Friday 5 April
Hall 3

Clinical pathology

Clinical pathology on a budget

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Getting more for no more from your laboratory

Ian Ramsey

The 'white heat' of modern veterinary science comes with a price tag that some owners are unable to contemplate. Clinical pathology can be a significant part of that final invoice. This cost can be reduced by careful case selection and choice of test. However, there are risks in this approach both in terms of patient welfare and practice economics. It is important to discuss all these costs and the risks of cost reduction with clients before embarking on any given course. Few in-house laboratories are cheaper than their commercial counterparts. Commercial laboratories can offer a more reliable service but there is an increased delay in getting results. Equally some tests are much cheaper if performed in practice (for example examination of a blood smear) but training, practice and experience are important.

Many external laboratories encourage the use of panels of tests. These selections can be beguilingly easy to choose, but some of the tests included within them are unnecessary in certain clinical situations. What constitutes an 'adequate work-up' should not be determined by a text book. The diagnostic criteria presented in research papers or used by referral practices may not be appropriate in a first-opinion practice. In general, the greater the number of clinical signs that are consistent with a diagnosis then the less rigorous the confirmation of the diagnosis is required to be. Large panels of tests increase the likelihood of results being outwith 'reference ranges' that may not be relevant to the patient, leading to misleading information and potentially incorrect diagnoses. Clinicians can save their clients considerable expense by contacting clinical pathologists to discuss the clinical history and the results of any diagnostic tests. All external laboratories provide telephone advice and can direct clinicians to further specialist advice on treatment if needed. Getting all the help and advice that you can reduces the risks of misdiagnosis and incorrect treatment.

The use of external laboratories also provides a huge opportunity for unofficial CPD. Examining blood smears and cytological preparations for yourself in those cases where samples are submitted allows you to better help those cases where owners cannot afford external expertise. Experience only comes with practice – and lack of practice damages self-confidence. Examining our own cytological preparations and blood smears allows us to better appreciate the comments made by external laboratories. Similarly, with biochemical results, a clinician is well advised to consider their own interpretation before looking at all interpretations that are provided both as an educational exercise and as critical control point. A clinician may never be as good as an expert in clinical pathology in an external laboratory, but that does not mean we can resign our responsibility for the final diagnosis. Therefore, we need to some extent to check that the laboratory report fits in with our clinical impression and not be led by the numbers or other people's interpretations.

Key to effective case selection is a detailed clinical examination with a focused patient history. This then needs to be communicated to the laboratory. The author believes that this communication is the attending clinician's responsibility and should not be delegated. However, this communication must also be brief – a PDF file of 78 pages of clinical notes is no more appropriate than one word. It is helpful to try to identify, then refine, the question that the clinician hopes the clinical pathologist will be able to help answer. 'What is wrong with my patient?' is too general!

KEY LEARNING OBJECTIVES

- You will know what is meant by the term 'clinical pathology on a budget' and the limitations and shifts in responsibilities that it imposes
- You will consider the range of outputs of external laboratories and how you can use these to their maximal effect in specific examples
- You will know the advantages of providing external laboratories with a good summary of the medical history and what is contained (and not contained) within that history

MULTIPLE CHOICE QUESTIONS

1. You are presented with a 13-year-old cat with a 3-month history of weight loss and lethargy but the owner reports no recent changes in appetite, thirst, urination and defecation. A detailed clinical examination is unremarkable. Your laboratory offers you an 'old thin cat profile' (consisting of T4, blood urea nitrogen (BUN), creatinine, alanine aminotransferase (ALT), alkaline phosphatase, glucose, urinalysis). What would be the most effective way of reducing the cost to the client?
   (A) Just test blood glucose first
   (B) Just test T4 first
   (C) Just test creatinine first
   (D) Just perform urinalysis first
2. Which group of clinical pathology tests is nearly always best done at an external laboratory?
   (A) Biochemistry
   (B) Endocrinology
   (C) Haematology
   (D) Urinalysis
3. What is the approximate incidence of abnormalities on a 'pre-anaesthetic profile' performed in otherwise healthy dogs such that anaesthesia would be postponed or the protocol changed?
   (A) 10%
   (B) 3%
   (C) 1%
   (D) 0.3%

Cytology: fine-needle aspirate techniques and solid lumps

Kathleen Tennant

Fine-needle aspiration of lumps and bumps can be a quick and relatively cheap way of screening lesions for those requiring more intensive treatment/assessment from those expected to have a more benign course.

