31 $^{c2, d2, e3, f3}$	188.9±6.39 $^{d1, e1, f2}$	165.7±5.48	171.8±3.99	162.8±6.42
FBS	95.9±1.38	98.4±1.76	93.6±1.83	147.7±11.34 $^{a2, b3, c2, e3, f3}$	98.9±1.25	109.8±5.00 $^{a1, c1, d2}$
Weight	58.2±1.64 $^{d1, e3, f3}$	64.1±3.18	59.3±2.13 $^{d1, e2, f2}$	66.9±1.71	66.3±1.15	67.2±1.97
Height	163.4±1.12	159.2±2.55	162.8±1.30	161.5±1.28	163.3±0.70	162.5±0.98
BMI	21.8±0.54 $^{b2, d2, e3, f3}$	25.3±1.12	22.3±0.68 $^{b1, d2, e3 f3}$	25.6±0.51	24.8±0.34	25.3±0.62
Waist	80.6±1.56 $^{b1, d2, e3 f3}$	88.8±2.40	83.1±2.22 $^{d2, e2, f2}$	91.9±1.33	89.8±0.97	91.2±1.71
Hip	91.7±1.09 $^{b1, d2, e3, f3}$	96.8±2.28	94.6±1.13 f1	97.0±1.12	96.5±0.66	97.9±1.09
WHR	0.88±0.01 $^{d2, e3, f2}$	0.92±0.02	0.88±0.02 $^{d2, e3, f2}$	0.95±0.01	0.93±0.01	0.93±0.01

Significant different from Control group: $^{a1}p<0.05$, $^{a2}p<0.005$

Significant different from HT: $^{b1}p<0.05$, $^{b2}p<0.01$, $^{b3}p<0.005$

Significant different from HC: $^{c1}p<0.05$, $^{c2}p<0.005$

Significant different from DM+HT+DLp: $^{d1}p<0.01$, $^{d2}p<0.005$

Significant different from HT+DLp: $^{e1}p<0.05$, $^{e2}p<0.01$, $^{e3}p<0.005$

Significant different from CHD: $^{f1}p<0.05$, $^{f2}p<0.01$, $^{f3}p<0.005$

Table 8 Percentage of number of healthy controls and subjects with various diseases according to frequency of exercise

Cases	Frequency of exercise (N)			
	Occasional	Frequent	Usual	Always
Control	30 (9)	20 (6)	36.67 (11)	13.33 (4)
HT	20 (2)	40 (4)	20 (2)	20 (2)
HC	15 (3)	20 (4)	40 (8)	25 (5)
HT+DLp	13.04 (9)	27.54 (19)	36.33 (25)	23.19 (16)
DM+HT+DLp	18.52 (5)	25.93 (7)	33.33 (9)	22.22 (6)
CHD	13.33 (4)	30 (9)	40 (12)	16.67 (5)

No significant different by chi-square test

Table 9 Means ± SEM of triglyceride in healthy controls and subjects with various diseases according to frequency of exercise

Cases	Frequency of exercise			
	Occasional	Frequent	Usual	Always
	← mg/dL →			
Control	87.0±9.58	99.8±14.70	101.6±6.79	74.5±9.53
HT	92.1±4.99	145.0±13.0	132.5±11.5	129.5±11.5
HC	109.7±13.38	118.5±8.17	131.8±11.27	129.2±13.09
HT+DLp	154.7±34.17	195.4±15.51	148.1±11.57	144.3±16.32 [a1]
DM+HT+DLp	143.4±26.78	116.6±9.08	364.2±214.78 [a1, b1, c1, d1]	180.5±31.91 [a1]
CHD	145.2±30.18	162.3±22.86 [a1]	136.3±16.40	153.4±29.72

Significant different from Control group: $^{a1}p<0.05$

Significant different from HC: $^{b1}p<0.05$

Significant different from HT+DLp: $^{c1}p<0.05$

Significant different from CHD: $^{d1}p<0.05$

Table 10 Means ± SEM of serum total cholesterol in healthy controls and subjects with various diseases according to frequency of exercise

Cases	Frequency of exercise			
	Occasional	Frequent	Usual	Always
	← mg/dL →			
Control	179.3±5.47	184.2±6.42	181.0±7.02	176.0±8.43
HT	175.5±17.5	186.2±5.23	180.5±17.5	175.5±14.5
HC	254.3±5.49 ^{a1}	276.0±7.73 $^{a2, b2}$	271.0±9.94 $^{a2, b2}$	288.8±18.14 $^{a2, b2}$
HT+DLp	251.9±14.59 $^{a2, b1}$	259.4±7.56 $^{a2, b2}$	251.5±7.00 $^{a2, b2}$	259.7±7.31 $^{a2, b2}$
DM+HT+DLp	258.0±6.42 $^{a2, b1}$	238.0±9.50 $^{a2, b2}$	247.7±12.16 $^{a2, b2}$	262.7±22.63 $^{a2, b2}$
CHD	224.2±53.16	247.7±7.91 $^{a2, b2}$	248.8±10.34 $^{a2, b2}$	242.2±14.25 $^{a2, b2}$

Significant different from Control group: $^{a1}p<0.05$, $^{a2}p<0.005$
Significant different from HT: $^{b1}p<0.05$, $^{b2}p<0.005$

Table 11 Means ± SEM of LDL-C in healthy controls and subjects with various diseases according to frequency of exercise

Cases	Frequency of exercise			
	Occasional	Frequent	Usual	Always
	← mg/dL →			
Control	101.4±6.84	115.7±6.54	107.7±6.84	105.8±6.31
HT	99.0±26.60	104.7±10.33	106.6±19.8	90.1±1.90
HC	167.4±9.83 ^{a1}	190.6±10.27 $^{a2, b2}$	180.8±12.21 $^{a2, b2}$	202.9±17.99 $^{a2, b2}$
HT+DLp	170.8±13.96 $^{a1, b1}$	175.5±9.12 $^{a2, b2}$	170.1±7.01 $^{a2, b1}$	171.3±8.44 $^{a2, b2}$
DM+HT+DLp	177.5±6.33 $^{a3, b1}$	158.8±8.87 $^{a2, b2}$	157.8±10.59 ^{a2}	170.9±19.46 $^{a2, b2}$
CHD	156.9±48.10 ^{a1}	168.4±8.5 $^{a2, b2}$	164.6±9.86 $^{a2, b1}$	158.5±12.8 $^{a2, b1}$

Significant different from Control group: $^{a1}p<0.05$, $^{a2}p<0.005$
Significant different from HT: $^{b1}p<0.05$, $^{b2}p<0.005$

Table 12 Means ± SEM of serum HDL-C in healthy controls and subjects with various diseases according to frequency of exercise

Cases	Frequency of exercise			
	Occasional	Frequent	Usual	Always
	← mg/dL →			
Control	60.5±3.34	48.5±4.59	53.0±2.02	55.2±7.48
HT	47.5±5.50	51.0±4.41	48.0±10.00	66.0±24.00
HC	65.0±3.05	61.8±6.10 ^{d1}	63.9±3.72 $^{c1, d1}$	60.0±3.63
HT+DLp	49.5±3.28 $^{a1, b1,}$	49.9±2.76	51.8±1.89	59.5±3.70
DM+HT+DLp	51.8±5.64	55.8±2.30	49.4±5.93	55.6±5.30
CHD	38.2±3.09 $^{a2, b2}$	46.8±4.86	57.0±5.31 $^{A1, B1}$	53.0±6.51 $^{A1, B1}$

Significant different from Control group: $^{a1}p<0.05$, $^{a2}p<0.01$
Significant different from HC: $^{b1}p<0.05$, $^{b2}p<0.01$
Significant different from DM+HT+DLp: $^{c1}p<0.05$
Significant different from HT+DLp: $^{d1}p<0.05$
Significant different from occasional: $^{A1}p<0.05$
Significant different from frequent: $^{B1}p<0.05$

Table 13 Means ± SEM of BMI in healthy controls and subjects with various diseases according to frequency of exercise

Cases	Frequency of exercise			
	Occasional	Frequent	Usual	Always
Control	21.5±1.17	22.2±1.08	22.9±0.86 $^{d1, e1}$	23.1±1.19
HT	22.8±0.39	25.9±2.58	26.2±2.62	24.1±3.17
HC	19.9±1.66	21.3±1.53 ^{b1}	22.9±1.33 $^{d1, e1}$	23.2±1.83
HT+DLp	25.1±1.14 $^{a1, c1}$	24.9±0.76	25.4±0.52	23.5±0.80
DM+HT+DLp	24.1±1.96	26.3±0.88 $^{a1, c1}$	25.4±1.04	25.3±0.88
CHD	22.9±1.14	24.5±0.95	26.1±1.08	26.3±2.03

Significant different from Control group: $^{a1}p<0.05$
Significant different from HT: $^{b1}p<0.05$
Significant different from HC: $^{c1}p<0.05$
Significant different from HT+DLp: $^{d1}p<0.05$
Significant different from CHD: $^{e1}p<0.05$

Table 14 Means ± SEM of WHR in healthy controls and subjects with various diseases according to frequency of exercise

Cases	Frequency of exercise			
	Occasional	Frequent	Usual	Always
Control	0.87±0.02	0.91±0.01	0.88±0.01 $^{c1, d1, e1}$	0.88±0.04
HT	0.92±0.01	0.94±0.02	0.89±0.08	0.94±0.02
HC	0.87±0.08	0.84±0.01 $^{a1, b1, c1, d1, e1}$	0.84±0.02 $^{c2, d2, e2, A1}$	0.94±0.01
HT+DLp	0.92±0.02	0.93±0.01	0.95±0.01	0.92±0.01
DM+HT+DLp	0.94±0.02	0.95±0.02	0.96±0.01	0.94±0.02
CHD	0.88±0.04	0.93±0.02	0.95±0.01	0.95±0.02

Significant different from Control group: $^{a1}p<0.05$

Significant different from HT: $^{b1}p<0.01$

Significant different from DM+HT+DLp: $^{c1}p<0.01$, $^{c2}p<0.005$

Significant different from HT+DLp: $^{d1}p<0.01$, $^{d2}p<0.005$

Significant different from CHD: $^{e1}p<0.01$, $^{e2}p<0.005$

Significant different from always: $^{A1}p<0.05$

Table 15 Percentage of number of healthy controls and subjects with various diseases according to smoking status

