Histology Group of Victoria Inc.

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Contents

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"The HGV aims to provide a dynamic continuing education program in which all persons with an interest in Histology and Histotechnology are freely invited to participate."

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Submissions:

Author enquiries and readers wishing to contribute articles or reports can contact the Editor – Neil O'Callaghan (98151588), email: editor@hgv.org.au or post directly to

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Please send articles on floppy-disc (preferably Microsoft Word format) for inclusion in the next edition.
All articles submitted for publication will then become the sole property of the Histology Group of Victoria

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From The Chair: A Blurb from the Bush

Well the National Histology conference is done and dusted for another two years. The South Australian Histology group is to be congratulated on presenting an outstanding meeting in all aspects. It was arguably the most professionally operated National conference and hosted an excellent array of renowned speakers and topics. The Saturday evening conference dinner was full of entertainment and frivolity, with good food and wine, a band and plenty of Histologists taking to the dance floor. None more impressive than the unique genre of moves that Greg Jenkins admits that only he is proficient.

Congratulations to Tania Marsden from Victoria who presented an excellent talk on Rhabdoid tumours and to Jarrod Phillips, who whilst now residing in WA, presented his findings of a study conducted at the Royal Children's Hospital on the importance of local perinatal autopsy standards.

Over 300 delegates registered for the workshops and conference, and it is without doubt that Histologists around the country see this event as the premier conference for Histology content, trade displays and networking. The 5th National Histology meeting is planned for 2011 and is to be hosted by the New South Wales Histology group.

Locally we have a scientific meeting in early June focussing on tissue processing, being presented by two old stalwarts, Geoff Rolls and Neville Farmer. Following this in early July the HGV is again putting on a cut-up workshop, focussing on lymphoid tissue, Lletz/Leep/Loop specimens, curetting, and POC. Due to costs associated with putting this workshop on, the committee has this year decided to levy a small $20 registration cost.

The trivia night is also fast approaching so start thinking about putting your workplace tables together. If you cannot fill a table, don't worry; we will accept any numbers and form tables of participants so nobody need miss out. Details for the trivia night and forthcoming scientific meeting and workshop are within and on our website www.hgv.org.au.

Our 2010 one day seminar plans are evolving. The date is 20th March and it will be held at St Vincent's Hospital Melbourne. Our aim is to bring a quality, diverse range of histology presentations and trade displays for a very reasonable cost to as many HGV members as possible. More information about program, workshops and of course the social events will be in subsequent editions.

Adrian Warmington
HGV President

Histochat:

HGV Inc. has introduced a bulletin board style discussion forum to their website - www.hgv.org.au. We hope this bulletin board "Histochat" will become a forum for the open exchange of information and ideas within the histology community.

Registration is required, as is email authentication, to access *Histochat*. No subscription fees are required and email addresses are used for correspondence and verification only. Registration is open to all. Students and junior staff are encouraged to participate. Free email clients such as hotmail may treat your authentication email as SPAM or JUNK MAIL, please check these folders if your authentication email does not arrive promptly. Authentication email needs to be responded to within 24 hours of registration. To those with online forum experience navigation should be relatively straight forward.

For those who need a little guidance YaBB have put together a step by step guide at www.yabbforum.com. Click on the "**Get Support"** link then click on "**Yabb Integrated Help**" There's no direct link on our web site as Yabb block direct linking to their help pages.

The forum layout is shown below:

There are a few broad forum topics. It's up to you to expand on them, ask questions, answer questions or just tell us your ideas. You can even upload images to assist with your discussions.

Sean Phefley, HGV IT Support

Meeting Report:

Meeting Report:

Scientific Meeting—ABPAS Test and Teach by Sonya Prasad and Erin Little of RCPA QAP

Sonya is the Technical Manager of the RCPA QAP and Erin is the Quality Representative, and together they gave a lively presentation of this Test and Teach on the Alcian Blue/Periodic Acid-Schiff technique.

