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In their opinion no which whether to embark on the medical laboratory science degree work for one year as a laboratory technician before deciding was very Female non-diabetic patients had a significantly higher prevalence of asymptomatic bacteriuria among out-patients of programme. Sue hopes that in the future this will attract the bright young ones for a career path in medical laboratory science.

Richard Omoregie and colleagues from Nigeria report on the prevalence of asymptomatic bacteriuria among out-patients of a tertiary hospital. They found a prevalence of 59.0% in diabetic patients versus a prevalence of 24.5% among non-diabetic patients. Female non-diabetic patients had a significantly higher prevalence of asymptomatic bacteriuria. Staphylococcus aureus was the most common uropathogen in patients.

In a Viewpoint Article, Sue Carnoutsis from Canterbury Health Laboratories reflects on her training as a medical laboratory scientist in the pre-degree era and states that the reason she started was very clear to her, namely a career path in medical laboratory science. She believes that with the advent of the degree based qualification this reasoning has changed in that current employees, upon graduation with a medical laboratory science degree, make career choices based on student loan debts, rather than what best matches their skills and education. This becomes a problem for employers in regard to recruitment and retention of staff. For this reason, Canterbury Health Laboratories has started a “gap year” programme in which potential future medical laboratory scientist work for one year as a laboratory technician before deciding whether to embark on the medical laboratory science degree programme. Sue hopes that in the future this will attract the bright young ones for a career path in medical laboratory science.

Gibbs and Read from North Shore Hospital present a case study of malignant meningitis consistent with a diffuse large B-cell lymphoma. The diagnosis was made possible by the examination of the CSF by a medical laboratory scientist with cytology experience who determined that the sample contained highly atypical lymphoid cells which upon cytological examination produced the diagnosis. They discuss the case and the advantages of cross disciplinary training in the laboratory.

In a Technical Communication, Christian presents a comparison of intact parathyroid hormone assays (iPTH) by two chemistry analysers he performed as a requirement for his BMLSc degree. He found a significant increase in the mean difference in higher iPTH with the Advia Centaur analyser compared to the Roche Modular E170 analyser and better internal reliability of iPTH assayed on the Advia Centaur analyser.

Med-Bio Journal Award

Med-Bio offers an award for the best article in each issue of the New Zealand Journal of Medical Laboratory Science. All financial members of the NZIMLS are eligible. The article can be an Original, Review or Technical Article, a Case Study or a Scientific Letter. Excluded are Editorials, Reports, or Fellowship Treatises. No application is necessary. The Editor and Deputy Editor will decide which article in each issue is deemed worthy of the award. If, in their opinion no article is worthy, then no award will be made.

Their decision is final and no correspondence will be entered into. Winner of the Med-Bio Journal Award from the April 2008 issue were Anna Denholm and David Patterson from the Haemostasis Laboratory, Canterbury Health, Christchurch for their article “Accurate diagnosis of high-affinity vWF-platelet disorders: a case study of pseudo von Willebrand disease”, N Z J Med Lab Sci 2008; 62 (1): 7-9.

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Are we the problem and is “Y” the answer?

Sue Carnoutsos, Operational Development Facilitator
Canterbury Health Laboratories, Christchurch

I love my job as a medical laboratory scientist! However if I sit down and try to analyse why, things get a little tricky – after all it is a big question!

As the daughter of a very much middle class New Zealand, born in the (very) late 50’s there were few career choices. I could be a teacher, or a secretary or even a nurse. A career in science was way outside the realms of possibility – and then there was the expectation of marriage and a family. But somehow I fell in to this profession and here I have stayed. When I say “fell” that is exactly what it seemed. Not much planning, just a phone call and a job offer as a laboratory assistant in the then Blood Bank at Christchurch Hospital – no interview, no expectations - just turn up Monday! I never had time to think what I wanted out of a job and if I had to analyse it now it would be that it interested me and offered job security. Aspects of pay never came in to it! I was not there to change the world or find a cure for cancer – I was there to work.

Along the way, the need for a career must have kicked in, along with the desire to study again, and I began training as a medical laboratory scientist (or technologist as we were known then) under the “work while you study” training available. You now hear this referred to as “the old system”. The reason I started as a trainee is still very clear to me – it was a career path in medical laboratory science. I can remember being very sure that this is what I wanted to do. After all I had spent time in labs, I knew the people and I knew the expectations of the position and the rewards that it could bring.

The advent of the degree based qualification has changed things somewhat. I have interviewed so many new graduates that when asked where they saw themselves in 5 years time, have stated “in your job”. When you look at the figures relating to average age in the laboratory workforce alongside the length of time new graduates are retained in the profession you can understand why this isn’t happening however you can’t knock the self belief!

So when does the disillusionment with medical laboratory science kick in? Is it before we even get to 1st year health sciences? Back as far as course selection at High School perhaps? Core sciences of chemistry, biology and physics are competing with exotic languages, art history and computing to name but a few. One careers advisor recently commented that core sciences were taken as a career path to medicine – surely he meant 1st year health sciences and surely he couldn’t be that narrow minded in terms of career options, could he?

So if course selection is eliminated as a barrier to a career in medical laboratory science, then perhaps it is in work expectation or the opportunity for a career path outside of a management role? Ask yourself what constitutes an ordinary day in the diagnostic laboratory. Yourself what constitutes an ordinary day in the diagnostic laboratory.

To my mind we have two distinct problems here – replacement of the current workforce by individuals who want to work in laboratories as a first career choice and the lack of a career pathway focusing on science.

Added to this they have the ability to work well in a team and especially with friends, they partner well with mentors, thrive on flexibility and space to explore, value guidance and expect respect. They have a desire to produce something worthwhile and seek to make a difference but are impatient in achieving their goals. Next month the generation of 2016 are going to ‘feel’ what’s wrong with NOW? Similarly “but we have always done it that way” is unlikely to appeal to our new recruits!

In this respect laboratories who fail to understand how to attract and retain Generation Y will see a dramatic negative impact as the baby boomers retire and this new generation enters the workplace – and decides to work in some other career or for another market employer. Thus they use their culture and values to ‘vote’ for employers by accepting employment and not the other way round that we have come to expect as part of our upbringing.

You may say that flexibility and working with like minded people who offer support and guidance while letting you explore the boundaries of your chosen profession is exactly what those currently within the laboratory workforce demographic want as well. So how is it different?

As baby boomers we entered the workforce with the tradition and work ethic of our parents. We were the first generation to have accepted as the norm the concept of both parents working outside the home. You worked for the boss within a well defined set of rules. We did not (and possibly still don’t) see ourselves as demanding flexibility in the workplace. We would like it but its okay if it is not available – we can wait until a position becomes available that suits our needs. We will ask for a change but if not forthcoming we are not likely to move to another city or another laboratory particularly in the current climate. While some have had a career change generally these individuals are few and far between. Those of us left have fond memories of the old days and the old faces.

Other than the rather cynical career path to medicine, ask yourself what is the current career path in medical laboratory science. From the outside looking in (and lets face it from the inside) it would appear to be a role in management, whether that be at Section Head/Team Leader level or at an operational level. What happened to the passion for science that we started off with? Where did it go to be replaced by budget lines and cost centres?

Who are they and what do they want from their careers? Aside from falling into an age demographic they are likely to also be the following:

- Lifestyle centered
- Self confident
- Optimistic
- Comfortably self reliant
- Techno savvy and connected 24/7

Added to this they have the ability to work well in a team and especially with friends, they partner well with mentors, thrive on flexibility and space to explore, value guidance and expect respect. They have a desire to produce something worthwhile and seek to make a difference but are impatient in achieving their goals. Next month the generation of 2016 are going to ‘feel’ what’s wrong with NOW? Similarly “but we have always done it that way” is unlikely to appeal to our new recruits!

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Now—as managers—perhaps it is time that we started to think about creating an environment that meets our needs as well as those of our future workforce. It has been said "organisations that can’t—or won’t—customise training, career paths, incentives and work responsibilities need a wake up call". Are we at this point in training our medical laboratory scientists of the future?

Those of you who read the last newsletter from the NZILMS and managed to read through to the back page will perhaps have noticed my previous foray into highlighting the problems with recruitment and retention of staff. The "gap year" programme initiated at Canterbury Health Laboratories in 2008 is seeking to address at least some of the issues highlighted above. Our four fixed term positions are designed to introduce suitable students—those with an aptitude and passion for science—to the world of medical laboratory science while measuring 24 hour urines, preparing cytology slides, looking for Trichomonas vaginalis, separating samples and loading an analyser. They also have to clean the benches, take out the rubbish and file the slides just like any other laboratory worker! In other words—they know what they are getting themselves into prior to a laboratory career and they are aware of the expectations of the various roles within the laboratory. And, after 6 months, all four are still really keen to complete a BMLSc!

The other aspect of course is promoting workplace loyalty. CHLabs is very aware of the need to attract suitable students for placement and subsequent vacancies, so the 12 months as a laboratory assistant also serves as a 12 month job interview in essence. Although this scheme will take 5 years to see some result it is hoped that at the very least we are seen as being proactive in the industry. It is just too easy to sit back and watch the workforce age increase and moan about the youth of today!

The career pathway is taking a little longer to get off the ground. My vision is one where a staff member with exceptional scientific skills is recognised outside of the traditional management career path. We have to move our focus away from the best scientist being promoted to management and allow that individual to progress within a pay scale based on scientific output. The ability to perform translational research has largely been lost due to our current career structure and this is, I am sure, causing some to shy away from work in medical laboratories. That and the fact that the typical Generation Y is not going to hang around waiting for the next 15 years for management to retire which undoubtedly they will and in large numbers!