Cases	Smoking status (N)		
	Never	Quit	Current
Control	37.84 (14)	21.62 (8)	40.54 (15)
HT	54.55 (6)	36.36 (4)	9.09 (1)
HC	64 (16)	20 (5)	16 (4)
HT+DLp	46.67 (35)	29.33 (22)	24 (18)
DM+HT+DLp	41.94 (13)	41.94 (13)	16.13 (5)
CHD	42.22 (17)	27.78 (10)	25 (9)

No significant different by chi-square test

Table 16 Means ± SEM of triglyceride in healthy controls and subjects with various diseases according to smoking status

Cases	Smoking status		
	Never	Quit	Current
	← mg/dL →		
Control	93.2±6.72	95.1±9.72	90.5±8.17
HT	91.3±13.80	158.8±28.43	103.0±0.00
HC	114.6±8.82	131.4±7.88	121.5±16.6
HT+DLp	140.3±9.75 ^{a2}	138.5±8.24	169.6±20.87
DM+HT+DLp	149.0±16.52 $^{a2, b1}$	311.1±48.43	126.6±22.46
CHD	135.2±12.01 ^{a1}	165.8±18.42	133.9±23.34

Significant different from Control group: $^{a1}p<0.05$, $^{a2}p<0.01$, $^{a3}p<0.005$

Significant different from HT: $^{b1}p<0.01$, $^{b2}p<0.025$

Table 17 Means ± SEM of serum total cholesterol in healthy controls and subjects with various diseases according to smoking status

Cases	Smoking status		
	Never	Quit	Current
	← mg/dL →		
Control	183.1±3.71	173.3±6.79	177.0±6.28
HT	187.8±3.49	170.0±9.40	191.0±0.00
HC	273.7±8.47 [a1, b1]	282.8±8.80 [a1, b1]	270.8±10.54
HT+DLp	238.1±6.81 [a1, b1]	258.5±13.06 [a1, b1]	229.1±21.11
DM+HT+DLp	251.7±12.05 [a1, b1]	241.3±8.10 [a1, b1]	267.6±14.62
CHD	256.2±5.28 [a1, b1]	256.7±8.05 [a1, b1]	247.9±8.73

Significant different from Control group: [a1]p<0.005
Significant different from HT: [b1]p<0.005

Table 18 Means ± SEM of serum LDL-C in healthy controls and subjects with various diseases according to smoking status

Cases	Smoking status		
	Never	Quit	Current
	← mg/dL →		
Control	111.6±4.37	101.7±7.02,	102.6±6.11
HT	111.4±7.95	94.5±11.53	89.4±0.00
HC	187.1±9.28 [a1, b2]	195.7±9.04 [a1, b2,]	188.2±12.56
HT+DLp	174.8±5.11 [a2, b2]	174.7±7.70 [a1, b2, A1]	165.7±10.30
DM+HT+DLp	166.3±9.27 [a2, b2]	154.0±7.66 [a1, b2]	189.2±10.26
CHD	156.2±6.16 [a1, b2, c2, d2]	181.4±12.9 [a1, b2]	151.1±17.81

Significant different from Control grooup: [a1]p<0.01, [a2]p<0.005
Significant different from HT: [b1]p<0.01, [b2]p<0.005
Significant different from HC: [c1]p<0.05, [c2]p<0.005
Significant different from HT+DLp: [d1]p<0.01, [d2]p<0.005
Significant different from current: [A1]p<0.05

Table 19 Means ± SEM of serum HDL-C in healthy controls and subjects with various diseases according to smoking status

Cases	Smoking status		
	Never	Quit	Current
	← mg/dL →		
Control	52.8±2.48	52.9±2.72	56.3±2.98
HT	58.2±7.22	43.8±3.22	41.0±0.00
HC	63.7±2.42 $^{a1, d1, e1}$	60.8±3.95 $^{b1, d1, e1}$	58.2±3.68
HT+DLp	53.3±2.29	54.2±2.36	48.2±2.52
DM+HT+DLp	55.6±3.26	47.2±3.73	53.0±5.30
CHD	54.8±3.24	43.9±2.34 ^{d1}	51.2±7.39

Significant different from Control group: $^{a1}p<0.05$
Significant different from HT: $^{b1}p<0.05$
Significant different from DM+HT+DLp: $^{c1}p<0.05$
Significant different from HT+DLp: $^{d1}p<0.05$
Significant different from CHD: $^{e1}p<0.05$

Table 20 Means ± SEM of BMI in healthy controls and subjects with various diseases according to smoking status

Cases	Smoking status		
	Never	Quit	Current
Control	22.6±0.81 ^{A1}	22.6±1.21	20.0±0.67
HT	25.1±1.84	26.6±1.19 ^{a1}	21.6±0.00
HC	22.3±0.83	23.9±1.14	20.4±2.18
HT+DLp	25.2±0.50 $^{a1, b2}$	25.2±0.59 ^{a1}	23.3±0.81
DM+HT+DLp	25.8±0.75 $^{a1, b2}$	25.5±0.75 ^{a1}	24.7±1.88
CHD	25.3±1.05 $^{a1, A1}$	26.7±0.91 ^{a2}	23.9±1.21

Significant different from Control group: $^{a1}p<0.05$
Significant different from HT: $^{b1}p<0.05$, $^{b2}p<0.01$
Significant different from current: $^{A1}p<0.05$

Table 21 Means ± SEM of WHR in healthy controls and subjects with various diseases according to smoking status

Cases	Smoking status		
	Never	Quit	Current
Control	0.88±0.01	0.89±0.02	0.85±0.01
HT	0.89±0.27	0.94±0.01	0.95±0.00
HC	0.86±0.02	0.90±0.01	0.90±0.03
HT+DLp	0.92±0.01 ^{b1}	0.94±0.01 $^{a2, b1}$	0.94±0.01
DM+HT+DLp	0.93±0.02 ^{b2}	0.95±0.01 ^{a1}	0.96±0.01
CHD	0.91±0.02 ^{b1}	0.95±0.01 ^{a1}	0.92±0.02

Significant different from Control group: $^{a1}p<0.05$, $^{a2}p<0.01$

Significant different from HC: $^{b1}p<0.05$, $^{b2}p<0.01$

Table 22 Percentage of number of healthy controls and subjects with various diseases according to frequency of high fat intake

Cases	Frequency of high fat intake (N)		
	Occasional	Frequent	Usual
Control	23.33 (7)	56.67 (17)	20 (6)
HT	-	80 (8)	20 (2)
HC	20 (4)	75 (15)	5 (1)
HT+DLp	20.59 (14)	69.12 (47)	10.29 (7)
DM+HT+DLp	26.92 (7)	69.83 (18)	3.85 (1)
CHD	13.33 (4)	76.62 (23)	10 (3)

No significant different by chi-square test

Table 23 Means ± SEM of triglyceride in healthy controls and subjects with various diseases according to frequency of high fat intake

Cases	Frequency of high fat intake		
	Occasional	Frequent	Usual
	← mg/dL →		
Control	89.3±8.50	94.9±7.44	85.7±13.34
HT	-	140.9±31.37	113.0±22.0
HC	98.8±10.15	132.1±6.54	126.0±0.00
HT+DLp	150.4±15.36	153.9±10.54 ^{a2}	152.0±28.21
DM+HT+DLp	433.7±75.07	146.2±12.15 ^{a1}	178.0±51.7
CHD	138.8±25.38	154.7±13.16 ^{a2}	110.7±8.41

Significant different from Control group: $^{a1}p<0.05$, $^{a2}p<0.005$

Table 24 Means ± SEM of serum total cholesterol in healthy controls and subjects with various diseases according to frequency of high fat intake

Cases	Frequency of high fat intake		
	Occasional	Frequent	Usual
	← mg/dL →		
Control	176.7±10.34	179.6±3.89	189.2±3.17
HT	-	181.1±5.20	179.5±18.50
HC	259.0±6.94 [a1]	278.9±7.96 [a1, b2]	259.0±0.00
HT+DLp	263.3±7.06 [a1]	257.1±5.33 [a1, b2, c2]	236.1±5.75
DM+HT+DLp	256.4±12.1 [a1]	245.8±7.23 [a1, b2, c2]	273.5±31.2
CHD	252.8±15.66 [a1]	244.0±10.39 [a1, b2, c2]	239.0±12.7

Significant different from Control group: [a1]$p<0.005$
Significant different from HT: [b1]$p<0.01$, [b2]$p<0.005$
Significant different from HC: [c1]$p<0.05$, [c2]$p<0.005$

Table 25 Means ± SEM of serum LDL-C in healthy controls and subjects with various diseases according to frequency of high fat intake

Cases	Frequency of high fat intake		
	Occasional	Frequent	Usual
	← mg/dL →		
Control	102.1±11.26	108.8±4.16	111.9±4.18
HT	-	98.9±7.41	109.4±17.40
HC	175.0±7.04 [a1]	190.2±9.24 [a1, b1]	172.8±0.00
HT+DLp	168.8±24.36 [a1]	164.1±9.18 [a1, b1, c1]	163.5±15.23
DM+HT+DLp	163.8±10.49 [a1]	163.7±6.88 [a1, b1, c1]	179.6±26.86
CHD	179.8±9.02 [a1]	173.9±5.49 [a1, b1]	150.3±8.49

Significant different from Control group: [a1]$p<0.05$
Significant different from HT: [b1]$p<0.05$, [b2]$p<0.005$
Significant different from HC: [c1]$p<0.05$, [c2]$p<0.005$

Table 26 Means ± SEM of serum HDL-C in healthy controls and subjects with various diseases according to frequency of high fat intake

Cases	Frequency of high fat intake		
	Occasional	Frequent	Usual
	← mg/dL →		
Control	56.7±4.33	51.8±2.36	60.2±4.17
HT	-	54.0±5.94	47.5±5.50
HC	64.3±3.84	62.3±2.59	61.0±0.00
HT+DLp	56.2±7.88	49.0±3.59	53.3±4.70
DM+HT+DLp	48.4±7.34	52.9±2.51	58.2±3.06
CHD	53.4±2.79	52.4±1.71	55.4±6.75

No significant different of serum HDL-C in healthy controls and subjects with various diseases according to frequency of high fat intake

Table 27 Means ± SEM of BMI in healthy controls and subjects with various diseases according to frequency of high fat intake

Cases	Frequency of high fat intake		
	Occasional	Frequent	Usual
Control	23.4±1.65	22.1±0.56	20.7±0.81
HT	-	24.5±1.47	27.1±0.28
HC	19.9±1.22	22.6±0.94	25.4±0.00
HT+DLp	23.8±0.83	25.1±0.44 $^{a2, b2}$	25.9±1.49
DM+HT+DLp	25.4±1.53	25.1±0.64 ^{a1}	27.1±0.95
CHD	24.2±1.05	25.6±0.79 ^{a1}	24.8±1.20

