VOLUME - II
ISSUE - XI
SEP / OCT 2005

The Crux

BIMONTHLY FORUM FOR THE LABORATARIANS

CONTENTS

Editorial

Hepatitis related investigations have come of age now. The knowledge now is exhaustive and precise. As some are self-limiting diseases and others leading to serious complications (that may be fatal), it is very important to identify the type of viral hepatitis. The relatively benign Hepatitis A can rarely end up as aplastic anaemia. Incidentally, we have hepatitis A, B, C, D, E, and even G as on date.

By the year 1970, investigations were available to diagnose both, Hepatitis A and Hepatitis B. However, at the same time cases that were neither Hepatitis A nor Hepatitis B were discovered. These were transfusion induced hepatitis cases. In the late 1980's a virally encoded antigen associated with non-A, non-B hepatitis (NANBH) was identified and labelled as Hepatitis C. It can over a period of time (usually decades) lead to cirrhosis and hepatocellular carcinoma. HCV is most often transmitted by percutaneous exposure to infected blood. Use of contaminated - blood or blood products, surgical implements, needles and sharps (in clinical practice or for IV drug use) are the most likely modes of transmission, though, it is also transmitted sexually too. Worldwide, over 170 million cases are known to exist. Mother to infant transmission is rare. Prevalence rate of HCV in India is said to be about 1.5 - 2%. HCV screening assumes great importance in relation to blood banking. Usage of contaminated blood is still the most important cause of HCV transmission. Availability of simple, quick, reliable and cost-effective diagnostic tools for HCV can reverse the trend. Fortunately, these are freely available now. Honesty exercised in the blood banks can block the spread of HCV. The DISEASE DIAGNOSIS section of this issue discusses at length, the percutaneous curse - namely Hepatitis C.

As tumour/cancer markers could not be discussed adequately in the previous two issues, the remaining overflow has been accommodated here under INTERPRETATION.

TROUBLE SHOOTING segment in this issue delves into the quality control aspects of a hematology laboratory. In fact, the quality assurance mechanisms mentioned can safely be transferred onto any branch of a pathology laboratory. Honesty towards one's work is by far the best quality control tool. The will to provide nothing but the best, automatically makes one move towards the quality control criteria. In the present era various agencies provide certifications pertaining to the perfect quality control measures being employed by anybody, including the clinical laboratories.

We are still laughing at ourselves in the BOUQUET. Wisdom is spelling success for you. Brainteasers as usual assess your histopathology prowess.

ISO 9001

Tulip Group

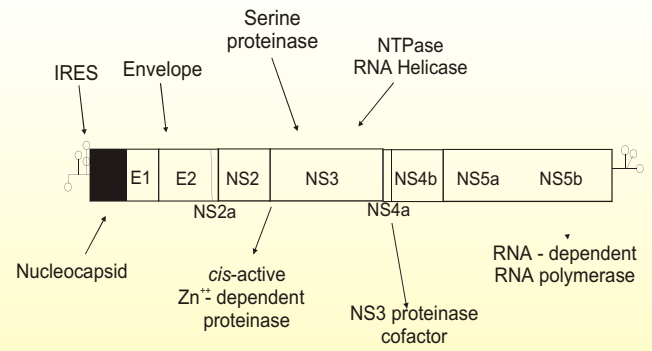
DISEASE DIAGNOSIS

VIRAL HEPATITIS C

INTRODUCTION

Primary viral infection of liver cells is caused by several hepatotropic viruses transmitted by oral and parenteral routes. While the clinical, epidemiological, pathological and immunological aspects of viral hepatitis have many common, yet subtle differences, the etiologies of liver disorders cannot be established on the basis of clinical signs and symptoms or on the basis of abnormal liver function tests alone. The virological and immunological properties of the five well established agents of viral hepatitis are summarized in the Table presented below.

