STUDY ON ASSOCIATION BETWEEN SERUM BILIRUBIN AND ACUTE ISCHEMIC STROKE AND ITS PROGNOSTIC SIGNIFICANCE

A Dissertation Submitted to

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For the Award of the Degree of

M.D. (GENERAL MEDICINE) - BRANCH – I

GOVERNMENT KILPAUK MEDICAL COLLEGE

CHENNAI

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BONAFIDE CERTIFICATE

This is to certify that "**STUDY ON ASSOCIATION BETWEEN SERUM BILIRUBIN AND ACUTE ISCHEMIC STROKE AND ITS PROGNOSTIC SIGNIFICANCE"** is a bonafide work done by **Dr. RAMYA. A**, Post graduate student, Department of General Medicine, Kilpauk Medical College, Chennai-10, under my guidance and supervision in partial fulfilment of rules and regulations of the Tamil Nadu Dr. M.G.R Medical University, for the award of M.D. Degree Branch I (General Medicine) during the academic period from May 2012 to April 2015.

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DECLARATION

I solemnly declare that this dissertation "**STUDY ON ASSOCIATION BETWEEN SERUM BILIRUBIN AND ACUTE ISCHEMIC STROKE AND ITS PROGNOSTIC SIGNIFICANCE"** was prepared by me at Government Kilpauk Medical College and Hospital, Chennai, under the guidance and supervision of **Prof. Dr. S. Ushalakshmi M.D., FMMC,** Professor and Unit Chief, Department of Internal Medicine, Government Kilpauk Medical College and Hospital, Chennai.

This dissertation is submitted to **The Tamil Nadu Dr. M.G.R. Medical University, Chennai** in partial fulfilment of the University regulations for the award of the degree of **M.D. Branch I (General Medicine)**.

Place: Chennai-10 **Dr. RAMYA.A**

Date:

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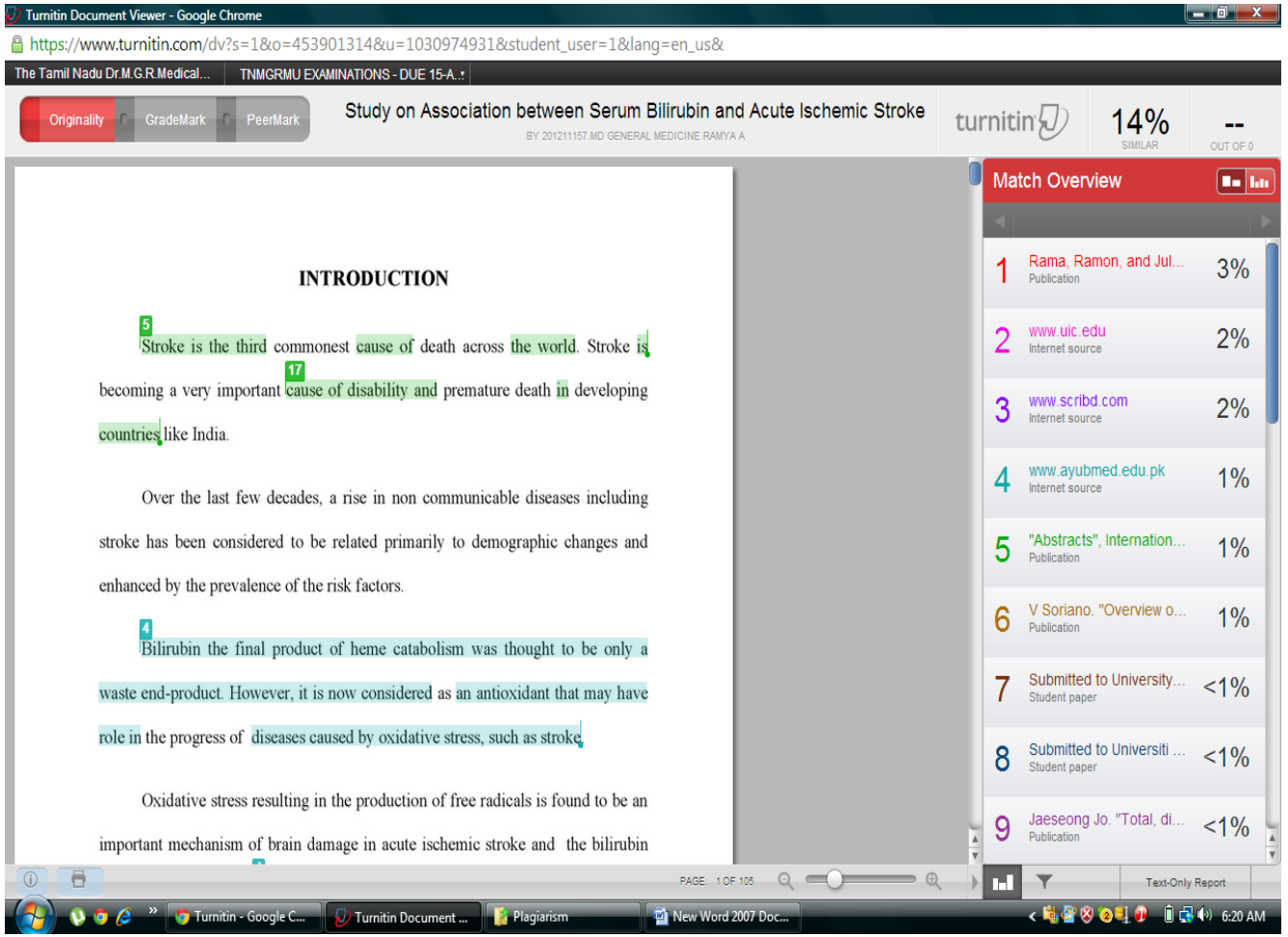
TABLE OF CONTENTS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INTRODUCTION
Stroke is the third commonest cause of death across the world. Stroke is becoming a very important cause of disability and premature death in developing countries like India.
Over the last few decades, a rise in non communicable diseases including stroke has been considered to be related primarily to demographic changes and enhanced by the prevalence of the risk factors.
Bilirubin the final product of heme catabolism was thought to be only a waste end-product. However, it is now considered as an antioxidant that may have role in the progress of diseases caused by oxidative stress, such as stroke.
Oxidative stress resulting in the production of free radicals is found to be an important mechanism of brain damage in acute ischemic stroke and the bilirubin
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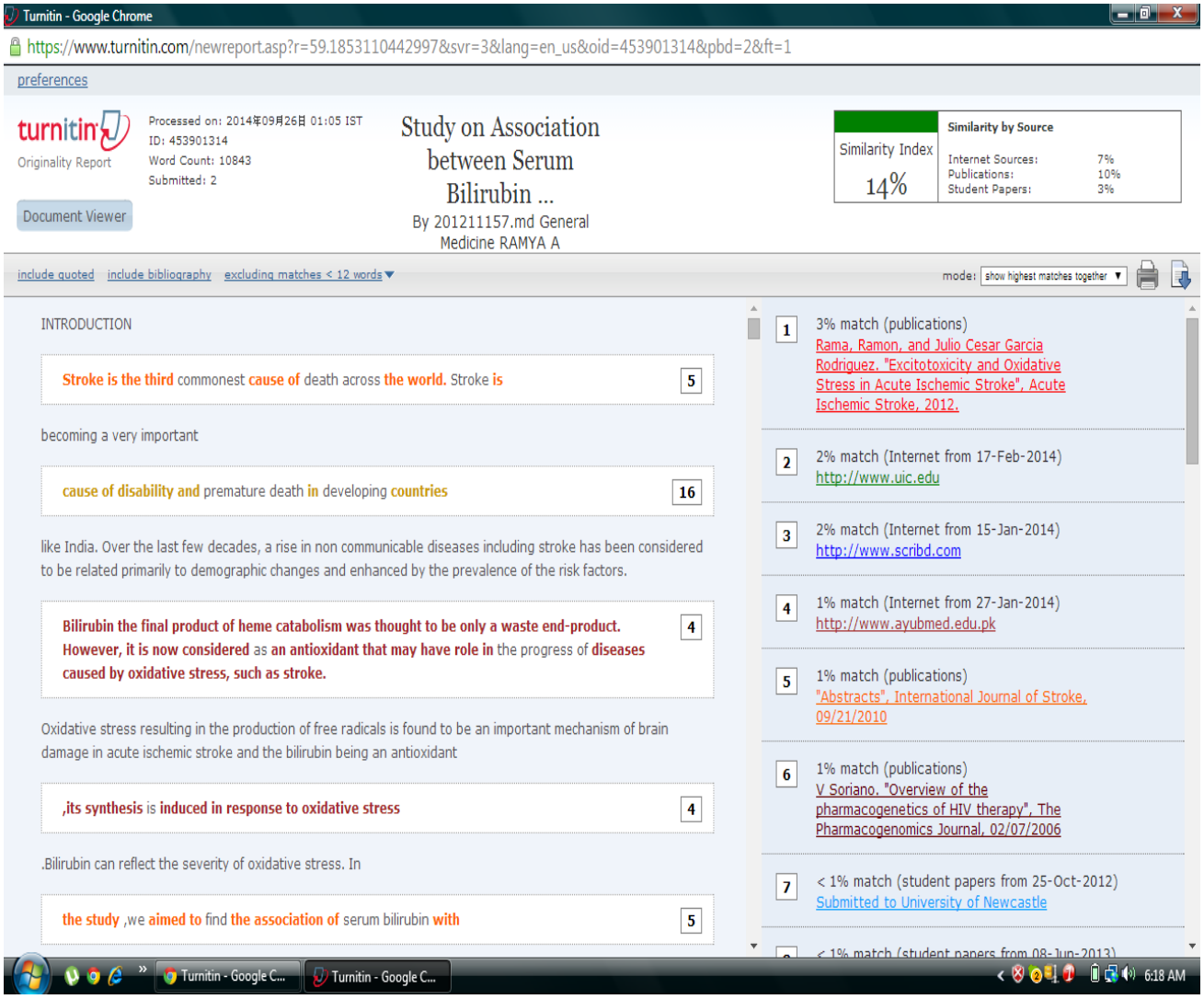
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.Bilirubin can reflect the severity of oxidative stress. In
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ABSTRACT

BACKGROUND

Bilirubin as a marker of oxidative stress can be increased in acute ischemic stroke.

OBJECTIVE

To find any association exists between serum bilirubin level and acute ischemic stroke and assess the usefulness of serum bilirubin in determining the severity and prognosis of stroke.

METHODS AND MATERIALS

Bilirubin and other biochemical parameters were measured in 50 cases (acute ischemic stroke) and 50 age, sex, comorbid conditions matched controls. NIHSS score was assessed at admission and MRS score was assessed after 7days of stroke. Serum total bilirubin levels were divided into 3 groups <0.6mg/dL,0.7-0.9mg/dL,≥1.0mg/dL. NIHSS score was divided into two groups ≥10(Severe Stroke) and <10. MRS score was divided into two groups <3(Good Outcome) and ≥3(Poor Outcome). The bilirubin level and its association with acute ischemic stroke and its correlation with stroke severity and prognosis was analyzed.

RESULTS

The level of serum total bilirubin and indirect bilirubin was significantly higher in acute ischemic stroke patients (p value < 0.001 and <0.001 respectively) than in the control group. The level of serum direct bilirubin didn't show any significant correlation. The level of serum total bilirubin (p value 0.0003 and 0.0002 respectively) and indirect bilirubin (p value 0.003 and 0.001 respectively) was significantly correlated with NIHSS ≥ 10and MRS ≥ 3.

CONCLUSION

In this study it was found that serum levels of total and indirect bilirubin were increased after acute ischemic stroke. Both serum total bilirubin and indirect bilirubin can reflect the severity and prognosis of stroke.

KEYWORDS:BILIRUBIN,ACUTE ISCHEMIC STROKE,SEVERITY, PROGNOSIS.

INTRODUCTION

Stroke is the third commonest cause of death across the world. Stroke is becoming a very important cause of disability and premature death in developing countries like India.

Over the last few decades, a rise in non communicable diseases including stroke has been considered to be related primarily to demographic changes and enhanced by the prevalence of the risk factors.

Bilirubin the final product of heme catabolism was thought to be only a waste end-product. However, it is now considered as an antioxidant that may have role in the progress of diseases caused by oxidative stress, such as stroke.

Oxidative stress resulting in the production of free radicals is found to be an important mechanism of brain damage in acute ischemic stroke and the bilirubin being an antioxidant,its synthesis is induced in response to oxidative stress .Bilirubin can reflect the severity of oxidative stress.

In the study ,we aimed to find the association of serum bilirubin with acute ischemic stroke.

Various studies conducted during the acute phase of ischemic stroke found inverse relationship between serum bilirubin and positive outcomes in stroke patients and bilirubin can act as marker of oxidative stress.

Yun Luo et al (2012) reported that both direct bilirubin and total bilirubin can reflect the severity of ischemic stroke.

Sandra Pineda et al (2008) reported an association between higher direct bilirubin on admission and greater stroke severity[1].

AIM AND OBJECTIVES OF THE STUDY

1. To study the association of serum bilirubin with acute ischemic stroke.
2. To assess the usefulness of serum bilirubin in determining the severity and prognosis of ischemic stroke.

REVIEW OF LITERATURE

BILIRUBIN

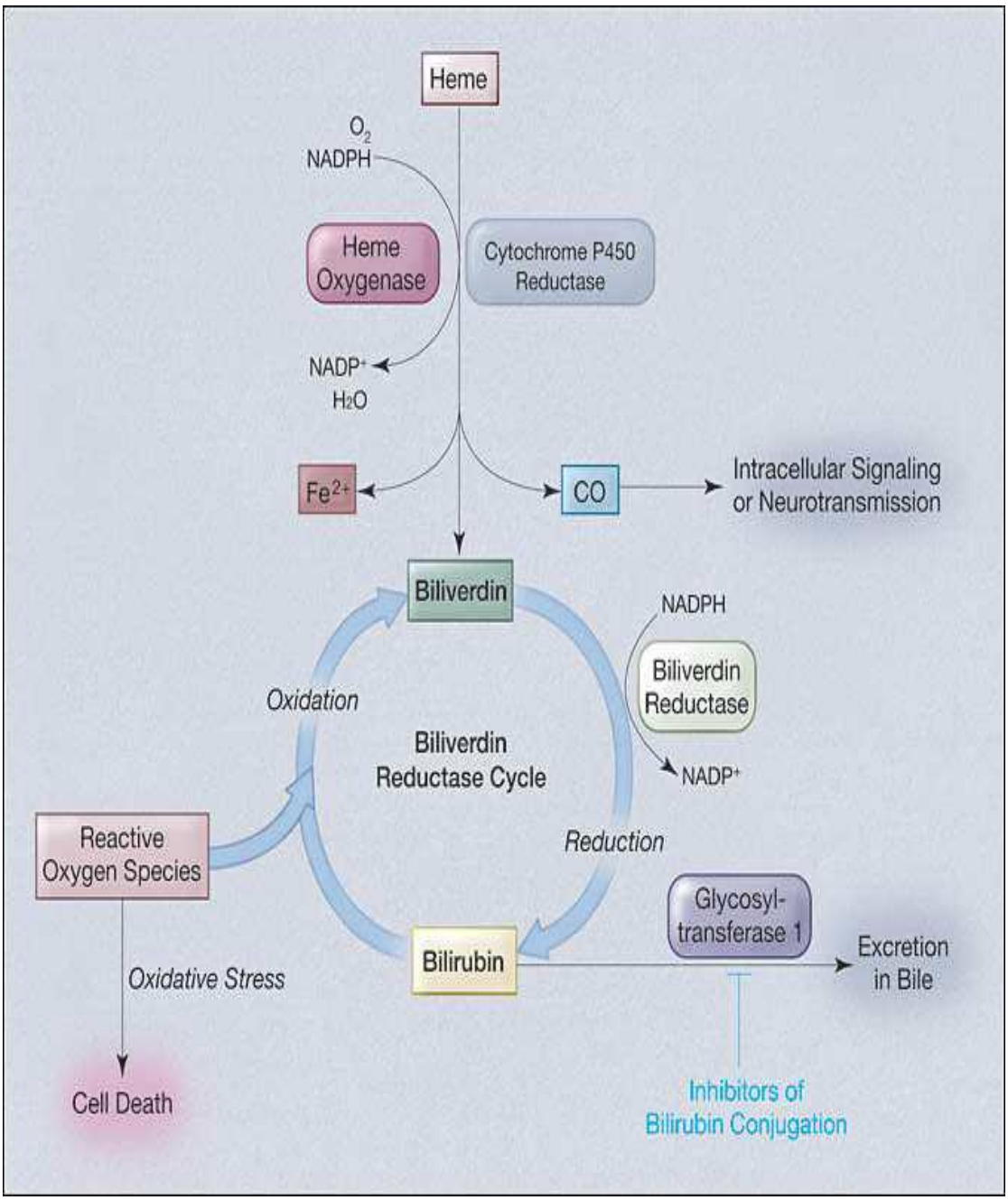
Formation

Bilirubin is formed by the breakdown of heme present in various forms which includes hemoglobin, myoglobin, peroxidase, catalase, tryptophan pyrrolase and cytochromes. Of these hemoglobin is the major source of bilirubin of around 80%[56].

The catabolism of heme is carried out by the complex enzyme system called hemeoxygenase. By the time the heme reaches this oxygenase system, the iron got oxidized to ferric form constituting hemin. In this system heme is formed again by reduction with NADPH. Oxygen is added to the alpha methylene bridge present between pyrroles I and II of the porphryin with the help of NADPH. Similarly ferric iron is formed by the oxidation of ferrous iron. On consequent addition of oxygen, there will be production of ferric iron, CO and also biliverdin which results from the splitting of tetrapyrin ring.

In mammals bilirubin is formed by the reduction of methyne bridge present between pyrrole III and IV in biliverdin with the help of the enzyme biliverdin reductase.

FIGURE 1: *BILIRUBIN FORMATION*



Conjugation

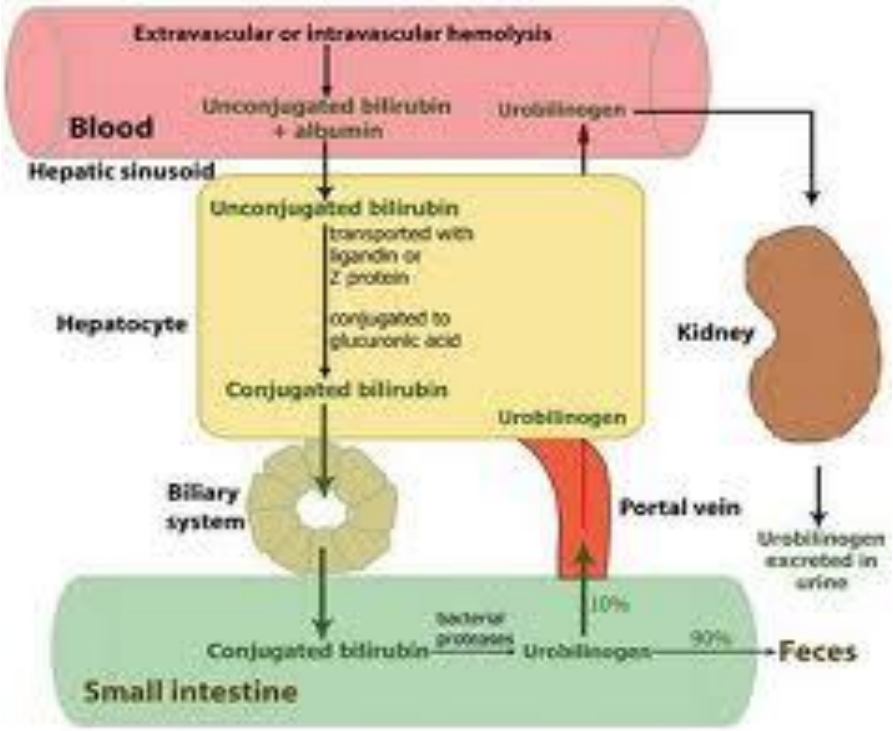
Bilirubin is non polar and is being converted to polar form in hepatocytes by the addition of glucuronic acid and this is called conjugation.

Conjugation can use polar molecules other than glucuronic acid (ex:sulphate). This process is catalyzed by specific glucuronosyl transferase(B-UGT) present chiefly in endoplasmic reticulum which uses UDP-glucuronic acid as the glucuronosyl donor. Bilirubin monolglucuronide is an intermediate and gets converted to bilirubin diglucuronide which form the major form of bilirubin in bile, whereas in pathological conditions bilirubin monoglucuronide are the predominant form in plasma. Bilirubin-UGT can be induced by number of drugs. Bilirubin is secreted into the bile by an active transport mechanism which is the rate limiting step is hepatic bilirubin metabolism. This process is mediated by the MRP-2 (Multidrug resistance like protein-2). The transport of conjugated bilirubin is inducible by the same drugs that can induce the conjugation of bilirubin.

Conjugated bilirubin is reduced to urobilinogen by intestinal bacteria. On reaching the terminal ileum and the large intestine, the glucuronide present in the conjugated bilirubin is removed by specific

bacterial enzyme (Beta-Glucuronidases) and is subsequently reduced by fecal flora to colorless tetrapyrrole compounds called urobilinogens.

FIGURE 2: *BILIRUBIN METABOLISM*



A small fraction of urobilinogen is reabsorbed in the terminal ileum and the large intestine and re-excreted through the liver which is called as called enterohepatic urobilinogen cycle .Under pathological conditions, like liver disease or excess bilirubin production, urobilinogen may be excreted in urine. Most of the urobilinogens in the colon are oxidized to urobilins colored one and are excreted in the feces .Oxidation of residual uribilinogen to urobilins cause darkening of feces on standing in air.

STROKE EPIDEMIOLOGY

Stroke incidence and also mortality are increasing as a result of modernization and increased life expectancy. Worldwide, each year 15 million people suffer from stroke[2]. Of those one third die and one third are left permanently disabled.[3]

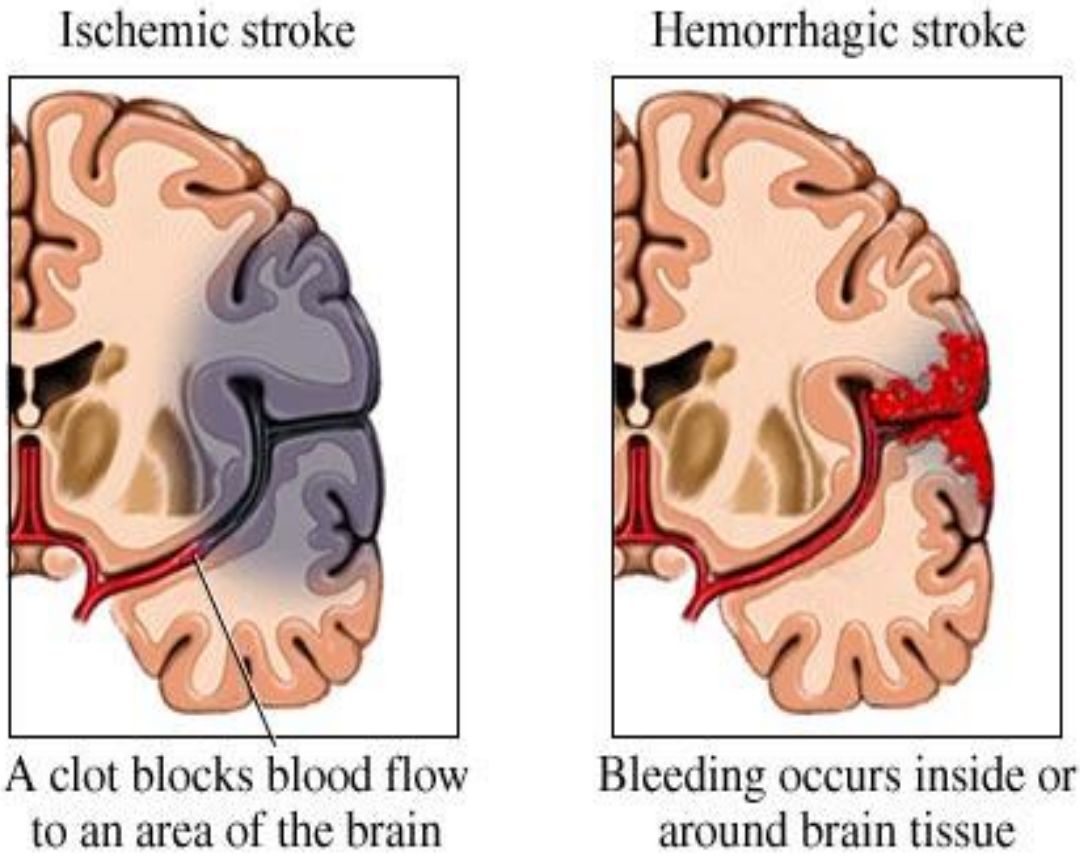
In developing countries there is decreasing trend of infectious and malnutrition related diseases, whereas stroke incidence is increasing in recent decades as a result of dietary changes, decreased physical activity, and increased tobacco use.