Sampling with a variety of needle sizes and smear techniques may lead to a better cell yield and preservation on some smears than others and increase chances of obtaining at least one diagnostic sample. Taking care of the sample (drying quickly, not exposing to formalin fumes or water vapour, not loading into slide carriers while still wet) also results in better samples for assessment.

Be familiar with the charging structure of the external laboratory you send your slides to and make the most of it.

Even if you do not receive a definitive diagnosis with your aspirate of a lump, sometimes determining what it is *not* can be enough to help you manage your case, whether this is treatment or further diagnostics.

- If the sample is inflammatory in nature, be patient and spend time specifically looking for a cause
- If the sample yields tissue cells, ask yourself if the type of cell is appropriate for that anatomical location – knowing which cells are expected to be present helps you to spot interlopers
- The appearance of cells often signals their behaviour. Cells performing the same task usually look similar to each other. Wide variation may signal a loss of growth inhibition and should be regarded with suspicion
- Cells also warn of imminent or current proliferation with the presence of coarser chromatin, nucleoli and mitotic figures – marked increases in these in what should be a slow-growing tissue may not be appropriate

In short, screening cytology samples may save time and money by directing treatment and rationalizing further diagnostics.

KEY LEARNING OBJECTIVES

- Pick the best sampling techniques for the aspiration of lumps and bumps
- Appreciate the common artefacts seen on cytology and how to avoid them
- Screen cytology samples for inflammatory and neoplastic processes

MULTIPLE CHOICE QUESTIONS

1. Which of these is NOT associated with malignancy?
   - **(A)** Marked variation in cell size within a single population
   - **(B)** Uniformity of size within a cell population
   - **(C)** Marked variation in nuclear size within a single population
   - **(D)** Increased numbers of mitotic figures
2. Which of the following is NOT a cytology 'sin'?
   - **(A)** Staining a slide before you send out a sample to ensure that you have cells
   - **(B)** Placing the slides in a plastic slide container while they are still wet
   - **(C)** Placing the cytology slides in the same plastic bag as the histopathology samples
   - **(D)** Blowing on the slides to ensure quick drying
3. Which of the following is required to be certain that a cytology sample demonstrates a septic bacterial cause?
   - **(A)** Many different bacterial types being present
   - **(B)** A monomorphic population of bacteria being present
   - **(C)** Bacteria being present inside macrophages
   - **(D)** Bacteria being present inside neutrophils

Cytology: don't forget the blood smear

Kathleen Tennant

Haematology in the practice or external laboratory involves the generation of numerical values and often graphs or scatter plots. Often these give a strong indication of underlying haematological abnormalities – however, there are still some clinically relevant findings which can only be identified on the blood smear.

Platelet counts through the machine are subject to many artefacts which may result in incorrect numerical values. Assessing the smear can lead to a very different interpretation of what may initially appear to be a very thrombocytopenic sample.

Although many erythrocyte abnormalities do result in abnormal smear values, there are some which will be missed entirely without smear examination. An example of this would be the presence of red cell shear injury products, which might be caused by micro-angiopathic damage from underlying conditions, such as disseminated intravascular coagulation or abnormal small vessels in some neoplasms. The presence of Heinz bodies secondary to oxidative damage to erythrocytes can be seen on the routine smear, but is more obvious on a New Methylene Blue smear – not only does the presence of these suggest possible underlying pathologies (e.g. diabetic ketoacidosis, toxicity),

but may also affect the haemoglobin measurement through the machine.

Leucocytes are counted by different machines using different methodologies, and while total counts are often reliable, differential leucocyte counts can be variable. Looking at the smear helps to confirm the presence of left shift and toxic change in neutrophils, allowing a full appreciation of the degree of demand for neutrophils. Abnormal nucleated cells present in the blood smear can be assessed for possible origin, although this is not always fruitful. Nucleated red cell precursors can be identified and counted differently in different machine types and being able to enumerate and assess these can avoid acceptance of a spurious leucocytosis.