Significant different from Control group: $^{a1}p<0.05$, $^{a2}p<0.01$

Significant different from HC: $^{b1}p<0.05$, $^{b2}p<0.01$

Table 28 Means ± SEM of WHR in healthy controls and subjects with various diseases according to frequency of high fat intake

Cases	Frequency of high fat intake		
	Occasional	Frequent	Usual
Control	0.90±0.03	0.88±0.01 [c1, d1, e2]	0.86±0.02
HT	-	0.92±0.02	0.96±0.01
HC	0.89±0.05	0.87±0.02 [c2, d2, e1]	0.86±0.00
HT+DLp	0.93±0.01	0.93±0.01	0.94±0.02
DM+HT+DLp	0.97±0.01 [a1, b1]	0.95±0.01	0.92±0.04
CHD	0.96±0.03	0.94±0.01	0.90±0.02

Significant different from Control group: [a1]$p<0.05$
Significant different from HC: [b1]$p<0.05$
Significant different from DM+HT+DLp: [c1]$p<0.01$, [c2]$p<0.005$
Significant different from HT+DLp: [d1]$p<0.01$, [d2]$p<0.005$
Significant different from CHD: [e1]$p<0.01$

Table 29 Means ± SEM of 16:0 in healthy controls and subjects with various diseases according to frequency of high fat intake

Cases	Frequency of high fat intake		
	Occasional	Frequent	Usual
	← Percent of total fatty acids →		
Control	22.07±0.89	25.51±1.39	26.94±4.42
HT	-	23.00±3.84	29.02±9.01
HC	23.02±4.50	25.69±2.47	46.49±0.00
HT+DLp	26.06±2.12	26.59±1.24	28.06±3.04
DM+HT+DLp	30.22±3.37	33.44±1.58 [a1, b1, c1, d1, e1]	33.49±2.50
CHD	23.68±3.80	27.22±1.93	24.36±6.98

Significant different from Control group: [a1]$p<0.005$
Significant different from HT: [b1]$p<0.05$
Significant different from HC: [c1]$p<0.05$
Significant different from HT+DLp: [d1]$p<0.05$
Significant different from CHD: [e1]$p<0.05$

Table 30 Means ± SEM of 16:1, n-7 in healthy controls and subjects with various diseases according to frequency of high fat intake

Cases	Frequency of high fat intake		
	Occasional	Frequent	Usual
	← Percent of total fatty acids →		
Control	1.14±0.21	1.21±0.15	1.33±0.28
HT	-	0.90±0.25 [b1]	2.45±1.49
HC	1.13±0.50	1.50±0.21 [a1]	1.41±0.00
HT+DLp	1.56±0.52	1.05±0.10 [b2]	0.86±0.16
DM+HT+DLp	1.12±0.45	0.76±0.13 [b3]	1.45±0.80
CHD	1.84±0.68	1.77±0.32	0.87±0.41

Significant different from DM+HT+DLpl: [a1]$p<0.005$
Significant different from CHD: [b1]$p<0.01$, [b2]$p<0.05$, [b3]$p<0.005$

Table 31 Means ± SEM of 18:1, *t* in healthy controls and subjects with various diseases according to frequency of high fat intake

Cases	Frequency of high fat intake		
	Occasional	Frequent	Usual
	← Percent of total fatty acids →		
Control	0.51±1.64	0.34±0.06	0.36±0.15
HT	-	0.23±0.07	0.67±0.41
HC	0.36±0.12	0.66±0.31 [a1, b1, c1, d1]	2.46±0.00
HT+DLp	0.31±0.05	0.33±0.04	0.32±0.08
DM+HT+DLp	0.28±0.09	0.21±0.07	0.27±0.13
CHD	0.25±0.12	0.30±0.04	0.25±0.14

Significant different from HT: [a1]$p<0.05$
Significant different from DM+HT+DLp: [b1]$p<0.01$
Significant different from HT+DLp: [c1]$p<0.05$
Significant different from CHD: [d1]$p<0.05$

Table 32 Means ± SEM of 18:2, n-6 in healthy controls and subjects with various diseases according to frequency of high fat intake

Cases	Frequency of high fat intake		
	Occasional	Frequent	Usual
	← Percent of total fatty acids →		
Control	7.29±0.46	10.05±1.68	14.13±4.17
HT	-	9.18±2.66	12.36±4.00
HC	13.95±5.05	12.26±2.36	9.37±0.00
HT+DLp	9.70±1.90	8.42±0.68 [a1]	6.29±0.35
DM+HT+DLp	8.70±1.18	10.26±1.4	7.46±0.69
CHD	9.70±2.90	12.56±1.66	6.90±0.35

Significant different from CHD: [a1]p<0.005

Table 33 Means ± SEM of 20:4, n-6 in healthy controls and subjects with various diseases according to frequency of high fat intake

Cases	Frequency of high fat intake		
	Occasional	Frequent	Usual
	← Percent of total fatty acids →		
Control	8.15±1.34	8.41±0.92	6.48± 1.53
HT	-	8.41±1.67 [b1]	7.13±5.75
HC	8.89±2.53	8.10±1.07 [b2]	0.76±0.00
HT+DLp	8.39±1.17	8.06±0.58 [a1]	7.95±2.36
DM+HT+DLp	6.01±2.25	4.40±0.52 [b2]	0.51±1.33
CHD	9.63±1.64	7.22±0.93 [b1]	10.22±2.02

Significant different from Control group: [a1]p<0.005

Significant different from DM+HT+DLp: [a1]p<0.005, [a2]p<0.01

Table 34 Means ± SEM of 18:3, n-3 in healthy controls and subjects with various diseases according to frequency of high fat intake

Cases	Frequency of high fat intake		
	Occasional	Frequent	Usual
	← Percent of total fatty acids →		
Control	4.49±1.06	2.91± 0.44	6.32±3.28
HT	-	10.61±2.41 [a1]	7.79±1.04
HC	3.02±1.22	4.42±0.81 [b1]	4.98±0.00
HT+DLp	3.86±0.58	4.20±0.45 [b1]	4.46±1.24
DM+HT+DLp	3.42±0.89	2.41±0.47 [b1, c1]	1.76±0.26
CHD	5.02±1.30	3.45±0.56 [a1, b1]	6.37±3.72

Significant different from Control group: [a1]$p<0.005$
Significant different from HT: [b1]$p<0.005$
Significant different from HT+DLp: [c1]$p<0.05$

Table 35 Means ± SEM of 20:5, n-3 in healthy controls and subjects with various diseases according to frequency of high fat intake

Cases	Frequency of high fat intake		
	Occasional	Frequent	Usual
	← Percent of total fatty acids →		
Control	2.99±1.51	1.21±0.46	1.13±0.70
HT	-	0.81±0.38	1.12±0.61
HC	0.81±0.18	0.48±0.11	0.28±0.00
HT+DLp	1.04±0.56	0.64±0.09 [b1]	1.34±0.67
DM+HT+DLp	0.38±0.10 [a1]	0.30±0.06 [b1]	0.40±0.18
CHD	0.41±0.08	1.43±0.65	0.50±0.15

Significant different from Control group: [a1]$p<0.005$
Significant different from CHD: [b1]$p<0.05$

Table 36 Means ± SEM of 22:5, n-3 in healthy controls and subjects with various diseases according to frequency of high fat intake

Cases	Frequency of high fat intake		
	Occasional	Frequent	Usual
	← Percent of total fatty acids →		
Control	4.89±0.71	3.58±0.56	3.13±0.99
HT	-	2.42±0.60	2.57±2.09
HC	3.10±1.27	2.54±0.56	2.68±0.00
HT+DLp	2.95±0.59 [a1]	3.73±0.36 [b2]	3.58±0.78
DM+HT+DLp	1.74±0.41 [a1]	2.28±0.47	4.08±1.65
CHD	2.07±0.76 [a1]	1.70±0.38 [a2]	0.49±0.35

Significant different from Control group: [a1]$p<0.05$, [a2]$p<0.01$
Significant different from DM+HT+DLp: [b1]$p<0.05$

Table 37 Means ± SEM of 22:6, n-6 in healthy controls and subjects with various diseases according to frequency of high fat intake

Cases	Frequency of high fat intake		
	Occasional	Frequent	Usual
	← Percent of total fatty acids →		
Control	12.04±1.19	9.68±1.13	3.72±1.40
HT	-	6.57±1.37	5.51±3.97
HC	7.27±2.47	6.10±1.12 [a1]	4.44±0.00
HT+DLp	8.71±1.41	8.33±0.60	7.03±0.79
DM+HT+DLp	4.84±1.67 [a2]	3.26±0.66 [a2, b1]	3.57±0.91
CHD	7.16±1.63	5.44±1.13 [a2, b1]	4.64±2.91

Significant different from Control group: [a1]$p<0.05$, [a2]$p<0.01$, [a3]$p<0.005$
Significant different from HT+DLp: [b1]$p<0.05$

Table 38 Percentage of number of healthy controls and subjects with various diseases according to alcohol intake

Cases	Alcohol intake (N)		
	Never	Quit	Current
Control	56.76 (21)	13.51 (5)	29.73 (11)
HT	36.36 (4)	27.27 (3)	36.36 (4)
HC	60 (15)	8 (2)	32 (8)
HT+DLp	54.67 (41)	16.67 (8)	37.67 (26)
DM+HT+DLp	56.67 (17)	16.67 (5)	26.67 (8)
CHD	63.89 (23)	13.89 (5)	22.32 (8)

No significant different by chi-square test

Table 39 Means ± SEM of triglyceride in healthy controls and subjects with various diseases according to alcohol intake

Cases	Alcohol intake		
	Never	Quit	Current
	← mg/dL →		
Control	96.1±5.84 [c1, d2, e1]	107.6±13.80	78.8±7.41
HT	99.8±19.95	173.0±66.48	142.0±36.80
HC	120.3±8.36	107.5±24.50	119.8±12.06
HT+DLp	140.9±8.65	124.1±18.27	164.1±14.40
DM+HT+DLp	155.5±15.95	171.4±19.32	379.0±243.72 [a1, b1, d1]
CHD	137.3±11.87	152.0±35.09	155.1±17.82

Significant different from Control group: [a1]$p<0.05$

Significant different from HC: [b1]$p<0.05$

Significant different from DM+HT+DLp: [c1]$p<0.01$

Significant different from HT+DLp: [d1]$p<0.05$, [d2]$p<0.01$

Significant different from CHD: [e1]$p<0.01$

Table 40 Means ± SEM of serum total cholesterol in healthy controls and subjects with various diseases according to alcohol intake

