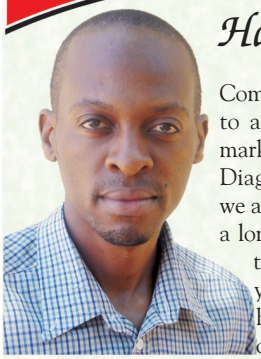
DIAGNOSTICS UPDATE .COM

NEWSLETTER
Version 8
February 2006

From My Keyboard



Happy New Year

Compliments of the new season to all our dear readers! 2006 marks the 2nd birthday for the Diagnostics-update newsletter we are proud to have come such a long way and are still around to share our knowledge with you. We thank all those who have been with us through out our journey and we hope you remain by our side as we venture along in this uncharted territory.

As we ring in the New Year, most of us weigh up ways to make this year healthier and happier than the last. I would like to encourage our readers not to set themselves up to fail this year. Its no secret that New Year's resolutions are always more often broken than followed. Maybe this is because we make them half-heartedly. Normally our intentions are always good but our willpower to live with those resolutions is always weak. However, if you have made some resolutions for this New Year, the approach you take in making those resolutions can mean the difference between success and failing to keep the resolutions. Below are a few pointers on how to be successful in following your resolutions as adapted from the Havard Health Publications:

1. Limit the number of resolutions

Even when we have the best intentions as we take our New Year's resolutions, one common mistake is to have too many goals. Attention and willpower get split so many ways that little

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Inside this Issue

Tiger Design and Graphics

DML Clinical Chemistry Department staff members pose with the new Hitachi Modular System Analyzer

DIAGNOFIRM MEDICAL LABORATORIES

Recent Events @ Diagnofirm

Compliments of the season good people! It is my sincere hope that you had a smooth roll-over into 2006. May this be your best year ever. From us here at DML, we say "Healthy 2006!!"

Since the last time you heard from us, quite a lot has been cooking here and we expect more this year.

Two more branches:

Towards the end of 2005, we opened two more branches for the convenience of our clients. People in the Tlokgweng and Village area do not need to come to the main laboratory for our services as they can now access them at Riverwalk Mall branch. People around the Fairgrounds Mall area can also now get our services at Medswana House.

Green and Red all around you

In line with our vision of striving to become the best analytical laboratory in Botswana and beyond, the beginning of 2006 has seen us painting Kanye village red and green. This means all surrounding villages like Jwaneng and Lobatse among others can get "pathology they can trust" closer to home.

Analyzer This!

In our quest to provide the best analytical laboratory services in Botswana, we at DML have recently acquired a state-of-the-art analyzer. The Hitachi Modular System, supplied by Roche Diagnostics is a massive machine measuring 1.5m by 5m by 1.2m and is only the 4th of its kind in Southern Africa. The other three are in South Africa.

This machine is composed of three modules. The E-170 does 170 endocrinology tests per hour; the P800 has capacity to do 800 Chemistry tests in an hour while the ISE-900 can run 900 Electrolyte tests per hour.

This modular system comes with new and improved methodologies and will certainly reduce average turn-around times

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THERAPEUTIC DRUG MONITORING

Basic assumptions underlying therapeutic drug monitoring (TDM) are that drug metabolism varies from patient to patient and that the plasma level of a drug is more closely related to the drug's therapeutic effect or toxicity than is the dosage.

TDM is the measuring of some drugs as a way to determine the most effective dose or to avoid toxicity. A lot of drugs are not monitored in similar way since they can be monitored by taking body temperature or blood pressure.

TDM is of importance because for some of these drugs, most of them work over a small range, below which they not effective and above which they become toxic to the patient.

Another factor is that people take more than one medication, which may interfere with the way that particular drug is absorbed or metabolized by the body.

At times patients default and do not take drugs as prescribed and hence TDM can also help identify if the patient is defaulting.

There are several drugs that are monitored and they fall into different categories.

Drugs with a **narrow therapeutic index**, where therapeutic drug levels do not differ greatly from levels associated with serious toxicity should be monitored. *(Example:* Lithium.)

Patients who have **impaired clearance** of a drug with a narrow therapeutic index are candidates for drug monitoring. The clearance mechanism of the drug involved must be known. *(Example:* Patients with renal failure have decreased clearance of gentamicin and therefore are at a higher risk for gentamicin toxicity.)

Drugs whose **toxicity is difficult to distinguish from a patient's underlying disease** may require monitoring. *(Example:* Theophylline in patients with chronic obstructive pulmonary disease.)

Drugs whose **efficacy is difficult to establish clinically** may require monitoring of plasma levels. *(Example:* Phenytoin.)

The associated table shows some of the drugs monitored and their use in medicine.

The proper timing of blood sampling for the purpose of TDM is quite essential. For most of the drugs listed in the table, blood is drawn right before the a dose is given. For others, the blood is collected after the dose is given at a specific time; for example 60 or 90 minutes later.

The level of the drug measured in the blood is used to determine if the patient is getting the correct amount of medication and to calculate the amount of drug that should be given the next time.

Some of the important factors to consider in TDM are:

1. Half Life

The half-life is the time it takes for the serum concentration of a drug to decrease by 50%. Knowing the half-life of the drug is important in that it helps determine the most advantageous dosing schedule for oral drugs, the intra dose fluctuations of the serum concentrations and the time the drug takes to reach a steady state.

2. Steady State

These are concentrations of the drug in serum that recur with each dose and that represent a state of equilibrium between the dosage of the drug administered and the amount of drug being excreted in a given time interval.

3. Peak Serum Concentration

This is the point of maximum concentration on the serum concentration versus time curve.

4. Trough serum concentration

This is the minimum serum concentration found during a dosing interval. The point is theoretically represented by the period immediately preceding administration of the next dose.

5. Absorption

This is the process by which a drug enters the body system. Drugs, which are injected intravascularly, are absorbed totally, but extra vascular administration will yield varying degrees and rates of absorption. The relation ship between rate of absorption and rate of excretion is what determines the drug concentration in the bloodstream.

6. Distribution

This is the dispersion of the drug from the intravascular space into extra vascular fluids and tissues and thus to the target receptor sites.

7. Therapeutic Range

The range of serum drug concentrations associated with a high efficacy, a low risk of toxicity. Some patients will show a response at serum levels below the lower limit of the range while others will require serum levels exceeding the upper limit for therapeutic benefit.

Correct timing of the sample collection is essential since drug therapy is often revised on the basis of serum concentration determinations.

It is important that the absorption and distribution phases of the drug be complete and a steady state concentration be attained before sample collection, otherwise results may be erroneously low. Hence, increasing a dose on such a result will produce toxic concentrations.

It is also very important to be consistent with the sampling time when making comparative measurements.

CATEGORY	DRUGS	TREATMENT USE
Cardiac Drugs	Digoxin, quinidine, procainamide, N-acetyl-procainamide	Congestive heart failure, angina, arrhythmias
Antibiotics	Aminoglycosides (gentamicin, tobramycin, amikacin), vancomycin, chloramphenicol	Infections with bacteria that are resistant to less toxic antibiotics
Antiepileptics	Phenobarbital, phenytoin, valphenytoin, valproic acid, carbamazepine, ethosuximide lamotrigine	Epilepsy, prevention of seizures
Bronchodilators	Theophylline, caffeine	Asthma, chronic obstructive pulmonary disorder (COPD), neonatal apnea
Immuno-suppressants	Cyclosporine, tacrolimus, sometimes sirolimus, mycophenolate mofetile	Prevent rejection of transplanted organs
Anti-cancer drugs	Methotresate	Various cancers, rheumatoid arthritis, non-hodgkin's lymphomas, osteosarcoma, psoriasis
Psychiatric drugs	Lithium, desipramine, some antidepressants (imipramine, amitriptyline, nortriptyline, doxepin)	Bipolar disorder (manic depression), depression

by Bozo and Ndlovu, Microbiology Department

CRYPTOSPORIDIUM INFECTION IN BOTSWANA

Symptomatic diarrhea infections are a major public health concern in all developing countries including Botswana. This is more pronounced in this era where many people, regardless of age, can be infected with HIV.

Cryptosporidium infections are often in immunologically deficient or immunologically compromised individuals. Human infection is zoonotic. In immuno-competent hosts, parasites are usually found in the stools of young children with watery diarrhea of mild or moderate severity. Some excretory of the organisms are asymptomatic.

In persons with severely weakened immune systems, especially those with AIDS, cryptosporidiosis can be serious, long lasting and sometimes deadly.

If your CD4 cell count is below 200, you are more likely to have diarrhea and other symptoms for a long time. If your CD4 count is above 200 and you get crypto infection, you may feel better in about 1-3 weeks, but you might still have the infection and be able to pass it to others even after you feel better. If you are still infected and your CD4 count later drops below 200, the crypto may act up again.

