HYPERHOMOCYSTEINEMIA AS A CARDIOVASCULAR RISK FACTOR IN YOUNG SOUTH INDIAN POPULATION

Dissertation submitted

In partial fulfillment of the requirements for the degree of

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GENERAL MEDICINE

TIRUNELVELI MEDICAL COLLEGE

THE TAMILNADU DR. M.G.R. MEDICAL UNIVERSITY

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April 2012

CERTIFICATE

This is to certify that the dissertation entitled **"HYPERHOMOCYSTEINEMIA AS A CARDIOVASCULAR RISK FACTOR IN YOUNG SOUTH INDIAN POPULATION"** submitted by **DR.PRAVEEN SINGH PETER.J** to The Tamilnadu Dr. M.G.R. Medical University, Chennai, in partial fulfillment for the award of Doctor of Medicine, is a bonafide work carried out by him under my guidance and supervision during the academic year 2009-2012. This dissertation partially or fully has not been submitted for any other degree or diploma of this university or other universities.

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DECLARATION

I, **Dr.PRAVEEN SINGH PETER.J, solemnly** declare that the dissertation titled **"HYPERHOMOCYSTEINEMIA AS A CARDIOVASCULAR RISK FACTOR IN YOUNG SOUTH INDIAN POPULATION"** has been prepared by me. This is submitted to the TamilnaduDr.M.G.R. Medical University, Chennai, in partial fulfillment of the regulations for the award of MD Degree Branch I (General Medicine). It was not submitted to the award of any degree/diploma to any University either in part or in full.

Dr.PRAVEEN SINGH PETER.J

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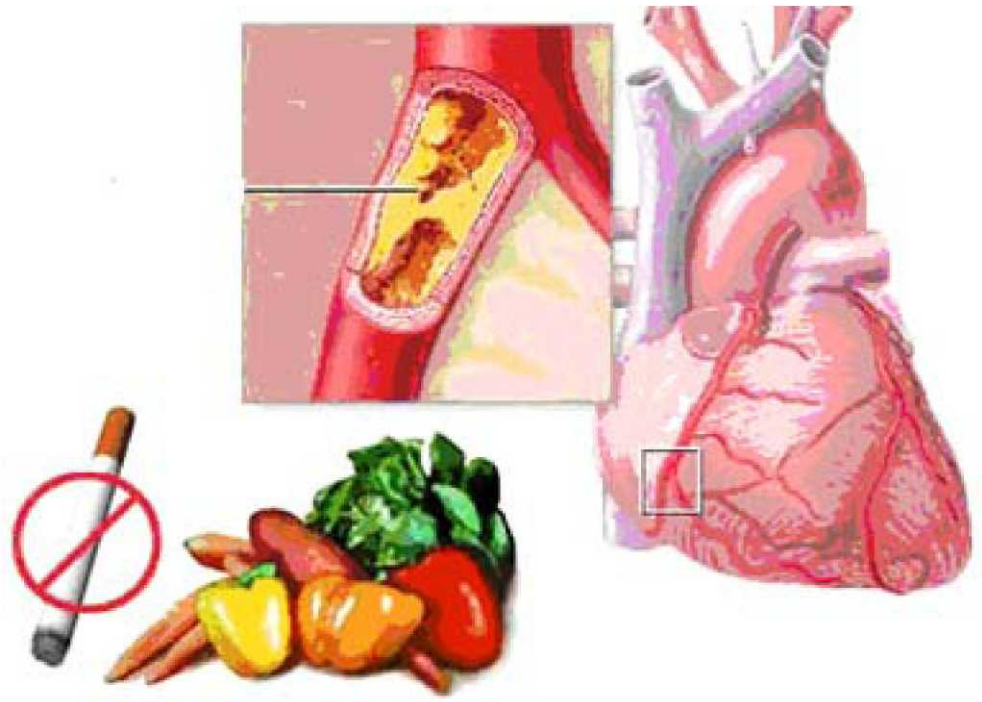
CONTENTS

ABBREVIATIONS

CAD	**-**	**Coronary Artery Disease**
DM	**-**	**Diabetes Mellitus**
Hcy	**-**	**Homocysteine**
tHcy	**-**	**Total homocysteine**
h/o	**-**	**History of**
MI	**-**	**Myocardial Infarction**



INTRODUCTION

INTRODUCTION

Myocardial Infarction generally occurs with the abrupt decrease in coronary blood flow that follows a thrombotic occlusion of a coronary artery previously narrowed by atherosclerosis. The injury is produced by or facilitated by factors such as cigarette smoking, hypertension, lipid accumulation, Diabetes and a number of other factors.

A quote from Berkeley heart lab - "Just being an Indian descent puts you at a high risk of coronary artery disease". The pattern of coronary artery disease is indeed changing in India.

It has been reported to be as follows:

a) Coronary artery disease appears a decade earlier in India than other countries.

b) Males are affected more than females

c) Heavy smoking is an important contributing factor

d) Hypertension and Diabetes account for about 40 percent of all cases.

The above findings were based on a clinical study done in Chandigarh in Persons of age greater than 30.

The risk of coronary artery disease in Indians is 3-4 times higher than white Americans, 6 times higher than Chinese and 20 times higher than Japanese Enas EA, Grag A, Davidson NA[1] et al - coronary artery disease risk factors in first generation immigrant Asian Indians to the USA).

Indians are prone as a community to coronary artery disease at a much younger age. The disease pattern is severe and diffuse. Premature coronary artery disease is defined as coronary artery disease occurring before the age of 40 years. Indians are affected by coronary artery disease 5-10 years earlier than the other communities and also they show higher incidence of hospitalization, morbidity, and mortality than other ethnic groups. The above data is based on a study by EnasEA, Dhawan J, Petkar S et al.[2] coronary artery disease in Asia Cigarette smoking has been the single factor very strongly associated with CAD in the young adult[3], Perhaps of equal interest is the lack of a strong association of other traditional risk factors to CAD in the young. Hypertension and lack of exercise are both firmly established risk factors for CAD in general. Observations in approximately 80 clinical and epidemiological studies suggested that elevated tHcy is a risk factor for atherosclerotic vascular disease and for arterial and venous thromboembolism. The aim of the present study is to evaluate the role of Hyperhomocysteine as a predictor of MI in young South Indian patient.

REVIEW OF LITERATURE

Myocardial infarction generally occurs with the abrupt decrease in Coronary blood flow that follows a thrombotic occlusion of a coronary artery previously narrowed by atherosclerosis.

NARY ATHEROSCLEROSIS

The earliest lesions of atherosclerosis can be found in young children and infants in the form of fatty streak whereas the advanced lesion, the fibrous plaques, generally appears during early adulthood and progress with age[4].The abdominal aorta is involved earliest. The aorta is usually most heavily involved at or near the orifice of its branches (particularly at the level of coronary arteries)[4]. In coronary arteries, raised lesions are most prominent in the main stems, the highest incidence being a short distance beyond the ostia. Atherosclerosis is usually always found in the epicardial (extramural) portions of the vessels, while the intramural coronary arteries are spared, coronary atherosclerosis is often diffuse. The degree to which the lumen is narrowed varies, but once the process has commenced, all the intima of the extramural portions of the vessels are usually involved. Typical atheromatous fibrous plaques also develop in saphenous vein, which is used for aortocoronaryby pass graft.

HYPOTHESES OF ATHEROSCLEROSIS

The response to Injury Hypothesis

The endothelial lining cells are exposed to repeated or continuous insults. Dysfunctional endothelial cells at the susceptible sites in the arterial tree would lead to exposure of the subendothelial tissue to increased concentrations of plasma constituents. This triggers a sequence of events including monocytes and platelet adherence, migration, platelet aggregation and formation of microthrombi and release of secretory products. This causes proliferation of smooth muscle cells at these sites of injury. Monocytes become transformed to foam cells. Thus a well developed plaque is formed[5]

Monoclonal Hypothesis

This states that the intimal proliferative lesion results from the multiplication of single, individual smooth muscle cells, as do benign tumors. According to this hypothesis, the intimal smooth muscle cell that proliferate to form an atheroma are normally under feedback control by mitosis inhibitors formed by the smooth muscle cells in the contiguous media, and this feedback control system tends to fail with age as these controlling cells die and are not adequately replaced[6].

RISK FACTORS

A number of factors are present more frequently in individuals who develop atherosclerosis much earlier than in general population and these factors have been termed as risk factors.

The risk factor concept implies that a person with at least one risk factor is more likely to develop a clinical atherosclerotic event and is likely to do so earlier than a person with no risk factors. The presence of multiple risk factors further accelerates atherosclerosis.

Hypercholesterolemia, hypertension and cigarette smoking may be the most potent factors for atherosclerosis[7]. Risk factors also vary in terms of their potential reversibility with current techniques of preventive management.

The 27th Bethesda conferences, conducted by the American College of Cardiology placed the risk factors into four categories. Modification of which will lower the incidence of coronary artery disease.

These categories are as follows:

CATEGORY I

CATEGORY II

CATEGORY III

CATEGORY IV

CATEGORY I

- Basic research and human observational study indicate a clear causal relationship.
- Intervention data demonstrates the magnitude of the benefit and risk.
- Interventions are cost-effective and have been proved to lower coronary artery disease risk.