Cases	Alcohol intake		
	Never	Quit	Current
	← mg/dL →		
Control	180.6±4.06	177.4±11.00	175.0±6.30
HT	186.8±4.31	178.0±8.89	176.2±10.87
HC	271.3±6.21	275.5±1.50 [a1, b1]	281.9±14.6 [a1, b1]
HT+DLp	245.3±6.48	241.0±19.51 [a1, b1]	230.6±24.79 [a1, b1]
DM+HT+DLp	247.3±9.47	247.4±17.7 [a1, b1]	252.9±11.81 [a1, b1]
CHD	255.6±5.52	268.0±11.6 [a1, b1]	248.2±6.49 [a1, b1]

Significant different from Control group: $^{a1}p<0.005$
Significant different from HT: $^{b1}p<0.005$

Table 41 Means ± SEM of serum LDL-C in healthy controls and subjects with various diseases according to alcohol intake

Cases	Alcohol intake		
	Never	Quit	Current
	← mg/dL →		
Control	107.1±4.10 [a3, b2, c2, d3]	102.3±13.5 [a1, b1, c3, d2]	104.6±6.04 [a2, b2, c2, d2]
HT	106.0±11.28 [a2, b1, c2, d3]	99.7±9.06 [a1, b1, c2, d1]	103.1±13.88 [a2, b1, c1, d1]
HC	182.8±7.38	194.5±0.10	199.2±14.40
HT+DLp	176.2±5.35	189.3±9.76	161.9±7.24
DM+HT+DLp	162.8±7.87	166.5±19.3	166.1±7.59
CHD	164.5±6.29	163.0±13.64	153.8±22.89

Significant different from HC: $^{a1}p<0.01$, $^{a2}p<0.005$
Significant different from DM+HT+DLp: $^{b1}p<0.01$, $^{b2}p<0.005$
Significant different from HT+DLp: $^{c1}p<0.01$, $^{c2}p<0.005$
Significant different from CHD: $^{d1}p<0.05$, $^{d2}p<0.01$, $^{d3}p<0.005$

Table 42 Means ± SEM of serum HDL-C in healthy controls and subjects with various diseases according to alcohol intake

Cases	Alcohol intake		
	Never	Quit	Current
	← mg/dL →		
Control	54.2±2.00	53.6±3.55	54.6±3.78
HT	60.8±10.93	43.7±2.18	47.8±4.66
HC	64.5±2.40 $^{a1, c1, d1, e1}$	59.5±6.50 ^{b1}	58.7±3.00
HT+DLp	51.4±1.79	53.9±3.29	53.5±2.83
DM+HT+DLp	53.4±2.56	46.6±3.50	48.5±6.14
CHD	53.3±3.63	48.0±3.06	45.8±3.73

Significant different from Control group: $^{a1}p<0.05$
Significant different from HT: $^{b1}p<0.05$
Significant different from DM+HT+DLp: $^{c1}p<0.05$
Significant different from HT+DLp: $^{d1}p<0.01$
Significant different from CHD: $^{e1}p<0.01$

Table 43 Means ± SEM of fasting blood sugar in healthy controls and subjects with various diseases according to alcohol intake

Cases	Alcohol intake		
	Never	Quit	Current
	← mg/dL →		
Control	96.7±1.83	97.0±3.48 $^{d1, f1}$	95.9±2.35
HT	100.0±4.62	98.0±1.53	97.0±2.12
HC	93.5±2.11	101.0±2.00	91.9±4.12
HT+DLp	98.8±1.80	95.6±2.43 $^{d1, f1}$	100.4±2.36
DM+HT+DLp	154.6±16.46 $^{a1, b1, c1, e2, f1}$	128.4±14.84	131.0±10.61 $^{a1, b1, c1, e1, f1}$
CHD	107.8±5.86	126.8±14.58	107.8±13.12

Significant different from control group: $^{a1}p<0.005$
Significant different from HT: $^{b1}p<0.01$
Significant different from HC: $^{c1}p<0.005$
Significant different from DM+HT+DLp: $^{d1}p<0.01$
Significant different from HT+DLp: $^{e1}p<0.05$, $^{e2}p<0.005$
Significant different from CHD: $^{f1}p<0.005$

Table 44 Means ± SEM of BMI in healthy controls and subjects with various diseases according to alcohol intake

Cases	Alcohol intake		
	Never	Quit	Current
Control	22.7±0.75 $^{c2, d2, e1, A1, B1}$	19.4±0.77 $^{c1, d1, e1}$	20.4±0.62 $^{a2, b1, c2, d3, e1}$
HT	26.1±2.72	23.8±1.79	25.6±1.34
HC	21.2±0.71 $^{a1, e3, d3, e2, B1}$	22.3±4.67	24.4±1.15
HT+DLp	25.1±0.44	23.8±0.91	24.6±0.72
DM+HT+DLp	26.2±0.58	24.5±1.36	25.4±0.99
CHD	24.7±0.82	26.8±1.21	26.3±1.43

Significant different from HT: $^{a1}p<0.05$, $^{a2}p<0.01$
Significant different from HC: $^{b1}p<0.05$
Significant different from DM+HT+DLp: $^{c1}p<0.05$, $^{c2}p<0.01$, $^{c3}p<0.005$
Significant different from HT+DLp: $^{d1}p<0.05$, $^{d2}p<0.01$, $^{d3}p<0.005$
Significant different from CHD: $^{e1}p<0.05$, $^{e2}p<0.01$
Significant different from quite: $^{A1}p<0.05$
Significant different from current: $^{B1}p<0.05$

Table 45 Means ± SEM of WHR in healthy controls and subjects with various diseases according to alcohol intake

Cases	Alcohol intake		
	Never	Quit	Current
Control	0.88±0.01 ^{c2,e2}	0.86±0.03 $^{a1, c1, d1, e1}$	0.86±0.01 $^{b1, c3, d2, e3}$
HT	0.89±0.02	0.97±0.01	0.90±0.03
HC	0.84±0.02 $^{c3, d2, e2, A1}$	0.93±0.01	0.93±0.03
HT+DLp	0.91±0.01	0.94±0.02	0.94±0.01
DM+HT+DLp	0.94±0.01	0.95±0.01	0.96±0.01
CHD	0.91±0.02	0.94±0.02	0.96±0.01

Significant different from HT: $^{a1}p<0.01$
Significant different from HC: $^{b1}p<0.05$
Significant different from DM+HT+DLp: $^{c1}p<0.05$, $^{c2}p<0.01$, $^{c3}p<0.005$
Significant different from HT+DLp: $^{d1}p<0.01$, $^{d2}p<0.005$
Significant different from CHD: $^{e1}p<0.05$, $^{e2}p<0.01$, $^{e3}p<0.005$
Significant different from current: $^{A1}p<0.05$

Table 46 Means ± SEM of 16:0 in healthy controls and subjects with various diseases according to alcohol intake

Cases	Alcohol intake		
	Never	Quit	Current
	← Percent of total fatty acids →		
Control	25.57±1.71	29.55±3.00	26.64±1.71
HT	25.05±6.16	28.67±8.02	17.62±2.24 [a1, d1, e1]
HC	28.86±2.53	18.01±1.10	24.58±3.31
HT+DLp	26.80±1.29	22.97±3.19	28.53±1.15
DM+HT+DLp	32.31±1.93 [a1, d1]	25.17±7.44	34.65±0.53 [a2, b1, c1, d1, e1]
CHD	27.50±2.28	27.04±3.85	26.86±3.77

Significant different from Control group: [a1]$p<0.05$
Significant different from HT: [b1]$p<0.05$
Significant different from HC: [c1]$p<0.05$
Significant different from HT+DLp: [d1]$p<0.01$
Significant different from CHD: [e1]$p<0.05$

Table 47 Means ± SEM of 16:1, n-7 in healthy controls and subjects with various diseases according to alcohol intake

Cases	Alcohol intake		
	Never	Quit	Current
	← Percent of total fatty acids →		
Control	1.12±0.14 [a1]	1.53±0.72	1.35±0.19
HT	0.78±0.44	2.06±1.01	0.89±0.15
HC	1.73±0.31	0.54±0.22	1.60±0.29
HT+DLp	1.03±0.08 [a1]	1.39±0.39	1.44±0.32
DM+HT+DLp	1.16±0.27	0.41±0.09	0.75±0.16 [a1]
CHD	2.94±1.30	1.61±0.55	2.38±0.72

Significant different from CHD: [a1]$p<0.05$

Table 48 Means ± SEM of 18:1, *t* in healthy controls and subjects with various diseases according to alcohol intake

Cases	Alcohol intake		
	Never	Quit	Current
	← Percent of total fatty acids →		
Control	0.41±0.08 [b1]	0.24±0.08	0.30±0.07 [a1]
HT	0.18±0.07	0.20±0.02	0.52±0.21
HC	0.47±0.15 [b1, c1]	0.21±0.05	0.90±0.56
HT+DLp	0.33±0.03	0.35±0.09	0.31±0.05 [a2]
DM+HT+DLp	0.20±0.04	0.40±0.25	0.09±0.02
CHD	0.25±0.04	0.30±0.07	0.43±0.10 [a2,]

Significant different from HC: [a1]$p<0.01$, [a2]$p<0.01$
Significant different from DM+HT+DLp : [b1]$p<0.05$
Significant different from CHD: [c1]$p<0.05$

Table 49 Means ± SEM of 18:2, n-6 in healthy controls and subjects with various diseases according to alcohol intake

Cases	Alcohol intake		
	Never	Quit	Current
	← Percent of total fatty acids →		
Control	9.70±1.37 [a1]	9.56±2.20	10.52±2.52
HT	9.52±2.83	14.31±5.99	5.23±1.47 [a1]
HC	15.06±2.75	7.73±3.61	15.52±3.58
HT+DLp	7.98±0.65 [a1, b1]	12.32±2.89	8.59±1.06 [a1]
DM+HT+DLp	9.18±1.01 [a1]	10.25±1.24	10.77±2.53
CHD	11.40±1.49	11.44±4.01	11.19±2.92

Significant different from HC: [a1]$p<0.01$
Significant different from CHD: [b1]$p<0.05$

Table 50 Means ± SEM of 20:4, n-6 in healthy controls and subjects with various diseases according to alcohol intake

