



CITYU VETERINARY DIAGNOSTIC LABORATORY

MESSAGE FROM THE DIRECTOR

Welcome to issue 4 of the 2019 CityU VDL newsletter. This is the final newsletter for 2019 and I would like to thank you for all your interesting submissions and support of the laboratory.

In this newsletter we highlight more new technology and tests available at CityU VDL in Hong Kong including additional E cuniculi test options, exotic animal haematology availability and an antibiotic sensitivity plate reader. We also profile Dr Jeanine Sandy, one of our registered specialist anatomic pathologists.

Best wishes for the Christmas and New Year festivities ahead.

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

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What's new at CityU VDL

BIOMIC V3

Recently the BIOMIC V3 system (figure 1) was installed in microbiology to improve the efficiency and accuracy of antibiotic sensitivity testing, continuing the CityU VDL policy of utilising the best technology and equipment for diagnostic testing.

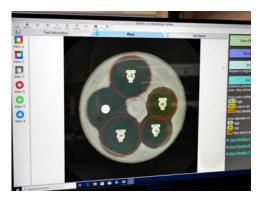


Figure 1: antibiotic sensitivity plate being analysed in the BIOMIC machine

NEW SUBMISSION FORM

Watch out for our revamped submission form including new tests, improved layout and increased information.

Encephalitozoon cuniculi tests

The test provides semi-quantitative detection of *E cuniculi* IgM or IgG antibodies in rabbit serum or plasma. Testing for both IgM and IgG antibodies provides a better indication of the infective status of an affected rabbit. A guide to using the results is given in table 1.

Table 1: user guide for interpreting IgG and IgM results

Titre result	Interpretation	Recommendations
Single negative IgG titre- healthy	No exposure, recent exposure, immature or immunosuppressed	Retest in 21 days to confirm non- exposure
Single positive IgG titre- healthy	Early infection (< 21 days), chronic infection, previous infection and recovery, reinfection	Test IgM
IgG elevated IgM negative	Chronic infection, exposure	No clinical signs,: monitor IgM (any increase treat) Clinical signs present; Treat
IgG negative IgM elevated	Acute infection (< 30 days)	Treat, retest in 4 weeks, IgG should increase
Both IgM and IgG elevated - healthy	Subclinical acute infection	Treat, retest in 4 weeks, retreat if still elevated
Both IgM and IgG elevated – clinical signs	Acute infection	Treat, retest in 4 weeks, retreat if still elevated
Both IgG and IgM negative- Clinical signs	Rules out infection	Revise diagnosis, consider otitis

References: Jeklova et al, Vet Parasitol, 170, 143-8, 2010. Mancinelli E, Vet Times, Aug, 2015.

IgM is indicative of a current active infection. IgM level will drop after 30 days post-infection.

IgG indicates long-term exposure, with levels continuing to rise steadily from 30 days post infection until they peak at 70 days post infection.

TESTING TIPS

Sample handling and submission

After collection of blood samples, the samples should be submitted to the laboratory for analysis as soon as possible (ideally within 2 hours of sample collection). If there is a delay, in vitro changes (or pre-analytical errors) occur that can alter the accuracy of the results. If the sample cannot be submitted immediately, proper handling and storage can minimize pre-analytical errors.

Plasma and serum

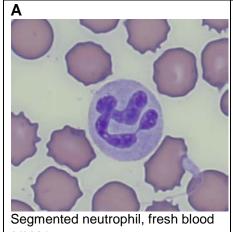
Plasma or serum for chemistry analysis should be separated from blood cells as soon as clotting has occurred, after centrifugation. The plasma or serum should then be transferred to a clean tube without any additives. Because of normal metabolic cellular processes, delayed separation of plasma or serum causes a false decrease in glucose and pH. This will also interfere with analytes such as free ionized calcium (increase) and bicarbonate (decrease). Leakage of cellular constituents such as potassium may also occur in animals with high, intra-erythrocytic, potassium concentrations (e.g. horses, cattle, and Asian dog

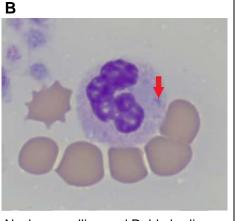
breeds). Importantly, the use of gel separator tubes does not prevent these pre-analytical errors and serum should also be collected from these tubes after centrifugation.

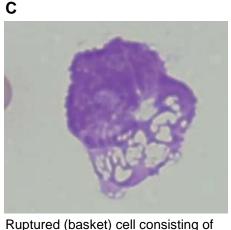
In small animals, clotting typically occurs within a few minutes of placing the blood in a tube without anticoagulant. In large animal species, clotting may take up to 30 minutes. Clotting can be enhanced by incubating the samples at 37°C for at least 10 minutes. Once harvested, the plasma or serum should be stored at 4°C until submission (most analytes are stable for up to 48 hours if kept refrigerated).