It is estimated that by 2040, in low and middle income countries around billion adults aged 65 years or older will be at risk for stroke[4]. In addition to the age, hypertension and tobacco use are the major risk factors worldwide.

MAJOR TYPES OF STROKE

Stroke occurs as a result of disruption of blood flow to a part of brain either because of blood vessel occlusion as in acute ischemic stroke (AIS) or blood vessel rupture causing bleeding either into the brain (Intracerebral hemorrhage-ICH) or around the brain (subarachnoid hemorrhage -SAH).

FIGURE 3: *ISCHEMIC STROKE & HEMORRHAGIC STROKE*



STROKE DEFINITIONS

Stroke is defined as the clinical syndrome of rapid onset of cerebral deficit (usually focal) lasting more than 24 hours or leading to death, with no apparent cause other than a vascular one.

Completed stroke is defined as the deficit which becomes maximal within 6 hours.

Stroke-in-evolution is defined as the progression of clinical symptoms and signs over first 24 hours.

In minor stroke patients usually recovers without significant neurological deficit within a week.

Transient ischemic attack (TIA) don't cause permanent brain damage and symptoms resolves spontaneously.

In TIA neurological symptoms lasts less than 24 hours, but the duration of most TIAs is between 5 and 30 minutes.

TIA is a warning sign indicating that a stroke may occur at any time consequently.

RISK FACTORS OF STROKE

TABLE 1: *NON MODIFIABLE AND MODIFIABLE RISK FACTORS OF STROKE* [5]

Non Modifiable	Modifiable- well documented
Gender Age Genetics Race/ethnicity Low birth weight	Cigarette Smoke Physical Activity Exposure to Poor diet Diabetes Hypertension Dyslipidemia Atrial fibrillation Carotid artery stenosis Postmenopausal hormone therapy Sickle cell disease

TABLE 2: *ISCHEMIC STROKE - ARTERIAL ETIOLOGIES*[6]

<table>
<tr><th>1. Thrombosis</th><th>2. Embolism</th><th>3. Luminal Obstruction</th></tr>
<tr><td>Atherosclerotic plaque
Lipohyalinosis of small vessel
Tumor invasion
TTP/DIC
Antiphospholipid antibody syndrome
Sickle cell disease</td><td rowspan="2">Cardioaortic
Cardiac thrombus
Cardiac vegetations
Cholesterol
Tumor
Artery-to-artery
Atheroma fragments
Decompression illness
Paradoxical
Amniotic fluid
Deep venous thrombus fragments
Cholesterol
Air</td><td rowspan="2">Vasculitis
Vasospasm
Subarachnoid hemorrhage
Meningitis
Drug-induced
Extrinsic artery compression
Masses
Herniation
Non inflammatory vasculopathy
Sickle cell disease
Migraine
Burger’s disease
Fibromuscular dysplasia
CADASIL
Moyamoya disease
Angiotrophic lymphoma
Lymphomatoid granulomatosis</td></tr>
<tr><td>4.Systemic Hypoperfusion
Massive MI
Shock
Cardiac arrhythmia
Severe hypotension
Hyperviscosity syndrome</td></tr>
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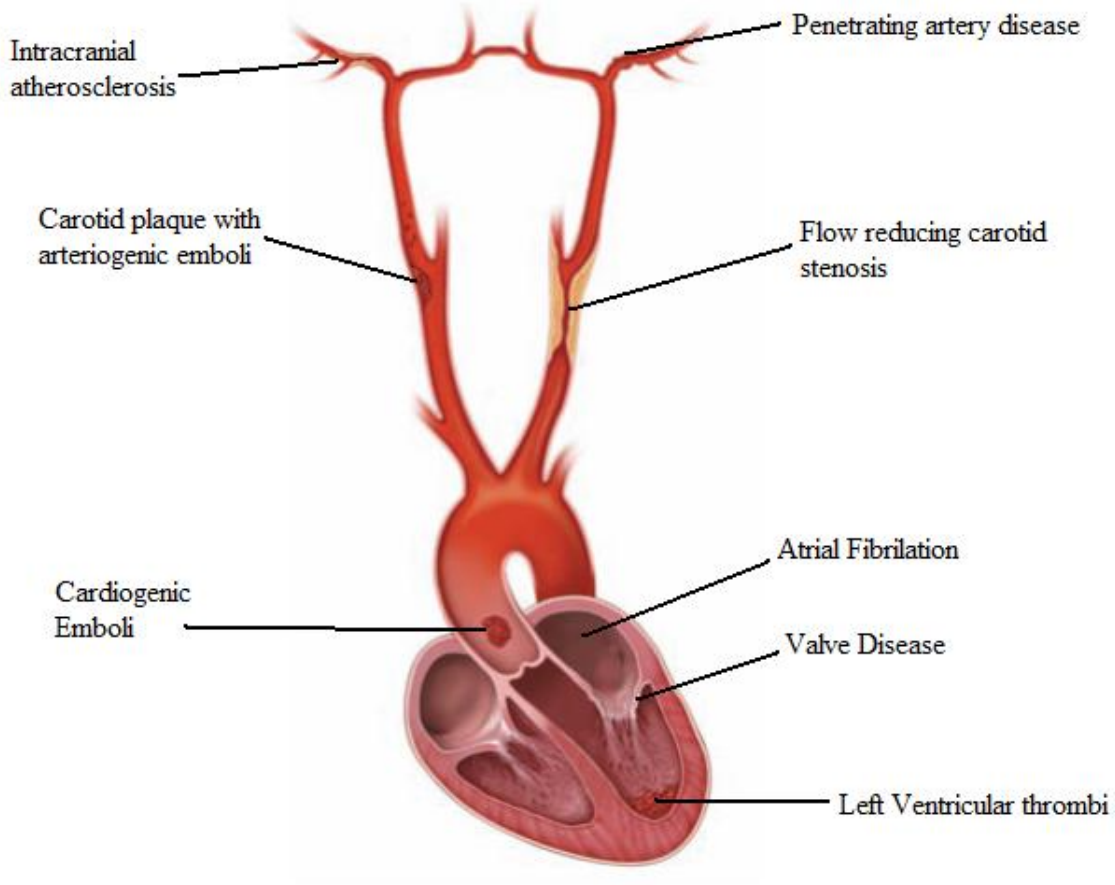
FIGURE 4: *MECHANISMS OF VESSEL OBSTRUCTION IN ISCHEMIC STROKE*[7]



1. Embolus arising from a distant site causing occlusion of intracranial vessel E.g. from sources such as carotid atherosclerotic plaque or atrial fibrillation.
2. Thrombosis of an intracranial vessel in situ, mainly in the small penetrating arteries.
3. Stenosis resulting in flow reduction of either intracranial or extracranial vessels mainly leading to watershed infarct.

ISCHEMIC STROKE - PATHOPHYSIOLOGY

Ischemic stroke occurs chiefly by 3 mechanisms which include:

- Thrombosis
- Embolism and
- Hypotension (global ischemia).

But all ischemic strokes need not fall into these 3 categories, there are number of other mechanisms causing ischemic stroke. However, the most infrequent causes of stroke are those caused by vasospasm (migraine, following SAH, hypertensive encephalopathy) and some form of "arteritis".

THROMBOSIS

Atherosclerotic lesion is the most common pathological form of vascular obstruction causing thrombotic stroke[8]. Ulcerations, thrombosis, calcifications, and intra-plaque hemorrhage are the secondary changes that can occur in plaque. The plaque structure, consistency and composition usually determine the tendency of the plaque to get disrupted or ulcerated.

Disruption of endothelium that can occur in the setting of any one of these pathological changes can result in a complicated process and activation of many destructive vasoactive enzymes.

Adhesion and aggregation of platelets to the vessel wall occurs resulting in the formation of small nidi of platelets and fibrin. Within one hour of the event, leucocytes at the site initiate an inflammatory response[9,10].

Thrombotic occlusion of a vessel can occur in pathological conditions other than atherosclerosis which include clot formation due to hypercoagulable state, arteritis (Giant cell and Takayasu), fibromuscular dysplasia and vessel wall dissection.

In contrast to large artery occlusion , occlusion of deep penetrating arteries that are 100 to 400 mm in diameter results in lacunar infarcts . The commonly affected sites are pons, basal ganglia and internal capsule.

Lacunar infarct sizes only about 20 mm in diameter. The small arteriole elongates most frequently because of chronic hypertension and becomes tortuous and undergoes subintimal dissections and micro-aneurysms which increase the susceptibility of arteriole to occlusion from micro-thrombi.

Lipohyalinosis resulting from fibrin deposition is the underlying pathological mechanism of lacunar infarct.

EMBOLISM

Embolic stroke (ES) results from dislodging of embolus from variety of sources in the central circulation.

Apart from atheromatous plaque other sources of embolus in the central circulation are fat, air, metastasis, foreign bodies and bacterial clumps.

The most frequent affected sites of emboli are superficial branches of cerebral and cerebellar arteries. Since 80% of the blood carried by the large arteries in the neck goes to the middle cerebral arteries, emboli lodges commonly in the middle cerebral artery distribution[11].

The most important sources of emboli are the cardiac chambers (left side) and large arteries (e.g. thrombus from the internal carotid artery).

Embolus acts as a vascular irritant and causes vasospasm which determines the outcome of the stroke in addition to vascular territory which gets affected. The vasospasm need not be limited to the site where the embolus seats, it can also affect the whole arterial tree.

Vasospasm is more common in young individuals comparing with elderly since they have pliable, less atherosclerotic vessels.

Most of the embolic strokes turn in to hemorrhagic infarction (HI). The pathogenesis of this hemorrhagic transformation of an infarct is a composite phenomenon which includes

1. During ischemia both the brain parenchyma and the blood vessels are injured. When the embolus either lyses automatically or breaks and moves distally, cerebral blood flow is restored to the ischemic microcirculation. This can result in a "red or hemorrhagic infarct, whereas poorly perfused are referred to as "pale" or "anemic infarcts."
2. Persistent occlusion can also results in bleeding, which indicates that hemorrhagic transformation need not always associated with migration of embolus. HI on the periphery of infarcts is caused by reperfusion from the leptomeningeal vessels forming collateral circulation which can reperfuse the ischemic area even when the main vessel is persistently obstructed.

In embolic stroke both hemorrhage and ischemia occurs together. Hemorrhagic transformation[12] of the infarct depends upon

- Size of the infarct,
- Collateral circulation richness, and
- Use of anticoagulants and thrombolytic agent.

Large infarctions are associated with a greater incidence of hemorrhagic transformation.

GLOBAL-ISCHEMIC OR HYPOTENSIVE STROKE

Hypotensive stroke is caused by the marked reduction in systemic blood pressure due to any cause. The susceptibility of neurons to ischemia is not uniform. The pyramidal cell layer of the hippocampus, the Purkinje cell layer of the cerebellar cortex and cerebral gray matter are most vulnerable. This is because of the abundance of glutamate in these neurons makes them more susceptible to ischemia.

“Boundary zone” or “Watershed area” is the area between the territories of the major cerebral and cerebellar arteries and this is the common site affected by the global ischemia.

The most commonly affected site is the parietal-temporal-occipital triangle which causes a clinical syndrome consisting of weakness and sensory loss predominantly the arm; the face is not affected and speech is spared. Watershed infarct constitutes 10% of all ischemic strokes and around 40% of these occur in patients having carotid stenosis or occlusion[13].

ISCHEMIC STROKE - PATHOGENESIS AT CELLULAR LEVEL

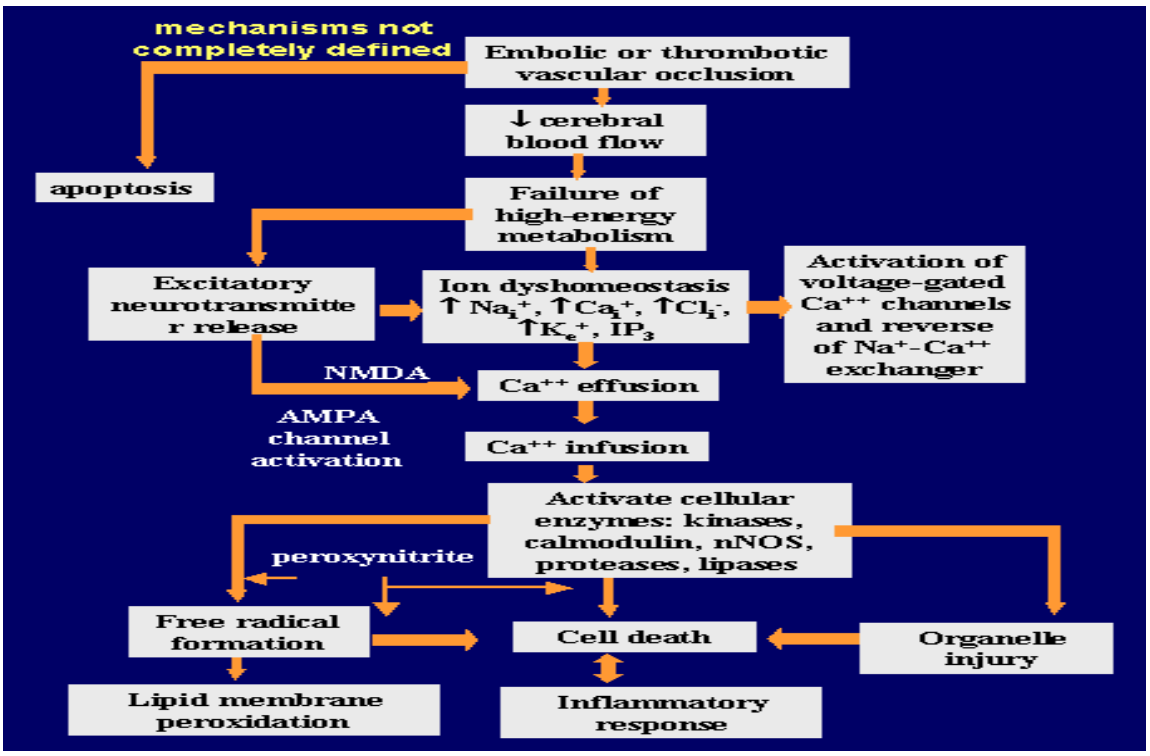
Ischemia causes a sequence of events that ultimately leads to neuronal injury and death irrespective of the mechanism responsible for the vessel occlusion[14]. Blood flow reduction decreases the formation of high energy phosphates.

This energy failure results in membrane depolarization and uncontrolled discharge of excitatory aminoacids, such as glutamate, in to the extracellular space which is called as excitotoxicity. This excitotoxic aminoacid glutamate exerts its action on various receptors e.g. NMDA and AMPA, finally causing calcium overload of neuronal cells This rise in calcium level results in the activation of proteolytic enzymes.

The activated enzymes degrade both intracellular and extracellular structures, and also other enzymes, i.e. cyclooxygenase and phospholipase A2 which can form free radicals. Neuronal NO synthase is calcium dependent enzyme and form nitric oxide, which can react with superoxide generating the highly reactive radical peroxynitrite[15]. Ischemia results in expression of proinflammatory genes so that several inflammatory mediators are released mainly tumor necrosis factor and interleukin 1ß.

In addition, adhesion molecules are also expressed which results in binding of neutrophils, monocytes and macrophages with endothelium causing microvascular occlusion and the blood cells crosses the vessel wall and penetrates in to the brain substance and exerts their inflammatory actions. The inflammatory cells can also form free radical.

FIGURE 5: *PATHOGENESIS OF ISCHEMIC STROKE*



Although excitotoxicity mainly leads to necrosis, there is evidence of apoptosis after cerebral ischemia and it has been proposed that both necrosis and apoptosis are triggered in parallel during ischemia and that the predominance of one mechanism will be determined by specific conditions.

STROKE SYNDROMES

Anterior Cerebral Artery Syndromes

Medial portion of the frontal and parietal lobes are supplied by anterior cerebral artery. Infarction of these areas results in contralateral hemianesthesia and hemiparesis that affects the leg more than arm/face due to the topographic arrangement of the homunculus.

In addition damage to the medial part of frontal lobe results in impairment of behavioral and executive functions which can cause abulia[6].

Anterior cerebral artery infarcts in dominant hemisphere may produce mutism, and whereas in nondominant hemisphere results in acute confusional state.

In bilateral ACA infarcts, severe abulia can be present as akinetic mutism along with bladder incontinence.

Middle Cerebral Artery Syndromes

The remaining frontal and parietal lobes, are supplied by middle cerebral artery which also supplies the superior part of the temporal lobe.

Stroke affecting the complete territory results in contralateral hemiparesis, hemianopia and hemianesthesia. Ipsilateral gaze preference with attention related frontal lobe dysfunctions results.

In dominant MCA lesions, impairment of language functions occurs resulting in motor aphasia with lesions at Broca's area which is present in the posterior-inferior portion of frontal lobe and as sensory aphasia with lesions at Wernicke's area which is present in the posterior-superior portion of temporal lobe.

Injury in the corresponding areas in the nondominant hemisphere results in subtle symptoms of language dysfunction in form of motor and sensory aprosodia. There is duplicative damage to sensory, motor, executive and language functions areas which occurs in total proximal MCA occlusions , by damaging both cortical representations and basal ganglia structures.

Lesions of the MCA in the distal part or at the level of bifurcation can allow blood flow in the lenticulostriate arteries sparing the internal capsule and basal ganglia. In this type of lesion the pattern of motor and sensory deficits may be incomplete and irregular and, sparing the leg function especially because of the topographic arrangement of homunculus [6].

Occlusion of superior division of MCA results in syndrome of frontal lobe dysfunction, with prominent motor language deficits with variable degree of sensory loss. Whereas in inferior division of MCA lesion sensory language deficit and hemianopsia results. Gerstmann

syndrome includes right-left confusion, acalculia, agraphia and finger agnosia which results from lesion in angular gyrus area.

Posterior Cerebral Artery Syndromes

The inferior temporal lobe and occipital lobe are supplied by posterior cerebral artery. The posterior cerebral artery lesions that don't involve early arterial branches to deep structures cause contralateral homonymous hemianopia. Alexia without agraphia is seen in dominant hemisphere lesion. In this conditon reading is impaired by the combination of

1. Impaired connection between the receptive language area and contralateral visual field and

2. Unilateral visual field defect

This occurs as a result of infarction of the fiber tracts passing posteriorly through the corpus callosal splenium.

Anton syndrome is characterized by confabulation, and cortical blindness as a result of damage to bilateral occipital lobes.

The combination of

1. Optic ataxia

2. Occulomotor apraxia and

3. Simultagnosia

is caused by bilateral PCA infarcts that affect the posterior parietal lobes. This condition is known as Balint syndrome.

TABLE 3: *LACUNAR SYNDROMES*[16]

LACUNAR SYNDROMES		
Pure Motor	Posterior limb of internal capsule or thalamus	Contralateral hemiparesis
Pure sensory	Posterior limb of internal capsule or thalamus	Contralateral hemisensory loss,,
Sensory motor	Posterior limb of internal capsule or thalamus	Contralateral hemisensory loss,hemiparesis
Ataxic hemiparesis	Pons,basal ganglia,internal capsule,Corona radiata,	Contralateral hemiparesis with ataxia
Hemiballismus	Subthalamic nucleus lesion	Contralateral hemiballismus
Dysarthria–clumsy hand	Pons,basal ganglia,internal capsule,corona radiate	Contralateral upper limb ataxia and dysarthria
Dejerine-Roussy	Thalamus	Contralateral hemibody pain with hemisensory loss

DIAGNOSIS OF STROKE:

CT Scan

CT scan images is used to identify or exclude hemorrhage as the cause of stroke. CT scans may not detect an infarction in the first 24 to 48 hours and appears normal in significant percentage .The main disadvantage of CT scan are small infarcts, infarcts in the posterior fossa may be easily missed because of bone artifact; small infarcts on the cortical surface may also be missed[7].

CT scans with contrast enhancement gives more details by enhancing subacute infarcts and allow clear visualization of venous structures .

CT angiography (CTA) can be performed with administration of iodinated contrast which is being coupled with newer multidetector scanners thus iodinated contrast allows better visualization of the cervical and intracranial arteries, and also intracranial veins. In this method, in one imaging session, aortic arch, and coronary arteries and intracranial veins can be visualized .

The ischemic penumbra can be detected by contrast study which delineates the area at risk of infarction surrounding the infarction. CT without contrast is the modality of choice in acute stroke patients because of its availability and speed, and CT perfusion imaging is also an useful adjunct.

TABLE 4: *CT SCAN FINDINGS*[17]

Time of Infarct	Findings
Hyperacute stage :within 12 hrs of onset of stroke	• 50-60% of patients shows no abnormal findings in this stage. • Presence of dense MCA sign. • Lenticular nucleus obscuration. • Insular ribbon sign. • Grey-white interfacement loss.
Acute stage:last 12 to 24 hrs	• In this stage basal ganglia will be hypodense. • Sulcal effacement.
Days :1day to 7 days	• Mass effect. • Wedge-shaped hypodense area in gray and white matter. • Hemorrhagic Transformation.
Weeks :1-8	• Resolution of mass effects. • Persistence of contrast enhancement
Months to years	• Encephalomalacia . Volume loss

MRI Scan

MRI clearly shows the location and extent of infarction in all areas, including cortical and posterior fossa structure .Regarding intracranial bleed it can identify but is not as sensitive as CT in the diagnosis of hemorrhagic stroke. More reliable and highly précised images can be obtained using higher field strength magnets. Diffusion-weighted imaging modality is more sensitive than CT for early brain infarction or standardMRI[7].

MR perfusion imaging can be done using gadolinium contrast. Areas with poor perfusion but appearing normal in diffusion sequence are considered as ischemic penumbra. Patients having large regions of this discrepancy may be taken for acute revascularization procedures. MRA is more sensitive in the detection of stenosis of extracranial and intracranial parts of internal carotid arteries. Comparing with conventional x-ray angiography, MRA overestimates the of stenosis severity. Extracranial or intracranial arterial dissection can be visualized by an sequence named as MRI with fat saturation. This technique detects even the clotted blood in the vessel wall .

The disadvantage of MRI are time consuming, cost ineffectiveness, less availability and more than these insensitive in detecting blood products comparing with CT. Claustrophobia is also an considerable disadvantage.

Most of the stroke protocols suggest CT because of these issues. But for clear description of extent of tissue injury after the acute period and distinguishing new from old infarction MRI is superior. In diagnosing TIA it has considerable significance .It is highly sensitive in detecting new infarction, which is a strong predictor of stroke occurrence subsequently.

TABLE 5: *MRI FINDINGS*

Time of Infarct	**Findings**
Immediate	Hyperintense on DWI. Contrast enhancement. Alterations in perfusion.
<12 hrs	Gyral edema, Sulcal effacement. Loss of gray-white interfaces (T1).
12 to 24 hrs	Hyperintensity (T2). Enhancement of meninges adjacent to infarct. Mass effect.
1 to 3 days	Enhancement of meninges begins to decline, Hemorrhagic Transformation Signal abnormalities on T1WI, T2WI.

Cerebral angiography

Conventional x-ray cerebral angiography is considered as the imaging modality for diagnosing atherosclerotic stenoses and also other vascular pathologies such as vasculitis, vasospasm, aneurysms, fibromusculardysplasia, arteriovenous fistula, intraluminal thrombi, and collateral channels.

Endovascular techniques may be used in performing balloon angioplasty , deploying stents ,treating aneurysms by embolization, and also in opening occluded vessels with mechanical thrombectomy devices during acute stroke.

In Conventional angiography there are risks of arterial damage, embolic stroke, groin hemorrhage, and renal failure.So it should be reserved where less invasive techniques are inadequate.

Ultrasound techniques

B-mode ultrasound image with a Doppler ultrasound can detect and quantify the stenosis present in the extracranial part of internal carotid artery especially at its origin. Transcranial Doppler (TCD) can be used in assessing flow in main cerebral arteries and also detecting stenosis.In addition TCD can assist thrombolysis and rtPA administration . MR angiography can be combined with transcranial and carotid ultrasound.

Perfusion techniques

Cerebral blood flow can be quantified by PET and xenon techniques but these are not usually applied in clinical practice, being used only in research purposes.

MR perfusion techniques and Single-photon emission computed tomography (SPECT) are other perfusion techniques which detects relative cerebral blood flow.

