INTRODUCTION

Human body is composed of delicate, contiguous, organ systems. In the healthy state of body, there is complete harmony in structure and functions among organs, thus organ systems. An acute disorder will temporarily derange the functioning of an organ system and its interrelated body part without having any permanent effect on the structure of organs. Acute viral infection of upper respiratory tract produces signs and symptoms for the short period, thereafter; body comes to an healthy state. The chronic disease will have its effect on body organs in terms of structure and functions. The repercussion of a chronic disorder is evident both in the organ system in which a disease starts as well as in the correlated system. Chronic Gastroesophageal reflex disease exhibits long lasting ramifications in the gastrointestinal system and also in the oral tissues due to acidic regurgitation.

Liver is the largest organ of human body having reddish brown colour. It weighs about 1-1.5 kg in adult human. It is a versatile organ involved in metabolism and various other biochemical functions. Liver diseases, in general can be classified into three categories.

1. Hepatocellular: This category encompasses diseases like viral hepatitis, alcoholic hepatitis, drug induced hepatitis etc. Hepatocellular diseases show the features of liver injury and necrosis.
2. Cholestatic (obstructive): The feature like bile flow obstruction predominates in the diseases like gall stone, malignancy and primary biliary cirrhosis.
3. Mixed type: The features of above two types of diseases are present in this category (Ghany, 2008).

NEED OF STUDY

1. In day toy clinical practice, physicians and surgeons come across patients with different ailments. The patients with compromised Liver and Renal functions may present perplexing moments for doctors to decide about the dose of drug, nature of drug, safety of drug, safety of anesthetics to be used and the protocol to be followed to handle bleeding tendency in these patients. The proposed study would help evaluate the renal function in liver diseased patients.
2. Previously, very few studies were conducted to assess the biochemical parameters in different liver disorders and their covariation in renal dysfunction, if any.

3. There is no study undertaken in the region of Bhatinda (Punjab), to assess the biochemical markers of liver and renal functions in healthy and diseased individuals. Bhatinda is a city with a population of mixed culture. Varied multi speciality and superspeciality medical centres cater to the medical needs of people. There is an urgent need to have a reference range of biomarkers of liver and renal functions in this region.

SCOPE OF THE STUDY

1. The study will provide useful information’s regarding the prevalence of renal dysfunctions, if any, in patients with liver diseases.

2. The study will also provide data about the biomarkers of liver and renal functions. This knowledge will be immensely helpful for doctors in daily clinical settings to stage and grade the liver diseases as well as to assess the renal status. The data will be helpful to modify medicine dose and adopt necessary measures in order to prevent drug toxicity and prolonged bleeding in liver and renal compromised patients.

3. Nevertheless, some new concepts and findings will emerge in this study that will expand the knowledge of surgeons and fellow researchers to carry out more intensive research work.

STATEMENT OF THE PROBLEM

To assess the renal function abnormality, if any, in patients inflicted with liver diseases and to find out if stage and grade of liver disorder (as determined by clinical biomarkers) effect the kidney functions. Therefore, the study problem is titled as “Assessing Renal function in patients suffering from liver diseases.”

AIM    OBJECTIVES         HYPOTHESES

AIM: Present study is to assess renal function in patients with liver diseases. OBJECTIVES: The following objectives are to be fulfilled by the present study.

1. To estimate Glomerular filtration rate of patients with liver disease.
2. To estimate Serum Creatinine of patients with liver disease.

3. To assess Blood urea of patients with liver disease.

4. To evaluate Blood urea nitrogen (BUN) to Creatinine ratio (BUN/C) of patients with liver disorder.

5. To estimate Serum Electrolytes of liver disease patients.

6. To assess liver functions by assaying biochemical parameters as serum bilirubin, serum albumin, serum aminotransferases, serum prothrombin, INR, haemoglobin.

**HYPOTHESES OF THE STUDY :**

1. Patients with advanced liver disease (ascites cirrhosis/decompensated cirrhosis) will significantly suffer from renal dysfunction.