Sonya began with a comprehensive review of the chemistry involved in staining acid and neutral mucins. In the PAS method, periodic acid oxidises adjacent glycol groups to form dialdehydes. These dialdehyde groups react with Schiff's reagent to form an insoluble magenta compound. Carbohydrates which stain with this method include polysaccharides, mucopolysaccharides, glycoproteins and glycolipids**.**

Alcian Blue, however, is a positively charged copper phthalocyanin basic dye. This attaches by adsorption to negatively charged acid mucins. In solution, acidic groups (carboxylic or sulfonic acids, etc.) will lose a proton and become negatively charged (anionic). Thus, the pH of the solution determines the extent to which any chemical group is protonated or deprotonated. In practical terms, this means that the staining pattern may be altered simply by altering the pH of the staining solution. Since most acid mucins are stained at pH 2.5, this is the pH mostly used in the ABPAS technique.
The discussion then went on to how the method is performed. The Alcian blue solution is used first to stain the acid mucins blue. Any acid mucins that are also Periodic Acid Schiff (PAS) positive will not react in the subsequent PAS reaction leaving only neutral mucins to be stained. The PAS reaction is then used to stain glycogen and other neutral mucosubstances magenta.
We then looked at some control material that showed mixed glands containing both AB and PAS positive material which stained purple. This was deemed not advisable and separate composite blocks were recommended in order to provide superior positive colour identification between the two types of mucins.

Erin gave a summary of the assessment criteria used at the QAP and gave an explanation of some of the comment codes. He also explained that the two recent technical exercises on the ABPAS were designed to aid laboratories in improving their results, make changes to their methods, and thus learning from the first exercise.
Several stained slides were shown and explanations were given as to their ratings and possible remedies for improvement for some of the less satisfactory stains. One of the things pointed out was the pink tinge sometimes found in the slides. This was deemed to be a result of insufficient washing off of the periodic acid prior to putting on the Schiff's reagent. A washing time of 5 minutes in running water was suggested.

A slide giving the results of the latest exercise came next which showed that 79% were satisfactory, 13% borderline and 8% unsatisfactory. Most laboratories used a manual method with in house prepared alcian blue stain and periodic acid, with a commercially produced Schiff's reagent, followed by Harris' or Mayer's haematoxylins as counterstains.
Only one slide was brought in for discussion, however it was difficult to assess the colours and it's quality using the microscope and laptop computer on hand.

Reported by Elizabeth Baranyai
Cabrini Health.

Trivia Night

Friday 31st July 2009

7pm – 11pm

Histology Group of Victoria Inc.

The Mount Erica Hotel
420 High St, Prahran

$20 per person or $200 per table.
(inc finger food, Drinks extra)
Tables of less accepted.
Why not amalgamate institutions? Or adopt your favourite trade rep for your table.

Who am i?
The rich List?
Musical challenge & More.
Chance for bonus points.
Prizes

Full payment must accompany this form. Payment will not be accepted on the night. No exceptions!!!Seats are limited, so book early to avoid disappointment. RSVP & payment due Friday 25th July. Contact Maria Chavez 9594 3493.
Please make cheque or money order to HGV Inc.
Return this section with payment and send to: Po Box 1461. Collingwood, Victoria 3066.

Contact Name .....Contact number.....
Institution/s.....Number of seats.....
Enclosed is a remittance for $.....

Roche

Article Review:

Grow Your Own Control Tissue

The field of tissue engineering took another significant step forward earlier this year with researchers from Stanford University School of Medicine publishing a paper in with they describe a method of enabling stem cells to thrive ex *vivo*[1].

In vitro generation of solid organs from stem cells has until now been limited by the lack of provision of a functional vasculature. Research groups have previously been able to generate some tissues and minor organs, such as blood vessels, and the first whole organ transplant, consisting of cells grown from the patients' own stem cells, was recently performed[2]. Tissue engineering companies are now even marketing technology for generating blood vessels from patients' stem cells with clinical trials under way[3].