COMPLICATIONS OF STROKE[5]

- ✓ Urinary tract infection
- ✓ Aspiration Pneumonia
- ✓ Bed sores
- ✓ Deep vein thrombosis
- ✓ Hypoxemia
- ✓ Hyperglycemia
- ✓ Hyponatremia and seizures
- ✓ Constipation
- ✓ Dehydration
- ✓ Frozen Shoulder and subluxation
- ✓ Contractures

TREATMENT OF ISCHEMIC STROKE

Stroke is an emergency condition irrespective of severity of neurological dysfunction.The priority should be given as to MI and serious trauma. Stroke management includes general care, specific treatment and treatment of complications.

First step in the management of stroke is confirmation of diagnosis as there are various mimics exists for stroke which includes

- Seizure
- Migraine with aura
- Hypoglycemia
- Wernicke's encephalopathy
- Hypertensive encephalopathy
- CNS tumor ,CNS abscess
- Drug toxicity and
- Psychogenic

GENERAL MANAGEMENT OF STROKE

- Fever and glycemic control
- Blood pressure management
- Fluid management
- Treatment of underlying etiology.

Blood Pressure Control[17]

If the systolic BP is between 185-220 mmHg or diastolic BP is between 105-120 mmHg no need of introducing antihypertensive medications unless there are conditions endangers life coexists which includes

- Acute renal failure
- Acute myocardial infarction/Left ventricular failure.
- Aortic dissection

If there is a plan of starting rtPA therapy BP >185/110 mmHg should be treated.

If the Systolic BP > 220 mmHg, diastolic BP 120-140 mmHg, antihypertensive should be immediately administered which includes sodium nitroprusside, nicardipine,captopril, nitroglycerine and labetalol.

SPECIFIC TREATMENT

- Recanalization
- Anticoagulant
- Aspirin
- Neuroprotective treatment
- Therapeutic hypothermia
- Hemodilution
- Craniectomy
- Rehabilitation

INTRAVENOUS THROMBOLYSIS

Treatment should be started within 3 hours of stroke onset. Treatment by this means can result in complete improvement at 24 hours and complete recovery or near normal at 3 months. The major risk is symptomatic bleeding in brain. But there is no mortality benefit by this treatment. Regimen for IV rt-PA treatment is infusion 0.9 mg/ kg over 1 hour, 10 % as bolus dose over 1 minute. Anticoagulants and antiplatelet agents should not be intiated for first 24 hours of fibrinolysis. This is recommended in the setting of early ischemic changes on CT, irrespective of its extent.

TABLE 6: *IV rt-PA INDICATIONS/CONTRAINDICATIONS*[7]

Indication	**Contraindication**
• Diagnosis of stroke clinically • Duration ≤3 h • CT scan – No hemorrhage or edema of >1/3 of the MCA territory • Age ≥18 years • Patient or surrogates Consent	• Rapidly improving symptoms • Minor stroke • Sustained BP >185/110 mm Hg inspite of treatment • Glucose <50 or >400 mg/dL; Platelets <100,000; HCT <25% • Heparin use within 48 h and prolonged PTT, or elevated INR • Prior stroke or head injury in preceding 3 months, prior ICH • Major surgery in preceding 14 days • GIT bleeding in preceding 21 days • Recent myocardial infarction • Coma or stupor

Antiplatelet

Aspirin (325 mg) should be initiated within 24-48 hours after stroke onset.

Anticoagulant

All patients with atrial fibrillation of non valvular origin and cardiac disease should be given anticoagulation. Anticoagulation is not recommended in preventing early recurrent stroke, or improving stroke outcome,and within 24 hours of treatment with IV rtPA. The contraindications are large infarction,uncontrolled BP,and advanced microvascular changes.

SURGERY

Decompressive evacuation of space-occupying infarction in cerebellum is effective in preventing and also treating herniation which results in brain stem compression.Decompressive surgery is also effective for malignant edema of cerebral hemisphere .

COMPLICATION MANAGEMENT

The major complication includes raised ICT, seizures and hemorrhagic transformation. Raised ICT can be managed by non pharmacologic measures including head end elevation of bed, avoiding hypotonic solution and hypoxia, and hyperventilation. Raised ICT can be

treated pharmacologically by IV mannitol, IV or oral glycerol,but furosemide and steroid is contraindicated. Decompressive surgery is also an option.

MEASUREMENT OF STROKE OUTCOMES

Stroke severity at presentation predicts stroke outcomes . National Institutes of Health Stroke Scale (NIHSS) a measure of stroke-related neurologic deficits, has been studied extensively in various clinical trials and found to be an useful predictor of stroke outcomes. The NIHSS is an excellent scale for clinicians to interpret the severity of a stroke. Physicians and also trained health care professionals caring for patients with strokes can asses this.NIHSSscore, may underestimate the severity of a posterior circulation stroke because most of the variables are related to symptoms of anterior circulation territory.Similarly ,it also underestimate the right middle cerebral artery stroke severity because of language function in left. There are different outcome scales which measure different dimensions of recovery and disability.

- Modified Rankin Scale (mRS) for assessement of functional independence.
- Glasgow Outcome Scale (GOS) assessememt of general level of disability and recovery.
- Barthel Index (BI) assessesment of ability of self-care and mobility.

NIH STROKE SCALE (NIHSS) SCORE[18]

1. Level of consciousness 5 points
2. Best gaze on eye movements 2 points
3. Field of vision 3 points
4. Facial movements 3 points
5. Hemiparesis and hemiplegia in extremities 4 points
6. Each limb is graded individually (4 points for each limb)
7. Ataxia in each limb 2 points
8. Sensation on both sides of the body 2 points
9. Language (presence of aphasia) 3 points
10. Dysarthria 2 points
11. Extinction (formerly 'neglect') (2 points)

Interpretation of NIHSS Score

0 – No stroke

1-4 – Minor stroke

5-15 – Moderate stroke

16-20 – Moderate to severe stroke

21-41 – Severe stroke

MODIFIED RANKIN SCALE[19]

It is the scoring system used for assessing the functional outcome after stroke.

0- patients don't have any symptoms

1- Inspite of symptoms patients don't have significant abnrormality; they can able to do usual daily activities.

2- patient have mild disability; not able to carry out all activities which could be done previously, but able to take care of them without assistance

3- patient have moderate disability; need some help, but can walk without assistance

4- Moderate to severe disability; can't walk without assistance and need help even for self body care.

5- Severe disability; patient will be bedridden, and need constant nursing care

6- Dead

Total score: 0 to 6

OXIDATIVE STRESS

Oxidative stress results from imbalance between the generation of reactive oxygen species and the antioxidant defense[20].

Increased ROS production through the entire course of acute ischemic stroke especially in the initial phase can induce the functional and structural damage of neuronal cells, playing an important role in the pathophysiology of brain injury[21] .

Low antioxidant activity in the plasma is associated with high neurological dysfunction in acute stroke.

Oxidative stress and acute ischemic stroke

High metabolic activity and oxygen consumption which results in the production of high levels of ROS, along with relatively low levels of endogenous antioxidant enzymes, mainly catalase make the neurons vulnerable to oxidative stress .

ROS reacts with lipids in brain to generate peroxyl radicals resulting in neuron membrane lipid oxidation . The combination of all these results in the increased vulnerability of CNS to oxidative damage.

A decrease in mitochondria redox potential resulting in ROS production from the ETC, mainly at cytochrome III becomes the chief source of free radical generation during ischemia[23-25].

Excitotoxicity after ischemia, results in excess cytosolic free Ca2+. This leads to overloading of the mitochondrial proton circuit, resulting in failure of oxidation along with increased ROS production .

Increased ROS formation in mitochondria leads to the impairment of the ETC, resulting in decreased ATP production, altered calcium homeostasis ,increased formation of free radicals, and mitochondrial dysfunction [22].

In the animal study, transient MCA occlusion results in ROS production and mitochondrial dysfunction . Over-expression of mitochondrial Hsp70/Hsp75 or antioxidant treatment, resulting in decrease ROS concentration attenuates mitochondrial dysfunction.

Excitotoxic pathways other than mitochondrial dysfunction are also important in inducing oxidative stress. It has been proposed that the primary source of superoxide synthesis following neuronal NMDAR activation is NADPH oxidase,which are transmembrane proteins involving in the transport of electrons across biological membranes. Usually, oxygen is the electron acceptor and O2−. is the product of the

electron transfer reaction . Thus the function of NOX enzymes is the production of ROS.

During the acute phase of stroke, nNOS gets activated as a result of high influx of Ca2+ through NMDA receptors resulting in the increased production of NO . Neuronal NO synthase (nNOS), enzyme is tethered to the NMDA receptor complex by a protein called postsynaptic density protein-95 (PSD95) .

During stroke, there is dramatic increase in nitric oxide production in the brain because of the increased activity of neuronal and inducible isoforms of NO synthases .

Peroxynitrite (ONOO−) and OH- are formed as a result of combination of NO with H2O2 and O2- which strongly contributes to brain damage during ischemia.

After ischemia ,NO-induced ONOO−causes mitochondrial dysfunction and subsequent increased production of free radicals leading to dysfunction of cellular membranes resulting in necrosis.

The ROS formed in the electron transport chain can react readily with nitric oxide to form highly reactive peroxynitrite, resulting in the damage of lipids, proteins and DNA [24-25].

It is considered that the mitochondrial Ca2+ overload, with generation of free radicals, and depression of energy metabolism are most important in the pathogenesis of ischemic brain damage , whereas the role of NO is also equally important.

In a study, after the occlusion of middle cerebral artery, the nitric oxide concentration in that ischemic area increased to micromolar levels. The gush in NO levels could be inhibited by glutamate receptor antagonists. It is suggested in the study that , increased nitric oxide metabolite in CSF was associated with greater brain injury and early deterioration of neurological function.

The H2O2 formed from O2− gets converted to hydroxyl radical (OH-) by the Haber-Weissreaction which is favoured by iron ions in Fenton reaction or in to water by oxidation of the small tripeptide glutathione (GSH) by means of a reaction which is catalyzed by the glutathioneperoxidase (GPX) or mutated to water and oxygen by catalase enzyme [28].More GSH is produced by the reduction of the oxidized glutathione (GSSG) by a reaction catalyzed by the enzyme glutathione reductase .GSH converts moreH2O2 to H2O . Superoxide, causes greatest oxidative damage because of its participation in peroxinitrite (ONOO−) formation[28].

Oxidative/nitrosative stress can lead to the damage of different cellular components by oxidation of membrane lipids , cell proteins and DNA and also initiates cascade reactions, resulting in mitochondrial dysfunction and caspases activation and also the activation of signal transduction pathways , finally leading to neuronal death. .

Result from various studies concluded that oxidative stress plays an important role in pathogenesis of ischemic stroke.

Mosher Muhammad Hussein Kossi et al concluded that oxidative stress is an important event in thrombotic stroke and may have unfavorable effect in stroke outcome[32].

Ayaka Ozkul et al 2007 proposed the harmful effects of oxidative stress in the outcome of acute ischemic stroke[31].

Jaspreet Kaur et al 2011concluded that oxidative stress contributes to the pathogenesis of acute ischemic stroke and TIA and also the imbalance between the oxidant and antioxidant may contribute to the severity of stroke[46].

Nai-Wen Tsai et al, 2014 concluded that large-vessel disease have higher oxidative stress but less antioxidant defense than small-vessel disease[33].

BILIRUBIN AS MARKER OF OXIDATIVE STRESS

Kazuhiro Utani 2001 et al proposed bilirubin metabolites in urine may act as a marker of oxidative stress in septic patients[30].

Mehmet Davutoglu et al 2008 concluded that bilirubin had significant positive correlation with MDA and NO and negative correlation with anti-oxidant enzyme activities[31].

Kenji Dohi 2003 proposed bilirubin levels serve as an useful marker of oxidative stress in patients with hemorrhagic stroke[43].

Nesrine salah el din abdul hamim et al (Cairo university 2001) concluded that bilirubin level increases as a response to oxidative stress and contributes to plasma antioxidant property[34].

ANTIOXIDANT ROLE OF BILIRUBIN

Various studies found that different forms of bilirubin are powerful antioxidants: Unconjugated ,conjugated, free and albumin-bound bilirubin were all found to be effective scavengers of peroxyl radicals .They are able to protect LDL against peroxidation [57].

Under physiological conditions, bilirubin may acts as a potent lipid chain-breaking antioxidant so that increased concentrations of

plasma bilirubin may reduce the formation of atheromatous plaque by the prevention of formation of oxidized LDL.

Various animal and human studies have concluded that bilirubin is a physiological antioxidant.

Yamaguchi and co-workers identified biotripyrrins (oxidative metabolites of bilirubin) in the urine of healthy humans and ascorbic acid depleted rats treated with endotoxin[35].

In the same study on feeding a documented physiological antioxidant , ascorbic acid, secretion of bilirubin metabolites was reduced and also there was suppression of the endotoxin-stimulated concentration of HO mRNA in liver.

In another animal study , ischemia and reperfusion of rat liver resulted in induction of HO-1and production of biotripyrrins . And there was attenuation of both HO induction and biotripyrrin production on feeding with ascorbic acid in this model.

These results denote that bilirubin serves as a strong physiological antioxidant in ischemia-reperfusion in vivo and protects against oxidative stress.

In an experimental study observed in pig hearts cardiac ischemia followed by reperfusion was associated with accentuated expression of HO-1mRNA and increased reactive vascular HO-1.

Dennery at al experiments using Gunn rats proposed the antioxidant role of bilirubin[47].

Another study of vascular balloon injury resulting in oxidative stress and intimal cell proliferation in rat carotid artery showed the protective role of bilirubin as an antioxidant.

And this study suggested that, increased HO activity and high bilirubin serve a protective role against injury-mediated proliferation of intimal cell .

Various human studies have led to similar conclusions that bilirubin play a role as an antioxidant.

For example, oxidative stress results in depletion of endogenous antioxidants like bilirubin and increased production of lipid hydroperoxides .

Infants with disorders of oxygen radical mediated injury, such as retinopathy of prematurity, intraventricular hemorrhage, bronchopulmonary dysplasia, and necrotizing enterocolitis, shows lower circulating bilirubin comparing with healthy controls .

Similarly, a direct correlation was found between antioxidant status and serum bilirubin concentrations in premature neonates.

BILIRUBIN AND INFLAMMATION

Role of bilirubin in inflammatory processes and immune reactions has also been documented. Nakagami et al. found that both bilirubin and biliverdin inhibit complement-mediated reactions and the administration of biliverdin inhibits Forssman anaphylaxis reaction in guinea pigs. These findings suggest the protective role of bile pigments by its anticomplement activity.

The correlation between inflammatory processes and bilirubin is supported by evidence that augmented activity of heme oxygenase [58]resulting in faster recovery of inflammation whereas attenuated activity of this enzyme resulting in inflammatory response augmentation.

Bilirubin exerts its anti-inflammatory actions by the following mechanisms:

- Decreasing vascular endothelial proliferation by inhibition of NFkB
- Inhibits oxidant-mediated activation of leukocytes
- Anticomplement activity
- Inhibition of leukocyte migration via suppression of VCAM[60].

BILIRUBIN AND ATHEROSCLEROSIS

Bilirubin offers protection against oxidation of lipoproteins and lipids and thereby reducing the formation of atheroma plaque[38,39]. So patients with low bilirubin concentrations may have augmented atherogenic plaque formation as a result of increase in lipids and lipoproteins oxidation[59].

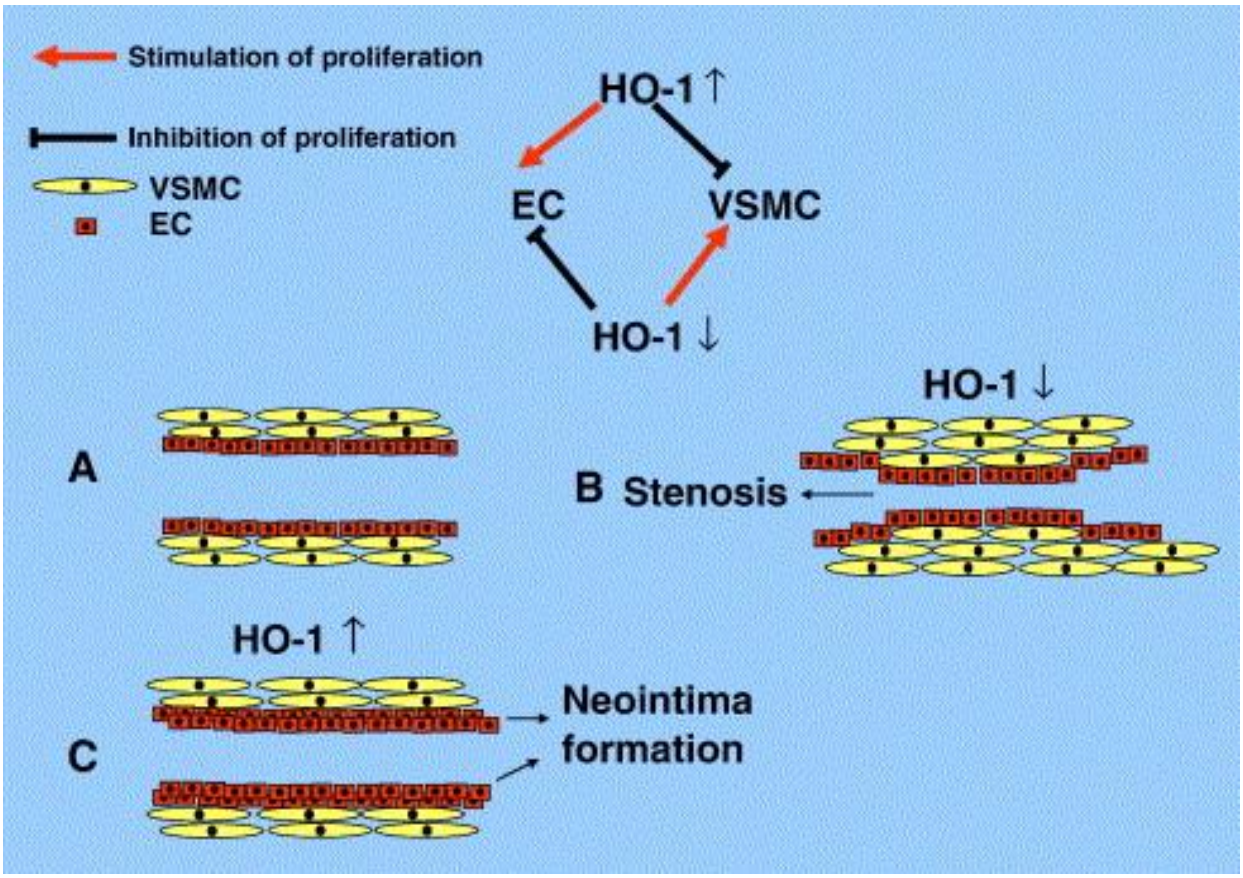
Bilirubin – Heme oxygenase activity

Increased HO activity may account for the antiatherogenic property of bilirubin. This is documented by increased heme oxygenase activity resulting in increased formation of CO, iron, and biliverdin and the pathophysiologyof atherosclerosis could be affected by changes in any one of these three metabolites.

For example, HO-1reduces the heme concentration thereby preventing heme mediated cell injury.

In addition,hemoglobin can act as a scavenger of nitric oxide that affects NO mediated vasodilatation.

FIGURE 6: *ILLUSTRATING ROLE OF HEME OXYGENASE IN STENOSIS AND NEOINTIMA FORMATION*



(A)Normal

(B) Vascular smooth muscle proliferation by low concentration of HO-1 which eventually results in stenosis.

(C) Increased HO-1, by inhibiting vascular smooth muscle proliferation inhibits neointmal formation.

SMOKING AND SERUM BILIRUBIN

Smoking induces oxidation of lipids due to exposure to LDL. It also increases the uptake of modified LDL by macrophages. It has already been established that smoking lowers serum bilirubin concentration in males.

HYPERTENSION AND SERUM BILIRUBIN

Ho Jun Chin et al 2009 concluded, that increase in bilirubin level but within the physiological range had a negative correlation with incidence of hypertension[49]. This effect of bilirubin was more evident in non-smokers and females.

PERIPHERAL VASCULAR DISEASE AND SERUM BILIRUBIN

The NHANES study concluded that higher the serum total bilirubin level lower the incidence of peripheral arterial disease.Whereas patients with low serum bilirubin levels had increased carotid intima-media thickness and also abnormal flow-mediated dilation which are useful in predicting cardiovascular disease in normal individuals.

All these findings gives the conclusion of increased bilirubin levels decrease the risk of acquiring cardiovascular disease in normal subjects.

CORONARY ARTERY DISEASE AND SERUM BILIRUBIN

Multiple studies showed inverse relationship between bilirubin concentration and incidence of coronary artery disease[38-41]. In the Framingham offspring study, it had been found that higher bilirubin levels were associated with decreased risk of acquiring cardiovascular disease in men

LauraJ.Horsfall, et al concluded that lower bilirubin is a risk factor for developing CAD and mortality.

RHEUMATOLOGICAL DISEASES AND SERUM BILIRUBIN

In various studies,inverse relationship was found between serum bilirubin levels and some rheumatological disorders.

- Wegener granulomatosis
- Systemic lupus erythematosus
- Rheumatoid arthritis

There is an inverse association between SLE and serum bilirubin levels. The prognosis of SLE patients is related to the efficiency of antioxidant defense systems. But the low serum bilirubin levels may be caused by the consumption of bilirubin during the pathogenesis of oxidative stress in SLE .This concept might apply for Wegener granulomatosis and Rheumatoid arthritis , where the same results have been found.

DEPRESSION AND SERUM BILIRUBIN

Wai Kwong Tang suggested that the bilirubin level is a novel, important biological marker for the risk of developing depression in ischemic stroke patients[42].

On univariate analysis it was found that more severe stroke was associated with higher bilirubin, reflecting the intensity of oxidative stress in the early phase. Thus severe the stroke ,greater the risk of post stroke depression.

In the study the association found between the bilirubin level and post stroke depression was independent of severity of stroke.So there must be some other possible mechanism for the association between these two.

Among the stroke patients high levels of psychological stress was noted. There are evidence that urine bilirubin metabolites correlates positively with psychological stress.

Thus , like high cortisol, high bilirubin level may denotes a higher level of perceived stress, resulting in increased risk of depression.

BILIRUBIN IN NEONATE

Transient increase in unconjugated bilirubin commonly occurs in newborns, which is called as as “physiologic jaundice,” .It usually further resolves without any consequences. Physiologic jaundice offers protection against oxidative stress causing damage to neonatal tissue.

But the unconjugated bilirubin levels may increase above the physiologic range from additional sources of hemolysis. Trauma during birth, G6PD deficiency and ABO or Rh blood incompatibilities are the additional sources of hemolysis.

This pathologic increase in bilirubin can be neurotoxic resulting in neonatal bilirubin encephalopathy(kernicterus). Basal ganglia and other brain stem nuclei are affected by acute bilirubin encephalopathy .

Thus bilirubin has been found to confer both neuroprotective antioxidant characteristics as well as neurotoxic properties . A complete knowledge of the interactions between bilirubin and central nervous system is needed since it may have profound clinical implications in the treatment modalities used in the critical care setting.

BILIRUBIN AND NEUROPROTECTION

Unconjugated bilirubin cant crosses intact blood brain barrier, which prevents its accumulation in the CNS, since most of the unconjugated bilirubin found in plasma is bound to albumin . When bilirubin acts as an antioxidant, biliverdin is formed by the oxidation of bilirubin, but again bilirubin is formed by the action of biliverdin reductase.

This explains even in low concentrations in neuronal cell cultures bilirubin exerts its powerful antioxidant property. HO-2constitutes the major form of heme oxygenase in the central nervous system, whereas HO-1 is found in specific cell types in the brain such as microglia and macrophages.

Recently in an animal study done in rats , after the occlusion of middle cerebral artery, propofol post-treatment there was evidence of attenuation of ischemic damage partly by up regulation of HO-1.

Similarly, in an experimental study done in mouse following cerebral ischemia showed greater damage of neurons in HO-2 knockout mice compared to normal counterparts, supporting the concept of neuroprotective role of bilirubin.

BILIRUBIN AND NEUROTOXICITY

Bilirubin not only serves protective role in neurological diseases , there are evidence stating the role of bilirubin in the progression of neurological dysfunction in various pathological conditions.

In most of these pathological states,there is increase in bilirubin levels above physiologic range,so that the toxic effects of bilirubin exceed the protective role. This effect will, results in damage to the central nervous system.

The neurotoxic effects of bilirubin starts above a certain micromolar concentrations, and when that level is reached it will aggregate and adhere to cellular membranes, resulting in the disruption normal function.

