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Diagnostic Update



American College of Veterinary Pathologists Board Certification - AVC Continues Great Track Record !

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By Shelley Burton, Veterinary Clinical Pathologist and Andrea Bourque, Veterinary Anatomic Pathologist

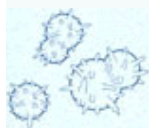
When calling our laboratory to discuss a case, do you ever wonder how the person on the other end of the phone got to be a pathologist? With the field of veterinary pathology rapidly growing and evolving, choosing this area as a specialty can be a very rewarding but daunting prospect! In this article, we hope to give you a bit of background on the training required to become a board certified pathologist and what challenges that person on the phone had to face.

The American College of Veterinary Pathologists (ACVP) was established in 1949 in Washington, D.C. Its goal was to set high standards for veterinary pathology, similar to that already established for human pathology. Through the efforts of the ACVP and the stringent examinations required for board certification, the hope was that the specialty would grow and that highly trained people would become involved in teaching, research and diagnostic pathology. A nucleus of approximately 50 experienced American veterinary pathologists founded the ACVP; it now includes approximately 1500 members from 17 countries.

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Veterinarians wishing to become ACVP boarded pathologists need to complete 3 years of training under the supervision of one or more ACVP board certified pathologists. Approximately one year of intense study is required to prepare for this examination, which is given each September over a 3 day period in Ames, Iowa. Candidates must review many images and glass slides, study the last 5 years of pathology articles in many journals and pore over several key textbooks. There are several sections to the examination, with all candidates writing the general pathology section covering topics such as carcinogenesis and inflammation. Clinical pathology candidates must complete essay, multiple choice and glass slide examinations



covering hematology, cytology, surgical pathology, biochemistry and urinalysis. Anatomic pathology candidates must complete multiple choice questions on clinical and toxicologic pathology as well as wildlife, zoo, exotic, domestic and laboratory animal diseases. A separate portion involves writing histologic descriptions and making diagnoses on 20 glass slide cases over a 4 hour period, while another is dedicated to viewing 100 images and answering questions regarding diagnoses, pathogenesis and etiology.

This examination is difficult and pass rates are low. In the last 3 years, the pass rate for 1st time anatomic pathology candidates varied from 26 - 53%. The pass rate for 1st time clinical pathology candidates is typically slightly higher. If a candidate passes 2 or 3 sections, they can return to write the remaining ones the following year. If they pass 1 or no sections, they must return and write all 4 sections again. While the process of studying and writing the examination is all-consuming and very stressful, it is acknowledged (grudgingly by some!) that this process can't help but strengthen pathology skills. It is indeed a badge of honor to be able to put the board certification letters (Diplomate ACVP or DACVP) behind one's name.

A group composed of 20 ACVP boarded pathologists make up the ACVP Examination Committee. Members spend many hours writing and reviewing questions over a 9 month period, then arrive in Iowa to proctor and mark the examination. Marks are finalized and letters are mailed to candidates directly from Ames, who generally find out their results within one week of writing. The Atlantic Veterinary College (AVC) has the distinction of now having 2 pathologists who have served on this committee - Dr. Lisa Miller (Anatomic Pathology) in 2006 and Dr. Shelley Burton (Clinical Pathology) in 2007.

The pathology group at the AVC places a high priority on training residents and has a terrific track record in producing successful candidates.

Pathologists currently at the AVC with board certification include Drs. Pierre Yves Daoust, Lisa Miller, Gerald Johnson, Barbara Horney, Paul Hanna, Shelley Burton, Andrea Battison, Sandra McConkey, Andrea Bourque, Les Gabor, Cora Gilroy and Maria Forzan. Over the last 20 years, training programs at AVC have produced 12 ACVP board certified pathologists!

Interpreting Low White Blood Cell Counts

by Barb Horney and Shelley Burton, Veterinary Clinical Pathologists

It can be challenging to interpret a low white blood cell count, but this article will provide some guidelines. First, it is critical that the white blood cell differential counts are interpreted based on the absolute counts ($\times 10^9/L$) compared to reference intervals, not on percentages! It is only the absolute counts that reflect the true number of the cell types in circulation.