1. Cigarette Smoking
2. LDL Cholesterol
3. High fat / high cholesterol diet
4. Hypertension
5. Left Ventricular Hypertrophy

CATEGORY II

> Studies indicate a causal relationship.

> Intervention data for large scale trials are limited.

> Cost effectiveness not ascertained but likely to lower coronary artery disease

1. Diabetes Mellitus
2. Physical inactivity
3. HDL Cholesterol
4. Triglycerides, LDL cholesterol
5. Obesity
6. Post menopausal status

CATEGORY III

> Studies indicate association

> Interventions have not been tested adequately

> Might lower risk

1. Psychosocial factors
2. Lipoprotein (a)
3. Homocysteine

4. Oxidative stress

5. No alcohol consumption

CATEGORY IV

> Factors associated with coronary artery disease risk, but cannot be modified.

1. Age

2. Male gender

3. Low socio economic status

4. Family history of early onset of coronary artery disease

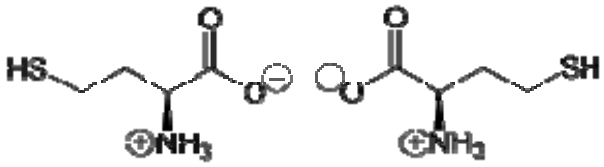
HOMOCYSTEINE

Homocysteine is a non-protein amino acid with the formula $HSCH_2CH_2CH(NH_2)CO_2H$. It is a homologue of the amino acid cysteine, differing by an additional methylene ($-CH_2-$) group. It is biosynthesized from methionine by the removal of its terminal C^{ε} methyl group. Homocysteine can be recycled into methionine or converted into cysteine with the aid of B-vitamins.

While detection of high levels of homocysteine has been linked to cardiovascular disease, lowering homocysteinelevels may not improve outcomes[8]

Structure

Homocysteine exists at neutral pH values as a zwitterion.



Betatine form of (*S*)-homocysteine (left) and (*R*)-homocysteine (right)

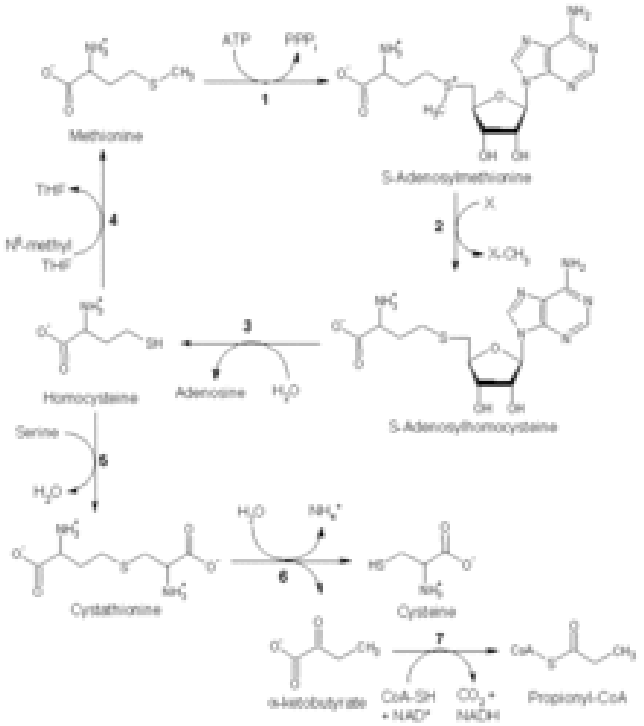
Biosynthesis and biochemical roles

Homocysteine is not obtained from the diet.[9] Instead, it is biosynthesized from methionine via a multi-step process. First, methionine receives an adenosine group from ATP, a reaction catalyzed by S-adenosyl-methionine synthetase, to give *S*-adenosyl methionine (SAM). SAM then transfers the methyl group to an acceptor molecule, (i.e., norepinephrine as an acceptor during epinephrine synthesis, DNA methyltransferase as an intermediate acceptor in the process of DNA methylation). The adenosine is then hydrolyzed to yield L-homocysteine. L-Homocysteine has two primary fates: conversion via tetrahydrofolate (THF) back into L-methionine or conversion to L-cysteine.[10]

Biosynthesis of cysteine

Mammals biosynthesize the amino acid cysteine via homocysteine. Cystathionine β-synthase catalyses the condensation of homocysteine and serine to give cystathionine. This reaction uses pyridoxine (vitamin B_6) as a cofactor. Cystathionine β-lyase then converts this

double amino acid to cysteine, ammonia, and α-ketobutyrate. Bacteria and plants rely on a different pathway to produce cysteine, relying on *O*-acetylserine.[11]



Two of homocysteine's main biochemical roles. (Homocysteine is seen in the left middle of the image.) It can be synthesized from methionine and then converted back to methionine via the SAM cycle or used to create cysteine and alpha-ketobuterate.

Methionine salvage

The methyl donor and cobalamin (vitamin B_{12})-related enzymes. More detail on these enzymes can be found in the Homocysteine can be recycled into methionine. This process uses N5-methyl tetrahydrofolate as article for Methionine synthase.

Other reactions of biochemical significance

Homocysteine can cyclize to give homocysteinethiolactone, a five-membered heterocycle. Because of this "self-looping" reaction, homocysteine-containing peptides tend to cleave themselves

Elevated homocysteine

Deficiencies of the vitamins folic acid (B_9), pyridoxine (B_6), or B_{12} (cobalamin) can lead to high homocysteine levels.[12]Supplementation with pyridoxine, folic acid, B_{12} or trimethylglycine (betaine) reduces the concentration of homocysteine in the bloodstream.[1314]Increased levels of homocysteine are linked to high concentrations of endothelial asymmetric dimethylarginine. Recent research suggests that intense, long duration exercise raises plasma homocysteine levels, perhaps by increasing the load on methionine metabolism.[15]Chronic consumption of alcohol may also result in increased plasma levels of homocysteine.[1617]

Elevations of homocysteine also occur in the rare hereditary disease homocystinuria and in the methylene-tetrahydrofolate-reductase polymorphism genetic traits. The latter is quite common (about 10% of the world population) and it is linked to an increased incidence of thrombosis and cardiovascular disease, which occurs more often in people with above minimal levels of homocysteine (about 6 μmol/L). These individuals require adequate dietary riboflavin in order for homocysteine levels to remain normal. Common levels in Western populations are 10 to 12 and levels of 20

μmol/L are found in populations with low B-vitamin intakes (e.g., New Delhi) or in the older elderly (e.g., Rotterdam, Framingham). Women have 10-15% less homocysteine during their reproductive decades than men, which may help explain the fact they suffer myocardial infarction (heart attacks) on average 10 to 15 years later than men. However, this phenomenon is more readily explained by higher levels of estrogen, which exerts a cardioprotective effect.

Blood reference ranges for homocysteine:

Sex	Age	Lower limit	Upper limit	Unit	Elevated	Therapeutic target
Female	12–19 years	3.3[11]	7.2[11]	µmol/L	> 10.4 µmol/L or > 140 µg/dl	< 6.3 µmol/L[12] or < 85 µg/dL[12]
		45[13]	100[13]	µg/dL		
	>60 years	4.9[11]	11.6[11]	µmol/L		
		66[13]	160[13]	µg/dL		
Male	12–19 years	4.3[11]	9.9[11]	µmol/L	> 11.4 µmol/L or > 150 µg/Dl	
		60[13]	130[13]	µg/dL		
	>60 years	5.9[11]	15.3[11]	µmol/L		
		80[13]	210[13]	µg/dL		

Cardiovascular risks and related medical studies

A high level of blood serum homocysteine "Homocystinemia" is a powerful risk factor for cardiovascular disease. However, one study which attempted to decrease the risk by lowering homocysteine was not fruitful.[18]This study was conducted on nearly 5000 Norwegian heart attack survivors who already had severe, late-stage heart disease. No study has yet been conducted in a preventive capacity on subjects who are in a relatively good state of health. However, Dr. Kilmer McCully has shown in several research studies that the development of arteriosclerosis requires elevated levels of homocysteine in the blood.

Studies reported in 2006 have shown that giving vitamins [folic acid, B_6 and B_{12}] to reduce homocysteine levels may not quickly offer benefit, however a significant 25% reduction in stroke was found in the HOPE-2 study [19] even in patients mostly with existing serious arterial decline although the overall death rate was not significantly changed by the intervention in the trial. Clearly, reducing homocysteine does not quickly repair existing structural damage of the artery architecture. However, the science is strongly supporting the biochemistry that homocysteine degrades and inhibits the formation of the three main structural components of the artery, collagen, elastin and the proteoglycans. Homocysteine permanently degrades cysteine disulfide bridges and lysine amino acid residues in proteins, gradually affecting function and structure. Simply put, homocysteine is a 'corrosive' of long-living proteins,

i.e., collagen or elastin, or life-long proteins, i.e., fibrillin. These long-term effects are difficult to establish in clinical trials focusing on groups with existing artery decline. The main role of reducing homocysteine is possibly in 'prevention' but studies in patients with pre-existing conditions found no significant benefit nor damage.[19, 20, 21] Hypotheses have been offered to address the failure of homocysteine-lowering therapies to reduce cardiovascular event frequency.[22] One suggestion is that folic acid may directly cause an increased build-up of arterial plaque, independent of its homocysteine-lowering effects. Alternatively, folic acid and vitamin B_{12} may cause an overall change in gene methylation levels in vascular cells, which may also promote plaque growth. Finally, altering methlyation activity in cells might increase methylation of l-arginine to asymmetric dimethylarginine which can increase the risk of vascular disease. Thus alternative homocysteine-lowering therapies may yet be developed which show greater effects on development and progression of cardiovascular disease.