Genomic Organisation of HCV



Characteristics	HAV	HBV	HCV	HDV	HEV	HGV
Family	*Picornaviridae*	*Hepadnaviridae*	*Flaviviridae / Togaviridae*	*Viroid*	*Caliciviridae*	*Flaviviridae*
Virion size (nm) & Shape Envelope	27, icosahedral	42, spherical	55, spherical	35,spherical	32, icosahedral	?
Genome	-	+	+	+	-	+
Type *	ss RNA	Partially ds DNA	ssRNA	ssRNA	ssRNA	ssRNA
Size (kb)	7.5	3.2	10	1.7	7.5	9.4
Replication	Positive - strand RNA	Positive - strand RNA intermediate	Positive - strand RNA	Helper HBV	Positive - strand RNA	Positive - strand RNA
Physical shape	Linear	Open circular	Linear	Closed circular	Linear	?
Polyadenylation	+	-	+/-?	-	+	?
Antigen(s)	HAV antigen	HBsAg, HBeAg, HBcAg	Env/NS1/E1,E2,core/NS3, NS4,NS5	HDV antigen	HEV antigen	NS2 / NS3 / Ns5 / E1,E2
Antibody(ies)	Anti- HAV -HAV IgM	Anti-HBs, -Hbe -Hbc IgM	Anti-core,E1,E2, -Ns2, NS3, NS4, Ns5	Anti- HDV	Anti- HEV	Anti-HGV
Gene amplification	RT-PCR	PCR	RT-PCR	RT-PCR	RT-PCR	RT-PCR

*ssRNa, single-stranded RNA : dsRNA, double-stranded RNA

BACKGROUND

Hepatitis C is an acute or chronic necro-inflammatory disease of the liver that is caused by a unique hepatotropic flavivirus. The disease was first recognized as early as the year 1970, when serological tests for Hepatitis A virus (HAV) and Hepatitis B virus (HBV) became generally available. It was noted at that time that most cases of transfusion associated hepatitis were not caused by either HAV or HBV, leading to the term non-A, non-B Hepatitis (NANBH). In the late 1980's, a virally encoded antigen associated with non-A, non-B hepatitis (NANBH) was identified and called the Hepatitis C virus (HCV).This finding rapidly led to the cloning of the viral genome, including the recognition of its proclivity to establish persistent infection, its strong association with chronic hepatitis, cirrhosis and hepatocellular carcinoma. Interestingly HCV was the first virus discovered by molecular cloning without the use of biological or biophysical methods. This was accomplished by extracting, copying into the cDNA and cloning all the nucleic acid from the plasma of a chimpanzee infected with NANBH by a contaminated factor XIII concentrate.

CLASSIFICATION OF THE HCV VIRUS

The HCV is a spheric, enveloped, positive strand RNA virus approximately 50 nm in diameter. Its structure, genomic organization and replication cycle, are similar to the members of the virus family *Flaviviridae,* yet sufficiently distinct to merit classification into a separate novel genus Hepacivirus.

HCV GENOTYPES, SUBTYPES AND QUASISPECIES

At the second International Conference of HCV and related viruses, a consensus nomenclature system was proposed for the future studies of the HCV genotypes and subtypes.

According to this system the HCV is classified on the basis of their nucleotide sequence into major genetic groups designated as "genotypes" and are assigned a number (arabic numerals) in the order of their discovery. The more closely related HCV strains within same genotypes are designated `subtypes" and are assigned lower case letters (in alphabetical order) in the order of their discovery. The complex of genetic variants found within an infected individual's isolate are termed as the "quasi species".It is hypothesized that the distinct viral

quasi species play a role in the pathogenesis and progressive HCV infection. Due to the sequence variability of the quasi species post infection, HCV is present in patients as a pool of viruses representing different epitopes. Modification of both B and T epitope patterns during HCV infection have been observed and clones contribute to HCV evasion from the immune system.

The phylogenetic grouping of HCV strain appears to be independent of the segment of genome that is analysed.

Substantial regional differences appear to exist in the distribution of HCV genotypes. Although genotypes 1, 2 and 3 appear to have a worldwide distribution, their relative prevalence varies from one geographical area to another.

HCV subtypes 1a and 1b are most common in USA and also predominant in Europe. Subtype 1b is responsible for upto 73% of the HCV infections in Japan. Subtype 2c is found commonly in north Italy whereas subtype 3a is particularly prevalent in IV drug users in Europe and North America. The genotype 4 is prevalent in North Africa and the Middle East and 5 and 6 seems to be confined to South Asia and Hong Kong respectively. Genotype 7, 8, 9 have been identified in Vietnamese patients and 10 and 11 have been identified in Indonesia. In India, genotype 1 and 3 are common to all parts of India, where as in the south of India, genotype 1 and 4 are responsible for infection.