Although unusual, some pathogens, can be seen on the blood smear. Although haemoplasmas are extremely difficult to identify, some larger organisms such as *Babesia* sp. can be demonstrated inside erythrocytes.

Assessing the smear is worthwhile where there are abnormalities in the machine values, and, where possible, in any clinically unwell animal to provide the full haematological picture and direct further management.

KEY LEARNING OBJECTIVES

- Assess some changes in erythrocyte morphology and appreciate how these may signal underlying pathology
- Identify certain pathogens on the blood smear
- Use the smear to confirm or refute platelet measurements through the machine

MULTIPLE CHOICE QUESTIONS

1. Which of the following is true of a blood smear showing red cell regeneration?
   - **(A)** Neutrophils will also be left shifted
   - **(B)** Microcytosis will be visible in the red cells due to younger red cell precursors being present
   - **(C)** Larger, more basophilic red cells will be visible on the smear
   - **(D)** Nucleated red cells are always present
2. Which of the following is true of platelets?
   - **(A)** Very low counts through the machine will be due to clots in the sample and should be ignored
   - **(B)** Platelet clumps in the sample means that the animal definitely has adequate platelet numbers
   - **(C)** Larger platelets can be associated with platelet regeneration
   - **(D)** All Cavalier King Charles Spaniels have larger platelets and their platelet mass cannot be assessed
3. Which of the following is NOT a sign of toxic change in a neutrophil?
   - **(A)** A U-shaped nucleus
   - **(B)** Cytoplasmic basophilia
   - **(C)** Cytoplasmic vacuolation
   - **(D)** Doehle bodies in the cytoplasm

Cytology: fluids

Kathleen Tennant

Cytology of fluid samples can come with its own challenges. Depending on the cellularity and viscosity of the fluid, direct 'slide over slide' techniques may be appropriate for higher cellularity or thick samples, but in many cases a degree of concentration of the sample may be appropriate to ensure the best cell yield. Line preparations or sedimentation techniques can be employed where the cells are too fragile to concentrate easily in the centrifuge.

Drying slides made from fluids is best done quickly – thick, slow-drying preparations (such as synovial fluid placed into slide holder while still wet) can result in artefacts which can severely hamper assessment.

When assessing the smear, attention should be paid not only to the cells present but to the background as well. The first question should always be 'Is this slide of diagnostic quality?' If the cells are difficult to assess, it may be worth stopping and making another preparation.

Low-cellularity fluids with a transudative origin, whether pleural, peritoneal or cerebrospinal fluid (CSF) can be challenging in terms of cell yield, but frequently have a limited number of cell types associated with them.

Addition of proteins or cells to fluids (e.g. in exudates) usually results in preparations which are easier to assess, but thicker areas should be avoided. If inflammatory cells are present, a careful search for possible causes is always worthwhile. As well as more obvious causes such as organisms in septic peritonitis/pyothorax, other causative substances such as bile pigment in biliary rupture can occasionally be seen on the smear. Degenerative change in neutrophils can occur with septic causes, but also with exposure to bile pigments, urine or pancreatic enzymes. It should be noted that neutrophils in septic synovial fluids rarely show degenerative change – and phagocytosed bacteria are extremely rarely noted.

Neoplastic populations can be easy to pick up in some conditions (such as large cell lymphomas), but may exfoliate poorly in others (e.g. haemangiosarcomas) and finding them requires patience.

The presence of reactive mesothelial cells in pleural, peritoneal and pericardial fluids can complicate the picture – these cells undergo changes which mirror criteria for malignancy, and it is often impossible to distinguish mesothelioma/carcinoma from a merely dysplastic, reactive population.