Since there are normally few eosinophils, monocytes and basophils present in blood, decreases in these cell types are unlikely to result in overall leukopenias. Neutrophils and lymphocytes are therefore the cells to focus on. The current AVC Diagnostic Services absolute count reference intervals, with our personal grey zone ranges (explained later) are provided on the top of the next page.

How new technology can affect results

The new laser technology for white cell counts is likely more accurate for values at the lower end than the older impedance methods and lower counts have been noticed. This means that patients that have a mild decrease in total WBC or segmented neutrophil counts (grey zones above) compared to previously established reference intervals may just reflect normal population variation - which overlaps with values that may occur in ill animals.

Species	WBC x 10 ⁹ /L	Segmented Neutrophils x 10 ⁹ /L	Lymphocytes x 10 ⁹ /L
Feline	5.5 - 19.5 <i>Grey zone:</i> 4.5 - 5.5	2.5 - 12.5 <i>Grey zone:</i> 2.0 - 2.5	1.5 - 7.0
Canine	5.4 - 14.3 <i>Grey zone:</i> 5.0 - 5.4	2.8 - 10.1 <i>Grey zone:</i> 2.5 - 2.8	1.0 - 4.8
Equine	5.5 - 12.5 <i>Grey zone:</i> 5.0 - 5.5	2.7 - 6.7 <i>Grey zone:</i> 2.5 - 2.7	1.5 - 7.5
Bovine	5.5 - 12.5 <i>Grey zone:</i> 5.0 - 5.5	0.6 - 4.0 <i>Grey zone:</i> 0.4 - 0.6	2.5 - 7.5

Our laboratory is in the process of revising all our reference intervals which will help clarify this, but this will take time. In the meantime:

When the WBC is low:

Step 1. Check for technical comments on sample quality. Do the technologists note that there is a clot in the sample or that cell clumping was noticed on the smear? If so, the reported count may be lower than what is truly in the animal's bloodstream. This is particularly true for cats, especially if the sample is over 12 hours old. If you have kept a well made blood smear for yourself as well as submitting unstained smears and the lavender-topped tube of blood to the laboratory, you can estimate the white blood cell count*, as is done by our technologists.

Step 2. If it is a canine sample, check the breed of dog: Healthy Greyhounds and Belgian Tervurens can have slightly lower WBC, segmented neutrophil and lymphocyte numbers than other breeds.

Step 3. Evaluate the differential:

If lymphopenia is the main change seen and segmented neutrophils are within the reference interval or mildly increased, stress is most likely. A mild monocytosis can also be seen in a classic stress response but we don't expect it in every case.

If the segmented neutrophils are low, is a left shift also present?

If there is no left shift: The patient could have very acute inflammation before a left shift becomes apparent; other possibilities are decreased marrow production or peripheral destruction. If it is a very mild neutropenia (grey zones above), normal patient variation could also be the reason. If there are no clues clinically, it is best to do another CBC in 1-2 days to see if the neutropenia persists or resolves. If it disappears, we can conclude that it was either artifact or a transient change on the first sample. If the neutropenia persists at a mild level, no left shift is seen and the patient is clinically normal, it may just be normal variability but further monitoring is wise. If the degree of neutropenia worsens, no left shift is seen and the patient status is not normal, bone marrow sampling may be indicated.

If there is a left shift: Inflammation is present in the body and the increased demand for neutrophils can be identified as the cause of the neutropenia. The challenge is then to identify the site clinically and to treat appropriately.

* Estimating the WBC from a blood smear:

The smear should be made at the time of collection from well mixed blood. It should have a good monolayer (area where the cells are not overlapping). Using 40x objective power, count the number of leukocytes in 10 fields, take the average and multiply by 2.5. Example: 40 cells total in 10 fields = 4.0 cells average: WBC = 10 x 10⁹/L.

New Hematology Analyzer - First North American Site

By Linda Ruschkowski, Veterinary Technologist

Diagnostic Services has a new hematology analyzer made by Sysmex. The instrument is called the XTV-2000i and has a limited veterinary software package. Diagnostic Services was the first location in North America to use the Sysmex in a veterinary setting. We were given the unique opportunity to work one-on-one with the distributors of Sysmex (Roche Diagnostics) to evaluate and adjust the instrument for the veterinary market and our own laboratory.