How is Cryptosporidiosis spread?

The most common ways one can get cryptosporidiosis are:

A. Touching mouth before washing hands after:
   1. Touching stool of infected people.
   2. Touching stool of infected animals.
   3. Touching soil or objects contaminated with stool.

B. Drinking water contaminated with stool or eating food contaminated with stool.

Prevention of cryptosporidiosis

Stool is a major source of the protozoan parasite. This information is helpful in deciding ways of prevention:

a. Avoid sex that involves contact with stool and don't kiss or lick the genitals or anus. Wash hands after touching your partner's anus or rectal area.
b. Avoid touching farm animals and if you do, wash your hands with soap before eating or preparing food.
c. Avoid touching the stools of pets. Use of gloves is encouraged.
d. Vegetables and fruits might be contaminated. Wash or cook the food.
e. Drink safe water i.e. boiled or filtered water.
f. When swimming in lakes, rivers, or public pools, avoid swallowing water.

Treatment of cryptosporidiosis

There is no drug that can cure it, although we do have drugs that reduce the symptoms. Oral rehydration therapy is necessary to avoid dehydration. Antibiotics, such as spiramycin and dicalzuril sodium, produce partial responses. Paromomycin has been shown to decrease the intensity of infection and improve intestinal function. It is a broad spectrum antibiotic similar to neomycin.

Laboratory Diagnosis

The diagnosis of Cryptosporidiosis in the laboratory is achieved by one of the following:

a. Demonstration of Cryptosporidium parvum oocysts in stool.
b. Demonstration of Cryptosporidium in intestinal fluid or small bowel biopsy specimens
c. Demonstration of Cryptosporidium antigen in stool
d. Molecular methods

Cryptosporidium parvum oocysts

Mature *C. Parvum* oocysts recovered from stool are round and contain four sporozoites within the thick walled oocysts. These can be identified easily using the staining methods such as modified Kinyoun's acid-fast method; hot Safranin stain and fluorescent dyes such as auramine/carbol fuchsine fluorescent method.

To maximize the recovery of oocysts, stool samples should be concentrated (formol ethyl acetate-FEA) prior to microscopic examination.

Studies have shown that one sample is enough to provide accurate diagnosis in 90% of the cases.

Cryptosporidium antigen in stool

ELISA: The specimens should not be concentrated prior to testing. This is a highly sensitive and specific technique, and is useful for screening large numbers of specimens in a short time period.

Also it does not rely on skills in microscopy. However, controls are necessary to determine the quality of commercially available reagents.

Immunoflurescence assay: This technique offers the highest combination of sensitivity and specificity and is considered the gold standard by many laboratories.

Molecular methods

PCR is used to detect *C. Parvum* in stool specimens. Faecal material must be stored in potassium dichromate or be frozen for detecting the DNA of the organism by PCR.

Most laboratories use the acid-fast staining methods. For greatest sensitivity and specificity, immunofluorescence microscopy is the method of choice (followed closely by ELISA).

Molecular methods are to be used mainly as a research tool.

Conclusion

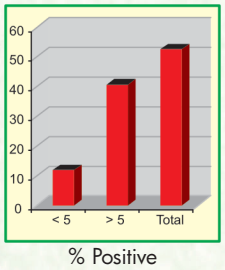
Chronic diarrhoea is one of the most common disorders and thus represents an important public health issue. In many cases physicians underestimate the prevalence of cryptosporidium, leading to more burdens to the already immunocompromised patients.

There is a strong suggestion to screen all patients with diarrhoea for cryptosporidium. ♥

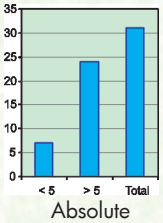
Over a period of one year (1/1/2005 to 31/12/2005), Diagnofirm Medical Laboratories tested 59 stool specimens for the presence of Cryptosporidium and the following results were obtained.

	POSITIVE		NEGATIVE	
AGE	Numbers %	Absolute Count	Numbers %	Absolute Count
< 5 yrs	11.9	7	25.4	15
> 5 yrs	40.7	24	22.0	13
TOTAL	52.6	31	47.4	28

Positives were 11.9% for patients below the age of 5 years and 40.7% for patients above the age of 5 years. With a total of 52.6%, these results are quite significant.



% Positive



Absolute Count

by Muhyaradzi Mangwendeza

Down Syndrome

The Down Syndrome is named after John Langdon Down, the first physician to identify this syndrome. Down syndrome is the most frequent genetic cause of mild to moderate mental retardation and associated medical problems and occurs in one out of 800 live births, in all races and economic groups.

Down syndrome is a chromosomal disorder caused by an error in cell division that results in the presence of an additional third chromosome 21 or "trisomy 21."

The Chromosomal Basis

To understand why Down syndrome occurs, the structure and function of the human chromosome must be understood. The human body is made of cells; all cells contain chromosomes, structures that transmit genetic information.

Most cells of the human body contain 23 pairs of chromosomes, half of which are inherited from each parent. Only the human reproductive cells, the sperm cells in males and the ovum in females, have 23 individual chromosomes, not pairs. Scientists identify these chromosome pairs as the XX pair, present in females, and the XY pair, present in males, and number them 1 through 22.

When the reproductive cells, the sperm and ovum, combine at fertilization, the fertilized egg that results contains 23 chromosome pairs.

A fertilized egg that will develop into a female contains chromosome pairs 1 through 22, and the XX pair. A fertilized egg that will develop into a male contains chromosome pairs 1 through 22, and the XY pair.

When the fertilized egg contains extra material from chromosome number 21, this results in Down syndrome.

Occurrence of Down Syndrome

Most of the time, the occurrence of Down syndrome is due to a random event that occurred during formation of the reproductive cells, the ovum or sperm. As far as we know, Down syndrome is not attributable to any behavioral activity of the parents or environmental factors.

The probability that another child with Down syndrome will be born in a subsequent pregnancy is about 1 percent, regardless of maternal age.

In 88% of cases, the extra copy of chromosome 21 is derived from the mother. In 8% of the cases, the father provided the extra copy of chromosome 21. In the remaining 2% of the cases, Down syndrome is due to mitotic errors, an error in cell division which occurs after fertilization when the sperm and ovum are joined.

The likelihood that a reproductive cell will contain an extra copy of chromosome 21 increases dramatically as a woman ages. Therefore, an older mother is more likely than a younger mother to have a baby with Down syndrome.

Many specialists recommend that women who become pregnant at age 35 or older undergo prenatal testing for Down syndrome

Prenatal Screening

Screening for Down syndrome has been a routine part of prenatal care for women over 35 years of age for the past 10-15 years. The current screening process involves the measurement of three maternal serum biochemical markers produced during the early second trimester between 15-20 weeks of a normal 40 week pregnancy.

These markers are produced by the foetus and the placenta. The levels of these markers, together with the age of the mother being screened, can help indicate the risk of the affected pregnancy.

Screening tests are not perfect. They can only assign risk of development, but cannot identify with certainity who is actually affected and who is not. However, good screening tests can identify the highest number of affected pregnancies while at the same time selecting the fewest number of unaffected pregnancies for follow up.

Second trimester screening is the widely used Down syndrome screening protocol. It is also known as a triple marker test, or just triple test. The maternal serum biochemical markers used are :

1. Alpha-pheto protein produced by the foetus liver,
2. Human chorionic gonadotrophin produced by the placenta, and
3. Uncojugated estriol (uE3) synthesized from steroid precursors that originate from the foetal adrenal gland and liver.

Alpha-feto protein is also used for screening of open neural tube defect. The performance of this method of screening is at 70% with a false positive screening of 5%.

Another marker known as inhibin A has also been found to be useful in the 2nd trimester screening. It is a product of the placenta and is also synthesized by the ovaries.

The levels of inhibin A in the 2nd trimester are, on average, 2 times higher in Downs syndrome than in unaffected pregnancies. Addition of inhibin A to the triple test increases the detection rate by 7-10%, giving the quad test a detection rate of 80% with 5% false positive.