KEY LEARNING OBJECTIVES

- Pick the best sampling, storage and preparation techniques for fluids
- Appreciate the common artefacts seen on fluid cytology and how to avoid them
- Screen fluid cytology samples for inflammatory and neoplastic processes

MULTIPLE CHOICE QUESTIONS

1. Which of the following is an acceptable way to handle fluids for cytological analysis?
   - **(A)** Make a direct preparation as well as a concentrated sample
   - **(B)** Routinely add a drop of formalin to fluid samples before making a preparation
   - **(C)** If there will be a delay in processing the sample, store in the freezer
   - **(D)** If there will be a delay in processing, store on the bench side
2. Which of the following is required for a confident diagnosis of septic pyothorax?
   - **(A)** Bacteria seen in the background
   - **(B)** Bacteria adherent to squamous epithelial cells
   - **(C)** Bacteria inside macrophages
   - **(D)** Bacteria inside neutrophils
3. Which of the following would NOT be a criterion for malignancy in a mesothelial peritoneal fluid population?
   - **(A)** A population in regular sheets
   - **(B)** A population showing variation in size
   - **(C)** A population with multiple nucleoli present in most cells
   - **(D)** A population with uneven multinucleation

When to test and when to treat?

Ian Ramsey

It is one of the perversities of life as a veterinary surgeon that the more experience of a particular condition that a clinician has, the cheaper the average total bill for that condition (author's unpublished observations). Some of this difference is the willingness of experienced clinicians to reduce the number and frequency of diagnostic tests, preferring to rely on therapeutic trials. Such trials are often highly effective management strategies. Preferring a treatment trial to a diagnostic test should not mean reducing the value placed on your time. The costs and benefits in treating dogs with a specific diagnosis need to be discussed with an owner before embarking on a therapeutic trial. Not all cases – even with a confirmed diagnosis – require treatment. Some cases may be monitored without treatment for months to years. Other cases may benefit from practical advice such as providing a dog-coat for dogs with non-pruritic alopecia.

Central to any discussion about treatment trials is to think about the necessity of diagnosis. Common presentations that do not usually require a diagnosis to be made include the first presentations of many acute clinical signs such as cough (without a heart murmur), vomiting, fever, diarrhoea, pruritis and stranguria. These presentations are noted for their ability to self-resolve, the availability of symptomatic treatment and one or two very common clinical causes. In contrast other presentations such as polyuria and polydipsia (PU/PD), yellow or white mucous membranes, ascites, abdominal masses and collapse do require some investigation to ascertain likely diagnoses and/or prognosis. Usually such presentations have no 'symptomatic treatment' options, may require urgent surgical intervention, or multiple possible differential diagnoses some of which may be rapidly progressive.

Therapeutic trials have their limitations. If the first attempt is unsuccessful then repeating them is unlikely to work. Second and subsequent presentations of many of the above clinical signs require at least an attempt at diagnosis. Cytological analyses of aspirates from fluids and masses are probably the most diagnostically useful form of clinical pathology. In contrast, haematology in non-anaemic, non-pyrexic animals is probably the least likely to be diagnostic. Urinalysis is best done in-house, whereas non-urgent biochemistry is often best done in an external laboratory.

It has been shown by several studies in several diseases that the clinical signs of a condition are often the best monitoring tools available. The better the owner monitors the clinical condition of the patient then the fewer blood tests etc. are needed. Asking owners to record water consumption, the time taken to eat the food, the faecal quality in an objective or semi-objective way will help to reduce the uncertainty factor which encourages clinicians to perform additional blood tests.

KEY LEARNING OBJECTIVES

- You will know a selection of case presentations that may be successfully managed without the use of diagnostic testing
- You will be able to develop a set of criteria for the assessment of any case to determine if diagnostic testing is likely to be productive
- You will appreciate the increased need for clinical monitoring with a reduction in diagnostic testing

