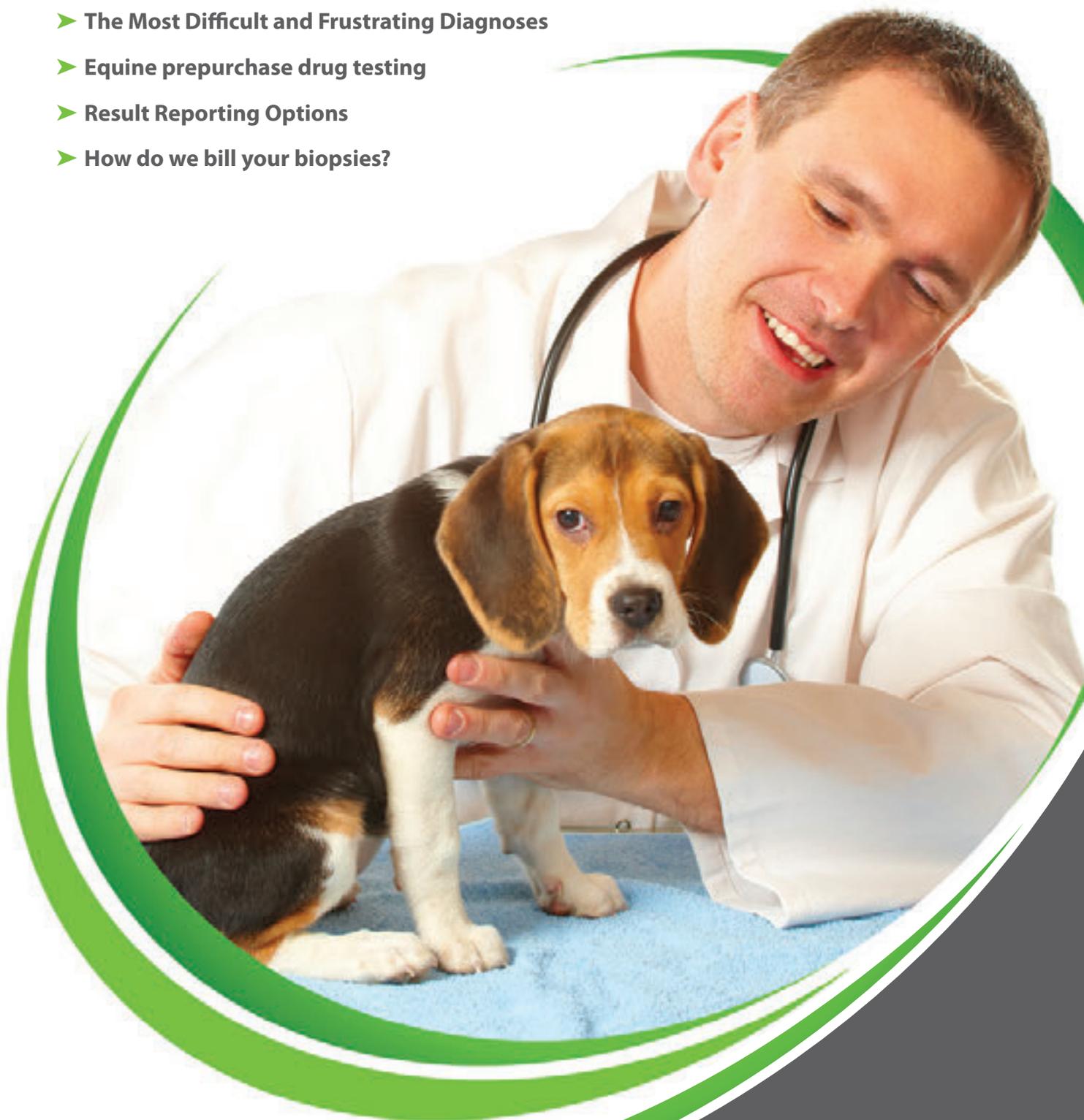


Partners in Practice

- **Adrenals:** Part 3: Routine Clinical Pathology
- **The Most Difficult and Frustrating Diagnoses**
- **Equine prepurchase drug testing**
- **Result Reporting Options**
- **How do we bill your biopsies?**



Welcome...

