Original Article

Correlation between lipid profile & carotid intima media thickness in cerebral ischemia

Debalina Sengupta[1]*, Jayati Bardhan[2], Anil Baran Singha Mahapatra[3], Joyshree Banerjee[4] and Jayanta Kumar Rout[5]

Departments of Physiology[1,3,4] and Biochemistry[5]
R.G. Kar Medical College,
Khudiram Bose Sarani, Kolkata – 700004
West Bengal, India

Department of Radiology[2]
Bankura Sammilani Medical College,
Bankura, West Bengal

Abstract

Cerebrovascular accident or stroke is defined by an abrupt onset of neurological deficit that is attributable to a focal vascular cause. Stroke is a major cause of morbidity and mortality worldwide. This may result from brain infarction or hemorrhage. Carotid atherosclerosis is a reasonable risk factor for cerebral ischemic stroke. Deranged lipid metabolism due to various modifiable and non-modifiable risk factors leads to the pathogenesis of atherosclerosis. This study is intended to find out any association between altered lipid metabolism (Cholesterol, Triglycerides, LDL : HDL ratio) and development of cerebral ischemia. An observational case control study was conducted with 50 cases of cerebral ischemia and 50 age & sex matched healthy controls within age group 50-70 years. After inclusion of cases and controls and taking informed consent they underwent history taking, proper clinical examination &biochemical investigations (lipid profile). Then data were collected and results were statistically analyzed using Chi-square test & Independent Sample "T-test". The study showed altered lipid profile is associated with cerebral ischemia by increasing carotid intima media thickness (IMT). There was significant (p<0.001) dyslipidemia (NCEP ATP III guidelines) in cases as compared to controls. Hence early diagnosis and monitoring of dyslipidemia and treatment of the high risk group with anti hyperlipidemic drugs will help to prevent the incidence of cerebral ischemic stroke thereby reducing morbidity and mortality.

*Corresponding author :

Dr. Debalina Sengupta, MD (Physiology), at present Assistant Professor, Department of Physiology, NRS Medical College, 138 AJC Bose Road, Kolkata – 700014, West Bengal, India; Cell: 9830434158; Off.: 033-22653213 Email: deblinagupta@hotmail.com

(Received on June 29, 2013)

Introduction

Cerebrovascular accident or stroke is defined by an abrupt onset of neurological deficit that is attributable to a focal vascular cause (1). Stroke is a major cause of morbidity and mortality worldwide. This may result from brain infarction or hemorrhage. Majority

(80%) are due to ischemic cerebral damage as a result of decrease in cerebral perfusion pressure (2). A fall in cerebral blood flow to zero causes death of brain tissue within 4-10 min; values <16-18 ml/100 g tissue per minute cause infarction within an hour ; and values < 20 ml/100 g tissue per minute cause ischemia without infarction unless prolonged for several hours or days (3). Even today cerebrovascular accident stands as the third major killer in western countries after coronary artery disease & cancer. Atherosclerosis is a progressive inflammatory disease characterized by accumulation of lipids and fibrous elements in the large arteries (4). The early lesions of atherosclerosis consists of sub endothelial cholesterol engorged macrophages called foam cells. More advanced lesions are characterized by the accumulation of lipid rich necrotic debris and smooth muscle cells. Carotid Intima-Media thickness (CIMT) is a non-invasive alternative marker of atherosclerotic disease that has been used extensively. CIMT is defined as the distance between the lumen-intima interface, which corresponds to the inner and outer echogenic lines seen on the B-mode ultrasound (Carotid Doppler) image. Increased CIMT has consistently been shown to predict future vascular events (5, 6). While there is an overwhelming amount of evidence relating high levels of serum total and LDL cholesterol and low levels of HDL cholesterol with coronary atherosclerosis, the relation between serum lipids, lipoproteins and cerebrovascular atherosclerosis is less clear. Studies of cholesterol levels in stroke patients have revealed results varying from insignificant changes to a moderate elevation. The meager reports available in Indian patients who have different social, living and dietary habits compared to western population, prompted us to undertake this study.

Thus this study is intended to correlate the Common Carotid Artery Intima Media Thickness (CCA-IMT) with lipid profile (Cholesterol, Triglycerides, LDL:HDL ratio) in cerebral ischemia.

Material and methods

An observational, case control type of study (7) was conducted in the Department of Physiology in collaboration with Department of Medicine, Radiology and Biochemistry at R.G. Kar Medical College, Kolkata from January, 2010 to July, 2011. After receiving approval from the Ethics committee for human experiments of the institute selection of cases and controls were done by simple random sampling. Study population included 50 stroke patients diagnosed by a neurologist as per WHO definition (8) and CT Scan (Computed Tomography) suggestive of cerebral infarction. Age sex matched 50 apparently healthy individuals were taken as "control group". All the volunteers were explained the purpose of the experiment, and a written consent was obtained from each of them.

Subject exclusion criteria :

- Severely ill subjects.

- Patients with cerebral hemorrhage.

- TIA without any CT Scan features of ischemic stroke.

- Intracranial neoplasm.

- Patients suffering from meningitis, neurocysticercosis or any infections.

- Head Injury.

- Intracranial aneurysms.

