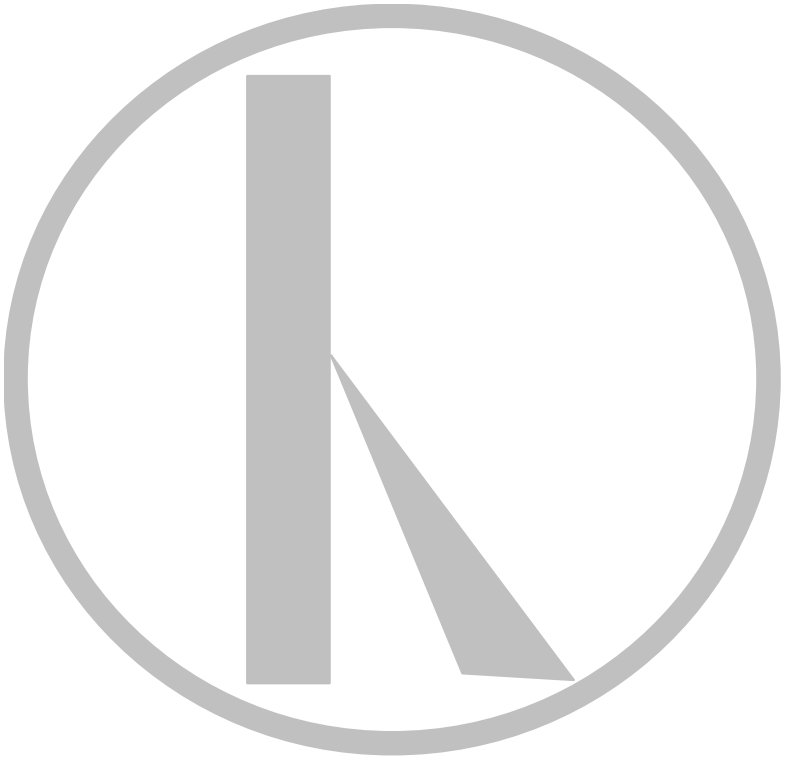
Histograph

www.histonsw.org.au

ISSUE 2
July 2013

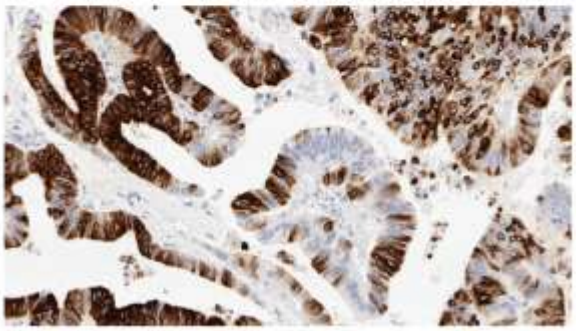


Newsletter of the Histotechnology Society of NSW

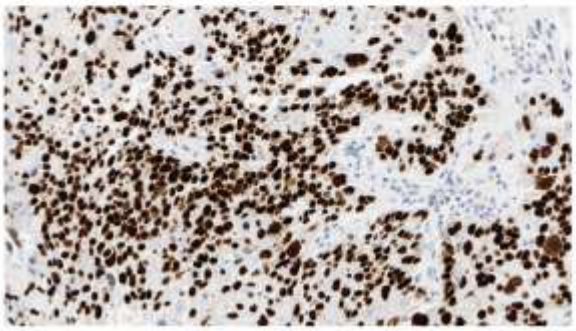
Dako
A Better Path™

Dako Expands the Antibody Portfolio

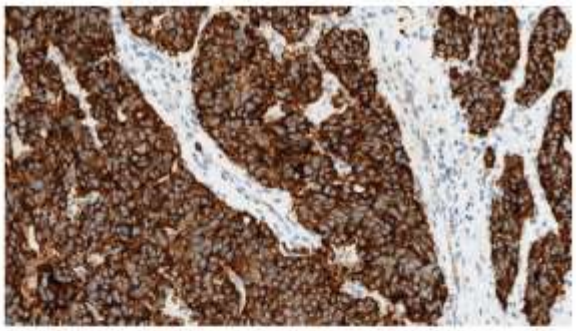
In June 2013, Dako strengthens the IHC menu of antibodies through the launch of: Anti-Synaptophysin, Anti-MUC5AC and Anti-p63 Protein*



MUC5AC staining. Pancreatic adenocarcinoma. The majority of the neoplastic cells show a moderate to strong cytoplasmic staining reaction.



p63 staining. Squamous cell lung carcinoma. The majority of the neoplastic cells show a strong nuclear staining reaction.



Synaptophysin staining. Lung atypical carcinoid. The neoplastic cells show a strong cytoplasmic staining reaction.