The VITATOPS trial (results presented in May 2010 by the lead investigator, Dr Graeme J Hankey of Royal Perth Hospital, Australia at the European Stroke Conference 2010, in Barcelona, Spain) has concluded that B-vitamin supplements, within 2 years, do not seem to significantly reduce subsequent stroke, MI, or vascular death in patients with a history of recent stroke and ischemic attack, despite lowering of homocysteine levels.[23]

Alzheimer's disease and homocysteine

Studies demonstrate the connection between elevated levels of homocysteine (hyperhomocysteinaemia) and occurrence of Alzheimer's disease (AD) besides other cognitive impairments. Researchers suggest that B-group-vitamin supplementation (including folate) may possibly decrease chances to develop AD.[24, 25, 26]

Bone weakness and breaks

Elevated levels of homocysteine have been linked to increased fractures in elderly persons. The high level of homocysteine will auto-oxidize and react with reactive oxygen intermediates and damage endothelial cells and has a higher risk to form a thrombus.[27, 28] Homocysteine does not affect bone density. Instead, it appears that homocysteine affects collagen by interfering with the cross-linking between the collagen fibers and the tissues they reinforce. Whereas the HOPE-2 trial[20] showed a reduction in stroke incidence, in those with stroke there is a high rate of hip fractures in the affected side. A trial with 2 homocysteine-lowering vitamins (folate and B_{12}) in people with prior stroke, there was an 80% reduction in fractures, mainly hip, after 2 years. Interestingly, also here, bone density (and the number of falls) were identical in the vitamin and the placebo groups.[29]

Vitamin supplements counter the deleterious effects of homocysteine on collagen. As they inefficiently absorb B_{12} from food, elderly persons may

benefit from taking higher doses orally such as 100 mcg/day (found in some multivitamins) or by intramuscular injection

Life style factor which increase Homocysteine levels include

a. Diet - A low diet with deficiency of nutrients particularly B-6, B-12, and folic acid.
b. Smoking
c. Alcohol - high alcohol intake reduce the absorption of vitamins.
d. Lack of physical exercise and excessive stress.
e. Obesity.
f. Drugs such as Oral contraceptive pills, L-Dopa, Methotrexate, Nicotinic acid, Theophylline, etc.

Renal failure, hypothyroidism, psoriasis and malignancies.

HYPERHOMOCYSTENEMIA AND VASCULAR PROBLEMS:

Hyperhomocysteinemia or **hyperhomocysteinaemia** is a medical condition characterized by an abnormally large level of homocysteinein the blood.

As a consequence of the biochemical reactions in which homocysteine is involved, deficiencies of the vitamins pyridoxine (B_6), folic acid(B_9), or B_{12} can lead to high homocysteinelevels.[30] Supplementation with pyridoxine, folic acid, B_{12}, or trimethylglycine (betaine) reduces the concentration of homocysteine in the bloodstream[31]. Hyperhomocysteinemia is a disease which increases risk of other artery or vein diseases.

Hyperhomocysteinemia usually occurs in people with at least one defective gene, which affects the breakdown of homocysteine. There are two common gene defects in the population. The first gene codes for an enzyme. This gene is known as methylenetetrahydrofolatereductase or MTHFR. The second common defective gene is methioninesynthetase or MS.

Hyperhomocysteinemia is a high level of homocysteine in human blood. A high level of homocysteine makes a person more prone to endothelial injury, which leads to vascular inflammation, which in turn may lead to atherogenesis and thrombophilic situation which can result in ischemic injury.[32] Hyperhomocysteinemia is therefore a risk factor for coronary artery disease. Coronary artery disease is when an atherosclerosis leads to occlusion of the lumina of the coronary arteries. These arteries supply the heart with oxygenated blood.

People who are most likely to get hyperhomocysteinemia are with two defective genes and having a diet low in folate and vitamin B_6 and B_{12}. The effect upon these people is greater than the people with only one defective gene. Hyperhomocysteinemia is linked to an increased risk of blood clots, heart attacks and strokes. It can cause miscarriage, pre-eclampsia and other birth defects in pregnant women.

The best way to prevent hyperhomocysteinemia is to eat foods which contain B_6, B_{12}, and folate such as potato, greens, beans and fish. The only natural sources of B_{12} are from animal products.

However, it is difficult to show that reducing your homocysteine levels will improve your health. In a study designed to see if lowering homocysteine will reduce heart ("vascular") and kidney problems, the results were just the opposite.[33] Patients with diabetes and known kidney disease ("diabetic nephropathy") were given commonly used doses of B vitamins: folic acid (2.5 mg/d), vitamin B_6 (25 mg/d), and vitamin B_{12} (1 mg/d). Diabetics usually have elevated homocysteine levels and a higher risk for heart attacks and strokes, so they make an ideal population to study homocysteine's effects on human health. Taking B vitamins did reduce homocysteine, as expected, but unfortunately doubled the patient's risk of serious vascular complications (heart attack, stroke, death) and worsened their kidney function.

MANAGEMENT OF HYPERHOMOCYSTEINEMIA

Genetic counseling in case of congenital disorders.

Once elevated Homocysteine has been diagnosed, the next step is a simple examination of diet. A healthy balanced diet containing at least five servings of fruits and vegetables a day will help to lower the Homocysteine level and keep within the ideal range.

Folic acid and vitamins B-6 and B-12 have been shown to have the greatest effect at breaking down harmful Homocysteine from within the body[34].

Foods rich in folic acid include:

a. Green leafy vegetables such as spinach, cabbage, etc.

b. Citrus fruits particularly orange and grape fruits.

c. Pulses such as black-eyed beans and chick peas.

d. Whole grain cereals.

Maintaining low plasma Homocysteine is also easily achieved through vitamin supplementation. Patients with elevated Homocysteine (> 14 **u** mol/L with no other risk factors, or > 11 **u** mol/L for patients with more than 2 risk factors) can be treated effectively with multivitamins containing 400 **u** g of folic acid supplemented with an additional 400 to 1000 **u** g of folic acid. After 6 to 8 weeks of vitamin supplementation, Homocysteine should be repeated. Continued elevated Homocysteine should be treated with increased doses of supplemental folic acid up to 5mg/d.[35] For many people, the daily intake of 500 **u** g TMG (Trimethyl glycine), 800 **u** g of folic acid, 1000 **u** g of vitamin B_{12} and 250 mg of choline, 250 mg of inositol, 30mg of zinc and 100mg of vitamin B6 will keep Homocysteine levels in the normal range..

CORONARY ARTERY DISEASE AND OTHER RISK FACTORS

The major conventional coronary risk factors are smoking, hypertension, sedentary life style, high LDL cholesterol heredofamilial factors and diabetes mellitus. The newer coronary risk factors are lipoprotein (a), psychosocial stress, etc.

A cohort, prospective study on coronary artery disease in the young; Here do familial or faulty life style or both was done in India. The study comprised 70 young (< 40 years) CAD patients. There were 56 males and 14 females, mostly belonging to 36-40 age groups. More than half of the patients were from

low socioeconomic group. It was found that 61.42% were chronic smokers and all of them were males. 18.8% gave history of CAD in their first degree relatives. Hypertension was detected in 51.42% cases, obesity in 35.71% cases and underweight in 14% subjects. Hypercholesterolemia was observed in 41.66% cases. Low HDL was the next most common dylipidemia. 7.14%cases were diabetic. Most ominous combination was smoking, low socioeconomic status, increased waist-hip ratio (WHR), dyslipidemia and hypertension. The study reported that both heredofamilial as well as faulty life style risk factors contribute to the development of CAD in young people.[36]

A case control study to assess the association of lipoprotein (a) [(Lp (a)] with CAD was conducted in Andhra Pradesh. 151 consecutive patients with clinical and angiographic evidence of CAD and 49 healthy controls were drawn for the study. Triglycerides, VLDL-C, total-cholesterol/high density cholesterol (HDL-C) ratio, low density cholesterol (LDL-C)/HDL-C ratio, and Lp (a) were found to be higher in pati In female sex and in those with family h/o CAD, higher total and LDL cholesterol levels were observed to be associated with higher Lp (a) levels. Significant difference in Lp(a) levels were observed between normal coronaries Vs single and triple vessel disease and also between single vs double and triple vessel disease. The findings suggested that a cut-off level of 25mg/dl for the determination of risk of CAD.[37]

A case-control study to determine the relationship between plasma lipoprotein (a) levels and their phenotypes in a group of South Indian patients

with coronary artery disease was done in CMC, Vellore. A total of 104 patients with angiographically proven coronary artery disease were compared with 104 age and sex-matched controls with no risk factors. Lipoprotein (a) levels were measured by ELISA method and its phenotyping was done by SDS agarose gel electrophoresis. Plasma lipoprotein (a) levels were significantly elevated in patients with coronary artery disease as compared to controls Lipoprotein (a) phenotyping showed that low-molecular weight isoforms were found only in 19.2% of the patients with coronary artery disease and their plasma lipoprotein (a) levels were significantly elevated compared to ents than controls.