Screening for Down's syndrome earlier in pregnancy is very desireable as it gives

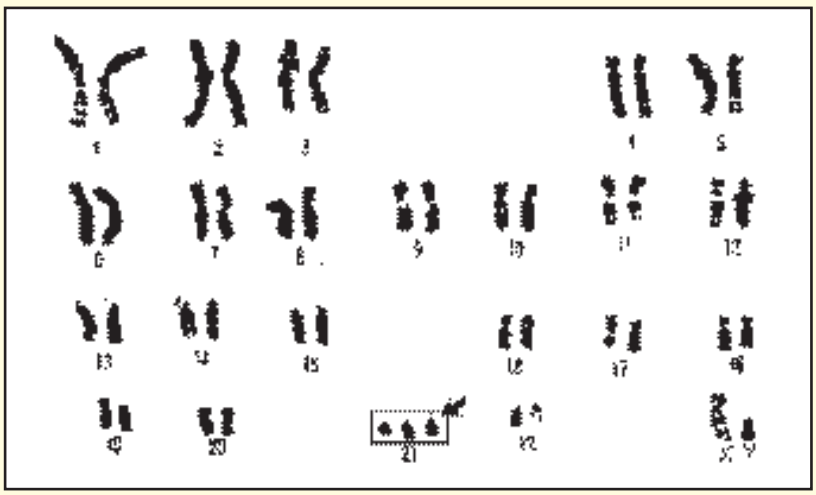


Image of the karyotype of a child with Down syndrome or Trisomy 21. The image shows the pairing up of the chromosomes, with chromosome 21 showing 3 chromosomes instead of 2.

the patient privacy and if chosen, safer termination.

First trimester screening tests combine maternal serum markers and a marker measured in an ultra sound exam, between 10-11 and 13 completed weeks of pregnancy.

The serum biochemical markers are: (a) pregnancy associated plasma protein A (PAPP-A) and (b) the free subunit of human chorionic gonadotropin (hcg). The levels of PAAP-A are lower in Down syndrome pregnancies and the levels of the free subunit of hcg are elevated.

First trimester screen also uses Nuchal translucency thickness, or NT, as measured by an ultrasound examination. The NT is increased in Down syndrome pregnancies.

Integrated Down syndrome screening tests are done with the 2 best 1st trimester markers NT and PAPP-A and the 2nd trimester quad markers. There is also a serum only version of the integrated test which is done without NT and uses only the PAPP-A plus the quad markers. This approach is based on the fact that if the markers are independent indicators of risk, then using all the 1st trimester and 2nd trimester together will produce a better screening test than using either the 1st trimester or second trimester tests alone.

The procedure for the tests is that a woman has the NT measurements and a blood drawn for PAPP-A measurements between 10-13 weeks and then a second blood draw at 15-16 weeks for the quad tests. The results for the NT and PAPP-A are reported together with those of the quad test to make one risk assessment.

The performance of the integrated tests is markedly more superior than the individual screening tests alone. Given the trend towards delaying pregnancy until a woman is in her 30s or 40s, it is imperative that such options are known to pregnant women as they evaluate their choices.

Diagnostic Testing

These are more reliable but risky tests used to confirm Down syndrome. They can identify chromosomal abnormalities with almost perfect acuuracy. However, they are invasive tests and increase the risk of having a miscarriage.

These tests include amniocentesis, chorionic villus sampling (CVS), and percutaneous umbilical blood sampling (PUBS). However, before undergoing any of these diagnostic tests, patients and their families should seek detailed genetic counseling to discuss their family history in relationship to the risks and benefits of performing these diagnostic procedures.

Amniocentesis, the removal and analysis of a small sample of fetal cells from the amniotic fluid, is widely available and involves a lower risk of miscarriage than chorionic villus sampling.

Amniocentesis cannot be done until the 14th to 18th week of pregnancy, and it usually takes additional time to determine whether the cells contain extra material from chromosome 21.

Chorionic villus sampling, conducted at 9 to 11 weeks of pregnancy, involves extracting a tiny amount of chorionic villi, tissue extensions that will eventually develop into a placenta. This tissue can be tested for the presence of extra material from chromosome 21. The villi can be obtained through the pregnant woman's abdomen or cervix. This type of sampling carries a 1-2% risk of miscarriage.

The third diagnostic method, percutaneous umbilical blood sampling or PUBS, is the most accurate method and can be used to confirm the results of CVS or amniocentesis. However, PUBS cannot be performed until later in the pregnancy, during the 18th to 22nd weeks, and carries the greatest risk of miscarriage.

Suggested Screening Guidelines

Pregnant women should be informed that:

1. If NT measurement is available, the full integrated test offers the highest screening performance for identifying Down syndrome pregnancy and should be offered to all pregnant women.
2. If NT measurement is not available, the serum only version of the integrated test will provide the best screening performance and should be offered.
3. If a woman wants the earliest available pre-natal diagnosis, and CVS is available, the 1st trimester screen with NT and serum markers should be offered. NT alone or serum markers alone should not be offered.
4. If a pregnant woman does not use prenatal screening before the 14th week, the 2nd trimester screening with quad markers should be offered. Second trimester screening with double markers should not be offered. ♥

DIAGNOSTIC TESTS FOR DOWN SYNDROME

AMNIOCENTESIS

- The removal and analysis of a small sample of fetal cells from the amniotic fluid.
- Cannot be done until the 14-18th week of pregnancy
- Lower risk of miscarriage than chorionic villus sampling

CHORIONIC VILLUS SAMPLING (CVS)

- Extraction of a tiny amount of fetal tissue at 9 to 11 weeks of pregnancy
- The tissue is tested for the presence of extra material from chromosome 21
- Carries a 1-2% risk of miscarriage

PERCUTANEOUS UMBILICAL BLOOD SAMPLING (PUBS)

- Most accurate method used to confirm the results of CVS or amniocentesis.
- The tissue is tested for the presence of extra material from chromosome 21
- PUBS cannot be done until the 18-22nd week
- Carries the greatest risk of miscarriage

Interlude

- When a problem gets too complicated for the physicists, they hand the problem to the chemists.
- When a problem gets too complicated for the chemists, it is handed over to the biologists.
- When the biologists think it is too complicated, they give the problem to the sociologists.

by Tapiwa Makunde

The Clinical Laboratory and Organ Transplantation

Organ transplantation is one of the most important advancement of modern day medicine. It involves the transfer of organ from one person to the other.

Most organs/tissues of humans can be transplanted. Some of the organs that can be transplanted are: kidney, heart, liver, lung, pancreas, cornea, skin, bone, and bone marrow among many others.

Laboratory Involvement

The laboratory is of importance in screening donors for infection; pre-transplantation donor and recipient testing; and long term monitoring of organ function and immunosuppressive therapy.

PRETRANSPLANTATION EVALUATIONS

Evaluation for Infection

All potential donors should be free from malignant tumors, sepsis, and other infectious diseases. The organ donor must be free of active infection as well. Donors with a recent history of infection documented by a positive blood, sputum, or urine culture must receive appropriate antibiotic treatment and should have negative culture results to be considered for donation. Prospective organ donors are usually screened for bacterial infections, Cytomegalovirus (CMV), HIV, Hepatitis B virus (HBV) and Hepatitis C virus (HCV) infections.

Evaluation for malignancy

Taking the impact of donor-transmitted malignancy on the outcome of organ transplantation, detection of malignancy is an important measure of donor suitability. However, as with other donor selection criteria, the acceptance of organs from donors at risk of transferring a malignancy to recipients must be weighed against the urgency of the transplant, and the recipient and/or the recipient's family must be informed of the potential risks.

Donor-Recipient Matching: ABO-Rh Compatibility Testing

ABO-Rh blood group compatibility of the donors and recipients should at all times be established. However, minor incompatibilities between donor and recipient do not necessarily preclude matching for transplantations since these can be countered by immunosuppression.

In most organ transplantations, compatibility of human leukocyte antigen (HLA) molecules is a routine exercise. One of the major biological functions of HLA molecules is to present antigenic peptides to T-cell receptors. Thus, allogenic HLA molecules in a transplanted organ induce strong immune reactions in the recipient that can require intervention to suppress. Preformed anti- HLA antibodies in the recipient (directed against donor HLA molecules) may be present due to previous pregnancy, multiple transfusions or previous transfusions. These antibodies should be screened for and identified so as to eliminate their adverse consequences

It is of particular importance to find the closest potential donor (in terms of ABO-RH and HLA compatibility) as possible in order to minimize chances of organ/ graft rejection.

POST TRANSPLANTATION MONITORING

Monitoring of success of transplantation

The recipient should be regularly monitored for transplant success through organ function analysis using serum or urine biochemical tests. Evidence of graft rejection should always be noted down.

Immunosuppressive Therapy

It is important to note that it's a very difficult, if not impossible, task to obtain a completely compatible match between the donor and the recipient of the organ. With this scenario in mind it is necessary to have immunosuppression in place in order to aid in organ survival. The general pattern of immunosuppression world wide for all organ transplants is to use a combination of agents, each in small doses, so as to get an added immunosuppressive effect, but without individual side effects of the different drugs.