CLINICAL RELEVANCE OF THE HCV GENOTYPES

The impact of HCV heterogeneity and genotypes on the day to day clinical management of chronic HCV infection has not been well established with regards to its role in progression of liver disease, the outcome of HCV infections and response to interferon therapy. However, the sensitivity and specificity of serologic and virologic assays for the detection of HCV may be influenced by the heterogenity of the HCV.

Potential role of HCV heterogeneity and genotypes has been suggested for mother to infant transmission and sexual transmissions. It is also suggested that genotypes 3a and 1a are closely associated with the IV drug usage and 1b in patients who acquire HCV through blood transfusion. Genotype 1b is reportedly associated with more severe disease and poor response to treatment.

Studies have provided some evidence that viral factors including the genotype may potentially play an important role in the development of chronic infection following exposure to HCV.

EPIDEMIOLOGY OF HCV

HCV is most often transmitted by percutaneous exposure to infected blood. The predominant modes of transmission may change over time and differ between and even within countries. HCV has been reported in virtually every country and the rough prevalence data estimates that more than 170 million people are infected worldwide. In developed nations the HCV prevalence is typically 1-2%, except for a few geographical regions such as Egypt, Japan, Taiwan and Italy where a high prevalence rate of infection, of between 10-30% have been reported. In India, the HCV prevalence rate is reported to be in the range of 1.5 -2%.

Folk remedies such as acupuncture, cutting with unsterilized knives and mass inoculation programs where frequently, unsterilized needles were often reused, have been suggested as likely modes of transmission. IV drug use, use of contaminated needles associated with illicit drugs, also remain one of the major causes of HCV infection worldwide.

BIOLOGICAL BASIS OF TRANSMISSION OF HCV

By using sensitive techniques, HCV RNA can be detected in blood (including serum and plasma), saliva, tears, synovial fluids, ascitic fluids, CSF and breast milk. However, information regarding potential infectiousness of body fluids is scarce. Without percutaneous exposure into blood stream, it is not clear as to how the virus reaches the liver, its primary site of replication. Ability of the virus to replicate in peripheral mononuclear cells has been also implicated. Mounting circumstantial evidence also suggests that HCV may be transmitted during sexual intercourse, though infrequently. HCV is uncommonly transmitted from mother to infant and the perinatal frequency of infection may vary between 0 -8%.

SPECTRUM OF CLINICAL HCV INFECTION

Chronic hepatitis C infection varies greatly in its course and outcome. At one end the spectrum are patients who have no signs or symptoms of liver disease and completely normal levels of serum liver enzymes (ALT). Liver biopsy usually shows some degree of chronic hepatitis, but the degree of injury is usually mild, and the overall prognosis may be good. At the other end of the spectrum are patients with severe hepatitis C infection who have symptoms, HCV RNA in serum, and elevated serum liver enzymes, and who ultimately develop cirrhosis and end-stage liver disease. In the middle of the spectrum are many patients who have few or no symptoms, mild to moderate elevations in liver enzymes, and an uncertain prognosis. Researchers estimate that at least 20% of patients with chronic hepatitis C develop cirrhosis, a process that takes 10 to 20 years post infection. After 20 to 40 years, a smaller percentage of patients with chronic disease may develop liver cancer.

It has been noted that coinfection with HBV accelerates the progression of the disease. HIV infection increases the level of HCV viremia and is associated with more rapid progression of the liver disease. Immunosuppression associated with agammaglobulinemia and organ transplantation also accelerates disease progression. In children asymptomatic infections as well as liver failures may occur. Elderly persons and males appear to be at a relatively higher risk for cirrhosis as well as liver cancer.

CLINICAL SYMPTOMS AND SIGNS

Many people with chronic hepatitis C have no symptoms of liver disease. If symptoms are present, they are usually mild, nonspecific, and intermittent. They may include :

- Fatigue
- Mild right upper-quadrant discomfort or tenderness
- Nausea
- Poor appetite
- Muscle and joint pains

Similarly, the physical examination is likely to be normal or show only mild enlargement of the liver or tenderness. Some patients have vascular spiders or palmar erythema.