- AV malformations.

- Any cardiac source of embolism.

- H/O intake of any lipid lowering drugs.

- H/O Smoking.

- Coronary Heart disease

Subject inclusion criteria :

1. Diagnosed stroke patients as per WHO definition (8) as "Rapidly developing clinical signs of focal or global neurological deficits lasting for 24 hours or longer or resulting in death with no apparent cause other than vascular origin".

2. Patients admitted with cerebrovascular accident confirmed to be ischemic in nature on CT scan.

3. Subjects with first attack of stroke were chosen.

4. Age: The interest of this study is focused on elderly age group. Studies conducted by Bhattacharya et al (9), Dalal et al (10) and Sridharan et al (11), showed that mean age for onset of stroke in India ranges from 63-65 for men and 57-68 for women. So this study was aimed at 50-70 yrs age group of subjects.

The controls were apparently healthy and age-sex matched with cases.

Study parameters:

■ History and Physical Examination with examination of the Nervous System in details.

■ Biochemical Tests: (1) Lipid Profile (after 12 hrs overnight fasting) for Cholesterol (CHOD/PAP Method), Triglycerides by (GPO/PAP Method) LDL: HDL ratio. (HDL by PEG Precipitation method and LDL by Friedewald's formula (Total Cholesterol) (Triglycrides/5) (HDL Cholesterol).

■ Carotid Doppler Study was performed only with appropriate high frequency transducers with patient in supine position. High resolution B mode, colour Doppler ultrasonography of both carotid arteries were performed with an ultrasound machine (Image Point HX)) equipped with a 10 MHz linear array transducer. After the carotid arteries were located by transverse scans the probe was rotated 90° to obtain and record a

longitudinal image of the anterior and posterior walls. The maximum IMT was measured at the near and far walls of the common carotid artery, the bifurcation, and the internal carotid arteries and was expressed as a mean aggregate value. In the study values greater than 0.8 mm (0.08 cm) were considered abnormal (12, 13, 28).

■ Plain CT scan of Brain

Plan of statistical analysis

■ Datas were compiled in MS excel worksheet (Office document 2007) and analysis done in SPSS VERSION 17 statistical software (for windows).

■ Results were statistically analyzed using following statistical tests :

    • Chi-square test.

    • Independent Sample "T-test".

    • Multiple Regression Analysis.

■ Mean and Standard deviation were calculated from the results of individual parameters (14).

■ P value of <0.05 was considered to be significant and P<0.001 as highly significant. The results were calculated within 95% confidence limits.

Results

An observational case control study was performed at R.G. Kar Medical College and Hospital, Kolkata.

TABLE I : Basic characteristics of subjects.

Basic characteristics	Case (N=50) Mean±SD	Control (N=50) Mean±SD	P value
Age (yrs)	61.72±6.93	59.38±5.74	>0.05
Systolic BP (mm of Hg)	154.60±16.13	135.48±14.45	>0.05
Diastolic BP (mm of Hg)	97.72±16.07	90.88±11.50	>0.05
Fasting Blood Sugar (mg/dl)	103.20±40.57	117.92±33.85	>0.05
Post Prandial Blood Sugar (mg/dl)	134.24±60.62	148.48±50.80	>0.05

P value <0.05 is considered to be significant.
P value <0.001 is considered to be highly significant.
BP - Blood Pressure, FBS - Fasting Blood Sugar, PPBS - Post Pandrial Blood Sugar.

TABLE II : Comparative study of lipid parameters between cases and controls.

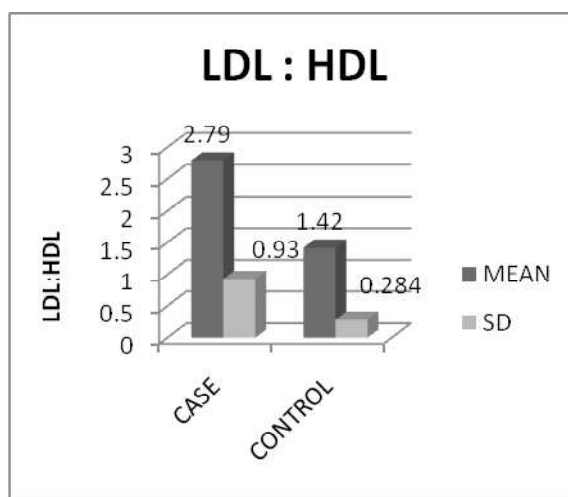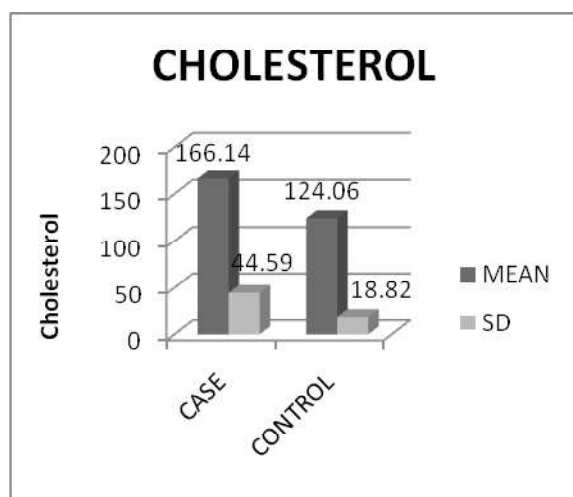| Lipid parameters	Case (N=50)	Control (N=50)	't' value	P value
Cholesterol (mg/dl)	166.14±44.59	124.06±18.82	6.148	p<0.001
LDL:HDL	2.79±0.93	1.42±0.284	10.024	p<0.001
Triglyceride (mg/dl)	194.44±48.22	91.04±19.81	14.025	p<0.001