2. Glomerular filtration rate (estimated by MDRD equation ) in patients with renal dysfunction will show significant correlation with serum albumin and serum sodium.

3. Hypoalbuminemia and Hyponatremia (as determined from serum albumin and serum sodium concentrations) will be significantly observed in ascitic cirrhosis patients.

4. BUN/ Creatinine ratio will be significantly elevated in ascites cirrhosis patients.

5. Hepatorenal syndrome, Spontaneous bacterial peritonitis and Acute tubular necrosis will be the major causes of renal dysfunction in ascites cirrhosis.

6. Anemia (as determined by decline in haemoglobin value) will be significantly observed in ascites cirrhosis patients.

7. Renal dysfunction will not be observed in patients suffering from acute hepatitis and non ascitic cirrhosis (compensated ascites).
LITERATURE REVIEW

Hepatitis: It is the inflammation of liver characterized by the presence of inflammatory cells in the liver. It can be caused by viruses, toxins, drugs or alcoholic consumption. This disorder is in general, self limiting, but may progress to cirrhosis (Firpi, 2006).

Viral agents inducing hepatitis can be virus A, B, C, D or virus E. The hepatitis A virus and hepatitis E virus are transmitted from person to person by ingesting contaminated food and water. The mode of infection is through oro-faecal route. This type of infection is self limiting, but usually has a tendency to relapsing of symptoms within 6 months after termination of acute condition in 15% of population. Hepatitis B and C viruses can spread through the contact of blood, semen, vaginal fluid of the infected persons. This category of infection has a tendency to progress into fibrosis of liver. Hepatitis D virus infection occurs as a coinfection with HBV or as a superinfection to chronic hepatitis B individuals. The HDV would complicate the outcome of HBV infection (Centre for Disease control, 2011; WHO, 2011; Tong et al., 1995; Wikipedia, 2011).

The early symptoms of hepatitis starts with low grade fever, malaise, anorexia, nausea, vomiting, abdominal pain. These features continue for 4-6 days, subsequently, followed by icterus stage characterized by yellowish discolouration of skin, mucosa and sclera along with dark urine.

CIRRHOSIS: The term cirrhosis was first of all introduced by LAENNEC in 1826

Cirrhosis is the chronic, diffuse disorder of varied etiologies and is characterized by hepatic cell necrosis, proliferation of connective tissues and nodular regeneration, thereby leading to abnormal reconstruction of lobular architecture and disturbed hepatic circulation. Alcoholism and post hepatic sequelae are the commonest causes of cirrhosis.

Metabolic disorders like Haemochromatosis, Wilsons disease and prolonged cholestasis would progress to cirrhosis. Obese patients demonstrate non alcoholic fatty liver disease.

Clinical features of cirrhosis vary according to the severity of cirrhosis as seen in non ascites cirrhosis (compensated cirrhosis) and ascites cirrhosis (decompensated cirrhosis). The common symptoms observed are indigestion, flatulence, loss of weight, anemia, haematemesis, jaundice, palmer erythema, oedema of extremities, distension of abdomen, delirium, flapping tremors of
hands (Gunter, 2009; University of Maryland, 2010; National Digestive disease information clearing House, 2010).

About 40% of the cirrhotics are asymptomatic. A damage of about 80% of the liver parenchyma will result into clinical manifestations of ascites, skin changes, renal dysfunction, encephalopathy and thus, will end up into a stage called decompensated cirrhosis. About half the ascites cirrhosis patients die if the liver transplantation is not done (Watring et al., 2007).

PORTAL HYPERTENSION IN CIRRHOSIS

PHT is the high blood pressure in portal vein and its tributaries and is defined as the portal pressure gradient (difference in blood pressure between Portal vein and Hepatic vein) of 10 mm or greater than 10 mm of hg. (Wikipedia, 2011).

