trūRapid[™] FOUR Test FAQs

What does trūRapid FOUR detect?

trūRapid FOUR is a rapid test designed to detect the presence of adult female *Dirofilaria immitis* (heartworm) antigens and antibodies to *Borrelia burgdorferi* (Lyme), *Anaplasma* spp. (*A. phagocytophilum and A. platys*), and *Ehrlichia* spp. (*Ehrlichia canis, Ehrlichia chaffeensis*, and *Ehrlichia ewingi*) in canine serum, plasma, or anticoagulated whole blood.

Which species can be assessed with trūRapid FOUR?

The test has been validated for use with dogs only.

How many tests are included in each test kit?

Each trūRapid FOUR test kit contains 25 tests (i.e., 25 foil-wrapped cartridges, 25 disposable sample pipettes, and a bottle of sample buffer). The catalog number is 100074.

Do test kits need to be refrigerated?

The trūRapid FOUR test kits can be stored at room temperature, 59–86°F or 15–30°C, and are valid until the expiration date listed on each test kit.

Are there any special requirements for handling the test kits?

The trūRapid FOUR test kits cannot be frozen or stored in direct sunlight.

What is the shelf life of the test kits?

The trūRapid FOUR test kits can be used up to the expiration date listed on the test kits. Test kits have at least two months of dating when shipped.

What is the recommended sample type for trūRapid FOUR?

The test can be performed using anticoagulated whole blood, serum, or plasma. Whole blood in EDTA or lithium heparin tubes may be preferred to avoid time involved in centrifugation and removal of plasma or serum. Testing with fresh samples is best. However, refrigerated or frozen samples can be used when following proper handling instructions.

Are there any sample handling requirements?

Samples can be tested fresh, or they can be stored in a refrigerator at 35–46°F or 2–8°C for up to one week. Refrigerated samples must come to room temperature prior to testing. If longer sample storage is required, serum or plasma samples may be stored at -4°F or -20°C. Frozen samples should be centrifuged prior to use.

What is the test process?

After collecting the sample, fill the provided sample pipette. (The collection syringe should not be used to deliver samples to the testing well.) Using the filled sample pipette, add one drop of the sample to each of the test wells on the test cassette, followed by two drops of the sample buffer to each sample well. Do not read the test before completing the 10-min incubation. Test results can then be manually entered into the data management system. NOTE: Test cassette should be identified with patient name/number.

Are there any interfering factors that can impact test result integrity?

There is no interference from lipemia or from mild to moderate hemolysis.

How long does it take to get a test result?

Test results should be interpreted 10 minutes after the addition of the sample and the buffer solution. Tests should not be interpreted before or after 10 minutes.

How are the test results interpreted?

trūRapid FOUR is a rapid lateral flow test that provides a positive or negative test result for *Dirofilaria immitis* (heartworm), *Borrelia burgdorferi* (Lyme), *Anaplasma* spp., and *Ehrlichia* spp. in one single test cassette. Test results should be interpreted 10 minutes after the addition of the sample and the buffer solution. Interpretation is done visually.

Each test cartridge contains three individual test strips: one for heartworm/Lyme, one for *Anaplasm*a, and one for *Ehrlichia*. Each test strip should be interpreted individually.

- Positive A pink/purple test line of any intensity appears next to Heartworm (H), Lyme (L), Anaplasma (A), and/or Ehrlichia (E) and in the individual test's corresponding control (C) area.
- Negative No test line appears next to Heartworm (H), Lyme (L), Anaplasma (A), and/or Ehrlichia (E), but a line is visible in the individual test's corresponding control (C) area.
- Invalid No control (C) line is visible for an individual test.

NOTE: Because each test strip is designed to be interpreted individually, it's possible to have varying combinations of positive, negative, and invalid test results on a single test cartridge. See the instructions for use for more details.

How does the test perform? What is the sensitivity/specificity?

trūRapid FOUR has excellent sensitivity and specificity when compared to laboratory tests and other commercially available in-house tests. Performance study details are documented in the trūRapid FOUR Product Bulletin.

Is it necessary to confirm a positive heartworm test result when using the truRapid FOUR?

American Heartworm Society (AHS) Canine Heartworm Guidelines (2024) recommends that all positive heartworm results should be confirmed with a secondary test that uses a different detection method than the original test¹. Secondary tests include microfilaria testing (Knott's), which can be ordered via Antech Reference Laboratory (Use test code T390 for individual test (CT390 in Canada) or ADD80 to add to profile).¹

Does Antech offer confirmatory testing for heartworm test results using trūRapid FOUR?

Please call Antech Specialty Products Technical Support at <u>800-464-3252, option 5</u> with any questions regarding trūRapid test results and to discuss follow-up testing options.

Are there any guidelines regarding confirmatory or follow-up testing for a positive Lyme test when using trūRapid FOUR?

The 2018 ACVIM consensus update on Lyme borreliosis in dogs and cats indicates the utility of testing for C6 antibodies for the detection of Lyme disease.² Refer to the Antech Lyme Clinical Algorithm for more detailed guidance.

Are there any guidelines regarding confirmatory or follow-up testing for a positive *Anaplasma* test when using trūRapid FOUR?

There is currently no consensus as to whether seropositive but clinically normal dogs require treatment. If the patient is asymptomatic but subclinical infection is a concern, consider a full minimum database, including a CBC (to evaluate for morulae and thrombocytopenia). If the patient has thrombocytopenia, confirm *Anaplasma* infection with a tick-borne lab method, evaluate for coinfections, and proceed to treat with doxycycline for 30 days. A recheck CBC is recommended to document the resolution of the thrombocytopenia. Year-round tick control strategies should be implemented or improved in the face of a positive result.³⁴

Are there any guidelines regarding confirmatory or follow-up testing for a positive *Ehrlichia* test when using trūRapid FOUR?

If the patient has thrombocytopenia, confirm Ehrlichia infection with a polymerase chain reaction (PCR) test, evaluate for coinfections, and proceed to treat doxycycline for 30 days.³⁴

How can truRapid FOUR test kits be ordered?

To order trūRapid FOUR test kits, call **800–464–3752, option 1, option 2**, or email **orders@heska.com**. You can also order via our customer ordering portal at **customerportal.heska.com**.

Who can we contact if we have questions about truRapid FOUR test performance?

For questions about trūRapid FOUR test performance, contact Antech's Specialty Products Technical Support at 800-464-3752, option 5.

- 1. American Heartworm Society Canine Guidelines for the Prevention, Diagnosis, and Management of Heartworm (*Dirofilaria immitis*) Infection in Dogs, 2024 Revised.
- 2. Littman, M. P., Gerber, B., Goldstein, R. E., Labato, M. A., Lappin, M. R., & Moore, G. E. (2018). ACVIM consensus update on Lyme *borreliosis* in dogs and cats. J Vet Intern Med, 32(3), 887–903. https://doi.org/10.1111/jvim.1508.
- 3. Companion Animal Parasite Council | Ehrlichia spp. and Anaplasma spp
- 4. Sainz Á, Roura X, Miró G, Estrada-Peña A, Kohn B, Harrus S, Solano-Gallego L. Guideline for veterinary practitioners on canine ehrlichiosis and anaplasmosis in Europe. Parasit Vectors. 2015 Feb 4;8:75. doi: 10.1186/s13071-015-0649-0. PMID: 25649069; PMCID: PMC4324656.