The Sysmex XTV-2000i is a fully automated analyzer capable of performing numerical analysis of red blood cells, white blood cells, and platelets as well as differentiating 5 white blood cell types in humans. Using flow cytometry via a semiconductor laser, the Sysmex



generates scattergrams in various combinations of scattered light to derive these counts. Automated reticulocyte counts are also achieved, which identify higher numbers of young erythrocytes in a rapid and accurate fashion. Many of you will be familiar with the old Coulter counter principle. Sysmex improves upon this technology by hydrodynamic focusing of cells in a sheath fluid which, when passed through an aperture, interrupts the electric current to register as cells counted. The sulfyl laurate solution for hemoglobin measurement is an improvement over older techniques; it is highly accurate and safe since no cyanide products are utilized.

Evaluating the new Sysmex analyzer for veterinary patients has been both challenging

and interesting. As you can see, Diagnostic Services continually strives to bring our clients the most accurate and up-to-date technology available.

Phenobarbital and KBr treatment of Atlantic Canadian dogs

By Cynthia Gaskill, Veterinary Clinical Pharmacologist

Currently the two most common drugs used to treat canine epilepsy are phenobarbital (PB) and potassium bromide (KBr). We recently compared PB and KBr monotherapies to see if one was superior in terms of safety and efficacy. Veterinarians throughout Atlantic Canada participated and we thank them for this. Here are the highlights:

The study was a year-long clinical trial involving 62 client-owned pet dogs with presumed idiopathic epilepsy. Dogs were randomly assigned to a treatment group: 30 dogs received PB, and 32 dogs received KBr. Data was collected prior to treatment and again 1, 4 and 12 months later. This data included results from physical examinations, medical and seizure histories, serum biochemical analyses, CBC analyses and serum drug concentrations. Additional data was collected if the dog became ill, had major side effects, or had poor seizure control at any time.

KBr monotherapy was associated with worse seizure control and a higher incidence of adverse effects. Adverse effects common to both PB and KBr groups included polyuria and polydipsia, polyphagia, anorexia, hyperactivity, lethargy, aggression, and skin problems. KBr treated dogs had a higher incidence of several of these compared to PB treated dogs. Additionally, KBr treated dogs had a higher incidence of vomiting and inappropriate defecation. No dogs in either group developed liver disease, but mean serum ALP activity was higher in the PB group, likely due to enzyme induction. During the study, 3 KBr treated dogs developed significantly increased amylase and lipase activities; one of the dogs developed fulminant clinical pancreatitis.

None of the PB treated dogs developed pancreatitis. Significantly more dogs in the KBr group were removed from the study and switched to alternative therapies due to poor seizure control or intolerable adverse effects (1 dog in the PB group; 7 dogs in the KBr group).

Results of this study indicate that PB is superior to KBr as the drug of first choice for the treatment of canine epilepsy. Phenobarbital usage was associated with better seizure control, fewer adverse effects, faster therapeutic response, and more predictable serum drug concentrations based on dosage than was KBr over the first year of treatment. In a few circumstances, KBr might be a better first choice: 1) if the dog has liver disease, as KBr is not metabolized by the liver and is not thought to cause liver damage, 2) if owner compliance is expected to be low, as KBr has a very long elimination half-life and very little serum drug fluctuation occurs if the owner misses occasional doses, and 3) KBr is generally cheaper than PB for treatment of very large dogs. As always, monitoring of serum levels of either drug is appropriate.

Client Survey Results

By Les Gabor, Veterinary Anatomic Pathologist

During early 2007, Diagnostic Services sent a questionnaire to all of our Atlantic Canadian clients. We received results on close to 50% of the surveys sent out. The staff at Diagnostic Services would like to thank all of our clientele for taking the time to give us the feedback that we require.

There are significant market forces present in Canada, and it is unrealistic for Diagnostic Services to match some elements offered by multinational corporations - such as extremely rapid turnaround time or costing. However, Diagnostic Services aims to offer unparalleled personal service - detailed, in depth, and up-to-date comments and diagnoses which reflect the academic depth of the AVC.