In normal liver, the intrahepatic resistance changes with the variations in portal blood flow. Since, cirrhosis eventually results into fibrosis, scarring and formation of regenerative nodules and altered state of hepatic circulation. These changes will raise intra hepatic resistance to flow of portal blood through liver [Carale, 2010]. These events predisposes to portal hypertension. Later on, GROSSMANN and co-worker changed this concept. They proposed the presence of increased number of specialized contractile elements in cirrhotic liver that would by way of their contraction, modulate the intrahepatic resistance to portal blood flow.

ROLE OF PERISINUSOIDAL HEPATIC STELLATE CELLS IN PORTAL HYPERTENSION

Hepatic stellate cells (HSC) are located in the perisinusoidal space of Disse. These cells constitute about 8% of total liver cells. They possess the properties of vitamin A storage, synthesis of matrix degrading metalloproteinase, cytokines, and growth factors under normal liver condition. In the diseased state, these cells undergo transition in their morphology and functions. The HSC, now in activated state, acquire myofibroblast like phenotype, thus having both pro-fibrogenic and contractile activities around sinusoidal space, it would thus narrow down the diameter of sinusoid. This phenomenon, collectively contribute to portal hypertension (Laleman, 2006; Carale, 2010). Blood from the portal vein is shunted away into systemic circulation by the formation of porto systemic collaterals. These collaterals drain away about 80% of the portal blood. Progressive shunting would aggravate hepatic hypoperfusion and a arterial and venous steal phenomenon is noticed (Newby et al., 2007).
SPLANCHNIC ARTERIAL VASODILATION

In cirrhosis, there is accumulation of vasodilating substances in the body. The most prominently influencing ones are, the Nitric oxide, Endothelin, Glucagon and Calcitonin gene related peptide (CGRP). These substances have pronounced vasodilating effect on the splanchnic circulation (Groszman, 1998). The assumption behind the elevated levels of vasodilating substances is the impaired hepatic metabolism and their escape through porto systemic collaterals into systemic circulation. The splanchnic vasodilation results into pooling of blood in the splanchnic vascular bed and subsequently, aggravates the portal hypertension (Bendt et al., 1991).

PERIPHERAL ARTERIAL VASODILATION HYPOTHESIS

It was proposed by SCHRIER and co worker in 1988. Peripheral arterial vasodilation is the earliest manifestation in cirrhosis and it would propose to renal sodium retention and diminished ability of kidney to excrete free water and thus, causing plasma volume expansion that would precede the development of ascites. This theory favours the “overfill hypothesis” (Schrier, 1988).

REDUCED EFFECTIVE CIRCULATORY VOLUME

Splanchnic and peripheral vasodilation reduce the effective circulatory blood volume, which in turn activates the high pressure baroreceptors located in carotid sinus and aortic arch. As a consequence, the sympathetic nervous system is stimulated and it induces the release of Norepinephrine from adrenal medulla. It is a potent vasoconstrictor and will affect the peripheral and renal circulation. The degree of activation of SNS is correlated to the severity of cirrhosis.

HYPERDYNAMIC CIRCULATION

Norepinephrine attaches to receptors located on S A node. It results into increased heart rate and cardiac output, decreased systemic vascular resistance, hypotension, and aggravates renal vasoconstriction. As the cirrhosis progresses, it favours splanchnic vasodilation, that interplays into a vicious circle to enhance the systemic vasodilation and renal vasoconstriction.

RENAL HYPOPERFUSION
State of reduced effective circulating volume influences the high pressure receptors in the juxtaglomerular apparatus of kidneys. Norepinephrine also aggravates the renal hypoperfusion by inducing renal vasoconstriction. This state of renal hypoperfusion and ischemia, activates Renin-Angiotensin-aldosterone system (RAAS). Despite the presence of circulating vasodilating substances, renal vasoconstriction continues and becomes intense as the cirrhosis advances and terminates into hepatorenal syndrome (Epstein et al., 1988).

The angiotensin II directly influences proximal tubule to reabsorb sodium and also induces the release of Aldosterone from adrenal cortex. It favours sodium retension through distal tubule. Anti diuretic hormone is released non-osmotically from hypothalamus-neurohypophysis. ADH results in water retension through collecting tubules.

