



# CITYU VETERINARY DIAGNOSTIC LABORATORY

# MESSAGE FROM THE DIRECTOR

Welcome to the third edition of volume three of the newsletter.

In this newsletter we welcome Dr Daniela Hernandez Muguiro back to CityU VDL in Hong Kong. As COVID-19 has disrupted the plans and travel of many of us so it was for Dr Hernandez Muguiro who was unable to take up a position in Canada. Fortunately for us, she has returned to her position here and picks up where she left off with her clinical pathology expertise. Also joining our team is Dr Steve Mills. Dr Mills is based in Montreal, Canada and supports us remotely for surgical biopsy and clinical pathology. We also profile our microbiology veterinarian, Dr Vidya Bhardwaj

- Dr Fraser Hill, Anatomic Pathologist, Director of CityU VDL

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#### LEPTOSPIROSIS ALERT

As the rainy season has returned so has leptospirosis. So far four cases have been identified by PCR testing, two in Kowloon and two on Hong Kong island. The major risk factor is swimming in water ways so prevention involves keeping dogs out of standing water as much as possible at this time of the year. To confirm a diagnosis take blood and urine samples and test both by PCR for leptospires. In acute infections, cases will be leptospiraemic (1-15 days of infection), then leptospiuric (10 days plus) as they clear infection. Unfortunately the mortality rate is high so rapid and aggressive intervention is required.

#### **NEW and RETURNING PATHOLOGISTS**

**Dr Steve Mills**DVM, DACVP



Dr Mills grew up in Calgary (western Canada) and completed his veterinary degree and pathology training at the University of Saskatchewan. He subsequently entered private practice, gaining valuable experience as a small animal clinician and clinic owner. He successfully completed his ACVP board certification in 2016. For the last three years he has focused primarily on diagnostic pathology, specializing in surgical pathology and all aspects of clinical pathology. Special interests include digital pathology and comparative features of cytology and surgical pathology as they relate to improved diagnostic reporting.

Dr Daniela Hernandez Muguiro BSc Vet Med, MClin Path, DACVP



Dr Hernandez Muguiro studied veterinary science at the Universidad de Guadalajara in Mexico before undertaking clinical pathology residency studies at the National Autonomous University of Mexico and the College of Veterinary Medicine, Cornell University. Dr Hernandez Muguiro successfully completed the American board examinations in veterinary clinical pathology in 2018.

Dr Hernandez Muguiro enjoys all aspects of clinical pathology with a special interest in leukaemia, acute myeloid leukaemia, cytochemistry and flow cytometry. She is pleased to be back in Hong Kong again, providing reports and assisting veterinarians with their cases.

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#### REQUESTS FROM THE SECTIONS

# Formalin fumes can damage cytology slides

Formalin and formalin fumes can affect unstained cells on cytology slides causing cell fixation and disintegration thereby preventing uptake of stains used to stain cytology slides.

If you are submitting tissue samples to CityU VDL in formalin for histopathology and there are any other cytology or haematology slides with the case, please take the following precautions to avoid slide degradation.

- Use only leak free containers (we can supply these)
- Promptly close all containers to limit release of formalin fumes and seal tightly
- Keep any cytology and hematology slides away from any areas where formalin is used and when preparing and storing histopathology samples
- Close the specimen container tightly, seal with Parafilm or cling wrap, and enclose in a sealed sample bag
- Submit any associated cytology or haematology slides securely enclosed in a labelled slide carrier, sealed in its own separate sealed sample bag

Formalin fumes may still permeate through plastic, so it is always important to keep formalin specimens away from cytology and hematology slides, even during transport to the laboratory.

Formalin will not affect blood or fluids inside of closed blood tubes.

# How to get the best output from your aerobic cultures?

By Dr Vidya Bhardwaj- Microbiology veterinarian

Culture results should always be interpreted alongside clinical findings. For example, a non-pathogenic *Staphylococcus* grown from a superficial skin lesion is unlikely to be significant, but the same organism isolated from a deep skin wound is more likely to be significant as an opportunistic pathogen.

A good history is thus invaluable in the interpretation of microbiology diagnostic testing. Apart from the signalment, the critical information to include in the submission form are:

(i) Location of lesion

Be as specific as possible.