Antibody	Description	Platform	Ordering number
Monoclonal Mouse Anti-Human MUC5AC, Clone CLH2	FLEX RTU 60 tests, 12 mL	Autostainer Link 48	IR66161
	Concentrate 0.2 mL	-	M731629
	Concentrate 1 mL	-	M731601
Monoclonal Mouse Anti-Human p63 Protein, Clone DAK-p63*	FLEX RTU 60 tests, 12 mL	Autostainer Link 48	IR66261
	Concentrate 0.2 mL	-	M731729
	Concentrate 1 mL	-	M731701
Monoclonal Mouse Anti-Human Synaptophysin, Clone DAK-SYNAP	FLEX RTU 60 tests, 12 mL	Autostainer Link 48	IR66061
	Concentrate 0.2 mL	-	M731529
	Concentrate 1 mL	-	M731501

*p63 will not be launched in the US.

If you would like further information about the new antibodies please call:
Natalie Fennell on 0402 405 226 or
Customer Care on 1800 653 103

Contents

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Editorial

Well at last this issue of Histograph has finally come together. We hope you find the articles informative and entertaining. The answer and accompanying discussion of the Test and Teach from the last issue is included as well pretty crystals, Vince Munro's Methylene blue for melanoma immunoperoxidase staining, thoughts on microtome blade saving and the important safety concern of the Pregnant Histotechnologist.

Hopefully, this will be my last issue as editor. After more than 10 years, it is time for me to hand over the reins (I hope). I have much to do in my twilight years including partying, travelling, completing my famous text book, did I mention partying? So see you at the AGM and the workshops

Tony Henwood,
Editor
anthonyh@chw.edu.au

Chairman's Report

Our Annual General Meeting is in the later part of August and we would like to see as many members and guests in attendance as possible. Apart from the AGM and the election of the new committee (we encourage new blood on the committee so consider becoming involved); we have an interesting presentation by Lawrence Young. This should be a great night and a time to meet and talk with fellow colleagues.

The focus on workshops continues with a " Frozen Section " workshop planned for September at Granville FAFE and a "Microscopy "workshop planned for early November, also at Granville TAFE.

Planning is well underway for our next NSW Conference in Orange, the 16th, 17th and 18th May, 2014. The venue will be the Orange Ex-Services Club with the associated Templers Mill Motel associated with the Ex Services Club Motel ,which has been block booked. Accommodation arrangements will need to be made by the attendees. There are also other motels nearby. For our Conference dinner we have a booking with Highland Heritage Estate Winery and function centre, which has an excellent dining room. Bus transport will be provided for those people staying close to our Conference venue. We will also have our usual trade display. Please note this in your diary. We will have a great weekend.

A reminder that membership fees are now due, and looking forward to seeing you at our AGM.

Cheers,

Trevor Hinwood.
Chairperson.
Histotechnology Society of NSW.

HOUSEPLANTS THAT WILL DETOXIFY THE AIR IN YOUR HOME

Areca Palm

removes indoor chemical toxins

Ficus Alii

removes toxins to purify the air

Lady Palm

improves indoor air quality

Dracaena Janet Graig

removes trichloreoethylene

Dwarf Date Palm

removes indoor air pollutants, particularly xylene

Bamboo Palm

removes traces of benzene, trichloroethylene and formaldehyde within the home

Boston Fern

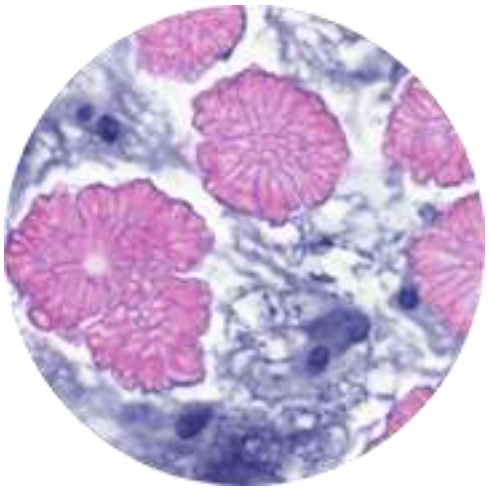
removes indoor air pollutants, particularly formaldehyde

Peace Lily

removes alcohols, acetone, trichloroethylene, benzene and formaldehyde from indoor air

Pretty Flowers -Tyrosine Crystals Histochemistry

Have you ever been shown a slide containing the extraordinary structures as show in the figure below?



These radially arranged, "petal-shaped" clusters of glossy, eosinophilic structures, surrounding a central core are tyrosine crystals. The periphery of the projecting structures was distinctly lobular, and the structures were refractile on in-and-out focusing (1).

Tyrosine crystals are floret-shaped eosinophilic extracellular structures that were first reported in a case of pleomorphic adenoma of the parotid gland in 1953. Despite the name, tyrosine crystals are thought to be composed of a mixture of proteins rather than pure tyrosine (2).

Several types of crystalline structures may be found in cytologic or histologic specimens of salivary glands. Tyrosine-rich crystalloids, non-tyrosine (alpha-amylase) crystalloids, and collagenous crystalloids are the most commonly encountered forms. By light microscopy, the large (5–500µm), geometrically shaped non-tyrosine crystalloids may be easily distinguished from the much smaller (30–60µm), floret-shaped tyrosine-rich crystalloids, and the radially arranged, needle-shaped clusters of collagenous crystalloids. Tyrosine-rich crystalloids are occasionally found in the stroma of benign (e.g., pleomorphic adenoma) and malignant (e.g., polymorphous low-grade adenocarcinoma, adenoid cystic carcinoma) salivary gland neoplasms, as well as in the lumen of non-neoplastic parotid gland cysts. On the other hand, non-tyrosine crystalloids have been observed only in benign and reactive salivary gland lesions, such as sialadenitis with sialolithiasis, lymphoepithelial cyst, pleomorphic adenoma, oncocytic cystadenoma, Warthin's tumor, and benign parotid gland cysts. Collagenous crystalloids have been described in association with both benign and malignant neoplasms, including pleomorphic adenomas, myoepitheliomas, and myoepithelial carcinoma, respectively (3).