Whole blood

Whole blood for a complete blood count (CBC) can be affected by in-vitro changes associated with delayed submission and sample aging. A commonly encountered preanalytical error is in-vitro cell swelling as cells begin to take up water from the plasma. This will typically manifest as a false increase in the red blood cell mean cellular volume (MCV) and/or low mean cellular haemoglobin concentration (MCHC) and this pattern mimics regenerative anemia. Additionally, this will also cause a falsely increased hematocrit. Morphologic aging changes also typically occur. Red blood cells can become echinocytic due to ATP depletion, and this is important as it may obscure more significant red blood cell morphologies. Leukocytes, particularly neutrophils, also show cell swelling. Nuclear lobation becomes less distinct and Dohle bodies appear (Figure A & B) mimicking a left shift and toxic change, respectively. Nuclei of lymphocytes become swollen, altering chromatin features, which may preclude accurate distinction between large reactive or neoplastic lymphocytes. With time, cells will rupture (Figure C).







smear.

Nuclear swelling and Dohle bodies (arrow), 48 h old blood

Ruptured (basket) cell consisting of free nuclear material

All these aging changes may alter the accuracy of the differential and the assessment of white and red blood cell morphology. Platelets may also start to clump, leading to falsely decreased counts or high mean platelet volume.

These pre-analytical errors can be minimized or circumvented by keeping the sample cool by refrigerating or storing with an ice pack (avoiding direct contact as this can cause freezing and hemolysis of the sample) and making at least one blood smear immediately after sample collection. Blood smears should be submitted unfixed and unstained along with the anticoagulated blood. Blood smears should be transported and stored at room temperature (do not refrigerate blood smears as this can cause cell lysis).

In conclusion, there are various and cumulative pre-analytical variables that can affect the accuracy of laboratory analyses. It is important and necessary to be aware of these preanalytical variables, how to prevent and minimize them, and how to take them into account when interpreting results.

Exotic haematology tests

CityU VDL has recently developed and validated a range of tests for exotic animal haematology including birds, reptiles and amphibians. Tests available include white blood cell (WBC) manual count or estimate and differential, packed cell volume (PCV), total protein (TP) by refractometry, thrombocyte estimate count, and blood smear examination.

Collect blood into EDTA (birds) or heparin (reptiles) tubes and ideally 1-2 unstained blood smears (do not refrigerate the slides).

Minimum sample volume is 0.3 mL (0.5-1 mL for ideal results).

Results will be available within 24 hours.

STAFF PROFILE

Dr. Jeanine Sandy- BVSc, PhD, Diplomate ACVP Registered Specialist Anatomic Pathologist

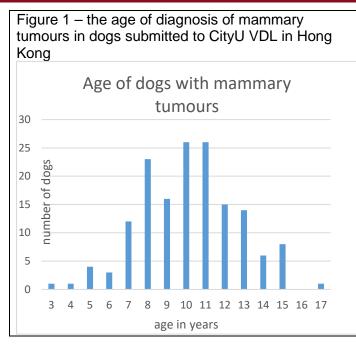


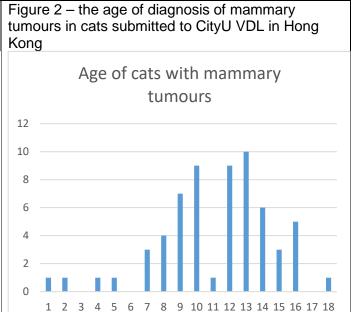
I have always wanted to be pathologist, since watching a television show about a forensic pathologist as a child. Veterinary pathology offers an exciting and challenging field of science, and one never feels as if you have "mastered" pathology, as something new and unusual comes across my microscope almost every week. Hong Kong offers a rich variety of pathologically interesting cases, and those I love most are when I get to interact with the clinicians on complex cases. Submission of CBC and biochemistry data is always useful for complex internal medicine cases;

submission of imaging results such as CT, MRI or radiographs for bone cases, and any gross photos of lesions, especially for dermatology cases are always welcomed. This additional information enables me to try and piece together what is really going on in the patient. This style of holist pathology, is what interests me the most.

Although my first love is anatomic pathology, dealing with tissue biopsies and post mortems, I have always had a love of clinical pathology. I have spent many years training in clinical pathology, and my ability with both anatomic and clinical pathology, as well as my previous experience at being a practicing veterinarian, helps me make sense of what I am looking at down the microscope. I am a great believer that the pathology should match the animal, so if you receive a diagnosis that does not fit with the patient clinically then never be afraid to pick up the phone or send an email to chat about the case.