The time is right and the time is NOW. The challenge for all in the industry is to get moving on attracting and then retaining our bright young things within a modified career structure that rewards good science alongside management roles.

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Observed changes in the prevalence of uropathogens in Benin City, Nigeria

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Abstract
Objective: Against the background of reports of changes in the prevalence of uropathogens, this study aimed to determine the prevalence of asymptomatic bacteriuria among out-patients of a tertiary hospital, the most prevalent uropathogen, and the distribution of uropathogens among both genders.

Methods: Clean-catch midstream urine were collected from 1,238 out-patients consisting of 1033 non-diabetic patients (451 males and 582 females) and 205 diabetic patients (93 male and 112 females). The diabetic patients consisted of 66 type 1 and 139 type 2 diabetes. Significant bacterial isolates were identified in the urine samples using standard techniques.

Results: There was a significant difference (p < 0.001) in the prevalence of asymptomatic bacteriuria between non-diabetic and diabetic patients (24.5% vs 59.0%). Type of diabetes did not affect the prevalence of asymptomatic bacteriuria (type 1: 53.03%; type 2: 61.87%; p > 0.05). Female non-diabetic patients had significantly higher prevalence of asymptomatic bacteriuria (p < 0.001). Staphylococcus aureus was the most common uropathogen (26.03%) as well as in both genders of diabetic and non-diabetic patients.

Conclusions: An overall prevalence of 30.29% of asymptomatic bacteriuria was found and Staphylococcus aureus was the predominant uropathogen in both genders of out-patients.

Keywords: Asymptomatic bacteriuria, prevalence, uropathogens, diabetes, Staphylococcus aureus


Introduction
Urinary tract infections (UTI) are among the most common conditions causing individuals to seek medical care (1). They are also among the most common bacterial infections in humans, both in the community and hospital settings, occur in all age groups, in both genders, and usually require urgent treatment (2). Urine is the most received and processed specimen in a clinical microbiology laboratory (3) and Escherichia coli has been reported as the most prevalent aetiological agent (1,2,4,5). However, two separate studies – one in Ibadan, Nigeria (6) and the other in Kampala, Uganda (7), in 1969, reported Staphylococcus aureus as the predominant isolate causing UTI among pregnant women. In 2003, Pseudomonas aeruginosa was reported as the predominant isolate causing asymptomatic UTI among residents of Zaria, Nigeria (8) and in 2006, Staphylococcus aureus was the most prevalent isolate causing asymptomatic UTI among pregnant women in Ibadan, Nigeria (9). Against this background, this study aimed to determine the prevalence of asymptomatic bacteriuria (AB) among out-patients attending various clinics in a tertiary hospital in Benin City, Nigeria, to determine the most prevalent uropathogen, and the distribution of uropathogens among both genders.

Materials and methods
Study population
The study was carried out at the University of Benin Teaching Hospital, Benin City, Nigeria for a period of six months. A total of 1,238 patients were studied. They comprised 1033 non-diabetic (non-DM) patients (451 males and 582 females) and 205 diabetes mellitus (DM) patients (93 male and 112 females). The DM patients consisted of 66 type 1 DM (30 males and 36 females) and 139 type 2 DM (63 males and 76 females) patients. The study subjects were out-patients attending various clinics. Exclusion criteria included signs and symptoms of UTI, antibiotic usage within one week and large fluid intake prior (less than one hour) before clinic attendance. Verbal informed consent was obtained from all patients prior to specimen collection. Approval for the study was given by the Ethical Committee of the University of Benin Teaching Hospital.

Specimen collection and processing
Clean-catch midstream urine was collected from each patient into a sterile screw-capped universal container, containing a few crystals of boric acid as preservative. The specimens were mixed, labeled and transported to the laboratory for processing.

A loop-full (0.001mL) of well mixed un-centrifuged urine was streaked onto the surface of blood agar and cystine lactose electrolyte deficient (CLED) medium (M6: Plasmatic Laboratories, United Kingdom). The plates were incubated aerobically at 37°C for 24 hours and counts were expressed in colony forming units (CFU) per millilitre (mL). A count of ≥ 105 CFU/mL was considered significant to indicate asymptomatic bacteriuria.

Ten mL of each well-mixed urine sample was centrifuged at 2000 g for 5 minutes. The supernatant was discarded and a drop of the deposit was examined microscopically at high magnification for pus cells, red blood cells, epithelial cells, casts, crystals yeast-like cells and Trichomonas vaginalis. Pus cells ≥ 5 per high power field were considered significant to indicate infection. The isolates were identified by standard microbiological methods (10).

Statistical analysis was by the Chi (X2) square test . A p value of <0.05 was deemed statistically significant.

Results
There was a significant difference in the prevalence of AB between non-DM and DM patients (24.59% vs 59.02% respectively; p<0.001). The prevalence of AB did not differ significantly between type 1 DM and type 2 DM patients (Table 1). Females showed a higher prevalence of AB than males, but only significant in the non-DM patients (Table 1).
A total of 484 microbial isolates were recovered and Staphylococcus aureus was the most predominant isolate irrespective of patient type (Table 2). Table 3 shows the distribution of uropathogens among gender. With the exception of females from non-DM and type 1 DM patients, Staphylococcus aureus was the predominant isolate in both genders. In females from non-DM patients, Candida albicans was the most prevalent isolate (29.65%) while in females with type 1 DM, both Candida albicans and Staphylococcus aureus predominated with a frequency of 30% each.

### Table 1. Distribution of asymptomatic bacteriuria among gender

<table>
<thead>
<tr>
<th>Subjects</th>
<th>Male</th>
<th>Female</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n</td>
<td>n tested</td>
<td>n infected (%)</td>
</tr>
<tr>
<td>Non-DMt</td>
<td>451</td>
<td>75 (16.63)</td>
<td>582</td>
</tr>
<tr>
<td>Type 1 DM</td>
<td>30</td>
<td>12 (40.00)</td>
<td>36</td>
</tr>
<tr>
<td>Type 2 DM</td>
<td>63</td>
<td>35 (55.56)</td>
<td>76</td>
</tr>
<tr>
<td>Total</td>
<td>544</td>
<td>122 (22.43)</td>
<td>694</td>
</tr>
</tbody>
</table>
| *Non-DM vs DM : p < 0.001. # Male vs Female : p < 0.001

### Table 2. Prevalence of uropathogens.

<table>
<thead>
<tr>
<th>Organism</th>
<th>Non-DM patients (%)</th>
<th>DM patients</th>
<th>Type 1 (%)</th>
<th>Type 2 (%)</th>
<th>Total (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Escherichia coli</em></td>
<td>65 (21.81)</td>
<td>9 (29.00)</td>
<td>29 (20.57)</td>
<td>103 (21.28)</td>
<td></td>
</tr>
<tr>
<td><em>Klebsiella species</em></td>
<td>16 (5.37)</td>
<td>4 (8.89)</td>
<td>15 (10.64)</td>
<td>35 (7.32)</td>
<td></td>
</tr>
<tr>
<td><em>Proteus species</em></td>
<td>31 (10.40)</td>
<td>1 (2.22)</td>
<td>11 (7.80)</td>
<td>43 (8.88)</td>
<td></td>
</tr>
<tr>
<td><em>Pseudomonas aeruginosa</em></td>
<td>16 (5.37)</td>
<td>1 (2.22)</td>
<td>8 (5.67)</td>
<td>25 (5.17)</td>
<td></td>
</tr>
<tr>
<td><em>Staphylococcus aureus</em></td>
<td>76 (25.50)</td>
<td>13 (28.89)</td>
<td>37 (26.24)</td>
<td>125 (26.03)</td>
<td></td>
</tr>
<tr>
<td><em>Coagulase negative Staphylococci</em></td>
<td>22 (7.38)</td>
<td>2 (4.44)</td>
<td>10 (7.09)</td>
<td>34 (7.02)</td>
<td></td>
</tr>
<tr>
<td><em>Enterococcus faecalis</em></td>
<td>1 (0.34)</td>
<td>5 (11.11)</td>
<td>9 (6.38)</td>
<td>15 (3.10)</td>
<td></td>
</tr>
<tr>
<td><em>Candida albicans</em></td>
<td>71 (23.83)</td>
<td>10 (22.22)</td>
<td>22 (15.60)</td>
<td>103 (21.28)</td>
<td></td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td>298 (61.57)</td>
<td>45 (9.30)</td>
<td>141 (29.13)</td>
<td>484 (100)</td>
<td></td>
</tr>
</tbody>
</table>