Welcome to the Autumn 2012 edition of the Vetnostics newsletter.

I hope you find the content interesting, topical and informative.

As always, please contact me on 02 9005 7272 or by email (doug.hayward@vetnostics.com.au) if you have any requests/ideas for future newsletters, or any other queries for that matter.

ADRENALS: What you won't find in a textbook

Dr Sue Foster BVSc, MVetClinStud, FACVSc **Vetnostics Small Animal Medical Consultant**

PART 3: ROUTINE CLINICAL PATHOLOGY

Hyperadrenocorticism (hyperA)

HAEMATOLOGY

1. Lymphopenia and absolute eosinopenia are the most frequently cited haematologic abnormalities (approximately 80% of dogs according to Feldman and Nelson 2004). However, they are not always present. Lymphocyte counts in hyperA dogs at Vetnostics quite often seem to be normal which would be consistent with the finding in one study that only 14% dogs with hyperA had lymphopenia (Peterson, 1984). Eosinopenia seems far more common than lymphopenia (consistent with the figure of 84% by Peterson, 1984). However, eosinophils can be normal to increased if there is a concurrent eosinophilic process (uncommon but occasionally seen).
2. Nucleated red cells are seen quite commonly (usually in low numbers, 1-3/100 WBC). Whilst this number would be classed as normal, it is more common in dogs with hyperA than in other dogs of similar age with no reported history or signs typical of hyperA.

BIOCHEMISTRY

1. Increased alkaline phosphatase (ALP) is widely cited as the most common routine laboratory abnormality but it is not always increased. A normal ALP does not rule out hyperA.
2. Increased ALP is largely due to induction of a specific ALP isoenzyme by glucocorticoids. Although this isoenzyme can be evaluated, it has been shown that an increase can be caused

by a variety of disorders and is not specific for hyperadrenocorticism (Solter et al 1993). The steroid-induced isoenzyme cannot be used to distinguish spontaneous or iatrogenic hyperA from liver disease or diabetes mellitus for example. It is often stated that 70-100% of the increase in hyperA dogs will be due to this isoenzyme but this is certainly not always the case with either iatrogenic or spontaneous hyperA (Feldman and Nelson 2004).

3. Lipaemia is very common in hyperA dogs. Most of the old studies that reported biochemistry findings did not report the frequency of hypertriglyceridaemia in hyperA dogs as veterinary laboratories have not typically run triglyceride concentrations. The triglyceride increases in hyperA dogs are often quite marked and the serum/plasma consequently often has a strawberry milkshake appearance, even on a fasted sample. HyperA should always be on the DDx list for fasting hypertriglyceridaemia in an otherwise well dog (or cat!). This also holds for breeds such as Miniature Schnauzers, not of all which will have familial hypertriglyceridaemia; Miniature Schnauzers do get hyperA (see Adrenal News 1).
4. Mild hyperglycaemia is reported as occurring in 45-60% of hyperA dogs (Peterson 1984, Feldman and Nelson 2004) but this would not be the case in my own patients or Vetnostics' cases. Whilst hyperglycaemia can occur, it would seem to occur at a much lower rate in our patients.

5. Alanine aminotransferase (ALT) is commonly increased but again, not necessarily. It is not usually increased to the same extent as ALP.
6. Urea concentration may be decreased due to polydipsia.
7. Hypokalaemia and hypernatraemia may occasionally be seen and are probably more common in dogs with adrenal tumours as the cause of their hyperA (presumably excessive mineralocorticoid secretion).
8. Bile acids test results may be increased in dogs with hyperA (Center et al 1985).
9. Serum lipase may be increased by exogenous corticosteroids (dexamethasone) thus possibly by endogenous glucocorticoids also. Although it would not be routinely measured in hyperA cases, this must be borne in mind when analysing lipase in potential pancreatitis cases. The effect of glucocorticoid excess on cPLI is currently unknown.

