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RESEARCH ARTICLE

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PLATELET AGGREGATION, FIBRINOGEN AND LIPID PROFILE IN CEREBROVASCULAR ACCIDENTS

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ABSTRACT

Platelet aggregation is the one of the initial haemostatic events that stops bleeding resulting from a vascular injury. It has been observed that increased collagen induced platelet aggregation in acute ischemic stroke patients. In view of the above, the present study was planned to determine the above said parameter in acute ischemic stroke patients. A total number of 60 subjects participated in the present study which included 30 acute ischemic stroke cases and 30 controls. Fasting plasma glucose, lipid profile, Plasma fibrinogen, Platelet aggregation were estimated.

Platelet aggregation was significantly lower in cases (acute ischemic stroke) as compared to controls; whereas collagen induced platelet aggregation and plasma fibrinogen were significantly higher in cases. No significant differences were found in triglycerides, HDL – cholesterol, VLDL, TG/HDL and Atherogenic Index. In present study we found an increased collagen induced platelet aggregation which is induced by plasma fibrinogen and positive correlation between atherogenic index and fibrinogen.

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KEY WORDS:

Platelet aggregation, Fibrinogen, Atherogenic index.

INTRODUCTION

Stroke(Cerebrovascular accident) is defined as the primary pathology of brain vascular system that temporarily or permanently affects brain functions as a result of ischemia or hemorrhage. Due to the consequent high mortality and morbidity rates, stroke is to become a major community health problem world wide and it is the third leading cause of death and long term morbidity in developed and developing countries [1].

Higher levels of serum total cholesterol, LDL cholesterol, lipoprotein (a) and lower HDL cholesterol have been implicated in the development of stroke and these lipid factors are known to be atherothrombotic. HDL may be protective against particularly nonfatal and ischemic stroke. In patients with coronary heart disease risk of ischemic stroke has been inversely related to the high HDL levels. But although a low HDL level is clearly a risk factor of CHD, the relationship with ischemic stroke is still unclear.

Although the various lipid risk factors (TC/HDL, TG/HDL, LDL/HDL, non HDL cholesterol (NHC) and atherogenic index (AI) are the predictors of cardiovascular vascular diseases, which contain both atherogenic and anti-atherogenic lipid components and these ratios has not been widely studied in association with ischemic stroke.

Platelets may play an important role in the pathogenesis of ischemic stroke since the atherosclerotic thrombus is made up mainly of fibrin, erythrocytes and platelets [2] and platelet emboli are the usual cause of transient ischemic attacks (TIA). The thrombus formation is considered to be a consequence of increased tendency of platelets to aggregate and there are several reports of enhanced platelet aggregability after cerebral ischemia. Platelet aggregation has been studied in vitro using agonist such as adenosine diphosphate (ADP), collagen and epinephrine. The Carephilly Prospective study demonstrates a paradoxical association of increased ischemic stroke with decreased whole blood ADP induced platelet aggregation [3]. It has been observed that increased collagen induced platelet aggregation in acute ischemic stroke patients. However, it is still controversial, whether platelets are hyperactive, unchanged, or hypoactive during the acute and chronic periods of stroke and whether the changes in platelet activity is the primary event in the pathogenesis of ischemic stroke.

Elevated levels of fibrinogen are associated with atherosclerosis and have been reported in patients with coronary heart disease, peripheral vascular disease, and carotid stenosis. Studies such as the Framingham study and Northwick Park Heart Study have demonstrated that increased fibrinogen content is an independent risk factor for atherosclerotic diseases such as myocardial infarction/stroke.

Even though fibrinogen levels, lipid parameters and platelet aggregability are reported in stroke patients, the correlation between these factors are not established. In view of the above, the present study was planned to determine the above said parameters in acute ischemic stroke patients.

MATERIALS AND METHODS

The study was conducted on patients who attended the Medicine department of JIPMER hospital (Puducherry) and from the general population around Puducherry. A total number of 60 subjects participated in the

present study which included 30 acute ischemic stroke cases and 30 controls. Informed consent was obtained from every control and from the patient's relatives. The study was approved by research and ethics committees of the institute.