### Table 3. Distribution of uropathogens among gender.

<table>
<thead>
<tr>
<th>Organism</th>
<th>Non DM</th>
<th>DM patients</th>
<th>Type 1</th>
<th>Type 2</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Escherichia coli</em></td>
<td>21 (21.21)</td>
<td>3 (20.00)</td>
<td>6 (20.00)</td>
<td>10 (18.87)</td>
</tr>
<tr>
<td><em>Klebsiella species</em></td>
<td>6 (6.06)</td>
<td>2 (13.33)</td>
<td>2 (6.67)</td>
<td>6 (11.32)</td>
</tr>
<tr>
<td><em>Proteus species</em></td>
<td>14 (14.14)</td>
<td>1 (6.67)</td>
<td>0 (0.00)</td>
<td>6 (11.32)</td>
</tr>
<tr>
<td><em>Pseudomonas aeruginosa</em></td>
<td>10 (10.10)</td>
<td>1 (6.67)</td>
<td>0 (0.00)</td>
<td>5 (9.43)</td>
</tr>
<tr>
<td><em>Staphylococcus aureus</em></td>
<td>31 (31.31)</td>
<td>4 (26.67)</td>
<td>9 (30.90)</td>
<td>11 (20.75)</td>
</tr>
<tr>
<td><em>Coagulase negative Staphylococci</em></td>
<td>4 (4.04)</td>
<td>1 (6.67)</td>
<td>1 (3.33)</td>
<td>4 (7.55)</td>
</tr>
<tr>
<td><em>Enterococcus faecalis</em></td>
<td>1 (1.01)</td>
<td>2 (13.33)</td>
<td>3 (10.00)</td>
<td>4 (7.55)</td>
</tr>
<tr>
<td><em>Candida albicans</em></td>
<td>12 (12.12)</td>
<td>2 (13.33)</td>
<td>3 (10.00)</td>
<td>7 (13.21)</td>
</tr>
</tbody>
</table>

### Discussion

Several reports exist, indicating changes in the prevalence of uropathogens (6-9). Against this background, this study focused on determining the prevalence of AB among out-patients (diabetic and non-diabetic) of a tertiary hospital as well as to determine the most prevalent uropathogen and the distribution of uropathogens among genders of the study population.

Our study showed a prevalence of AB of 30.29%. Non-DM patients had a significantly lower prevalence than DM patients and this agrees with earlier reports (4,11). High glucose concentration in urine which may create a culture medium for pathogenic microorganism and immunologic impairment resulting in lower host defense system, have been reported to increase the risk of AB in diabetics (4).
DM patients on insulin therapy have been reported to have a higher prevalence of AB (4,11), severity of diabetes, which may be indicated by insulin use (4), is a possible reason for this difference. However, in this study, type 2 DM patients had a higher prevalence of AB than type 1 DM patients, though the difference failed to reach statistical significance.

The finding that females had higher prevalence of AB than males agrees with earlier studies (1,12). However, in this study, statistical significance was observed among non-DM patients only ($p < 0.001$). Close proximity of the female urethral meatus to the anus, shorter urethra, and sexual intercourse have all been reported as factors that influence the higher prevalence in females (1,2).

The aetiological agents associated with AB are similar in both DM and non-DM patients. A total of 484 isolates were recovered from 375 specimens with AB, indicating mixed infections in some patients. Staphylococcus aureus was generally, the most common isolate and in both DM and non-DM patients. Also, Staphylococcus aureus was the most prevalent isolate in both genders, with the exception of female non-DM patients, where it was second in prevalence to Candida albicans. Staphylococcus aureus is a normal flora of female perineum and vulva (13) and during sexual intercourse can easily be carried into the urethra by a massaging process. Also, staphylococci are part of vagina flora (13) and manipulations that alter vaginal flora, such as insertion of a contraceptive device – a known risk factor for UTI (14), can result in opportunistic UTI infection with this organism.

The reason for the high prevalence of Staphylococcus aureus in males is not clear, though lack of circumcision, receptive anal intercourse and HIV infection are recognized risk factors for UTI in males (2).

We also noted that these observed changes in the prevalence of uropathogens are mostly found in Africa as Escherichia coli remains the most common aetiological agent in North America (15). This may indicate that these changes occur in some geographical locations. The changes may also be transient as they were first reported in 1969 (6,7) and later in 2003 and 2006 (8,9). However, these will require further investigations to verify.

The observed changes in this study have serious implications as most clinicians treat patients without recourse to laboratory guidance (2). Such treatments are usually based on known aetiological agents and susceptibilities. This observed change in the prevalence of uropathogens may lead to change in antimicrobial susceptibility and ineffective treatment. Therefore, clinicians should rely on laboratory guidance before therapy as this will overcome the problem of mistreatment and reduce the emergence of resistant uropathogens.

In conclusion, our study revealed a prevalence of 30.29% of AB among out-patients of University of Benin Teaching Hospital, Benin City. The prevalence was higher in DM patients and Staphylococcus aureus was the most prevalent uropathogen in both genders of DM and non-DM patients. Further studies are needed to ascertain if this change in the prevalence of uropathogens are transient and restricted to certain geographical locations.

Acknowledgement
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Pulmonary mucormycosis diagnosed by brushing cytology. A case report

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Abstract
Pulmonary mucormycosis, an uncommon fungal infection occurs among immunocompromised individuals. It is clinically rapidly progressive and often fatal, early diagnosis is critical to patient survival. Bronchoscopy with bronchial brushing is extremely useful for diagnosing pulmonary mucormycosis in immunocompromised hosts as these organisms are morphologically distinct and are infrequently isolated from clinical samples.

We report a case of pulmonary mucormycosis diagnosed on bronchial brushing cytology in a 47 year old male patient presented with pneumonia, acute renal failure and type II diabetes. The Papanicolaou stained filter preparation from the specimen showed broad, ribbon like asapect fungal hyphae with right angled branching. The characteristic cytologic features permitted a diagnosis of mucor infection. This case demonstrates that brushing cytology is a useful technique in the rapid diagnosis of pulmonary mucormycosis. Familiarity with the cytologic appearances of these fungi assist in the correct diagnosis of this disease. This case is of interest also as it showed associated needle shaped crystals in rosette formations and bilateral renal mucormycosis.

Key words: mucormycosis, zygomycosis, phycomycosis, Rhizopus, brushing cytology


Introduction
Pulmonary mucormycosis is an aggressive fungal infection, usually associated with vascular invasion and infarction and can be fatal if not treated quickly (1). Early diagnosis is essential to successful treatment. We report a case that demonstrates the usefulness of bronchial brushing cytology in the diagnosis of this infection.

Case report
A 49 year old male presented with the clinical features of a severe left upper lobe (LUL) community acquired pneumonia, acute renal failure, diabetic ketoacidosis with a pH on admission of 6.99 and type II diabetes. He was treated with intravenous fluids, intravenous antibiotics, cefuroxime and erythromycin. He improved on antibiotics and became afebrile, and was discharged after seven days.

Five days following discharge, he presented with abdominal pain, left sided pleuritic chest pain, fever, shortness of breath and cough. His sputum was mucopurulent and blood stained with a heavy growth of coagulase negative Staphylococcus, no acid fast bacilli were seen. A blood culture gave no growth after 48 hours incubation. His urine showed only mixed bowel flora on culture. Intravenous cefuroxime and metronidazole were commenced with a provisional diagnosis of an unresolved pneumonia.

His symptoms did not improve after ten days and he was referred for chest x-ray and computed tomography (CT) revealing a large cavitating lesion in the LUL and bilateral lower lobe atelectasis/collapse. Abdominal ultrasound showed echogenic bilaterally enlarged kidneys with mild hydronephrosis.

Seventeen days after admission fungal growth was noted on the urinary and sputum specimens and fluconazole commenced. Bronchoscopy was performed that day and revealed a left upper lobe completely replaced by necrotic tissue. Bronchial washing, brushing and biopsy of left lower lobe were performed via the bronchoscope. Bronchial washing and brushing cytology revealed fungal hyphae consistent with mucormycosis. The biopsy also showed fungal hyphae.

A diagnosis was made of pulmonary and bilateral renal mucormycosis. The patient was treated with amphotericin B intravenously. After full lung function testing, the patient was considered not fit for curative cardiothoracic surgery due to the unacceptably high perioperative risk. Medication was discontinued and the patient was discharged for palliative care in home environment. He died twelve days later.

Material and methods
Bronchoscopy derived bronchial washing and brushing specimens were collected in cytology containers with 30% ethyl alcohol in physiologic saline. The filter preparations were made from both specimens on size 5 μm Sartorius membrane filter (AG-cellulose acetate, Germany) using the cytosieve method and stained by Papanicolaou method.

Results
Cytologic findings
Papanicolaou stained filter preparations showed numerous acute inflammatory cells, reactive bronchial cells and broad, ribbonlike asapect fungal hyphae with right angled branching at irregular intervals consistent with Rhizopus mucor infection (Figures 1A and 1B). In addition, a few rosettes of needle shaped crystals were identified (Figure 2).

Histologic findings
Histologic examination of the bronchial biopsy taken during bronchoscopy confirmed the diagnosis of mucor infection. H & E stained sections of the biopsy showed an aggregates of broad, asapect hyphae with right angled, irregular branching (Figure 3) and a few rosettes of needle shaped crystals (Figure 4).

Discussion
Mucormycosis, also known as zygomycosis or phycomycosis, is an infection caused by one of a variety of fungi belonging to the class phomycetes, order mucorales. The common genera causing this infection are Rhizopus, Mucor and Absidia (2,3,6,7). Although these fungi are usually saprophytic and ubiquitous in nature, being found in soil and mouldy bread, they can cause serious and rapidly fatal infection in man as opportunistic disease agents (4).

Infection typically occurs in individuals who are immunosuppressed (5) and have underlying systemic disease such as uncontrolled diabetes (especially diabetic ketoacidosis), haematologic malignancies, renal failure and malnutrition (1-8). The patient presented in this case clearly had predisposing clinical conditions, uncontrolled diabetes with severe diabetic ketoacidosis and acute renal failure.
In diabetes mellitus, acidosis plays an important role in the development of fungal infection. With the fall in blood pH, there is increased release of iron from transferrin that enhances hyphal growth (2). In addition, diabetes causes neutrophil dysfunction, a key defense against fungi.

