New Immunodiagnostic/Parasitology Tests Added to Orderable Catalog

by: Nicole Grosjean, BS, LVT

The Immunodiagnostic/Parasitology Section has added several new tests. Although previously available by special request, we have now included these in our regular test catalog in response to increased demand.

PCR Assays

Epizootic Hemorrhagic Disease Virus (EHDV) - Code 60071
EHDV infected thousands of deer during the summer and fall of 2012 resulting in the largest-ever outbreak in Michigan. Several other states were also impacted by EHDV during the season as well. DCPAH had over 130 cases submitted in just 3 months.

Tritrichomonas foetus - Code 60072
This PCR assay is done on bovine samples for reproductive disease and on feline samples for diarrhea. Due to the ease of sample submission, and faster turn-around-time, the diagnostic PCR assay is being requested much more frequently than the pouch culture assay that we have offered for years.

Equine Herpesvirus-5 - Code 60070
EHV-5 is a herpesvirus of horses and has been linked with Equine Multi-nodular Pulmonary Fibrosis (EMPF), a recently described respiratory disease of horses. Several PCR assays are available to detect this virus in clinical specimens, biopsies, and formalin fixed tissues.

Mycoplasma haemolamae - Code 60073
This PCR assay detects the agent, Mycoplasma haemolamae, which causes anemia in camelids such as llamas and alpacas. The disease is commonly termed Epe because it had been thought that an Eperythrozoon bacterium was the cause of the anemia.

IFA Titer

Brucella canis - Code 60067
This assay is available for dogs that will be breeding, or for clinical cases of reproductive disease. However, this assay does not satisfy most export requirements. We began offering this assay in response to the unavailability of the serum agglutination kits.

Therefore, the Brucella canis Antibody test (Code 90009) is unavailable until kits are available for purchase.

For complete information regarding sample submission, pricing, and other details on these assays, please visit our website at animalhealth.msu.edu and click Available Tests. If you have further questions, please contact the Immunodiagnostic/Parasitology section at 517.353.1683.
Diagnostic Utility of Vitamin D Metabolites – When Should I Measure Vitamin D?

By: Patricia A. Schenck, DVM, PhD

The regulation of serum calcium concentration is complex and requires the integrated actions of parathyroid hormone (PTH), and vitamin D metabolites. PTH and calcitriol (1,25-dihydroxyvitamin D) are the main regulators of calcium homeostasis. The intestine, kidney, and bone are the major target organs affected by calcium regulatory hormones. When serum ionized calcium concentration falls, PTH secretion is stimulated. PTH exerts direct effects on bone and kidney and indirect effects on the intestine through calcitriol. PTH increases synthesis of calcitriol by activating renal 1-alpha-hydroxylation of 25-hydroxyvitamin D from the circulation. Calcitriol then increases calcium absorption from the intestine, and also participates with PTH to stimulate osteoclastic bone resorption. Both PTH and calcitriol will cause increased tubular reabsorption of calcium from the glomerular filtrate, preventing calcium loss in urine.

Vitamin D is ingested from the diet as either vitamin D₂ (ergocalciferol) of plant origin, or vitamin D₃ (cholecalciferol) of animal tissue origin. Unlike humans, most animals have very little production of vitamin D in the skin with exposure to UV light. Vitamin D is stored in adipose tissue and is released into the circulation for conversion by the liver to 25-hydroxyvitamin D. 25-hydroxyvitamin D is the predominant circulating metabolite of vitamin D and is a good measure of vitamin D status. Circulating 25-hydroxyvitamin D undergoes glomerular filtration and is delivered to renal proximal tubules where it undergoes reabsorption by endocytosis. In the kidney, 1-alpha-hydroxylase converts 25-hydroxyvitamin D to calcitriol, the vitamin D metabolite with the highest biologic activity.

Measurement of vitamin D metabolites can be helpful in diagnosing disorders of calcium homeostasis. 25-hydroxyvitamin D and calcitriol are both of clinical interest for detection of hypovitaminosis D, hypervitaminosis D, and in renal disease. The metabolites of vitamin D are chemically identical in all species, and are stable during refrigeration and freezing.