**HAEMODILUTION**

The renal retension of free water and sodium raises the plasma volume, decreases plasma osmolarity and cause haemodilution. This favours a fall in serum albumin level, haemoglobin conc. The total body water rises more than total body sodium, favouring a rise in extra cellular volume, oedema formation and ascites (Gines et al., 2005). **ROLE OF NATRIURETIC PEPTIDES IN CIRRHOSIS** Atrial natriuretic peptides and brain natriuretic peptides are released in high concentration in cirrhosis. These peptides have natriuretic, vasorelaxing, RAAS-ADH-SNS inhibitory effect (Flora, 1990). The effects of these peptides are blunted in cirrhosis partly due to increased renal endopeptidase activity which degrade them, and partly to down regulation of renal natriuretic peptides receptors.

**ACUTE RENAL FAILURE**

Acute renal failure is characterized by rapid decline in glomerular filtration rate (GFR) over hours to days. It is usually asymptomatic and is diagnosed when biochemical investigation of hospitalized patients reveals a rise in blood urea nitrogen, serum creatinine concentrations. In general, the causes predominantly contribute to acute renal dysfunction are:

1. Renal hypoperfusion resulting in decreased glomerular filtration rate without any histological alteration in kidneys (Pre Renal ARF ≈55%).

2. Disorder that affect renal parenchyma (intrinsic ARF≈40%).
3. Disorder that cause urinary tract obstruction (Post renal ARF≈5%)  (Kathleen, 2008)

Renal failure was designated as renal dysfunction by ADQI work group in 2004. In an Indian tertiary hospital, the most common cause of acute renal dysfunction was Acute Tubular Necrosis (ATN≈44.4%), followed by pre renal azotemia (36.6%) and HRS (19.9%) (Madaan & Mehta, 2004).

HEPATORENAL SYNDROME

It is the functional renal failure that develops in patients with advanced cirrhosis of liver. It is characterized by intense vasoconstriction leading to low renal perfusion and fall in glomerular filtration rate. Renal ability to excrete free water and sodium is decreased in cirrhosis. Patients exhibit dilutional hyponatremia. Renal histology shows no lesion. Detailed description of HRS was given in 1950 by Sherlock, Paper, Vessin. Their studies emphasized the functional nature of renal failure, along with the co-existence of circulatory disturbances and poor prognosis of HRS. Investigations carried out during 1960-70 particularly by Epstein, showed renal failure in HRS due to extreme vasoconstriction in renal circulation.

HRS is a frequent problem seen in patients suffering from advanced liver disease and ascites. The incidence of HRS is estimated to be about 8% annually. It is associated with intense renal vasoconstriction and decreased renal perfusion and glomerular filtration rate. The renal ability to excrete sodium and free water is reduced and patients show dilutional hyponatremia (Arroyo et al., 2008).

Types of HRS:

HRS – I   This type has features of rapid onset and poor prognosis and is characterized by serum creatinine level more than 2.5 mg/dl in less than two weeks.

HRS – II  This type has features of slow onset and better prognosis and characterized by serum creatinine level more than 1.5 mg/dl (Gines et al., 1993).

SPONTANEOUS BACTERIAL PERITONITIS

It is an acute infection of ascitic fluid and is a common complication in ascites cirrhotic patients. The major proportion (70%) of the SBP patients fall in class C of Child Pugh Score. In ascites,
the frequency of SBP may be 18-30% in hospitalized patients. This disorder was first of all recognized by HAROLD CONN in 1960.

Enteric organisms have been isolated from more than 90% of SBP patients. The preponderance of enteric organisms in SBP has favoured the hypothesis of Bacterial Translocation where direct transmural migrations of microbes from an intestinal lumen take place. But the exact mechanism of bacterial inoculation of ascetic fluid is still a controversy. The key predisposing factors are attributed to lowered immune response in cirrhotic patients, impaired phagocytic function and decreased activity of reticulo endothelial system collectively result in proliferation of microbes and their reduced clearance from blood that would promote their migration into and proliferation within ascetic fluid (Syed et al., 2007).