CAD patients with higher molecular weight isoforms. It was concluded that plasma lipoprotein (a) levels are significantly elevated in CAD patients and the commoner phenotype in a south Indian population is the larger apolipoprotein (a) in which the lipoprotein (a) levels are lower.[38]

A case-control study on the comparative account of serum lipids, lipoproteins and Apolipoprotein-B in patients of coronary artery disease was done in Punjab. Serum total lipids (cholesterol and triglycerides), lipoproteins (VLDL, LDL and HDL) and Apolipoprotein-B levels of normal healthy individuals (n=25) and coronary artery disease patients (n=25) were estimated.

The objective of the present study was to ascertain the role of apo-B in causation and inheritance of CAD. It was observed that on an average serum total cholesterol and triglyceride more than 200mg/dl bring the individuals to a risk of CAD irrespective of the age. CAD patients achieved this value at an

early age (35-45 years). Similarly VLDL and LDL levels were found to be raised and HDL levels were found to be lower in CAD patients compared to age-matched controls. Serum apo-B levels were significantly raised in CAD patients and patient with positive family history of CAD. A positive coefficient of correlation was between serum apo-B and LDL levels suggesting that more the number of Apo-B particles, more will be the synthesis of atherogenic particles (LDL). The study concluded that elevated apo-B levels turn out to be a genetic factor responsible for causative factor of coronary artery disease.[39]

A prospective, case-cross over study was done in Israel on negative emotions, anger and sudden changes in posture as independent triggers for ischemic stroke. 200 consecutive patients hospitalized with an ischemic stroke or a transient ischemic attack were interviewed one to four days after stroke onset using a validated questionnaire. Reported exposure to potential triggers, such as negative and positive emotions, anger, sudden postural changes in response to a starting event, strenuous physical exertion, heavy eating and sudden temperature changes during a two-hour period prior to stroke onset were compared with the same period during the preceding day and to average exposures in the previous year. Anger and other negative emotions were rated on a seven-point scale and were said to be present if the score was 5 or higher. During 2 hours before stroke 76 patients experienced at least one of the study triggers. Thus the study demonstrated that apart from the conventional risk

factors, there are factors that may trigger the premature onset of stroke and this is an important area of potential invention.

AIM OF STUDY

The aim of the present study is

> to estimate the plasma Homocysteine levels in young patients presenting with myocardial infarction

> to assess the possible role of Homocysteine as non-traditional risk factor in these patients

> to study the association of Homocysteine with traditional risk factors.

STUDY PLAN

The present study was planned to estimate the plasma Homocysteine in young patients presenting with MI, and to associate with Homocysteine with non-traditional risk factor and finally to assess the role of Homocysteine as a new predictor of MI in young patients with signs of myocardial infarction, admitted in the Tirunelveli medical college as outlined below :

> Selection of patients based on inclusion criteria.

> Study of demographic and disease presenting characteristics and correlation with MI.

> Collect patient family history and family medication history and to associate with the diagnosed indications.

> Estimation of Homocysteine levels in MI patients and compare it with healthy subjects.

> To study the association of Homocysteine and disease.

To explore the possible influence of Homocysteine on traditional risk factors.

MATERIALS AND METHODS

Setting

Department of Medicine and Department of Cardiology, Tirunelveli medical collegehospital,Tirunelveli.

Study design

This is a cross-sectional, Case control study carried out in young MI patients.

Study protocol

Patients admitted in Intensive Care Unit of Tirunelveli medical college hospital for MI were enrolled in the study. Patients were interviewed and a designed questionnaire was used to collect demographic details, previous history of MI, presenting symptoms, the earlier medication and food supplementation used.

Ethical Committee Approval

The study protocol was duly approved by the Ethics Committee of Tirunelveli medical college hospital.

Collaborating Department

1. Department of Medicine and Department of Cardiology,Tirunelveli medical college hospital.
2. Path care lab Tirunelveli.

Study Criteria

Selection of patients: Inclusion Criteria:

The following categories of patients from cardiology department are included in the study.

> Inpatients admitted for myocardial infarction.

> Patients with age 45 years and below only.

Exclusion criteria:

> Patients with age above 45 years.

> Patients suffering from diseases such as renal failure, hypothyroidism, psoriasis, any malignancies and psychiatric disorders.

> Patients taking drugs such as Methotrexate, oral contraceptive pills, L-dopa, nicotinic acid and Theophylline.

> Patients taking folic acid or any vitamin supplement.

SOURCE OF DATA

Department of Cardiology, Tirunelveli medical college hospital. The data were obtained from

> Patient interview

> Treatment chart

> Prescriptions

METHOD OF COLLECTION OF DATA

This is a cross-sectional, case control study carried out from Dec 2010 to Dec 2011, to analyze the Homocysteine levels in young MI patients and its association with other risk factors.

Proforma was prepared as an aid for collecting the demographic data. Model of Proforma is given in Appendix.

ESTIMATION OF HOMOCYSTEINE

Estimation of Homocysteine was performed by automated immuno assay analyzer - IMX ABBOTT System (USA).

DATA ANALYSIS:

Graph-Pad software was used for statistical analysis.

OBSERVATION AND RESULTS

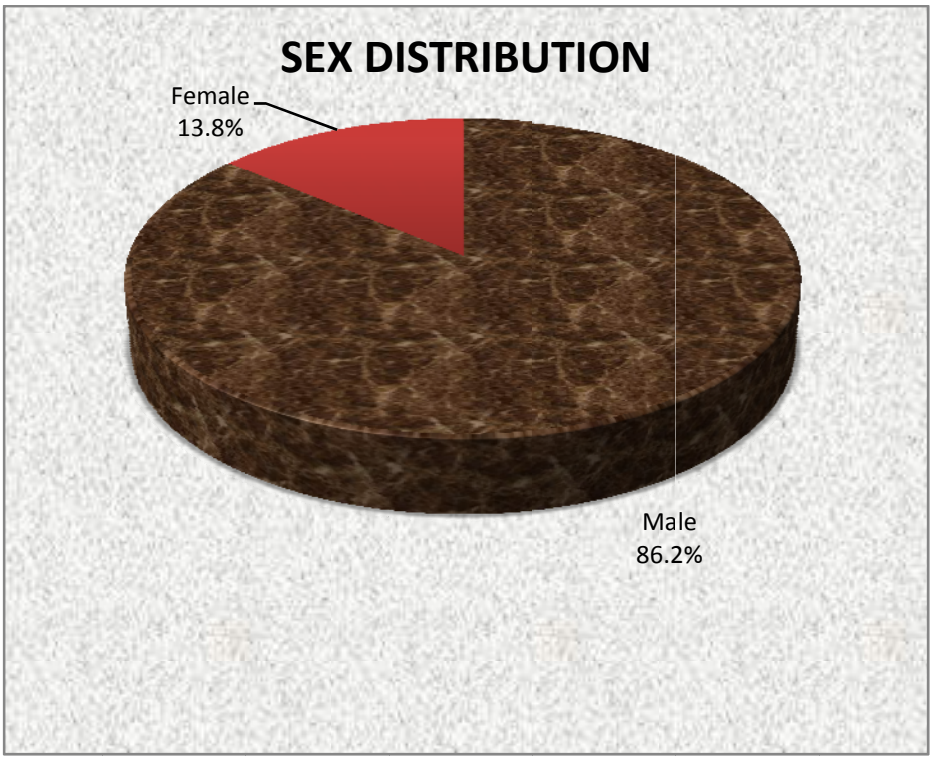
1. SEX DISTRIBUTION IN STUDY GROUP

Sex	Case		Control	
	N	*%*	*N*	*%*
Male	36	90.0	20	80.0
Female	4	10.0	5	20.0
Total	40	100.0	25	100.0

$\chi^2 = 1.290\ df = 1$ $p = 0.256$

In our study group males (86.2%) occupy the major portion compared to females and there is no significance regarding sex on comparing cases and controls(p=0.256).