Drugs can compete with bilirubin for albumin-binding sites ,resulting in increase of plasma bilirubin levels.

For example, bilirubin can be displaced from albumin by fatty acid components which leads to amplification of bilirubin related neurotoxicity in susceptible patients.

NEUROTOXICITY OF INDIRECT BILIRUBIN

But Maria Alexandra Brito et al observed the role of unconjugated bilirubin in promoting lipid peroxidation, ROS formation and protein oxidation in synaptosomal membrane systems[37].

Similarly,in another study it has been proposed that the pathogenesis of encephalopathy by hyperbilirubinemia is due to action of unconjugated bilirubin by induction of oxidative stress[48].

Cristina Bellarosa 2011 proposed that increased unconjugated bilirubin (UCB) can result in bilirubin encephalopathy.Oxidative and Endoplamic Reticulum stress are suggested to be involved bilirubin induced neurotoxicity[36].

NEUROTOXICITY OF BILIRUBIN AND HEMORRHAGIC STROKE

When a weakened blood vessel ruptures hemorrhagic stroke occurs, which results in bleeding into the brain substance and neuronal injury subsequently. There are additional complications in hemorrhagic

stroke patients resulting in secondary damage which occurs days after the initial event such as cerebral ischemia and vasospasm.

HO-1 in the brain is induced by the blood presence locally resulting in increased production of unconjugated bilirubin .

There are clinical evidence which supports the concept , that the environment immediately around the hematoma is highly contributing to oxidative reactions, augmenting the conversion of bilirubin into bilirubin oxidation products.

BOXes(bilirubin oxidation products) in CSF have temporal relationship with the time of onset of cerebral vasospasm, and proved to be vasoactive.

These findings collectively gives a conclusion that BOXes can either cause or contribute to vasospasm and also the resulting delayed neurologic dysfunction following hemorrhagic stroke[43].

ISCHEMIC STROKE AND SERUM BILIRUBIN

In 1971 herishanu et al in a study found that hyperbilirubinemia was prevalent in patients with acute ischemic stroke during the first 48 hours after the onset of stroke symptoms., whereas liver enzymes and other values were normal in the same patients,but they couldn't substantiate this discrepancy.

There are studies supporting the evidence of protective role of bilirubin in the incidence of cardiovascular and cerebrovascular diseases[51-52,44].

But ,it has been proposed by various studies that during the acute phase of ischemic stroke bilirubin levels get elevated .

And patients with higher level of bilirubin had severe disease and worse outcomes.

Bilirubin causes brain damage by

- Mitochondrial enzymes inhibition,
- Disrupt of DNA synthesis, and
- Attenuation of protein production.

Nicholas V Mendez1 [45]et al 2013 explained about the role of bilirubin in ischemic stroke as follows,

In Ischemic stroke when blood flow to a part of brain is obstructed either by thrombus or embolus ,there will be hypoxic ischemic state, which produces downstream hypoxic-ischemic conditions resulting in increased oxidative stress.

In these conditions, HO-1 induction takes place resulting in amplification of bilirubin formation. It has been proposed that the serum bilirubin level is a biomarker of the degree of ischemic damage following stroke.

These findings suggest that the bilirubin may be used as an early prognostic marker of stroke severity and outcome during the management of patients having ischemic stroke.

MATERIALS AND METHODS

SETTING:

Patients admitted in Kilpauk Medical College and Hospital, Chennai.

COLLABORATING DEPARTMENTS :

Neurology, Radiology, Biochemistry

PERIOD OF STUDY:

April 2014 to September 2014

STUDY DESIGN:

Case control Study (Ischemic stroke patients as cases and non stroke persons as control group)

STUDY POPULATION:

All patients presenting with new onset neurological deficit following ischemic stroke within 48 hours of onset of stroke admitted in the medical wards were included in the study.

An equal number of age, sex and comorbid conditions[Diabetes Mellitus, Systemic Hypertension] matched persons not having stroke attending OPD were included in the study as control group

DEFINITIONS FOLLOWED IN THIS STUDY

STROKE

In this study,Stroke was defined as rapid onset of focal neurological deficit lasting for more than twenty four hours or resulting in death with no cause apparently other than vascular etiology.

HYPERTENSION

Hypertension was diagnosed when it was documented in medical records or who were on regular intake of drugs for hypertension.

DIABETES

Diabetes was diagnosed with past medical records or when patients were on oral anti-diabetic drug or insulin

ALCOHOLISM

Patient was defined as alcoholic when there was a history of alcohol consumption in past 5 years.

SMOKER

Patients was diagnosed as smoker when there was smoking history in past 5 years.

NIHSS SCORE: NIHSS scoring was done on the basis of the clinical parameters. In this study, patients with score ≥10 were considered as having severe stroke

MRS SCORE: MRS score of <3were considered as Good Outcome and scores of ≥3were considered as Poor outcome.

INCLUSION CRITERIA

All patients presenting with new onset neurological deficit following ischemic stroke admitted within 48 hours of onset of stroke.

EXCLUSION CRITERIA

Patients with

- H/O alcoholism
- Known liver disease
- Known Chronic Kidney disease
- Known Coronary artery disease
- Known Malignancy
- Known Connective Tissue Disorders
- Hemodynamic instability – BP<90/60mmHg
- Hepatotoxic drug intake in past 30 days
- Infection identified through history and clinical examination
- Haemorrhagic Stroke Patients (ICH,SDH,EDH) - with the aid of CT/MRI scan

STUDY METHODS

This study is to compare the serum total bilirubin levels between acute ischemic stroke patients and non stroke subjects and its significance among them. And also to identify whether there is any association between stroke severity and prognosis with serum total bilirubin level.

Patients/relatives were explained about the study. Patients were included in the study after obtaining written informed consent.

Fifty patients who had acute ischemic stroke and fifty controls were included for study. Patients who got admitted within 48 hours of stroke onset only were taken under study. Then complete relevant medical history, neurological examination, relevant blood investigations and CT/MRI scan were done and all data were recorded in a standardized proforma.

CT/MRI scan was done for the exclusion of hemorrhagic stroke. Blood sample for Serum total and direct bilirubin and other baseline investigations was taken as soon as patient got admitted . National Institute of Health Stroke Scale (NIHSS) scoring was assessed at the time of admission and these patients were grouped according to score of <10 and ≥ 10

These patients were treated according to standard protocols. None of the patients were thrombolysed.. Modified Rankin Scale was assesed to know the functional recovery of the patient after 7 days either in ward or in review opd if got discharged.

Similarly subjects in control group were studied by relevant medical history,and clinical examnation and relevant blood investigations.

All the subjects in control group were selected in respect to age(group match),gender,co-morbid conditions (diabetes and hypertension) matched with cases.

BIOCHEMICAL ANALYSIS

Blood samples were taken as soon as the patient admitted in case group and in control group randomly. Serum bilirubin was measured by spectrophotometry method in the laboratory using Jendrassik-Grof allied methods.

DATA ANALYSIS

Continuous variables like age and bilirubin level were expressed as mean (Standard Deviation). Association between ischemic stroke and total bilirubin level was tested by comparing serum bilirubin levels in ischemic stroke patients with that of controls using unpaired (Independent) t test. A P value of <0.05 was considered as statistically significant.

Severity of stroke on admission was assessed by NIHSS severity scale (NIHSS$\geq$ 10 was considered as Severe stroke) and the prognosis of stroke on 7^{th} day of stroke was be assessed by Modified Rankin functional outcome Scale (MRS $\geq$3was considered as poor prognosis). Total bilirubin levels were divided in to three groups, (Group 1: $\leq$ 0.6mg/dl, Group 2: 0.7 to 0.9mg/dl, Group 3: $\geq$1.0 mg/dl). Severity of stroke, prognosis of stroke was expressed as percentages and its correlation with three groups of bilirubin was tested by Chi Square analysis. A P value of <0.05 was considered as statistically significant.

RESULTS AND ANALYSIS

TABLE 7: *AGE DISTRIBUTION IN STUDY GROUP*

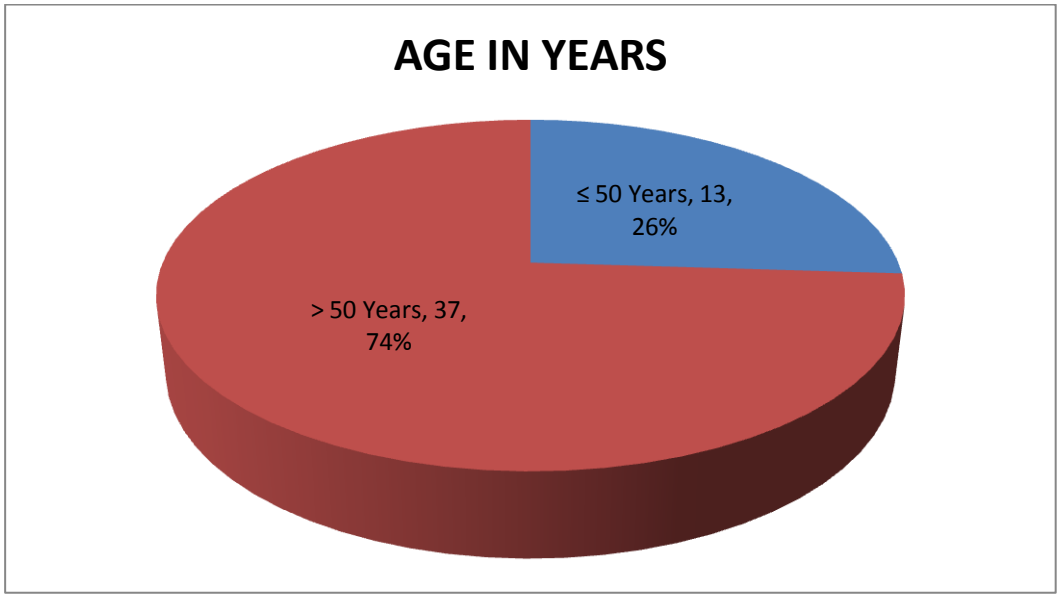
Parameter	Cases	Control	P value
Mean age in Years (SD)	59.7 (11.97)	58.28 (10.1)	0.523

There is no statistically significant difference in respect to age among case and control group.

TABLE 8: *AGE DISTRIBUTION IN STUDY GROUP*

AGE (IN YEARS)	NO.OF CASES(n)	NO.OF CASES(%)
≤ 50	13	26%
> 50	37	74%
TOTAL	50	100%

FIGURE 7: *DISTRIBUTION OF CASES IN AGE GROUP*

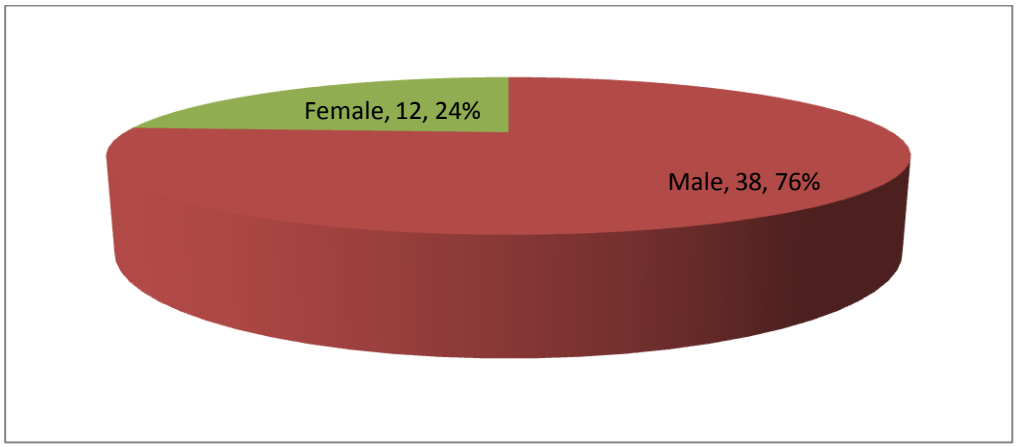


Out of 50 cases, 26%, 13 cases were in the age group ≤50 years and 74%, 37 were in the age group >50 years.

TABLE 9: *GENDER DISTRIBUTION IN STUDY GROUP*

Gender	**Cases (n=50)**	**Controls (n=50)**
Male	38 (76%)	38 (76%)
Female	12 (24%)	12 (24%)
Total	50 (100%)	50 (100%)

FIGURE 8: *DISTRIBUTION OF CASES IN GENDER GROUP*

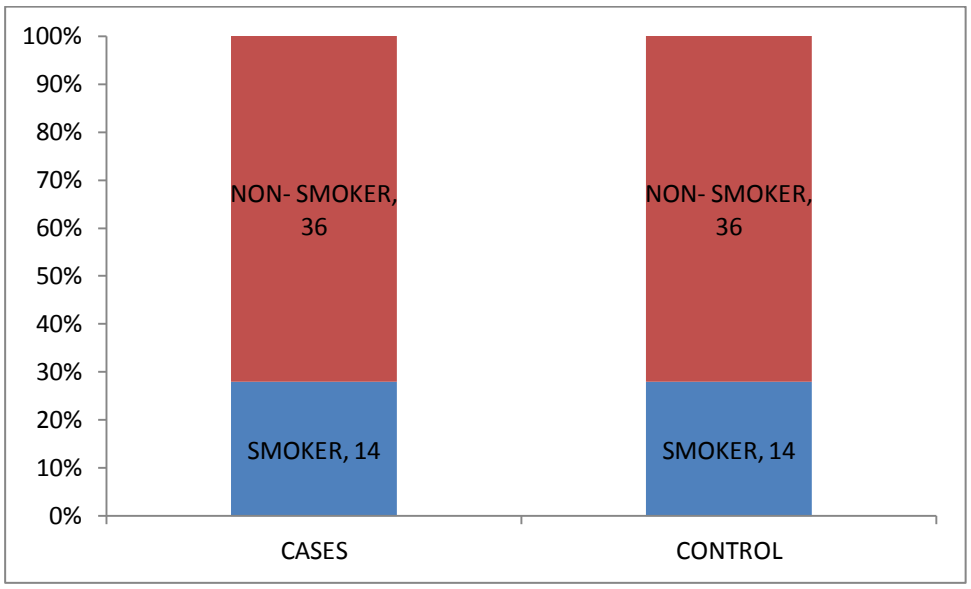


In the study, among the 50 cases ,76% ,38 cases were males and 24 %, 12 cases were females ,as the controls were sex matched equal numbers found in control group.

TABLE 10: *DISTRIBUTION OF TOBACCO SMOKING IN STUDY GROUP*

Gender	**Cases(n=50)**	**Controls(n=50)**
Smoker	14 (28%)	14 (28%)
Non smoker	36 (72%)	36 (72%)
Total	50 (100%)	50 (100%)

FIGURE 9: *DISTRIBUTION OF TOBACCO SMOKING IN STUDY GROUP*



In the study population, both among the case and control group 28%,14 were smokers and 72%,36 were non smokers.

TABLE 11: *BODY MASS INDEX OF CASE AND CONTROL GROUP*

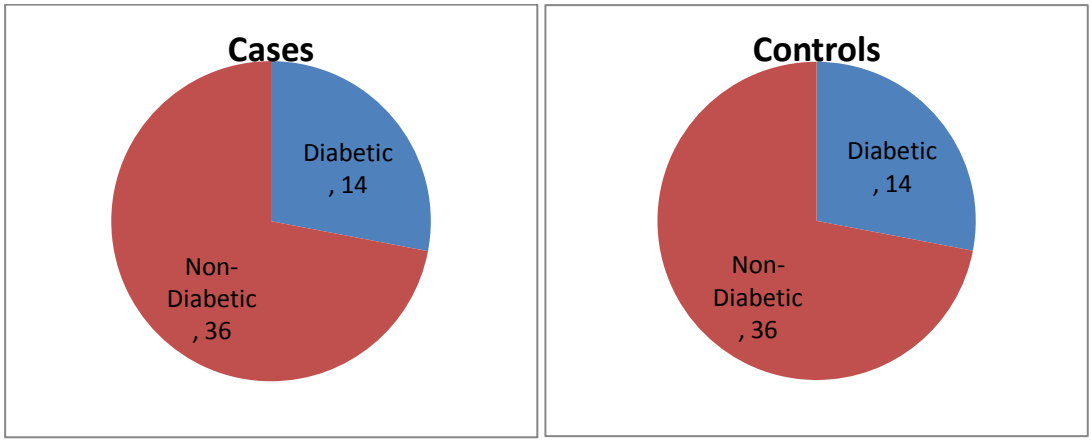
BMI	**CASE, n (%)**	**CONTROL, n (%)**
<18.5	0	5 (10)
18.5-22.9	27 (54)	26 (52)
23-24.9	12 (24)	11 (22)
≥25	11 (22)	8 (16)
Total	50 (100)	50 (100)

There is no major difference in BMI among case and control. Majority of the both cases and control were in the range of 18.5 to 22.9 BMI.

TABLE 12: *DISTRIBUTION OF DIABETES AMONG CASES & CONTROL*

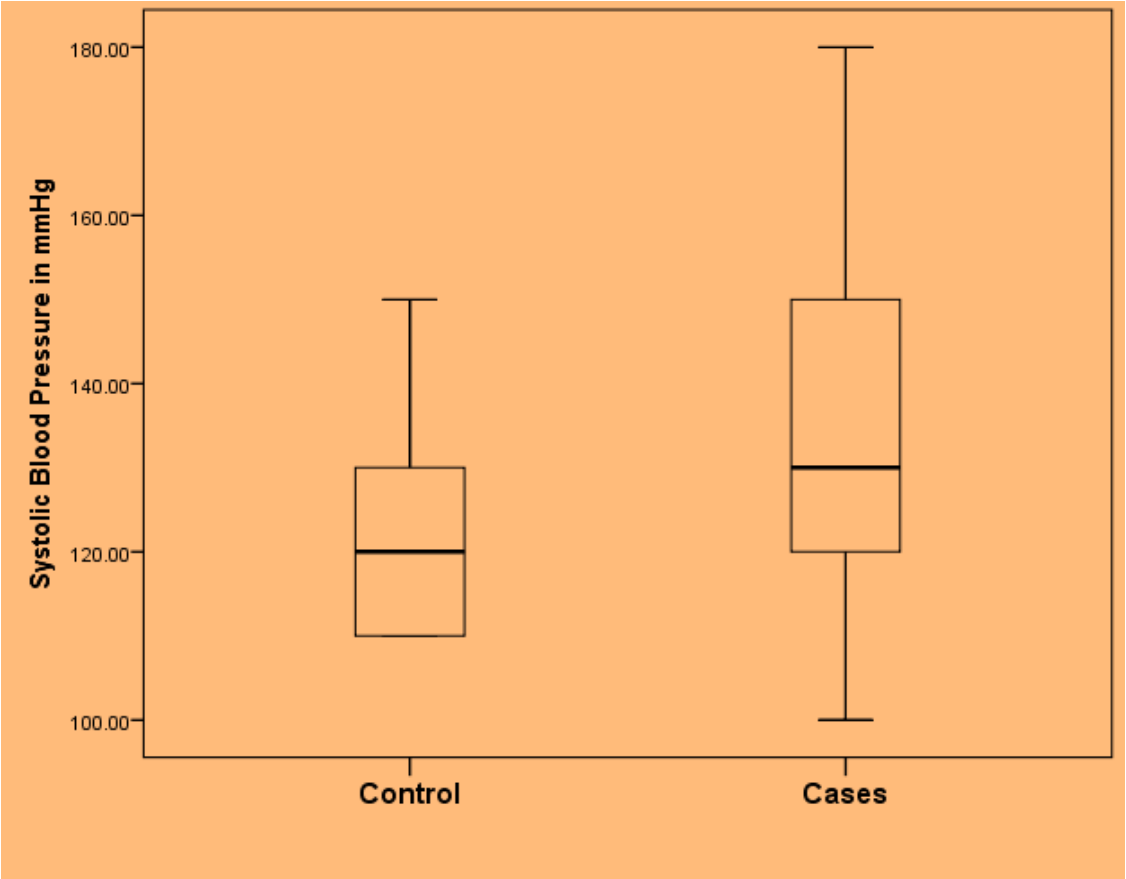
PARAMETERS	**CASE**	**CONTROL**
Diabetic	14(28%)	14(28%)
Non-Diabetic	36(72%)	36(72%)

FIGURE 10: *DISTRIBUTION OF DIABETES AMONG CASES & CONTROL*



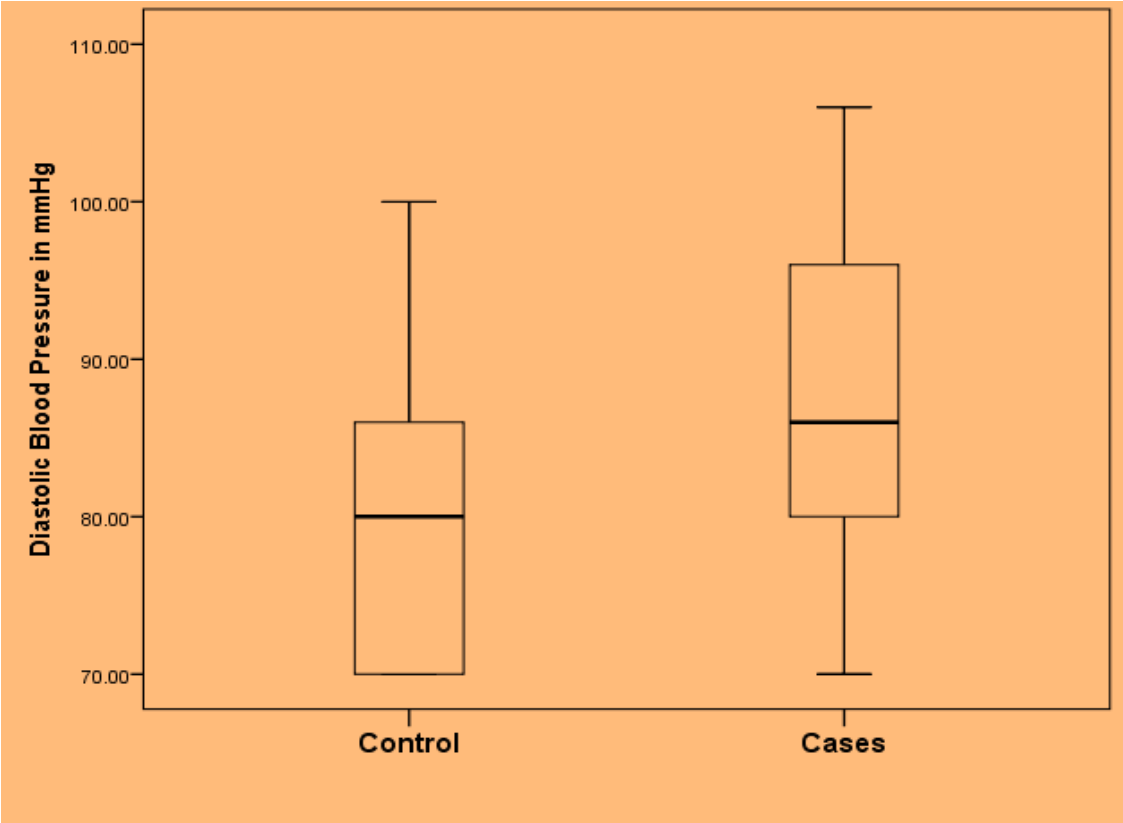
In the study population, 28%, 14 were diabetics and 72 %, 36 were non diabetics in both case and control group as controls were matched for diabetes.

FIGURE 11: *DISTRIBUTION OF SYSTOLIC BLOOD PRESSURE IN STUDY GROUP.*



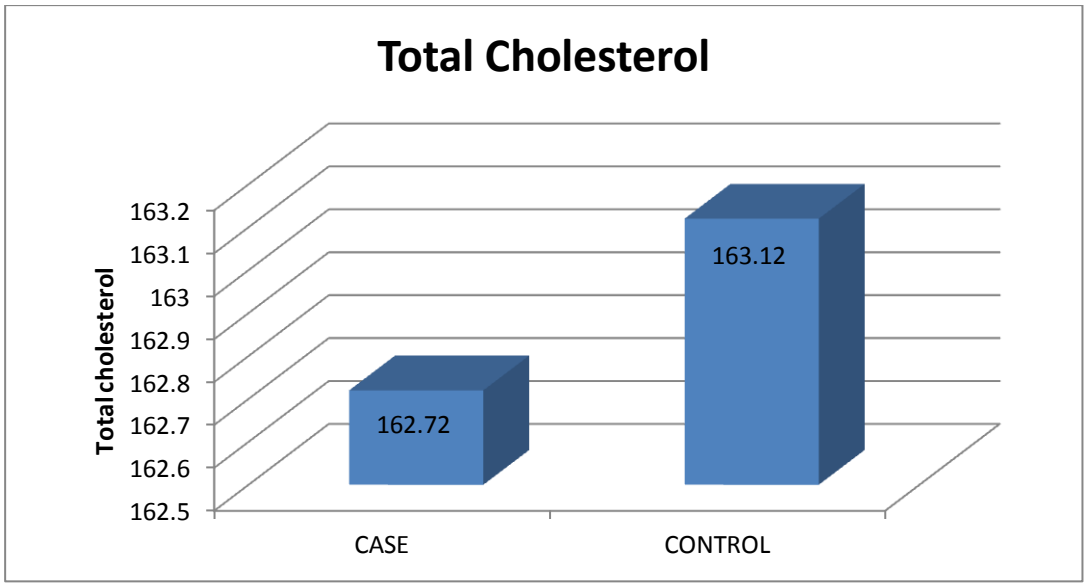
Box plot of systolic blood pressure shows that median systolic blood pressure was 120 and 130 among controls and cases respectively.