The collagenous crystalloids are composed largely of radially arranged short bundles of collagen that form a stellate structure. The major periodicity of the collagen determined by electron microscopy of dehydrated tissue is similar to that of Type I collagen. The structures have the tinctorial properties of collagen with van Gieson's and Masson's trichrome stains. However, other connective tissue components also appear to be present. The pale blue coloration with alcian blue at pH 2.5 implies the presence of polyanions such as glycosaminoglycans. Intense argyrophilia seen with the methenamine silver stain implies the presence of carbohydrate. The PAS reaction is positive with the inclusions of smallest diameter and only faintly positive or entirely negative with

those of greatest dimension. One possible interpretation of this reaction to the PAS reagent is that some carbohydrate component is lost as the structures mature and become larger (1).

Non-tyrosine crystalloids are chemically composed of protein consistent with crystallized alpha-amylase produced by salivary gland cells in supersaturated saliva. It has been noticed that there is a relationship between non-tyrosine (alpha-amylase) crystalloids and cells with oncocytic features. It has been speculated that non-tyrosine crystalloids might represent a secretory product of salivary gland oncocytes. Some examples of non-tyrosine (alpha amylase) crystalloids may have been erroneously reported as tyrosine-rich because they showed histochemical positivity for Millon's reagent, used to detect the presence of tyrosine. However, a complex protein such as alpha-amylase contains some tyrosine and may therefore exhibit histochemical positivity for Millon's reagent (3).

	Tyrosine-Rich Crystalloids	**Collagenous Crystalloids**	**Oncocyte/Cyst-Associated Crystalloids**
Birefringent	No	Yes	No
Millon reaction	Pink	Negative (colourless)	Pink
DiPAS	Colourless to pale pink	Smaller structures bright pink: Larger structures pale pink or negative	Colourless
Alcian blue (pH 2.5)	Negative (colourless)	Pale blue	Negative
Methenamine silver	Negative (colourless to pale orange)	Black	Negative
Van Gieson's method	Negative (brilliant yellow)	Bright red	Negative
Verhoeffs Elastic	Black	Light grey	Black
Reticulin	Negative; with some black material between crystals	Grey to black	Negative
Masson's trichrome	Deep purple	Green	Deep Purple

So what is this Millon reaction that has been mentioned as the stain of choice for tyrosine? It was shown by Meyer in 1864 that a red colour is produced when tyrosine is treated with mercuric chloride in the presence of potassium nitrite in acid solution. Millon's test depends in reality on a similar reaction, nitrite being formed in the preparation of the reagent (4).

It is believed that the reaction proceeds in two stages. A nitrosophenol is formed first, by the substitution of -NO for -H in a position ortho- or meta- to the hydroxyl of the phenol. The red compound is then formed, apparently by the inclusion of mercury in a new ring that also includes the nitrogen of the nitroso-group. Nearly all phenols, except those that are doubly substituted in the ortho or meta positions, react positively, though thymol provides a partial exception. Tyrosine is naturally positive (4).

Unfortunately, the major issue with this stain is the use of the highly toxic mercury salt. The following is given for completeness (5):

1. Deparaffinize sections and bring to distilled water.
2. Place slides in 5% mercuric acetate in 30% trichloroacetic acid for 10 minutes at 40^{o}C.
3. Transfer immediately to another bath of the same solution, with 0.05% sodium nitrite added, for 1 hour at 30^{o}C.
4. Put slides directly into 70% ethanol for 10 minutes.
5. Give two additional changes of 70% ethanol, 10 minutes each.
6. Dehydrate sections in 95% and absolute ethanol, 3 to 5 minutes in each.
7. Clear in xylene or toluene, and mount.

Glenner (6) has described a nitrosophenol reaction for tyrosine. Nitrosation of tyrosine is performed in the presence of one of a number of divalent cations to produce an ortho nitrosotyrosine metal chelate compound. The nitrosotyrosine metal chelate is stable to light anti unless prolonged exposure to nitrous acid occurs, no diazonium salt is fornued. The reaction is performed at 50^{o}C to promote chelate formation

Solution:

Copper Sulphate	1g
Sodium Nitrite	1g
Acetic Acid	2ml
Distilled water	98ml

Method:

1. Dewax and hydrate paraffin sections to water
2. Place in pre-heated (50^{o}C) solution for 7 hours
3. Wash in water 5 minutes
4. Dehydrate, clear and mount.

Results paralleled those of the diazotization-coupling (D-C) reaction with negative reactions for tryptophan and gramicidin and positive reactions for tyrosine, gelatine anti zein. Though this technique is useful as a confirmatory technique for the localisation of protein bound tyrosine, its sensitivity is markedly less than that of the diazotization-coupling reaction (6).