Fig 1

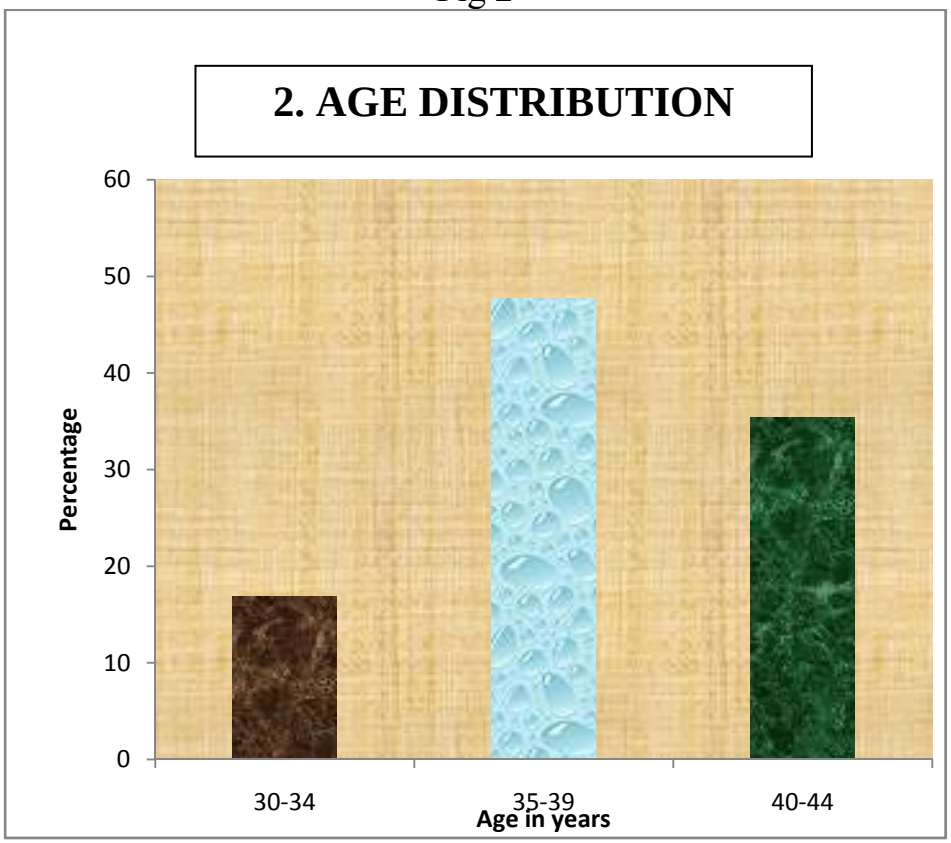


2. AGE DISTRIBUTION

Age in years	Case		Control	
	N	*%*	*N*	*%*
30-34	7	17.5	4	16.0
35-39	19	47.5	12	48.0
40-44	14	35.0	9	36.0
Total	40	100.0	25	100.0

$\chi^2 = 0.026$ $f = 2$ $p = 0.967$

Fig 2



In our study group major portion of patients are in age group 35-39 yrs(47.5%) and there is no significance regarding age on comparing cases and control(p=0.967).

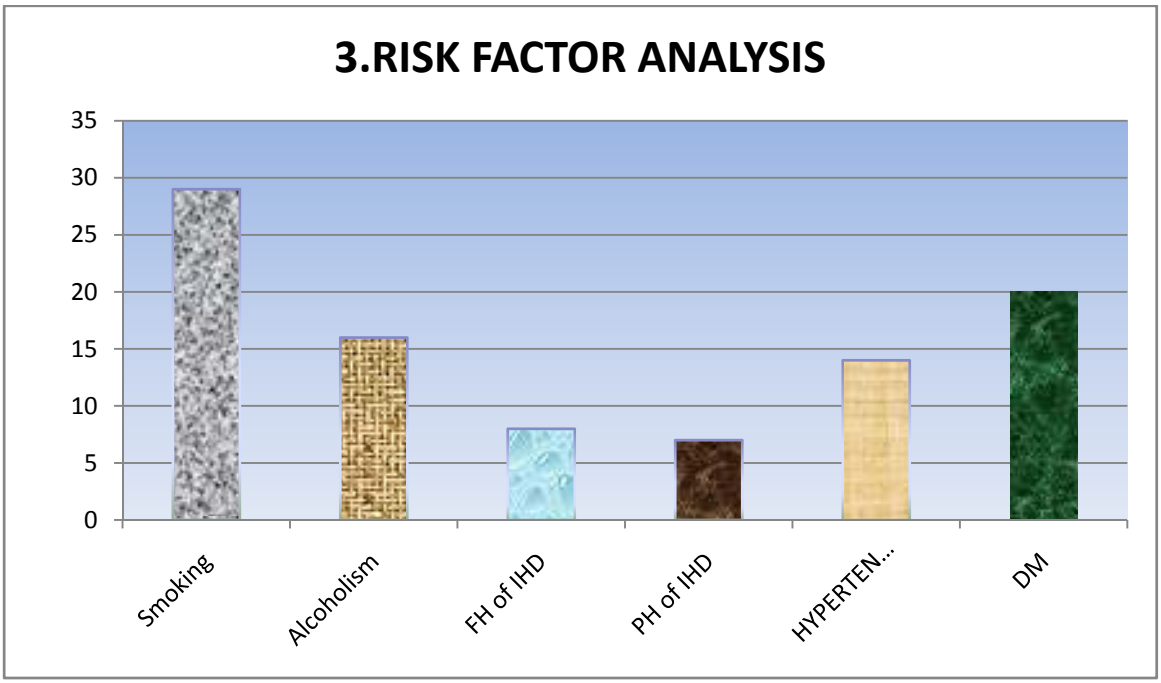


Fig 3

In our study group smokers and diabetics occupy the major proportion.

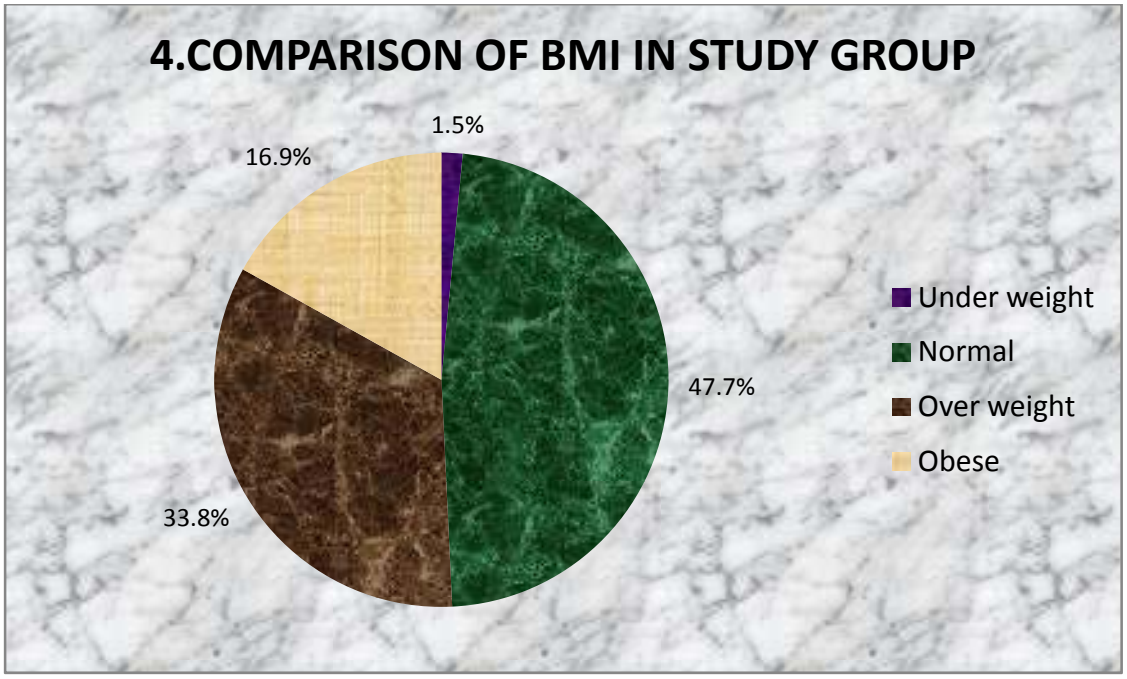


Fig 4

BMI	case		Control	
	N	*%*	*N*	*%*
Under weight	1	2.5	0	.0
Normal	23	57.5	8	32.0
Over weight	11	27.5	11	44.0
Obese	5	12.5	6	24.0
Total	40	100.0	25	100.0

$\chi^2 = 5.162$ *df* $= 3$ $p = 0.160$

In our study group major portion of our patients are in normal BMI (57.5%)and there is no significance on comparing cases and controls(p=0.160).

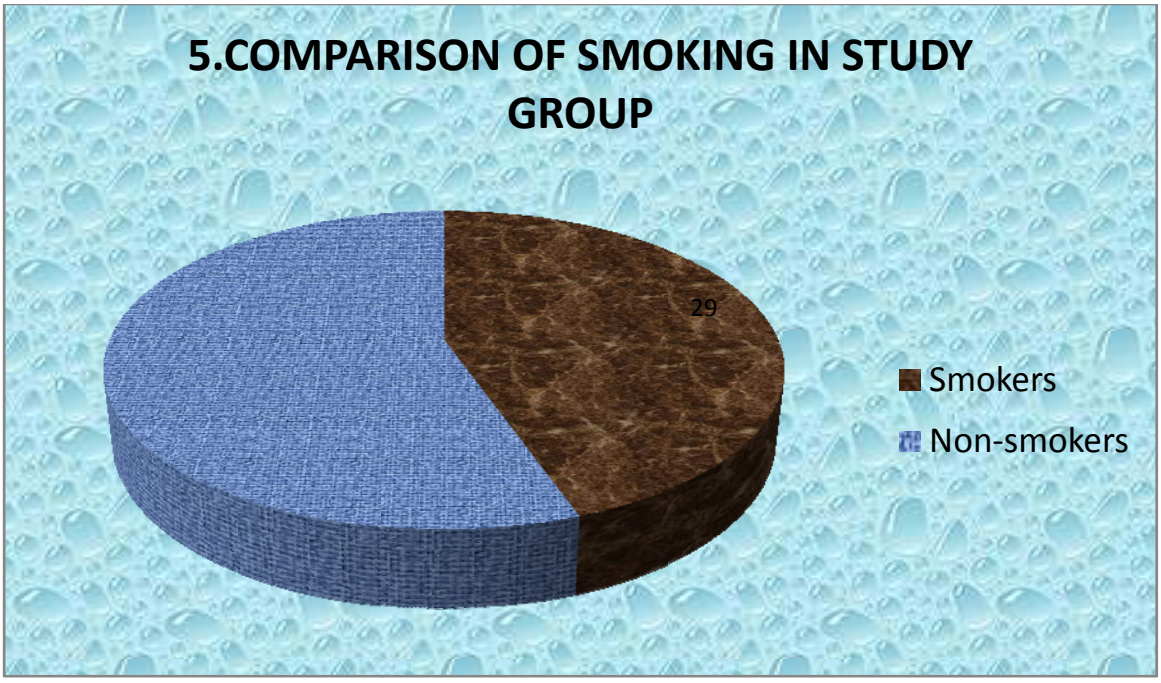


Fig 5

Smoking	case		Control	
	N	*%*	*N*	*%*
Yes	21	52.5	8	32.0
No	19	47.5	17	68.0
Total	40	100.0	25	100.0

$\chi^2 = 2.616$ $df = 1$ $p = 0.106$

in our study group major portion of our cases are smokers(52.5%) and there is no significance on comparing cases and controls(p=0.106).