Cases	Alcohol intake		
	Never	Quit	Current
	← Percent of total fatty acids →		
Control	7.36±0.84	4.65±1.65	7.36±1.25 ^{b1}
HT	8.59±1.99	5.00±4.08	11.16±1.56 ^{b1}
HC	6.57±1.04	13.16±2.83 ^{a1}	8.26±1.53 ^{b1}
HT+DLp	7.82±0.76	7.02±177	7.17±0.71 ^{b1}
DM+HT+DLp	5.60±0.91	5.18±1.53	3.32±0.51
CHD	7.41±0.88	9.17±2.35	6.68±1.95

Significant different from Control group: $^{a1}p<0.01$
Significant different from DM+HT+DLp: $^{b1}p<0.05$
Significant different from never: $^{A1}p<0.05$

Table 51 Means ± SEM of 18:3, n-3 in healthy controls and subjects with various diseases according to alcohol intake

Cases	Alcohol intake		
	Never	Quit	Current
	← Percent of total fatty acids →		
Control	4.33±1.03 ^{a2}	2.25±0.58 ^{a1}	3.19±0.46 ^{a2}
HT	9.87±2.84	8.56±5.64	9.59±3.51
HC	3.48±0.44 ^{a2}	3.68±3.30	4.65±1.46 ^{a2}
HT+DLp	4.16±0.48 ^{a2}	3.95±1.16	3.75±0.47 ^{a2}
DM+HT+DLp	2.78±0.55 ^{a3}	2.80± 0.97	1.71±0.18 ^{a3}
CHD	4.07±0.88 ^{a2}	2.79±1.06	4.40±1.33 ^{a2}

Significant different from HT: $^{a1}p<0.01$, $^{a2}p<0.01$, $^{a3}p<0.005$

Table 52 Means ± SEM of 20:5, n-3 in healthy controls and subjects with various diseases according to alcohol intake

Cases	Alcohol intake		
	Never	Quite	Current
	← Percent of total fatty acids →		
Control	1.79±0.62	2.08±0.93	1.49±0.70
HT	1.35±0.69	0.74±0.50	0.46±0.08
HC	0.51±0.07	0.75±0.33	0.63±0.18
HT+DLp	0.78±0.14	1.40±0.49	0.92±0.34
DM+HT+DLp	0.36±0.06 [a1]	0.35±2.22 [a1]	0.17±0.04
CHD	1.58±0.73	0.50±0.10	1.19±0.51

Significant different from Control group: [a1]p<0.01

Table 53 Means ± SEM of 22:5, n-3 in healthy controls and subjects with various diseases according to alcohol intake

Cases	Alcohol intake		
	Never	Quit	Current
	← Percent of total fatty acids →		
Control	3.49±0.49 [b2]	3.64±1.16	4.07±0.76 [a1, b2]
HT	1.92±0.41	0.79±0.52	4.35±0.42 [a1, b2]
HC	2.53±0.62	2.86±0.86	1.58±0.70
HT+DLp	3.51±0.38 [b2]	2.39±0.88	3.42±0.41 [a1, b2]
DM+HT+DLp	2.55±0.51	2.12±0.38	2.48±0.89
CHD	1.71±0.38	1.19±0.46	1.20±0.48

Significant different from HC: [a1]p<0.01

Significant different from CHD: [b1]p<0.05, [b2]p<0.01

Table 54 Means ± SEM of 22:6, n-3 in healthy controls and subjects with various diseases according to alcohol intake

Cases	Alcohol intake		
	Never	Quit	Current
	← Percent of total fatty acids →		
Control	8.37±1.08	7.50±2.95	8.36±1.34
HT	6.76±2.18	2.91±1.26	9.35±0.97
HC	6.39±1.17	9.57±0.48	3.41±1.10 [a1, b1, c1]
HT+DLp	8.33±0.62	3.73±1.18	8.06±0.93
DM+HT+DLp	3.74±0.66 [a2, c3]	3.41±1.51	2.37±0.79 [a2, b1, c2, d1]
CHD	4.22±0.86 [a2, c3]	4.77±1.84	7.34±2.37

Significant different from Control group: [a1]$p<0.01$
Significant different from HT: [b1]$p<0.05$
Significant different from HC: [c1]$p<0.05$, [c2]$p<0.01$, [c3]$p<0.005$
Significant different from CHD: [d1]$p<0.05$

Table 55 Regression equations (N=222)

Independent variables	Dependent variables	R	R^2	Regression equations	p-value
Smoking status	HDL-C	-0.135	0.018	y = 57.17-0.135x	0.048
Smoking status	BMI	-0.205	0.042	y = 25.76-0.205x	0.003
Alcohol intake	WHR	0.185	0.034	y = 0.891+0.185x	0.007

CHAPTER V
DISCUSSION

1. Serum fatty acids

The present study demonstrated that some serum fatty acid concentrations in subjects with various diseases were significantly different from healthy controls. The fatty acid profile showed that myristic acid (14:0), palmitic acid (16:0) in subjects with DM and HT and DLp were significantly lower and higher than healthy controls and oleic acid (18:0) in subjects with DM and HT and DLp and subjects with HT and DLp also was significantly higher than healthy controls. Although, all of the level of serum saturated fatty acids in all groups of subjects with various diseases were not significantly different from healthy controls but their levels tended to be higher than healthy controls. These results were similar to the report from Miettinen and co-workers (77), they reported that palmitic (16:0) and stearic acid (18:0) of phospholipids were significantly higher, and the linoleic acid; LA (18:2, n-6) and most polyunsaturated fatty acids were lower in CHD patients when compared with controls and they also considered fatty acid pattern of serum phospholipids to be an independent risk of coronary heart disease. These findings reflected that subjects with risk factors of coronary heart disease had high level of serum saturated fatty acids as a result of their high intake of saturated fat, which was a dietary risk factor for coronary heart disease.

Dietary fatty acids composition not only influences serum fatty acid composition but also serum cholesterol profile. Migration studies and international comparisons suggest a strong positive association between saturated fat intake and risk of coronary heart disease (83). In metabolic studies, diets high in saturated fat and low polyunsaturated fat increase blood cholesterol concentration (57, 83). However, different classes of saturated fatty acids can have different effects on plasma lipid and lipoprotein concentrations. Saturated fatty acids with 12-16 carbon atoms tend to

increase plasma total and LDL-C concentrations, whereas stearic acid (18:0) does not have a cholesterol-raising effect compared with oleic acid (18:1). However, stearic acid (18:0) may lower HDL-C and increase lipoprotein (a) [Lp(a)] concentrations. Among the cholesterol-raising saturated fatty acids, myristic acid appears to have cholesterol-rising effect more than lauric acid (12:0) or palmitic acid (16:0) (84, 85).

The differential effects of specific saturated fats on plasma lipids and lipoproteins imply that these fats may have different effects on the risk of coronary heart disease. So far, epidemiologic studies have assessed the association of risk of coronary heart disease with total saturated fatty acids. Hu, 1997 (28) found a modest positive association between saturated fat intake and risk of coronary heart disease in women participating in the Nurse's Health Study.

One of the purposes of this study was to determine total fatty acid concentrations especially n-3, n-6 and *trans* fatty acids of healthy controls and subjects with various diseases. The results in Table 6 showed that serum ALA (18:3, n-3) in subjects with HT was significantly higher than healthy controls. Serum EPA (20:5, n-3) in subjects with HC, subjects with DM and HT and DLp and subjects with HT and DLp was significantly lower than healthy controls and the serum DHA (22:6, n-3) in subjects with DM and HT and DLp and subjects with CHD was significantly lower than healthy controls. The results mentioned above showed the n-3 fatty acids especially DHA and EPA contents in CHD and the risk group of coronary heart diseases to be lower than healthy controls. The results in this study were similar to the study of Das (85), reporting the abnormality of fatty acids level in some cases that the level of EPA and DHA were lower in patients with CHD. In the patients with type2 diabetes mellitus, the level of ALA and DHA were also lower when compared with control group (85). In addition, the results of this report were similar to the study from Yli-JAMA and co-worker (70) that investigated the associated between composition of serum free fatty acids fraction and risk of first myocardial infarction (MI) in 103 patients and 104 population controls from Norway. They found that the mean percentage content of DHA (22:6, n-3), EPA (20:5. n-3) in serum free fatty acid fraction was significantly lower in cases than in controls and suggested that increased percentage content of total n-3 fatty acids in serum free fatty acid fraction was associated with decreased risk of MI. In addition, this study was similar to the study

from Hojo and co-worker (72) that compared the serum lipid profiles of patients with or without significant coronary stenosis and control subjects. They founded that level of HDL-C and EPA were significantly lower in patients with significant coronary stenosis than in the control subjects. It suggests that HDL-C and EPA may have a protective effect on the progress of coronary atherosclerosis.

Some fatty acids are considered to be indirect biomarkers of the consumption of individual foods or food group (59-61, 86). This is the case when the food source is the primary source of the fatty acid and the fatty acid is stable in the sampling medium. Examples of this are the level of short chain fatty acids present in milk (i.e., vacinic acid) and the n-3 polyunsaturated fats found primarily in marine animals (EPA, DHA). The correlations between dietary reports of fish consumption and serum EPA were 0.58-0.75. (50). So the high concentration of EPA and DHA in this study may be altered from long consumption of EPA and DHA rich sources, since EPA and DHA mainly derive from the diet. Epidemiological and dietary intervention studies showed that the fatty acid composition of platelet, plasma phospholipids and serum reflect from the type of dietary intake and therefore may be used to assess an individuals dietary fat intake. The phospholipids fatty acid composition in plasma and the fatty acid composition in serum may be influenced by diet in the short-term, whereas a relatively longer period of time in required for the diet to influence the platelet phospholipids fatty acid composition and the fatty acid composition of one's usual diet is reflected in the composition of the fatty acid pattern in serum and serum fatty acid composition is one of the valuable markers to determine their actual fat intake (86). For example the study from Lemaitre and co-worker (87) that investigated the associations of plasma phospholipids concentration of DHA, EPA and ALA as biomarkers of intake of incident fatal ischemic heart disease (IHD) and incident nonfatal myocardial infraction in older adults. They found significantly lower plasma phospholipids concentration of combined DHA and EPA and a higher concentration of LA than match control. They mentioned that higher plasma phospholipids concentrations of the long chain n-3 polyunsaturated fatty acids DHA and EPA were associated with a lower risk of fatal IHD, whereas the intermediate chain n-3 PUFA ALA was associated with a tendency to lower risk.

This finding may indicate that high intake of EPA and DHA was a benefit effect to protect the control group from CHD and related disease. There are some evidence confirmed that n-3 fatty acids especially EPA and DHA may associated with a low prevalence of cardiovascular disease such as the incidence of heart disease is low in Greenland Eskimos, Alaskan Eskimos and Japanese had a lower intake of saturated fat and high intake of monounsaturated fat and n-3 PUFAs (88-90).