ACUTE TUBULAR NECROSIS

It is the most common cause of acute renal dysfunction in cirrhosis. This disorder is the continuum of renal hypoperfusion as observed in pre renal failure. The prolonged renal ischemia results in renal tubule epithelial cell injury and cell death. Proximal tubule and ascending loop of Henle are the major areas of ischemic cell necrosis. ATN is manifested in three phases as initiation, maintenance and recovery phases. The initiation phase is characterized by apical blebs, loss of brush border margins, loss of polarity and integrity of tight junctions (Lerma, 2011).

CAUSES OF RENAL FAILURE IN CIRRHOSIS

Following causes based on clinical findings and analysis would classify renal failure as:

1. Renal failure associated with infection: In this category, the renal failure is considered secondary to an ongoing infection in the absence of other causes of renal failure. The most common infection is the spontaneous bacterial peritonitis and is diagnosed clinically by the presence of polymorphonuclear neutrophil count of more than 250/cumm in the ascites fluid (Rimola et al., 2000)

2. Hypovolumia related renal failure: It is diagnosed when patients had history of fluid losses (GIT and Renal losses) in the preceding days and absence of other causes of renal dysfunction (Morean and Lebrec, 2007).
3. Intrinsic renal failure: The cause of this form of renal failure is the alterations in the renal parenchyma. This is diagnosed clinically either by the presence of proteinuria > 500mg/24h, or RBC count > 50 under HPF, or presence of Casts as seen in urine sediment analysis (Pham et al., 2005).

4. Hepato renal syndrome: It is diagnosed when serum creatinine conc. rises > than 1.5 mg/dl, along with the exclusion of all other causes of renal dysfunction.

MATERIALS AND METHODS

RESEARCH DESIGN

Prospective, Observational, Single Centre, Case Control Study.

TIME SPAN OF RESEARCH WORK

The research work is expected to be complete within a duration of approximately two years starting from January, 2010.

PLACE OF STUDY

Study will be conducted in the Department of Gastroenterology and Hepatology, in Delhi Heart Institute and Research Centre, located at Bathinda (Punjab). It is a super-speciality centre.

SAMPLING DESIGN

Proportional stratified sampling method would be used to recruit study samples from the population.

PATIENT SELECTION CRITERIA

Patients would be diagnosed on the basis of case history, general physical examination and confirmation will be done by laboratory estimation of biochemical markers of liver and renal functions. The following criteria will be adopted for the inclusion and exclusion of patients as under:

Patient Inclusion Criteria:
1. Patients with acute hepatitis and cirrhosis of liver.

2. Patients of 18 years and above.

3. Patients of both gender will be eligible.

Patient Exclusion Criteria:

1. Patients of cholestatic liver disorder, liver Malignancy and non alcoholic fatty liver disorder.

2. Patients with history of any other systemic, metabolic or endocrinal disease.

3. Patients with history of AIDS.

4. Pregnant women.

5. Patients who will be involved in any other interventional clinical trial.

**PATIENTS STRATIFICATION CRITERIA**

Patients would be stratified on the basis of biochemical parameters of liver functions.

**CRITERIA FOR SELECTION OF CONTROLS**

Participants, equal to patients selected, from among the individuals who will accompany patients and volunteers will be selected as controls. The controls will be age and sex matched.

Control Inclusion Criteria

1. Absence of history of any systemic, metabolic or endocrinal disease.

2. Absence of any liver disease.

3. Controls will be from the hospital population.

4. Controls will be Age and Sex matched.

Control Exclusion Criteria

1. Presence of history of blood transfusion.

**STUDY VARIABLES**

Liver function parameters would be independent variables whereas Renal function parameters that would assess renal dysfunction, if any, will be taken as Dependent variables.