FIGURE 12: *DISTRIBUTION OF DIASTOLIC BLOOD PRESSURE IN STUDY GROUP.*



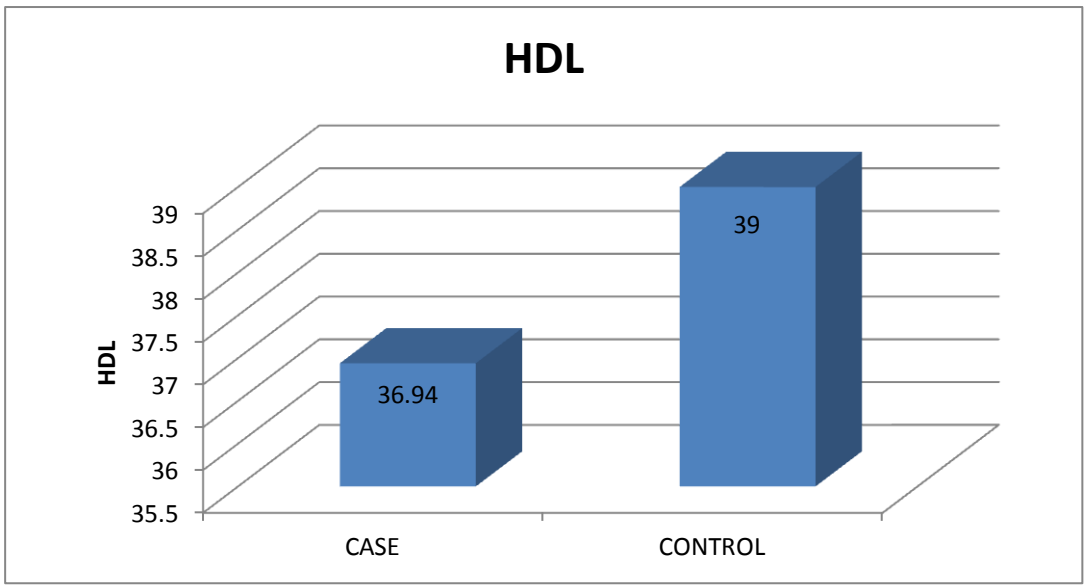
Box plot of diastolic blood pressure shows that median diastolic blood pressure was 80 and 86 among controls and cases respectively.

FIGURE 13: *TOTAL CHOLESTEROL AMONG CASE AND CONTROL GROUP*



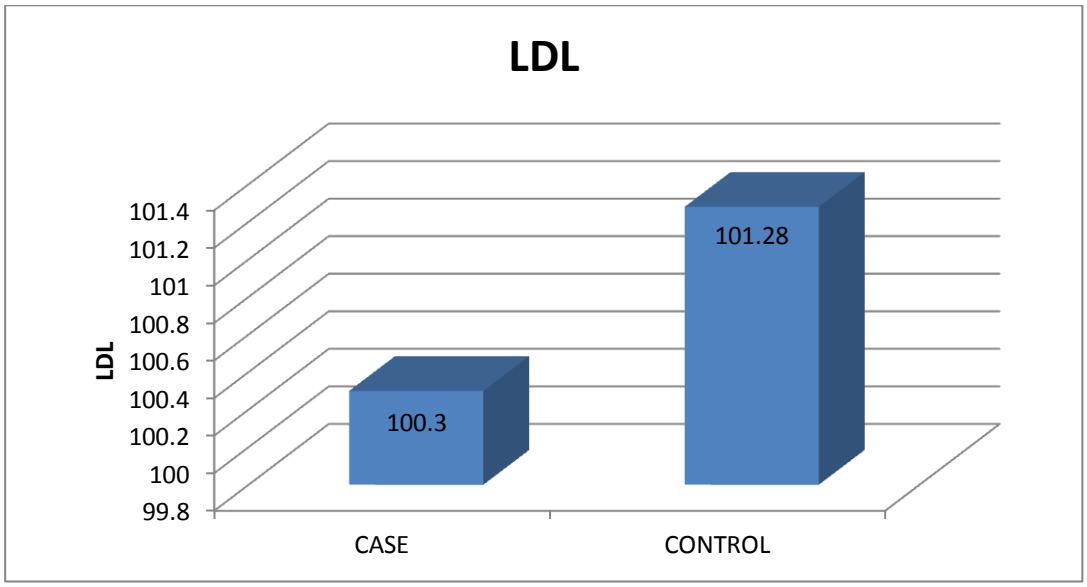
The total cholesterol among two groups doesn't show statistical difference.

FIGURE 14: *HDL AMONG CASE AND CONTROL GROUP*



The HDL among two groups doesn't show statistical difference.

FIGURE 15: *LDL AMONG CASE AND CONTROL GROUP*



The LDL among two groups doesn't show statistical difference.

FIGURE 16: *TG AMONG CASE AND CONTROL GROUP*

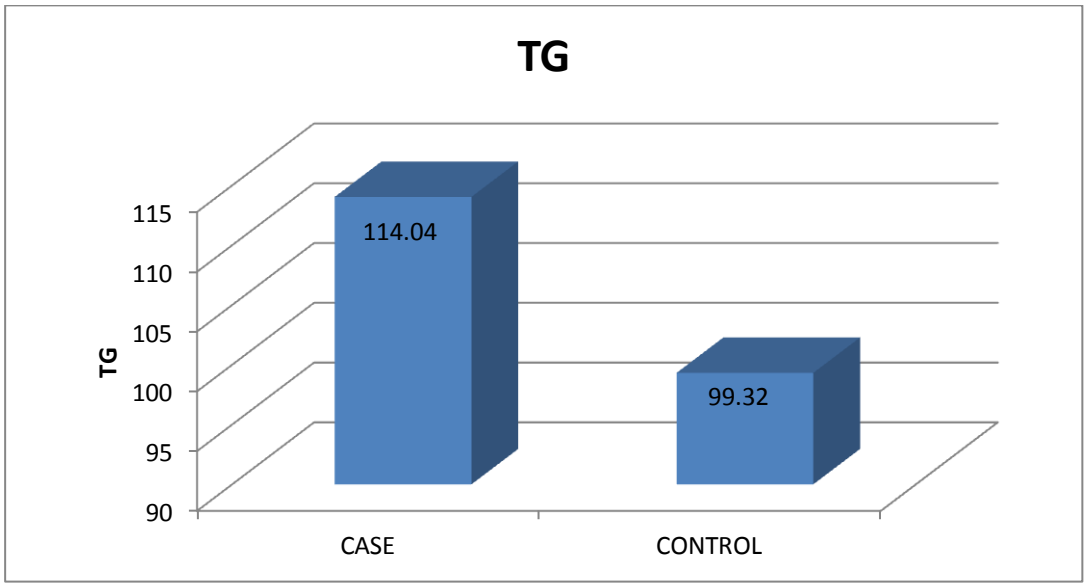
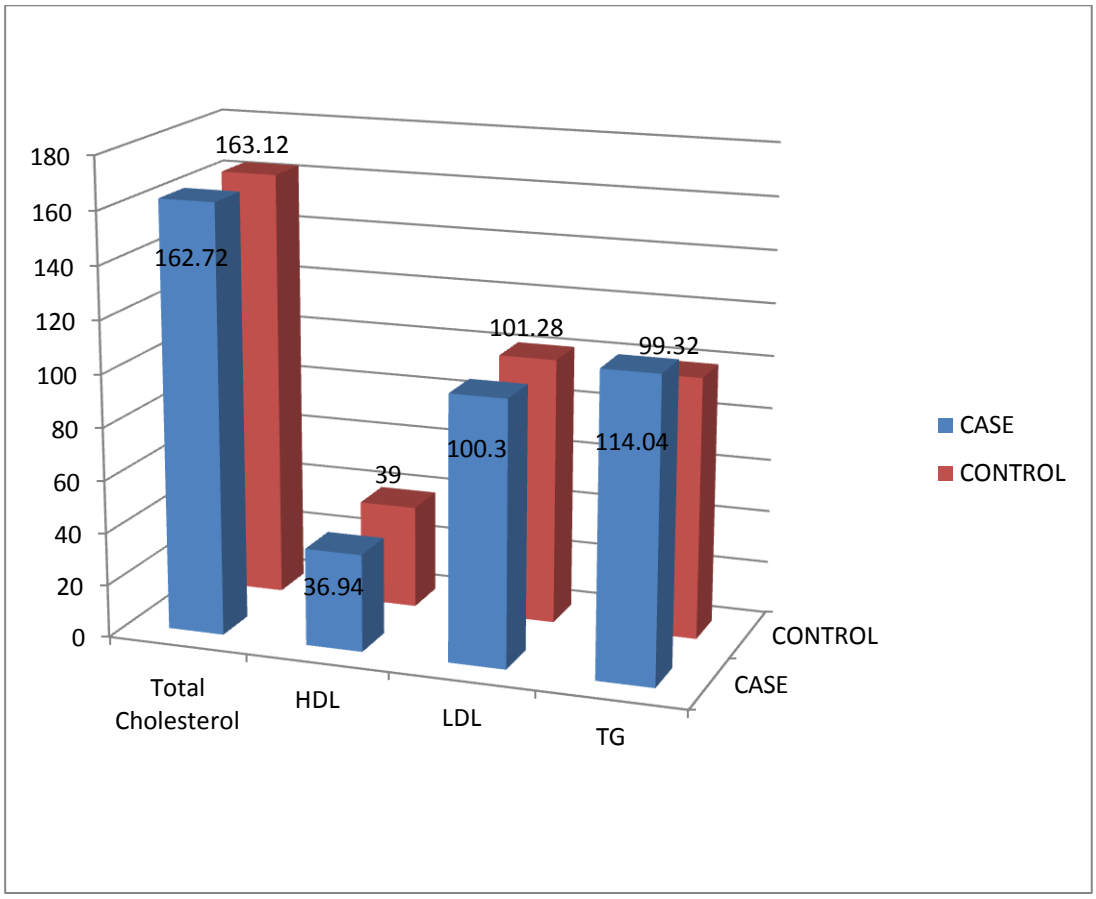


TABLE 13: *LIPID PROFILE OF CASE AND CONTROL GROUP*

Parameter	**CASE**	**CONTROL**	**p value**
Total Cholesterol	162.72 (35.48)	163.12 (29.61)	0.951
HDL	36.94 (8.44)	39 (6.53)	0.175
LDL	100.3 (27.18)	101.28 (20.76)	0.84
TG	114.04 (43.81)	99.32 (34.19)	0.064

The lipid profile parameter does not imply any significant differences in total cholesterol, HDL, LDL, TG among the case and control group.

FIGURE 17: *LIPID PROFILE OF CASE AND CONTROL GROUP*



There were no significant differences in total cholesterol, HDL, LDL, TG among the case and control group.

TABLE 14: *LIVER ENZYMES IN CASE AND CONTROL GROUP*

Liver Enzymes	Case	Control	p value
AST	14.82 (7.6)	16.76 (11.2)	0.314
ALT	22.82 (11.18)	23.92 (11.6)	0.63

The difference in the liver enzyme Aspartate Transaminase(AST), Alanine Transaminase(ALT) among the case and control group is not statistically significant.

FIGURE 18: *LIVER ENZYMES IN CASE AND CONTROL GROUP*

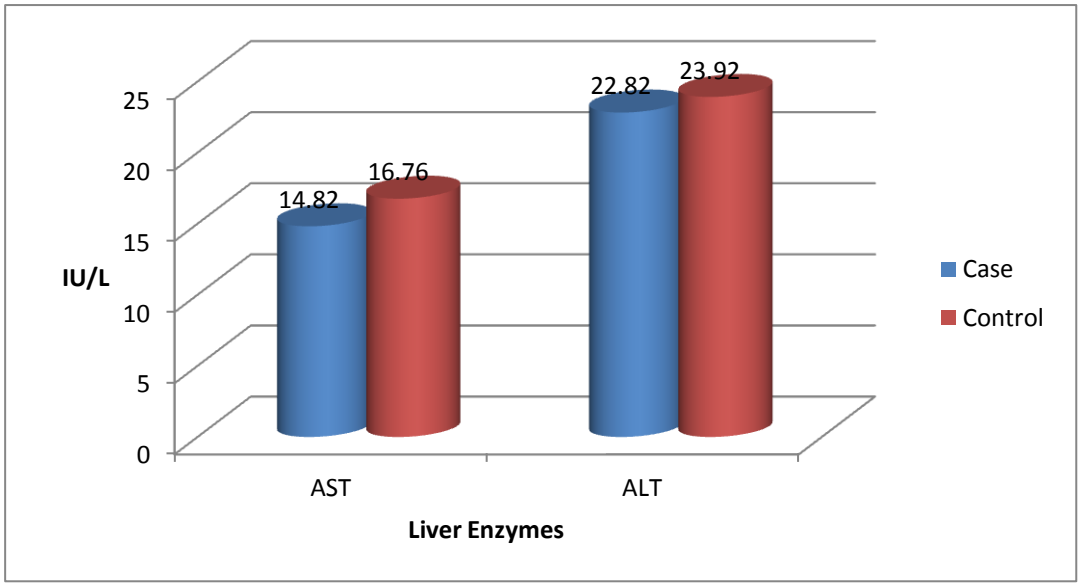
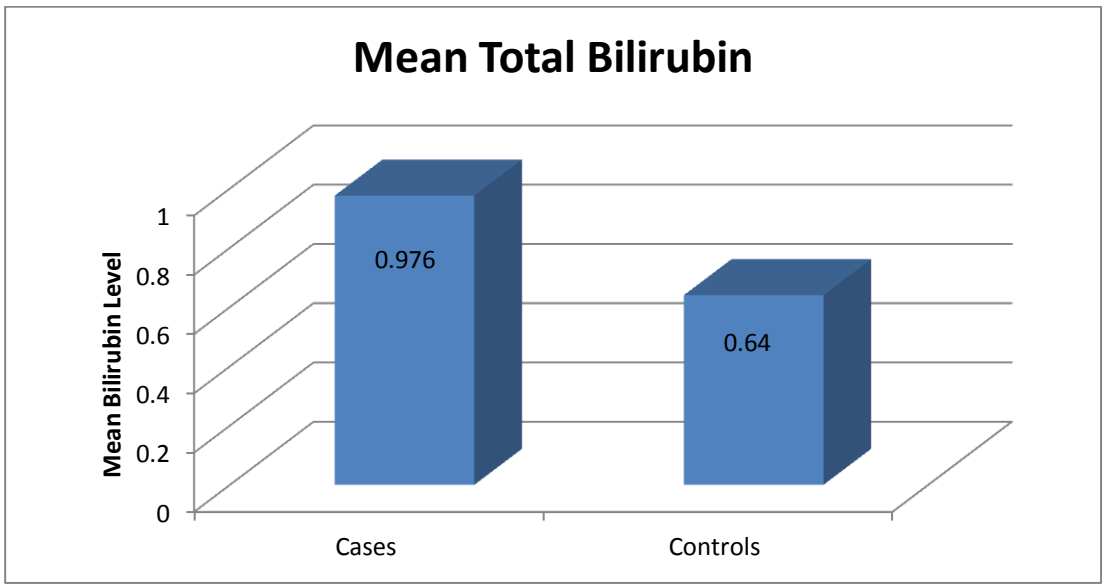


TABLE 15: *MEAN (SD) TOTAL BILIRUBIN IN CASE AND CONTROL GROUP*

Parameter	Cases (n=50)	Controls (n=50)	p value
Mean Total Bilirubin (SD)	0.976 (0.328)	0.64 (0.249)	<0.001

FIGURE 19: *MEAN (SD) TOTAL BILIRUBIN IN CASE AND CONTROL GROUP*

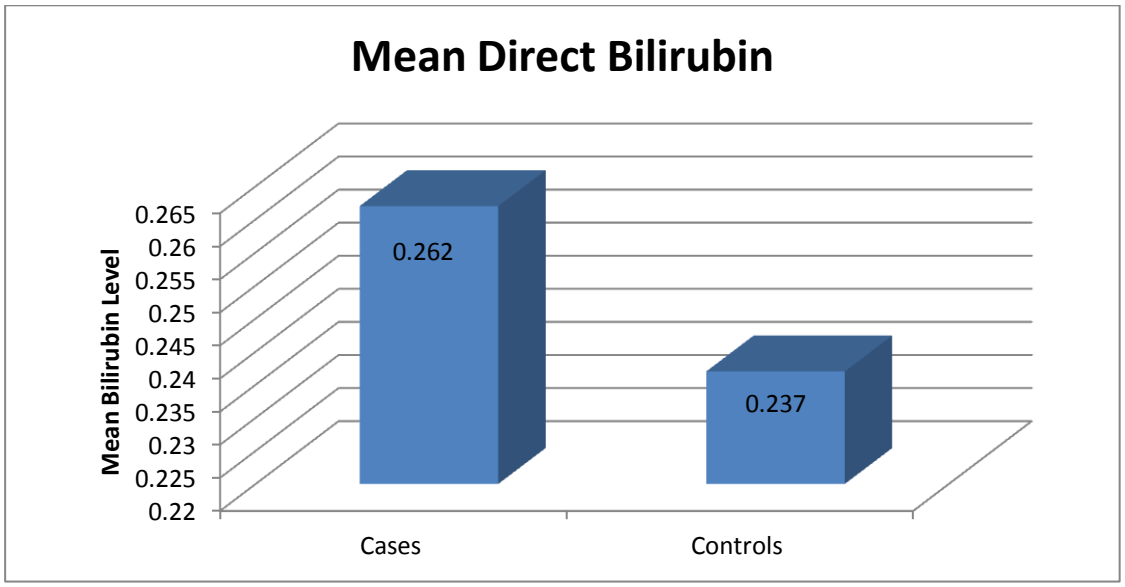


The mean total bilirubin in cases was 0.976, whereas in control group it was 0.64. Thus there is positive correlation between total bilirubin and acute ischemic stroke and it is statistically significant.

TABLE 16: *MEAN (SD) DIRECT BILIRUBIN IN CASE AND CONTROL GROUP*

Parameter	Cases (n=50)	Controls (n=50)	p value
Mean Direct Bilirubin (SD)	0.262 (0.199)	0.237 (0.123)	0.452

FIGURE 20: *MEAN (SD) DIRECT BILIRUBIN IN CASE AND CONTROL GROUP*

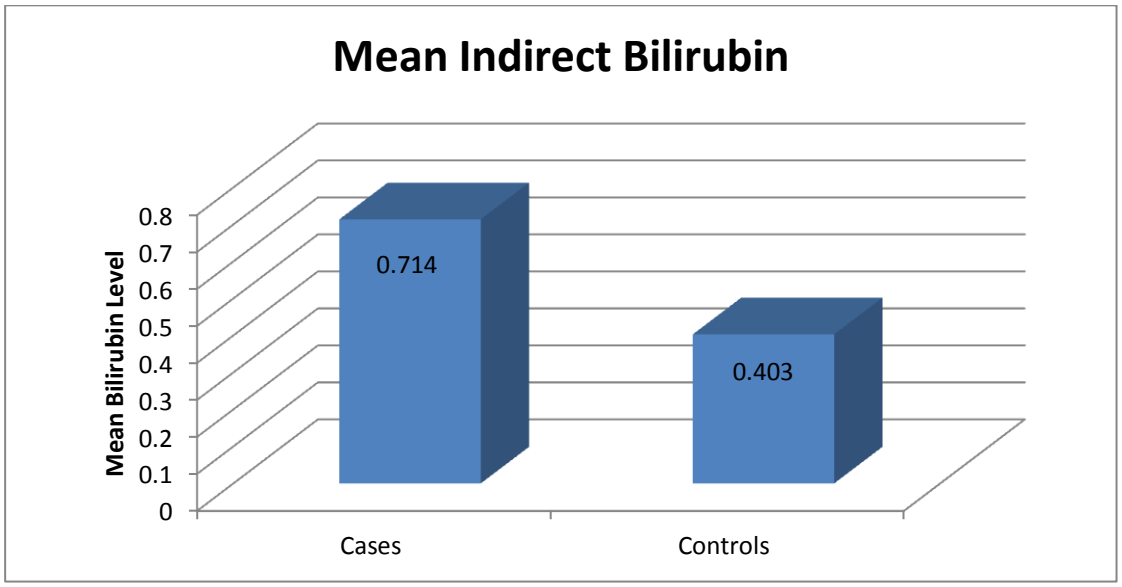


The mean direct bilirubin in cases was 0.262 and in control group is 0.237, so there is no correlation between direct bilirubin and acute ischemic stroke

TABLE 17: *MEAN (SD) INDIRECT BILIRUBIN IN CASE AND CONTROL GROUP*

Parameter	**Cases (n=50)**	**Controls (n=50)**	**p value**
Mean Indirect Bilirubin (SD)	0.714 (0.286)	0.403 (0.186)	<0.001

FIGURE 21: *MEAN (SD) INDIRECT BILIRUBIN IN CASE AND CONTROL GROUP*



The mean indirect bilirubin in cases was 0.714 and in control group was 0.403,so there is a positive correlation between indirect bilirubin and acute ischemic stroke and it is statistically significant.

TABLE 18: *TOTAL BILIRUBIN IN CASE AND CONTROL GROUP*

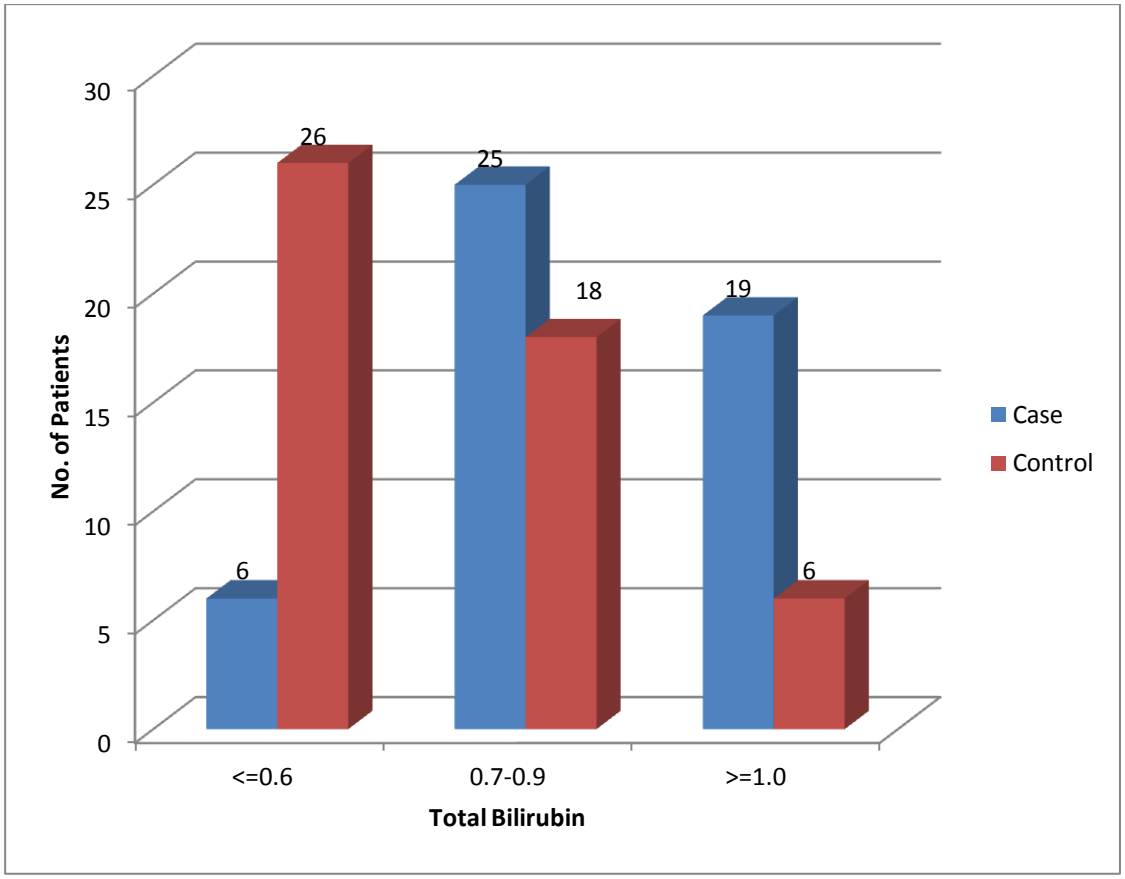
Total Bilirubin	**Case, n (%)**	**Control, n (%)**
≤0.6	6 (12%)	26 (52%)
0.7-0.9	25 (50%)	18 (36%)
≥1.0	19 (38%)	6 (12%)
Total	50 (100%)	50 (100%)

P value: <0.001

In the study,out of 50 cases 12%,6 cases had total bilirubin in the range ≤ 0.6,50%,25 cases had total bilirubin in the range of 0.7 - 0.9,38%,19 cases had total bilirubin in the range ≥1.0.