CLINICAL FEATURES OF CIRRHOSIS

Once a patient develops cirrhosis or if the patient has severe disease, symptoms and signs are more prominent. In addition to fatigue, the patient may complain of muscle weakness, poor appetite, nausea, weight loss, itching, dark urine, fluid retention, and abdominal swelling.

Physical findings of cirrhosis may include:

- Enlarged liver,
- Enlarged Spleen
- Jaundice
- Muscle wasting
- Excoriations
- Ascites
- Ankle swelling

EXTRAHEPATIC MANIFESTATIONS

Complications that do not involve the liver develop in 1- 2 % of people with hepatitis C. The most common is cryoglobulinemia, which is marked by

- Skin rashes, such as purpura, vasculitis, or urticaria
- Joint and muscle aches
- Kidney disease
- Neuropathy

- Cryoglobulins, rheumatoid factor, and low complement levels in serum

Other complications of chronic hepatitis are:

- Glomerulonephritis
- Porphyria cutanea tarda

Diseases that are less well documented to be related to hepatitis C are:

- Seronegative arthritis
- Kerato-conjunctivitis sicca (Sjogren's syndrome)
- Non-Hodgkin's type B-cell lymphomas
- Fibromyalgia
- Lichen planus

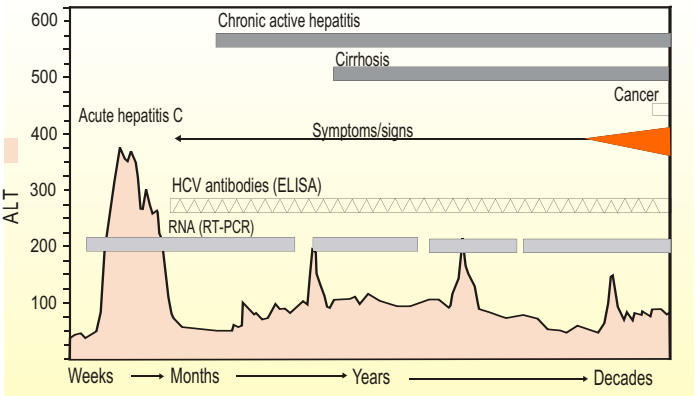
There is a poor correlation between necro-inflammatory liver injury, serum ALT levels, HCV RNA and extent of fibrosis. It is not possible to predict which HCV patient will develop cirrhosis and which will go to clinically decompensated liver disease.

The major complication of HCV infection is the development of hepatic fibrosis progressing to cirrhosis. Primary hepatocellular carcinoma is typically a late complication of chronic hepatitis C infection.

DIFFERENTIAL DIAGNOSIS

The major conditions that can be confused clinically with chronic hepatitis C include:

- Autoimmune hepatitis
- Chronic hepatitis B and D
- Alcoholic hepatitis
- Nonalcoholic steatohepatitis (fatty liver)
- Sclerosing cholangitis
- Wilson's disease
- Alpha- 1-antitrypsin-deficiency-related liver disease
- Drug-induced liver disease



BASIS FOR DIAGNOSIS OF HCV INFECTIONS

Acute HCV infection is typically mild and often not diagnosed till it becomes chronic. The two basic methods for HCV laboratory diagnosis are based on:

(a)Serological identification of anti-HCV antibodies from the plasma/serum of infected patients

(b) Detection of HCV RNA in the serum of infected patients

Most of the current diagnostic tests for detection of anti-HCV antibodies cannot differentiate between chronic and acute HCV infections because anti-HCV IgM occurs variably in acute infection and is also detected at high rates in patients with chronic HCV infection. However, double antigen sandwich based fourth generation immunoassays that detects IgG & IgM simultaneously, provides clinical decision makers an advanced tool to diagnose HCV infection at all stages of the disease. Anti-core and anti-NS3 may be the first antibodies to appear during acute phase (defined by increased ALT levels andA symptoms). Anti-NS5 appears somewhat later while anti-NS4 is the last antibody to be detected in an acute self limited infection.

HCV RNA in serum or liver appears to be the earliest detectable marker of acute HCV infection preceeding the appearance of anti-HCV by several weeks. HCV viremia may persist despite normalization of serum ALT levels. In acute self limited HCV infection the HCV RNA in serum usually last for fewer than four months.