Azathioprine, Cortico-steroids and Cyclosporin are usually used together. Each has different main side effects. Cortico-steroids stunt growth and cause a round face and impair the healing of wounds. Azathioprine can inhibit the bone marrow and cause anaemia and a low white cell count. Cyclosporin can cause increase in growth of hair and damage the kidney. When these three agents are used together the patient can generally tolerate the immunosuppression well. In this case the laboratory is important in therapeutic drug monitoring to ensure that the immunosuppressive drugs being taken by the patient do not reach toxic levels and remain within the therapeutic range.

Unfortunately acute rejection crises can still occur and are usually treated with a short course of high dose steroids or anti-lymphocyte globulin preparations. This powerful immunosuppression for rejection prevention may lead to infection particularly with viruses, causing severe cold sores and may activate CMV infection, which can cause a high temperature and specific bad effects on a variety of different organs. All immunosuppression methods predispose a patient to infection and tumour formation.

Despite these worries, most patients do well with organ grafts and after a time can be maintained with relatively low dosage of immunosuppression. With few exceptions however, stopping immunosuppression usually leads to acute rejection and chronic rejection

The clinical laboratory thus plays an important role in organ transplantation from the initial screening stages to the monitoring of organs transplanted into the recipient. ♥

Interlude

- A woman worries about the future until she gets a husband.
- A man never worries about the future until he gets a wife.
- A successful man is one who makes more money than his wife can spend.
- A successful woman is one who can find such a man.

HAPPINESS

- To be happy with a man, you must understand him a lot and love him a little.
- To be happy with a woman, you must love her a lot and not try to understand her at all.

Acute Coronary Syndromes (ACS):

Biomarkers in the prognostic stratification of non ST elevation acute coronary syndromes

Four components can be identified in acute coronary syndromes:

1. The presence of vascular damage;
2. The activation of inflammatory cells;
3. The formation of a sub-occlusive or occlusive thrombus;
4. The development of transient or irreversible myocardial dysfunction resulting in haemodynamic stress.

Biomarkers reflecting the severity of these four components, predict the risk of major adverse cardiac events and their predictive value is additive.

At the present time, however, only troponin levels impact in-hospital clinical decision making.

A - Components of ACS

1. Presence of vascular damage

Coronary atherosclerosis is highly prevalent in the population, yet it may remain clinically silent for many years or all through life. Nevertheless the greater the extent and severity of vascular damage, the higher the risk of acute coronary events happening and recurring.

2. Activation of inflammatory cells

The transformation of a stable atherosclerotic plaque into an unstable atherosclerotic plaque is frequently associated with the activation of inflammatory cells. This transformation is not limited to the culprit stenosis (responsible for clinical symptoms), but it can be widespread -throughout the whole coronary circulation and may even involve remote arterial districts as well as the myocardium.

3. Formation of a subocclusive or occlusive thrombus

The more intense and persistent the activation of inflammatory cells, the greater the thrombogenic stimulus. Thrombus formation is influenced not only by the intensity of inflammation, but also by the intensity of the thrombotic response.

4. Development of transient or irreversible myocardial dysfunction resulting in hemodynamic stress

The more intense the thrombotic response, the greater the risk of persistent coronary occlusion and of subsequent recurrences. The consequence of transient or permanent coronary thrombosis, in the absence of compensatory collateral circulation recruitment, is transient or irreversible myocardial dysfunction. The greater is the extent of myocardial dysfunction, the worse is the outcome for the patient.

B - Risk stratification of patients with non ST elevation ACS

Symptoms, signs and ECG findings

For many years physicians have based prognostic stratification of patients with non ST elevation acute coronary syndromes on symptoms, signs and ECG findings.

These tools remain of paramount importance, as they allow the identification of very high risk patients; for instance those with recurrent ischaemic episodes in spite of maximal medical treatment or those who develop evidence of heart failure during myocardial ischaemia, in particular in the presence of ST depression. In this subset of patients a step up of treatment is dictated by the clinical presentation and no further risk stratification is needed.

Biomarkers : an additive prognostic value

In the past ten years several studies have consistently demonstrated that in the remaining patients risk stratification can be based on biomarkers reflecting the severity of the four components of acute coronary syndromes mentioned above.

Indeed, lower values of creatinine clearance - a marker of vascular damage- are associated with a worse outcome.

Furthermore, higher serum levels of several markers of activation of inflammatory cells, including C-reactive protein (H-CRP) *, myeloperoxidase, plasma-associated pregnancy protein A and placental growth factor , are associated with a worse medium and long-term outcome, in particular mortality.

With regard to thrombus formation, higher serum levels of troponins*, caused by coronary microembolisation reflecting the thrombotic burden, or higher serum levels of sCD40 ligand - a marker of platelet activation- have consistently been associated with a higher recurrence rate of coronary instability.

Finally, raised levels of brain natriuretic peptide (Pro-BNP)* -a marker of ischaemic burden- as well as of the degree of underlying impairment of left ventricular function, hence a marker of haemodynamic stress, have been associated with a worse short and medium-term outcome, including mortality and recurrence of coronary instability.

One most note that the prognostic value of biomarkers of the four components of acute coronary syndromes is additive.

Biomarkers : variable clinical utility

However, the clinical utility of these four classes of biomarkers varies.

1. Biomarkers of vascular damage

Biomarkers of vascular damage do not impact on in hospital clinical decision making, but they are important for long term management.

2. Biomarkers of inflammation

Biomarkers of inflammation do not impact on clinical decision making yet. Indeed, the increased risk conferred by inflammation is not abated by currently available therapeutic regimens, including a very early invasive approach.

Recent circumstantial evidence suggests that it might be partially reduced with high doses of statins, thanks to their anti-inflammatory action; however, this notion needs to be tested in prospective studies. More importantly, a better knowledge of the triggers and molecular mechanisms of inflammation associated to acute coronary syndromes is warranted in order to design specific anti-inflammatory treatments.

3. Biomarkers of thrombotic burden

Biomarkers of thrombotic burden impact on in-hospital clinical decision making and should be measured in all patients with a

Continued on Page 14

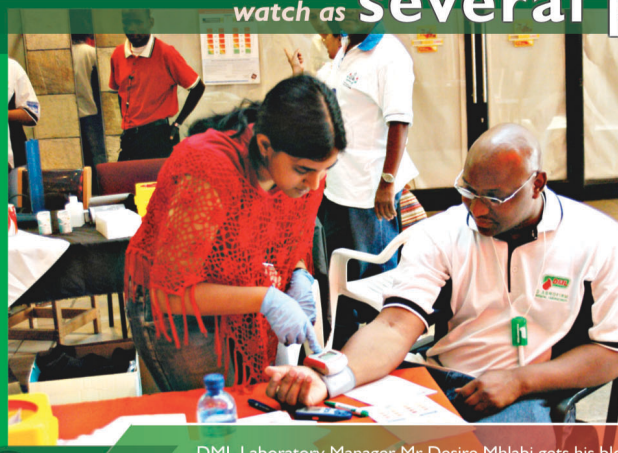
"Picture diagnosis..."

Mr. Mulamuli Moyo from DML Chemistry Dept. gives out information to the UB Students during the UB Health Awareness week

Ms. Peggy Le
holding a clo

watch as **several**

DML's finest Malebogo, Tracy, and Leslie



DML Laboratory Manager Mr Desire Mhlabi gets his blo
checked by Zakiyyah Chand

Mr. Silas Nunu and Mr. Chishamiso Mudenyanga
on World Diabetes day in Lobatse

The representative from WHO gives her speech on
Stanbic Bank World AIDS day

Mr. Silas Nunu from the DML Clinical Chemistry taking blood
from a patient to check his glucose level

burru from Stanbic Bank
ck donated by DML.

Mirriam, Leslie and Zakiyyah with a patient on World Diabetes Day - Riverwalk

Mrs W. Chand poses with DML Phlebotomist Getrude

people go through tests....

ood pressure

Leslie Rahman puts a couple under the hammer as she checks their glucose, cholesterol and blood pressure

DML QA Manager Mr Munyaradzi Mangwendeza giving his speech during the Stanbic Bank World AIDS day commemoration.

Mr. I. Chand donating a clock to Lobatse Town Council.

Mr. Silas Nunu Explains glucose testing results to a patient

Mrs. B. Mudenda explaining test procedures to a patient.

Mr. I. Chand donating glucometers to Lobatse Town Council

by Dr. D. Gangopadhyay, Cardiac Clinic.

MICROALBUMINURIA IN TYPE 2 DIABETICS AND IN HYPERTENSIVES: AN IMPORTANT, OVERLOOKED CARDIOVASCULAR RISK FACTOR

Increased attention has been given to the role of microalbuminuria as a cardiovascular risk indicator, particularly in patients with diabetes and hypertension.