Serum LA concentration in subjects with HC was significantly higher than healthy controls and serum AA level in subjects with DM and HT and DLp was significantly lower than healthy controls (Table 6). The results were similar to Das who found the abnormality of n-6 fatty acids in plasma phospholipid fraction (85). He found that patients with hypertension had significantly higher ALA and GLA and significantly lower LA and AA. In patients with CHD and patients with diabetes, a significantly lower level of AA and dihomo-gamma-linolenic acid; DGLA (20:3, n-6) were found when compared with normal controls. This showed that AA levels relate to coronary heart disease because of the effect on blood coagulation rather than the progress of atherosclerosis because AA is the precursor of thromboxane A_2. In addition, many researches confirmed that the protective effect of serum LA and AA on coronary heart disease and related disease because of many proposed mechanism. First, dietary intake of n-6 fatty acids may reduce the risk of coronary heart disease and related disease by blood pressure-lowering effect. Second, a previous study showed that dietary intake of LA may improve glucose tolerance and has a cholesterol lowering effect. Finally, n-6 fatty acids also platelet aggregation and enhances erythrocyte deformation (56-58).

In this study, LA was higher than ALA in healthy controls and subjects with various diseases except subjects with HT. Higher amounts of LA will suppress the conversion of ALA to EPA and DHA because both LA and ALA use the same enzyme set for elongation and desaturation to AA, EPA and DHA although, the conversion of ALA to EPA and DHA occurs to a lesser extent about 10-15% in the adult human body (91) From the results, all groups of subjects showed a higher AA than EPA. Perhaps the lack of EPA contributed to the increases platelet aggregation through the absence of competition with AA for the cyclooxygenase pathway in which

AA will liberate more TXA, which can increase platelet aggregation, making a blood clot at the site of the plaque, leading to myocardial infraction and death. (51)

A major role of fatty acids in the body is as structural components of membranes. *Trans* fatty acids that cannot be synthesized by man, may provide an indication of membrane fatty acid composition, in addition present a reliable estimate of the dietary intake of these fatty acids in the period surrounding the blood sampling. Apart from blood lipid and lipoprotein concentrations, *trans* fatty acids might also influence CHD risk via effect on membrane function, prostaglandin synthesis and homeostasis.

The pattern of *trans* fatty acids formed in the partial hydrogenation of vegetable oils differs from that found from ruminant sources. Much of the *trans* fatty acids from ruminants is *trans* vaccenic acid, and the main *trans* fatty acid found in hydrogenated vegetable oil is elaidic acid, although other *trans* fatty acids are also present. Given the evidence for adverse effects of *trans* fatty acids from hydrogenated vegetable oil on blood lipid and lipoprotein concentration and the results of studies indicating that a higher intake of *trans* fatty acids from hydrogenated vegetable oil is associated with increased risk of MI and CHD, a positive association between elaidic acid and other *trans* fatty acids found in hydrogenated vegetable oils, and CAD might be expected. It has been suggest that the combined results of metabolic and epidemiological studies provide strong evidence for a causal relationship between *trans* fatty acids intake and CHD risk.

In this current study, the intake of *trans* fatty acids was not assessed but it has been estimated by the mean of trans fatty acids in serum because of high intake of *trans* fatty acids was reflect to high content of them in serum. Elaidic acid, *t* concentration in subjects with various diseases except subjects with DM and HT and DLp were similarly to control group. This result was similar to the study from Lucy and co-worker that performed a study in CHD patients, and measured the *trans* fatty acid concentration of the plasma phospholipids fraction between a cases group and control group. They founded that saturated fatty acids was slightly though significantly higher in cases with CHD than controls. For all other fatty acids, *trans* fatty acids (16:1,n-7, 18:1, n-9, 18:2, n-6) values were similar in cases with CHD and controls (81) and also was similar to Jonathan (92), that measured platelet trans fatty acid and

assessed the relationships between trans fatty acids and degree of CAD in 191 non diabetic patients. They founded the level of elaidic acid was similar level with the result in this study and founded the positive association between elaidic acid and the extent score of CAD. But this result was not similar to the study from Roberts and co-worker (93), that studied a population case-control study of sudden cardiac death to test the hypothesis that *trans* isomers of oleic acid and LA increase the risk of sudden cardiac death due to CAD. In adipose tissue obtained at necropsy from 66 cases of sudden cardiac death and taken from 286 healthy age and sex match controls. In cases, the mean percentage of total *trans* fatty acids (18:1 plus 18:2) was significantly lower in healthy controls. However, *trans* oleic acid was negatively associated with risk of sudden cardiac death, whereas no association with trans form of LA was seen. Roberts et al study does not support the hypothesis that *trans* isomers increase the risk of sudden cardiac death (93).

2. Serum lipid and some anthropometric parameters

Triglyceride

TG was recognized as a risk factor for CHD especially when accompanied by a low serum HDL-C. In this study, subjects with DM and HT and DLp and subjects with HT and DLp had significantly higher serum TG level than healthy controls. Serum TG levels in subjects with DM and HT and DLp, subjects with HT and DLp and healthy controls were 211.8, 149.5 and 91.8 mg/L, respectively. Base on the NCEP criteria (94) in the second ATP report, the desirable TG level was less than 150 mg/dL, in this study, only subjects with DM and HT and DLp had TG level higher than 150 mg/dL and the level was higher than the report of Tanphaichitr in CEGAT study in 1995 (95).

Total cholesterol

According to the NCEP (94), the desirable level of serum TC was below 200 mg/dL. Results of this study showed that healthy controls and subjects with HT had TC level lower than 200 mg/dL but another group of subjects had high serum TC, mean serum TC was 275.0, 250.7, 254.3 and 242.1 mg/dL among subjects with

HC, subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD. Means serum TC among subjects with various diseases was higher than rural subjects in ampole Khon Kaen, Thailand in 1993 with TC level was 162 mg-dL (96).

High density lipoprotein

According to the NCEP (94), serum HDL-C level less than 35 mg/dL was associated with high risk for CHD. It was an independent and powerful predictor for CHD incidence. This study showed significantly higher mean serum HDL-C level between subjects with HC than healthy controls but mean of HDL-C level in every group of subjects in this study were above 35 mg/dL (Table 7), similar to Tanphaichitr in 1995 (95).

Low density lipoprotein

According to the NCEP (94), the desirable serum LDL-C was indicated at below 100 mg/dL. This study showed that every group of subjects had LDL-C above optimal level (100 mg/dL), especially subjects with HC, subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD had high serum LDL-C level, which were 188.9, 165.7, 171.8 and 162.8, respectively. This results was similar to Tanphaichitr in 1995 (95) that found EGAT subjects had LDL-C level higher than optimal level (100 mg/dL).

Fasting blood glucose

According to American Diabetes Association (97), the desirable serum FBG level higher than 126 mg/dL was associated with high risk of CHD. Table 7 showed that every group of subjects had serum FBG level under the optimal level (126 mg/dL) except the subjects with DM and HT and DLp, who had serum FBG level higher than optimal level, which was 147.7 mg/dL. Framingham data suggest that hyperglycemia was an independent risk factor. It has been determined that patients with DM carry an increased risk for CHD (97).

Obesity

Body Mass Index (BMI), which described relative weight for height, was significantly correlated with total body fat content. The BMI should be used to assess overweight and obesity ant to monitor changes in body weight. In addition, measurements of body weight alone could be used to determine efficacy of weight loss therapy. Weight classifications by BMI, based on NHLBI expert panel (98); BMI of<18.5 kg/m^2, 18.5-24.9 kg/m^2, 25.0-29.9 kg/m^2, 30-34.9 kg/m^2, 35-39.9 kg/m^2, >40 kg/m^2 were classified as underweight, normal weight, overweight, obesity class 1, obesity class 2, obesity class 3 or extreme obesity. Table 7 showed that healthy controls, subjects with HC and subjects with HT and DLp had normal BMI but subjects with HT, subjects with DM and HT and DLp and subjects with CHD had overweight, which was 25.3, 25.6 and 25.6 kg/m^2. Obesity predisposes to CVD in general and to stroke in particular. However, obesity prevalence increased with advancing age and obesity was associated with increased blood pressure, blood sugar and blood lipid.

Waist Hip ratio

WHR, which described relative waist circumference for hip circumference, was significantly correlated with intra-abdominal fat. The desirable WHR lower the 0.77 and 0.9 in women and men, respectively. The most subjects in this study were men therefore 0.9 was used to as a cut-off value for WHR. Table 6 showed that healthy controls and subjects with HC were had normal WHR but WHR in subjects with HT subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD were higher than cut-off value, considered centrally obese which are positively associated with risk for CHD.

3. Lifestyle

According to the frequency of exercise, there were no significant different among frequency of exercise pattern among healthy controls and subjects with various diseases. Frequency of exercise had no effect on serum TG, TC, LDL-C and BMI in healthy controls and subjects with various diseases. Although, subjects with CHD who often exercise and subjects with DM and HT and DLp who usual and

always exercise and subjects with HT and DLp who always exercise had significantly higher serum TG level than healthy controls. However, the result also showed that subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD had higher TG level than healthy control and subjects with HT only. Table 10-11 showed subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD had significantly higher serum TC, LDL-C level than healthy controls and subjects with HT. The result from table 12 and 13 showed that frequency of exercise had influent on the level of serum HDL-C in subjects with CHD. Subjects with CHD who had usual and always exercise had significantly higher level of HDL-C than subjects who had occasional and frequent exercise. However, the subjects with HC who always exercise had significantly higher WHR than the subjects who usually had exercise.

According to cigarette smoking, smoking status was not significantly different between healthy controls and subjects with various diseases. Smoking status affected only serum LDL-C in subjects with DM and HT and DLp that was subjects with DM and HT and DLp who current smoking had significantly higher level of LDL-C than those who quit smoking (Table 17-21). Smoking status was also effect on BMI in healthy controls and subjects with CHD. Subjects with HC, subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD who never smoke and quit smoking had serum TC, LDL-C level significantly differ from healthy controls and subjects with HT. They tended to have higher level of serum TG too.