Among the control group,52%,26 subjects had total bilirubin in the range ≤0.6, 36%,18 subjects had total bilirubin in the range of 0.7 - 0.9, 12%,6 cases had total bilirubin in the range of ≥1.0.

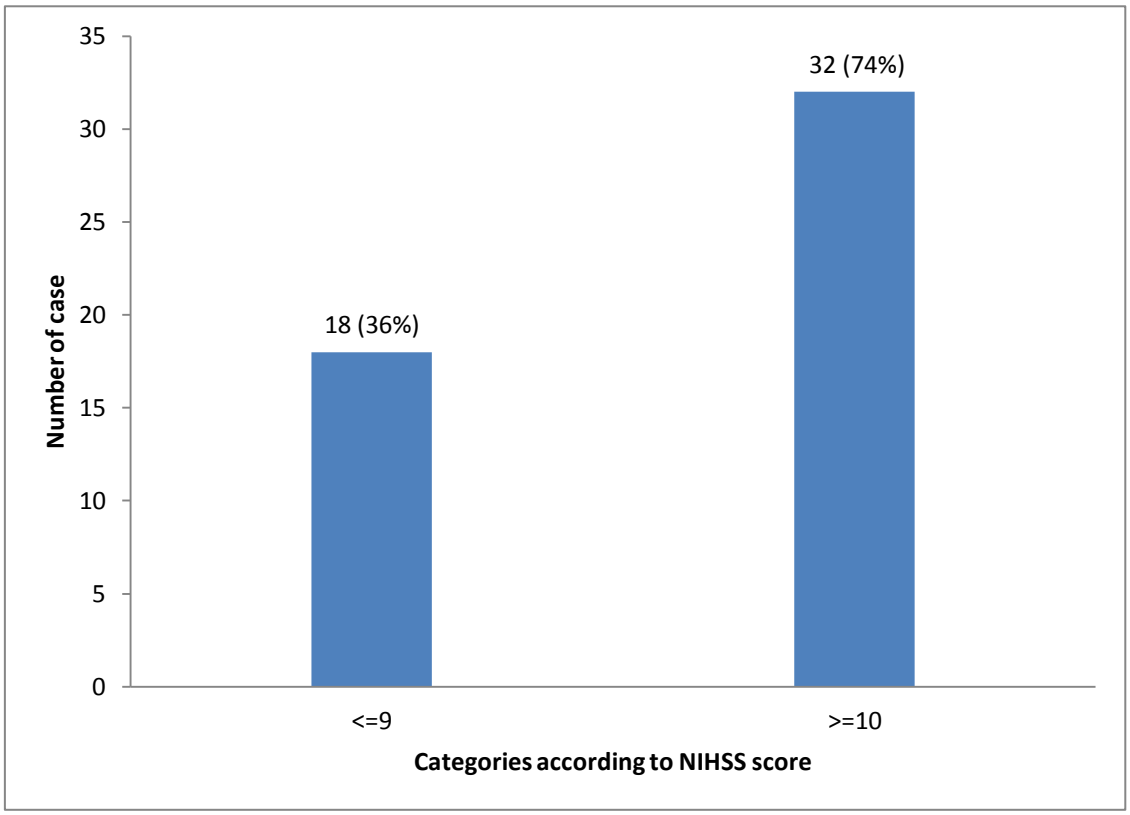
FIGURE 22: *TOTAL BILIRUBIN IN CASE AND CONTROL GROUP*



Thus 44 cases had total total bilirubin in the range ≥0.7 and only 6 cases had total bilirubin ≤0.6.

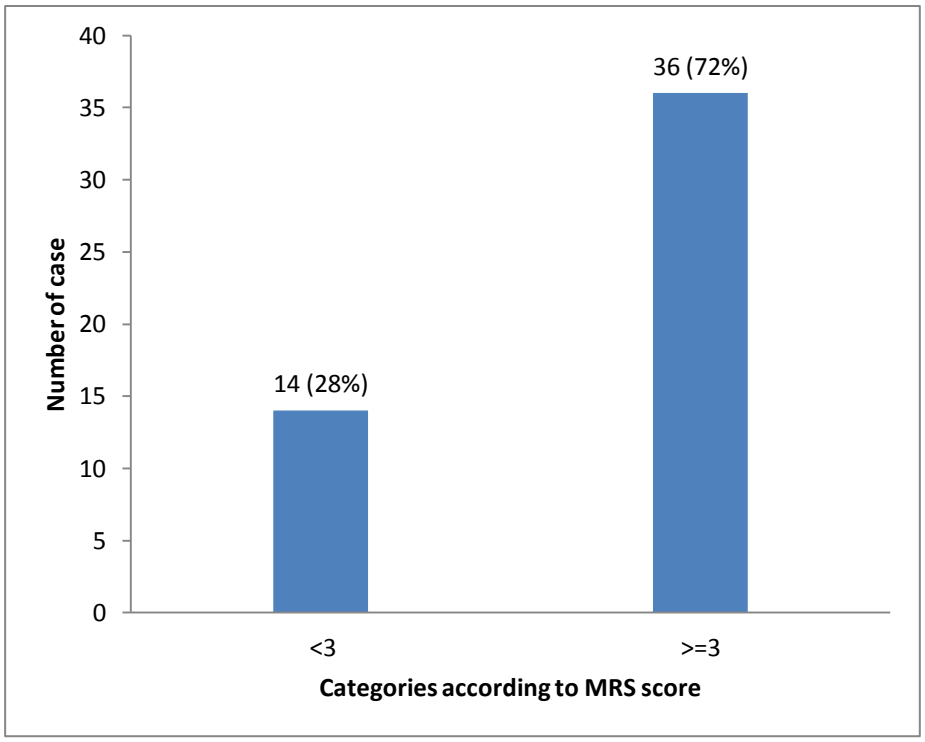
So the association between the acute ischemic stroke and admission total bilirubin is statistically significant comparing with control group.

FIGURE 23: *CATEGORIES ACCORDING TO NIHSS*



In the study, out of the 50 cases (36%) 18 cases had NIHSS ≤9 at admission and (74%) 32 cases had severe stroke NIHSS ≥10 at admission.

FIGURE 24: *CATEGORIES ACCORDING TO MRS*



In the study ,out of the 50 cases (28%) 14 cases had good outcome after 7 days of onset of stroke(MRS <3) and(72%)36 cases had poor outcome after 7 days of onset of stroke(MRS≥3)

TABLE 19: *ASSOCIATION OF TOTAL BILIRUBIN WITH NIHSS SCORE AMONG CASES*

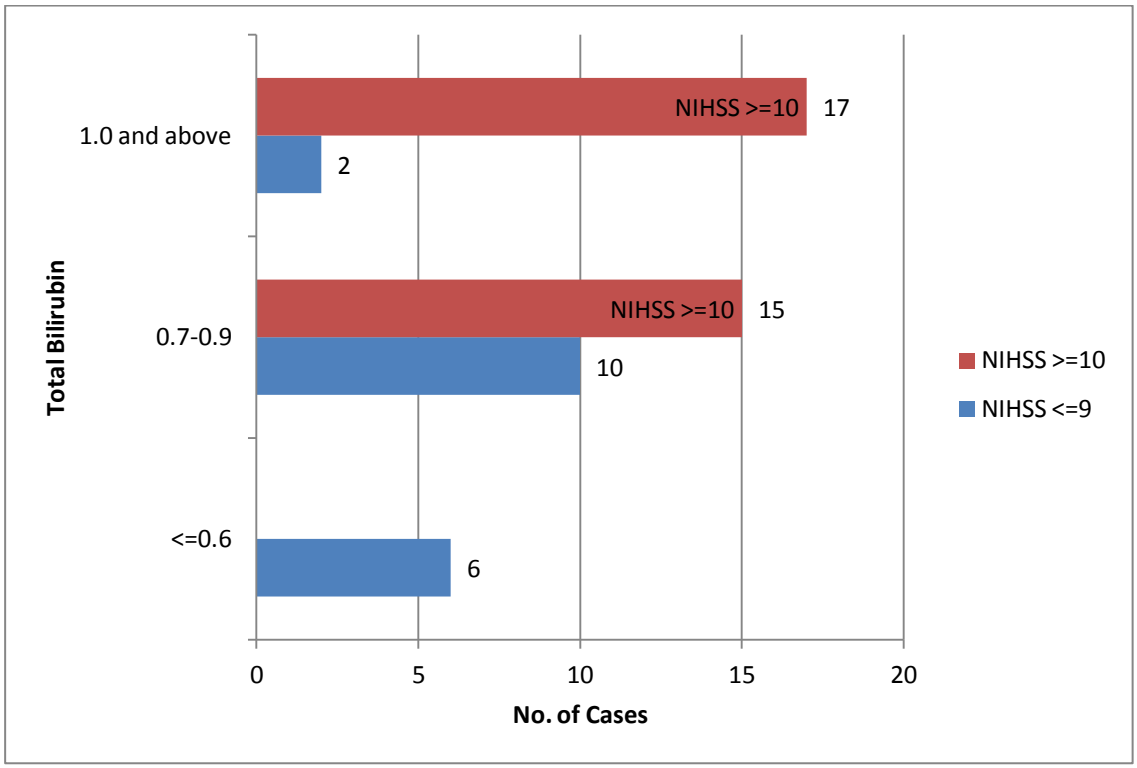
Total Bilirubin	**NIHSS**		
	≤9	**≥10**	**Total**
≤0.6	6(100%)	0(0%)	6(100%)
0.7-0.9	10 (40.0%)	15 (60.0%)	25 (100%)
1.0 and above	2 (10.5%)	17 (89.5%)	19 (100%)
Total	18 (36%)	32 (64%)	50 (100%)

P value= 0.0003

In the study, among 18 cases (36%) who had (NIHSS ≤9), six cases had total bilirubin in the range of ≤ 0.6, 10 cases had total bilirubin in the range of 0.7-0.9 and only 2 cases had total bilirubin ≥ 1.

Among 32 cases (64%) who had (NIHSS≥10), none of them had total bilirubin in the range of ≤ 0.6, 15 cases had total bilirubin in the range of 0.7-0.9 and 17 cases had total bilirubin ≥ 1.

FIGURE 25: *ASSOCIATION OF TOTAL BILIRUBIN WITH NIHSS SCORE AMONG CASES*



Thus, 32 cases with severe stroke(NIHSS≥10) had total bilirubin ranging ≥0.7. So, the association between the admission total bilirubin level and NIHSS at admission is statistically significant.

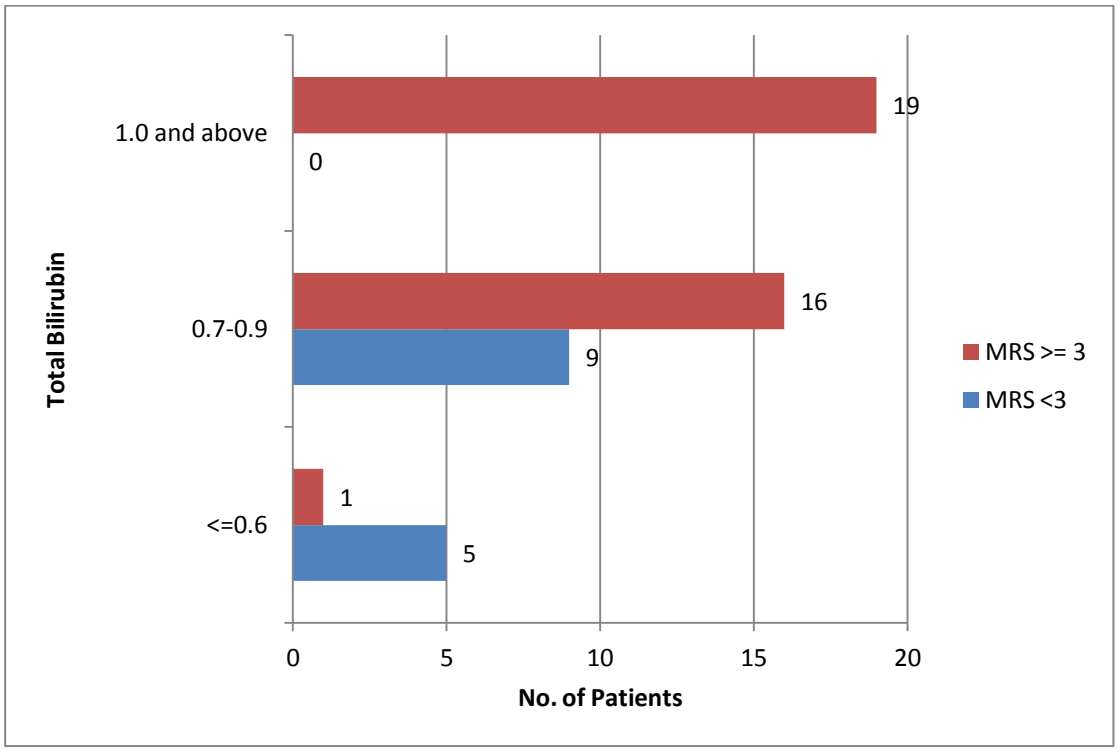
TABLE 20: *ASSOCIATION OF TOTAL BILIRUBIN WITH MRS SCORE AMONG CASES*

Total Bilirubin	**MRS**		
	<3	**≥3**	**Total**
≤0.6	5(80%)	1(20%)	6(100%)
0.7-0.9	9 (36%)	16 (64%)	25 (100%)
1.0 and above	0(0%)	19 (100%)	19 (100%)
Total	14 (28%)	36 (72%)	50 (100%)

p value: 0.0002

In the study, among the 14 cases (28%) who had good outcome after 7 days (MRS<3), 5 cases (80%) had total bilirubin in the range of ≤0.6, 9 cases(36%) had total bilirubin in the range of 0.7 -0.9 and none of them had total bilirubin 1 or above, whereas among the 36 cases(72%) who had poor outcome after 7 days(MRS≥3),only 1 case had total bilirubin in the range of ≤0.6, 16 cases(64%) had total bilirubin in the range of 0.7 -0.9 and 19 cases (100%) had total bilirubin ≥ 1.

FIGURE 26: *ASSOCIATION OF TOTAL BILIRUBIN WITH MRS SCORE AMONG CASES*



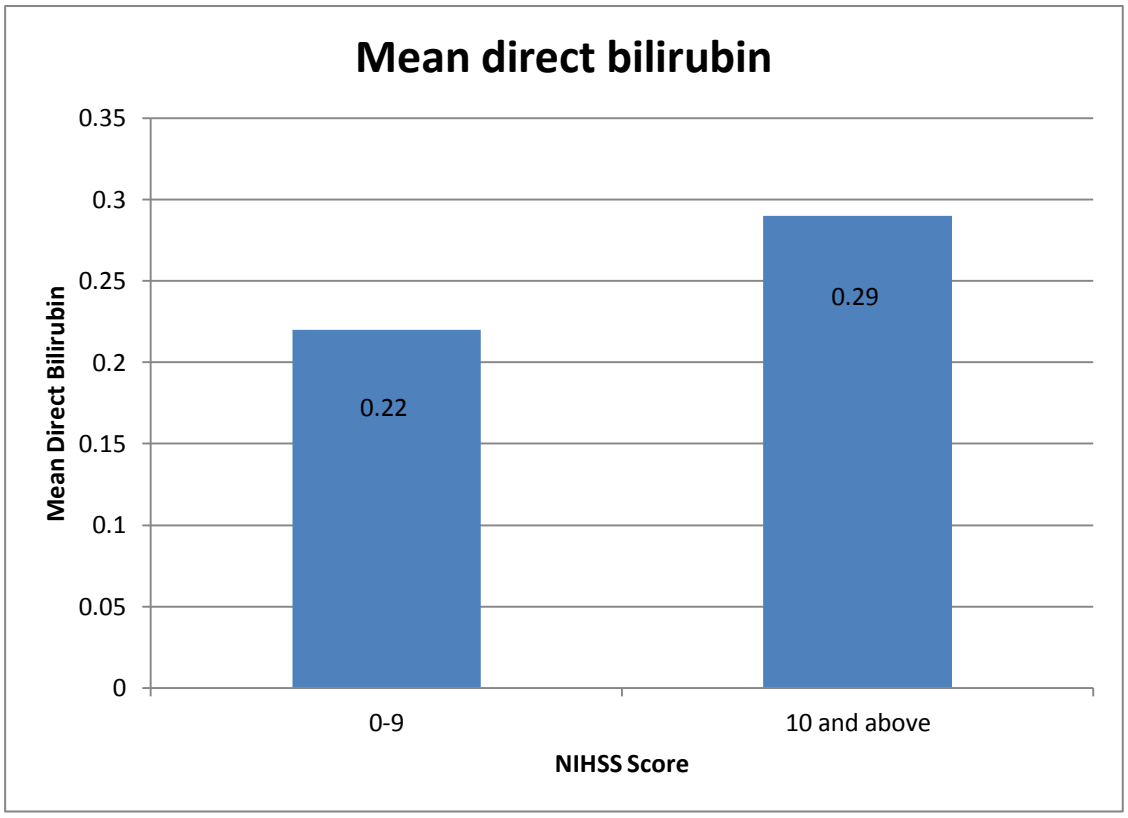
Thus 35 cases who had poor outcome after 7 days had total bilirubin in the range of 0.7 and above at the time of admission. So, the association between the admission total bilirubin and stroke outcome after 7 days is statistically significant

TABLE 21: *ASSOCIATION OF MEAN DIRECT BILIRUBIN WITH NIHSS SCORE AMONG CASES*

NIHSS group	**Mean direct bilirubin (SD)**	**P value**
0-9	0.22(0.15)	0.23
10 and above	0.29 (0.22)	

The difference in Mean Direct Bilirubin between the two groups is not statistically significant.

FIGURE 27: *ASSOCIATION OF MEAN DIRECT BILIRUBIN WITH NIHSS SCORE AMONG CASES*



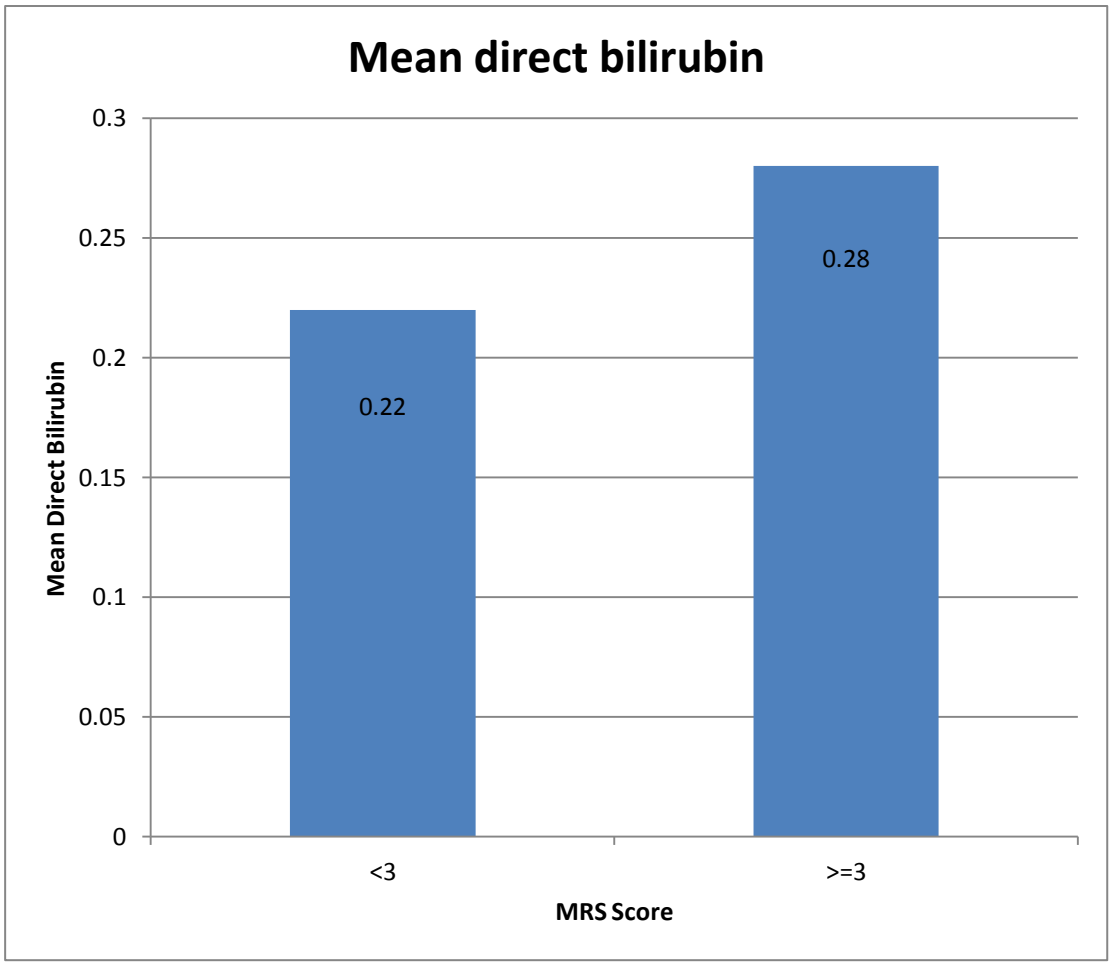
The Mean Direct Bilirubin in severe stroke patients (NIHSS≥10) is 0.29 whereas in another group(NIHSS≤9) is 0.22 and the difference is not statistically significant.

TABLE 22: *ASSOCIATION OF MEAN DIRECT BILIRUBIN WITH MRS SCORE AMONG CASES*

MRS group	**Mean direct bilirubin (SD)**	**P value**
<3	0.22(0.14)	0.37
≥3	0.28 (0.18)	

The difference in Mean Direct Bilirubin between the two groups is not statistically significant.