URINE

1. Urine in dogs with hyperA is usually isosthenuric or hyposthenuric and in one old study (Meijer 1980), 80-85% of dogs had a USG <1.013. However, as we often pick up hyperA much earlier (ie before they become textbook classics) that figure is probably an overestimate in modern medicine. Not all hyperA dogs have polydipsia/polyuria as presenting signs; only 82% of 300 hyperA dogs had PU/PD in one report (Peterson 1984). In addition, many dogs can concentrate their urine reasonably after being in a hospital (see Adrenal News 2) so the urine concentration measured at any one moment, could be hyposthenuric, isosthenuric or concentrated.
2. Urinary tract infection (UTI) reportedly occurs in 40-50% of hyperA dogs (Feldman and Nelson 2004). Again, I think it would be interesting to review that figure. Frequency of UTI has probably decreased with earlier detection of the disease but as urine culture is not routine, this is impossible to assess.
3. It is important to remember that a) hyperA dogs with UTIs may not have any pyuria or haematuria (presumably because of the anti-inflammatory effect of excess glucocorticoids) and b) routine sediment examination on a wet preparation may



fail to detect white cells and bacteria in dilute or weakly concentrated urine. A stained, air-dried smear will increase detection of both white cells and bacteria but culture is usually required to detect UTIs in dogs with hyperA. UTIs may well be undiagnosed in hyperA dogs.

Hypoadrenocorticism (hypoA)

HAEMATOLOGY

1. Lack of a stress leucogram in a sick dog can be an indication of hypoA and may be the only clinicopathologic abnormality in dogs with glucocorticoid deficient (atypical) hypoA. When I ask vets about the leucogram in suspected hypoA cases, the common response is "Everything is normal". Remember, a normal leucogram can be quite abnormal for a collapsed dog and each count should be assessed with respect to the dog.
2. Lymphocytosis is not always present.
3. Lymphopenia is a good "rule-out" for hypoA. I have never seen a hypoA case with lymphopenia however, a recent study on lymphocyte counts in dogs with hypoA (Seth et al 2011) did identify a few low lymphocyte counts. In this study, 100% of hypoA dogs had a lymphocyte count >0.75x10⁹/L and 92% had lymphocyte counts >1.00x10⁹/L.
4. Eosinophilia is also not always present.
5. I have never yet seen a hypoA dog with an eosinophil count of 0, thus an eosinophil count of 0 would be a good "rule-out" for hypoA. Let me know if you have a hypoA dog with an eosinophil count of 0!

ADRENALS: Part 3 continued

BIOCHEMISTRY

1. A Na:K ratio of <27 is NOT diagnostic for hypoA. In one study only 24% of dogs with Na:K ratio <24 had hypoA and 41% of dogs had renal disease (Roth and Tyler 1999). It is worth noting that all dogs in that study with Na:K ratios <15 had hypoA. Other diseases causing low Na:K ratios include whipworm (and other gastrointestinal diseases), renal disease, pancreatitis, diabetes mellitus, pyometra and body cavity effusions. Another larger, more recent study showed that whilst hypoA was the most common cause of a Na:K ratio <27 , only 16.7% of dogs with Na:K ratio <27 had hypoA (and that was after the cases with suspected EDTA contamination had been removed from the sample population).

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GENERAL

1. Combining the Na:K ratio with the lymphocyte count is diagnostically superior as a screening test than either alone (Seth et al 2011).
2. A faecal flotation test should be performed in all dogs with a low Na:K ratio. Occasionally dogs have both whipworm and hypoA!!
3. A manual differential white cell count (of at least 100 cells) is mandatory in order to detect hypoA leucocyte "patterns" with any accuracy. Most of the in-house analysers would not perform with high enough accuracy (Papasouliotis et al 1999, Bienzle et al 2000, Papasouliotis et al 2006) to detect the subtle "patterns" of hypoA

Result Reporting Options

After generating results for the various submissions received, it is obviously imperative to get the results out to you as efficiently as possible. With this in mind, we have multiple options available for reporting of Vetnostics laboratory results (please see below).

