## Nu.Q<sup>®</sup> on the Element i+<sup>™</sup> Immunodiagnostic Analyzer Frequently Asked Questions

#### SUMMARY

Nu.Q<sup>®</sup> rapidly detects and quantifies nucleosomes in canine plasma. An elevated nucleosome concentration can serve as a biomarker to raise suspicion for certain types of cancer, especially lymphoma and hemangiosarcoma.<sup>1-3</sup>

- Nu.Q<sup>®</sup> detects nucleosomes which can act as a biomarker for current/active cancer in otherwise healthy patients. Nu.Q<sup>®</sup> cannot
  predict future cancer development.
- Nu.Q<sup>®</sup> is not a confirmatory test for cancer. Elevated Nu.Q<sup>®</sup> results should act as an impetus to perform additional diagnostics to obtain a diagnosis.
- Not all cancer presentations cause nucleosome elevation. Certain cancer types and stages do not cause an elevated Nu.Q® result.
- Other conditions can elevate nucleosome concentrations, including but not limited to: immune-mediated disease, sepsis, trauma, and other inflammatory conditions. This test cannot differentiate between inflammatory diseases and cancer.

#### WHAT AND WHEN

#### What does Nu.Q® measure?

DNA within cells is wound tightly around histones (proteins) in assemblies called nucleosomes, which form structures resembling beads on a string along each chromosome. When a patient has a type and stage of cancer that causes higher than normal amounts of cell death, nucleosomes from those cancer cells are released into the blood and can be measured. Nu.Q<sup>®</sup> measures the level of nucleosomes that are freely circulating in the blood.

#### In which species can I use Nu.Q<sup>®</sup> testing?

Nu.Q® on the Element i+® is only validated for use in dogs. There are currently no published studies for its use in other species.

#### When should I perform Nu.Q<sup>®</sup> testing?

Nu.Q<sup>®</sup> is best suited to be performed during wellness checks for healthy dogs who are 7 years and older. It is also indicated as a part of routine wellness diagnostics for dogs who are 4 years and older with a predisposition to developing cancer, including dogs with a family history of the disease and/or certain breeds with higher incidence rates (e.g., Labrador retrievers, French bulldogs, golden retrievers, German shepherds, beagles, Rottweilers, boxers, Pembroke Welsh corgis, Siberian huskies, Bernese mountain dogs, bullmastiffs, Irish water spaniels, flat coated retrievers, and English setters).<sup>4</sup>

#### Does the patient need to be fasted?

Fasting is not required prior to running the Nu.Q(R ball) test. However, if a non-fasted patient returns a Moderate or High Suspicion result, consider collecting a new sample after fasting and repeat the test. If the result remains elevated, further diagnostic investigation is warranted.

#### Can I run this test on a patient with concurrent disease or showing signs of illness?

Nu.Q<sup>®</sup> is suited for apparently healthy, asymptomatic dogs during routine wellness visits. Other conditions can elevate nucleosome concentrations, including but not limited to: immune-mediated disease, sepsis, trauma, and other inflammatory conditions.<sup>5,6</sup> This test cannot differentiate between inflammatory diseases and cancer. For this reason, we do not recommend using Nu.Q<sup>®</sup> to screen for cancer in patients who could have these types of diseases. Mild/chronic and well-managed conditions are less likely to cause elevated nucleosomes and therefore less likely to impact the results of the Nu.Q<sup>®</sup> test.



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### Can Nu.Q<sup>®</sup> be used to test patients on medications?

Certain medications like corticosteroids may affect nucleosome concentration and could therefore impact Nu.Q<sup>®</sup> results. Volition, the company that developed Nu.Q<sup>®</sup>, has suggested that other medications such as trazodone and nonsteroidal anti-inflammatories do not appear to interfere.

#### SAMPLE HANDLING

# What if I don't centrifuge the sample within 1 hour of the draw or don't pipette the plasma off immediately following centrifugation?

Delaying centrifugation or not separating the plasma from the remaining blood components immediately may cause erroneous Nu.Q<sup>®</sup> results. If a delay in these steps occurs, it is recommended to draw a new sample and centrifuge it within 1 hour. Immediately after centrifugation, harvest the plasma using the Antech-provided yellow 50 µL pipette.

#### What if I can't perform the Nu.Q® test right away?

For best results, harvested plasma should be assayed immediately. If that isn't possible, it is recommended to immediately harvest the plasma from the other layers after centrifugation and place it in a no additive tube. The sample should then be stored at 4°C (39.2°F). Harvested plasma can be stored at 4°C (39.2°F) for up to 24 hours. When ready to run the assay, remove the plasma from refrigeration, centrifuge it again for 10 minutes on the blood setting, and then run it immediately.

### Can I use different sample types other than EDTA plasma?

EDTA plasma is required for Nu.Q<sup>®</sup> testing using the Element i+<sup>®</sup> analyzer. Do not use whole blood, serum, or lithium heparin plasma for this test.

#### Will hemolysis, lipemia, and/or icterus affect results?

Moderate protein, bilirubin, cholesterol, and triglycerides in the sample will not interfere with the Nu.Q<sup>®</sup> test. Hemoglobin levels of up to 3 g/dL have shown no interference. Please see table below.