Microalbuminuria typically is defined as a urinary albumin excretion (UAE) rate (or albumin excretion rate [AER]) of 30-299mg/day. In cross-sectional studies, the prevalence of microalbuminuria was 20-40% in patients with diabetes and in middle-aged individuals without diabetes.

Microalbuminuria is an independent predictor of stroke, death, and myocardial infarction. Moreover, in persons with diabetes and microalbuminuria, the risk of cardiovascular morbidity and mortality is estimated to be 1.8 times greater than for those with normoalbuminuria.

Although pharmacotherapies that reduce or prevent progression of microalbuminuria lower risk of progression to overt nephropathy, their effects in lowering subsequent cardiovascular morbidity and mortality have yet to be documented.

Many clinical factors complicate the understanding of the association between microalbuminuria and risk of cardiovascular morbidity and mortality. A key issue in understanding the relationship is to recognize that more than one risk factor (hyperglycemia, hypertension) may modify and contribute to a cardiovascular event.

Studies that examine the association are important for obtaining insight into the future role of pharmacotherapies that directly or indirectly alter albumin levels. However, the extent to which a progressive graded relationship exists is unclear. Furthermore, the definition of microalbuminuria is based on its ability to predict diabetic nephropathy (microalbuminuria, clinical proteinuria) rather than on cardiovascular morbidity and mortality.

Microalbuminuria: A marker for Cardiovascular Disease

In type 2 diabetics, microalbuminuria is the earliest clinical sign indicating vascular damage in the glomerulus, which is reflective of vascular disease throughout the body. Although only 20-40% of type 2 diabetics with microalbuminuria progress to overt nephropathy and ESRD, this figure would likely be much higher if these patients did not die from myocardial infarctions or strokes first. In other words, these patients do not live long enough to develop renal failure.

Epidemiologic evidence indicates that the presence of albuminuria is predictive of increased cardiovascular morbidity and mortality independent of other cardiovascular risk factors. There is a near linear relationship between increasing urinary protein excretion and both myocardial infarction and stroke in patients with type 2 diabetes.

These observations are also valid in patients without type 2 diabetes. Thus, screening for albumin, or protein in the urine, (even with lower levels below 150 mg/d) can have important predictive value for identifying patients who are more likely to experience a cardiovascular event.

Gerstein and colleagues analyzed information from the Heart Outcomes Prevention Evaluation (HOPE) study and noted that microalbuminuria was a powerful predictor for major cardiovascular events (myocardial infarction, stroke, or cardiovascular death) and all-cause mortality in patients with and without diabetes.

Of the more than 9000 high-risk patients who were screened for this study, an abnormal baseline urine albumin to creatinine ratio measurement was detected in 33% of patients with diabetes and in 15% of patients without diabetes.

A linear graded relationship was observed between the urine albumin and the creatinine ratio and cardiovascular morbidity and mortality. The large number of study participants allowed the investigators to note that the graded relationship between albumin to creatinine ratio and cardiovascular mortality was evident well below the standard microalbuminuria threshold (30 mg/g creatinine), indicating that screening should occur early so preemptive cardiovascular risk reduction therapy can be planned.

Microalbuminuria is also commonly associated with cardiovascular risk clustering phenomena. This has been most commonly called the metabolic cardiovascular syndrome and indicates the association between high blood pressure, left ventricular hypertrophy, central obesity, microalbuminuria, dyslipidaemia, glucose intolerance, and loss of nocturnal dip of blood pressure.

Consequently, the interplay of these risk factors leading to blood vessel damage may be best determined by the early appearance of microalbumin in the urine. Hence, a simple, clinical measurement of urine albumin is useful for not only identifying cardiovascular risk clustering phenomena, but also for identifying increased risk of cardiovascular events.

Microalbuminuria: A marker of Renal Injury

The presence of microalbumin in the urine heralds the onset of kidney disease in diabetics. It is not known whether it indicates greater risk for kidney disease in patients without diabetes.

The presence of albumin indicates early damage to the glomerular blood vessels; however, in diabetics it also could indicate failure of renal autoregulation. Although there is no direct clinical evidence to support this theory, microalbuminuria is evidence of generalized impairment of vascular function, and one of the critical responsibilities of the afferent glomerular arteriole is to assist in the regulation of glomerular hemodynamics. This may be an important concern in patients with diabetes.

Consequently, improved control of glomerular capillary pressure with better systemic blood pressure control with drugs that block the renin-angiotensin system have increasing importance to avoid progressive renal injury.

The relationship between albuminuria and progressive renal injury is not completely understood.

On one hand, the albuminuria could be indicative of elevated glomerular capillary pressures that over time could lead to mechanical stretch and strain and resultant vascular injury of the filtering apparatus.

On the other hand, there is experimental evidence to indicate that renal epithelial cell uptake of filtered proteins may lead to an abnormal accumulation of proteins in the endolysosomes and endoplasmic reticulum that could stimulate nuclear signals for vasoactive and inflammatory genes.

The subsequent release of these vasoactive and inflammatory substances

within the renal interstitium could lead to myofibroblast proliferation, fibrogenesis, and subsequent scarring.

The presence of glomerular capillary hypertension, albuminuria, and renal tubular epithelial cell reabsorption of filtered proteins may be the necessary combination of events to serve as a final common pathway for progressive renal injury.

These observations also raise questions about optimal strategies to protect kidney function and whether specific antimicroablbuminuric or antiproteinuric regimens are as important as, or more important than, other cardiovascular risk reduction strategies, such as lowering blood pressure or lipids, in preventing progression or renal disease.

Screening for Microalbuminuria

Microalbuminuria can be defined by different measurements as shown in the Table. There are three methods for screening for microalbuminuria: measurement of spot urinary albumin to creatinine ratio, 24-hour urine collection with albumin and creatinine, and a timed 8-hour or overnight urine collection to measure the albumin to creatinine ratio.

The spot determination is the easiest to use in an office setting and often provides accurate information. It has been demonstrated to be at least 90% sensitive for determining microalbuminuria compared with a 24-hour urine collection, even after adjusting for age and gender.

It is best performed with first morning void urine because of the known diurnal variation in albumin excretion.

If urine collections are used, the ratio of the albumin (in milligrams) to creatinine (in grams) should be measured, as the timed collection of urine specimens is often difficult to perform accurately. Because urine creatinine production is constant on a daily basis (it reflects the muscle mass and its breakdown products) the only variability is the urine albumin excretion.

This obviates the need for a timed specimen and encourages the use of spot specimens for screening. The spot specimen can also serve as a means for determining response to anti-microalbuminuric therapy. The simplicity of the measurement allows for its use for monitoring purposes.

Because of the variability in urinary albumin excretion, two or three specimens collected within a 3-to-6-month period should be abnormal before considering the patient to have crossed one of the diagnostic thresholds of albuminuria. As discussed previously, there is a continuum of risk based on increasing albumin in the urine even below the traditionally reported thresholds, as outlined in the Table.

Note that normal is defined as < 30 mg/g creatinine. Microalbuminuria is 30-299 mg/g creatinine, and overt nephropathy or clinical albuminuria is > 300mg/g creatinine. One can also use 24-hour collections in milligrams per 24 hours or timed collections in micrograms per minute.

However, for simplicity, spot collections evaluating milligrams of albumin per grams of creatinine should be used as the standard. Also, please note that some of the variability in urinary albumin excretion can be based on exercise, infection, or fever within the previous 24 hours.

In addition, poor glycaemic control, marked hypertension, congestive heart failure, urinary tract infection, protein and salt intake, and even haematuria may elevate urinary albumin excretion over baseline.

Screening should always be performed by measuring microalbuminuria, not by measuring protein. Protein measurements in the urine are not sensitive at lower determination levels and are unreliable. Screenings with reagent tablets or dipsticks may also be used since they show acceptable sensitivity and specificity when carried out by trained personnel.

However, the reagent strips only indicate concentration and do not correct for creatinine as the spot urine albumin to creatinine ratio does. Thus, there is greater potential for errors in quantification of albumin in the urine based on variations in urine concentration. Any positive test by a reagent strip or tablet should be confirmed by a more specific spot urine albumin to creatinine ratio for quantitation purposes.

Table: Diagnostic Criteria for Albuminuria

TYPE	SPOT SPECIMEN (NOT TIMED)	24-HR SPECIMEN (TIMED)
Normo Albuminuria	< 30 mg/g creatinine	< 30 mg/day (< 20 µg/min)
Micro Albuminuria	30-299 mg/g creatinine	30-299 mg/day (20-200 µg/min)
Macro Albuminuria (overt nephropathy)	≥ 300 mg/g creatinine	≥ 300 mg/day (≥ 200 µg/min)

Implications for Clinical Practice

Earlier identification and management of patients with microalbuminuria or clinical proteinuria is imperative. Microalbuminuria is a much more easily treatable clinical problem than proteinuria in that the blood pressure tends to be lower, renal function is usually less impaired, and less medication to reduce albuminuria is required.