The organism is ubiquitous in the environment and may be acquired several ways. Intravenous drug users are at risk most likely due to direct inoculation by needles contaminated with the organisms (6). In organ transplant recipients immunosuppression is a major predisposing factor either by activation of latent infection or by greatly reducing lethal dose of the organism (2). This infection may occur in immunocompetent patients due to traumatic implantation of soil contaminants (7).

Mucormycosis most commonly involves the rhinocerebral, pulmonary, gastrointestinal and cutaneous tissues (1,3,6,7). A generalized or disseminated infection may also be seen and renal infection is usually seen in this context. Isolated pulmonary mucormycosis without evidence of dissemination is quite uncommon (2).

The symptoms of pulmonary mucormycosis are fever, cough and shortness of breath, and initially signs of consolidation, which mimics bronchitis and pneumonia. As the organism invades blood vessels, necrosis of the parenchyma leads to cavitation, seen on x-ray. Patients are usually treated with broad spectrum antibiotics, which fail to slow the disease progression – demonstrated by this patient. Without systemic antifungal therapy death occurs in two to three weeks (2). The best chance of survival occurs with early diagnosis, and bronchial brushings are the key investigation to allow this.

Bronchial brushing is an established tool for diagnosis of various pulmonary neoplastic and non neoplastic diseases, and is particularly sensitive in diagnosing peripheral lesions. Sputum culture in pulmonary mucormycosis can fail to give a positive result in over 50% (2) and takes considerable time before a result is available. Transthoracic lung biopsy, thoracentesis, percutaneous needle biopsy and bronchoalveolar lavage are less successful than bronchial brushing in obtaining diagnostic material (2) and are invasive diagnostic procedures unsuitable for in severely ill patients with possible fungal vascular invasion and infarction. By using multiple brushings the diagnostic sensitivity is increased and the need for, but also invasive procedures, is decreased. In addition, bronchial brushing cytology gives a rapid diagnosis facilitating early treatment (7).

The cytologic diagnosis of mucormycosis is based on the recognition of the characteristic mucorales fungal hyphae. Regardless of the species, the hyphae fragments are quite similar and treatment is the same. The hyphae are quite variable in size with average 15-20μm in diameter and aseptate, unlike Aspergillus, branching at irregular intervals and at right angles as opposed to acute angles as in Aspergillus. The tendency of the hyphae to fold and wrinkle gives them a ribbon like appearance which is very helpful in identification. In the case presented here, the diagnosis was made solely by cytomorphology and supported by biopsy findings.

As well as fungal hyphae a few rosettes of needle shaped birefringent (polarizable) crystals were noted. Various types of crystals such a calcium oxalate, monosodium urate and calcium salt of fatty acids can be associated with the fungal infection (4,9). These crystals were not analysed, but morphologically are probably calcium oxalate, known to be produced occasionally as a metabolite byproduct of mucor infections, especially in high oxalate environments such as in renal failure. After reviewing the literature, we believe that this report is possibly the third case of pulmonary mucormycosis diagnosed on brushing cytology in the English literature and first case with associated crystals on brushing cytology.

**Conclusion**

This case demonstrates the ability to diagnose pulmonary mucormycosis by bronchial brushing cytology which is less invasive than biopsy and fine needle aspiration biopsy and allows for more rapid identification of fungal organisms than histologic examination or and culture.
Figure 4. Histology section of bronchial biopsy showing a rosette formation of needle shaped crystals (Hematoxylin-eosin stain X 400)

NZIMLS Journal Prize

Council of the NZIMLS has approved an annual Journal prize for the best case study accepted and published in the Journal during the calendar year. The prize is worth $200.

Case studies bring together laboratory results with the patient’s medical condition and are very educational. Many such studies are presented at the Annual Scientific Meeting, SIG meetings, and the North and South Island Seminars, yet are rarely submitted to the Journal for wider dissemination to the profession. Consider submitting your case study presentation to the Journal. If accepted, you are in consideration for the NZIMLS Journal Prize and will also earn you additional CPD points. Please contact the Editor or any Editorial Board Member for advice and help. Contact details are on the NZIMLS web site (www.nzimls.org.nz) as are instructions to authors.

No formal application is necessary but you must be a financial member of the NZIMLS during the calendar year to be eligible. All case studies accepted and published during the calendar year (April, August and November issues) will be considered. The Editor, Deputy Editor and President of the NZIMLS will judge the eligible articles in December each calendar year. Their decision will be final and no correspondence will be entered into.

Winner of the 2007 NZIMLS Journal Prize was Rossi Holloway, formally from PathLab Bay of Plenty, now LabPlus Auckland for her article “Sideroblastic anaemia secondary to chronic alcoholism: a case study and review”, N Z J Med Lab Sci 2007; 61 (3): 69-70.

Acknowledgment

The authors acknowledge Louise Goosens for her excellent photographic assistance and Ian Tompson for graphical assistance.

References


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The competitive immunoassay on either the Roche Modular E170 and vitamin D and the first 34 amino acids of the receptors on target tissues and at the requirement of high be is does not be.

Methods: K3-EDTA plasma samples from 24 patients were collected and analyzed in duplicate on both Roche Modular E170 and Advia Centaur analyzers.

Results: The paired duplicate data from the Roche Modular E170 and the Advia Centaur gave the internal reliability (CV) of 7% and 3% respectively. The correlation coefficient (r=0.995) showed good association between the two analyzers, however, the Bland and Altman difference graph demonstrated a significant increase in mean difference in higher iPTH concentrations with the Advia Centaur compared to the Roche Modular E170 ( t-test: 3.77; p<0.05)

Conclusions: The iPTH assay on the Advia Centaur showed better internal reliability compared to the Roche Modular E170. Despite the requirement of high plasma volume, the Advia Centaur can be the analyser of choice to perform iPTH assay.

Key words: intact parathyroid hormone, non-competitive immunoassay, electrochemiluminescence, chemiluminometric, Advia Centaur, Roche Modular E170

Introduction
Parathyroid hormone (PTH) is synthesized by four parathyroid glands which are located close to or on the posterior surface of the thyroid gland. However, additional parathyroid glands may be located elsewhere such as in the neck or within the thymus in the superior mediastinum (1). PTH is a single-chain polypeptide of 84 amino acids with molecular weight of 9500 Daltons. Intact PTH (iPTH) is the form stored in the glands and the principal form secreted into the blood stream. The plasma half-life (1/2) of iPTH is less than 10 minutes on which iPTH is cleaved in the region of amino acids 33-37 and elsewhere in peripheral tissues, principally the liver and kidney, to an amino acid (N-terminal) fragment of at least 34 amino acids, carboxy-terminal (C-terminal), and mid peptide fragments (1,2). The biological activity of iPTH resides in the first 34 amino acids of the N-terminal, therefore, both iPTH and N-terminal fragments posses full biological activity.

Together with vitamin D and calcitonin, iPTH binds to type 1 PTH receptors on target tissues and regulates the ionized or free calcium (CaI) (1). The level of iPTH also affects the concentration of plasma ionized phosphate (Pi), even though the plasma Pi concentration does not control iPTH secretion directly. Elevated plasma iPTH which causes hypercalcaemia is found in primary hyperparathyroidism and secondary hyperparathyroidism. On the other hand, low plasma iPTH, which causes hypocalcaemia, is found in primary hypoparathyroidism and secondary hypoparathyroidism.

The plasma concentration of iPTH can be measured by non-competitive immunoassay on either the Roche Modular E170 or Advia Centaur analyzers. On the Roche Modular E170 analyzer, electrochemiluminescent technology is used to measure iPTH level, on which a biotinylated monoclonal antibody and a monoclonal antibody labelled with ruthenium are applied to bind the N-terminal fragment (1-37) and C-terminal fragment (38-84) respectively. After the addition of streptavidin-coated microparticles, the complex becomes bound to the solid phase via interaction of biotin and streptavidin. The reaction mixture is aspirated into the measuring cell where the microparticles are magnetically captured onto the surface of the electrode. Unbound substances are then washed off, followed by the application of a voltage to the electrode to induce chemiluminescent emission which is measured by a photomultiplier.

The Advia Centaur analyzer uses a similar principle as the Roche Modular E170 analyzer. However, instead of using electrochemiluminescent detection technology, the Advia Centaur analyzer uses chemiluminometric detection technology on which dispensing of both acid reagent and base reagent induces the chemiluminescent emission. The Advia Centaur analyzer uses a polyclonal goat anti-human PTH antibody labelled with an acridum ester and a biotinylated polyclonal goat anti-human PTH antibody to bind the N-terminal (1-34) and the C-terminal (39-84) respectively.

The objective of this study was to compare the reliability of iPTH assays between the Roche Modular E170 and the Advia Centaur analyzers, and to determine whether both analyzers can be used interchangeably.

Methods
K3-EDTA plasma samples were collected over the period of four weeks from 24 different patients reported by Canterbury Health Laboratory (CHL) Christchurch to have iPTH levels between 1.7 and 135.0 pmol/L. The whole blood samples were centrifuged at 3700 rpm for seven minutes, plasma separated, and stored at -20°C in plastic Kahn tubes. On the day of analysis, the samples were thawed and mixed using a vortex mixer then centrifuged for seven minutes prior to analysis.