(A)

(B)

(C)
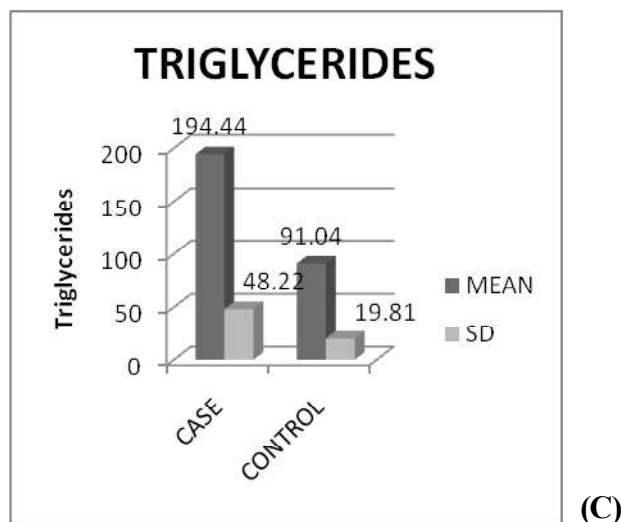
Diagram 1 : Table II and Diagrams 1A, 1B, 1C shows a significant difference (p<0.001) of lipid profiles (Cholesterol, LDL:HDL and Triglyceride) between cases and controls.

On an average about 100 patients of cerebral ischemia got admitted every year over the past 3 yrs. As the duration of this study was about 1 yr 6 mths and due to single person constraint hundred cases of stroke (CVA) were selected from the Indoor Department of Medicine and among them 50 patients were included in this study following strict inclusion

& exclusion criteria. Results were evaluated and

TABLE III : Comparative study of Avg. Imt between cases and controls.

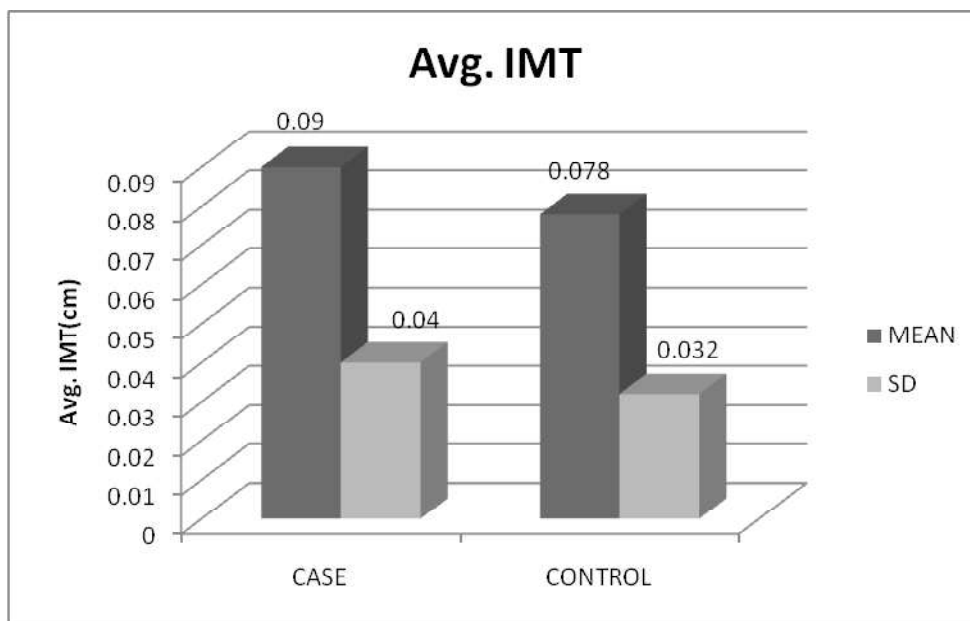| Parameter	Case	Control	't' value	P value
Avg. IMT (cm)	0.09±0.04	0.078±0.032	2.093	p<0.05

Diagram 2 :   There is a significant difference (p<0.05) of Avg. IMT between cases and controls.

TABLE IV :   Correlation between lipid profile parameters and average IMT.

Lipid profile parameters	Cases (N=50)		Controls (N=50)	
	Pearson'sCorrelation Coefficientwith Avg. IMT (r)	Significance (2-tailed)	Pearson'sCorrelation Coefficientwith Avg. IMT (r)	Significance (2-tailed)
Cholesterol (mg/dl)	0.675	p<0.001	0.626	P<0.001
LDL:HDL	0.664	p<0.001	0.560	p<0.001
Triglyceride (mg/dl)	0.594	p<0.001	0.584	p<0.001

analysed on the basis of the specific objectives of the study.