Protein diazotization followed by alkaline coupling with a naphthol has been successfully applied for the histochemical localization of the protein bound tyrosine in tissues (7). Prolonged nitrous acid treatment in the cold is required (18-24 hours at 3^{o}C). The best coupling reagent so far tried is 11-amino-8-napthol-4-sulphonic acid (S acid) and coupling succeeds best in a 1 % potassium hydroxide in 70 % alcohol, allowing 1 to 4 hours at 3^{o}C. Topochemical results parallel those of the Millon reaction, with greater colour intensities and superior histologic definition.

Solutions:

- Nitrosate Solution

Sodium nitrite	6.9g,
Glacial acetic acid	5.8ml
Add distilled water to make	100ml

- S acid Solution

1-amino-8-napthol-4-sulphonic acid (S acid)	1g,
Potassium hydroxide	1g,
Urea	2g,
70% ethanol	100ml

1. Use 5um paraffin sections of material fixed in 4% formaldehyde or other suitable fixative.
2. Deparaffinise and hydrate as usual through warm xylene and graded alcohols.
3. Place sections in Nitrosate Solution 18 hours in refrigerator (3^{o}C.)
4. Wash in 3 changes of ice cold distilled water, 5 seconds each.
5. Couple in S acid Solution for 1 hour at 3^{o}C
6. Wash in 3 changes 10N hydrochloric acid, 5min each

7. Wash in water 10 minutes
8. Dehydrate, clear and mount.

Results: Hair cortex and the soft keratins of the epidermis, pharyngeal, esophageal and proventricular epithelia reddish purple. Background is pale pink to red depending on levels of tyrosine.

References

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2. Amanda H, Jerad G, Doug P, David M, Mark E (2013) "Chondroid Syringoma (Mixed Tumor) with Tyrosine Crystals" J Cutan Path 40(6):528-529.
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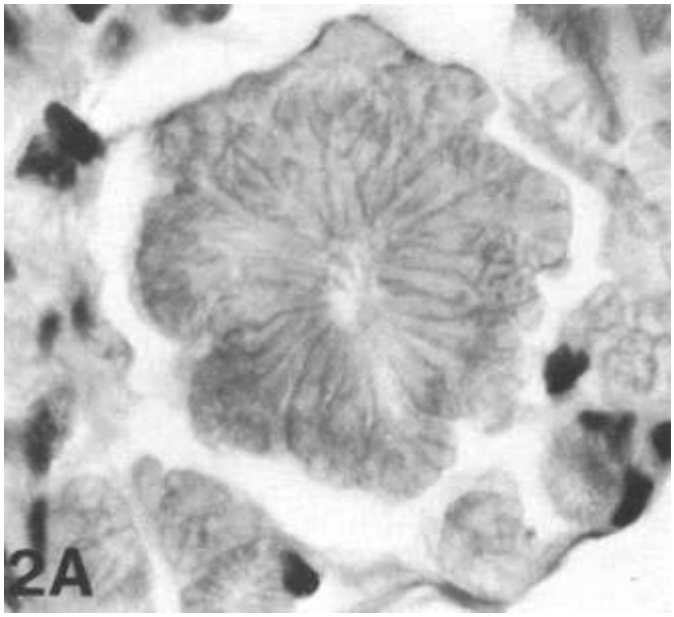


HISTOTECHNOLOGY SOCIETY OF NSW
POSTAL ADDRESS

PO BOX 871
SEVEN HILLS, NSW 1730

Cryotomy and Immunofluorescence Workshop

Place: Granville TAFE

Date Saturday 14th September 2013

Time 9-12

Cost $60

In this workshop participants will experience frozen sectioning using the latest cryotomes, perform direct immunofluorescence staining and review the results using modern fluorescent microscopes.

The workshop will contain a detailed lecture, explaining the theory and application of these techniques as well as examples of renal and skin frozen and immunofluorescent stained sections. Workshop notes will be provided.

Several companies including ThermoFisher, Leica, and Olympus will have Cryotomes and Fluorescent Microscopes available for use.

Places are limited; please contact

Bharathi Cheerala, Douglass Hanly Moir Pathology
Ph: 02 9855 6271 Email: bcheerala@dhm.com.au

Methylene Blue for Skin Biopsies Stained for Melanoma Markers

Anne Prins, following on from the last issue's discussion on differentiating melanin from DAB, has kindly supplied the follow technique. Anne, while working at St Vincents Hospital in Sydney, remembers that Dr Vince Munro insisted on a methylene blue solution for the counterstain on any skin immunohistochemistry as it stained the melanin green.