Calibrations and quality controls were analyzed for both analyzers according to the manufacturers’ instructions. The Roche Modular E170 was calibrated using Elecsys PTH CalSet (0.005 pmol/L and 477 pmol/L), while the Advia Centaur was calibrated using intact PTH (iPTH) calibrator (2.68 pmol/L and 88.30 pmol/L). The quality control materials used were PreciControl Bone 1, 2, and 3 for the Roche Modular E170, and Ligand plus 1, 2, and 3 for the Advia Centaur. The patients K3-EDTA plasma samples were assayed in duplicate on both analysers. The coefficient of variance (CV) was calculated from the paired duplicate data. The correlation coefficient and regression analysis was used to determine the association of the results measured by two different analysers. The difference or bias between two analysers was evaluated by plotting the Bland and Altman difference graph and by performing 95% confidence interval (p<0.05) paired t-test using n-1 degree of freedom.

Results
The paired duplicate data from the Roche Modular E170 and the Advia Centaur analyzers gave the internal reliability (CV) of 7% and 3% respectively. The correlation between the two analysers
is shown in Figure 1, a significant difference was demonstrated between the two analyzers (t=3.77, p<0.05).

![Figure 1. Correlation of iPTH analysis between the Roche Modular and the Advia Centaur analyzers.](image1)

Figure 1. Correlation of iPTH analysis between the Roche Modular and the Advia Centaur analyzers.

From the slope value of 1.6047, it can be concluded that the Advia Centaur gave higher results compared to the Roche Modular with the average difference between two means of 12.32 pmol/L. This difference increases as the iPTH concentration increases (Figure 2).

![Figure 2. Bland and Altman difference graph demonstrating bias between the Roche Modular and Advia Centaur analyzers.](image2)

Figure 2. Bland and Altman difference graph demonstrating bias between the Roche Modular and Advia Centaur analyzers.

Discussion

Even though the correlation analysis (r² = 0.995) showed that the results from both analyzers were associated, it is clear from the regression equation in Figure 1 that iPTH assays had higher values on the Advia Centaur analyzer when compared with the Roche Modular E170 analyzer. As correlation analysis measures association rather than agreement (3), this demonstrates that both analyzers are designed to measure the same substance but not necessarily be in numerical agreement. Therefore, paired t-test was undertaken to show the agreement between two data sets. The paired t-test (t = 3.77, p<0.05)) indicated that the average difference between two methods was significant. Therefore, both analyzers should not be used interchangeably. As expected, the paired t-test between iPTH levels obtained by Roche Modular E170 and CHL Christchurch (who use this method) showed no significant difference between two results (t=1.89, p<0.05). Furthermore, the paired t-test between iPTH levels obtained by Advia Centaur and CHL Christchurch showed that there was a significant difference between two results (t=3.64, p<0.05) confirming the results in this study.

The significant difference in results obtained using both analyzers may be explained by the fact that both analyzers use different calibrator materials thereby producing differences in the expected values of iPTH. This is reflected in the reference ranges of the two analyzers where the Roche Modular E170 analyzer reference range was 1.6-6.9 pmol/L while on the Advia Centaur, the reference range was 1.18-8.43 pmol/L.

The difference in these results between both analyzers may be caused by two different features found in both assays. First, unlike the Roche Modular E170 that uses electrochemiluminescent technology, the Advia Centaur uses direct chemiluminescent technology and second, the Roche Modular E170 uses monoclonal antibodies while the Advia Centaur uses polyclonal antibodies.

A previous study on iPTH assays by Santini et al (2004) indicated that the use of more standardized method of calibration and antibodies that recognize only the biologically active PTH molecule may decrease the wide gap between results obtained from a range of different analyzers (4).

This study was limited by a small number of samples as well as low sample volume as the the Advia Centaur required a 200μL sample volume while the Roche Modular E170 required only 50μL of sample. This limited the opportunity to undertake further evaluation such as a sensitivity analysis and a wider range of iPTH values.

In conclusion, iPTH assays on either analyzer took approximately 18 minutes per sample and assay costs were comparable. However, despite a good correlation, the average difference of iPTH assay between the Roche Modular E170 and the Advia Centaur is significantly different from zero. Therefore, they should not be used interchangeably. Comparison of the two analyzers showed that the Advia Centaur had better reliability. Despite a higher plasma volume requirement, the Advia Centaur can be the analyzer of choice for iPTH assay in the clinical biochemistry laboratory.

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References


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Journal article questionnaire
for the Haematology Special Interest Group

Questions:
1. What are the two major applications of molecular genetics in chronic lymphoid malignancies?
2. Mature B cell neoplasms are malignancies of a monoclonal population of B cells- True or False?
3. How is immunoglobulin diversity achieved? Explain.
4. What is the method used as Gold standard for demonstration of a clonal population of B cells?
5. Where is the IGH locus located?
6. What surrogate marker is useful in CLL for assessing IGVH mutation status?
7. Detection of ZAP 70 is an indicator of a poor prognosis- True or False?
8. What is the consequence of t(11;14) translocation in Mantle cell lymphoma?
9. Which test is more useful for detecting t(11;14) in Mantle cell lymphoma?
10. What is the role of BCL2 gene in Follicular lymphoma?
11. Other than BCL2 mutation, what other genes are involved in transformation of follicular lymphoma to Diffuse large B cell lymphoma?
12. What is the most common Non Hodgkins lymphoma?
13. Name the two main subtypes of DLBCL?
14. What is the premalignant stage of Multiple Myeloma called?
15. Most T cell express a TCR molecule composed of chains- True or False?
16. What other conditions show an increase in clonal T cells?
17. What are the two syndromes classified as Cutaneous T cell lymphomas (CTCL)?
18. Significant proportion of Anaplastic large cell lymphoma (ALCL) are characterised by aberrant expression of anaplastic lymphoma kinase (ALK) protein. What is its function?
19. What causes Adult T cell leukaemia / lymphoma (ATLL)?
20. Majority of cases of NK cell malignancies show infection with what virus?

Questions prepared by Shalini Reddy, Haematology Dept. North Shore Hospital. For a copy of the journal article, ph 09 486 8327, extn 2327 or email shalini.reddy@waitetomatahb.govt.nz.

Answers on page 45
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An unusual fluid.
The advantages of cross discipline training

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Abstract
A 70 year old male with a history of Non-Hodgkin’s large B-cell lymphoma was admitted to North Shore Hospital (NSH) with symptoms of confusion, headache, nausea and vomiting. Cerebrospinal Fluid (CSF) was examined in the microbiology laboratory and showed a lymphocytic pleocytosis. An initial diagnosis of probable viral meningonecphalitis was made. A subsequent CSF sample examined by a medical laboratory scientist with cytology experience determined that the sample contained highly atypical lymphoid cells. Cytological examination produced a diagnosis of malignant meningitis consistent with a diffuse large B-cell lymphoma. This article discusses the case and the advantages of cross disciplinary training in the laboratory.

Keywords: lymphoma, meningonecphalitis, cytology

Introduction
A 70 year old male was admitted to NSH with a two week history of intermittent confusion, occipital headache, nausea, vomiting and periods of expressive aphasia. In 2005 he had been diagnosed with Non-Hodgkin’s diffuse large B cell lymphoma involving the pleura and bone marrow. This had been successfully treated with chemotherapy and he had been in complete remission for 2 years. On this current admission he was afebrile. Cardiovascular, respiratory and abdominal examinations were normal. Neurological examination revealed fluctuating confusion and intermittent expressive dysphasia. A computerised tomography (CT) scan of the head was normal.

A CSF sample was obtained and sent to the NSH microbiology laboratory for examination and culture. This showed a white blood cell (WBC) count of 223x106/L (normal range 0-5x106/L) with a differential of 97% lymphocytes and 3% polymorphonuclear neutrophils (PMN), a protein of 0.75g/L (normal range 0.15-0.45g/L), and glucose of 0.7mmol/L (normal range 2.8-4.4mmol/L). Based on these results, and the lack of growth on culture media, samples were sent off for numerous additional tests including polymerase chain reaction (PCR) for TB and HSV. Two days later another CSF sample was obtained. This showed a WBC count of 504x106/L, a protein of 1.09g/L, and glucose of 0.6mmol/L, again with a lymphocytic predominance. In this case the medical laboratory scientist involved in the CSF examination had a background in cytology and recognised the cells in the differential as abnormal and referred the sample to cytology.

Cytological examination produced a diagnosis of malignant meningitis with a diffuse large B-cell non-Hodgkin’s lymphoma, consistent with a relapse of his previously treated malignancy. The patient has subsequently been treated with intrathecal chemotherapy with evidence of response.

Basics of cytology
Although it takes years to become fully proficient in cytology a few basic rules of thumb usable even on Gram stains and differential stains which yield nuclear detail, can indicate if a sample should be forwarded to cytology. (Note these pictures are from samples of the patient’s CSF, the basic criteria can be applied to all cells, but in this case lymphocytes are predominant).

1) Cellularity- Is the sample more cellular than normal (containing more cells), are there unusual cells, or clusters of cells, and are they chaotically arranged? Some samples, such as CSF, are usually almost acellular, although a few WBC or RBC is not uncommon. This sample shows far more cells than normal (Figure 1).

2) Cellular morphology- Are the cells typical of what should be in the sample? Variations in cellular size and shape are indicative of an abnormality; some cells may be highly atypical (Figure 2).