- Online results with optional email alerts in real time via Path-Way online results system. Results viewed on Path-Way can be saved as a colour PDF or printed. Register online at www.path-way.com.au or email laverty.edi@laverty.com.au for further details.
- Direct downloads via secure download direct to patient management systems including: RXWorks, Vetcare, Vetlink SQL and Vision VPM (Vision VPM currently in development).
- Other delivery options such as fax or direct email are available on request.



Please contact a veterinary pathologist or the appropriate IT department (laverty.edi@laverty.com.au) if you would like to discuss these options further.

What is your diagnosis?

In this edition of 'What is your diagnosis?', we focus on a blood film from an 8-year old male neutered cat presenting with inappetance, dehydration and pale mucous membranes.

Amongst other changes, the FBC revealed a markedly severe, non-regenerative anaemia. In the image to the right (figure 1), what abnormalities, apart from crenation and anisocytosis, can you see associated with the erythrocytes and what is unusual about the finding in this case particularly?

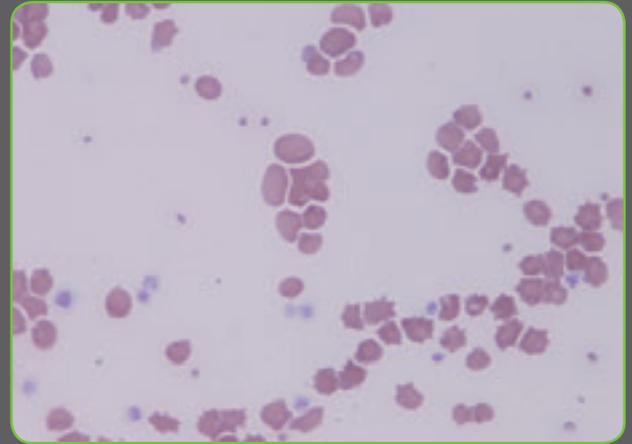


Figure 1: Wright's stained blood film (x100 oil)

Answer...

The vast majority of the erythrocytes revealed the presence of Heinz bodies, pale-staining round structures identified toward the periphery (predominantly) of the erythrocytes (see figures 2 and 3). These reflect the presence of oxidised, precipitated haemoglobin, identified in cats in increased numbers (increased where greater than 5% of erythrocytes involved) with conditions such as acetaminophen toxicity, onion/garlic toxicity,

lymphoma, diabetes mellitus and hyperthyroidism. The unusual finding in this case is the very high numbers of Heinz bodies present (>90% of the erythrocytes) and yet the anaemia being non-regenerative (Heinz body haemolytic anaemias are expected to be regenerative). The findings in this case may perhaps have reflected a prerenal phase of a haemolytic anaemia.

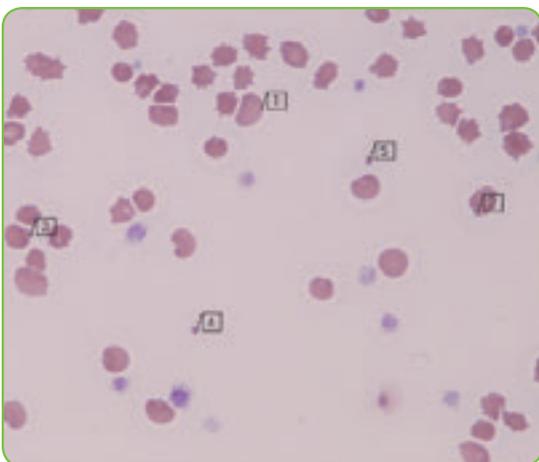


Figure 2: Same case as in figure 1. Number 1 indicates a normal erythrocyte, numbers 2 and 3 indicate Heinz bodies within erythrocytes and numbers 4 and 5 indicate free Heinz bodies within plasma. (Wright's stain, x100 oil)