The Stanford University team used explanted micro-circulatory beds (EMBs) as the scaffolding into which they seeded a number of types of stem cells. The article describes EMBs as "consisting of an afferent artery, capillary beds, efferent vein, and surrounding parenchymal tissue"[1]. The EMBs are maintained in a bioreactor and perfused with an oxygenated "soup" of finely tuned nutrients. The team plan to grow significant cell populations and eventually hope to create whole organs. However, they do concede that other methods may come along which may be more effective[4].

The application of stem cells in therapy and transplants may soon be routine but what about histology? Maybe the ability to create our own control tissues is not too far off into the distant future either…

References:

1) Chang *et al*. (2009) Tissue engineering using autologous microcirculatory beds as vascularized bioscaffolds. *The FASEB Journal*. 23:906-915.
2) Macchiarini *et al*. (2008) Clinical transplantation of a tissue-engineered airway. The Lancet. 372-9655; 2023-2030.
3) www.cytograft.com
4) http://www.sciencedaily.com/releases/2009/02/090226110657.htm

Simon Davies
Leica Microsystems

Have Your Say:

The HGV would love to hear from you and let you have your say! Email your thoughts to editor@hgv.org.au along with your name or pseudonym, as we would like to publish some of your issues or responses in our forth coming editions. Or pose a question, what would you would like see discussed.
Editor

Embed with Madonna:

Wouldn't you know it – just when you thought there was absolutely **NO** reason to open Paraffinalia – an old friend reappears. I know, I know, a lot of you have been wondering – just where did Madonna go …. and why? When last you heard from me, I was swapping my white lab gown for gowns of better quality (and colour) and forging a new life for myself minus the formaldehyde and paraffin wax.

Not merely content at being a Mrs, I decided to take it one step further and become a mother – twice over! No one prepared me for the consequences of a night of way too many Long Island Ice T's and Sex & The City re-runs. One kid, ok – but TWO, Jeeeeeezzzzz, I really should have cancelled the FOXTEL subscription

However daunting it all seemed at first, I must say that working in a Histology Laboratory somewhat prepared me for the onslaught of nappies, sleepless nights and emissions I only though possible in the Post Mortem Room.

I knew it wasn't going to be easy – but then again, neither is the Bielschowsky! 'Baby' days can be rather like days in the lab – you just have to roster yourself through each area.

Take Specimen Reception for example. You never know really what you are likely to be confronted with when undoing a nappy – sort of like opening a fresh bowel left in Theatre overnight.

Emissions, from either the proximal or distal end of the infant, are equally powerful projections of fluid that seem to reappear on a wall, on the carpet or even more disturbingly, on your clothes, hours later. Instead of the ubiquitous white T's and three quarter pants, a lab gown would be more appropriate!

You then fix/wash, process/clothe and embed/feed the infant – and just like the lab on a busy Friday, you get to do it all again, three hours later (remind you of those dreaded short cycles anyone?).

Special Stains is likened only to the Mother's Group get together when some unsuspecting 'first timer mother' (like myself) brings along the coloured textas and Playdoh. Why colour in on the paper when drawing on each other is so much more fun? And does ANYONE know how to get ground in Playdoh out of cream carpet?

The on-call roster is what really prepared me for the sleepless nights. You know the feeling – it's a Friday night and you have a date, then the open lung biopsy call at 6pm. The surgeons don't really know what time it'll happen – same as for babies. If *they* don't wake you up screaming, your boobs almost self combust.

Dealing with the toddler is just like dealing with a Consultant who is ticked off at having to come into Cut-up to fix up a Registrar's macro and the tediousness of watching a 3 year old eat is only likened to watching a brand new Reggie dissecting their first Gall Bag – the frustration is so crippling that you physically hold yourself back from ripping the scalpel/fork out of their hands and doing it yourself.

