Screening for vector-borne disease

IDEXX 4Dx® Plus Test clinical reference guide
Every dog, every year

The Companion Animal Parasite Council (CAPC) Guidelines recommend annual comprehensive screening for pathogens transmitted by ticks and mosquitoes.1 Adding annual cycle of comprehensive testing and year-round prevention to your practice benefits your patients, clients, and practice in 3 important ways:

1. **React to changing prevalence**
   Mosquitoes and ticks are constantly on the move, and annual testing is the most reliable way to determine if new infections are threatening pets in your area. Pets move too, of course; without comprehensive testing, you sacrifice the ability to detect and treat mosquito and tick-borne infections acquired in other locations.

2. **Detect and treat coinfection**
   Comprehensive testing lets you assess a dog’s risk of having more than one infection.2

3. **Measure the effectiveness of prevention protocols**
   Only comprehensive testing helps you know if your prevention protocols are working. Even a negative result is valuable; it’s an opportunity to celebrate the pet owner’s role in successfully preventing these infections and keeping their pet healthy.

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Know more with every result

With the IDEXX 4Dx® Plus Test, a positive result can also be an indication of ticks and the pathogens they carry.

**When you use the IDEXX 4Dx Plus Test as a screening tool, you may**

<table>
<thead>
<tr>
<th>pathogens</th>
<th>carried by</th>
<th>that may also transmit other pathogens and infections to dogs and people</th>
<th>Geographic tick distribution as of 20213</th>
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<tr>
<td>Ehrlichia ewingii</td>
<td>Lone star tick</td>
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<td>Anaplasma phagocytophilum</td>
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<td>Ehrlichia canis</td>
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<td>American dog tick (Dermacentor variabilis)</td>
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</tr>
</tbody>
</table>

Anaplasma spp.
Babesia spp.
Rocky Mountain spotted fever
Tularemia

Geographic tick distribution as of 2021:

- **Brown dog tick (Dermacentor variabilis)**
- **Lone star tick**
- **Black-legged tick (deer tick)**
- **Lone star tick**
- **Brown dog tick**
- **Rocky Mountain spotted fever**
- **Tularemia**
Lyme disease

Transmitted by the black-legged tick (deer tick), Lyme disease is caused by the bacterium Borrelia burgdorferi. Clinical signs may not appear until several months after infection. Lyme disease has been found throughout North America with cases that have mild to severe disease.

**Did you know?**

- Dogs testing positive for antibodies to the C6 peptide had 43% increased risk of having chronic kidney disease (CKD) compared to seronegative dogs.
- The C6 peptide used in the IDEXX 4Dx® Plus Test and Lyme Quant C6® Antibody Test does not cross-react with the antibody response to commercially available Lyme vaccines.
- Dogs with seroreactivity to both Borrelia burgdorferi and Anaplasma phagocytophilum may have two times the risk of developing clinical illness than singularly infected dogs.

**Borrelia burgdorferi**

**Primary vectors**
- Ixodes scapularis or Ixodes pacificus

**Pathology**
- Localizes in tissues of infected dogs
- Synovitis (may be subclinical)
- Lyme nephritis

**Clinical presentation**
- Chronic infection with clinical signs that may present acutely:
  - Fever, anorexia
  - Polyarthritis, lameness
  - Rapidly progressive renal failure
  - Neurologic syndromes

**Laboratory abnormalities**
- Elevated C6 antibody level ≥ 30 U/mL
- May have proteinuria
- May have IDEXX SDMA™ Test result > 14 µg/dL

**CKD monitoring**
- Chemistry panel with SDMA – Recommended to evaluate for proteinuria
- CBC with blood film evaluation – Recommended as part of a minimum database

**Monitoring considerations**

1. If C6 antibody level drops ≥ 50% from initial level, treatment was successful.
2. If C6 antibody level drops < 50% from initial titer, differentials include:
   - Noncompliance with treatment. Consider retesting.
   - Reinfection. Reevaluate tick control; consider retesting.
   - Chronic infection.