IgM CLASS ANTI HCV ANTIBODIES

The primary IgM class anti-HCV response is usually against the core polypeptide and is detectable in patients with acute HCV infections and is still the first active antibody response. The IgM anti-HCV is rapidly followed by the IgG response. IgM anti-HCV is however not limited to the acute phase of the disease and is also found for protracted periods in long term HCV chronic patients. However, there is a strong association of the presence of anti-HCV IgM with HCV RNA positivity, patients progressing to ESRD, ESLD, in HCV disease reactivation patients and non responders to therapy.

Thus anti-HCV IgM activity is useful as a serological marker for ongoing, persistent and active HCV replication and HCV infection.

Serological assays that can detect anti-HCV IgM along with anti-HCV IgG class of antibodies may more consistently detect and indicate acute as well as chronic HCV seroresponse during the spectrum of the HCV infection. Since, during early phase of acute infection, patients are usually asymptomatic, from transfusion point of view, assays detecting an evolving IgM response may add value to the detection of infected blood.

SUPPLEMENTARY ASSAYS

Supplementary assays such as recombinant immunoblot assays and HCV RNA based assays are used to reconfirm the positive results of EIAs and other first line rapid immunoassays.

RECOMBINANT IMMUNOBLOT ASSAYS

Immunoblot assays are used to confirm anti-HCV reactivity with screening tests. These tests are also called "Western blots". Serum is incubated on nitrocellulose strips on which recombinant viral proteins are blotted /sprayed. Color changes brought about by reaction of enzyme conjugated to Anti-human IgG with the substrate indicate that antibodies are adhering to the blotted proteins on the solid phase. An immunoblot is considered positive if two or more proteins react and is considered indeterminate if only one positive band is detected. In some clinical situations, confirmatory testing by immunoblotting is helpful, such as for the patients with anti-HCV detected by EIAs who test negative for HCV RNA. The EIA anti-HCV reactivity in such cases could represent a false-positive reaction, or recovery from hepatitis C, or continued virus infection with levels of virus too low to be detected (the last occurs only rarely when sensitive PCR assays are used). If the Immunoblot test for anti-HCV is positive, the patient has most likely recovered from hepatitis C and has a persistent antibody to the virus. If the immunoblot test is negative, the EIA result was probably a false positive. Immunoblot tests are routine in blood banks when anti-HCV positive sample is found positive by EIA.

Immunoblot assays are highly specific and valuable in verifying anti-HCV reactivity. Indeterminate tests require further follow-up testing, including attempts to confirm the specificity by repeat testing for HCV RNA.

HCV RNA BASED ASSAYS

HCV RNA can be detected in plasma and serum by RT PCR's and branched DNA (bDNA) based assays. These assays vary in their ability to detect HCV RNA but RT PCR's are generally considered to be more

sensitive than bDNAbased assays. Detection of HCV RNA indicates ongoing infection whereas clearance from serum of HCV RNA spontaneously or after treatment correlates with ALT normalisation and improvement in liver histological findings. Testing for HCV RNA is particularly useful for diagnosis of HCV when ALT is normal or when the values are slightly elevated, or when anti HCV is negative or when several other causes of liver diseases are not implied. HCV RNA estimation is especially helpful in diagnosis of infections in immunosuppressed, immunocompromised, organ transplant or patients having chronic renal failure.

Since the maternal antibodies are passively transferred to infants, the diagnosis of HCV infection in infants must be based on viral DNA based tests. The anti HCV antibodies persist in the infants even after 18 months of age. Since viremia can be intermittent in the first year of life and some HCV RNA positive infants never develop anti-HCV antibodies, infection in infants can be excluded only based on repeat HCV RNA test findings.

It must however be understood that HCV RNA assays have an intrinsic variation. Additionally there is a lack of a quantitative gold standard for HCV RNA based assays and the reporting units differ. Serial measurement of viremia must be performed using the same tests and preferably using the same laboratory setup.

SEROLOGICAL DIAGNOSIS OF HCV: OPEN ISSUES

In spite of the various technological developments in HCV serodiagnosis, the following issues will require continued resolution:

- Improvement of analytical sensitivity and specificity of antibody and antigen assays.
- Detection of low antibody titers in immunosuppressed patients
- Earlier detection of seroconversion.
- Serological assays to unequivocally distinguish between acute/chronic and resolved infections.
- Serological marker for immunity.