E.g.: Instead of "wound", describe the lesion as a "superficial skin wound" or a "surgical site over the flank" or "dog bite wound".

(ii) Method of sample collection

This is especially important for urine samples. The bacterial population expected in a voided urine sample is very different to what we expect to see in a cystocentesis sample.

(iii) Usage of antibiotics or antiseptics

Ideally, antibiotics and antiseptics should not be used within 3 days of sample collection as this hinders the growth of bacteria in the laboratory. However, if the test cannot wait, please mention the antibiotic usage in the submission form so we can adjust our methods accordingly.

It is important to remember that the presence of bacteria does not necessarily imply disease.

To fully understand the clinical significance of the bacteria cultured, supportive tests such as cytology and histology are invaluable.

## How to submit formalin fixed material to CityU VDL

#### By Dr Jeanine Sandy

Formalin fixed tissues are the backbone of histopathology. There are some basic guidelines to ensure that tissues are fixed well, with minimal tissue distortion, using safety conscious containers.

#### 1. **Formalin:tissue ratio** (Figures 1-2)

The guideline is 10 parts of formalin to 1 part tissue. This allows proper fixation of tissues. Insufficient formalin leads to only the periphery of tissues being fixed, leaving the centre unfixed.

#### 2. Wide necked containers (Figure 3)

It is best to place samples in a wide necked container. Narrow necked containers present problems as unfixed tissues can be slide into the container but fixed tissues become firm and cannot be cannot be removed. The container then has to be dismantled, which is often difficult to achieve.

#### 3. Containers with lids

Secure the lid firmly then place the container in a ziplock/leakproof bag. Avoid sticky tape around the rim as removing tape from containers can be difficult and exposes our staff to risk of injury.

#### 4. Plastic containers only.

Please do not use glass containers. Glass can break during transportation, posing a hazard for our couriers and accessioning team.

#### 5. Labelling

Ensure that the containers are labelled, so that the information matches the submission form. E.g. container labelled "1: Right liver lobe".

#### 6. **Tiny tissue samples** (Figure 4).

Samples less than 2 mm in diameter are problematic. If you are collecting small or fragile tissues, it is best to use a cassette with two foam pads, sandwiching the specimen in between, to keep the tissues intact. Tiny floating tissues in a container are otherwise difficult to process and impossible to orient.

CityU VDL can provide suitable containers, cassettes and foam pads- please call the laboratory on 3442 4849 for any supplies.



Figure 1. Excellent formalin: tissue ratio. Wide necked container.



Figure 2. Inadequate formalin.



Figure 3. Narrow necked container. Tissues are impossible to remove once fixed.



Figure 4. Tiny tissue samples can be placed into cassettes, sandwiched between pads

# **TESTING TIPS**

# Ovarian remnant syndrome- has this animal been spayed?

Diagnosing ovarian remnant syndrome in cats and dogs can involve a combination of behavioural signs, hormone measurements and histopathology.

The behavioural signs of oestrus may prompt the owner to request a consultation and further investigation.

For both cats and dogs, testing Anti-Mullerian Hormone (AMH) and progesterone on a plain tube (no additive) serum sample is a convenient test. The sample needs to be kept at 4C after collection and centrifuged within 4 hours of collection so arrange to collect the serum sample just prior to our courier arrival.

A positive AMH test is consistent with the presence of an ovarian remnant, but a negative AMH test does not rule out the presence of a remnant. The progesterone level is especially important when the AMH is negative. The cell population starts producing progesterone instead of AMH after ovulation, so high progesterone levels with an inconclusive or negative AMH is also diagnostic for an ovarian remnant.

Final confirmation is by surgical removal of suspected tissue and histopathology.

# Diagnosis of Feline Infectious Peritonitis (FIP) in cats

Ante mortem diagnosis of FIP in cats without significant effusion can be difficult. Evaluation of signalment, history, clinical signs and laboratory testing all need to be combined together for evaluation to arrive at the diagnosis.

Haematological abnormalities can include anaemia, neutrophila and thromobocytopaenia. A range of biochemical changes are possible but the most common abnormality is hyperglobulinaemia reported in 89% of cats with FIP and often hyperbilirubinaemia in cats with effusions.

In cats with effusions, examination of the fluid can reveal a viscous, straw coloured fluid forming clots or strings because of the high protein content.