Methylene Blue for skin biopsies stained with S100 (or other immunohistochemical markers)

Staining Solution:

2.38gm Sodium acetate
4.7ml Acetic acid
5gm Methylene Blue
Make up to 1 litre with distilled water

Procedure:

1. After immunostaining, wash slides in tap water
2. Stain in Methylene Blue solution for 2 minutes
3. Wash well in water
4. Counterstain in Haematoxylin, wash well & blue as usual.
5. Dehydrate, clear and mount

Results:

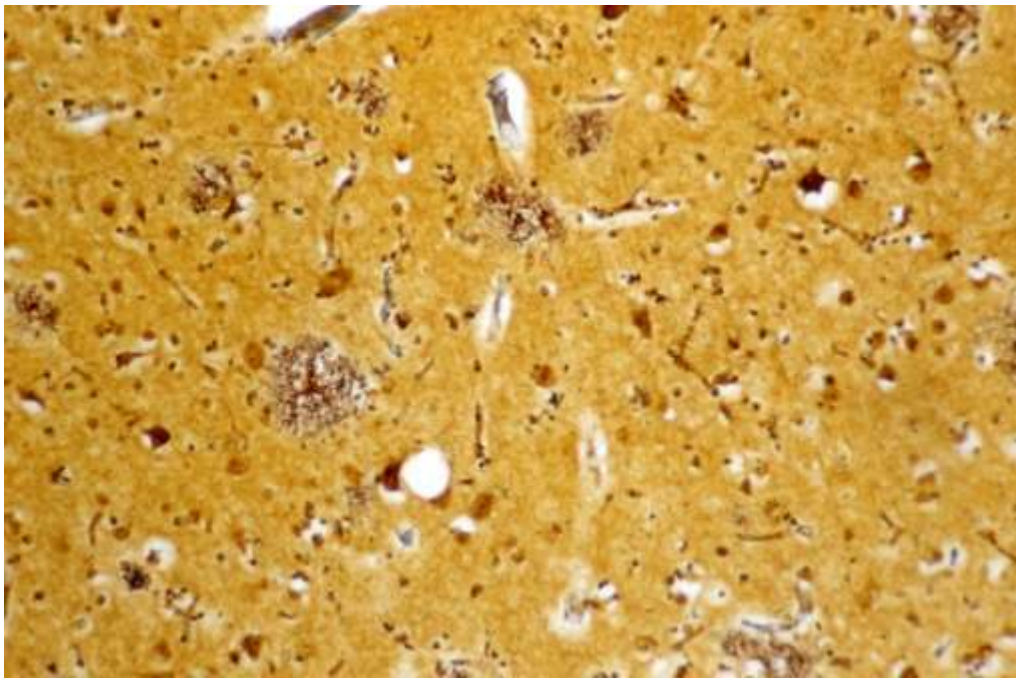
Melanin should stain green-blue.

Anne also brought to my attention that the Azure A method presented in the last issue was incomplete. As Anne has pointed out, "Solution 2 contains 600 ml of sodium acetate – no strength/molarity given and added to 3.4 ml 0.1M acetic acid and 27 ml of AD. Doesn't that sound odd to you? I did check the article on-line and that's what it said so I'm surprised there was no query. I would have thought that 600 ml of 'x' sodium acetate would nullify 3.4 ml of acetic acid, regardless of how strong it was".

So after searching the cited articles from the paper, I hope that the correct formulation is as follows (from Kamino H, Tarn ST (1991) Immunoperoxidase technique modified by counterstain with azure B as a diagnostic aid in evaluating heavily pigmented melanocytic neoplasms. Journal of cutaneous pathology, 18(6):436-439):

After the slides were immunostained and rinsed in distilled water, they were stained with a mixture of the following solution: 100 mg of Azure blue (Sigma) in 4 ml of distilled water. Solution 2 was prepared as follows: 600µl 0.1M sodium acetate and 3.4 ml 0.1 M acetic acid were added to 27 ml distilled water. Both solutions 1 and 2 were combined and 5 ml of acetone was added. The slides were incubated for 60 min at room temperature, differentiated in 95% ethanol and dehydrated in several changes of absolute ethanol, followed by clearing in xylene with subsequent mounting

What Stain is this?



What Stain is this? Bielschowsky's silver staining

What is the tissue? Brain

What is being demonstrated? Senile Plaques. The Bielschowsky's silver staining is commonly used to demonstrate axons, neurofibrillary tangles, nerve fibers and senile plaques.

Senile Plaque Update

Senile plaques (SPs) and neurofibrillary tangles (NFTs) are regarded as the main neuropathological changes in Alzheimer's disease (AD). They mainly occur in the parietal temporal neocortex and the hippocampus, but also in subcortical areas and in the cerebellum. SPs and NFTs can be detected by a number of silver stains such as Gallyas, Bielschowsky, or Campbell, with immunological methods using antibodies against Aß4 and Tau-protein as well as by the amyloid specific Congo red stain. Usually, post mortem histopathological proof of SPs and NFTs can confirm a clinically based presumption of AD (1).

The Bielschowsky silver stain impregnates both the amyloid and neuritic components of the senile plaques and reliably stains

neurofibrillary tangles (3). However, Bielschowsky method is not a specific method for plaques and neurofibrillary tangles since blood vessels also stain (4). Although the exact mechanism still remains to be clarified, it is plausible that prior treatment with silver nitrate provides active foci for subsequent silver deposition in the ammoniacal silver solution. It is reasonable to suppose that this stepwise impregnation (silver nitrate-ammoniacal silver) improved the staining probably by increasing the amount and the size of silver precipitates (5). The presence of a huge number of modifications, however, indicates that this method is highly unstable and still awaits improvements (5).