We were pleased to note that between 97-100% of customers were highly satisfied with a number of our services, most strikingly cytology and clinical pathology. Our assessment is that the turnaround time in more labour intensive tests, such as histology, need to be improved, and we are striving to change this. In short, we acknowledge that a number of elements of the service do need improvement, and Diagnostic Services is committed to making the changes necessary. We will continue, however, to strive to maintain a personal connection with clinicians. Thank you for your support, and please, continue to contact us by phone to discuss cases or issues at any time.

Staff Focus

Hats off to Bob Maloney!!

By Linda Ruschkowski, Veterinary Technologist

Bob Maloney, the parasitology technologist at the AVC Diagnostic Services, will be retiring in the summer of 2008. Bob has had a long and colorful career and he will be greatly missed.

Bob has been working in laboratories since 1970 when he finished his training with the Canadian

Armed Forces.

In 1972, he was selected by the Department of Health and Welfare to work with a medical team in Kampala, Uganda. It was during his tour of duty in



Africa that Bob developed a keen interest in parasitology and decided to make it his specialty. Upon his return to Canada, he continued to work in various laboratory disciplines, but his main focus became parasitology. In 1980, he was sent to the Center for Disease Control in Atlanta, Georgia, for advanced training in blood and intestinal parasites.

For 4 years after his return from Atlanta, Bob taught parasitology to technologists as well as military and civilian physicians. In 1988, Bob joined Diagnostic Services at the AVC. Since then, he has furthered his knowledge of all types of endo and ecto parasites in the veterinary world.

Bob has always been physically active and we often see him running at lunch time on a nearby trail. He frequently rides his bike to work and cycles on weekends with his wife, Pam, who works in our Chemistry department. For many years, he has organized the AVC recreational curling teams. Bob has a dry sense of humor and probably has the world's largest collection of parasite jokes! When he retires, Bob plans to continue his physical fitness regime, become involved in volunteer work and take exotic cycling trips.

Bob's expertise will be greatly missed; he is considered an expert in parasitology and he will not be easily replaced. Throughout his career, his professionalism has been highly regarded. He is an excellent teacher and his dedication to the AVC veterinary students is unrivaled, as he often comes in after hours and on weekends to impart his knowledge to them through microscope sessions. Many thanks to Bob Maloney, our "go-to guy" in parasitology. When he retires, we will miss him and we thank him for his dedication to Diagnostic Services. Good luck, Bob!

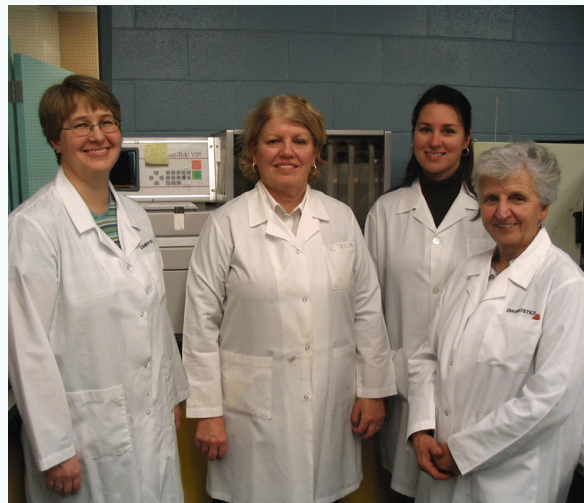
Spotlight on Histology

By Linda Ruschkowski, Veterinary Technologist

In this issue, we would like to highlight the Histology section of Diagnostic Services. Charge technologist Ramona Taylor is a lively gal who loves antiquing, movies and singing barbershop music. Her colleagues include Dianne O'Connor, Sarah Bernard and Karen Blackburn.

Dianne, a grandmother of 3, enjoys keeping fit and travelling. Sarah works part-time in Histology and Bacteriology and loves family life with her daughter, Cara, and husband, Kimble. Karen, our most devoted animal lover, provides a home for 8 cats and 2 dogs.

Left to right: Karen, Ramona, Sarah and Dianne



Our pathologists greatly appreciate the expertise and quick turn around time provided by these dedicated people!

Reader Feedback: The *Diagnostic Update* group invites comments or suggestions for future topics in the newsletter. Please submit your comments to *Dr. Cora Gilroy* (cgilroy@upeu.ca), Diagnostic Services, Atlantic Veterinary College, UPEI, Charlottetown, PE, C1A 4P3 and they will be forwarded appropriately.