Patience, that is what twenty years of histology taught me and only now can I draw upon my experiences. But there are some good sides of being a parent – when they smile it's like cutting the best frozen section ever, and when they reach up and take your hand it really is like looking at the perfect renal PASM.

Don't get me wrong, they can turn at any minute, just like the Lab Manager fresh out of a Budget Meeting and there are days I would rather cut 100 HIV infected brain frozen's than deal with toilet training and finding that lost Bob the Builder toy.

One day I will return back to Lab life, but not in the foreseeable future.

A wise Cytologist once told me 'no matter what, always slap some lippy on in the morning' (Chanel No. 142 for those interested) something I do religiously and I try to have at least one cup of (hot) liquid a day, although my days of lattes and lunches at hip cafes are numbered – for the time being…

From the land of Bugaboo's, Bonds and 4 wheel drives, until we next 'embed' together….. Madge x

Intermediate
Cut Up Workshop 2

Histology Group of Victoria Inc.

2nd July 2009

Michael Chamberlain Lecture Theatre
St Vincents Hospital
Victoria Parade East Melbourne

Program
This is the second of many workshops aimed to educate our members in some of the specimens classified as "**non complex**".

Session One 6.30 pm to 7.15 pm
This session will cover the theory and practices behind the description, protocols and dissection of lymphoid tissue, with reference to the associated testing. Presented by Dr Stephen Lade, Peter Mac.

Refreshments 7.15 to 7.35

Session Two 7.35 to 8.20
This session will cover the description, protocols techniques and challenges that surround cervical tissue(including LLETZ, Loops & Leeps), Currettings and POC's. Presented by Dr Stephen Chan, Dorevitch Pathology.

Registration is essential to secure a place at the workshop.

To Register

To secure a seat, for catering purposes and production of printed material it is essential to register for the intermediate Cut up Workshop.

Early Bird Registrations close: 5th June 2009. Cost $20
Final registrations by: 25th June 2009. Cost $25

Email Registrations Only

Please email membership@hgv.org.au including:-

1) Quote "Cut Up Workshop Registration"
2) Your name
3) Institution
4) Contact Number
5) Please indicate if you **do not** wish to receive other electronic information from the HGV.

The HGV will send you an invoice **after** you have emailed your registration. This will include payment instructions

Article Review:

Are survival predictions reliable? Hospital volume versus standardisation of histopathologic reporting for accuracy of survival estimates after pancreatoduodectomy for adenocarcinoma.

Westgaard A et al., Eur J Cancer (2009), doi:10.1016/j.ejca.2009.03.019

The prognosis for adenocarcinoma in the pancreatic head is poor and histopathologic reporting is often not standardised. In order to investigate new treatment plans and have accurate survival estimates it is important to ensure that histopathologic reporting of resected specimens is of the highest calibre.

Non standardised reporting could lead to inaccuracies in survival estimates in either direction. By non-reporting of the resection margin or lymph node involvement, it could skew results in a negative way. Positive skewing could result from including non- pancreatic tumours with a better prognosis in the report due to difficulty in diagnosis. Standardisation of reporting increases the quality of reports and ensures that clinical trials would then be comparing the same information.

A comparison of non-standardised reporting with standardised reporting using the prognostic factors, tumour size, lymph node involvement, resection margin involvement, and tumour origin would examine the importance of standardisation for survival outcomes. Previous comparison of survival outcomes has not been reported according to the authors. Age, gender, surgery date and cancer origin site were obtained from the cancer registry.

The study hospital with standardised reporting used a template which included gross examination, specimen dissection, tissue sampling and microscopic examination.
The comparison hospitals did not follow any guidelines and there were no national or regional rules.

This study showed that standardised reporting led to an increase in number of blocks taken and more lymph nodes studied. Tumour size, resection margins, small vessel and perineural involvement were all consistently included.