Many researchers have their own preferred demonstration method and several have compared different stains in order to determine the most sensitive method.

Lamy et al (2) compared seven staining methods for senile plaques and neurofibrillary tangles. They found that Yamamoto & Hirano's Modified Bielschowsky method revealed both amyloid and neurites. Cross, Bodian and Gallyas methods stained neurites preferentially and were more sensitive for neurofibrillary tangles than for senile plaques. Silver methenamine revealed amyloid in much the same way as thioflavine S. The highest count of senile plaques and neurofibrillary tangles was obtained with the Yamamoto & Hirano's modified Bielschowsky method. This technique is, however, expensive and difficult to perform. It stains a large number of normal structures and the recognition of the lesions is more subjective than with other selective stains. Lamy et al (2) therefore found it difficult to recommend this technique for routine use.

Litchfield & Nagy (3) found that the Bielschowsky stain is highly sensitive to changes of the ambient temperature. They found that the most reliable and easily reproducible modification of the Bielschowsky silver impregnation was one that used incubations at 5^{o}C.

Mavrogiorgou et al (1) found that the commonly used Bielschowsky method turned out to be insufficient in marking SPs and NFTs. They found that SPs were most effectively stained using the method of Campbell and the Gallyas stain was best for NFTs (1).

References

1. Mavrogiorgou, P., Gertz, H. J., Ferszt, R., Wolf, R., Bär, K. J., & Juckel, G. (2011). Are routine methods good enough to stain senile plaques and neurofibrillary tangles in different brain regions of demented patients?. Psychiatria Danubina, 23(4.), 334-339.

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HISTOTECHNOLOGY SOCIETY OF NSW

ANNUAL GENERAL MEETING (AGM)

23rd August Friday 2013 at 6.30pm

The Hitchhiker's Guide to the Laboratory

~ by Lawrence Young ,Dako An Aligent Technologies Company

What does a third generation, Australian born Chinese do when he is not only asked to transfer to Dako's global organization, but also move to China to help set up a Dako subsidiary in Shanghai? Whilst I did manage to learn some rudimentary Mandarin, and became more fluent in Cantonese, I also got the added opportunity to observe histotechnology in practice in more than 50 countries around the world.

"Best Practice" is definitely relative to the countries that you visit. We all aim to achieve the same outcome; it's just that we get there in all manner of different ways!

And….I had some of the most rewarding experiences imaginable.

*"In every job that must be done, there is an element of fun. You find the fun and – SANP- the job's a game," **~ from Mary Poppins***

***WHERE:** North Ryde RSL, 27/41 Magdala Rd*
Cnr of Pittwater and Magdala Rds, North Ryde

Finger food provided at the conclusion of the meeting.

RSVP: 16th AUGUST FRIDAY 2013

***Contact:** Bharathi Cheerala, Douglass Hanly Moir Pathology*

***Ph:** 02 9855 6271* ***Email:** bcheerala@dhm.com.au*

The Pregnant Histotechnologist

Safety Corner

Certain chemicals are known or suspected to harm fetuses or reproductive health of adults. Some examples of reproductive toxins are: anaesthetic gases, arsenic and certain arsenic compounds, benzene, cadmium and certain cadmium compounds, carbon disulfide, ethylene glycol monomethyl and ethyl ethers, ethylene oxide, lead compounds, mercury compounds, toluene, vinyl chloride, xylene, and formamide. The first trimester of pregnancy is a period of high susceptibility. Often a woman does not know that she is pregnant during this period. Individuals of childbearing potential are warned to be especially cautious when working with such reproductive toxins. These individuals must use appropriate protective apparel (especially gloves) to prevent skin contact. Pregnant women and women intending to become pregnant should seek advice from knowledgeable sources before working with substances that are suspected to be reproductive toxins (1).

The relative risks to the fetus change with the various stages of pregnancy

- From 0-3 weeks: the woman may be unaware that she is pregnant, but this is a period of relative safety for the fetus.
- From 3-12 weeks: the period of greatest vulnerability for the fetus. It is during this period that the arms, legs and major organ systems are being formed.
- From 3-9 months: toxic substances encountered during this period tend to produce less severe defects, such as growth retardation or premature birth.

Highly toxic compound have the ability to cause a harmful effect after a single exposure. Acutely toxic agents can cause local toxic effects or systemic toxic effects. Among the most useful parameters for assessing the risk of acute toxicity of a chemical are its LD50 and LC50 values, the mean lethal dose or lethal concentration causing death in animals. A substance is considered highly toxic if the oral LD50 for rats is less than 50 mg/kg, or if the LC50 is less than 200 ppm for one hour. Examples of highly toxic compounds include diisopropylflurohposphate, hydrofluoric acid, hydrogen cyanide, osmium tetroxide, and tetrodotoxin.