**Test results**

- A Lyme Quant C6® Antibody Test is recommended to evaluate for active infection and to establish a baseline to assess response to treatment if indicated.
- A Urinalysis with Reflex UPC is recommended to evaluate for proteinuria.

**Test interpretation**

- A C6 antibody level > 30 U/mL is considered clinically significant and consistent with a clinical Lyme disease infection.
- An elevated UPC > 0.2 may be evidence of secondary glomerulonephritis.

**Asymptomatic dogs may not have specific hematologic or biochemical changes but should be monitored for proteinuria or development of clinical signs.**

**Retest at 6 months using Lyme Quant C6 Antibody Test.**

The IDEXX 4Dx Plus Test can be used to identify a subset of asymptomatic dogs at risk for developing clinical illness and would require follow-up Lyme Quant C6 Antibody Test to evaluate bacterial response.

**Prevent**

Evaluate tick prevention strategies and reinforce the value of year-round protection.

**SeroLOGY is typically used to diagnose Lyme disease. B. burgdorferi localizes to the tissues and is therefore rarely detectable in the blood by PCR.**

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* Lyme disease is transmitted by the black-legged tick (deer tick). Lyme disease is caused by the bacterium *Borrelia burgdorferi*. Clinical signs may not appear until several months after infection. Lyme disease has been found throughout North America with cases that have mild to severe disease.

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Heartworm disease

Dirofilaria immitis, the causative agent of heartworm disease, is transmitted when mosquitoes infected with D. immitis larvae feed on (or bite) a healthy dog. Heartworm disease has subtle or mild clinical signs in the early stages, making preventive measures so much more important—especially as advanced infection may result in death.

Did you know?
• Despite availability of monthly preventives, prevalence rates of canine heartworm have remained consistent nationwide.7
• The American Heartworm Society (AHS) and the Companion Animal Parasite Council (CAPC) recommend testing all dogs for both antigen and microfilariae annually.7,8
• For more information and current recommendations on treating canine heartworm disease, go to heartwormsociety.org or capcvet.org.

Dirofilaria immitis

- Primary vector: Mosquito
- Pathology: Infective larvae (L3) mature to adult worms in the heart and pulmonary arteries
- Clinical presentation: Asymptomatic at first, later developing:
  • Mild, persistent cough
  • Lethargy
  • Exercise intolerance
  • Reduced appetite
  • Weight loss
- Laboratory abnormalities that may be seen:
  • Eosinophilia
  • Azotemia
  • Increased liver enzymes
  • Proteinuria

Clinical signs

Does the dog have one or more of these signs?
• Cough
• Dyspnea
• Syncope
• Abnormal heart or lung sounds

IDEXX 4Dx® Plus result

- Negative
- Positive
  • A negative result is unexpected when heartworm infection is suspected.
  • Heartworm Antigen by ELISA with Heat Treatment (test code 7232) should be considered.

Next step considerations

Heat treatment aids in the detection of heartworm antigen in an infected dog when immune complexes are present. A positive result following heat treatment is consistent with the presence of immune complexes blocking the detection of heartworm antigen.

The American Heartworm Society recommends that a confirmatory test be run on all positive antigen test results prior to therapy, especially when a positive test result is unexpected.

- Retest for heartworm antigen with Heartworm Antigen by ELISA or retest using the IDEXX 4Dx® Plus Test or SNAP® Heartworm RT Test
- Evaluate for microfilaria.
- Consider the following:
  • Thoracic radiographs
  • CBC
  • Chemistry panel with SDMA
  • Urinalysis with UPC

- A negative result is unexpected when heartworm infection is suspected.
- Heartworm Antigen by ELISA with Heat Treatment (test code 7232) should be considered.

Monitor

Recommend treating confirmed positive results with adulticide.*
Retest in 9–12 months following adulticide treatment.*
Retest in 12 months.

Prevent

Evaluate vector-borne disease prevention strategies and reinforce the value of year-round protection.