**INDEPENDENT VARIABLES**

Serum total Bilirubin, Serum Glutamate-Pyruvate Transaminase (SGPT), Serum Glutamate-Oxaloacetate Transaminase (SGOT), serum Albumin, INR, Haemoglobin concentration and Platelets count.

**DEPENDENT VARIABLES**

Glomerular filtration rate (by MDRD Equation), Serum Creatinine, Blood Urea, BUN/Creatinine ratio, Serum Sodium, Serum Potassium,

**DATA COLLECTION METHODS AND INSTRUMENTS**

Primary data will be collected from patients admitted in the ward of Gastroenterology, and from individuals, who will escort patients as well as from volunteers, for control group.

**INSTRUMENTS**

1. Structured interview schedule will be used for the collection of demographic data.

2. Direct observation and structured observation schedule will be utilized to collect data regarding clinical signs and symptoms.

3. Laboratory estimation of biochemical parameters will be done and data will be collected by pre-structured proforma.

**METHODS FOR ESTIMATION OF LIVER AND RENAL FUNCTION PARAMETERS**

1. **Estimation of Serum Bilirubin**: It will be done by Diazotised sulphanilic acid reaction method described by Van den Bergh.
PRINCIPLE: Serum is treated with Diazotized sulphanilic acid and this forms Azobilirubin complex. The direct bilirubin (conjugated) will react with diazo reagents directly whereas the unconjugated bilirubin will react only in the presence of an accelerator like caffeine-benzoate reagent. The azobilirubin so produced is purple in colour in acid medium. This colour is changed into blue by the addition of alkaline tartarate solution. This reaction is terminated by the addition of ascorbic acid (Ochei & Kolhatkar, 2000).

2. Estimation of Serum Albumin: It will be done by Bromocresol Green (BCG) method.
PRINCIPLE: In this method, serum albumin binds to Bromocresol green specifically, under acidic conditions. This reaction will produce a green coloured, albumin-BCG complex. The absorbance is read at 640 nm (red filter). The intensity of the colour of complex produced is directly proportional to the amount of albumin present in serum (Chawla, 2008).

3. Estimation of Serum Aminotransferases (SGPT and SGOT): It will be done by the method of Reitman and Frankel.
PRINCIPLE: In this method, to determine SGPT (ALT), the serum is treated with alanine and alpha ketoglutarate. These two compounds act as substrates, whereas to determine SGOT (AST), the serum is treated with aspartate and alpha ketoglutarate as substrates. Two keto acids namely, pyruvate and oxaloacetate are produced in these reactions respectively. These new keto acids are treated with 2,4-dinitrophenylhydrazine. The resultant compound, dinitrophenylhydrazone formed is of brown colour. The absorbance is read at 505nm (blue-green filter) (Chawla, 2008).

4. Estimation of Blood Urea: It will be estimated by urease enzymatic method described by Nessler.
PRINCIPLE: In this method, urea is converted to ammonia by the action of enzyme urease. The ammonia so produced reacts with Nessler’s reagent (potassium mercuric iodide). The brown coloured compound is read at 450 nm (Sood, 1999).

5. Estimation of Serum Creatinine: It will be estimated by Jaffe’s alkaline picrate method.
PRINCIPLE: Creatinine reacts with picric acid in alkaline medium and it will form a creatinine
picrate complex. This has red colour. It is read at 520 nm. The intensity of colour produced is dependent on the amount of creatinine in serum (Sood, 1999).


PRINCIPLE: In this method, for estimation of serum sodium and potassium, the emission flame photometry is used. The diluted serum is sprayed as fine droplets into the flame. The flame gets coloured due to emission of sodium or potassium ions. The amount of light emitted is dependent upon the strength of metallic ions in the serum (Chawla, 2008).


PRINCIPLE: Haemoglobin is treated with hydrochloric acid and it gets converted into acid haematin. The brown colour of the compound is matched visually by comparing with a standard in the sahli comparator (Ochei & Kolhatkar, 2000).