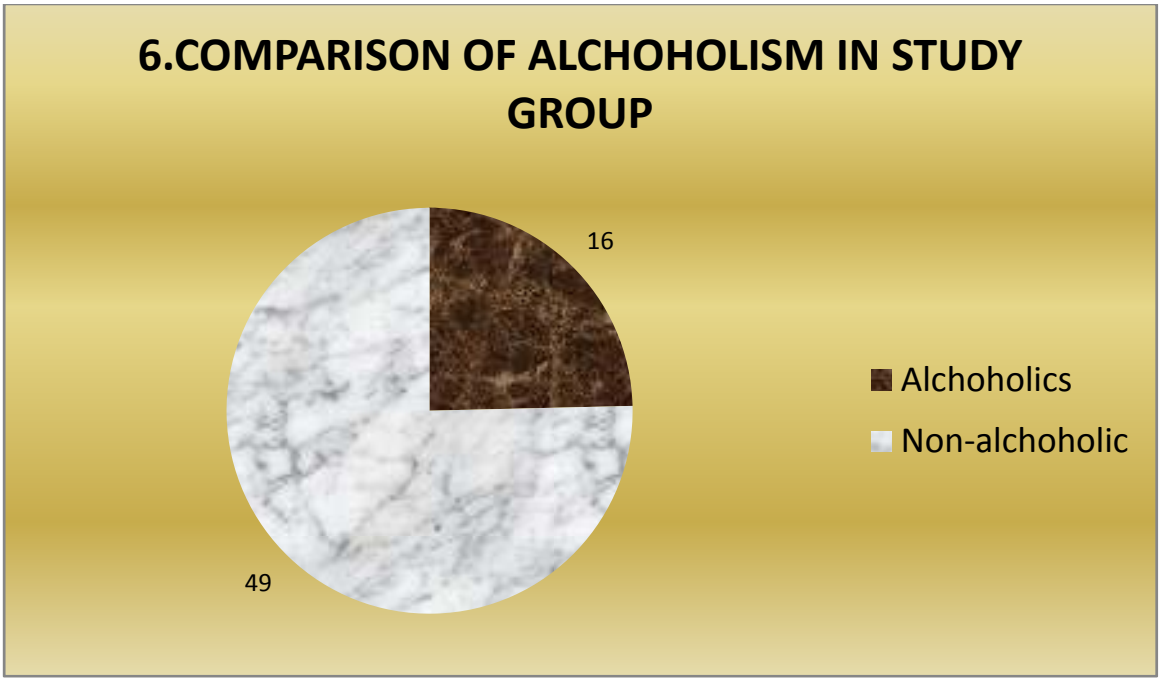


Fig. 6

Alcoholism	case		Control	
	N	*%*	*N*	*%*
Yes	13	32.5	3	12.0
No	27	67.5	22	88.0
Total	40	100.0	25	100.0

$\chi^2 = 3.484$ *df* $= 1$ $p = 0.062$

In our study group major portion of our cases alchoholics(32.5%). There is no significance on comparing the cases and control(p=0.062).

7. COMPARISON OF FAMILY HISTORY OF MI IN STUDY GROUP

FH	case		Control	
	N	*%*	*N*	*%*
Yes	7	17.5	1	4.0
No	33	82.5	24	96.0
Total	40	100.0	25	100.0

$\chi^2 = 2.598$ *df =1* *p=0.107*

In our study group there is no significant association on comparing the family history of MI between cases and controls.

8. **COMPARISON OF PAST HISTORY OF MI IN STUDY GROUP**

PH	case		Control	
	N	*%*	*N*	*%*
Yes	7	17.5	0	.0
No	33	82.5	25	100.0
Total	40	100.0	25	100.0

$\chi^2 = 4.903$ *df =1* *p=0.027*

In our study group there is no significant association on comparing the past history of MI between cases and controls.

9. COMPARISON OF HYPERTENSION IN STUDY GROUP

HT	case		Control	
	N	*%*	*N*	*%*
Yes	12	30.0	2	8.0
No	28	70.0	23	92.0
Total	40	100.0	25	100.0

$\chi^2 = 4.406$ *df =1* *p=0.036*

In our study group there is significance on comparing hypertension between cases and controls(p=0.036).

10. COMPARISON OF DIABETES IN STUDY GROUP

DM	case		Control	
	N	*%*	*N*	*%*
Yes	15	37.5	5	20.0
No	25	62.5	20	80.0
Total	40	100.0	25	100.0

χ^2 *=2.212 df =1* *p=0137*

In our study group there is no significance on comparing diabetes between cases and controls.

11.COMPARISON OF TOTAL CHOLESTEROL IN STUDY GROUP

TC	case		Control	
	N	*%*	*N*	*%*
>200	4	10.0	0	.0
<200	36	90.0	25	100.0
Total	40	100.0	25	100.0

χ^2 *=2.664 df =1 p=0.103*

12.COMPARISON OF LDL IN STUDY GROUP

LDL	case		Control	
	N	*%*	*N*	*%*
>100	4	10.0	0	.0
<100	36	90.0	25	100.0
Total	40	100.0	25	100.0

χ^2 *=2.664 df =1 p=0103*

In our study group there is no significance on comparing Total cholesterol and LDL between cases and controls.

13.COMPARISON OF HDL IN STUDY GROUP

HDL	case		Control	
	N	*%*	*N*	*%*
Abnormal	38	95.0	22	88.0
Normal	2	5.0	3	12.0
Total	40	100.0	25	100.0

χ^2 *=1.062 df =1* *p=0.303*

In our study group there is no significance on comparing HDL between cases and controls.

14. COMPARISON OF TGL IN STUDY GROUP

TGL	case		Control	
	N	*%*	*N*	*%*
>150	15	37.5	2	8.0
<150	25	62.5	23	92.0
Total	40	100.0	25	100.0

χ^2 *=6.932 df =1 p=0.008*

In our study group there is significance on comparing TGL between cases and controls(p=0.008)

15.COMPARISON OF HOMOCYSTEINE IN STUDY GROUP

Homocysteine	Case		Control	
	N	*%*	*N*	*%*
<10	9	22.5	16	64.0
10-15	15	37.5	9	36.0
15-20	13	32.5	0	.0
>20	3	7.5	0	.0
Total	40	100.0	25	100.0

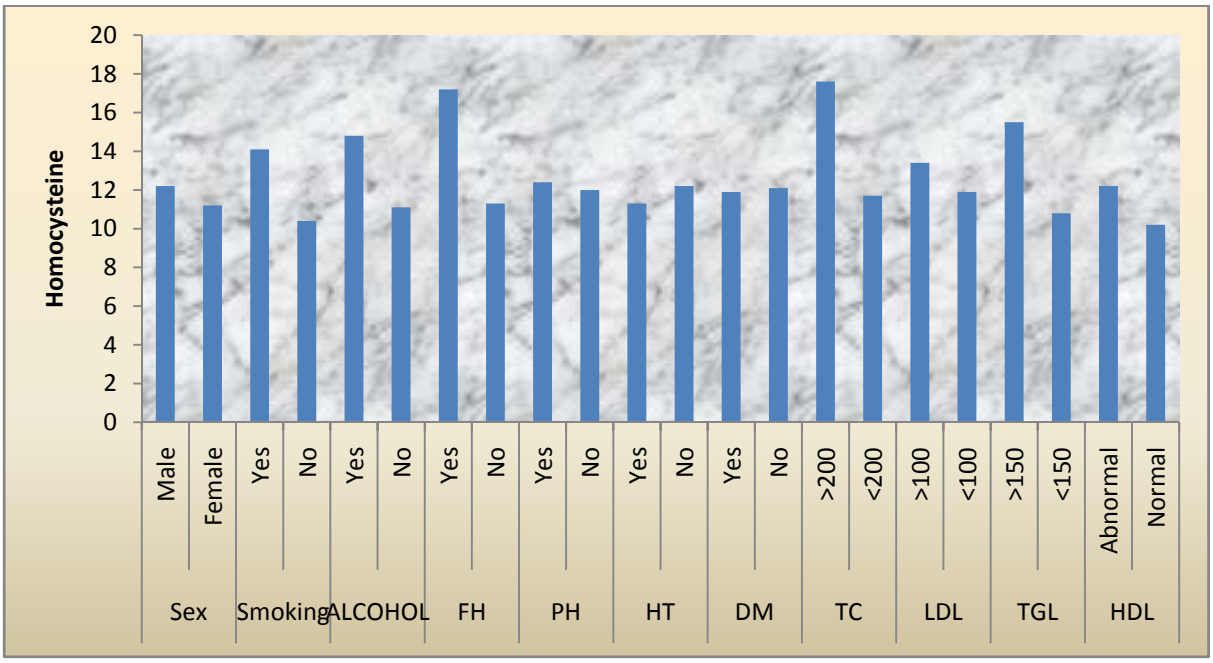
χ^2 =16.898 df =3 p=0.001

In our study group major portion of cases having homocysteine more than 15mc mol(40%). On comparing the homocysteine between cases and control there is significant association(p=0.001).