Reproductive toxins are defined by the OSHA Laboratory Standard as substances that cause chromosomal damage (mutagens) and substances with lethal or teratogenic (malformation) effects on fetuses. Reproductive toxins may affect the conceptus at any stage of its development, from fertilization to birth, although damage is most likely during the first 8 to 10 weeks of pregnancy. Teratogens are chemical and physical agents that interfere with normal embryonic development. Teratogens differ from mutagens in that there must be a developing fetus. Reproductive toxins may produce congenital malformations or death of the fetus without inducing damage to the pregnant woman. In general, carcinogenic, mutagenic and teratogenic chemicals should be considered as hazards to reproductive health. Even though OSHA has established exposure limits of hazardous materials, a developing fetus may be adversely affected by lower doses than those considered safe for adults. Thus,

exposures must be kept as low as reasonably achievable to minimize reproductive health hazards. For example the North Carolina Regulations for Protection Against Radiation has established a radiation dose limit of 500 millirems for the conceptus during the entire gestation period.

Examples of Reproductive Toxins

Dibromochloropropane	Lead	Arsenic	Benzene
Cadmium	Ethylene glycol monomethyl (and ethyl) ethers	Antimony	Carbon disulfide
Ethylene thiourea	Ethylene oxide	Mercury compounds	Toluene
Xylene	Polychlorinated biphenols (PCBs)	Nitrous oxide	Formaldehyde
Ethylene dibromide	Ionizing radiation		

Further Reading

1. Khattak, S., Guiti, K., McMartin, K., Barrera, M., Kennedy, D., & Koren, G. (1999). Pregnancy outcome following gestational exposure to organic solvents. JAMA: the journal of the American Medical Association, 281(12), 1106-1109.
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DO NOT USE TOO MANY DISPOSABLE MICROTOME BLADES!

"I work in a hospital, there are three of us on this particular shift and we cut approx. 200 blocks, give or take a few. Our histo lab manager is telling us we should only be using one pack of blades (50 per pack) a month". Well, what do our colleagues on Histonet have to say?

That's penny pinching right there. I'd say fine, you tell me when to change blades and then put through the crap slides they get. Just avoid the calculi and staples at all costs... sometimes these number crunchers don't think.

Sorry, I tell my techs a knife is cheaper than a complaint from the pathologists so, please don't abuse. However; don't hesitate to use what you need. As the old saying goes for us "A happy Pathologist is a happy life"!! A dull is bad for sectioning and is dangerous overall. I can't imagine telling my Histologist to cut corners on knives and they know me well enough that they would think I had lost my mind!! So many other places to cut cost and not affect section quality.

From a manager's point of view, that is a poor way to try to cut expenses. It will only lead to recuts and possible loss of important tissue. For the techs to understand the necessity to conserve is important but the tech needs to use their discretion as to when a blade needs changing.

You need to tell your manager that you cannot do your job without proper tools. Only the tech cutting knows how many blades he or she needs to cut a day's work. These micro managers need to do some bench work and get a reality check. Unbelievable!

Is your manager a Tech? Sure does not sound like one. Anyone that is willing to compromise the quality of diagnostic slides to save a dollar should not be in a management position.

What total rubbish, what planet is this manager from??? Good luck.....

Certainly techs should be conscious of not wasting supplies- but I have never known this to be an issue. If the goal is to reduce costs, rationing blades has to be one of the worst and least effective means to achieve that. I can think of so many other ways to reduce waste (cost) that will have a greater positive impact on the bottom line. The re-work alone might become ridiculous.

Wow!!! It is pretty obvious this person has never worked as a HT. There are some things you can cut corners but some things you cannot. Do they want unreadable slides? I would without hesitation say to this manager the reason why this is not a good idea and would he want substandard slides if it was his tissue or someone he loves. Augh!!!! Stuff like this just makes me angry.

The price of a few blades is nothing compared to a lawsuit for intentional neglect and endangerment to patient care. Not to mention some blades are "bad" when they come out of the box. This is a very good example of "pound foolish and penny wise". Only the tech cutting knows when they need to change a blade. I encourage my techs to change blades often. It's all about getting good sections. When our slides go out to other institutes (for consults) it is a reflection on our work. Not to mention the cost for recuts (if the tissues hasn't been lost). Tech time is expensive too. One must weigh the whole situation in terms of cost.
I always tell my techs to think of each specimen as one of their family member's.

I concur with the all the responses. It seems your lab manager is not grasping the technical reality you must work under. If you are being asked to save money on blades why not try some different brands or negotiate some better pricing? That is something the lab manager can work on. Also, I would think you are doing your best optimize the use of each blade. You should be able to get 3 good cutting areas per blade before they're spent. Another consideration is having some blades for facing in only.
I'm guessing the manager is being pressured to cut cost. I would look in other areas and at other

items. Blades are of too critical importance to mess around with much.

At 200 blocks/day x 5 days/week x 4 weeks/month = 4000 blocks/month which means that the 3 of you will have to use 1 blade every 80 blocks including trimming and sectioning which is ABSOLUTELY RIDICULOUS!
The cost of blades, especially the better ones, are going up and you can save by using one blade to trim and another to make the final section, but at the rate your manager wants the quality will be compromised.
The "norm" (if there is a norm at all) is that a histotech will probably change blades every 5 to 10 blocks if the infiltration is good and there are no decals involved in the process.
Let's assume that you can hold to 1 blade every 10 blocks, that will mean that during 1 month you will use 400 blades = 8 blades boxes.
Find out how many you are actually using now and you will have an idea of your present blades usage.
Additionally dull blades not only compromise the quality of the sections but also reduce sectioning productivity. What you may be saving in blades is going to increase histotech time and total section production costs.