ACE inhibitors and angiotensin II receptor blockers have been convincingly demonstrated to have renoprotective benefits. The majority of the data with the ACE inhibitor is in type 1 diabetics and in patients with nondiabetic renal disease. The more recent data with angiotensin II receptor blockers is confined to patients with type 2 diabetes; however, more often than not these patients will require multiple drugs to achieve the lower levels of blood pressure, which may be important in correcting glomerular capillary hypertension and optimally reducing proteinuria.

Newer strategies to use combinations of drugs will likely prove to be important. Early studies indicate that using an ACE inhibitor and an angiotensin II receptor blocker together may be better than using either one alone in the approved dosing range to reduce microalbuminuria or proteinuria.

The addition of a thiazide diuretic or a nondihydropyridine calcium antagonist may also amplify the antiproteinuric effects of drugs that block the renin-angiotensin system. Moreover, most of the renal outcome trials with ACE inhibitors or angiotensin II receptor blockers included diuretic support and/or calcium antagonists to achieve better blood pressure control.

More studies are also needed to evaluate higher doses of ACE inhibitors and angiotensin II receptor blockers beyond those which have been shown to be useful for high blood pressure, to see whether better antiproteinuric effects and enhanced renal protection can be achieved.

Additionally, clinicians need to be aware that modification of dietary salt consumption is crucial to optimize the antiproteinuric effects of both ACE inhibitors and angiotensin II receptor blockers because increasing dietary sodium will offset both the antihypertensive and antiproteinuric effect of these drugs.

Continued on Page 14

by Silas Nunu

Systemic Rheumatic Diseases and the Laboratory

Testing for systemic rheumatic diseases (SRD), which can be highly variable diseases, can be complex and is often misunderstood. In today's healthcare environment, it is critical that the proper tests be performed not only from the point of view of reducing patient care costs, but also to avoid their mis-diagnosis through the use of just clinical evaluation as a basis.

Some patients are difficult to diagnose, especially those still in the early stages of the disease or those that have overlapping symtoms.

The disease manifestations are highly variable, may progress, and in some cases evolve into or coalesce with other autoimmune diseases in many different combinations (generally called "overlap syndromes").

The most common SRDs are systemic lupus erythematosus (SLE), Sjogren's syndrome (SjS), systemic sclerosis, rheumatoid arthritis (RA), autoimmune vasculitis, and mixed connective tissue disease (MCTD).

As might be expected, the symptoms range in severity and may progress or evolve from being extremely mild to severe and protracted to even fatal.

While no single pathogenic mechanism has been elucidated for autoimmune diseases, they share the commonality of autoantibodies, which are a diagnostic hallmark of all SRDs. These are distinct profiles of auto antibodies directed to intracellular antigens can be detected in the system of the afflicted individual. These can be used individually or in combination to characterise a patient's specific disease.

Because of the wide and often confusing reactivity of the major rheumatic autoantibodies – double-stranded DNA (dsDNA), chromatin, Sm, ribonucleoproteins (RNP), SS-A, SS-B/Ro, Scl-70, Jo-1 and CCP – intepretation of the results by the laboratory can be important in the specific diagnosis of SRDs.

Several studies have suggested that overuse of common serum rheumatic tests, including antinuclear antibody (ANA) and rheumatoid factor (RF) measurements, leads to unnecessary referrals and further laboratory work-ups.

Failure to use these tests in a knowledgeable and thoughtful manner can result in diagnostic confusion and increased costs. It is thus important that we look at the individual common SRDs and the tests that can be purposefully used in diagnosing them.

SLE – Systemic lupus erythematosus

SLE is an autoimmune disorder striking every organ system. Its many presentations can mimic many diseases, thus making its initial presentation highly variable. This disease requires close cooperation between the treating physician and the laboratory.

In patients with suspected SLE, appropriate screening laboratory studies include a complete blood count, erythrocyte sedimentation rate, urinalysis, biochemical profile and antinuclear antibodies.

There are 11 criteria for the diagnosis of SLE, and in order for a definitive diagnosis to be made; a patient must meet at least four. Two of the criteria are positive ANA and the detection of antibodies to Smith (Sm), double-stranded DNA (ds-DNA) or cardiolipin.

The presence of antibodies to dsDNA is also one of the criteria for the diagnosis of SLE and these antibodies are associated with active disease. 30-70% of patients with SLE will be anti-dsDNA positive.

Another commonly occurring marker for SLE is the presence of antibodies to chromatin. These occur more often than antibodies to dsDNA, and they are also associated with glomerulonephritis. These antibodies are responsible for the LE cell phenomenon in which nuclei are phagocytosed by mature polymorphonuclear leucocytes and digested.

So in general the tests to be used to get a conclusive diagnosis of SLE are dsDNA, chromatin, Sm, RNP, SS-A and SS-B.

Scleroderma (Systemic Sclerosis)

This disease usually affects people 30 to 50 years old, and is characterised by Raynaud's phenomenom and excess deposition of collagen in the skin and organs. Hence the disease has extensive organ involvement.

The scleroderma antibodies Scl-70 and centromere can be used to differentiate between limited and diffuse scleroderma. Patients with limited scleroderma have skin changes limited to the extremities whilst those with diffuse scleroderma have skin changes affecting the trunk and the extremities.

The more typical antibodies seen in Scleroderma are:

1. **Anticentromere Antibodies:** these are found in up to 90 percent of patients with limited systemic sclerosis. In contrast, they are found in only 10 percent of patients with diffuse scleroderma. The patients, who have limited Scleroderma, are more likely to have this antibody. People with Raynauds phenomena, who have positive anticentromere antibodies, are more likely to develop limited Scleroderma.
2. **Anti- Scl-70 antibodies:** This is the antibody to the topoisomerase-1 antigen. Topoisomerase-1 is involved in the uncoiling of DNA doing transcription. It is seen in up to 40 percent of patients with diffuse Scleroderma. It is not usually seen in limited Scleroderma.
3. **Other antibodies,** including Rheumatoid factors may be seen in a small percentage of patients.

Sjorgrens's Syndrome

Clinically it is characterized by dry eyes and mouth. The eyes may become red, burn and itch and the dryness of the mouth can cause difficulty in swallowing and talking.

The patients also have autoimmune symptoms. Patients with Sjogren's syndrome typically produce a myriad of autoantibodies, these include ANAs, which are present in nearly all patients.

The other typical antibodies that are found in most, but not all patients are SS-A and SS-B antibodies, rheumatoid factor and thyroid antibodies.

Sjogrens syndrome can be classified into either primary or secondary SjS. These are different in that primary does not show symptoms of other SRDs whilst secondary shows symptoms associated with RA, SSc, and SLE. These two can be differentiated by that antibodies to SS-A occur alone in secondary SjS whilst they occur together with anti SS-B in primary SjS.

Rheumatoid Arthritis (RA)

RA is characterized by bilaterally symmetrical polyarthritis involving three or more joints and morning stiffness lasting more than one hour.

Approximately 80% of patients with RA have antibodies to RF. While some patients

with RA have a mild disease, a majority of patients diagnosed with RA progress to an erosive arthritis therefore early diagnosis of rheumatoid arthritis and early, aggressive treatment can help prevent joint damage and deformity.

The traditional markers associated with a poor prognosis in RA include high titers of RF, increased sedimentation rates, presence of C-reactive protein, and the development of erosions as determined by X-ray.

Another useful test for the diagnosis of RA is the anti- cyclic citrullinated peptide (anti- CCP) test. If present in a patient at a moderate to high level, it not only confirms the diagnosis but also indicates that the patient is at increased risk for damage to the joints.

While the RF is more common in RA patients, many patients with a positive RF do not have RA. Furthermore, the presence of the RF has less prognostic significance than the CCP.

The current trend in rheumatology is to identify those patients that may develop more severe RA early in the course of the disease so that more aggressive intervention can be started hence the importance of anti-CCP.

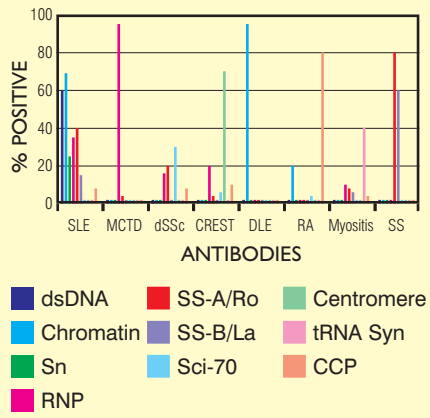


Figure 1 - The chart shows the percent of patients who are positive for a specific antibodyin each of the major SRDs. These include SLE, MCTD, dSSC(diffuse scleroderma), DLE(drug induced lupus), RA, myositis, and SjS

What has been illustrated here is the complexity involved with autoimmune disorders and the role, which the laboratory can play in alleviating possible problems arising from these complexities and helping the clinician come up with a valid diagnosis.