Discussion

Stroke is an important cause of mortality and morbidity worldwide. Carotid atherosclerosis is a reasonable risk factor for cerebral ischemic stroke. Deranged lipid metabolism due to various modifiable and non-modifiable risk factors leads to the pathogenesis of atherosclerosis. Gradual deposition of lipoproteins in the carotid arteries leads to increased carotid artery Intima Media Thickness (IMT) and gradually plaque formation occurs. Rupture and dislodgement of a plaque leads to embolism and subsequent ischemic stroke. With this background we have done an observational study with 50 cases of cerebral ischemia and 50 age and sex

matched controls based on the three specific objectives of the study. After collecting data, results and statistical analysis was done. Regarding the basic characteristics from Table I there is insignificant (p>0.05) difference of age, SBP, DBP, FBS, PPBS between cases & controls. So cases and controls are matched according to age, SBP, DBP, FBS, PPBS. Earlier studies of Bhattcharya S et al (9), Dalal P et al (10), Sridharan S et al (11) showed that the mean age of onset of stroke for men in India ranges from 63-65 for men and 57-68 for women, hence we have taken the same range of age as study population.

Comparative study of lipid parameters between cases and controls showed that mean Cholesterol of cases is 166.14 mg/dl with SD 44.59 whereas that of controls is 124.06 mg/dl with SD18.82; mean LDL:HDL of cases is 2.79 with SD 0.93 and that of controls
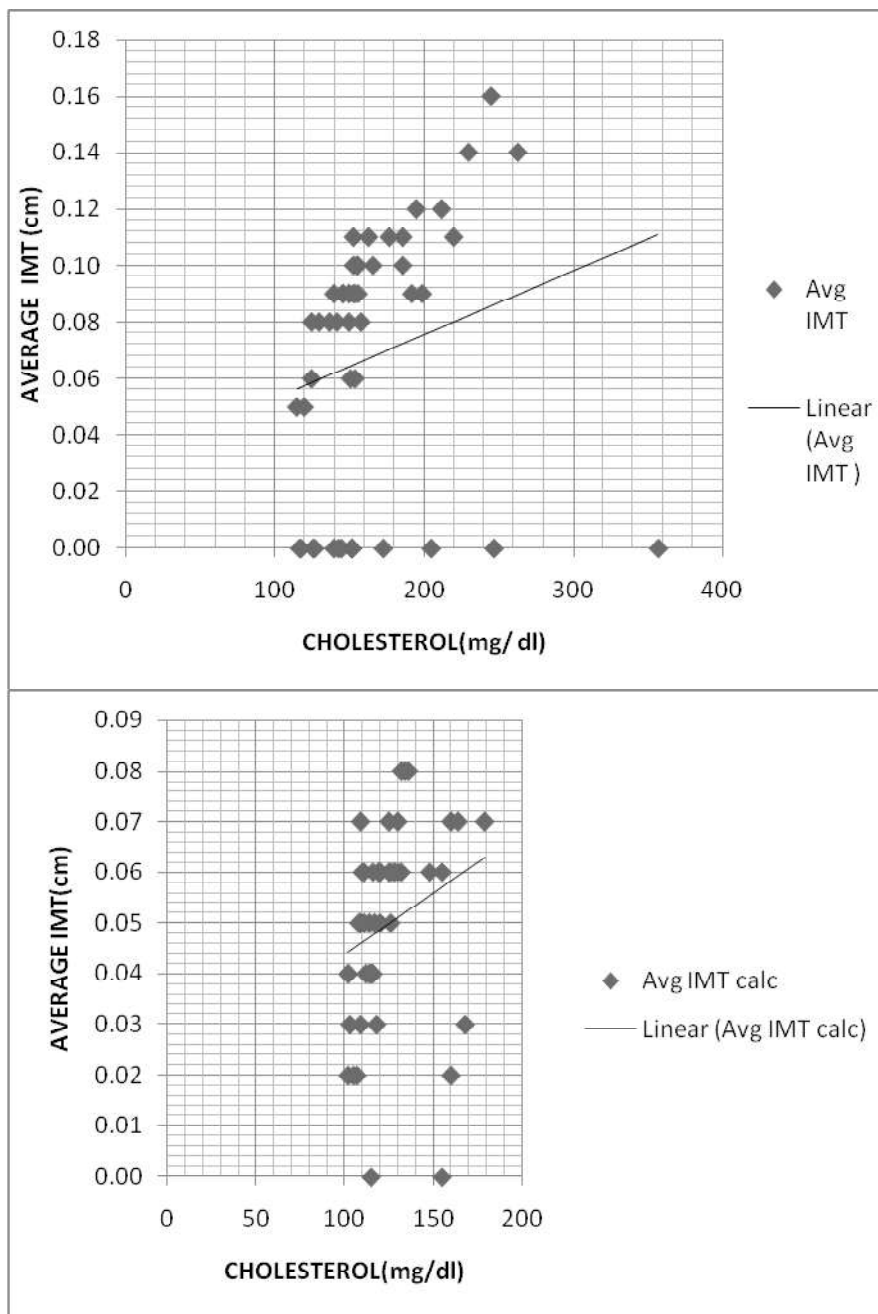
Diagram 3 : Scatter plot in between cholesterol (mg/dl) and AVG. IMT of Case (A) (N=50) & Controls (N=50) (B). IMT - Intima Media Thickness (cm).