FIGURE 28: *ASSOCIATION OF MEAN DIRECT BILIRUBIN WITH MRS SCORE AMONG CASES*



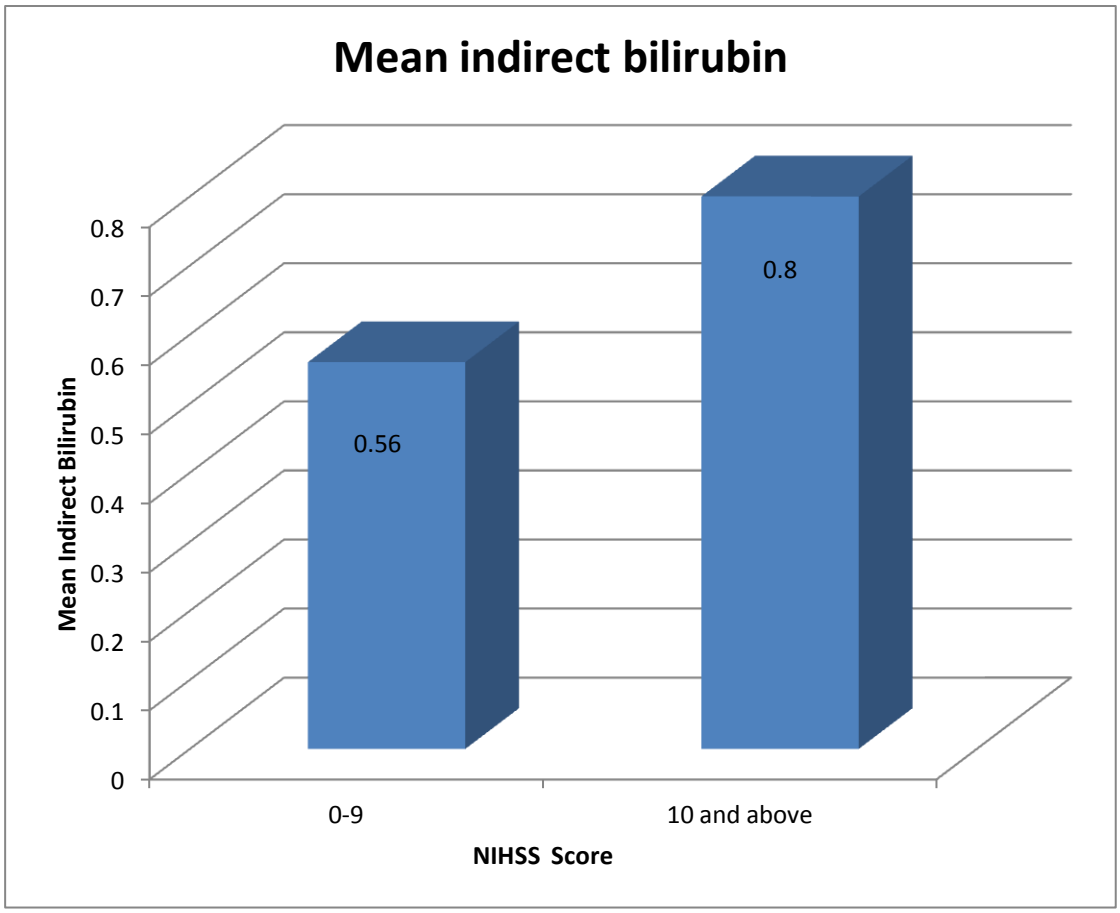
The Mean Direct Bilirubin in poor outcome patients (MRS≥3) is 0.28 whereas in another group (MRS<3) is 0.22 and the difference is not statistically significant.

TABLE 23: *ASSOCIATION OF MEAN INDIRECT BILIRUBIN WITH NIHSS SCORE AMONG CASES*

NIHSS group	**Mean indirect bilirubin (SD)**	**P value**
0-9	0.56(0.16)	0.003
10 and above	0.80 (0.31)	

The difference in Mean Indirect Bilirubin between the two groups is statistically significant.

FIGURE 29: *ASSOCIATION OF MEAN INDIRECT BILIRUBIN WITH NIHSS SCORE AMONG CASES*



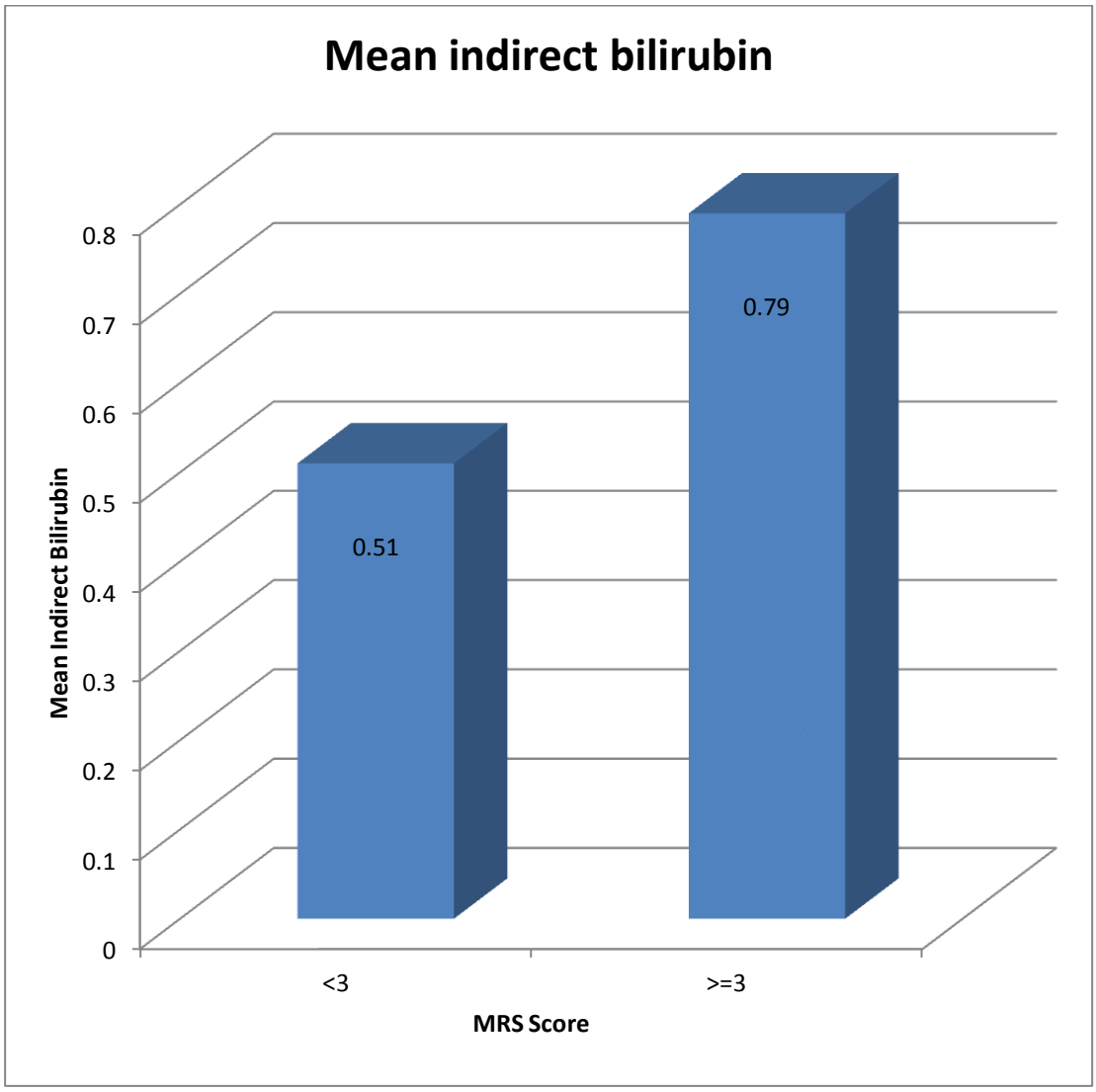
The Mean Indirect Bilirubin in severe stroke patients (NIHSS≥10) is 0.8 whereas in another group (NIHSS≤9) is 0.56 and the difference is statistically significant.

TABLE 24: *ASSOCIATION OF MEAN INDIRECT BILIRUBIN WITH MRS SCORE AMONG CASES*

MRS group	Mean indirect bilirubin (SD)	P value
<3	0.51(0.16)	0.001
≥3	0.79 (0.26)	

The difference in Mean Indirect Bilirubin between the two groups is statistically significant.

FIGURE 30: *ASSOCIATION OF MEAN INDIRECT BILIRUBIN WITH MRS SCORE AMONG CASES*



The Mean Indirect Bilirubin in poor outcome patients (MRS≥3) is 0.79 whereas in another group(MRS<3) is 0.51 and the difference is statistically significant.

TABLE 25: *ASSOCIATION OF NIHSS AND MRS*

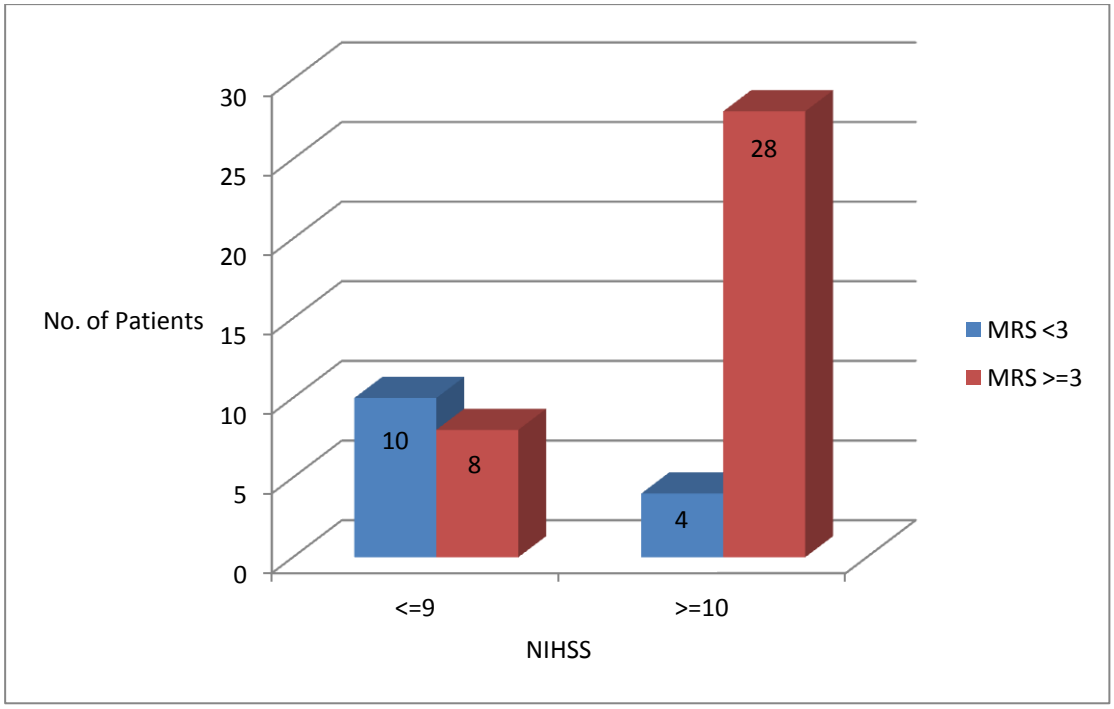
NIHSS	**MRS**		
	<3	**≥3**	**Total**
≤9	10 (55.6)	8 (44.4)	18 (100)
≥10	4 (12.5)	28 (87.5)	32 (100)
Total	14 (28)	36 (72)	50 (100)

p value= 0.001

In the study, among 18 cases who had NIHSS ≤9 at admission, 10 cases (55.6%) had good outcome (MRS <3) after 7 days and 8 cases (44.4%) had poor outcome (MRS ≥3) after 7 days.

Among 32 cases who had NIHSS ≥10 at admission, only 4 cases (12.5%) had good outcome (MRS <3) and 28 cases (87.5%) had poor outcome (MRS ≥3) after 7 days.

FIGURE 31: *ASSOCIATION OF NIHSS AND MRS*



Thus the association between NIHSS at admission and MRS after 7 days is statistically significant.

DISCUSSION

Bilirubin once considered to be the toxic waste product, in recent days gained importance because of its antioxidant property.

Various studies proposed about the role of bilirubin in oxidative stress mediated diseases like stroke, coronary artery diseases, cancer etc.

There are studies which concluded that greater admission serum bilirubin levels were associated with greater stroke severity and poor short term outcome.

Several studies have proved that the synthesis of bilirubin is induced in response to oxidative stress.

So the high bilirubin level at the admission found in ischemic stroke patients may be simply due to oxidative stress pathway induction and bilirubin may play no role in protection against neurological damage.

In this Case control Study (Ischemic stroke patients as cases and non stroke persons as control group) conducted in Kilpauk Medical College during the period of April 2014 to September 2014, I analysed 50 patients who had Ischemic stroke were taken as cases, 50 patients who did not have stroke were taken as control group.

All the cases and controls were subjected to complete investigation and were analyzed for difference in serum total bilirubin levels between cases and controls.

Further analysis was done to find any association exists between total bilirubin and stroke severity at admission and prognosis after 7 days.

In the study, mean total bilirubin was higher in cases than controls and the difference is stastically significant (p <0.001) and also the indirect bilirubin was also higher in cases significantly but the direct bilirubin didn't showed such results.

Among the cases, 74 % had severe stroke on admission and 72% had poor functional outcome after 7 days.

Patients with higher total bilirubin level (i.e in the second and third group of total bilirubin) at admission had severe stroke(p0.0003) and it was positively correlated with significance.

This is supported by the study conducted by Tian Xu et al , concludes that elevated serum total bilirubin positively correlates with stroke severity

Patients with higher total bilirubin level (i.e in the second and third group of total bilirubin) at admission had poor outcome and it was positively correlated with significance. (p 0.0002)

Most of the studies conducted on ischemic stroke and bilirubin found statistically significant association between admission bilirubin and short term clinical outcome.

Arsalan et al found stastically significant correlation between bilirubin and short term outcome.

In the study, direct bilirubin levels were not correlated both with severity and functional outcome. But this result is not supported by any of the previous similar studies.

Sandra Pineda et al concluded higher admission direct bilirubin is associated with greater stroke severity.

In the study, indirect bilirubin levels were elevated in cases significantly(p0.001) and also correlates with stroke severity(p0.003) and functional outcome.(p0.001).

In the study the mean AST, ALT between two groups didn't have any stastistically significant differences. This association has not been analysed extensively.

In the study, there is a positive correlation between severe stroke and poor outcome and it is statistically significant.

Above findings support the hypothesis that serum bilirubin might act as the endogenous antioxidant, but during the acute phase of stroke it acts as the marker of oxidative stress damage.

Whatever the preventive or destructive function in ischemic stroke, bilirubin is elevated in ischemic stroke which is related to stroke severity and poor functional outcome and bilirubin can act as the marker of oxidative stress.

So further study in the role of bilirubin in the pathogenesis of ischemic stroke is warranted, as it may have numerous clinical implications.

LIMITATIONS OF THE STUDY

But the limitation of the study are hepatitis virus carriers were not excluded from the study considering the low incidence of carriers who didn't developed jaundice at least once in the life time in local population and second is the small study group.

CONCLUSION

In summary ,this study found that serum total and indirect bilirubin levels are higher in acute ischemic stroke patients. Serum total and indirect bilirubin levels were correlated positively and significantly with stroke severity NIHSS≥10 and poor short term functional outcome MRS ≥ 3.

CLINICAL IMPLICATIONS

Since bilirubin acts as the marker of oxidative stress in stroke patients, measures to reduce oxidative stress in patients with higher bilirubin will be taken as therapeutic measure to reduce morbidity and mortality.

And also with this basic investigation, severity and prognosis of stroke can be assessed and Ischemic stroke patients with higher bilirubin levels can be monitored to avoid complications.

BIBLIOGRAPHY

1. Association of Serum Bilirubin with Ischemic Stroke Outcomes. Sandra Pineda, BS, Oh Young Bang, MD, PhD and Bruce Ovbiagele, MD J Stroke Cerebrovasc Dis. 2008; 17(3): 147–152.
2. UN Chronicle Health Watch. Atlas of Heart Disease and Stroke. 2005; 0105: 46.
3. Warlow C, Sudlow C, Dennis M, Wardlaw J, Sandercock, P. Stroke. Lancet 2003; 362: 1211–24.
4. Reddy K, Yusuf S. Emerging epidemic of cardiovascular disease in developing countries. Circulation 1998; 97:596–601.
5. Davidson's principles and practices of Medicine, 20th edition.
6. Ischemic Stroke: Pathophysiology and Principles of Localization Editor: Alireza A tri, MD, PhD www.turner –white .com. Hospital Physician Board Review Manual
7. Harrisons Principles of Internal Medicine Volume 2, Page: 3275
8. Challa V. Atherosclerosis of the Cervicocranial arteries. In Toole JF (ed) erebrovascular disorders. 5th edition. Lippincott Williams and Wilkins, Philadelphia, 1999.
9. Glaglov S, Zarins CB. What are the determinants of plaque instability and its consequences? J Vasc Surg 198; 9: 389-390. 1.
10. Falk E. Why do plaques rupture? Circulation 1992; 86:30-42.

11. Garcia JH, Ho Khang-Loon, Pantoni L. Pathology in Barnett, Henry JM, Mohr JP, Stein BM, Yatsu FM (eds), Stroke Pathophysiology, Diagnosis and Management. Third edition, Philadelphia, PA: Churchill Livingston; 1998.

12. Lyden PD, Zivin JA. Hemorrhagic transformation after cerebral ischemia:Mechanisms and incidence. Cerebrovasc Brain Metab Rev.1993; 5:1-16.

13. Garcia JH, Anderson ML: Circulatory disorders and their effects on the brain.pp 715-822. In Davis RL, Robertson DM (eds): Textbook of Neuropathology, 3rd edition. Williams & Wilkins, Baltimore 1997

14. Fisher M., Schaebitz W. An overview of acute stroke therapy. Past, present, and future. Arch Intern Med 2000;160:3196-3206.

15. Lee J-M., Zipfel G.J., Choi D.W. The changing landscape of ischaemic brain injury. Nature 399:A7-A14; 1999

16. Labovitz DL, Boden-Albala B, Hauser WA, Sacco RL. Lacunar infarct or deep intracerebral hemorrhage: who gets which? The Northern Manhattan Study. Neurology 2007;68:606–8.

17. Bansal recent concept of stroke , Medicine update 135 -137 Guidelines for the Early Management of Patients With Acute Ischemic Stroke AHA/ASA Stroke guidelines

http://stroke.ahajournals.org/lookup/doi/10.1161/STR.

18. NIHSS, National Stroke Association Vol. XVI Issue 1, Jan,Feb 2006
19. MRS, The Lancet Neurology, Volume 5, Issue 7, Pages 603 - 612, July 2006. doi:10.1016/S1474-4422(06)70495-1
20. Sies H. Oxidative stress: introductory remarks. In: Oxidative Stress. New York: Academic Press, pp. 1–8 (1985).
21. Halliwell B. Antioxidant defence mechanisms: from the beginning to the end (of the beginning). Free Radical Res 31: 261-272 (1999).
22. Piantadosi CA, Zhang J. Mitochondrial generation of reactive oxygen species after brain ischemia in the rat. Stroke 27: 327-331 (1996).
23. Niizuma K, Endo H, Chan PH. Oxidative stress and mitochondrial dysfunction as determinants of ischemic neuronal death and survival. J Neurosci 109 (Suppl. 1): 133-138 (2009)
24. Piantadosi CA, Zhang J. Mitochondrial generation of reactive oxygen species after brain ischemia in the rat. Stroke 27: 327-331 (1996).
25. Niizuma K, Endo H, Chan PH. Oxidative stress and mitochondrial dysfunction as determinants of ischemic neuronal death and survival. J Neurosci 109 (Suppl. 1):133-138 (2009)
26. Orrenius S, Gogvadze V, Zhivotovsky B.: Mitochondrial oxidative stress: implications for cell death. Annu Rev Pharmacol Toxicol 47: 143-183 (2007).
27. Andreyev AY, Kushnareva YE, Starkov AA. Mitochondrial metabolism of reactive oxygen species. Biochemistry 70: 200-214 (2005).

28. Cooper AJL, Kristal BS.: Multiple roles of glutathione in the central nervous system. Biol Chem 378:793-802 (1997).

29. Kinuta Y, Kikuchi H, Ishikawa M, Kimura M, Itokawa Y. Lipid peroxidation in focal cerebral ischemia. J Neurosurg 71: 421-429 (1989). J Surg Res. 2001 Mar;96(1):44-9.

30. Increased urinary excretion of bilirubin oxidative metabolites in septic patients: a new marker for oxidative stress in vivo. Otani K[1], Shimizu S, Chijiiwa K, Yamaguchi K, Kuroki S, Tanaka M

31. Journal of Clinical Neuroscience Volume 14, Issue 11, November 2007, Pages 1062–1066. Oxidative stress in acute ischemic stroke. Ayca Ozkul, Ali Akyol, Cigdem Yenisey, Esra Arpaci**,** Nefati Kiylioglu**,** Cengiz Tataroglu

32. Oxidative Stress in the Context of Acute Cerebrovascular Stroke Mohsen Muhammad Hussein El Kossi and Madeha Mahrous Zakhary Stroke. 2000;31:1889-1892, doi:10.1161/01.STR.31.8.1889

33. Association between Oxidative Stress and Outcome in Different Subtypes of Acute Ischemic Stroke . Nai-Wen Tsai, Ya-Ting Chang, BioMed Research International Volume 2014 (2014), Article ID 256879, 7 pages

34. Oxidative stress markers and antioxidant level in neonatal hyperbilirubinemia , Nesrine Salah El-Din Abdel Hamid , Nahed Fahmy Helal ,

35. Yamaguchi T, Horio F, Hashizume T, Tanaka M, Ikeda S, Kakinuma A, Nakajima H. Bilirubin is oxidized in rats treated with endotoxin and acts as a physiological antioxidant synergistically with ascorbic acid in vivo. Biochem Biophys Res Commun 1995;214:11-19.

36. Cristina Bellarosa, Unconjugated bilirubin mediated oxidative stress, ER stress and activation of Nrf2 pathwayQaisiya M.; Coda Zabetta C., Bellarosa C., Tiribelli C. "Bilirubin mediated oxidative stress involves antioxidant response activation via Nrf2 pathway" Cellular Signalling , Volume 26, Issue 3, March 2014, Pages.

37. Maria Alexandra Britoa, Dora Britesa, D. Allan Butterfieldb. Centro de Patoge'nese Molecular-UBMBE, Faculdade de Farma'cia, University of Lisbon, Av. Das Forc.as Armadas, 1600-083 Lisbon, Portugal Department of Chemistry, Center of Membrane Sciences and Sanders-Brown Center of Aging, University of Kentucky, Lexington, KY 40506, USA. Accepted 31 July 2004, Available online 13 September 2004512-520

38. Schwertner HA, Jackson WG, Tolan G. Association of low serum concentration of bilirubin with increased risk of coronary artery disease. Clin Chem 1994;40:18-23.

39. Wu TW. Is serum bilirubin a risk factor for coronary artery disease?. Clin Chem 1994;40:9-10.

40. Hopkins PN, Wu LL, Hunt SC, James BC, Vincent GM, Williams RR. Higher serum bilirubin is associated with decreased risk for early familial coronary artery disease. Arterioscler Thromb Vasc Biol 1996;16:250-255.

41. Breimer LH, Wannamethee G, Ebrahim S, Shaper AG. Serum bilirubin and risk of ischemic heart disease in middle-aged British men. Clin Chem 1995;41:1504-1508.

42. Psychiatry Clin Neurosci. 2013 May;67(4):259-64. doi: 10.1111/pcn.12051.Association between high serum total bilirubin and post-stroke depression. Tang WK, Liang H, Chu WC, Mok V, Ungvari GS, Wong KS.

43. Dohi K et al. Transient elevation of serum bilirubin level in hemorrhagic stroke - bilirubin is marker of oxidative stress. Acta Neurochir Suppl. 2003;86:247-9

44. Stocker R, Yamamoto Y et al. Bilirubin is antioxidant for possible physiological importance. Science. 1987 Feb 27;235(4792):1043-6

45. Clinical Implications of Bilirubin-Associated Neuroprotection and Neurotoxicity. Nicholas V Mendez, Jeffrey A Wharton, Jenna L Leclerc, Spiros L Blackburn, Martha V Douglas-Escobar, Michael D Weiss, Christoph N Seubert and Sylvain DorÃ

46. Role of Oxidative Stress in Pathophysiology of Transient Ischemic Attack and Stroke. Jaspreet Kaur , Sarika Arora, Bhawna Singh ,LC Thakur , J Gambhir , KM Prabhu Int J Biol Med Res. 2011; 2(3): 611-615.

47. Antioxidant and cytotoxic effects of bilirubin on neonatal erythrocytes. Mireles LC[1], Lum MA, Dennery PA. Pediatr Res. 1999 Mar;45(3):355-62.

48. Brain Res. 2004 Nov 5;1026(1):33-43. A link between hyperbilirubinemia, oxidative stress and injury to neocortical synaptosomes. Brito MA, Brites D, Butterfield DA.

49. The Bilirubin Level is Negatively Correlated with the Incidence of Hypertension in Normotensive Korean Population. Ho Jun Chin, Young Rim Song, and Suhnggwon Kim. J Korean Med Sci. Jan 2009; 24(Suppl 1): S50–S56. Can J Neurol Sci. 2013 Jan;40(1):80-4.

50. Association of serum bilirubin with stroke severity and clinical outcomes. Xu T, Zhang J, Xu T, Liu W, Kong Y, Zhang Y.