*Administer treatment according to American Heartworm Society protocol.
Canine anaplasmosis

Canine granulocytic anaplasmosis is caused by the bacterium *Anaplasma phagocytophilum* (transmitted by the black-legged tick [deer tick]). *Anaplasma platys* (transmitted by the brown dog tick) is the cause of infectious cyclic thrombocytopenia.

**Did you know?**
- Many mammalian species, including humans, are susceptible to *A. phagocytophilum* infection.
- Dogs coinfected with *Anaplasma* and other bacterial pathogens may have more complex disease presentations and respond more slowly to therapy.
- *A. platys* infects canine platelets and is frequently seen as a coinfection with *Ehrlichia canis*.

**Clinical signs**
- Fever
- Anorexia
- Lethargy
- Polyarthritis, lameness
- Neurologic signs

**Pathology**
- Infects neutrophils
- Infects platelets

**Clinical presentation**
- Can present acutely:
  - Fever
  - Anorexia
  - Lethargy
  - Polyarthritis, lameness
  - Neurologic signs

**Laboratory abnormalities**
- Thrombocytopenia
- Anemia
- Lymphopenia
- Increased liver enzymes
- Other findings may be seen:
  - Decreased albumin
  - Increased globulin
  - Increased ALP and ALT
  - Proteinuria
  - Decreased Urine SG
  - Increased UPC

**Note**
- Previous infection may not prevent reinfection and persistent infections are possible.**9,10**

**IDEXX 4Dx® Plus result**
- Negative
- Positive
- Positive
- Negative

**Test results**
- Thrombocytopenia
- Anemia
- Lymphopenia
- Increased liver enzymes
- Other findings may be seen:
  - Decreased albumin
  - Increased globulin
  - Increased ALP and ALT
  - Proteinuria
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  - Increased UPC

**Test interpretation**
- Look for evidence of other tick-borne diseases.
- The results are consistent with a clinical anaplasmosis disease diagnosis. Consider treatment with appropriate therapy.
- A positive *Anaplasma* result indicates the presence of *Anaplasma* antibodies. In a dog without clinical signs and a normal CBC, this may reflect prior exposure to a tick infected with *Anaplasma*.

**Monitor**
- Evaluate clinical response and repeat CBC in 7 days. If no improvement, pursue other differentials for clinical and hematology abnormalities.

**Prevent**
- Evaluate tick prevention strategies and reinforce the value of year-round protection.
Canine ehrlichiosis

Canine ehrlichiosis is caused by the bacteria *Ehrlichia canis* (transmitted by the brown dog tick) and *Ehrlichia ewingii* (transmitted by the lone star tick). Canine *Ehrlichia* infections may progress to the subclinical phase or may become chronic infections.

**Did you know?**

- Dogs coinfected with *E. canis* and *A. platys* were found to have more severe anemia and thrombocytopenia than dogs with either single infection.11
- In a study of healthy dogs with antibodies to *E. canis*, 39% were thrombocytopenic.12
- Chronic *E. canis* infections, if left untreated, can lead to bone marrow dysfunction or kidney disease.
- Dogs with *Ehrlichia* antibodies in *E. canis* endemic areas had a 112% increased risk of developing chronic kidney disease (CKD).13
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**Laboratory abnormalities**

- Anemia
- Thrombocytopenia
- Hyperglycemia
- Proteinuria

Other clinical findings may include:
- Decreased albumin
- Increased globulin
- Mild increased ALT and ALP
- Increased SDMA
- Creatinine
- Decreased urine specific gravity, proteinuria
- Increased urine protein creatinine (UPC) ratio

**Note**

Previous infection may not prevent reinfection, and persistent infections are possible.14

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**CKD monitoring**

- Chemistry panel with SDMA - Recommended to evaluate for secondary kidney disease.
- Urinalysis with UPC - Recommended to evaluate for proteinuria.
- CBC with blood film - Recommended as part of a minimum database.