MULTIPLE CHOICE QUESTIONS

1. You are presented with an 8-year-old Cocker Spaniel with a 2-day history of frequent bilious vomiting, lethargy and anorexia. There is no history of toxin or foreign body ingestion. A detailed clinical examination only suggests mild abdominal pain and dehydration. Your laboratory offers a Vomiting Dog screen (consisting of blood urea nitrogen (BUN), creatinine, alanine aminotransferase (ALT), alkaline phosphatase, electrolytes, qualitative pancreatic lipase). How would you manage this case if the client had a limited budget?
   - **(A)** Don't test but rather treat with fluids, maropitant and pain relief for 24–48 hours and then reassess
   - **(B)** As in a. and submit samples for the Vomiting Dog screen
   - **(C)** As in a. and also perform diagnostic imaging but not clinical pathology
   - **(D)** As in a. and just perform an in-house qualitative pancreatic lipase
2. What is the most effective method of monitoring a 12-year-old Minature Pinscher with diabetes?
   - **(A)** Clinical examination
   - **(B)** Fructosamine
   - **(C)** Glucose curve
   - **(D)** Owner monitoring of urination, thirst, appetite and weight
3. You are presented with a 9-year-old neutered female Dobermann with a 2-day history of stranguria and the owner has noticed blood in the urine (which the dog is still able to pass in small volumes frequently). What is the most appropriate symptomatic treatment?
   - **(A)** Marbofloxacin and buprenorphine
   - **(B)** Amoxicillin and increased water added to the food
   - **(C)** N-acetyl glucosamine and hyaluronic acids
   - **(D)** A prescription diet formulated to regulate urinary pH and time

Point-of-care meters

Ian Ramsey

In the last 20 years many point-of-care (POC) meters have been introduced to the veterinary market. These differ from in-house biochemical analysers in offering a smaller range of tests that are designed to be taken to the patient, used repeatedly and provide near-instant results.

POC devices include meters not only for glucose (with which many primary care clinicians are already familiar) but also for blood gases, electrolytes, ammonia, lactate, haemoglobin, ketones, coagulation tests and many others. These devices often have their origins in human medical POC tests. Such devices are subject to statutory regulation in the human healthcare market ensuring that certain performance criteria are met. No such regulations exist within the veterinary market and therefore it is important that practices are aware of the performance characteristics of these devices, so they can interpret results from them effectively.

POC devices offer economies of scale that make individual tests much cheaper than conventional in-house or external laboratory testing. However, as they often only measure one parameter they are usually best seen as monitoring devices rather than diagnostic tools. Their convenience can be offset by a reduction in the quality of the results, but providing the quality is still sufficient then the immediacy of those results may be of significant clinical benefit. In some cases the quality of the results is a significant improvement on those available from in-house analysers (e.g. ammonia). In others the quality is better than existing alternatives (e.g. haemoglobin compared to the traditional spun packed cell volume).

These devices can be very accurate and reliable – but they can be inaccurate or, worse, inconsistent both between samples and when used repeatedly on the same sample. There are published papers in respected peer-reviewed journals on the performance parameters for some POC devices, however some POC devices have never been independently evaluated in the clinical setting and performance data are limited and hard to obtain. Veterinary practices need to know how their devices perform over time and wherever possible against other machines and clinical expectations.

These devices can be easy to use – and easy to use wrongly. Incorrect use may not produce error alerts. Instructions provided with the device are not always clear or well illustrated. Veterinary practices using such devices must therefore have clear training protocols (and it is good working practice to have a list of nominated trainers and who have been trained to use the device).

The convenience of POC testing will mean that the number and range of these devices is likely to increase in the future. Newer technologies will expand the range of tests available and the number that any one device can deliver.

KEY LEARNING OBJECTIVES

- You will appreciate the current range, value and limitations of point-of-care (POC) meters with specific examples
- You will be able to develop protocols within your own small animal practice that ensure the optimal value and use of these meters
- You will be aware of the likely future for POC meters in small animal practice and be better prepared for their introduction in your working life

MULTIPLE CHOICE QUESTIONS

1. Which of the following tests is available on portable POC devices?
   - **(A)** Bile acids
   - **(B)** Ammonia
   - **(C)** Liver enzymes
   - **(D)** Bilirubin
2. Which one of the following statements about the sensitivity of POC devices is correct?
   - **(A)** Sensitivity is calculated by dividing the number of affected animals by the number of affected and unaffected animals in a population
   - **(B)** Sensitivity is more important than specificity when considering a test that, if positive, will likely result in the euthanasia of an animal
   - **(C)** Sensitivity is affected by the frequency of disease in a population and so needs to be determined in each population
   - **(D)** Sensitivity increases the accuracy of the measurement of the analyte in question
3. Why are traditional PCV measurements better than POC devices for haemoglobin in some primary care practices?
   - **(A)** They produce less sharps and are therefore safer
   - **(B)** They are less susceptible to operator error
   - **(C)** They are a better indicator of the oxygen-carrying capacity of the blood
   - **(D)** The senior partners still use the Fahrenheit scale and don't speak to doctors

Small mammals

John Chitty

Although haematology and serology will also be discussed in the talk, this is a basic guide to interpretation of biochemistries in small mammals (Figure 1).