51. Low Serum Bilirubin Level as an Independent Predictor of Stroke Incidence. A Prospective Study in Korean Men and Women Heejin Kimm, MD, PhD; Ji Eun Yun, PhD; Jaeseong Jo, BS; Sun Ha Jee, PhD

52. Perlstein TS, Pande RL, Beckman JA, Creager MA. Serum total bilirubin level and prevalent lower-extremity peripheral arterial disease: National Health and Nutrition Examination

53. Ayub Med Coll Abbottabad 2011;23(2) Prognostic Significance Of Serum Bilirubin In Stroke Arsalan, Muhammad Ismail, Muhammad Bilal Khattak, Faramoz Khan,Muhammad Jalil Anwar, Zeeshan Murtaza, Abuzar Khan

54. Elevated bilirubin after acute ischemic stroke linked to the stroke severity Yun Luo, Jingwei Li, Junfeng Zhang, Yun Xu Show more DOI: 10.1016/j.ijdevneu.2013.08.002

55. Correlation of Serum Total Bilirubin Levels and the Severity of Acute Ischemic Stroke. Shin SK, Lee YB, Shin DJ, Park HM, Park KH, Seong YH, Kim JH, Lim EK, Park CW.

56. Lipincott – Medical Physiology – 2nd Edition

57. Wu TW. Is serum bilirubin a risk factor for coronary artery disease?. Clin Chem 1994;40:9-10.

58. Platt JL, Nath KA. Heme oxygenase: protective gene or Trojan horse. Nat Med 1998;4:1364-1365.

59. Ross R. Atherosclerosis–an inflammatory condition. N Engl J Med 1999;340:115-126.

60. Molla, M., M. Gironella, R. Miquel, V. Tovar, P. Engel, A. Biete, J. M. Pique, J. Panes. 2003. Relative roles of ICAM-1 and VCAM-1 in the pathogenesis of experimental radiation-induced intestinal inflammation. Int. J. Radiat. Oncol. Biol. Phys.

ANNEXURES

ABBREVIATION

Accronym	Abbreviation	Accronym	Abbreviation
ACA	Anterior Cerebral Artery	MDA	Malondialdehyde
AIS	Acute Ischemic Stroke	MI	MyocardialInfarction
ALT	Alanine Transaminase	MRA	Magnetic Resonance Angiography
AST	Aspartate Transaminase	MRI	Magnetic Resonance Imaging
BOX	Bilirubin Oxidative Metabolites	MRS	Modified Rankin Scale
B-UGT	Bilirubin-UDP Glucouronosyl Transferas	NIHSS	National Institure of Health Stroke Scale
CAD	Coronary Artery Disease	NMDAR	NMDA Glutamate Receptor
CADASIL	Cerebral autosomal dominant arteriopathy with subcortical infarcts and leukoencephalopathy	nNOS	Neuronal Nitric Oxide Synthase
CNS	Central Nervous System	NO	Nitric Oxide
CO	Carbon Monoxide	NOX	NADPH Oxidase
CSF	CerebroSpinal Fluid	O2-	Super Oxide Anion
CT	Computed Tomography	OH-	Hydroxyl Radical
CTA	Computed Tomography Angiography	ONOO-	Peroxy Nitrite
DB	Direct Bilirubin	PCA	Posterior Cerebral Artery
DIC	Disseminated Intravascular Coagulation	PET	Postitron Emission Tomography
DM	Diabetes Mellitus	PTT	Partial Thromboplastin Time
EDH	Extradural Hemorrhage	ROS	Reactive Oxygen Species
ES	Embolic Stroke	rt-PA	Recombinant Tissue - Plasminogen Activator
ETC	Electron Transport Chain	SAH	Sub Arachnoid Hemorrhage
G6PD	Glucose 6 Phosphate Dehydrogenase	SDH	Subdural Hemorrhage
HDL	High Density Lipoprotein	SHT	Systemic Hypertension
HI	Hemorrhagic Infarction	SLE	Systemic Lupus Erythematosus
HO	Hemeoxygenase	SPECT	Single-photon emission computed tomography
IB	Indirect Bilirubin	TB	Total Bilirubin
ICH	Intra Cranial Hemorrhage	TC	Total Cholesterol
ICT	Intra Cranial Tension	TG	Triglyceride
INR	International Normalized Ratio	TIA	Transient Ischemic Attack
LDL	Low Density Lipoprotein	TTP	Thrombotic Thrombocytopenic Purpura
MCA	Middle Cerebral Artery	VCAM	Vascular Cell Adhesion Molecule

PROFORMA

NAME	IP NO	SL. NO:

AGE :

SEX :

OCCUPATION :

ADDRESS :

DATE AND TIME OF STROKE :

DATE AND TIME OF ADMISSION:

DATE OF DISCHARGE :

CT/MRI BRAIN:INFARCT : Y/N

LOCATION : ACA/MCA/PCA [Water Shed,

/Global/Lacunar/Others]

Bilirubin:

TOTAL BILIRUBIN	
DIRECT	
INDIRECT	

NIHSS SCORE AT THE TIME OF ADMISSION (Severity Assessment):

MRS AT 7TH DAY OF STROKE(Prognosis Assessment):

MASTER CHART: CASES

S.No	NAME	AGE	SEX	SMOKING	BMI	DM	SHT	BP	LIPID PROFILE				LIVER ENZYMES		BILIRUBIN			TERRITORY	NIHSS	MRS
									TC	HDL	LDL	TG	AST	ALT	TB	DB	IB			
1	KATHARAVAYAN	60	M	-	23.5	✓	-	130/80	213	34	130	265	38	50	0.9	0.5	0.4	MCA	5	1
2	ARUMUGAM	52	M	-	21.5	✓	-	120/80	122	35	70	54	16	24	1.1	0.3	0.8	MCA	9	3
3	ASAITHAMBI	45	M	✓	21.8	-	-	120/70	97	24	65	92	12	16	0.9	0.2	0.7	MCA	8	3
4	RADHA	73	F	-	23.8	-	-	130/70	159	38	97	100	14	28	1.4	0.8	0.6	MCA	28	6
5	MANI	48	M	-	20.3	-	-	140/96	150	50	76	72	18	22	0.5	0.2	0.3	MCA	7	1
6	PADMAVATHI	70	F	-	23.4	✓	✓	150/96	154	36	82	106	36	50	1.6	0.2	1.4	MCA	30	5
7	SUDHAKAR	33	M	-	29.8	✓	-	140/86	216	30	146	218	10	14	0.8	0.1	0.7	MCA	25	4
8	RAVI	55	M	-	21.5	-	✓	160/90	220	56	130	150	17	26	0.6	0.1	0.5	ACA	9	2
9	KRISHNAMOORTHY	60	M	✓	30.4	✓	✓	160/86	226	35	164	132	12	12	1.5	0.3	1.2	VBA	29	4
10	JAYAKUMAR	48	M	-	22	-	✓	140/96	196	48	126	146	16	8	0.9	0.2	0.7	MCA	7	2
11	SEKAR	45	M	✓	23.2	-	✓	150/90	156	36	98	160	14	18	0.9	0.1	0.8	MCA	12	3
12	THIRUMAL	73	M	-	19.5	-	✓	160/100	140	30	94	86	16	28	1.4	0.2	1.2	MCA	26	4
13	KRISHNAMOORTHY	65	M	-	18.6	-	✓	140/90	186	40	128	112	9	16	0.8	0.1	0.7	MCA	11	5
14	SEKAR	55	M	✓	26.5	-	-	130/86	216	30	146	148	30	42	0.8	0.2	0.6	MCA	10	2
15	DURAISAMY	72	M	-	21	-	-	150/100	150	45	86	72	9	16	0.8	0.1	0.7	MCA	8	3
16	SATHYA	37	M	✓	22	-	-	160/90	194	34	124	74	12	15	1.5	0.8	0.7	MCA	28	4
17	SIVALINGAM	66	M	-	28.5	-	-	120/76	176	40	112	124	16	20	1.0	0.5	0.5	MCA	25	5
18	DHEVASIGAMANI	57	M	✓	22.4	-	-	140/96	142	30	100	112	12	24	1.0	0.5	0.5	VBA	8	3

19	KUPPAMMAL	50	F	-	20	✓	✓	160/100	226	34	160	136	28	48	0.6	0.1	0.5	MCA	5	3
20	ARUMUGAM	70	M	-	26.6	-	✓	150/96	174	38	98	96	10	16	1.3	0.7	0.6	ACA+MCA	28	6
21	MANGAMMAL	75	F	-	20	✓	-	120/80	156	44	96	230	13	24	1.2	0.2	1	MCA	26	6
22	MUNUSAMY	70	M	-	21	-	-	110/86	98	28	68	104	12	28	0.7	0.4	0.3	MCA	10	2
23	AJEETHA BI	80	F	-	19.5	✓	✓	130/94	196	36	146	168	34	46	0.9	0.1	0.8	MCA	9	3
24	SELVARAJ	55	M	-	23	-	-	110/70	174	60	85	44	10	24	0.7	0.2	0.5	MCA	10	3
25	SRINIVASAN	57	M	✓	23	-	-	120/80	100	26	48	164	11	19	0.7	0.4	0.3	MCA	31	4
26	KANNAN	75	M	-	19.5	-	-	110/76	97	24	64	96	10	28	0.9	0.1	0.8	MCA	13	3
27	PADMA	65	F	-	24	-	-	120/86	138	30	84	164	10	16	1.0	0.1	0.9	MCA	31	5
28	SRIDHAR	55	M	✓	21.5	✓	-	140/96	146	32	86	140	16	18	0.8	0.1	0.7	MCA	16	3
29	BASKAR	39	M	✓	20.4	-	-	110/80	146	30	94	104	12	14	0.7	0.1	0.6	MCA	8	4
30	SYED MEERA	69	F	-	26	-	-	110/86	114	35	72	56	12	24	0.8	0.4	0.4	MCA	9	1
31	PAZHANI	48	M	-	21	✓	-	120/84	136	39	74	78	18	20	0.9	0.1	0.8	MCA	21	3
32	CHENGAN	83	M	-	28.4	-	✓	150/90	150	34	102	101	10	28	0.9	0.2	0.7	MCA	7	2
33	SALAMUDEEN	55	M	-	23.8	✓	✓	160/90	146	34	80	110	12	24	0.8	0.2	0.6	VBA	15	5
34	RAVI	60	M	-	23.5	✓	-	120/80	160	36	120	94	34	54	1.7	0.3	1.4	VBA	31	4
35	RAMEEZ B	60	F	-	21	-	✓	160/100	194	40	80	104	12	24	0.8	0.1	0.7	MCA	19	3
36	RAMACHANDRAN	76	M	-	28.4	-	-	140/96	120	30	64	96	9	12	0.9	0.2	0.7	MCA	25	4
37	SRINIVASAN	68	M	-	22.5	-	-	120/70	184	38	124	96	14	14	0.6	0.1	0.5	MCA	6	2
38	VELU	55	M	✓	20.5	-	-	160/100	194	46	120	104	10	22	1.0	0.1	0.9	ACA	30	4
39	RAJENDRAN	50	M	-	23.5	-	-	110/80	206	44	142	96	8	10	0.8	0.4	0.4	MCA	11	2
40	PARVATHI	60	F	-	26.6	-	✓	170/106	148	30	90	74	18	16	1.8	0.4	1.4	MCA	27	6

41	ANDAAL AMMA	80	F	-	19.5	-	-	120/70	198	42	100	74	20	30	1.5	0.8	0.7	MCA	30	5
42	GOPAL	50	M	-	24	-	✓	180/100	150	30	108	76	12	22	1.1	0.3	0.8	VBA	22	3
43	SUBRAMANI	60	M	-	22	-	-	100/80	124	26	84	142	9	12	0.5	0.1	0.4	MCA	4	1
44	GOTHANDAM	50	M	✓	23.5	-	-	120/76	140	30	94	112	12	10	1.0	0.2	0.8	MCA	30	4
45	MANIKKAM	84	M	-	24	-	-	110/80	180	60	92	76	9	18	0.9	0.5	0.4	MCA	8	3
46	ARUMUGAM	56	M	✓	19	-	✓	140/96	136	50	76	80	11	24	0.8	0.1	0.7	VBA	9	2
47	KANNAGI	50	F	-	22.5	-	-	110/70	186	36	80	142	13	28	1.0	0.2	0.8	MCA	26	3
48	MALLIGA	54	F	-	32.5	✓	✓	160/96	184	38	122	112	10	9	1.7	0.3	1.4	VBA	29	5
49	GOTHANDAM	58	M	-	26.7	✓	-	130/84	190	44	108	90	9	12	0.9	0.1	0.8	MCA	10	2
50	KANNAN	54	M	-	23.5	-	✓	104/80	132	32	80	120	6	18	0.5	0.1	0.4	ACA+MCA	7	1

Data	Representation	Parameters	Units	Parameters	Units
✓	PRESENT	TC	mg/dL	AST	IU/L
-	ABSENT	HDL	mg/dL	ALT	IU/L
		LDL	mg/dL		
		TG	mg/dL		
		TB	mg/dL		
		IB	mg/dL		
		DB	mg/dL		

MASTER CHART: CONTROLS

S.No	NAME	AGE	SEX	SMOKING	BMI	DM	SHT	BP	LIPID PROFILE				LIVER ENZYMES		BILIRUBIN		
									TC	HDL	LDL	TG	AST	ALT	TB	DB	IB
1	ANNAMALI	59	M	✓	24.6	✓	-	110/70	194	46	108	93	40	54	0.9	0.2	0.7
2	KUPPUSAMY	54	M	-	23.4	✓	-	120/70	180	38	118	116	38	43	1.2	0.3	0.9
3	CHINARASU	48	M	-	17.9	-	-	110/80	146	42	100	68	14	20	0.4	0.1	0.3
4	PONNAMAL	76	F	-	17.8	-	-	130/70	102	36	64	74	9	14	0.7	0.3	0.4
5	DEVARAJ	50	M	-	23.7	-	-	126/76	156	37	98	102	16	20	0.4	0.2	0.2
6	MANIMEGALAI	69	F	-	21.6	✓	✓	120/70	206	38	110	126	9	18	0.8	0.3	0.5
7	KARUNAKARAN	38	M	-	30.4	✓	-	110/80	166	39	106	114	12	30	0.8	0.2	0.6
8	MUTHUSELVAN	58	M	✓	24.6	-	✓	130/90	176	38	112	10	22	30	0.6	0.3	0.3
9	RAMALINGAM	58	M	-	24.8	✓	✓	120/90	174	51	106	91	26	38	0.5	0.1	0.4
10	SUBBAYA	45	M	-	21.8	-	✓	130/86	196	32	134	186	16	19	0.9	0.3	0.6
11	SIVAKUMAR	42	M	✓	26.8	-	✓	140/90	156	38	102	76	8	17	0.5	0.2	0.3
12	PURUSHOTHAMAN	70	M	-	27.2	-	✓	120/70	154	34	101	104	14	22	0.4	0.2	0.2
13	PICHAIMUTHU	68	M	✓	22.6	-	✓	110/70	132	32	84	70	6	12	0.8	0.2	0.6
14	MATHIAZHAGAN	56	M	-	22.4	-	-	120/80	162	52	84	77	20	24	0.4	0.1	0.3
15	CHAKRAVARTHY	70	M	✓	20.6	-	-	110/80	168	42	102	111	16	22	0.5	0.1	0.4
16	VISWANATHAN	38	M	-	23.8	-	-	120/86	140	50	86	71	24	40	1.0	0.5	0.5
17	THANDAPANI	67	M	✓	20.6	-	-	130/80	136	48	84	92	9	16	0.8	0.5	0.3
18	SUDHAKAR	56	M	-	21.2	-	-	110/80	190	38	124	96	9	12	0.7	0.3	0.4
19	TAMILARASI	49	F	-	24.8	✓	✓	130/80	199	32	120	102	12	24	0.7	0.3	0.4
20	THANGAPPAN	68	M	-	19.8	-	✓	140/90	146	34	96	121	18	28	0.3	0.1	0.2

21	INDIRANI	70	F	-	21	✓	-	120/70	186	32	98	100	20	24	1.1	0.5	0.6
22	SUBBURAYAN	69	M	✓	23.8	-	-	120/76	140	48	86	76	20	24	0.6	0.3	0.3
23	NADIYAMMAL	72	F	-	26	✓	✓	140/96	176	34	104	78	6	18	0.7	0.3	0.4
24	SIVALINGAM	53	M	-	22.7	-	-	130/80	140	50	76	84	12	16	0.4	0.1	0.3
25	GURUMOORTHY	54	M	-	22.3	-	-	110/74	104	37	54	88	14	18	0.4	0.1	0.3
26	KUMAR	72	M	-	24	-	-	120/84	179	35	120	94	18	30	0.5	0.1	0.4
27	SUNDARI	64	F	-	16.6	-	-	110/70	138	34	88	61	16	20	0.4	0.3	0.1
28	KANNAN	53	M	-	22.7	✓	-	120/80	250	29	104	136	12	16	1.1	0.2	0.9
29	BALAMURUGAN	38	M	✓	19.4	-	-	120/70	144	30	92	102	10	14	0.6	0.3	0.3
30	MUTHAMMAL	62	F	-	23.4	-	-	110/74	218	50	138	90	12	18	0.8	0.2	0.6
31	GURUMOORTHY	50	M	-	21.2	✓	-	120/86	180	32	141	116	8	12	0.3	0.1	0.2
32	CHENGAN	80	M	✓	20	-	✓	140/90	180	40	116	168	6	14	0.7	0.3	0.4
33	JEYARAJ	59	M	-	22.6	✓	✓	130/76	198	42	128	156	48	56	0.4	0.1	0.3
34	KALIMUTHU	54	M	-	21.2	✓	✓	110/70	190	38	144	190	12	20	0.4	0.2	0.2
35	RASIYA BEGAM	58	F	-	23	-	✓	120/86	112	32	70	100	14	18	1.3	0.3	1.0
36	SUBRAMANI	70	M	✓	21.2	-	-	110/70	148	30	94	134	9	22	0.7	0.3	0.4
37	NATARAJAN	65	M	-	19.2	-	-	120/84	150	44	89	64	22	40	0.5	0.1	0.4
38	THANIGACHALAM	53	M	-	21.2	-	-	110/76	154	42	104	77	16	22	0.7	0.3	0.4
39	DEVENDRAN	48	M	✓	18.8	-	-	130/94	142	46	86	74	14	18	0.4	0.1	0.3
40	SUBBULAKSHMI	58	F	-	29.6	-	✓	110/70	170	36	109	112	42	36	0.9	0.4	0.5
41	SEETHAMMAL	72	F	-	19.2	-	-	120/70	130	38	84	66	13	12	0.4	0.1	0.3
42	DURAI	48	M	-	18.2	-	✓	130/86	132	34	82	88	24	40	0.7	0.3	0.4
43	JEYASEELAN	58	M	✓	18.8	-	-	110/74	160	30	96	74	12	38	0.8	0.5	0.3
44	KANNIAPPAN	48	M	-	19.3	-	-	110/86	130	40	76	72	9	12	0.5	0.2	0.3

45	SATHYAMOORTHY	62	M	✓	17.3	-	-	140/86	146	44	64	76	8	14	0.3	0.15	0.15
46	PANNEER SELVAM	58	M	-	19.4	-	✓	110/70	164	50	112	70	12	24	1.0	0.5	0.5
47	LAKSHMI	56	F	-	26.4	-	-	130/90	204	38	140	94	6	13	0.4	0.1	0.3
48	SUNDARAMMAL	55	F	-	30.5	✓	✓	150/100	170	44	112	164	60	54	0.6	0.2	0.4
49	RAJA SEKAR	60	M	-	23.3	✓	-	110/70	146	30	92	112	16	18	0.7	0.3	0.4
50	SRIDHARAN	56	M	✓	29.4	-	✓	130/96	196	44	126	150	9	12	0.4	0.1	0.3

Data	**Representation**	**Parameters**	**Units**	**Parameters**	**Units**
✓	PRESENT	TC	mg/dL	AST	IU/L
-	ABSENT	HDL	mg/dL	ALT	IU/L
		LDL	mg/dL		
		TG	mg/dL		
		TB	mg/dL		
		IB	mg/dL		
		DB	mg/dL		

நோயாளி ஒப்புதல் படிவம்

ஆராய்ச்சியின் விவரம்: குருதி ஊட்டக்குறை பக்கவாத நோயாளிகளின் இரத்த நீர்த்த பித்தச்செம்பசை அளவு (சீரம் பில்லிரூபின்) மற்றும் அவற்றின் தொடர்பினை ஆராய்யும் ஆய்வரிக்கை.

ஆராய்ச்சி மையம்: அரசு கீழ்பாக்கம் மருத்துவக் கல்லூரி மருத்துவமனை

நோயாளியின் பெயர்: நோயாளியின் வயது:

பதிவு எண்:

நோயாளி கீழ்கண்டவற்றுள் கட்டங்களை (✓) செய்யவும்

1. மேற்குறிப்பிட்டுள்ள ஆராய்ச்சியின் நோக்கத்தையும் பயனையும் முழுவதுமாக புரிந்துகொண்டேன். மேலும் எனது அனைத்து சந்தேகங்களையும் கேட்டு அதற்கான விளக்கங்களையும் தெளிவுபடுத்திக் கொண்டேன். ☐
2. மேலும் இந்த ஆராய்ச்சிக்கு எனது சொந்த விருப்பத்தின் பேரில் பங்கேற்கிறேன் என்றும், மேலும் எந்த நேரத்திலும் எவ்வித முன்னறிவிப்புமின்றி இந்த ஆராய்ச்சியிலிருந்து விலக முழுமையான உரிமை உள்ளதையும், இதற்கு எவ்வித சட்ட பிணைப்பும் இல்லை என்பதையும் அறிவேன். ☐
3. ஆராய்ச்சியாளரோ, ஆராய்ச்சி உதவியாளரோ, ஆராய்ச்சி உபயத்தாரரோ, ஆராய்ச்சி பேராசிரியரோ, ஒழுங்குநெறி செயற்குழு உறுப்பினர்களோ எப்போது வேண்டுமானாலும் எனது அனுமதியின்றி எனது உள்நோயாளி பதிவுகளை இந்த ஆராய்ச்சிக்காகவோ அல்லது எதிர்கால பிற ஆராய்ச்சிகளுக்காகவோ பயன்படுத்திக்கொள்ளலாம் என்றும் மேலும் இந்த நிபந்தனை நான் இவ்வராய்ச்சியிலிருந்து விலகினாலும் தகும் என்றும் ஒப்புக்கொள்கிறேன். ஆயினும் எனது அடையாளம் சம்பந்தப்பட்ட எந்த பதிவுகளும் (சட்டபூர்வமான தேவைகள் தவிர) வெளியிடப்படமாட்டது என்ற உறுதிமொழியின் பெயரில் இந்த ஆராய்ச்சியிலிருந்து கிடைக்கப்பெறும் முடிவுகளை வெளியிட மறுப்பு தெறிவிக்கமாட்டேன் என்று உறுதியளிக்கின்றேன். ☐
4. இந்த ஆராய்ச்சிக்கு நான் முழுமனதுடன் சம்மதிக்கின்றேன் என்றும் மேலும் ஆராய்ச்சிக் குழுவினர் எனக்கு அளிக்கும் அறிவுரைகளை தவறாது பின்பற்றுவேன் என்றும் உறுதியளிக்கின்றேன். ☐
5. இந்த ஆராய்ச்சிக்குத் தேவைப்படும் அனைத்து மருத்துவப் பரிசோதனைகளுக்கும் ஒத்துழைப்பு தருவேன் என்று உறுதியளிக்கின்றேன். ☐
6. இந்த ஆராய்ச்சிக்கு யாருடைய வற்புறுத்தலுமின்றி எனது சொந்த விருப்பத்தின் பேரிலும் சுயஅறிவுடனும் முழுமனதுடனும் சம்மத்திக்கின்றேன் என்று இதன் □ லம் ஒப்புக்கொள்கிறேன். ☐

நோயாளியின் கையொப்பம் / பெருவிரல் கைரேகை:

இடம்: தேதி:

ஆராய்ச்சியாளரின் கையொப்பம்:

இடம்: தேதி:

INSTITUTIONAL ETHICAL COMMITTEE
GOVT.KILPAUK MEDICAL COLLEGE,
CHENNAI-10
Ref.No.1589/ME-1/Ethics/2014 Dt:06.03.2014.
CERTIFICATE OF APPROVAL

The Institutional Ethical Committee of Govt. Kilpauk Medical College, Chennai reviewed and discussed the application for approval "A Study on association between serum bilirubin and acute ischemic stroke and its prognostic significance" - For Project work submitted by Dr.A.Ramya, (21.07.1986), MD (GM), PG Student, KMC, Chennai-10.

The Proposal is APPROVED.

The Institutional Ethical Committee expects to be informed about the progress of the study any Adverse Drug Reaction Occurring in the Course of the study any change in the protocol and patient information /informed consent and asks to be provided a copy of the final report.

GOVT. KILPAUK MEDICAL COLLEGE
29 MAY 2014
Kilpauk, Chennai-10.

29/5/14

CHAIRMAN,
Ethical Committee
Govt.Kilpauk Medical College,Chennai