16.COMPARISON OF HOMOCYSTEINE WITH OTHER RISK FACTORS

		N	Homocysteine		t	P
			Mean	Sd		
Sex	Male	56	12.2	6.7		
	Female	9	11.2	4.5	0.409	0.684
Smoking	Yes	29	14.1	8.3	2.376	0.021
	No	36	10.4	3.8		
ALCOHOL	Yes	16	14.8	4.7	2.007	0.049
	No	49	11.1	6.7		
FH	Yes	8	17.2	14.0	2.516	0.014
	No	57	11.3	4.3		
PH	Yes	7	12.4	4.6	0.152	0.880
	No	58	12.0	6.7		
HT	Yes	14	11.3	3.9	-0.446	0.657
	No	51	12.2	7.0		
DM	Yes	20	11.9	4.1	-0.087	0.931
	No	45	12.1	7.3		
TC	>200	4	17.6	4.1	1.829	0.072
	<200	61	11.7	6.4		
LDL	>100	4	13.4	5.0		
	<100	61	11.9	6.5	0.447	0.656
TGL	>150	17	15.5	9.5		
	<150	48	10.8	4.5	2.734	0.008
HDL	Abnormal	60	12.2	6.6		
	Normal	5	10.2	3.9	0.662	0.510

In our study on comparing the elevated homocysteine with other risk factors it shows significant relation with smokers(p=0.021), Family history of MI(p=0.014), Alchoholics(p=0.049) and elevated TGL levels(p=0.008)



DISCUSSION

Cardiovascular disease is a major global health problem reaching epidemic proportions in the Indian sub-continent accounting for 78% of all deaths. Higher risk of cardiovascular disease have been reported among south Asians regardless of whether they live overseas or in their native lands. Presence of conventional risk factors such as Smoking, diabetes mellitus, Hypertension, dyslipidemia are clearly associated with coronary artery disese. Prevalence of risk factors is higher and rapidly raising in Indian population. This predisposition to accelerated atherosclerosis seems to have genetic predisposition but is being enhanced by changing lifestyle, dietary and cultural preferences, and suboptimal application of health care. Even though the environmental and behavioral responses to urbanization and westernization appear to be consistent across cultures, the genetically determined metabolic response to these environmental changes and resulting CAD risk profiles may vary in different ethnic groups.[42] The extent to which these variations in different ethnic groups are due to genetic or environmental factors remains unclear.[43]

According to the Indian studies, the prevalence rate of CAD was 9.7% in Delhi, 3.5% in Rajasthan and 11% in urban population of South India.[44]

Among all the ethnic groups studied, the standard mortality ratio for men born in the Indian subcontinent and aged between 20 and 69 years was by far the highest.[45]

In the analysis by Balarajan, the excess mortality seen among immigrants from the subcontinent steadily increased with decreasing age. Men in the age group 20-29 and 30-39 years had SMRs of 313 and 210, respectively. The rate of first MI was five times higher among Indian men compared to the Europeans in the study.[46]

Further, the mean age at first MI was about 5 years lower for the Indian men (50.2 v 55.5 years). More South Asian patients (38% v. 22%) were found unsuitable for surgery because of extensive disease. CAD tends to occur earlier in life among people of Indian descent.[47]

In addition to the conventional risk factors such as smoking, HT, DM, hypercholesterolemia, obesity and reduced physical activity Hyperhomocysteinemia was found to be independent risk factor of CAD.

Identification of new markers such as homocysteine associated with an increased risk of CAD may provide a better insight into the pathology of coronary atherosclerosis and facilitate the development of preventive and therapeutic measures.

Normal values of plasma total homocysteine are between 5 and 15 u mol/L. In our study, the mean plasma homocysteine in healthy subjects was 14.0 ± 7.4 and in CAD patients, the total plasma homocysteine was 8.8 ± 2.1 (P = 0.001) indicating that homocysteine level is an independent marker of CAD. A two fold increase in likelihood of MI among persons with a total Hcy

concentration more than or equal to 15 u mol/L has been noted in an United States study.[49]

An Indian study which gave a mean value of 22.81 ± 13.9 in cases and 7.77 ± 7.3 in controls showed a significant difference of homocysteine values between the two groups and hence showed increased homocysteine levels among patients with coronary artery disease.[50]

However, another Indian study with a mean homocysteine value of four cases as 18.30 ± 10.08 u mol/L and control 18.04 ± 10.65 reported a negative association between homocysteine with MI.[51]

Homocysteine and Smoking:

CAD patients who were smokers had a mean Hcy level of 14.1 ± 8.0 as compared to 10.4 ± 3.8 (p=0.021) in CAD patients who are non-smokers. This higher level of Hcy may be due to the influence of smoking in homocysteine metabolism. Among secondary causes of hyperhomocysteinemia, smoking plays an important role by interfering with the synthesis of pyridoxal phosphate, which is important for the conversion of homocysteine to cystathione by the enzyme cystathione b synthase.[52]

Homocysteine and alcoholism:

Alcoholic intake interferes the absorption of vitamins and interferes with the metabolism of homocysteine, leading to hyperhomocysteinemia. In our study mean Hcy of alcoholics was found to be 14.8 ± 4.7 as compared to 11.1 ± 6.7(p=0.049) in non-alcoholics.

Homocysteine and Hypertension:

A number of studies have shown a correlation between hyperhomocysteinemia and HT. However, a contradictory report was obtained from a western study with a mean value of tHcy 10.4 ± 53 in hypertensives.[53]

In my study also it shows that no significant co-relation between hyperhomocysteinemia and hypertensives(p=0.657). So further studies have to be conducted to establish relationship between hyperhomocysteinemia and hypertension.

Homocysteine and Diabetes Mellitus:

Varied results were observed among diabetes as well. **Munshi et al.** demonstrated elevated homocysteine levels in non-insulin dependent diabetes subjects compared to age matched controls[54] and **Hoffman et al,** reported that hyperhomocysteinemia is more common in type 1 diabetic patients with nephropathy.[55] **Deepa et al.** did not reveal any significant increase in homocysteine level in diabetes with CAD.[56] In our study population, there was a small difference in Hcy levels among diabetes and non diabetes patients (11.9 and 12.1)

Homocystiene and hypercholesterolaemia

In the **Hordaland study** in Norway, increasing plasma levels of cholesterol, triglycerides and smoking were associated with increasing levels of homocysteine. In our study, only 4 patients had hypercholesterolemia and there is no significant co-relation between hyperhomocysteine and

hypercholesterolemia(p=0.072). Lowered Hcy may be due to change in life style adopted by patients as part of treatment for high cholesterol. In addition majority of our patients were manual workers, not having sedentary life style pattern.

But in our study there is significant co-relation between hypertriglyceridemia and hyperhomocysteinemia(p=0.008). So further studies should be done to evaluate to co-relation between dyslipedemia and hyperhomocysteinemia.

Homocysteine and Obesity:

In our study, 1.5% were underweight with Hcy value of 12.2, 47.7% normal body weight with Hcy value (12.2 ±8.6) and 33.8% over weight patients with (11.3 ± 3.5)Hcy, 16.9% obese patients having Hcy value (12.8 ± 4.2). Elevated levels of Hcy in under weight subjects could be due to malnutrition and its impaired effect on Hcy metabolism.

Homocysteine and family history of CAD:

It is also interesting to note that Hcy levels were higher (17.2) in patients with a family history of MI in comparison of patients without such risk factor (Mean Hcy 11.3).

Homocysteine and previous history of CAD:

Many studies have shown that patients with MI had significantly elevated tHcy levels compared to patients without such history as well as to controls.[59] However, in our study it was found that tHcy of patients with previous history

was lower that of patients with previous history of MI (12.4 Vs 12).

This may be due to the effects of administration of lipid lowering agents following the previous MI events. It has been reported in a study that lower levels of tHcy were observed amongst patients taking lipid-lowering agents.[57]

Homocysteine and gender:

In this study, no significant difference was found in the total homocysteine levels of men and women (p=0.256).

The study matched with an Indian study where large number of women had high total homocysteine and were deficit in vitamin B12.[58]

Hyperhomocysteinemia as a risk factor for MI may have profound public health implications because total homocysteine can be lowered inexpensively and easily by modification of dietary patterns and vitamin supplements. But a Western study had shown that vitamins did not reduce the risk of major cardiovascular events in patients with vascular disease.[59]

The results of ongoing clinical trials to determine the effect of multivitamin therapy on CAD are awaited. If casually related, our findings suggest that young people with high risk of MI may particularly benefit from interventions that lower total homocysteine concentrations.