Parameter	High	Low
Glucose	Very variable in an animal that 'continuously' feeds on high-carbohydrate foods. Not reliable enough to diagnose diabetes mellitus unless levels are persistently elevated and accompanied by persistent glucosuria. Even then it may simply reflect excessive feeding of simple carbohydrate. Glycosylated haemaglobin and fructosamine *may* assist Stress!!!! Pain!!! May allow assessment of pain	Important critical care parameter in moribund animals – hypoglycaemia may be accompanied by ketosis
Protein parameters – total, albumin, globulin	Similar interpretation to other mammals – ideally should be assessed by electrophoresis. Diet and husbandry play a major part in determining plasma albumin levels	Low albumin most likely to result from nutritional problems though liver or renal disease may play a part
Cholesterol/triglyceride	Potentially of great use given the frequency with which hepatic lipidosis is seen. However, diurnal rhythms and feeding changes make these hard to interpret (need to use fasted sample) However, in the anorexic rabbit elevated values and/or lipaemia can be important prognostic indicators	
Liver enzymes	Alanine aminotransferase (ALT) – sign of hepatocellular damage but not organ specific and has short half-life Aspartate aminotransferase (AST) – less specific and will rise with muscular exertion or tissue damage Gammaglutamyl transferase (GGT) – biliary stasis. Also found in the kidneys but not always increased in renal disease Alkaline phosphatase (ALP) – nearly all tissues Glutamate dehydrogenase (GLDH) – specific but not very sensitive	

Bile acids	Diurnal rhythm. Fasting samples not easily possible, so dynamic testing not likely Very high single samples may indicate hepatic dysfunction	
Urea	Renal disease (sensitive??) Also, prerenal – dehydration, water deprivation, cardiac disease, etc Also gut effects	Impaired hepatic function??
Creatinine	Renal disease – not very sensitive	
Calcium	Found as bound and unbound (ionized) forms. Values vary greatly compared to other species. High values not necessarily abnormal	Hypocalcaemic tetany seen in pregnant/lactating does. Must measure ionized form! Renal disease may cause raised or lowered levels of total calcium
Phosphate	May rise in renal disease	May fall in renal disease – effects dependent on diet
Electrolytes	Probably the same as in other mammals	Don't forget effects of anaesthesia and lipaemia In rabbits, sodium levels may be used as a prognostic indicator in gut stasis cases – lower levels are associated with a guarded prognosis

Figure 1: Interpretation of biochemistry results in small mammals

NB. *Guinea pigs*. Hyperthyroidism is well recognised in older animals showing weight loss and a rapid heart rate. Enlarged thyroids may be palpable. Normal ranges seem similar to cats, and measurement of total T4 is recommended in these cases.

In all species a euthyroid sick syndrome may be recognized. Hypothyroidism is rare in all these species.

KEY LEARNING OBJECTIVES

- To understand blood sampling methods in small mammals
- To understand interpretation of biochemical and haematology parameters
- To understand use and possibilities of faecal, urinary and bacteriology sampling in small mammals

MULTIPLE CHOICE QUESTIONS

1. Which of the following is CORRECT about serology?
   - **(A)** Provides information regarding exposure to infectious agents
   - **(B)** Confirms infection with an agent as cause of disease
   - **(C)** Confirms an animal will show signs of that disease in the future
   - **(D)** Guarantees the animal is not a carrier of that disease
2. Which of the following is CORRECT about a glucose level >30 mmol/l in a rabbit?
   - **(A)** Confirms that surgery is needed in a gut stasis case
   - **(B)** Suggests that obstruction of the gut is a strong possibility
   - **(C)** Is of no help at all in determining severity of disease in these cases
   - **(D)** Confirms that fluids and analgesia are all that will be required in this case
3. A rabbit is sneezing. What does the finding of *Pasteurella* spp. from an adult rabbit nasal swab show?
   - **(A)** This rabbit has pasteurellosis
   - **(B)** Is of no significance – *Pasteurella* is a commensal
   - **(C)** Should only be interpreted in the light of radiological and cytological findings
   - **(D)** Means the rabbit should be isolated from other rabbits