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Erythropoietin Therapy to Treat Anaemia in Chronic Heart Failure

Anaemia has been found to affect nearly a quarter of heart failure patients. It is associated with a worse prognosis, an increased risk of hospitalisation and reduced exercise tolerance. Although the mechanism is unclear early reports suggest there may be some benefit to correction of anaemia by erythropoietin therapy. We await the outcomes of randomised controlled trials to know if this line of treatment is effective.

Introduction

Chronic heart failure (CHF) is a multi-organ syndrome with changes in the periphery affecting muscle and blood vessels, neurohormonal activation and metabolic and hormonal changes. It is now recognised that anaemia is also frequently part of this syndrome.

Epidemiology and pathophysiology

- If we define anaemia as a haemoglobin (Hb), less than 12.0 g/dl anaemia affects between 10-25% of CHF patients with more in the sicker patients' population.
- The cause of anaemia in chronic heart failure is not known.
- In many cases anaemia in chronic failure shows similarities to the anaemia of chronic disorders, in which there is a defect in iron utilisation.
- The commonly recognised causes of anaemia in a general population such as iron, folate or B12 deficiency can exist in CHF and should be looked for.
- Blood loss can also be excessive in patients on anti-platelet and/or anticoagulant regimes for their primary cardiovascular disorder.
- In most CHF cases with anaemia, however, there is no identifiable cause.
- Aetiological factors are thought to include bone marrow dysfunction secondary to poor perfusion, impaired renal function and the effects of cytokine activation -especially TNF-alpha- which can induce both reduced erythropoietin production and increased resistance to its effects.
- In severe heart failure immune activation and pro-inflammatory cytokines correlate strongly with the severity of anaemia.

Consequences

Anaemia has an independent effect on exercise intolerance by further reducing an already impaired oxygen delivery to the exercising muscle groups.

Studies in large heart failure populations including cohorts from several of the large outcome trials in CHF have shown an independent detrimental effect of anaemia on the risk of death or admission to hospital for worsening heart failure.

These findings suggest that treatment of anaemia might have the potential to improve outcomes.

Treatment

Where an identifiable cause of anaemia in chronic heart failure is found it should be treated appropriately.

We know that both erythropoietin or IV Iron treatment can increase haemoglobin levels in CHF and in early reports this has been associated with improved well-being and exercise tolerance.

Whether this is with an acceptable safety profile and indeed whether it reduces the risk of mortality and morbidity is presently unknown. This exciting new treatment option awaits proper randomised outcome trials in the next few years.

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Continued from Page 1

From My Keyboard

headway is made toward reaching any goal. So if you've made 10 New Year's resolutions, trim them back to three.

2. Be specific

Another formula for failure is to set your sights on behaviors that are too vague. Be specific on what you really want to do and how you will do it.

3. Be realistic

A common pitfall is setting goals that are too superior. For example, in terms of exercising resolving to run several miles a day when you haven't been exercising at all is a definite way of falling short of your goal. Instead find advise from expert on how you can set up achievable goals.

4. Choose what is important to you

You're more likely to keep a resolution if the motivation is coming from you, not someone else. You must also match your interests and values to your goals. People find it easier to reach structured, self-set goals.

5. Pick a goal AND a strategy

And remember, it's not enough to just have a goal, you must come up with a strategy for reaching a goal that's rooted in practical steps. Figure out ways to make the desired behaviors more or less automatic.

The most important thing is that you make tangible strategies about how you will go about achieving your goals. The result could be a happier, healthier new year for you.

Wish you all the best in the New Year and may all your dreams come true.

Stay informed.

Munyaradzi Mangwendeza
moonya@diagnostics-update.com

Continued from Page 1

Recent Events @ Diagnofirm

for test results. Before its purchase, two separate machines were in use for the tests the modular can do.

The Hitachi Modular system comes with a state-of-the-art software system, the PMS system, that allows the user to view all results from all other analyzers in the laboratory. This allows for improved result management as all the patient's results can be reviewed and validated at once. This can also serve as a mini-server of results.

We hope this machine will help us cope with the ever-increasing demand for medical laboratory services. For the benefit of our clientele and in the name of quality service provision, we shall continue to keep abreast with technological advances.

DML and You!

Though 2005 was a challenging year for most, it did not deter DML from fulfilling its social obligation of helping the needy. During the commemoration of World Diabetes Day, we donated 50 glucometer machines to the Diabetes Association of Botswana. Also on World AIDS Day we joined the nation in marking this day by providing free voluntary counseling and testing at Tsholofelo Park in Gaborone.

Led by our Managing Director, Mr Chand, the DML team worked hard on that day to attend to a total of 300 volunteers. All of them walked away with free caps and T-shirts courtesy of DML.

Graceful Touch!

Other places that have felt our graceful touch include the University of Botswana where we did free cholesterol and glucose testing and Lobatse Town Council where we provided free glucose testing.

Over 120 people were tested and to cap it all DML donated 10 glucometer machines to the town council. On the morning of Saturday 11th November, shoppers at Riverwalk were pleasantly surprised to find the DML road show offering free glucose, cholesterol and blood pressure tests to the public. Over 150 people participated and walked home with vital information about their wellness.

Devotional Day

The morning of Sunday 12th December saw DML participating in the BCL HIV/AIDS Devotional Day in Selebi Phikwe. The day started with a 7K walk from the BCL mine to Makhubu Cricket Club. Mr Chand, Desire and Mula represented DML in the march.

The function was also attended by Mrs Chand, Mohamed Chand (Jnr), as well as Blessing and Nicholas from our Selebi Phikwe branch. DML generously donated HIV testing kits and caps and T-shirts.

Healthy 2006!

Mulamuli Moyo

Continued from Page 11

Microabluminuria

Other antihypertensive medications are capable of reducing microalbuminuria/proteinuria beyond what would be expected by their antihypertensive effects. Perhaps the most useful are the nondihydropyridine calcium antagonists.

These drugs, either alone or in combination with other drugs like ACE inhibitors, help lower both blood pressure and microalbuminuria/proteinuria. Most of the clinical trials were performed in patients with clinical proteinuria. There has been some concern that dihydropyridine calcium antagonists may increase or not change proteinuria in patients when used without a renin-angiotensin-system-blocking drug. However, when used with ACE inhibitors or angiotensin II receptor blockers, this is not a concern.

Conclusions

Screening for microalbuminuria should be performed on a yearly basis in every patient with type 2 diabetes.The presence of microalbuminuria indicates that a patient is at greater cardiovascular risk and consequently will require lower blood pressure goals (less than 130/80mmHg) and more specific attention to lipids, glucose, diet, and platelets, as well as antimicro-albuminuric therapies.

Continued from Page 7

ACS

non ST elevation acute coronary syndrome clinical decision making. Indeed, Patients with raised serum levels of troponins exhibit a better outcome if treated with a potent antithrombotic regimen and an invasive approach, while patients with normal levels are not penalised if treated with a cheaper and more comfortable conservative approach.

To measure this biomarker, different essays are commercially available and it is impossible to define a fixed cut off value. However the cut off value for each essay should exceed the concentration corresponding to a total analytical imprecision of 10%.

4. Hemodynamic stress

Biomarkers of haemodynamic stress of do not impact on in hospital clinical decision making, but they are important for long term management.

*** HCRP, Pro BNP and troponin T testing is all available through Diagnofirm® laboratories.**

by Munyaradzi Mangwendeza

Helicobacter Pylori

Helicobacter pylori are gram-negative microaerophilic bacteria, which normally resides in the stomach. The bacteria, *H. pylori*, usually don't cause problems in childhood. However, if left untreated the bacteria can lead to digestive illnesses, including gastritis (the irritation and inflammation of the lining of the stomach), peptic ulcer disease (characterized by sores that form in the stomach or the upper part of the small intestine, called the duodenum), and even stomach cancer later in life.

Even though the bacteria can cause these illnesses, most *H. pylori* infections produce no symptoms. The bacteria are found everywhere in the world, but especially in developing countries, where up to 10% of children and 80% of adults can have laboratory evidence of an *H. pylori* infection, usually without having any symptoms.

The most important risk factor is low socioeconomic status characterized by poor sanitation and overcrowding.

Anyone, including a child, can have an *H. pylori* infection without knowing it. When symptoms do appear, they're usually either symptoms of gastritis or peptic ulcer disease.