Overall 5 year survival rate at the study hospital was increased even though regional lymph node involvement and resection margin involvement was also reported more often. The study hospital also had increased survival rates when node free resection was reported but this did not occur with node positive resection. Similarly tumour size less than 2.5cm reported at the study hospital had a better outcome than in the comparison hospitals. Cancer origin site and differentiation of the tumour had similar outcomes in the two groups. Standardised reporting of lymph node involvement is the most important factor for accurate survival rates.

Accuracy of diagnosis and prognostic outcomes are significantly improved with standardised histopathologic reporting.
The authors note that template reports provide a checklist for the pathologist and accuracy and consistency for the clinician.

Raelene Houwen
Dorevitch Pathology

Australian Institute of Medical Science (AIMS) Preliminary Flyer and Date claimer

AIMS Tropical Division

and Partners

25th Annual North Queensland Conference

11th to 14th June 2010

Celebrating 100 Years of Tropical Medicine in Townsville

On behalf of the Organising Committee I extend you an invitation to attend our 25th Annual Conference to be held from 11th to 14th June 2010 at Jupiter's Hotel & Casino.

Contact:
David Porter
+61 7 47962400
david_porter@health.qld.gov.au
www.aims.org.au

Professional Partners to date;

- *The Australasian College of Tropical Medicine (ACTM).*
- *North Queensland Centre for Cancer Research (NQCCR)*
- *Histotechnology Group of Queensland*
- *Australian Association of Clinical Biochemists (AACB)*
- *Australian Phlebotomy Association*
- *Australian Society for Microbiology*
- *James Cook University*

Article Review:

Prognostic gene signatures for non-small-cell lung cancer.

Boutros P. C., et al. 2009. Proceedings of the National Academy of Sciences. 106(8), pp2824-2828.

Non-small-cell lung cancer (NSCLC) is principally treated by resection and this can cure the patient. However, in patients with stage IB to IIIA there is a significant number (30-60%) of patients whom experience recurrence and die within 5 years. Adjuvant chemotherapy has been demonstrated clinically to benefit patient outcomes at stages IB to IIIA but it is unclear if the treatment of Stage IA patients is beneficial. Within the Stage IA patients there appears to be a subgroup that have more aggressive tumours and would therefore benefit from adjuvant therapy. Different research groups have attempted to identify the at-risk subgroup of Stage IA patients using mRNA expression profiles of the removed tumours. However, there has been little overlap in results between research groups primarily due to the diversity of biological responses requiring thousands of samples to be taken to develop robust and reproducible results. The aim of this paper was to develop an alternative statistical analysis method for examining mRNA expression data that would enable patients with NSCLC cancer to be divided into distinct prognostic groups.

Using their own previously published real time-PCR NSCLC dataset of 158 genes from 147 patients a modified steepest decent (mSD) algorithm was devised by combining unsupervised pattern recognition with a gradient descent optimisation.

From the original dataset a prognostic signature containing six genes was identified: syntaxin 1A (*STX1A*), hypoxia inducible factor 1A (*HIF1A*), chaperonin containing TCP1 subunit 3 (*CCT3*), MHC Class II DP beta 1 (*HLA-DPB1*), v-maf musculoaponeurotic fibrosarcoma onco-gene homolog K (*MAFK*) and ring finger protein 5 (*RFP5*). This enabled patients to be divided into two groups with significantly different clinical outcomes.

The six gene signature was then tested on four publicly available datasets from independent groups and was found to be prognostic for all four datasets, even when one of the genes from the signature set was missing from the test dataset. A further 589 patients from other older 8 datasets were also analysed. Again the six gene signature separated out the patients into two prognostic groups. This also occurred when only the Stage I patients were examined.

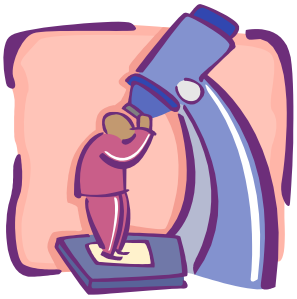
Boutros et al 2009 also questioned whether there may be other six gene sequences that could be used to differentiate prognosis. By cross matching genes from their original study and those of the four other studies they found 113 common genes. These gave rise to 10 million permutations of six gene combinations. The original six gene set was 99.999% superior to any other combination in the groups original data set and 99.98% superior in the other four data sets from other groups.