DEFINITIONS

1. Hepatitis: It is the inflammation of liver characterized by the presence of inflammatory cells in the liver.

2. Cirrhosis: Cirrhosis is the chronic diffuse disorder of varied etiologies and is characterized by hepatic cells necrosis, proliferation of connective tissues and nodular regeneration, thereby leading to abnormal reconstruction of lobular architecture and disturbed hepatic circulation.

3. Renal dysfunction: It is characterized by rapid decline in glomerular filtration rate over hours to days. It is usually asymptomatic and is diagnosed when biochemical markers in hospitalized patients reveal a rise in blood urea and serum creatinine levels. Serum creatinine concentration of > 1.5 mg/dl is considered as a cut off to decide renal dysfunction (Salerno et al., 2007).

4. Jaundice: It is the yellowish discoularation of skin, mucosa and conjunctiva. It is the hallmark symptom of liver disease and is clinically noticeable at serum bilirubin level >3 mg/dl (Ghany et al., 2008).
5. Ascites: it is the accumulation of fluid in the peritoneal cavity. It is clinically, observed as an increase in girth of abdomen. Diagnosis will be made by physical examination as patients have bulging flanks and shifting dullness (Mailliard et al., 2008).

6. Spontaneous bacterial peritonitis: It will be confirmed by the presence of Polymorphonuclear Neutrophil count > 250/mm³ (by laboratory analysis of ascitic fluid).

7. Acute Tubular Necrosis: This disorder will be confirmed by the presence of renal tubule epithelial cell casts as seen in examination of urinary sediments (Medline plus, 2011).

REFERENCE RANGE AND CUT OFF POINTS

1. eGFR: Glomerular filtration rate is estimated by MDRD equation utilizing serum creatinine, age, sex, race as variables. Reference range (90-120 ml/min/1.73m²). cut off point (75 ml/min/1.73m²)(Bellomo et al., 2004; Peter, 2011).

2. Serum Creatinine: reference range (0.6-1.2 and 0.5-1 mg/dl for male, female). cut off point(≥1.5 mg/dl) (Fernandez, 1995).

3. BUN: reference range (10-20 mg/dl) cut off point >20mg/dl (Ochei & Kolhatkar, 2000).

4. BUN/CREATININE: cut off point ≥20 mg.dl (Chertow, 2008).

5. Serum Bilirubin: reference range (0.2-1.2mg/dl) cut off point 1.2 mg/dl (Sood, 1999).

6. Serum Albumin: reference range (3.5-5 mg/dl). Cut off point 3.4 mg/dl (Sood, 1999).

7. Serum Sodium: reference range (135-145 meq/L) cut off point 134 meq/L (Sood, 1999).

8. INR: reference range (0.8-1.2 ) cut off point (1).

9. Serum Aminotransferases: reference range for SGPT (up to 45 IU), SGOT (up to 40 IU). Cut off point is >300 IU and SGPT >SGOT, in acute hepatitis whereas cut off point is < 300 IU and SGOT > SGPT in cirrhosis (Pratt, 2008).

10. Haemoglobin: reference range (15±2.5 and 14±2.5 mg/dl in male and female), cut off point 13 mg/dl (Ochei & Kolhatkar, 2000).
11. Platelets count: reference range (1.500000-3.500000/c mm) cut off point 1.500000 cmm (Ochei & Kolhatkar, 2000).

12. Serum Potassium: reference range (3.5-5 meq/L) cut off point 3.5 meq/L (Sood, 1999).

**STATISTICAL DESIGN**

The following statistical techniques will be used for analysis of data.

1. Classification and Tabulation of data: All data will be presented in the form of Tables, numbered in Arabic numerals.

2. Descriptive Statistics: Data will be expressed in terms of Mean and Standard Deviation as the means of central tendency and dispersion. Categorical data corresponding to demographic characters, clinical signs - symptoms and laboratory results of random urine sample and ascetic fluid will be presented in terms of (number of individuals) percentage.

3. p value of ≤ 0.05 will be implied as significant for all statistical tests.