In children, symptoms of gastritis may include nausea, vomiting, and frequent complaints about pain in the abdomen. However, these symptoms are seen in many childhood illnesses.

H. pylori can also cause peptic ulcers (commonly known as stomach ulcers). In older children and adults, the most common symptom of peptic ulcer disease is a gnawing or burning pain in the abdomen, usually in the area below the ribs and above the navel. This pain typically gets worse when someone with ulcers has an empty stomach and improves as soon as he or she eats food, drinks milk, or takes antacid medicine.

Children who have peptic ulcer disease can have ulcers that bleed, causing hematemesis (bloody vomit or vomit that looks like coffee grounds) or melena (stool that's black, bloody, or looks like tar).

Younger children with peptic ulcer disease may not have symptoms as clear-cut as those of older children, and their illness may be harder to diagnose.

H. pylori infection may be contagious, because the infection seems to run in families and is more common where people live in crowded or unsanitary conditions.

The diagnosis of an *H. pylori* infection by using many different types of tests.

1. The Gold Standard

The gold standard is the culturing of the bacteria from a biopsy. The bacteria are fastidious in growth requirements, as well as, the transport to the laboratory. The biopsies should be transported in saline if they will be processed within two hours but a transport medium containing glycerol if the biopsy will be processed later.

After successful culture, the morphology on the agar plates will be: tiny colonies < 1 mm diameter, glistening, translucent convex with entire edges and with a small area of alpha haemolysis.

On staining, the bacteria are gram negative curved or horseshoe shaped cells. The biochemical tests used are urease, catalase and oxidase.

2. Non-invasive urea breath test

The test is based on the fact that urease activity of the bacteria splits carbon dioxide from the ingested labeled urea. After ingesting labeled urea, labeled carbon dioxide can be detected in the patient's breath.

The test is used mainly to detect infection and determine successful treatment. It is an economical, non-invasive test and rapid test, which can be done in an office to detect *H.pylori*. The disadvantage is that it requires a high density of bacteria to give a reliable test.

3. Histology biopsy

Normal gastric mucosa does not contain inflammatory cells. *Helicobacter pylori* infection is associated with a high infiltration of inflammatory cells. These can be seen on a histological examination of an endoscopic biopsy specimen using special stains.

4. Rapid urease test

The test is based on the principle that *H. pylori* contains a large amount of enzyme urease, which splits urea into ammonia and carbon dioxide. When gastric containing *H. pylori* is placed into a urea-containing medium, the ammonia produced by the bacteria will increase the pH, which is easily detected by the colour change of the pH indicator.

5. Serologic tests

Detection of serum IgG against Helicobacter produces a reliable assessment of the current or prior infection. The antibody test remains positive even after successful treatment and a positive result cannot be used to ascertain whether the patient has been adequately treated or not. Its major limitation is the inability to distinguish active infections from past infections.

Doctors treat *H. pylori* infections using antibiotics. Because a single antibiotic may not kill the bacteria, a patient may be given a combination of antibiotics.

There is no vaccine against *H. pylori*. And because transmission isn't clearly understood, prevention guidelines aren't yet available. However, it's always important to make sure that:

- Wash your hands thoroughly.
- Eat food that's been properly prepared.
- Drink water from a safe source.

All patients with peptic ulcer disease should be tested for *H. pylori* and those with *H. pylori* infection should be treated. Positive identification should be followed by treatment. ♥

Interlude

ROMANCE MATHEMATICS

Smart man + smart woman = romance
Smart man + dumb woman = affair
Dumb man + smart woman = marriage
Dumb man + dumb woman = pregnancy

OFFICE ARITHMETIC

Smart boss + smart employee = profit
Smart boss + dumb employee = production
Dumb boss + smart employee = promotion
Dumb boss + dumb employee = overtime

Assisted Reproductive Technologies and the role of the Clinical Laboratory

There are millions of couples that are unable to conceive their own children and are seeking ways to improve their fertility status. Advances have been made in assisted reproductive technologies, allowing more couples to have their own children than ever before.

In 1978, there was the first successful pregnancy from in vitro fertilization (IVF). These technologies range from fertilization of retrieved eggs in vitro and placement of the fertilized eggs directly into the uterine cavity, to direct injection of a single sperm into the cytoplasm of an oocyte to achieve direct fertilization in vitro.

Now that assisted reproductive technologies are becoming more widely available, the need for laboratory services that support these techniques is growing.

Certain abnormal anatomical structures, and sometimes disease, in people lead to infertility due to the effect they have on the steps of pregnancy development. This results in the person not being able to conceive a child and hence experience normal pregnancy.

The main causes of infertility have been cited as ovulatory disorders (30%); abnormal sperm production and function (25 %); reproductive tract defects, involving the cervix, uterus, and fallopian tubes (22%); and endometriosis (5%).

It has also been noted that infertility occurs more in females than in males since there are more factors that contribute to the process of pregnancy in females than in males.

When it comes to males, it has been noted that a reduction in sperm count, as well as the decrease in the number of sperms with normal morphology, are the principal reasons for poor pregnancy rates attributed to males.

The laboratory plays a very central and pivotal role in all the technologies currently used for in vitro fertilization. A successful pregnancy cannot be attained without the involvement of the laboratory. For example, the timing of unfertilized egg harvesting and the timing of implantation of the fertilized egg are all dependent on the results attained from the laboratory. The timing and monitoring of each stage of the in vitro fertilization (IVF) with accurate and precise hormone measurements is important in the success rate of IVF technologies.

The measurements of the follicle stimulating hormone (FSH), leutinizing hormone (LH), human chorionic gonadotropin (HCG), oestradiol (E2) and progesterone form the basis of the support provided by the laboratory for IVF.

The measuring of E2, FSH and LH is used to establish the adequate baseline in the woman before the use of ovulation induction methods. It is also possible to tell the number of fertilizable oocytes and the pregnancy rate per IVF attempt from the ratio of LH to FSH which correlates with the E2 concentration.

Another important measurement is that of FSH on the 3rd day of the menstrual cycle. A recorded FSH level less than 15mIU/ml is associated with a clinical pregnancy rate of approximately 23% per cycle. Levels higher than 25 mIU/ml are associated with a pregnancy rate less than 5% per cycle.

During the process of ovulation induction, an accurate and precise measurement of serum E2 is important in monitoring the process of follicular development and for determining exactly when to apply the ovulation stimulant.

The observed pattern of oestradiol rise during this period will give a good indication of how successful the pregnancy will be, and when used together with ultrasound, will reflect the number of follicles that would have ripened.

Hyper stimulation of the ovaries will occur when the E2 levels have exceeded 2000 pg/ml and when more than 15 follicles reach a diameter of 12 mm. When the fertilized eggs have been transferred to the uterus, the luteal phase adequacy must be closely monitored with HCG and progesterone measurements.

It is important to note that the success rate per IVF cycle really depends on the type of IVF procedure employed as well as the method of ovary stimulation used to provide a retrievable oocyte. However, the method which currently gives the best results is pulsate stimulation of the ovary with GN-RH (gonadotropin releasing hormone) used together with the use of gonadotropins.

The oocytes produced by this method have the highest fertilization and successful pregnancy rate.

Of late, it has been noted that multiple gestations have been on the rise with twin babies up by 33% and those of the higher order like triplets and quadruplets up by 178%.

Multiple gestations are basically a result of the number of fertilized eggs transferred to the fallopian tubes or the uterus. Sometimes this cannot be avoided as the success rate of IVF is increased when multiple fertilized eggs are transferred and hence the increased probability of a multiple gestation pregnancy.

The results of a twin gestation pregnancy can be undesirable at times. These include prematurity (39%), low birth weight infants (18%) and perinatal mortality rate (3.5%).

It is important to note that these statistics are much higher for triplets and other higher numbers of offsprings. About 20% of IVF pregnancies will experience a spontaneous abortion, while 5% of fallopian tube transfers will result in an ectopic pregnancy.

If successful, early pregnancy will need to be monitored using transvaginal sonography and serial HCG levels. The IVF rate for congenital abnormalities is 3.5 %, which is slightly higher than conventional pregnancies.

One major concern for IVF is the cost. Assisted reproductive technologies are not covered by medical aid and patients will have to pay cash for the services. Cost will vary widely depending on the number of reproductive cycles attempted to induce ovulation and the number of cycles needed for normal implantation. It is also much more expensive for women who cannot produce their own viable oocytes and have to use donor eggs.

Without good laboratory methods for the hormone measurements and semen analysis, all the IVF programs would not boast such excellent success rates.

Reference: Laurence M. Demers, PhD (DABCC): Clinical Laboratory News.