3) Nucleocyteplasmic ratio- Neoplastic cells show an increase in nuclear size relative to the total cell size, compared to normal cells of that type; some cells will show little or no cytoplasm. Note that this is not as helpful with lymphocytes, as they can intrinsically show a variable N/C ratio.

4) Multinucleation- Cells normally only have a single nucleus. Actively replicating cells may show more than one, but multiple cells showing multinucleation is atypical, any multinucleation of lymphocytes in the CSF may be regarded as abnormal (Figure 3, C). NB- lymphomas do not normally exhibit multinucleation, and you should endeavour to be certain the cell is not a histiocyte.

5) Nuclear outline- Typically nuclei are rounded with a smooth regular outline, neoplastic cells often show irregular nuclear outline with a rough nuclear membrane (Figure 4, D and E).

Typical abnormalities include “clefts” (voids between lobes of a nucleus as seen at D), “rat bites” (deep indentations in the nuclei as if a bite has been taken out), and “blebs” (large bulges out from the nuclei).

6) Nucleoli- These represent active cells and appear as darker blue patches in the nucleus in these images, and are not in themselves indicative of neoplasia; however the presence of multiple nucleoli, especially those with irregular outlines, and those showing variation in size and number of nucleoli within nuclei are significant (Figure 4, E).

7) Mitotic figures- These are indicative of rapidly dividing cells. These are unusual in normal cells, and the numbers are indicative of the severity of malignancy. Any mitoses in lymphocytes in the CSF can be regarded as abnormal. (Figure 5, F).

Discussion
This case illustrates the advantage of cross disciplinary training in the laboratory. When the patient’s CSF was initially examined in microbiology, a diagnosis of viral meningitis was suspected. When the second sample was then examined by a medical laboratory scientist with experience in cytology, the lymphocytes in the CSF were recognised as being highly abnormal and inconsistent with the diagnosis of viral meningonecphalitis. The specimen was then formally examined by the cytology department and the correct diagnosis of abnormal cells consistent with a malignant meningitis was made. The patient was then able to receive appropriate therapy. Although the patient appears to have responded to treatment for his relapsed NHL, the diagnosis of malignant meningitis was somewhat delayed and he was subjected to a number of expensive and unnecessary tests in the interim.
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Figure 1. Even at 100x the highly cellular nature of the sample is evident.

Figure 3. Note the cell marked (C) at 400x exhibiting 3 nuclei.

Figure 2. At 400x the variations in both size and shape are more pronounced, note the large difference in size between a "typical" lymphocyte (A) and the neoplastic cells (B).

Figure 5. 400x micrograph.

Figure 4. 400x micrograph.
Lymphoma

Lymphomas are neoplasms (cancers) of lymphatic cell origin. They can arise from B or T cell lineages. Mature B-cell neoplasms are clonal proliferations of cells at various stages of B-cell differentiation and the progenitor cells can range from mature plasma cells to naive B-cells. Each stage of differentiation leads to a specific form of lymphoma. B cell lymphomas are most commonly follicular or diffuse large cell lymphomas, and comprise over 90% of lymphoid neoplasms worldwide (1). Diffuse large B-cell lymphomas constitute 30-40% of adult non-Hodgkin's lymphomas, with the median age at diagnosis being 70-80 years, although the disease can be seen in children. Morphologically they can be divided into centroblastic, immunoblastic, histiocytic rich or anaplastic varieties, depending on the cell line of origin (1), and while aggressive may be successfully treated with multiagent chemotherapy.

Lymphomas may be characterised through immunophenotyping, a process by which the cells are distinguished through the attachment of labelled antibodies raised to cell surface antigens (typically Cluster Designation or CD antigens), to these antigens. This is useful to distinguish between malignant and benign lymphoid proliferations, between B- and T-cell processes, and between sub-categories of B- and T-cell lymphomas. However in this case as it is a recurring lymphoma it would probably not be necessary to repeat it.

Malignant meningitis

This is also known as meningeal carcinomatosis, and can be caused by primary neuroectodermal tumours such as neuroblastoma, tumours of the haematopoietic system such as lymphoma, or solid tumours metastasizing to the leptomeninges (e.g. lung, breast, gastrointestinal tumours, and melanoma (2, 3)). In up to 50% of patients the site of the primary tumour is unknown and the disease may manifest up to 20 years after a primary tumour.

Clinical findings of malignant meningitis may include pain (radicular discomfort, headache, neck or back pain), mental state abnormalities, weakness and occasionally seizures (3, 4). Radiographic studies such and contrast-enhanced MRI may give a clue to the diagnosis. The diagnosis is most often made fortuitously when CSF is obtained to investigate such neurological symptoms (2)

The classic CSF findings include a high opening pressure, a high protein level and low glucose (3) and raised white blood cell count. The microscopy may show tumour cells; these are often bizarre and may occur in clumps. The finding of such malignant cells in the CSF is pathognomonic, however the sensitivity of CSF is 80-95% and it may be necessary to obtain further samples to confirm the diagnosis. It is important that large volumes of CSF are withdrawn (10mls minimum), and specimens should be processed promptly (within 1hr to ensure well preserved cells are available for evaluation to improve the yield. If the patient has a known primary tumour then proteins specific to this primary tumour may be sought.

The prognosis of metastatic meningitis is poor, primarily because the malignant disease is well advanced. The treatment that is principally used is craniospinal irradiation and intrathecal chemotherapy, as the blood brain barrier prevents accumulation in the CSF of therapeutic concentrations of most drugs administered by other routes (2).

Conclusions

Although malignant meningitis is an unusual diagnosis, medical laboratory scientists, especially those in microbiology, routinely deal with fluids that may contain neoplastic cells. They should be trained to recognise abnormal cytological features as described previously, such as unusually high cellularity that would then prompt them to seek a formal cytological examination for the specimen. The ability to determine that a fluid may contain neoplastic cells, and that it should be forwarded for cytological exam, is important in making a rapid and accurate diagnosis.

Acknowledgments

Dr Stephen Allpress, Dr Jan Craik, and Dr Sanjeev Chunilal, North Shore Hospital, Auckland.

References

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Phlebotomy Special Interest Group

In order to apply for an Annual Practising Certificate in 2009, QPs need to undertake a minimum of 3 hours of Professional Development Activities during 2008.

In May we were treated to presentations by peers and professionals covering a wide range of topics at the Phlebotomy/Specimen Services SIG meeting as part of the North Island Seminar in Palmerston North.

Let me tell you about my day and a few of the snippets from my notes. We had a really pleasant flight from Auckland to PH (1 hr, not counted). We piled into a minibus and chattered our way to the Travelodge, checked in and received our programme booklet which included information on all the topics with abstracts and note-taking space. A friendly crowd gathered around the hot coffee and muffins, as we broke the ice. The weather was cool but not icy, but a great way to start (1 hr, not counted). The sessions had 15 minute slots for speakers with a break in the middle for lunch and afternoon tea, and a few minutes between speakers to fluff our pillows.

Session 1 was combined with our NZIMLS colleagues from all disciplines (2hrs, counted). It was nice to be included and learn from topics of common interest and relevance.

- Warfarin: past, present & future by Dr Paul Harper: warfarin has been around for over 50 years now and is the most widely used anticoagulant in clinical practice, but have you heard that there are newer drugs undergoing clinical trials that may replace warfarin with its high risk of bleeding?
- CPD audit & new COPD requirements for MLTs by Jillian Broadbent. This includes QPTs. (That's us.) Do you know that you may be required to provide more evidence for audit by MLSB than just a certificate of attendance? (So taking notes was a good idea!)
- Have you wiped? An environmental search for Chlamydia by Roger Athersuch: Chlamydia trichomonas is the most prevalent STD in NZ. The study tested for molecules of CT in the environment, some were found on the bed remote! (I wonder if any of our equipment could be contaminated?)
- Cord blood banking in NZ: A really interesting talk by Kasey Kime on the significance of cord blood banking, its collection, and medical application. More parents are arranging storage of their babies' cord blood for stem cell treatment in cases of brain injury, cerebral palsy, etc. Did you know that CordBank has recently been granted permission from the MOH to extend the definition of Cord Blood Collectors to include Phlebotomists? (That's us again)
- TRALI & “male only” FFP: by Melissa Nelson & Trent Jones, BMLSc students. Transfusion related acute lung injury is often caused by HLA antibodies and since female donors have a higher chance of having these in their plasma because of pregnancy, NZBS now uses only male fresh frozen plasma. (Now that's a rare thing – something a male is better for!)
- Charging ahead - private specialist referred testing in Wellington: by Keliy Belton. Changes in funding policy impacted on their community laboratory operation, from collections to result reporting. This is likely to affect all of us sooner or later. Phlebotomists and specimen receptionists are going to need increased knowledge and skill to cope with this when it hits your town.