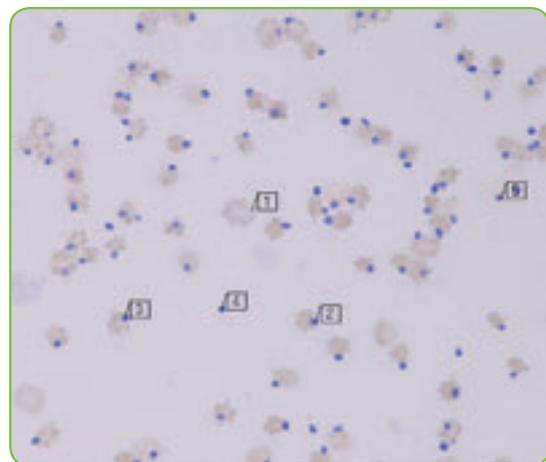


Figure 3: As for figure 2. (Methyl violet stain, x100 oil)

Equine prepurchase drug testing



Due to increased demand, we have now increased the range of tests available for prepurchase drug testing in horses.

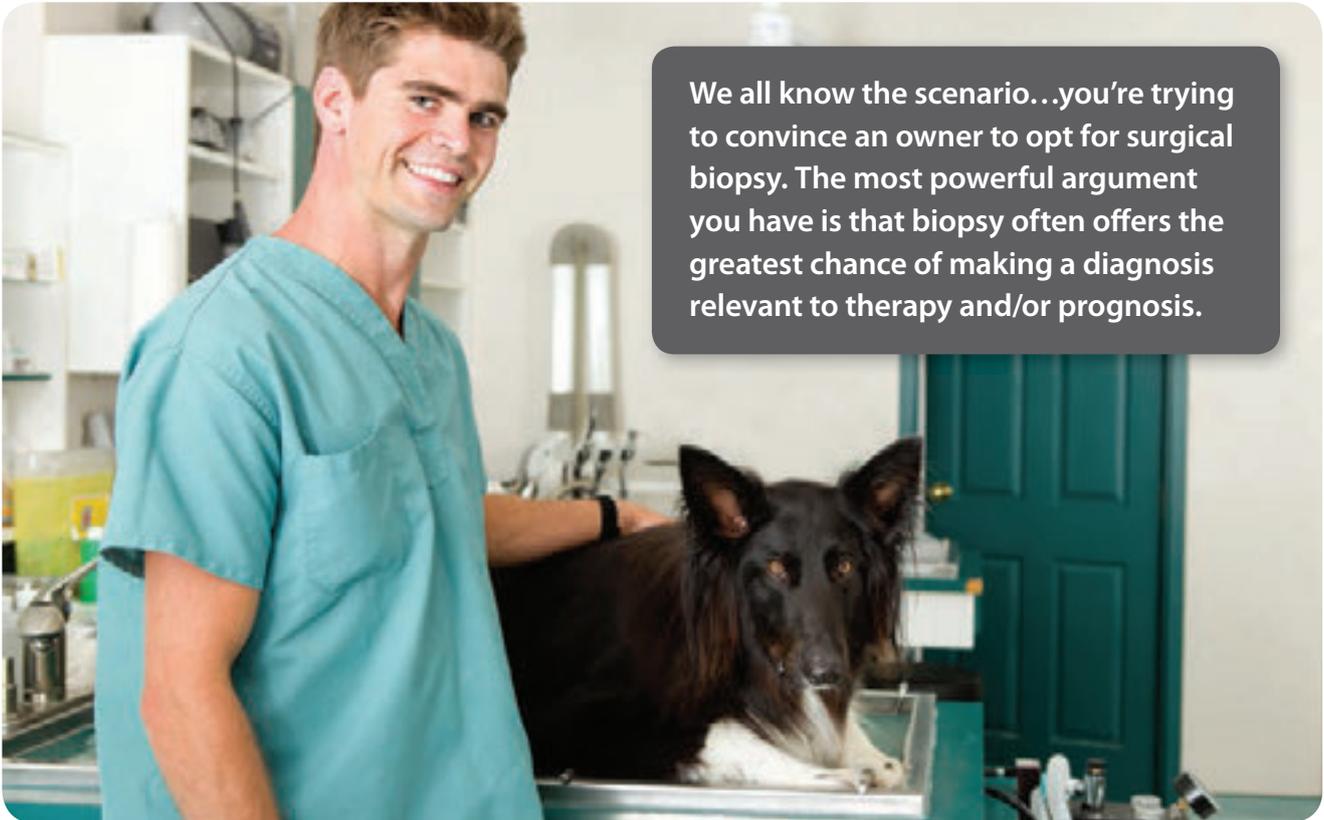
In addition to non-steroidal anti-inflammatory (NSAID) drug testing, we are now able to offer testing for sedatives/tranquilisers and anabolic steroids on a routine basis. All testing is performed by Gas chromatography–mass spectrometry (GC-MS).