Reptiles and Birds

John Chitty

Clinical pathology is an essential tool in exotic pet medicine. While it can only ever be an aid to diagnosis rather than diagnostic in its own right, the generalized signs with which these animals are often presented and their ability to hide signs, means that 'tools' other than physical examination and history are helpful.

Good reliable results depend on many factors.

- Correct sample taking. Sites for sampling of birds and reptiles will be discussed as will the need for sedation/anaesthesia and handling techniques. The volumes suitable for collection will be discussed as will choices of tests for very small samples. In summary, the following sites are generally used:
  - birds – right jugular vein, brachial vein, caudal tibial vein;
  - chelonia – jugular vein, dorsal tail vein, subcarapacial vein, brachial vein;
  - lizards – ventral tail vein, heart;
  - snakes – ventral tail vein, heart
- Good-quality samples. Correct anticoagulant selection and sample handling are vital
- Choice of laboratory. As a minimum, in-house laboratories should be able to check packed cell volume (PCV), glucose, electrolytes, ionized calcium and cytology. External laboratories should be selected on the basis of experience with exotics' haematology, interest in exotics and range of tests available
- Choice of tests. Few tests have been evaluated in all the species that may be seen. In many cases tests used in other species are 'applied' to exotics. In addition, few 'normals' using large numbers of individuals have been generated. As such, choice of test and interpretation of results can be difficult
  - In all cases, full haematology is essential and often provides more information than biochemistries
  - Protein assessment is also extremely useful though it is recommended that electrophoresis is used to measure this as it is not only more accurate (especially in birds where a pre-albumin fraction may confuse differentiation of globulins) but provides an assessment of any inflammatory response
  - Electrolytes should be measured (sodium, potassium, chloride, ionized calcium) wherever possible and may be done patientside or on a spun heparin gel sample
  - Similarly, total calcium and phosphorus assessment are required in many cases
  - Creatinine kinase (CK) is often included as a marker of tissue damage, as many of the liver enzymes (e.g. aspartate aminotransferase (AST), lactate dehydrogenase (LDH)) are non-specific to liver, comparison to CK may assist in determining whether an enzyme rise is due to hepatocellular damage or tissue damage
  - Beta-hydroxybutyrate (especially chelonia), cholesterol and triglycerides are often used to assess nutritional state and risk of hepatic lipidosis

KEY LEARNING OBJECTIVES

- Understand sampling methods in birds and reptiles
- Understand interpretation of biochemical and haematology parameters
- Understand use and possibilities of faecal, urinary and bacteriology sampling in birds and reptiles

MULTIPLE CHOICE QUESTIONS

1. You have a 2-year-old grey parrot showing twitching and neurological signs. You suspect hypocalcaemia. Which test is most likely to be diagnostic?
   - **(A)** Total calcium
   - **(B)** Total calcium + phosphate
   - **(C)** Ionized calcium tested patientside
   - **(D)** Ionized calcium tested at an external laboratory using posted spun serum
2. A faecal sample from a healthy Mediterranean tortoise reveals some ciliated protozoa and a number of flagellates. What is the significance of this?
   - **(A)** These are a potential cause of diarrhoea and metronidazole should be given
   - **(B)** These are a potential cause of diarrhoea and probiotics should be given
   - **(C)** These are normal commensals
   - **(D)** The tortoise may be infectious to others
3. What is the significance of azurophils in snakes?
   - **(A)** Raised in chronic inflammatory conditions, analogous to monocytes in mammals
   - **(B)** Artefact
   - **(C)** Part of the acute inflammatory response
   - **(D)** Of no significance in interpretation