4. Dietary habit

According to high fat intake, frequency of high fat intake pattern was not significantly different from healthy controls and subjects with various diseases by chi-square test. Frequency of high fat intake did not affect on serum lipid and serum fatty acids in healthy controls and subjects with various diseases. Serum LDL-C and serum TC in subjects with HC, subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD who never smoke and quit smoking were significantly different from healthy controls and subjects with HT and they tended to have high level of triglyceride and BMI but there was no significant difference from serum HDL-C in healthy control and subjects with various diseases. The clupadonic

acid and DHA in subjects with CHD who frequent had high fat intake were significantly different from healthy control and subjects with various disease tended to have lower level of this fatty acids concentrations than healthy controls.

According to alcohol consumption, frequency of alcohol intake pattern was not significantly different between healthy controls and subjects with various diseases by chi-square test. Alcohol intake affected BMI and WHR in subjects with HC, subjects with HC who never had alcohol intake had lower BMI and WHR than those who current had alcohol intake. Serum LDL-C in subjects with HC, subjects with DM and HT and DLp, subjects with HT and DLp and subjects with CHD who never smoke, quit smoking and currently smoke had alcohol intake significantly higher than healthy control and subjects with HT. This was similar to the results of serum TC and FBG. The clupadonic acid and DHA in subjects with various diseases were tended to be lower than healthy controls. These findings indicated that the frequency of exercise, smoking habit and alcohol consumption had influence affect on serum lipids and fatty acids pattern.

CHAPTER VI
CONCLUSION

This study investigated and compared the serum fatty acids concentration, lifestyle and dietary habit among healthy controls, subjects who had risk factor of coronary heart disease and subjects with coronary heart disease. The significant findings were as follow:

1. Serum fatty acids and dietary habit were important risk factors of coronary heart disease and can be use to assess the risk of coronary heart disease.
2. Serum DHA (22:6, n-3) and Clupadonic acid (22:5, n-3) content in healthy control were significantly higher than subjects with coronary heart disease.
3. Frequency of exercise influenced the serum level of HDL-C in subjects with CHD and WHR in subjects with HC.
4. Smoking status affected only serum LDL-C in subjects with DM and HT and DLp and BMI in healthy controls and subjects with CHD.
5. Frequency of high fat intake did not have any effect on serum lipid and serum fatty acids in healthy controls and subjects with various diseases.
6. Alcohol intake affected BMI and WHR in subjects with HC.
7. The positive correlation was found in the alcohol intake and WHR.
8. There were negative correlation between smoking habit and BMI and HDL-C.

REFERENCES

1 Swales SD, de Bono DP. Cardiovascular risk factors. New York: Gower Medical Publishing, 1993.

2 Reeder B, Layzon R, Nair C, Petrasovis A. Cardiovascular disease in Canada. Ottawa: Heart and Stroke Foudamantal of Canada, 1991.

3 Connor Sl, Gustafson JR, Artaud-Wild SM, et al. The cholesterol/saturated fat index; an indication of the hypercholesterolemia and atherogenic potential; of food. Lancet 1986; 2: 1229-32.

4 Pan American Health Organization. Health in the Americans. Washington, D.C.: Publication no. 569. PAHO, 1998.

5 Ministry of Interior. Number and percentage of deaths from heart disease by sex and age group, 1996-2000, available at http://203.157.19.191./Pla22 html.

6 Kannel WB. Contributions of the Framingham Study to the conquest of coronary artery disease. Am J Cardiol 1988; 62: 1109-12.

7 Levy D, Wilson PW, Anderson KM, Castelli WP. Stratifying the patient at risk from coronary disease: new insights from the Framingham Heart Study. Am Heart J 1990; 119: 712-7.

8 National Institutes of Health. Clinical guidelines on the identification, evaluation and treatment of overweight and obesity in adults- the evidence report. Obes Res 1998; (suppl): s51-209.

9 Despr′es JP. Abdominal obesity and the risk of coronary artery disease. Can J Cardiol 1992; 8: 561-2.

10 Slawta JN, Wilcox AR and et al. Health behavior, body composition and coronary heart disease risk in women with multiple sclerosis. Arch Phys Med Rehabil 2003; 84: 1823-1830.

11 Konishi M, lida M, Naito Y, et al. The trend of coronary heart disease and its risk factors based on epidemiological investigations. Jpn Circ J 1987; 51: 319-341.

12 Yoshiike N, Matsumura Y, Lwaya M, Sugiyama M, Yamaguchi M. Nation nutrition survey in Japan. J Epidemiol 1996; 6(3 suppl): S189-200.

13 Bang HO, Dyerberg J, Sinclair HM. The composition of the Eskimo food in north western Greenland. Am J Clin Nutr 1980; 33: 2657-61.

14 Hirai A, Hamazaki T, Terano T, et al. Eicosapentaenoic acid and platelet function in Japan. Lancet 1980; 2: 1132-3.

15 Erkkila AT, Lehto S, Pyorala K and Uusitupa MI. N-3 fatty acids and 5-y risks of death and cardiovascular disease events in patients with coronary artery disease. Am J Cli Nutr 2003; 78: 65-71.

16 Hu FB, Bronner L, Willett WC, et al. Fish and omega-3 fatty acid intake and risk of coronary heart disease in women. JAMA 2002; 287: 1815-21.

17 Bucher HC, Hengstler P, Schingler C, Meier. N-3 Polyunsaturated fatty acids in coronary heart disease: a meta analysis of randomized controlled trials. Am J Med 2002; 112: 298-304.

18 Burr ML, Fehily AM, Gilbert JF, et al. Effects of changed in fat, fish, and fiber intakes on death and myocardial infraction: diet and infraction trail (DART). Lancet 1989; 2: 757-61.

19 Harris WS. Fish oils and plasma lipid and lipoprotein metabolism in humans: a critical review. J Lipid Res 1989; 30: 785-807.

20 Bang H, Dyerberg J, Hjorne N. The composition of food consumed by Greenland Eskimos. Acta Med Scand 1976; 200: 69-73.

21 Dyerberg J, Bang HO, Stofferson E, Moncada S, Vane JR. Eicosapentaenoic acid and prevention of thrombosis and atherosclerosis. Lancet 1987; 2: 117-9.

22 Kromhout D, Bosschieter EB, Coulander C. The inverse relation between fish consumption and 20-year mortality from coronary hear disease. N Eng J Med 1985; 312: 1205-9.

23 Oomen CM, Feskens EJM, Rasanen L, et al. Fish consumption and coronary heart disease mortality in Findland, Italy and the Netherland. Am J Epidemiol 2000; 151: 999-1006.

24 Shekelle RB, Missel L, Paul O, Shryck AM, Stamler L. Fish consumption and mortality from coronary heart disease. N Eng J med 1985; 313: 820 (latter).

25 Schmit EB and Dyerbergb J. Omega-3 fatty acids. Current status in cardiovascular medicine. Druge 1994; 47: 405-24.

26 Hu FB, Stampfer MJ, Manson JE, et al. Dietary fat intake and the risk of coronary heart disease in women. The New England Journal of Medicine 2003; 337: 1491-99.

27 Willett WC, Stampfer MJ, Manson JE, et al: Intake of *trans* fatty acids and risk of coronary heart disease among women. Lancet 1993; 341: 581-5.

28 Hu FB, Stampfer MJ, Manson JE, et al: Dietary fat intake and the risk of coronary heart disease in women. N Eng J Med 1997; 337: 1491-9.

29 Ascherio A, Katan MB, Zock PL, et al: Trans fatty acids and coronary heart disease. N Engl J Med 1999; 340: 1994-8.

30 Mensink RP, Katan MB: Effect of dietary trans fatty acids on high-density and low-density lipoprotein cholesterol levels in healthy subjects. N Engl J Med 1990; 323: 439-45.

31 Mensink RP, Zock PL, Katan MB, et al: Effect of dietary cis and trans fatty acids on serum lipoprotein[a] levels in humans. J Lipid Res 1992; 33: 1493-1502.

32 WHO MONICA PROJECT. WHO MONICA project: Objectives and design. Int J Epidemiol 1989; 18: 3(Suppl): S29-34.

33 Tanphichitr V. Dyslipidemia and CHD. Intern Med 1987; 3: 188.

34 Tanphichitr V, Leelahagul P, Pakpeankitvatana R. Dietary prescription of protein and fat. In Kim WH, Lee YC, Lee KY, Ju Js, Kim SH, eds. Proceedings of the 14th International Congress of Nutrition. Vol I. Seoul: Ewha Womans University 1989 p.553-6.

35 Ross R. The pathogenesis of atherosclerosis-an update. N Eng J Med 1986; 314: 488-500.

36 Steinberg D. Lipoproteins and atherosclerosis: a look back and a look ahead. Arteriosclerosis 1983; 3: 288-301.

37 Munro JM and Cotran RA. The pathogenesis of arthrosclerosis: arthrogenesis and inflammation. Lab Invest 1988; 58: 249-61.

38 Blumgart HL. Blood lipids and atherosclerosis. Am J Med 1951; 11: 271-3.

39 Swank RL and Grimsgaard A. Multiple sclerosis: the lipid relationship. Am J Clin Nutr 1988; 48: 1387-93.

40 Kannel WB. Update on the role of cigarette smoking in coronary artery disease. Am Heart J 1981; 101: 319-28.

41 Neaton JD and Wentworth D. Serum cholesterol, blood pressure, cigarette smoking and death from CHD. Arch Intern Med 1992; 152: 56-64.

42 Reed PB. Nutrition:An Applied science. St. Paul: West publishing company; 1980. p.70-81.

43 Spiller GA. Handbook of Lipid in Human Nutrition. New York: CRC Press, Inc; 1996. p.3.

44 Mason P. Fatty acid: which ones do we need?. The Pharmaceutical Journal 2004; 273: 750-2.

45 Allison DB, Denke MA, Dietschy OM, Emken OE, Kris-Etherton PM and Nicolosi RJ. Trans fatty acids and coronary heart disease risk. Report of the expert panel on trans fatty acids and coronary heart disease. Am J Clin Nutr 1998: 62(3); 655S.

46 Kreutler PA. Nutrition in perspective.New Jersey: Prentice-Hall, Inc. 1980. p.87.

47 Leaf A and Waber PC. Cardiovascular effect of n-3 fatty acids. N Eng J Med 1988; 6: 31-35.

48 Wijendran V and Hayes KC. Dietary n-6 and n-3 Fatty acid balance and cardiovascular health. Annu. Rev. Nutr. 2004; 24: 597-615.

49 Nicolosi RJ, Stucchi AF, Kowala MC et al. Effect of dietary fat saturation and cholesterol on low density lipoprotein composition and metabolism. I. In vivo studies of receptor and non-receptor mediated catabolism of LDL in Cebus monkeys. Arteriosclerosis 1990; 10: 119-28.