Currently at CityU VDL, I am conducting research on canine and feline mammary tumours. If your clinic has submitted a mammary tumour to us in the past, then look forward to a phone call in the near future for further information about the patient. I am looking at various aspects of canine and feline mammary disease including age of dogs and cats who develop mammary tumours shown in the graphs below (Figures 1 and 2).





WORLD ANTIBIOTICS AWARENESS WEEK

CityU VDL participated in the World Health Organisation Antibiotics Awareness week in November. In conjunction with the Jockey Club College of Veterinary Medicine and Life Sciences, CityU VDL highlighted the key role we all play in responsible antibiotic stewardship.

Laboratory systems and professionals are key to identifying, mapping, quantifying and communicating antimicrobial resistance (AMR). CityU VDL is actively involved in global ventures aimed at tackling AMR, such as participation in World Antibiotic awareness Week. Dr Vidya Bhardwaj, Microbiology Veterinarian, has been invited to sit on the panel of mentors at an upcoming AMR event in Europe, Hacking AMR 2019. This event, organised by the Joint Programming Initiative on Antimicrobial Resistance, aims to bring together scientists, designers, patients, developers, innovators, students and entrepreneurs to collaborate, dream up & create solutions to real-world problems to improve human, animal and environmental health and well-being with regards to Antimicrobial Resistance.

Two of the images from the week are reproduced below:













INTERESTING CASE

Feline Lung Digit Syndrome

Dr. May Tse

A 12-year-old female spayed Domestic Short Hair cat presented for veterinary attention because of forelimb lameness. On physical examination, the left elbow was swollen and firm. Over the next six weeks, the lesion progressed significantly to involve more of the limb and a radiograph of the thorax revealed an opaque, ill-defined area in the caudal dorsal lung lobe. The decision was made to amputate the left foreleg and submit it to CityU VDL for gross examination and histopathology.

After cross-sectioning the limb and lesion with the band saw, representative sections were decalcified before cutting and staining. On microscopic examination, the periosteum of the distal femur, the joint capsule, proximal radius and bone marrow were expanded and replaced by an unencapsulated, poorly demarcated, infiltrative, moderately cellular neoplasm (figure 1). Neoplastic polygonal to cuboidal cells formed tubules, ducts, and islands on a variable amount of loose to dense fibrovascular stroma. The neoplasm was surrounded by massive amounts of new bone formation and lesser amounts of desmoplastic fibrous tissue.

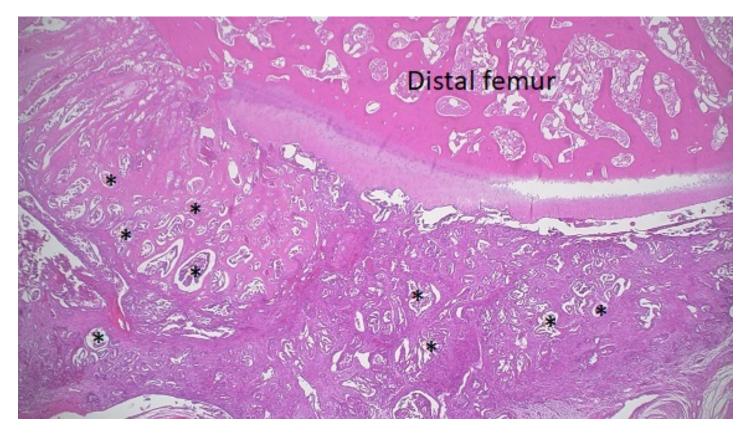


Figure 1: image of the lesion within the bone characterised by tubules, ducts and islands of neoplastic cells (*) HE staining x 100

Considering the clinical findings and the histopathologic changes, a primary lung adenocarcinoma metastatic to the left elbow is highly likely in this case, and would be consistent with the feline lung-digit syndrome.

Feline lung-digit syndrome describes an unusual pattern of metastasis seen with various types of primary lung tumours, particularly bronchial and bronchioalveolar adenocarcinomas. Tumour metastases are found at atypical sites, notably the distal phalanges of the limbs, but multiple-digit and multiple-limb involvement is also common. Other sites of metastases include the skin, eyes, skeletal muscle and bone, as well as multiple thoracic and abdominal organs. The metastatic lesions are thought to arise from direct arterial embolization of tumour cells. The prognosis is generally grave for cats with this syndrome, with a mean survival time of only 58 days after presentation.

Reference:

- 1. Goldfinch N and Argyle D. 2012. Feline Lung-digit syndrome unusual metastatic patterns of primary lung tumors in cats. JFMS: 14, 202-208.
- 2. Caswell JL and Williams KJ. 2016. Respiratory system. In: Maxie MG, ed. Jubb, Kennedy, and Palmer's Pathology of Domestic animals. Vol 2. 6th ed. St. Louis, MO: Elsevier, 2016: 497.

CityU VDL thanks the submitting veterinarian for the interesting case.

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