Samples were collected from admitted patients on requisition for routine investigations. 5 ml of venous blood was collected, 2ml added to sodium citrated plastic tubes for platelet aggregation studies and fibrinogen the remaining blood added to the bottles containing EDTA. Plasma was collected by centrifuging EDTA blood at 3000g for 5 minutes. The plasma obtained was used for estimation of glucose and lipid profile and remaining plasma stored at -70^{o}C until analysis.

Fasting plasma glucose and lipid profile were estimated using the reagent kits from Agappe Diagnostics (Kerala, India) adapted to 550 Express plus Batch Auto Analyser (CIBA CORNING, BAYER DIAGNOSTICS).Plasma fibrinogen estimated by immunoturbidometric method [4] .Platelet aggregation was measured with impedance method, using model 700-2 chronolog aggregometer (West Park road, Havertown, PA) [5].

STATISTICAL ANALYSIS

Results were shown as Mean ± SD. Statistical significance of difference between control, and case group was evaluated using Students't' - *test*. Correlation between the clinical parameters was estimated by Pearson's Correlation Co-efficient**.** A P-value less than 0.05 were considered statistically significant. All calculations were performed using the SPSS version 13.0 for windows.

RESULTS

Table 1
***General characteristics and plasma glucose levels in controls and acute ischemic stroke patients*.**

Sl.No	Parameters	Control(n=30)	Case(n=30)
1.	Age (years)	53.40±9.49	56.51±10.57
2.	SBP (mm of Hg)	105.22±13.01	107.58±13.40
3.	DBP (mm of Hg)	71.14±8.14	70.86±7.79
4.	Plasma glucose (mg/dl)	93.37±21.02	130.86±56.05*

* ***P < 0.05 is statistically significant***

Table 1 shows that, plasma glucose was significantly higher in cases (acute ischemic stroke) as compared to controls. No significant differences were found in age, SBP and DBP.

Table 2:
Platelet aggregation and fibrinogen levels in controls and acute ischemic stroke patients.

Sl.No	Parameters	Control(n=30)	Case(n=30)
1.	Platelet aggregation (ADP) (ohms)	8.77±3.70	1.65±2.27*
2.	Platelet aggregation (Collagen) (ohms)	17.96±3.68	24.82±4.49*
3.	Fibrinogen(mg/dl)	298.96±85.97	362.41±89.56*

P < 0.05 is statistically significant

Table 2 shows that, ADP induced platelet aggregation was significantly lower in cases (acute ischemic stroke) as compared to controls, whereas collagen induced platelet aggregation and plasma fibrinogen were significantly higher in cases.

Table 3:
Lipid profile and atherogenic indices in controls and acute ischemic stroke patients.

Sl.No	Parameters	Control(n=30)	Case(n=30)
1.	Triglycerides (mg/dl)	135.14±60.09	158.00±72.87
2.	Total Cholesterol (mg/dl)	188.81±32.83	211.48±39.86*
3.	LDL- Cholesterol (mg/dl)	117.70±24.03	136.51±31.92*
4.	HDL -Cholesterol (mg/dl)	44.18±12.03	43.72±13.12
5.	VLDL (mg/dl)	27.02±12.01	31.60±14.57
6.	TC/HDL	4.48±1.16	5.24±1.81*
7.	TG/HDL	3.33±1.87	4.16±2.62
8.	LDL/HDL	2.82±0.90	3.42±1.38*
9.	NHC (mg/dl)	144.62±29.66	166.75±39.99*
10	Atherogenic Index	0.45±0.24	0.52±0.30

* ***P < 0.05 is statistically significant***

Table 3 shows that, Total cholesterol, LDL – cholesterol, TC/HDL, LDL/HDL, and NHC were significantly higher in cases (acute ischemic stroke) as compared to controls. No significant differences were found in triglycerides, HDL – cholesterol, VLDL, TG/HDL and Atherogenic Index.

Table 4:
Correlation coefficient among platelet aggregation, fibrinogen and lipid profile in acute ischemic stroke patients.