is 1.42 with SD 0.284 and mean Triglyceride of cases is 194.44 mg/dl with SD 48.22 whereas that of controls is 91.04 mg/dl with SD19.81. Hence there is a significant difference (p<0.001) of lipid profiles (Cholesterol, LDL:HDL and Triglyceride) between cases and controls. It is well documented that increasing levels of total plasma cholesterol and low density lipoprotein cholesterol and decreasing level

of high density lipoprotein cholesterol, are strong risk factors for coronary heart disease, while the relation between risk of blood lipids and stroke is much weaker (15, 16, 17). However recent studies clarified the relationship between lipids and stroke, as well as showing that the risk of stroke and amount of carotid atheroma can be reduced with cholesterol lowering drugs (18, 19, 20, 21, 22). On
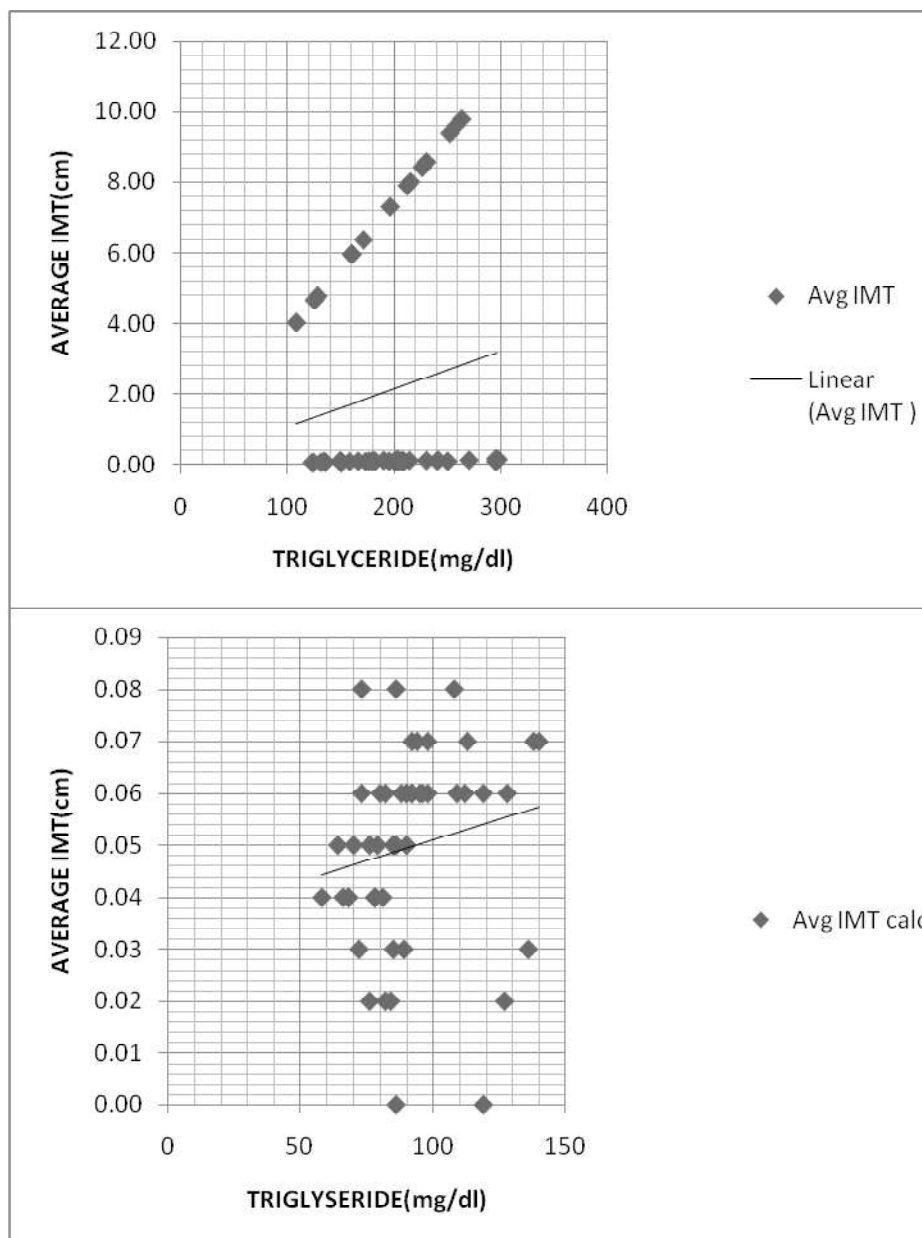
Diagram 4 : Scatter plot in between triglyceride (mg/dl) and AVG. IMT of Case (A) (N=50) & Controls (N=50) (B).

the other hand an inverse association between total cholesterol and cerebral hemorrhage had been reported (17, 23).

Our study also shows a strong positive correlation (p<0.001) of Avg IMT(cm) with Cholesterol (mg/dl) both in cases (r=0.675) and controls is (r=0.626): Avg IMT(cm) with LDL:HDL both in cases (r=0.664) and controls is (r=0.560) : Avg IMT(cm) with triglyceride both in cases (r=0.594) and controls is (r=0.584). Scatter diagrams support

the findings.