Vitamin D Toxicity
Measurement of circulating 25-hydroxyvitamin D is beneficial in cases of possible vitamin D toxicity. Vitamin D intoxication may result from excessive vitamin D supplementation, errors in commercial diet formulation, ingestion of rodenticides containing vitamin D compounds, ingestion of toxic plants that contain glycosides of calcitriol (Cestrum diurnum, Solanum malacoxylon, and Trisetum flavescens), or ingestion of topical ointments containing vitamin D analogues (calcipotriene). These conditions are typically characterized by an elevated serum ionized calcium concentration (hypercalcemia) with low PTH and elevated serum phosphorus concentration. Serum 25-hydroxyvitamin D and calcitriol concentrations will be elevated in cases resulting from excessive vitamin D supplementation or ingestion of rodenticides; they will not be elevated in cases of plant ingestion or with Dovonex™ (a human anti-psoriasis cream containing calcipotriene) ingestion, as these substances contain calcitriol analogues that do not have cross-reactivity with the current 25-hydroxyvitamin D or calcitriol assays.

Vitamin D Deficiency
In cases of suspected vitamin D deficiency, serum 25-hydroxyvitamin D should be measured. With vitamin D deficiency, nutritional secondary hyperparathyroidism develops, characterized by a normal to low serum ionized calcium and elevated PTH concentration. Vitamin D deficiency most commonly occurs when diets low in calcium, low in vitamin D, or high in phosphorus (all meat diets) are fed.

Available Tests
We can help you measure vitamin D metabolites for your patients. DCPAH’s Endocrinology Section offers the following tests:

- **Vitamin D Profile** – Code 20035 (includes PTH, ionized calcium, and 25-hydroxyvitamin D)
- **25-Hydroxyvitamin D** – Code 20001
- **Calcitriol** – Code 20524 (new assay added in September 2012)

Each test includes interpretation of results by a veterinary endocrinologist.

Any pet fed a homemade diet should be evaluated routinely for vitamin D deficiency.

Nutritional secondary hyperparathyroidism can also occur when severe gastrointestinal disease is present, or when there has been prolonged anorexia. In these cases, the 25-hydroxyvitamin D concentration is low, reflecting inadequate intake or intestinal absorption of vitamin D.

DCPAH is a full-service veterinary diagnostic laboratory offering more than 800 tests in 11 service sections. Any news or information you’d like to see here? Please contact Courtney Chapin at chapinco@dcph.msu.edu.
Granulomatous Inflammation & Lymphoma
Calcitriol measurement may be useful in granulomatous inflammation and in some cases of lymphoma. Hypercalcemia can result from calcitriol synthesis by activated macrophages during granulomatous inflammation. Calcitriol is elevated in blastomycosis, nocardiosis, nodular panniculitis, and potentially other causes of granulomatous inflammation. In most cases of lymphoma with associated hypercalcemia, calcitriol concentration is decreased. However, some lymphocytes contain 1-alpha-hydroxylase that converts 25-hydroxyvitamin D to calcitriol, and in these cases, calcitriol concentration is elevated.

Vitamin D-Dependent Rickets
Measurement of serum calcitriol is also very useful in differentiating between type 1 and type 2 vitamin D-dependent rickets (VDDR). Affected animals are typically hypocalcemic, with an elevated PTH concentration (secondary hyperparathyroidism). Clinically, these patients are young, and have lameness, bone malformations and fractures, despite having been fed complete and balanced diets. Vitamin D-dependent rickets type 1 is characterized by a deficiency of 1-alpha-hydroxylase which converts 25-hydroxyvitamin D to calcitriol. The serum concentration of 25-hydroxyvitamin D is usually within normal limits, and serum calcitriol concentration is low. Vitamin-D dependent rickets type 2 is characterized by end-organ resistance to calcitriol. Serum concentration of 25-hydroxyvitamin D is usually within normal limits, and serum calcitriol concentration is elevated.