Either blood (serum or heparin plasma) or urine is adequate for NSAID and sedative/tranquiliser testing. Anabolic steroids can currently only be tested for on

urine samples (minimal urine sample volume 50ml). Routine turnaround time is approximately 1 week. The cost for each broad group if requested individually is \$230 excl GST. Combination of NSAID and sedative/tranquiliser testing is available for \$400 excl GST. Prepurchase drug testing kits for submission of samples are available at an additional cost if required (please contact a vet pathologist).

The most difficult and frustrating diagnoses

Dr David Taylor BVSc, Dip ACVP Veterinary pathologist



We all know the scenario...you're trying to convince an owner to opt for surgical biopsy. The most powerful argument you have is that biopsy often offers the greatest chance of making a diagnosis relevant to therapy and/or prognosis.

It probably rates as one of life's more embarrassing moments, therefore, when the report comes back with very inconclusive findings. You ask yourself, 'Is the pathologist incompetent?' Probably not! To support my case I thought it might be useful to review in this and subsequent Newsletters some of the conditions for which biopsy interpretation has greater-than-usual likelihood of being inconclusive. As you will see, the reasons vary a great deal, but nonetheless most pathologists would agree with the following difficult and frustrating diagnoses.

Splenic haemangiosarcoma. I find it particularly frustrating when I am unable to confirm a diagnosis of suspected splenic haemangiosarcoma. It is, to many pathologists, the most difficult and frustrating diagnosis in all of surgical pathology because of the "needle in a haystack" phenomenon. The problem is that the neoplastic endothelial cells frequently represent

only a small percentage of the large mass, the majority of which is haematoma resulting from the extreme fragility of the neoplastic vascular channels. In some specimens, good fortune results in an easy diagnosis in the very first section, while in other cases multiple sections reveal only haematoma and necrosis. Hint: If there are omental or mesenteric implantations in the form of tiny red nodules, submitting these as well as the spleen will increase the probability of diagnosis because these small nodules are metastases of "pure" tumour. However, I remain reluctant to diagnose splenic haematoma as I know through the literature that most haematomas were proven, later, to have metastasized!

Next Newsletter... chronic dermatitis.

How do we bill your biopsies?

Dr David Taylor BVSc, Dip ACVP **Veterinary pathologist**

The general rules are:

One tissue or lesion = V1H = one biopsy fee

Two tissues or lesions that are different = V2H = two biopsy fee

Three tissues or lesions = V3H = three biopsy fee

Four tissues or lesions = V4H = four biopsy fee

Five or more tissues or lesions = VMH = multiple biopsy fee

Tissues or lesions well in excess of 5 will be charged the multiple biopsy fee plus an additional fee (please call to discuss).

Some examples (by no means exhaustive)...

1. Cat with itchy skin: 3 biopsies collected – 1x normal and 2x affected skin = one biopsy fee.
2. Investigation of chronic vomiting: Biopsies of stomach, small intestine, large intestine and liver = 4 biopsy fee. (Note multiple biopsies of the stomach, small intestine etc are not charged again and are actually encouraged).
3. Splenic mass: 5 tissues from the mass submitted = one biopsy fee.
4. Dog with itchy skin (as above) plus a separate lump on hock = two biopsy fee.
5. Post mortem performed in-clinic and sections of heart, lung, kidney, liver, stomach, intestine, spleen, brain submitted = multiple biopsy fee.
6. Three separate masses from different sites all submitted in one formalin pot = 3 biopsy fee.

As always, if you would like to discuss sampling and biopsy billing issues please call me on 02 9005 7714.