O'Leary DH et al. (24) reported that an increased carotid IMT is associated with a higher risk of stroke and acute myocardial infarction in an elderly population and it is also a more powerful predictor of cardiovascular disease than the conventional risk factors for atherosclerosis. Similarly, Touboul PJ et al. (25) demonstrated a greater CCA IMT in patients with all major cerebral infarction subtypes compared with controls.
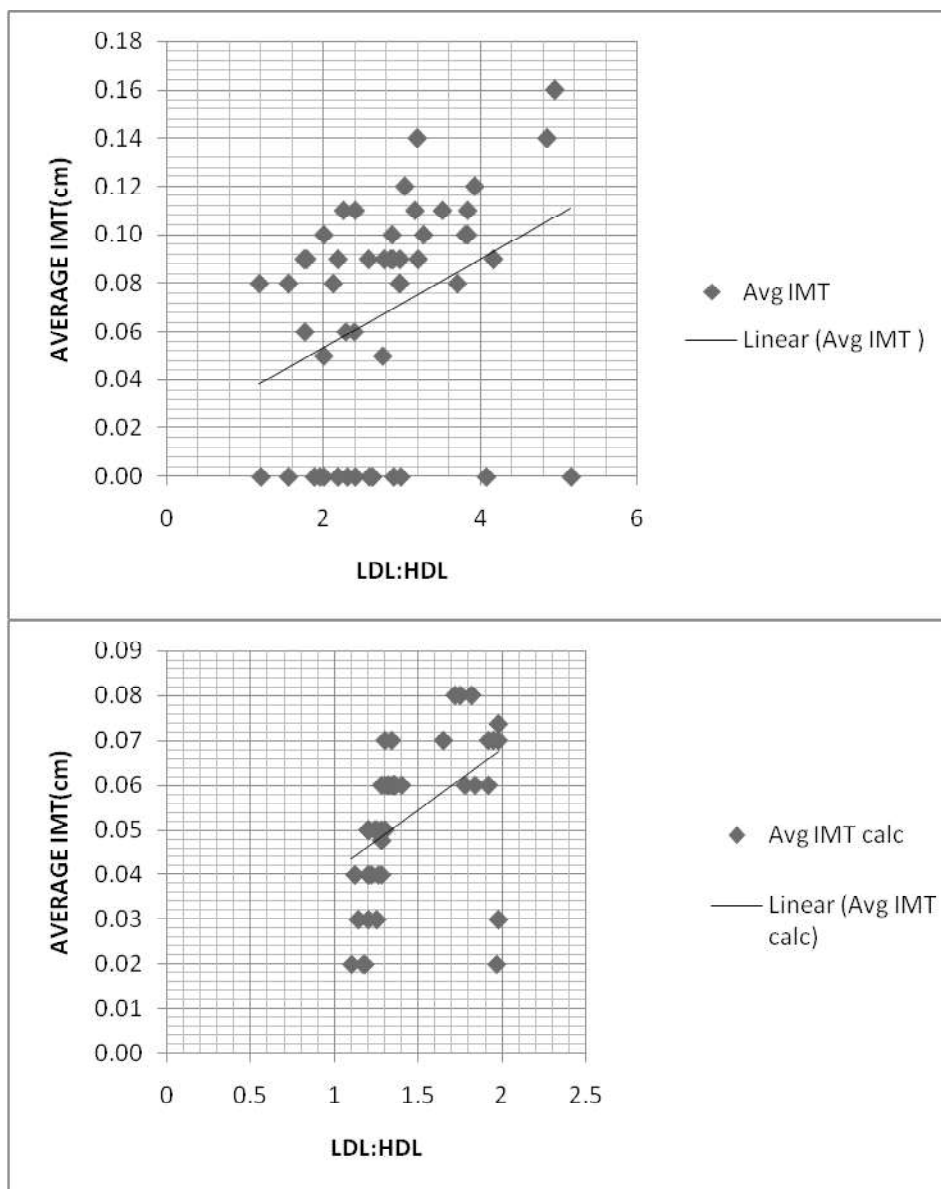
Diagram 5 : Scatter plot in between LDL:HDL and AVG. IMT of Case (A) (N=50) & Controls (N=50) (B).

From the Table IV and Scatter Plot diagrams 3A/B, 4A/B, 5A/B it is evident that there is a significant correlation (p<0.001) between Cholesterol, Triglyceride, LDL:HDL and Avg IMT of cases and controls.

TABLE V : Multiple regression analysis between different lipid profile parameters with Average Imt of cases (N=50).

Model	Standardized coefficients Beta	t	Significance
Constant		0.287	0.776
Cholesterol	+0.367	1.918	0.061
LDL: HDL	+0.383	2.106	0.041 (p<0.05)
Triglyceride	−0.043	−0.349	0.729

Predictors: Constant: Triglyceride, LDL:HDL & Cholesterol
Dependent Variable: Avg. IMT
From the above table it is evident that among all the lipid parameters taken in the study LDL:HDLratiohas a significant correlation (p<0.05) with Avg IMT.