Finally, they examined gene enrichment for the 113 common genes. Three of the genes, STX1A, HIF1A and CCT, were found to be among the top 10 enriched genes. This suggests that they could particular to NSCLC or to the stage of disease raising the possibility for the six gene sequence together with some housekeeping genes to be used as a prognostic indicator for patients.

Review by Nardia Baxter – St John of God Pathology Ballarat

Under the Microscope:

reported by Maria Chavez



Cameron Skehan
Grade 2 Scientist
Monash Medical Centre

1.What was your first job?
My first job was in the Anatomical Pathology department at the Alfred Hospital as a lab assistant

2. What attracted you to Histology?
I would like to say that there had alway been a yearning to become a histologist but realistically it was the first job that I got. Having said that though, once at the Alfred both Barb Thomas and John Hall set me on the right path and inspired a love of histology in me.

3 .What is the worst decision you have ever made?
If my friends were to read this they would probably disagree but I don't think that I make really bad decisions, just small hiccups that I remember not to repeat. Je ne regrette rien!!!!

4. What is the best decision you have ever made?
I have two best decisions: 1) Taking up teaching histology to students at Holmesglen Institute: It's really rewarding when you end up having some of them eventually come back to work with you. 2) Learning to become a cytology screener because now I am able to work in all areas within anatomical pathology not just histology (hope that doesn't sound like I am bagging histology).

5. Who would you most like to have dinner with and why?
Karl Lagerfeld would definitely be the guest of honour, the man is an absolute creative genius and style icon. Madonna, just because I have been harbouring some weird obsession with her for a while now, plus I think she would be good company for Karl because if both of these people turned up for dinner I would have massive stage fright. Last but definitely not least is Ms Sonya Prasad, surely I don't need to explain why!

6. What music do you enjoy listening to?
I really listen to a wide variety of music but my all time favourite band would be the Pixies with the Strokes coming a close second.

7. What is your favourite stain?
Besides the PAP stain (sorry that was the cytologist in me speaking) I am quite fond of the AFG. I really like the look of the elastic lamina in temporal artery biopsies stained using this technique.

8. What is your favourite food/Restaurant?
I don't really have a preferred food style, I will give anything a go once with the exception of certain forms of offal. My favourite restaurant ever is a Michelin two star in Lyon called Christian Tetedoie, it has been my most memorable dining experience so far.

9. What are you reading at the moment?
At the moment I'm reading DeMays' Art and Science of Cytopathology because I am studying for my CT(ASC) exam.

10. What is the best conference you have ever attended?
The 2008 ASC conference in Sydney. It the perfect mixture of education and entertainment.

11. Are there any current projects you are working on at the moment?
Currently I am working on protocols for the processing of non gynae specimens on our newly purchased ThinPrep processor. I am especially trying to perfecting the art of producing an air dried smear using this instrument.

Future Scientific Meetings: 2009

Histology Group of Victoria Inc.

5th March
~~Scientific Meeting – Series of Short Presentatior~~
~~Venue – PeterMac~~

30th April
~~Scientific Meeting – ABPAS Test and Teach~~
~~Venue – PeterMac~~
~~Speaker – Sonya Prasad RCPA QAP~~

8th – 10th May
~~4th National Histology Conference~~
~~Hosted by Histology Group of South Australia~~

4th June
Scientific Meeting – Tissue Processing
Venue – PeterMac

2nd July
Cut – Up Workshop
Venue – St Vincents

31st July
Social Event – Trivia Night
Venue – Mt Erica Prahran

3rd September
Scientific Meeting – Liver Biopsy Scoring System
Venue – PeterMac

12th November
Scientific Meeting – New Antibodies & AGM