Chronic Kidney Disease
Secondary hyperparathyroidism can also occur in chronic kidney disease (CKD). A deficit of calcitriol is an important factor leading to the uncontrolled secretion of PTH resulting in secondary hyperparathyroidism. As renal proximal tubule cells exhibit decreased function, calcitriol synthesis decreases, resulting in decreased intestinal calcium absorption leading to hypocalcemia. As the ionized calcium concentration decreases, the secretion of PTH is stimulated. The excess PTH effectively resorbs bone in an attempt to raise ionized calcium, resulting in bone loss. Concentrations of both 25-hydroxyvitamin D and calcitriol are significantly lower in CKD as compared to healthy animals. 25-hydroxyvitamin D concentrations can be low in CKD patients due to inadequate dietary intake, but also from failure to adequately reclaim 25-hydroxyvitamin D following glomerular filtration in those with advancing CKD. It is important to monitor serum ionized calcium, PTH, 25-hydroxyvitamin D, and calcitriol concentrations in CKD. Renal secondary hyperparathyroidism can occur prior to the development of azotemia, suggesting that routine monitoring of calcium status may be beneficial in older patients or those at risk of CKD. Serum total calcium measurement alone is not adequate since elevations of PTH and decreases of calcitriol can occur while serum total calcium and phosphorus remain within normal limits. Periodic measurement of serum calcitriol can document decreases over time, and replacement therapy can be initiated early in the course of CKD to prevent the development of secondary hyperparathyroidism.

Table: Conditions in which measurement of 25-hydroxyvitamin D (25-(OH)D) and/or calcitriol may be helpful.

<table>
<thead>
<tr>
<th>Measure 25-hydroxyvitamin D</th>
<th>Measure Calcitriol</th>
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| Excess Vitamin D Suspected – 25-(OH)D elevated  
- Vitamin D oversupplementation  
- Possible cholecalciferol rodenticide exposure | Excess Suspected – Calcitriol elevated  
- Overdosage of calcitriol therapy  
- Possible Granulomatous Disease  
- Some cases of lymphoma |
| Vitamin D Deficiency Suspected – 25-(OH)D low  
- Pets fed homemade diets  
- Severe gastrointestinal disease  
- Prolonged anorexia | VDDR type 1 – Calcitriol usually low  
VDDR type 2 – Calcitriol usually elevated |
| VDDR type 1 or 2 – 25-(OH)D usually normal | Obtain baseline calcitriol concentration in older pets prior to development of CKD  
Renal disease – Calcitriol low |
| Renal disease – 25-(OH)D low to normal | |

Client Education Resources Available!
A new guide designed to help clinicians educate pet owners about living with a pet diagnosed with chronic kidney disease is available at animalhealth.msu.edu. Other guides on canine leptospirosis and ticks and tick-borne diseases are also available. More client education resources are coming. Please let us know if you have topics you’d like us to cover by contacting DCPAH communications manager Courtney Chapin, chapinco@dcpah.msu.edu.
Full AAVLD Accreditation Extended
DCPAH again achieved full accreditation / all species by the American Association of Veterinary Laboratory Diagnosticians (AAVLD) after a recent site review. AAVLD extended our full accreditation for an additional five years, 2012-2017. We have an ongoing commitment to quality and continuous improvement and were proud to receive this important validation of the high quality diagnostic service we provide to our clients.

Find our quality assurance letter and accreditation certificate online at animalhealth.msu.edu under Quality Assurance.

It's Almost Conference Time
This winter we’ll be in the exhibit hall of the three conferences listed below. If you plan to attend one, please stop by.

Meet the people behind the testing you and your clients rely on. Ask us questions. Give us feedback. Pick up informational materials and giveaways. Mention that you read this newsletter and get a special token of our appreciation!

We enjoy meeting our clients and always receive great ideas when we have the opportunity to interact with them face-to-face. We’d love to meet you.

North American Veterinary Conference
Orlando, Florida
January 19-23, 2013
Booth 530

Michigan Veterinary Conference
Lansing, Michigan
January 25-27, 2013
Booth 78-79

Midwest Veterinary Conference
Columbus, Ohio
February 21-24, 2013
Booth 522

Please Note Our Holiday Hours
DCPAH will be open on the following university holidays:
We will be closed December 25 and January 1.

Holiday hours of operation, including specimen receiving and telephone coverage, are 8:00 a.m. - 1:00 p.m.
All of us at DCPAH wish you a happy and healthy holiday season!