The multiple regression analysis between different lipid profile parameters and Avg IMT of cases showed

- For Cholesterol β = +0.367 (β – Standardized Coefficient)

- For LDL:HDL β = +0.383

- For Triglyceride β = −0.043

Thus it is evident that among all the lipid parameters taken in the study LDL:HDL ratio has maximum correlation (p<0.05) with avg IMT.

In the Helsinki Study, a 5-year clinical trial of more than 4,000 middle-aged men with elevated lipids, the LDL-C/HDL-C ratio had more prognostic value than LDL-C or HDL-C alone (260. In a cross-sectional study conducted by Enomoto M et al (27), a significant association between IMT and proatherogenic lipoprotein measurements was seen and the results of multiple linear regression analysis showed that the LDL-C/HDL-C ratio was the strongest predictor for IMT progression.

Summary

Cerebrovascular accident or Stroke is a major cause of morbidity and mortality worldwide. This may result from brain infarction or hemorrhage. Among all the neurological disorders of adult life, stroke ranks first in frequency & importance. Carotid atherosclerosis is a reasonable risk factor for cerebral ischemic stroke. Deranged lipid metabolism due to various modifiable and non-modifiable risk factors leads to the pathogenesis of atherosclerosis.

Thus this study is intended to correlate the Common Carotid Artery Intima Media Thickness (CCA-IMT) with lipid profile (Cholesterol, Triglycerides, LDL:HDL ratio) in cerebral ischemia.

On this background three specific objectives of our study were –

- To find out any association between altered lipid metabolism and development of cerebral ischemia.

- To find out the correlation between carotid IMT changes and altered lipid profile.

An observational case control study was conducted at the Dept. of Physiology, R.G. Kar Medical College & Hospital in collaboration with Dept. of Radiology, Dept. of Biochemistry for the period of January 2010 to July, 2011. After Ethical Committee clearance, 100 cerebral infarction cases, were randomly selected from stroke patients attending Medicine department R.G. Kar Medical College. Among them 50 patients were included in this study following strict inclusion & exclusion criteria. Age & sex matched 50 healthy

controls were included according to inclusion & exclusion criteria from the Department of Physiology & Biochemistry. After inclusion of cases & controls, every subjects were explained about the whole procedure and consent formed duly signed. After that they underwent history taking, proper clinical examination,& special investigations (lipid profile, carotid doppler). Then data were collected, grand charts are formed, and then by using SPSS (version-17) software's statistical analysis were done. Following results were obtained.

Mean age group of stroke patients were 61.72 with SD 6.93, & range is 50 years to 70 years. There were equal number of male female, as we excluded more male patient due to smoking history. Most of the patient were hypertensive. Mean SBP is 154.60 mm of Hg & DBP is 97.72 mm of Hg. Mean of cholesterol, LDL:HDL, Triglyceride were 166.14, 2.79, 194.44 respectively. There was significant ($p<0.001$) dyslipidemia (NCEP ATP III guidelines) in cases as compared to controls. There was significant positive correlation of average IMT with levels of Cholesterol ($r=+0.675$, $p<0.001$), LDL:HDL ($r=+0.664$, $p<0.001$) & Triglycerides ($r=+0.294$, $p<0.05$) in cases. Even we found that average IMT increases with increasing the level of cholesterol, LDL:HDL, triglyceride in control. So dyslipidemia is an important risk factor for increasing average IMT in high risk group. Multiple Regression Analysis shows that among the multiple lipid profiles, the most important factor for increasing average IMT is LDL:HDL.

Conclusion

From the above study it can be concluded that altered lipid profile is associated with cerebral ischemia by increasing carotid intima media thickness (IMT). Among the multiple lipid profiles, the most important factor for increasing average IMT is LDL:HDL.LDL:HDL had already been an important marker for cardiovascular disease but our study shows its importance in relation to cerebrovascular disease also. Gradual deposition of lipoproteins in the carotid arteries leads to increased carotid artery Intima Media Thickness (IMT) and gradually plaque formation occurs. Rupture and dislodgement of a soft plaque leads to embolism and subsequent ischemic stroke.

Further studies are necessary to establish whether immediately after an ischemic event initiating an aggressive medical (statin) or surgical therapy will reduce the risk of stroke and the risk of subsequent ischemic events.

So monitoring the lipid profile at regular intervals may prevent many complications due to accelerated atherosclerosis in many patients. Hence early diagnosis of dyslipidemia and treatment of the high risk group with anti hyperlipidemic drugs will help to prevent the incidence of cerebral ischemic stroke thereby reducing morbidity and mortality.

Limitations of the study

A. Small sample size.

B. Study period was only one year.

C. Follow up was not possible.

Acknowledgements

References

1. Wade S Smith, Joey D, English S. Claiborne Johnston. Cardiovascular diseases, Harrison's principles of Internal Medicine, Vol. II, Ed 17th; 364: 2513–2530.

2. Uma N, Chugh SK, Harshwardhan, Geol A, Gopal D. Echocardiography in patients with cerebral infarction. *JAPI* 1999; 47: 291–293.