**Clinical signs**

- Fever
- Lethargy
- Anorexia
- Lameness
- Epistaxis
- Lymphadenopathy
- Petechiae
- Neurologic signs

**Test results**

- Thrombocytopenia
- Anemia
- Positive PCR result
- With or without neutrophilia
- With or without monocytosis

- Moderate to marked pancytopenia may be seen in the severe chronic phase of ehrlichiosis.

**Test interpretation**

- Look for evidence of other tick-borne diseases.
- The results are consistent with a clinical ehrlichiosis disease diagnosis. Consider treatment with appropriate therapy.
- A positive *Ehrlichia* result indicates the presence of *Ehrlichia* antibodies. In a dog without clinical signs and whose CBC results do not support ehrlichiosis, this may reflect prior exposure to a tick infected with *Ehrlichia* spp.
- Evaluate clinical response and CBC in 7 days. If no improvement, pursue other differentials for clinical and hematology abnormalities.
- A chemistry panel with SDMA and Urinalysis with Reflex UPC if indicated are recommended to evaluate for secondary kidney disease.

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**Clinical signs**

- Does the dog have one or more of these signs?
  - Fever
  - Anorexia
  - Lethargy
  - Lameness
  - Epistaxis
  - Lymphadenopathy
  - Petechiae
  - Neurologic signs

**IDEXX 4Dx® Plus result**

- Negative
- Positive

**Next step considerations**

- CBC with blood film evaluation
- Tick/Vector Comprehensive (RealPCR®) Panel
- Chemistry panel
- Urinalysis with Reflex UPC
- A CBC with blood film evaluation on a fresh whole blood specimen is recommended to evaluate for evidence of clinical ehrlichiosis.
- Infection is unlikely. Review benefits of tick prevention. Return for tick-borne disease annually.

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**Test results**

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- Anemia
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- With or without monocytosis

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**Prevent**

- Evaluate tick prevention strategies and reinforce the value of year-round protection.
Serology and PCR for sick patients

For dogs presenting with clinical signs consistent with a vector-borne disease, using serology and PCR together improves your ability to make an accurate diagnosis.

Benefits and limitations of each diagnostic method

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<tr>
<th>Measures</th>
<th>Serology</th>
<th>Polymerase chain reaction (PCR)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serology</td>
<td>Antibody response of host</td>
<td>Nucleic acid (DNA) from pathogen</td>
</tr>
<tr>
<td>Benefits</td>
<td>Useful for screening as well as diagnosis of infection</td>
<td>Specifically identifies pathogens indicating active infection</td>
</tr>
<tr>
<td>Limitations</td>
<td>Clinical signs may precede a measurable antibody response</td>
<td>A negative PCR result does not necessarily rule out infection</td>
</tr>
</tbody>
</table>

Dogs with ehrlichiosis and anaplasmosis may present with clinical signs at different times after infection. Which sick dog are you dealing with?

When to use the IDEXX vector-borne disease RealPCR™ panels

- Sick patients with clinical signs and/or laboratory abnormalities consistent with a vector-borne illness
- Patients with subclinical infections based on history, physical examination, serology, and clinical laboratory findings

“"No single test is sufficient for diagnosing an infectious disease in a sick patient.""

Edward Breitschwerdt, DVM, DACVIM*
Professor, Internal Medicine
College of Veterinary Medicine, North Carolina State University

*Dr. Breitschwerdt has a business relationship with IDEXX pursuant to which he receives compensation from IDEXX from time to time. The views expressed in this guide are solely those of Dr. Breitschwerdt.
Debendositivity and improved sensitivity.

Available in-clinic or from IDEXX Reference Laboratories

SNAP® technology uses a proprietary three-step process to deliver dependable sensitivity and specificity.

Bidirectional flow:
Sample flows across the test and binds with capture reagents. Activating the test unleashes a second flow that drives the sample back across the capture reagents, providing another opportunity for binding and improved sensitivity.

Wash:
To enhance specificity, a wash step removes unbound debris that could interfere with results. It also cleans the window so you can easily read the result.

Amplification:
For maximum sensitivity, a chemical reaction amplifies the signal and generates the SNAP® test’s distinctive blue dots, allowing you to see even low-level positives.

References