SIMPLE FLOW THROUGH AND LATERAL FLOW ICTs ALONG WITH RAPID VISUAL ELISA FORMATS ARE GOOD ENOUGH TOOLS FOR ROUTINE CLINICAL PRACTICE.

PREVENTION OF HCV INFECTION

The key to reducing the incidence of HCV is by decreasing exposure to contaminated blood and reducing post transfusion HCV infection rates. Nosocomial HCV transmission can be controlled through adherence to universal precautions and infection control protocols diligently.

BOUQUET

In Lighter Vein

"Doctor, Doctor, You've got to help me - I just can't stop my hands shaking!"
"Do you drink a lot?"
"Not really - I spill most of it!"

A man speaks frantically into the phone, "My wife is pregnant, and her contractions are only two minutes apart!"
"Is this her first child?" the doctor queries.
"No, you idiot!" the man shouts. "This is her husband!"

A man walks into a doctor's office. He has a cucumber up his nose, a carrot in his left ear and a banana in his right ear.
"What's the matter with me?" he asks the doctor.
The doctor replies, "You're not eating properly."

A young woman went to her doctor complaining of pain.
"Where are you hurting?" asked the doctor.
"You have to help me, I hurt all over", said the woman.
"What do you mean, all over?" asked the doctor, "be a little more specific."
The woman touched her right knee with her index finger and yelled, "Ow, that hurts." Then she touched her left cheek and again yelled, "Ouch! That hurts, too." Then she touched her right earlobe, "Ow, even THAT hurts", she cried.
The doctor checked her thoughtfully for a moment and told her his diagnosis, "You have a broken finger."

Wisdom Whispers

- If you find it in your heart to care for somebody else, you will have succeeded.
- There are trivial truths, and there are great truths. The opposite of a trivial truth is plainly false. The opposite of a great truth is also true.
- Destiny is not a matter of chance; but a matter of choice. It is not a thing to be waited for, it is a thing to be achieved.
- To expect defeat is nine-tenths of defeat itself.
- Defeat never comes to any man until he admits it.
- Anyone who has never made a mistake has never tried anything new.
- The greatest test of courage on earth is to bear defeat without losing heart.
- Mistakes are the portals of discovery.
- Try , and try again; success usually follows.

Brain Teasers

1.What type of necrosis is seen in tuberculosis?
A.Gummatous B. Coagulative C. Caseation D. Liquefactive

2.Parasitic inflammation would show a predominance of
A.Eosinophils B. Neutrophils C. Lymphocytes D. Monocytes

3. Auer rods may be observed in
A.Plasma cell B. Myeloblast C. Lymphoblast D. Stem cell

4. Melanocytes may occasionally be found in
A. Ovary B. Adrenal medulla C. Urinary bladder
D. Substantia nigra of brain

Answers 1. C, 2. A, 3. B, 4. ABCD

INTERPRETATION

CANCER MARKERS (TUMOR MARKERS)...Contd

hCG (human chorionic gonadotropin)

Reference interval

Serum/plasma,

hCG and hCG + hCG:

Men and premenopausal women	< 5 IU/L
Postmenopausal women	< 10 IU/L
hCG	< 0.2 IU/L

Indication

The tumor markers hCG and hCG are used for diagnosis, follow-up, and monitoring of therapy.

Absolute indications

- Germ cell tumors
  - Hydatidiform mole and choriocarcinoma in women
  - Testicular cancer in men
- Extragonadal germ cell tumors

Relative indications

Patients with increased risk for germ cell tumors:

- Cryptorchidism
- Healthy, monozygotic twin of a patient with testicular cancer
- Patients in complete remission after therapy for testicular cancer; due to increased risk of development of a contralateral second tumor.
- Nontrophoblastic tumors secreting hCG; in this setting more sensitive and more specific tumor markers are usually available.

Other indications

- Early diagnosis of eutopic and ectopic pregnancy
- Diagnosis of spontaneous abortion
- Diagnosis of chromosomal anomalies (trisomy 21)

MCA (mucin-like cancer-associated antigen)

Reference interval

Serum, plasma 15 U/mL

MCA is useful for monitoring the disease course in patients with metastatic breast cancer. The test is not suited as a screening test or adjunct to the diagnosis because the clinical sensitivity is too low for localized disease and elevated levels are often associated with benign breast diseases and with other organ cancers. Furthermore, it should be considered that all mucin markers including MCA, should be combined with another type of marker, e.g. with CEA in breast cancer, but not with each other.