3. Adams & Victor, Principles of Neurology 3rd Ed.

4. Luiss AJ, Atherosclerosis, *Nature* 2000; 407: 233–241.

5. Dijk JM, van der, Graaf Y, Bots ML, Grobbee DE, Algra A. Carotid intima-media thickness and the risk of new vascular events inpatients with manifest atherosclerotic disease: the SMART study. *Eur Heart J* 2006; 27: 1971–1978.

6. Stein JH, Korcarz CE, Hurst RT, Lonn E, Kendall CB, Mohler ER, Najjar SS, Rembold CM, Post WS. Use of carotid ultrasound to identify subclinical vascular disease and evaluate cardiovascular disease risk: a consensus statement from the American Society of Echocardiography Carotid Intima-Media Thickness Task Force. Endorsed by the Society for Vascular Medicine. *J Am Soc Echocardiogr* 2008; 21: 93–111.

7. Park K. Textbook of Preventive and Social Medicine. Principles of epidemiology and epidemiologic methods, analytical epidemiology; Ed 3: 2009; p. 67.

8. Hatane S. Experience from Multicentre stroke register a preliminary report. *WHO* 1976; 54: 541–553.

9. Bhattacharya S, Prasarsaha S, Basu A, Das K. A 5 year prospective study of incidence, morbidity and mortality stroke profile on stroke in a rural community of Eastern India. *J Indian Med Assoc* 2005; 103 (12): 65510.

10. Dalal P et al. Population-bases stroke survey in Mumbai, India: Incidence and 28-day case fatality. *Neuroepidemiology* 2008; 31: 254–261.

11. Sridharan S et al. Incidence, types, risk factors and outcome of stroke in a developing country: the Trivadrum Stroke Registry. *Stroke* 2009; 40: 1212–1218.

12. Jadhav UM, Kadam NN. Carotid I-M thickness as an independent predictor of coronary artery disease. *Indian Heart J* 2001; 53: 458–462.

13. Polak JF. Carotid Ultrasound. *RCNA* 2001; 39(3): 569–589.

14. Mahajan BK. Methods in Biostatistics 2006; 6: 8–9.

15. Hokanson JE, Austin MA. Plasma triglycerides level is a risk factor for cardiovascular dis. Independent of HDL-cholesterol level: a meta-analysis of population-based prospective studies. *J. Cardiovasc. Risk* 1996; 3: 213–219.

16. Law MR, Wald NJ, Wu T et al. Systematic underestimation of association between serum cholesterol concentration and ischemic heart disease in observational studies: data from the BUPA study. *BMJ*, 1994; 308: 373–379.

17. Lindenstorm E, Boysed G, Nyboe J. Influence of total cholesterol, high density lipoprotein cholesterol, and triglycerides on risk of cerebrovascular disease: the Copenhagen city heart study. *BMJ* 1994: 309: 11–11.

18. Blauw GJ, Lagaay AM, Smelt AH, et al. Stroke, statins and cholesterol a meta-analysis of randomized, placebo-controlled, double-blind trials with HMG-Co-A reductase inhibitors. *Stroke* 1997; 28: 946–950.

19. Bucher HC, Griffith LE, Guyatt GH. Systematic review on the risk and benefit of different cholesterol-lowering interventions. *Arterioscler Thromb Vasc Biol* 1999; 19: 187–195.

20. Hodis HN, Mack WJ. LaBree L, et al. Reduction in carotid arterial wall thickness using lovastatin and dietary therapy, a randomized controlled clinical trial. *Ann Intern Med* 1996; 124: 54S–556.

21. Shepherd J, Cobbe SM, Ford I, et al. Prevention of coronary heart disease with pravastatin in men with hypercholesterolemia: West of Scotland Coronary Prevention Study Group. *N Engl J Med* 1995; 333: 1301–1307.

22. Vaughan CJ, Delanty N. Neuroprotective properties of statins in cerebral ischemia and stroke. *Stroke* 1999; 30: 1969–1973.

23. Ueshima H, Iida M, Shimamolo T, et al. Multivariate analysis

of risk factors for stroke: eight-year follow-up study of farming villages in Akila, Japan. *Prev Med*. 1980; 9: 722–740.

24. O'Leary DH, Polak JF, Kronmal RA, et al. Thickening of the carotid wall: a marker for atherosclerosis in the elderly ? *Stroke* 1996; 27: 224231.

25. Touboul PJ, Elbaz A, Koller C, et al. Common carotid artery intima-media thickness and brain infarction. *Circulation* 2000; 102: 313318.

26. Manninen V, Tenkanen L, Koskinen P, Huttunen JK, Manttari M, Heinonen OP, Frick MH. Joint effects of serum triglyceride and LDL cholesterol and HDL cholesterol concentrations on coronary heart disease risk in the Helsinki Heart Study. Implications for treatment. *Circulation* 1992; 85: 3745.

27. Enomoto M, Adachi H, Hirai Y, LDL-C/HDL-C Ratio Predicts Carotid Intima Media Thickness Progression Better Than HDL-C or LDL-C Alone. *Journal of Lipids* 2011; ID 549137: 6.

28. The Extracranial Cerebral Vessels, Edward I. Bluth, Barbara A. Carroll, Diagnostic Ultrasound, 4th Ed, Vol 1, Chap 25, pg. 951, Carol M. Rumack et al.