We grabbed a bite to eat for lunch from a yummy smorgasbord of savories, sandwiches, wraps, fruit, and slices, with coffee or tea and used the opportunity to make new friends and catch up with the old. The networking at NZIMLS functions is always a good time to find out how others are doing this or that. (A very worthwhile 1 hour, counted)

Sessions 2 & 3: The room was divided, with half attending the seminar and the other half attending our Special Interest Group. (Fairly significant that we comosed half of this NZIMLS function.) (4hr, counted.) Topics were:

- Lean: is about doing more of what matters by eliminating what doesn't. Keiry Belton applied lean thinking to phlebotomy with examples of practical process improvement. The overall aim is to do the right tests on the right patient, with the right results being reported to the right requestor at the right time. We play quite an important role in that. Our personal aim could be to 'identify and improve one thing in our area every day'.
- Specimen transport: Dave Kendall gave us a look at local and international requirements for sample transportation – things you need to know if you’re sending your sample away to be analysed, like how large the letters need to be on the label (6mm), and that packages in dry ice that must permit the release of gas during transport (so your package doesn’t blow up!).
- Cryoglobulins: Maire Bray gave a presentation on these proteins which are associated with diseases such as myeloma, Hep C, CLL, SLE or RA, whose symptoms may be skin lesions, vascular purpura, Raynaud’s phenomenon, arthralgia and arthritis, and which can precipitate out at different temperatures below 37°C. Gelpacks, sand or water can be used to pre-heat tubes for collection and for transport of samples to the 37°C centrifuge.
- Smart learning: Jane Kendall passed on some smart learning strategies to get organised and be prepared for the exam. Take rests during study, relax – stress is a barrier to learning. Create multi sensory memories like flash cards, mnemonics, mind maps, notes, flowcharts, pictures. Organise – have a year planner, a weekly plan, and a ‘things to do today’ list. Exams – know your exam (topics, length, marks, format, understand the questions) and keep out of panic mode!
- Case study – arterial puncture: Maree Southee presented a case study following an accidental arterial puncture, a scenario that everyone present will wish to avoid. Moral of the story – check all veins before venipuncture, and the basilic vein should always be last choice.
- Workplace health & safety: Clare Lynn shared some of her occupational health work experiences. Some hazards we face daily – cranky colleagues, work stress, fiddly equipment, family flutters, etc. Workplace incidents are best dealt with by identifying patterns and using a “no blame” approach. Each of us has responsibilities to eliminate, isolate, or minimise hazards.
O1. Demonstrations of the clonal nature of lymphoid cells and the detection of pathogenetically important rearrangements.

2. True.

3. Combinatorial diversity and junctional diversity. Combinatorial diversity results from random selection of V-D-J segments into the rearranged gene. Junctional diversity results from deletion/insertion of nucleotides at boundaries between the segments.

4. Southern Hybridisation

5. Chromosome 14q32.

6. ZAP70 (Zeta-associated protein 70).

7. True.

8. Aberrant over expression of CCND1 (cyclin D1) - important for G1-S transition during cell cycle.

9. FISH - based approach is the test of choice.

10. Inhibition of apoptosis, and also acts as a modulator of cell cycle progression.

11. 5' regulatory region of BCL6 and MYC gene.


14. MGUS.

15. False - most T cells are composed of and chains.

16. Age, viral infections or immunodeficiency.

17. Sezary syndrome and Mycosis Fungoides.

18. ALK is a receptor tyrosine kinase that is normally only expressed with the nervous system.

19. Infection of HTLV-1 virus.

20. EBV.
Medical laboratory science news

Massey University

NZIMLS Prize

The NZIMLS Prize for the best third year BMLSc student in 2007 was awarded jointly to Mr Eamon Karalus and Ms Anthea Povall. Both Eamon and Anthea have done extremely well in their studies and the Examinations Committee could not distinguish between them. They are currently enrolled in the fourth year of the course and we look forward to following their careers in Medical Laboratory Science.

MSc Completions

Congratulations to the following who have recently completed their research projects for the MSc in Medical Laboratory Science:

Mr Maqhawe Ndovu on the “Isolation and genetic comparison of Escherichia coli strains from women with urinary tract infections and their pet dogs.”

Ms Jodi Heaven (nee Hopkins) on the “Development of a tandem mass spectrometry procedure for the measurement of itraconazole in human serum.”

Ms Kate Marson on the “Development of a flow cytometry method for the measurement of Zap 70 in chronic lymphocytic leukaemia.”

Maqhawe and Kate join a growing group of graduates who have successfully completed their masterates extramurally while employed full time. Jodi, who is currently living in the United Kingdom, has just one more paper to finish this year.

New staff

Dr Matthew Perrott has recently joined us as a Senior Lecturer in Histopathology. Matthew completed his PhD at Massey in 1999 studying viral agents of pox and has continued to work with infectious and epidemic disease since then. Matthew worked on the foot and mouth disease epidemic of sheep and cattle in the UK (2001). For his postdoctoral research in the USA, he developed a ferret model of chronic wasting disease, an infectious transmissible spongiform encephalopathy of deer and elk (2001-06). More recently, in Australia, he was involved with infectious laryngotracheitis and other viral agents of poultry (2007-08). His particular research and teaching interests are in histopathology, histological techniques, immuno-histochemistry and western blot.

Ms Louise Shaw has taken up the position of BMLSc Technician. Louise has worked for Ag Research, Masterton Hospital Laboratory and, most recently, Gribbles Veterinary Pathology.

Ms Liz Burrows has accepted the position of Microbiology Technician. Liz also comes to us from Gribbles Veterinary Pathology and, like Louise, has extensive experience in diagnostic laboratory work.

We are delighted to have Matthew, Louise and Liz join the “BMLSc team” and look forward to the contributions they will undoubtedly make to the course.

Otago University

The 2007 academic year ended successfully with graduation and the awarding of prizes. For the NZIMLS prizes awarded to the top student in each BMLSc year, the second year prize went to Lisa Stevens, third year prize to Charlotte Hughes and the fourth year prize to Malles Kovoert. Malles was also awarded the Colin Watts Prize for the best overall student and was a nominee through the Division of Health Sciences for the Prince of Wales Prize in Health Sciences. Tania Feary was awarded the James Le Grice prize for the top student in Clinical Biochemistry.

It was a sad start to 2008 however, with the death of Professor Sandy Smith from the Department of Microbiology and Immunology after a long illness. Sandy was a strong advocate for the BMLSc and took an active interest in promoting microbiology and academic standards in the BMLSc course. Dr Heather Brooks will replace Sandy; Heather was a medical laboratory scientist before taking up a position in the university.

The new second year health science programme was introduced this year and has proceeded very well. The new programme has provided more time for our second year students to have a better introduction to medical laboratory science prior to third year professional disciplines.

On the research front Dr Erin Cawston (BMLSc graduate, 1999) graduated with her PhD and will take up a new position at the Mayo Clinic, USA doing research on signalling mechanisms in cancer, at the end of 2008. Dr Jade Hollis-Moffatt (BMLSc graduate, 1998) was awarded the TrustPower Ashburton Rotary Club Young Achiever Award for her research relating to the genetics of diabetes and Dr Alasdair Russell (BMLSc graduate, 1998) is now working for the Cancer Research Unit at Cambridge, UK with the Mammary Stem Cell Biology Research Group.

It is very pleasing to acknowledge the award of Fellowship of the Institute of Biology, UK to Rob Siebers at the Otago School of Medicine and Health Sciences, Wellington. Rob is a member of the Board of Studies for Medical Laboratory Science at the University.

Associate Professor Mike Legge (Director, Medical Laboratory Science Programme) is a member of the Medical Laboratory Science “Think Tank” investigating the possible extended role of medical laboratory scientists and was recently appointed to the Editorial Board of the journal, ‘Theoretical Biology Insights’. He was also appointed as a specialist editor for the Mosby Dictionary of Medicine, Nursing and Health Professions. Finally I would like to take the opportunity to thank all the Diagnostic Pathology laboratories that accepted our Fourth Years for clinical placements.
NZIMLS Council News
June 2008

COUNCIL MEETING

The NZIMLS Council met in Palmerston North on the 8th and 9th May, 2008. Apologies were received from Jillian Broadbent (for Thursday only).

This Newsletter outlines the key issues from the Council meeting and includes other information relevant to our profession.

BITS AND BOBS AND FROM AROUND THE REGIONS

Council has been notified of the retirement of Errol Crutch, Senior Scientist, Coagulation Laboratory, Wellington Hospital. Errol had a long career in Medical Laboratory Science, and we wish him well in his retirement.

We encourage members who have anything of interest for this segment of the newsletter to contact their local Council representative.

PROFESSIONAL AFFAIRS

Professional Development for Medical Laboratory Technicians

Under section 41 of the HPCA Act, the Medical Laboratory Science Board has set a recertification programme for medical laboratory technicians, including phlebotomists. MLTs will be required to undertake a minimum of 8 hours of professional development (PD) in 2008 and their supervisor will have to declare on the technician’s APC application for the 2009/2010 year that this has taken place. The Board plans to fund an independent audit of 10% of practising technicians each year. Those technicians audited will have to complete the MLSB PD form supplied by the Board plus supply evidence (documentation) of their claim.

A number of MLTs are currently members of the NZIMLS CPD Recertification programme. Council has recently sought clarification from the Board on the validity of documented CPD activities in the Institute’s programme for the purpose of verification of a technician’s CPD activity, if audited by the MLSB. In response, the Board has stated that they have not approved recertification programmes other than the Board’s programme for MLTs, but this does not prevent an MLT joining other programmes. However, if a technician’s hours of professional development are audited, they must use the MLSB PD form supplied with
their APC this year and available on the Board’s website. That is, the
Board cannot accept the points tally sheets from the Institute’s
programme. However, the Board has indicated that there is no objection to the MLTs supplying the
NZIMLS CPD form as part of their documentation, but it will be the MLSB form that is audited. Council
encourages MLTs to enroll in the NZIMLS CPD programme as it provides access to opportunities for
professional development, such as the NZIMLS classroom and journal-based questionnaire.