Indications

Monitoring the outcome of treatment and disease course in patients with breast cancer.

NSE (neuron-specific enolase) -enolase

NSE is useful for monitoring the outcome of treatment and disease course in patients with neurcendocrine tumors, in particular small cell lung cancer and neuroblastoma. The test is not suited as a screening test or adjunct to diagnosis because of low clinical sensitivity and specificity.

Reference interval

Serum, CSF(values in g/L)

Adults:	Serum	10 or 20
	Cerebrospinal fluid	0-3.7 or 20
Children	During the first year of life	25
	During the 1st-5th year of life	20
	During the 6th-8th year of life	18

Indication

- Monitoring the outcome *of* treatment and disease course in patients with neuroendocrine tumors and APUDomas
- Absolute indications: small cell lung cancer (SCLC), neuroblastoma
- Relative indication: medullary thyroid carcinoma

PSA (prostate-specific antigen)

PSA is a very useful parameter for prostate cancer screening. It is used in combination with digital rectal examination and possibly transrectal ultrasound examination in asymptomatic men > 50 years of age. Furthermore, PSA is suitable for staging purposes, monitoring of treatment and the disease course in patients with primary or metastatic prostate cancer.

Reference interval

Serum, plasma 4 µg/L

For grey-zone values , free PSA must also be evaluated.

Indication

- Screening of asymptomatic men > 50 years of age for the presence of prostate cancer, together with digital rectal examination and transrectal ultrasound examination/biopsy depending on the individual findings
- Supplement to the staging process prior to the treatment of prostate cancer
- Monitoring treatment and clinical course in patients with prostate cancer

SCC (squamous cell carcinoma antigen)

SCC is not a specific tumor marker for squamous cell cancer and is not suited for screening purposes due to a lack in clinical sensitivity and specificity. However, it can be recommended for monitoring of disease course and response to therapy in primary and current squamous cell cancer.

Reference interval

Serum, plasma 3.0 µg/L

Indication

Monitoring of disease course and response to therapy in patients with squamous cell cancer of the cervix, lung, esophagus, anus, and head-neck region.

TROUBLE SHOOTING

QUALITY CONTROL IN HEMATOLOGY

Quality control in medical laboratories encompasses a set of procedures which ensure that reliable and timely test results are received by the users *of* laboratory service. Reliability implies both precision and accuracy.
There are four components of quality assurance program:
A. Internal quality control (IQC)
B. External quality control (EQA)
C. Standardization.
D. Proficiency surveillance.

A. Internal Quality Control

Since now most of the tabs are dependent on automated machine, it has become extremely important to maintain good internal quality control, which is done by:

1. Testing Control Sample

The best known method is by testing a control sample along side the routine specimen in each batch of test. Control material is either obtained commercially or prepared individually, but its stability and homogeneity should be ensured.

2. Control Chart (Levy-Jennings or L-J chart)

In this process when a batch *of* samples is dispensed (after being run along a control sample), the mean and standard deviation of each parameter is obtained and linear graphs are ruled, showing the +2 standard deviation (SD) limits. Statistically, not more than 1 in 20 samples should fall outside these limits if the system is in control.

3. Cusum Analysis

Cumulative Sum (Cusum) Charting was introduced in 1960s. Deviation from the largest is plotted in a cumulative manner so that each point represents the sum of all the deviations to date from the mean or target value. This method of plotting exaggerates trends in the data. And makes shifts of the mean much more obvious than by other plots. The rules for using the cusum system for quality control are less well defined than for the L-J system.

4. Duplicate Tests

A well known method for checking precision in clinical analysis is duplicate testing. In this process a few of the specimens that were measured in an earlier batch, are rechecked with the next batch control

5. Inbuilt Quality Control

This includes:
- Use of cumulative reports of a single patient
- Clinical correlation: if a physician can not interpret a report on clinica grounds, a repeat test with a fresh specimen is indicated.
- Red cell indices: if reports are giving erroneous rise or fall in the red cells indices, this usually points to an error in analysis.
- Blood film examination ultimately helps in double checking the analysis done by the instrument.