It is one thing to say "not a good idea" but as has been said, if you can count it you can manage it, so it's the numbers that will convince people.
Numbers help in the discussion by making the situation real. How about some numbers from everyone about how many blades they think is reasonable?

A stingy person is called an Iron Rooster (tie gong ji) in Chinese. An iron rooster won't turn loose of even one feather. The scratches that appear on slides cut with blades that should have been changed --we call them iron rooster tracks. Sort of a pun, since many people think Chinese characters look like chicken tracks. We don't worry about what the blades cost. Our clients demand good answers, and we send pictures of the lesions with our reports, so we need good results.

I run an academic lab on a very tight budget. A paper towel used to dry washed hands is used again. Out-dated dye solutions are adsorbed onto a small pile of old paper towels to save on waste disposal costs. (A quarter-pound of solid waste costs less to dispose of than 2 liters of aqueous liquid waste.) Disposable pipettes are washed and reused until the numbers wear off. I make up Vector's ImmPact SG 1.7 ml a time, store it in the fridge, and use it all week. I don't save on microtome blades. Dull blades leave holes in 4 micron sections. Sections cut with a dull blade have the annoying habit of exploding on the water bath. Dull blades tease out collagen fibers and drape them over the cells I'm trying to study. When a blade is dull it goes into the sharps box.

200 blocks per day / 3 techs = 66 blocks per day per tech.
66 blocks X 20 workdays a month = 1320 blocks a month.
1320 blocks / 50 pack of blades = 26 blocks per blade.
Reasonable or not? You decide.

I don't think it's a matter of being reasonable or not, though I think 26 blocks is a bit high.
The major issue is taking away the discretion of the tech to make a decision as to the quality of the slide they are producing.

"26 blocks per blade"

That's good. At the price we pay for blades that would be less than $11.00 per day per tech (150 blocks/day average), OR LESS THAN 20 MIN OF PAY PER TECH!! If they were limiting their blades, how many minutes would they spend trying to get good sections, or how expensive would mistakes be if something was missed due to poor sections?

However, I think some labs do try to save money this way. Our pathologists often comment how much better our output is than the consult slides we get -especially after we recut outside blocks and compare to the original slides.

There is no reason to cut quality of the slides by rationing blades. You have just made your techs job harder, which will affect TAT and now will probably get numerous recuts not to mention phone calls from the pathologist reading the slides. I tell my techs use what you need. I purchase them the accu-edge blades so for me cost is not an issue. But this is coming from a tech turned manager so my stand is a little different.

Desperately trying to salvage something positive out of this justifiably acrimonious thread...may I suggest the following blade conservation strategy that though perhaps well-known, hasn't come up in this discussion yet.
By using one blade as a trimming blade, the 'edge' on the next blade will be conserved for actual sectioning. Similarly, when cutting levels, one-half of a blade can be used for rough trimming, then the same blade pushed across into the cutting zone for the actual sectioning. Also, if during trimming a hard/calcified/stapled section is found, perform microtomy on that block last, after trying to minimize the negative effects on cutting. Perhaps by conserving blades in these and other ways, some cost-savings can be found for the penny-wise manager!

Just wondering if your manager will be seeing any of these responses?

I really appreciate everyone's constructive comments regarding my post on blade rationing. Lots of you said there are many other ways to cut costs in the lab. I would like to hear some of your suggestions so I can take them back to my manager. I'd like to give her some legitimate alternatives to her proposal. Would like to contribute to solving the problem of cutting costs.

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From 2014, we are planning a different format for our Surgical Cut Up Workshops. Instead of running the Workshops on four separate Saturdays as we have done since 2008, we are going to run them over three consecutive days (Wednesday to Friday), still to be held at the CIT in Bruce, ACT. This will mean one trip to Canberra – it will make the Workshops more feasible for interstate attendees.

The format would be similar – commencing at 9 am and running til 5 pm, with morning tea, lunch & afternoon tea provided. We envisage that the fees will remain the same at $400.00 per Workshop for those who pre-pay and $440.00 for those who pay on attendance. Payment is by cash, cheque or direct debit as we **do not** have credit card facilities.

There is no accommodation within walking distance of the CIT but you can find information on www.visitcanberra.com.au – Belconnen Premier Inn (3.3 km), Canberra Motor Inn (4.2 km) and Belconnen Way Motel (6.4 km) are three suggestions.

There would be a minimum of 6 attendees and a maximum of 12 per Workshop. We envision running 2 perhaps 3 Workshops next year, depending on interest and suggested times are April, July and October. We are seeking expressions of interest in attending on this basis and this will not be considered binding.

If you are interested, please write your name, workplace and contact details on the sheet indicating which, if any, time of year would suit you best.

Many thanks,

Penny & Anne

penelope.whippy@act.gov.au and anne.prins@anu.edu.au

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