JOURNAL

The journal-based questionnaire continues to be a popular route for obtaining CPD points. For the
April 2008 issue there was a record number of submissions, with 651 in total. Previous issues of
the journal attracted approximately 500 submissions. In most instances, members were able to
successfully submit on-line. However, a few members experienced problems with their submissions
failing to transmit to the Editor. These problems may relate to IT issues, such as on-line sessions
timing out in an employee’s organisation if the employee is on-line for some time. In cases like that it
may be prudent to write the answers in a word document and then cut & paste onto the web site.

A reminder of the annual NZIMLS prize ($200) for the best case study accepted and published in the
Journal. Increasingly, good case studies are presented at SIG meetings and the North and South
Island Seminars. Consider submitting your case study to the Journal and reaching a wider national
and international audience.

CPD PROGRAMME

By now most of you will have been issued with your 2008-2009 Annual Practising Certificates (APCs)
by the Medical Laboratory Science Board (MLSB).
If you haven’t received yours, and the hold-up is due to a lack of CPD points because you have
forgotten to enter them from previous years, send an email to cpd@nzimls.org.nz with the specific
details of the points you would like added.

The CPD Programme booklet has been reprinted and copies have recently been sent out to all
practitioners enrolled in the NZIMLS CPD programme who are financial for 2008. If you haven’t
received your booklet, and you believe you ARE financial, please send an email to cpd@nzimls.org.
nz so we can follow up for you.

The April Journal questionnaire has now closed, but watch out for the questionnaire in the next
journal in August – this is a great way to earn your CPD points!
QMLT / QPT/ QSST EXAMINATIONS

The Examination Committee has reviewed the Common Syllabus and is currently making sure that all syllabi are up to date.

Plans are in progress to produce logbooks for all QMLT examination subjects.

Because of the amount of work involved in the preparation and review of examination documentation, Council will be offering to pay a fee to those members offering to assist with this important task. The schedule of payments is:- $150 for syllabi review, $300 for producing a discipline specific logbook or $150 for the review of an existing log book.

MLS EDUCATION

Moderation of the university BMLSc courses is an on-going process and the NZIMLS is always interested to hear from anyone who would like to be involved in the moderation process. Council is currently seeking moderators for the subjects due to be moderated in 2009, which are Haematology, Biochemistry and Cytology. Members who have an interest in participating in this important function are welcome to contact the Executive Office. $600 is paid for each subject moderated.

SPECIAL INTEREST GROUPS

We are still seeking a Convenor for the Biochemistry SIG as well as a Microbiology SIG Convenor. Please contact the Executive Office if you are interested in taking on this important and rewarding role, or know of someone who could be approached.

Council is looking at the possibility of forming a POCT Special Interest Group. This would depend upon sufficient numbers to form a viable group for a meeting. To gauge numbers could those who are interested in attending a POCT SIG please email Sandy.Woods@cdhb.govt.nz to express interest and Council will discuss further at the next meeting.
FELLOWSHIP

The NZIMLS Fellowship Committee have reviewed the Fellowship Regulations and Council has approved a change to regulation 6.1. The amended rule now states: “the Fellowship Committee shall consist of three members appointed by Council, with the three appointees being financial Fellows of the NZIMLS. The committee will be reappointed annually at the November Council Meeting”. Council is very pleased to have 5 applicants for the current Fellowship examination.

UPCOMING EVENTS

- Immunology SIG Seminar
  Museum Hotel, Wellington, June 21st

- Microbiology SIG Seminar
  Beavan Lecture Theatre, Christchurch Hospital, July 18th - 20th

- NZIMLS Annual Scientific Meeting
  University of Otago, Dunedin, August 25th - 29th

- Histology SIG Seminar,
  Devon Hotel, New Plymouth, November 7th - 8th
Happenings at the PPTC and in the Region
We have run a Haematology Course here at the PPTC while over the past few months there have been several international and regional meetings taking place that are of importance to laboratories in the Pacific Region.

Haematology and Blood Cell Morphology Course
In April a very successful Haematology and Blood Cell Morphology Course was run here at the PPTC with seven students. We had a first with this course with a first ever participant from Timor Leste, Luscendar Fernandes Alves. It was great having her active participation in the course and we learnt a lot about Timor Leste. We are hoping to get some photos of the laboratory in Dili to put on our website soon. The other participants were Falekaka Tomu from Vaiola Hospital Lab, Tonga; Sam Amos, Santo, Vanuatu; Ricky Lee, Vila Central Hospital, Vanuatu; April Solang, Belau National Hospital Lab, Palau; Eric Fole, National Referral Hospital Lab, Solomon Islands and Roland Waki, Kiliuufi Hospital Lab, Solomon Islands. During the course staff from Roche Diagnostics spoke to the students about maintenance and trouble shooting on Sysmex analysers, which many of them have, and Alison Bond from BD spoke about blood sample collection, the correct use of vacuum tubes plus the changes and advances that are occurring, and the importance of this whole pre-analytical area of testing.

We thank the two tutors Marilyn Eales and Phil Wakem for all the hard work they put into the course and we have received positive feedback from a number of the students and their supervisors about how they are putting into practice what they learnt while here.

Joint WHO-CDC International Conference on Health Laboratory Quality Systems
This meeting was held in Lyon, France from 9 – 11 April with around 200 delegates present from all areas of the world; the PPTC was represented by John Elliot.

As the background paper to the conference stated: “The critical importance of health laboratory quality is now more widely recognized than ever before, and greater demands arise for implementing laboratory quality systems, including the setting up of national laboratory quality standards at country level. The poor quality of laboratory results can result not only in inappropriate medical treatments for individual patients but also in constraining timely and effective investigation and response of epidemic-prone diseases. At global level, the International Health Regulations (IHR) logically require all the WHO Member States to establish a mechanism ensuring quality laboratory results allowing reliable and timely laboratory identification of any aetiological agents and substances likely to cause public health emergencies of international concern. … it was identified that it is integral for all the Member States [of WHO] to develop national quality standards, which are reasonably achievable and clearly show the minimal standards that all the health laboratories in the country should conform to….”

As all countries in the Pacific are members of WHO this will mean that they will all have to develop a national quality standard for their laboratory and you as laboratory staff will have to be involved in this.

Joint Consultative Meeting on HIV Testing in the Pacific
This meeting was held in Pago Pago early in May with the objective of making recommendations on an algorithm for HIV testing. A large number of different organisations involved in HIV/AIDS and STIs were present and John Elliot attended as the PPTC representative. There were a number of presentations and reports on new rapid tests that will become available in the near future. I will give a full report on this meeting in the next Pacific Way but in the meantime don’t make any changes to your HIV testing protocols or to the tests you are using unless advised by us, WHO or SPC.
Journal-based questionnaire

Below are 10 questions based on articles in this, the August 2008 issue of the Journal. Read the articles fully and carefully, most questions require more than one answer.

Answers are to be submitted through the NZIMLS web site. Make sure you supply your correct email address and membership number. The site will remain open until Friday 12 September 2008. You must get a minimum of 8 questions fully right to obtain 5 CPD points.

Journal questions

1. Morphologically, into which varieties can diffuse large B-cell lymphomas be divided into.
2. What are the classic CSF findings in malignant meningitis.
3. What did the H & E stained sections of the bronchial biopsy of the pulmonary mucormycosis case show.
4. What are the common fungi genera causing mucormycosis.
5. In which tissues is mucormycosis normally involved in.
6. What are the main symptoms of pulmonary mucormycosis.
7. What is the cytologic diagnosis of mucormycosis based on.
8. Overall, what was the predominant uropathogen isolated in Benin City, Nigeria and what were the next two predominant uropathogens.
9. What has been reported to increase the risk of asymptomatic bacteriuria in diabetics.
10. What is the plasma half-life of intact parathyroid hormone and where does its biological activity reside in.

Questions and answers for the April 2008 journal-based questionnaire.

1. What are the main criteria for authorship. Substantial contribution to design and conception, or data acquisition, or analysis and interpretation. Drafting manuscript or critical revision. Final approval.
2. What is the genetic basis of methicillin resistance of MRSA and what does it render MRSA resistant to. Acquisition of mecA gene. All B-lactam antibiotics.
3. What does the methicillin resistance gene encode and what is it carried on. Methicillin-resistant penicillin-binding protein, PBP2a. Carried on a mobile genetic element, the staphylococcal cassette chromosome mec (SCCmec).
4. In how many health workers was MRSA isolated and which strains were they. Four. EMRSA-15 and WSSP1.
5. What is the ‘gold standard’ for identification and confirmation of MRSA isolates. Presence of mecA gene by PCR.
6. Why are patients at a higher than normal risk of acquiring S. aureus infection. In-patient population tends to be older, sicker and weaker.
7. Pseudo (platelet type) von Willebrand disease is a rare disorder arising from what. Arising from a gain of function mutation in the gene for GP1BA.
8. What is the recommended drug therapy for risk of bleeding during childbirth and if bleeding occurs despite this, what further treatment is recommended. DDAVP. 1500 units of Biostate.
9. To differentiate Type 2B vWD from pseudo vWD which mixing studies are performed. Ristocetin Induced Platelet Aggregation (RIPA) mixing studies.
10. What is Type 2B vWD caused by. Functionally defective vWF with high affinity for GP1BA resulting from a mutation in the vWF gene located on chromosome 12.
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