Antibody tests cannot differentiate between feline coronavirus and FIP and even high antibody titres are not a specific indicator for FIP.

RT-PCR testing on blood and effusions offers testing options for diagnosis. The best test is the PCR test on an effusion. In some studies there is 100% sensitivity and specificity (compared to histopathology) when ascitic fluid was tested. The sensitivity for whole blood is lower (varies from 20-75%) while the specificity is high at 100% so a positive result is a true positive but a negative result may be a false negative. Even then, the detection of coronavirus does not confirm a diagnosis of FIP as the PCR test detects all coronavirus so the overall clinical picture and all the laboratory findings need to be considered together.

CityU VDL offers both tests but PCR on an effusion is the recommended option for confirmation.

Immunohistochemical (IHC) staining of FIP antigen within histopathological lesions is the gold standard diagnostic technique and a mainstay of diagnosis at CityU VDL with excellent results available.

Reference: Felten S and Hartmann K. Diagnosis of FIP: a review of the current literature. Viruses, 11, 1068, doi:10.3390/v11111068, 2019

### STAFF PROFILE

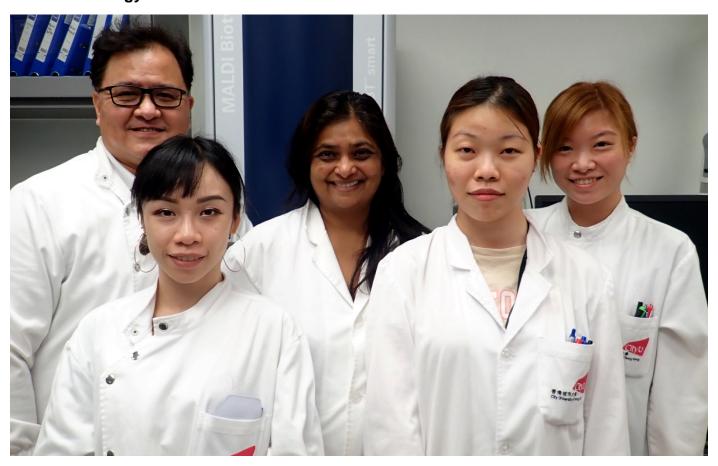
## Dr Vidya Bhardwaj – BVSc, MVS, PhD, Microbiology Veterinarian



Dr Vidya Bhardwaj graduated with a BVSc from The University of Sydney, and an MVS from The University of Melbourne before completing a PhD in microbiology at The University of Sydney. In addition, she has extensive veterinary clinical, biotechnology and continuing education experience in Australia and Hong Kong, and heads the microbiology department at the CityU VDL. She is passionate about antimicrobial resistance and responsible antibiotic use.

She is a committee member of the Hong Kong Veterinary Association and recently became an elected member of the Veterinary Surgeons Board of Hong Kong.

#### The Microbiology Team



Headed by Dr Vidya, the microbiology team comprises four registered medical laboratory technologists (MLT) including Mr Clive Chan, a Grade 1 MLT with many years of veterinary microbiology experience, assisted by Miss Joanne Wong, Miss Mirami Wong and Miss Maggie Fung.

To contact our veterinary staff, call 3442-4849 and ask to be connected, or email:

**Pathologists** 

Dr Jeanine Sandy Dr Andrew Ferguson

Email: <u>i.sandy@cityu.edu.hk</u>
Email: <u>andrew.ferguson@cityu.edu.hk</u>

Dr May Tse Dr Fraser Hill

Email: maypy.tse@cityu.edu.hk Email: fraser.hill@cityu.edu.hk

Dr Daniela Hernandez Muguiro Dr Steve Mills

Email Daniela.hernandez@cityu.edu.hk Email: infovdl@cityu.edu.hk

Microbiology Veterinarian

Dr Vidya Bhardwaj

Email: bhardwaj.vidya@cityu.edu.hk

# Contact Us

Phone: (852) 3442-4849

(For specimen pickups, consumable purchases, submission forms, specimen bags, and pricelist request)

Fax: (852) 3442-0819

Email: infovdl@cityu.edu.hk

Address:

Y1710, Yeung Kin Man Academic Building City University of Hong Kong 83 Tat Chee Avenue Kowloon, Hong Kong

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