50 Hu FB, Manson JE and Willett WC. Types of dietary fat and risk of coronary heart disease: A critical review. J Amer Coll Nutr 2001; 20: 5-19.

51 Holub BJ. Clinical nutrition: 4. Omega-3 fatty acids in cardiovascular care. CMAJ 2002; 166: 608-14.

52 Weber PC, Leaf A. Cardiovascular effect of omega-3 fatty acids. Atherosclerosis risk factor modification by omega-3 fatty acids. World Rev Nutr Diet 1991; 66: 8-32.

53 Valek J, Hammer J, Kohout M, Grafnetter D, Vondra K, Topinka V. Serum linoleic acid and cardiovascular death in postinferction middle-aged men. Atherosclerosis 1985; 54: 111-8.

54 Lovejoy J. and DiGirolamo M. Habitual dietary intake and insulin sensitivity in lean and obese adults. Am J Clin Nutr 1992; 55: 1174-9.

55 LoveJoy JC. Dietary fatty acids and insulin resistance. Curr Atheroscler Rep 1999; 1: 215-20.

56 Hayes KC. Dietary fatty acids, cholesterol and the lipoprotein profile. Br. J. Nutr. 2000; 84: 397-9.

57 Hegsted Dm, McGandy RB, Myers ML and Stare FJ. Quantitative effects of dietary fat on serum cholesterol in men. Am J. Clin. Nutr 1965; 17: 281-95.

58 Grundy SM and Denke MA. Dietary influences on serum lipids and lipoproteins. Journal of Lipid Research 1990; 31: 1149-72.

59 Simopoulos AP. Essential fatty acids in health and chronic disease. Am J Clin Nutr 1999; 70(Suppl): 560S-9S.

60 Larque E, Zamora S and Gil A. Dietary trans fatty acids in early life: a review. Early Human Development 2001; 65(Suppl): S31-S41.

61 Judd JT, Clevidence BA, Muesing RA, Wittes J, Sunkin ME, Podczasy JJ. Dietary trans fatty acids: effects of plasma lipids and lipoproteins of healthy men and women. Am J Clin Nutr 1994; 59: 861-68.

62 Holman RT**,** Caster WO and Wiese HF. Estimation of Linoleate Intake of Men from Serum Lipid Analysis**.** Am. J. Clin Nutr 1964; 14: 193-196.

63 Schmidt EB, Arnesen H, de Caterina R, Rasmussen LH and Kristensen SD. Marine n-3 polyunsaturated fatty acids and coronary heart disease available at http:intl.elsevierhealth.com/journal/thre.html.

64 Bang Ho, Dyerberg J and Niesel AB. Plasma and lipid and lipoprotein pattern in Greenlandic West Coast Eskimos. Lancet 1971; 1: 1143-6.

65 Gesdes Lu, Schmidt EB, Klausen IC et al. Plasma concentration levels of apolipoprotein A-I and lipoprotein (a) in Greenland Inuit (Eskimos). J Int Med 1992; 231: 623-5.

66 Norell SE, Ahlbom A, Feychting M and Pederson NL. Fish consumption and mortality from coronary heart disease. Br Med J 1986; 293: 426-30.

67 Dewailly E, Blanchet C, Gingras S, Lemieux S and Holub BJ. Cardiovascular disease risk factors and n-3 fatty acids status in the adult population of James Bay Cree. Am J Clin Nutr 2002; 76: 85-92.

68 Dewailly E, Blanchet C, Gingras S, Lemieux S, Sauve L, Bergeron J and Holub BJ. Relation between n-3 fatty acid status and cardiovascular disease risk factors among Quebecers. Am J Clin Nutr 2001; 74: 603-11.

69 Dewailly E, Blanchet C, Lemieux S, Sauve L, Gingras S, Ayotte P and Holub BJ. N-3 Fatty acids and cardiovascular disease risk factors among the Inuit of Nunavik. Am J Clin Nutr 2001; 74: 464-73.

70 JAMA PY, Meyer HE, Ringstad J and Pedersen JI. Serum free fatty acid pattern and risk of myocardial infarction: a case-control study. J Intern Med 2002; 251: 19-28.

71 Hongtong K, Boonnim D, Pakpeankitwatana R, Hamroongroj T, Chantaranipapong Y and Changbumrung S. Platelet fatty acids in coronary heart disease, dyslipidemia, hypertension and healthy controls. Southeast Asian J Trop Med Public Health 2003; 34: 675-80.

72 Hojo N, Fukushima T, Isobe A, Gao T, Shiwaku K, Ishida K, Ohta N and Yamana Y. Effect of serum fatty acid composition on coronary atherosclerosis in Japan. Intern J Cardiol 1998; 66: 31-8.

73 Lewis B. Comparison of plasma cholesterol ester in relation to coronary heart disease and dietary fat. Lancet 1958; ii: 71-3.

74 Kingrbery KJ, Morgan DM, Aylott C, Borton P, Emmerson R and Robinson PJ. A comparison of polyunsaturated fatty acids of the plasma cholesterol ester and subcutaneous depot fats of atheromatous and normal people. Cli. Sci. 1962; 22: 161-70.

75 Schrade W, Boehle E. and Biegler R. Humoral changes in arteriosclerosis investigation on lipid, fatty acids, ketone bodies, pyruvic acid, lactic acid and glucose in the blood. Lancet 1960; ii: 1409-16.

76 Brouwer DA, van der Dijs FP, Leerink CB, Steward HN, Kroon TA, Suverkropp GH, Romer JW, van Doormaal JJ and Muskiet FA. The dietary fatty acids of patients with coronary artery disease and controls in Curscao, Implications for primary and secondary prevention. West Indian Med J. 1997; 42: 53-6.

77 Miettinen TA, Naukkarinen V, Huttunen JK, Mattila S and Kumlin T. Fatty acid composition of serum lipids predicts myocardial infarction. Br Med J (Clin Res Ed). 1982; 285: 993-6.

78 Smith CB, Woodward M, Fenton S, Brown CA. Dose dietary trans fatty acid intake relate to the prevalence of coronary heart disease in Scotland? Eur Heart J 1996; 17: 837-45.

79 Kim KM and Campos H. Intake of trans fatty acids and low-density lipoprotein size in Costa Rican population. Metabolism 2003; 52: 693-8.

80 Baylin A, Kabagambe EK, Ascherio A, Spiegelman D and Campos H. High 18:2 trans fatty acids in adipose tissue are associated with increased risk of nonfatal acute myocardial infarction in Costa Rican adults. Amer Soc for Nutr Science 2003; 2: 1186-91.

81 Lucy PL, van de Vijver, Poppel GV, Houwelingen AV, Kruyssen DA and Hornstra G. Trans unsaturated fatty acids in plasma phospholipids and coronary heart disease: a case-control study. Atherosclerosis 1996; 126: 155-61.

82 Lepage G and Roy CC. Direct transesterification of all lipid in a one step reaction. J Lip Res 1986; 27: 114-20.

83 Kato H, Tillostson J, Nichamen MZ, Rhoads GG and Hamiton HB. Epidemiologic studies of coronary heart disease and stroke in Japanese men living in Japan, Haiwaii and California: serum lipids and diet. Am J Epidemiol 1973; 97:372-85.

84 Kris-Etherton PM and Yu S. Individual fatty acid effects on plasma lipids and lipoprotein: human studies, Am J Cli Nutr 1997;65(suppl):1628-44.

85 Das UN. Essential fatty acid metabolism in patients with essential hypertension, diabetes mellitus and coronary heart disease. Pros Leuk Essen 1995; 52: 387-91.

86 Ribili E, Ronnholm H and Saracci R. Biological markers of diet. Cancer Serv 1987; 6: 687-718.

87 Lemaitre RN, King IB and Mazaffarian D. n-3 polyunsaturated fatty acids, fatal ischemic heart disease and nonfatal myocardial infraction in order aldults: the Cardiovascular Heart Study. Am J Clin Nutr 2003; 77: 319-25.

88 Bjerregaard P and Dyerberg J. Mortality from ischemic heart disease and cerebrovascular disease in Greenland. Int J Epidimol 1988; 17: 514-19.

89 Newman WP, Propst MT and Middaugh JP. Atherosclerosis in Alaska natives and nonnatives. Lancet 1993; 3: 1056-7.

90 Harris WS. Fish oils and plasma lipid and lipoprotein metabolism in human: a critical review. J Lipid Res 1989; 30: 785-807.

91 Emken EA, Adlof RO and G ulley RM. Dietary linoleic acid influences desaturation and acylation of deuterium-labeled linoleic and linolenic acids in young adult males. Biochim Biophys Acta 1994; 1213: 177-188.

92 Hodson JM, Wahlqvist ML, Boxall JA and Balaxs ND. Pletelet trans fatty acids in relation to angiographically assessed coronary atery disease. Atherosclerosis 1996; 120: 147-154.

93 Robert TL, Wood DA, Riemersma RA, Gallagher PJ and Lampe FC. Trans isomers of oleic and linoleic acids in adipose tissue and sudden cardiac death. Lancet 1995; 345: 278-82.

94 Third Report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evalustion and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III) Executive Summary National Cholesterol Education Program National Heart, Lung and Blood Institute National Institutes of Health 2001 NIH publication No. 01-3670.

95 วิชัย ตันไพจิตร, รัตนา พากเพียรกิจวัฒนา และปรียา ลีฬหกุล. ความผิดปกติของระดับไขมันในเลือดของคนไทยในเขตเมือง. โภชนศาสตร์คลินิก 2538: 4; 4105-11.

96 Bhuripanyo K, Tatsanavivat P, Matrakool B, Bhuripanyo P, Harnthaveesompol S. A prevalence survey of lipid adnormalitise of rural area in Amphoe, Khon Kean. J Med Assoc Thailand 1993; 46(2): 101-8.

97 American Diabetes Association: consensus statement. Detection and management of lipid disorders in diabetes. Diabetes Care 1993; 16(suppl): 106-21.

98 NHLBI Obesity Education Initiative. The Practical Guide Identification, Evaluation and Treatment of Overweight and Obesity in Adults. Institutes of Health, National Heart, Lung and Blood Institute; 2000 NIH Publication Number 00-4084.

BIOGRAPHY

NAME	Khakhanang Ratananikom
DATE OF BIRTH	July 11, 1980
PLACE OF BIRTH	Bangkok
INSITUTIONS ATTENDED	
	Burapha University, 1998-2001
	B.Sc. (Horn. 2 Biochemistry)
HOME ADDRESS	491 Maungtong 2 Project 3, Phattanakan 53 Road, Suanloang, Bangkok 10250, Thailand