CONCLUSION

At the threshold of this millennium, CAD is looming large as the new epidemic afflicting Indians at a relatively younger age. Of the 40 MI patients, 24(60%) were having desirable plasma homocysteine levels (<15umol/L), 16(24%) were having undesirable homocysteine levels (>15umol/L).

In the study, homocysteine was identified as a nontraditional risk factor for MI. Among the study population of MI significant proportion of patients were found to have higher homocysteine than control. In the study, the mean plasma homocysteine level in healthy subjects was 8.8 umol/l and in CAD patients the total plasma homocysteine was (14+7.4umol/l)(P=0.001) indicating that homocysteine level is an independent marker of CAD.

It was also found that homocysteine levels were higher in patients who had additional conventional risk factors when compared to patients without such risk factors except hypercholesterolemia.

However, as the study was limited to a small population due to financial constraint, analysis of a larger group would definitely give an insight into the various causative factors leading to hyperhomocysteinemia and its role in CAD among the young South Indian population.

As such Asian Indians are genetically predisposed to CAD. Hence it is mandatory to determine homocysteine levels in people after thirties so that preventive measures such as vitamin supplementation and life style modifications could be undertaken and thus reduce the incidence and mortality due to CAD.

PROFORMA

Name of the patient : **Age :**

Marital Status : **Body Weight (in kg) :**

BMI : **Food habits : Veg:**

If non-veg, how frequently? **Sex:**

Height (in cms.): **Non-veg:**

SYMPTOMS:

- **Giddiness**
- **Palpitation**
- **Chest discomfort**
- **Headache**

Habits : *a. Smoker*

If yes, how long? :

Number of cigarettes/day :

If discontinued, whether after MI:

b. Alcoholic

If yes, how long? :

Frequency (mild/moderate) :

If discontinued, whether after MI:

Concomitant disease:

Kidney failure:

Psoriasis:

Hypothyroidism:

Diabetes Mellitus:

Hypertension:

CAD:

FAMILY HISTORY:

Diabetes mellitus :

CAD :

HYPERTENSION :

CKD :

Drug history

a.Oral contraceptive pills:

b.L-Dopa:

c.Methotrexate:

d.Nicotinic acid:

e.Theophilline:

f. Exposure to nitrous oxide:

Any folic acid / vitamin being taken:

General examination:

g.Pallor

h.Jaundice

i. Pedal edema

j. External markers of hyperlipidemia

k.PR:

l. BP:

m.Systemic examination: CVS :

n. RS :

o. ABDOMEN:

CNS :

Diagnosis:

Investigation:

Haemoglobin%:

ESR:

Blood sugar:

Serum creatinine:

Serum urea:

Lipid profile:

i. Total Cholesterol:

ii. Triglycerides:

iii. HDL:

iv. VLDL:

v. LDL:

ECG

ECHO:

PLASMA HOMOCYSTIENE:

MASTER CHART

Cases

S. No.	Age	Sex	Smoking	Alcohol	FH	PH	HT	DM	BMI	TC	HDL	LDL	TGL	HOMOC
1	33	M	N	Y	N	N	Y	N	18.1	149	42	103	160	12.2
2	32	M	Y	Y	N	N	Y	N	23.6	153	43	85	182	13.1
3	35	M	Y	N	N	N	N	N	22.5	165	44	74	175	11
4	40	M	Y	Y	N	N	N	N	23.5	173	39	95	143	16.2
5	36	M	N	N	Y	N	N	Y	24.4	185	38	75	180	12
6	38	M	Y	Y	N	N	N	N	20.6	205	40	90	170	18.2
7	40	M	N	N	N	N	N	N	21.8	172	38	83	191	16.3
8	41	M	Y	N	N	N	N	Y	28.8	185	40	75	180	15.5
9	42	F	N	N	Y	Y	N	Y	22.7	152	42	86	209	10
10	43	M	N	N	N	N	Y	N	19.8	149	43	96	172	11
11	39	M	Y	N	Y	N	Y	Y	18.8	163	40	69	123	12
12	38	M	Y	N	Y	N	N	N	20.8	185	39	90	205	50.4
13	39	M	N	Y	N	N	N	N	28.7	201	45	70	125	12
14	35	M	Y	N	N	N	N	N	29.1	192	44	65	133	11
15	43	M	Y	Y	N	Y	N	Y	25.6	230	41	103	145	18.3
16	44	M	N	N	N	Y	Y	Y	20.6	171	39	64	130	13
17	37	M	Y	N	N	N	Y	Y	21.6	163	40	72	141	14
18	40	F	N	N	N	N	N	N	22.8	190	42	110	151	16.2
19	41	M	Y	Y	N	N	N	Y	23.5	182	40	72	162	17
20	43	M	N	N	N	N	N	N	24.8	153	39	106	110	7
21	38	F	N	N	N	N	N	Y	19.6	149	38	98	120	6
22	40	M	Y	N	N	N	Y	Y	19.4	142	40	68	131	15.8

23	39	M	N	Y	N	N	N	N	21.9	139	41	75	133	18
24	44	M	N	N	N	Y	N	N	22.5	166	43	78	210	10
25	43	M	Y	N	Y	Y	N	N	21.6	173	39	64	121	11
26	38	M	N	N	N	N	N	N	23.3	149	38	80	123	19
27	41	M	Y	Y	Y	N	N	Y	27.9	240	36	85	135	22
28	36	M	Y	Y	N	N	Y	N	21.6	182	35	81	125	15.8
29	37	M	Y	N	N	N	N	Y	23.6	171	40	63	141	11
30	40	M	N	N	N	N	N	N	24	169	43	79	152	9
31	42	M	N	N	N	N	Y	Y	22.3	135	41	71	115	5
32	42	M	Y	N	N	Y	N	N	24.5	142	42	91	190	18.3
33	33	M	Y	Y	N	N	N	N	20.9	165	37	85	113	19
34	36	M	N	N	N	N	Y	Y	21.6	172	36	61	126	7
35	35	M	Y	N	N	N	N	N	19.8	160	37	69	129	8
36	40	F	N	N	N	N	Y	N	22.3	172	35	88	132	19.5
37	41	M	N	Y	Y	N	N	Y	24.4	151	40	77	115	11
38	32	M	Y	Y	N	N	N	N	20.6	141	40	90	117	20.5
39	42	M	N	N	N	Y	Y	N	18.6	188	43	87	105	6
40	39	M	Y	N	N	N	N	N	24.5	190	41	95	120	9.5

Controls

S. No.	AGE	SEX	Smoking	Alcohol	FH	PH	HT	DM	BMI	TC	HDL	LDL	TGL	HOMOC
1	32	M	Y	N	N	N	N	N	20	136	42	70	133	6
2	40	M	N	N	N	N	N	N	22	130	40	75	126	7.5
3	35	M	Y	Y	N	N	N	N	21	152	39	64	132	6.8
4	38	M	N	N	N	N	N	N	20.5	140	38	63	128	5.5
5	42	F	N	N	N	N	N	N	21.6	160	40	92	115	6
6	41	M	N	N	N	N	N	N	23.2	153	41	73	120	7.2
7	43	F	N	N	N	N	N	N	20.9	145	45	72	132	7.7
8	39	M	Y	Y	N	N	N	N	23	156	44	69	135	8.1
9	38	M	N	N	N	N	N	N	22	166	41	68	140	6
10	35	M	N	N	N	N	N	N	21	170	42	62	115	7
11	39	M	Y	Y	N	N	N	N	24	192	39	72	122	8
12	41	F	N	N	N	N	N	Y	25	183	42	97	140	11
13	38	M	N	N	N	N	Y	N	23.6	178	43	90	135	12
14	44	M	Y	N	N	N	N	Y	26	197	42	90	130	10
15	40	M	N	N	N	N	Y	N	25.4	182	41	88	122	9.1
16	43	M	Y	N	N	N	N	Y	24.8	189	38	85	111	11
17	38	M	N	N	N	N	N	N	23.6	179	41	90	140	10.2
18	33	F	N	N	N	N	N	Y	25.4	182	39	98	160	12.2
19	40	M	N	N	N	N	N	Y	23.8	185	41	89	143	11.3
20	42	M	Y	N	N	N	N	N	24.4	178	40	78	135	9.3
21	41	F	N	N	N	N	N	N	25.5	190	38	95	145	12.2
22	43	M	N	N	N	N	N	N	23.3	181	39	92	155	11.4
23	36	M	N	N	Y	N	N	N	24	190	38	95	148	9
24	39	M	N	N	N	N	N	N	24.8	178	39	87	130	8
25	37	M	Y	N	N	N	N	N	25.2	176	38	88	132	7.8

HT - HYPERTENSION

DM- DIABETES MELLITUS

BMI- BODY MASS INDEX

TC- TOTAL CHOLESTROL

PH - PAST HISTORY OF IHD

HDL - HIGH DENSITY LIPOPROTEINS

LDL- LOW DENSITY LIPOPROTEINS

TG-TRIGLYCERIDES

HOMOC-HOMOCYSTEINE

FH- FAMILY HISTORY OF IHD

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