	Platelet aggn(ADP)	Plat aggn (collagen)	Fibrinogen	TG	TC	LDL	HDL	NHC	AI
Platelet aggn(ADP)		.337	0.256	-.124	-.098	-.060	-.007	-.095	-.036
Platelet aggn (collagen)	.		.376*	-.160	.279	.361	.162	.225	-.103
Fibrinogen				.301	.166	.151	-.163	.219	.372*
TG					.535**	.385*	-.419*	.671**	.852**
TC						.933**	.155	.946	.287**
LDL							-.034	.941**	.298
HDL								-.174	-.783**
NHC									.543**
AI									

** P < 0.05 is statistically significant*
*** P < 0.01 is statistically significant*

Table 4 shows that there is significant positive correlation between collagen induced platelet aggregation and that induced by fibrinogen. Atherogenic index positively correlates with fibrinogen, triglycerides, total cholesterol (TC), and non HDL cholesterol (NHC) and significant negative correlation with HDL cholesterol. LDL cholesterol shows positive correlation with NHC and with total cholesterol. Triglyceride shows positive correlation with TC, LDL, and NHC and negative correlation with HDL cholesterol.

DISCUSSION

Platelet aggregation is the one of the initial haemostatic events that stops bleeding resulting from a vascular injury. Aggregation is mediated by the binding of plasma protein fibrinogen to a platelet cell receptor, glycoprotein IIb-IIIa (GPIIb-IIIa) [6]. Platelet hypersensitivity to aggregating agents has been associated with IHD and ischemic stroke. Enhanced platelet activation has been found in subjects with myocardial infarction (MI) and stroke [7]. The Carephilly Prospective study demonstrates a paradoxical association of increased ischemic stroke with decreased whole blood ADP induced platelet aggregation. It has been observed that increased collagen induced platelet aggregation in acute ischemic stroke patients [8].

Our present study shows decreased whole blood ADP induced platelet aggregation and an increased collagen induced platelet aggregation in acute ischemic- stroke patients. Decreased whole blood ADP induced platelet aggregation may be due to an existence of sub-

clinical endothelial disease and compensatory mechanisms, which is down regulating ADP induced aggregation sensitivity in ischemic stroke. We did not find any correlation between platelet aggregation and lipid profile and with other lipid risk factors. However, it is still controversial, whether platelets are hyperactive, unchanged, or hypoactive during the acute and chronic periods of stroke and whether the changes in platelet activity is the primary event in the pathogenesis of ischemic stroke.

Fibrinogen, an acute phase protein and a clotting factor, appears to be an independent risk factor for cardiovascular disease [9], but the mechanism(s) are still uncertain. Fibrinogen is the primary circulating ligand for GPIIb/IIIIa in vivo and has itself been associated with increased risk of IHD and stroke (24), which in turn is a precondition for platelet aggregation in vivo [10].

In the present study we found an elevated fibrinogen levels in acute ischemic stroke patients as compared to controls and we have that found there is a positive correlation between plasma fibrinogen level and collagen induced platelet aggregation. We did not find any correlation between fibrinogen and lipid profile but it shows positive correlation with the atherogenic index.

In the present study there was a significant difference in total cholesterol and LDL cholesterol of ischemic stroke patients compared to controls. LDL cholesterol showed positive correlation with non HDL cholesterol (NHC) and with total cholesterol (TC) in stroke patients. Though we did not find significance in serum triglycerides, but there was an increased TG level in ischemic stroke patients. Triglyceride shows a positive correlation with TC, LDL, and NHC levels and negative correlation with HDL cholesterol in stroke patients.

Although the various lipid risk factors (TC/HDL, TG/HDL, LDL/HDL, non HDL cholesterol (NHC) and atherogenic index (AI) are predictors of cardiovascular vascular diseases and these ratios has not been widely studied in association with ischemic stroke.

In present study we studied all lipid risk factors (TC/HDL, TG/HDL, LDL/HDL, NHC and atherogenic index (AI) and we found significant differences in TC/HDL ratio, NHC, LDL/HDL ratio between acute ischemic stroke patients and controls. Atherogenic index positively correlates with fibrinogen, triglycerides, total cholesterol (TC), and non HDL cholesterol (NHC) and significant negative correlation with HDL cholesterol. Non-HDL cholesterol may be a stronger predictor of CVD than LDL cholesterol or triglycerides considered alone because it correlates highly with atherogenic lipoproteins [11]. In present study NHC shows positive correlation with LDL, TG.

In present study we found an increased collagen induced platelet aggregation which is induced by plasma fibrinogen and positive correlation between atherogenic index and fibrinogen. Further studies are clearly warranted to explore the role of fibrinogen and platelet hyper aggregability in the pathogenesis of acute ischemic stroke in a larger patient population.

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