B. External Quality Assessment

The college of American Pathologist first introduced "proficiency testing" survey program in 1960. In the late 60 The British Committee for Standards in Hematology, finally developed the National External Quality Assessment Scheme (NEQAS) for Hematology. Such methods are used by various laboratories all over the world to keep up with international standards.

C. Standardization

Modem diagnostic systems depend on a calibration procedure for accurate performance. Calibrators or testing standards are commercially prepared products, made by a direct comparison with a primary international standard. They are used for accuracy and inter-laboratory harmonization of test results. The calibrator has an assigned value as close to the true value as can be established.
The WHO (World Health Organization) provides a wide range of biologically important international reference standard material. Some examples of these which are available for use in hematology are:
a) Hemoglobin preparation.
b) Hemoglobin A2 and F.
c) Thromboplastin.
d) Blood typing sera.
e) Various coagulation factors.

D. Proficiency Surveillance

This is concerned with the pre-analytical and post-analytical parts of the process that require control, if tests are to be reliable and effective. This involves following a standard guideline at various steps of a laboratory analysis.
The steps are:
1) Standard of blood collection tube
2) Phlebotomy technique
3) Identification of sample with special reference to hazardous specimens
4) Maintenance of transportation standards
5) Data processing of results
6) Establishing normal reference values, assessment of the significance of results and taking decisions for further tests.

TECHNICAL PROFICIENCY HAS ALWAYS BEEN THE CORNER STONE OF THE LABORATORY, BUT IN RECENT YEARS WITH THE ADVENT OF SOPHISTICATED INSTRUMENTS AND AUTOMATION, QUALITY CONTROL HAS ASSUMED AN EVEN MORE IMPORTANT ROLE IN GOOD LABORATORY PRACTICE.
IT IS THE DUTY OF THE LABORATORY STAFF TO ENSURE THAT THE TESTS WHICH ARE CARRIED OUT ARE APPROPRIATE AND TO PROVIDE RELIABLE ANALYTICAL RESULTS.

TULIP NEWS

TULIP GROUP EXPANDS ITS INSTRUMENTATION DIVISION

In the same section of the Mar/Apr 2004 issue (Vol-I, Issue-II)we had discussed about the INSTRUMENTATION DIVISION of TULIP GROUP OF COMPANIES along with the range of instruments offered. To complement its ELISA reagents range viz. QUALISA-HIV1& 2, QUALISA-HBsAg and QUALISA-HCV, TULIP GROUP goes one step further to introduce state-of-art brands in ELISA automation. Two novel instruments - Lisaquant (ELISA plate reader-3000) & Lisawash (ELISA plate washer-3000) have been introduced.

Lisaquant
ELISA PLATE READER-3000

Lisaquant is a user friendly, versatile micro-plate reader, designed to measure and interpret enzyme immunoassay results, both monochromatically and bichromatically. Lisaquant works on different menu-driven modes such as absorbance mode, single calibrator mode, cut-off mode, multistandard mode and % absorbance mode. The pre-programmed calculation modes in this instrument help to facilitate data processing of enzyme immunoassays. Some additional features of this instrument are that it operates on a wide voltage range(90-270V) thereby eliminating the need of external voltage stabilizer. Lisaquant has the advantage of accomodating flat as well as round bottom configuration and can be connected to any external IBM PC compatible printer.

Lisawash
ELISA PLATE WASHER-3000

Lisawash is a versatile, user friendly & rugged instrument designed for use as a ELISA plate washer, with programmable soak time, wash cycles and dispensing volume. The special features of the instrument are wash bottle full indicator with audible alarm, inbuilt stabilizer and battery backed up memory for 35 tests.

NEW INTRODUCTIONS

Printed and published by D.G. Tripathi, Edited by Dr. R.J. Sood M.D. (Path.) for and on behalf of Tulip Diagnostics Private Ltd, Gitanjali, Tulip Block, Dr. Antonio Do Rego Bagh Alto Santacruz, Bambolim Complex Post Office, Goa - 403202, INDIA. Fax: (0832) 2458544, E-mail: tulip@sancharnet.in. Website: www.tulipgroup.com