Interfering Substance	Conditions Tested	Nu.Q <sup>®</sup> Sample Concentrations Tested	Result
Protein	7 g/dL		
Bilirubin	15 mg/dL		
Cholesterol	350 mg/dL	45 ng/mL and 95 ng/mL	No significant effect
Hemoglobin	Up to 3 g/dL		
Triglycerides	250 mg/dL		

## Will it affect the results if the sample does not reach the EDTA tube's minimum fill line, or if, in the absence of a minimum fill line, the EDTA tube is less than ½ full?

Yes. As with most other diagnostics tests, it's vitally important for the sample tube to be filled properly to ensure accurate results.



## Can I use a different pipettor or different pipette tips?

It is recommended to use only the Antech-provided 50 µL pipette for Nu.Q<sup>®</sup> testing on the Element i+<sup>®</sup> analyzer. The Nu.Q<sup>®</sup> test cartridge was designed and validated with this specific pipette.

#### **RESULT INTERPRETATION**

#### Can Nu.Q® confirm cancer?

No. Nu.Q<sup>®</sup> cannot provide a definitive cancer diagnosis. Other conditions can elevate nucleosome concentrations, including but not limited to: immune-mediated disease, sepsis, trauma, and other inflammatory conditions.<sup>5,6</sup> Elevated Nu.Q<sup>®</sup> results should act as an impetus to perform additional diagnostics to obtain a diagnosis.

#### Can Nu.Q<sup>®</sup> rule out cancer?

No. Not all cancer types, stages, and presentations cause nucleosome elevation. A Low Suspicion Nu.Q<sup>®</sup> result doesn't rule out cancer, but it does suggest a lower likelihood of the presence of cancers that have been shown to increase nucleosome levels (e.g., lymphoma and hemangiosarcoma). If the Nu.Q<sup>®</sup> result is Low Suspicion and the clinician still suspects cancer, further diagnostics should be pursued. Nu.Q<sup>®</sup> results cannot predict whether a dog will develop cancer in the future or detect previously treated cancer.

### Can Nu.Q<sup>®</sup> tell me what type of cancer my patient has?

No. Nu.Q<sup>®</sup> cannot provide a definitive cancer diagnosis or information on cancer type. If cancer is suspected, it is recommended to continue down the diagnostic pathway to locate and diagnose the cause of the patient's elevated nucleosomes. Refer to the instructions in your Nu.Q<sup>®</sup> kit for more interpretation guidance.

## Can Nu.Q<sup>®</sup> detect all types of cancers and at all stages?

No. Not all cancer types, stages, and presentations cause nucleosome elevation.

### What types of cancer has Nu.Q® been able to detect?

Nu.Q<sup>®</sup> test has been shown to detect a variety of cancers – most notably lymphoma (77%), hemangiosarcoma (82%), and histiocytic sarcoma (54%) at 97% specificity.<sup>3</sup> Data also suggests the Nu.Q<sup>®</sup> test can detect some instances of mast cell tumors, osteosarcoma, oral melanoma, and soft tissue sarcoma. Please reach out to your Antech sales representative to access the current published studies.

### Is the baseline Nu.Q® test level prognostic?

Nu.Q<sup>®</sup> is not a prognostic test. The numerical value of a positive Nu.Q<sup>®</sup> test result does not necessarily directly correlate with disease stage or severity. Additionally, Nu.Q<sup>®</sup> results cannot predict whether a dog will develop cancer in the future. A dog's Nu.Q<sup>®</sup> value may also change over time as a result of cancer treatment or progression.<sup>7</sup>

#### How does Nu.Q® compare to CADET® BRAF?

Nu.Q<sup>®</sup> is a test that can detect and quantify nucleosomes in canine plasma. A high nucleosome concentration can indicate the presence of multiple types of cancer, but especially lymphoma and hemangiosarcoma.

CADET<sup>®</sup> BRAF is a PCR-based test that evaluates urine for a genetic mutation corresponding to urothelial carcinoma (also called transitional cell carcinoma).

Nu.Q<sup>®</sup> is intended for presumably healthy dogs with an increased risk of developing cancer and is best used as a targeted cancer screening test. But if there is a concern for cancer in the urinary tract, CADET<sup>®</sup> BRAF is warranted.



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## How does Nu.Q<sup>®</sup> compare to OncoK9<sup>®</sup>?

OncoK9<sup>®</sup> is no longer available, but like Nu.Q<sup>®</sup>, it was a test that evaluated canine blood samples for certain biomarkers that can indicate the presence of cancer. And similar to Nu.Q<sup>®</sup>, it was indicated for use in dogs at a high risk of developing cancer (e.g., older dogs and certain predisposed breeds).

Nu.Q®	OncoK9®	
Testing performed in-clinic, using the Element i+® analyzer	Samples sent to reference lab	
Low price	Moderately priced	
Results in minutes	Results in 10 – 14 days	
Requires 50 µL sample and uses standard EDTA/purple-top tubes	Required 15 mL sample and special Roche® tubes	
Tests for elevated nucleosome concentrations	Tested for cancer-associated genomic alterations in DNA	
Most likely to identify lymphoma and hemangiosarcoma	Most likely to identify lymphoma, hemangiosarcoma, and osteosarcoma	
Best suited for asymptomatic, presumably healthy dogs during wellness visits	Could be used to evaluate presumably healthy dogs and also to aid in diagnosing dogs in which cancer was suspected based on clinical findings	

## What should I do if I have questions about the results when I get them?

If you have any questions or concerns about the Nu.Q<sup>®</sup> test cartridge, patient results, or the function of the Element i+<sup>®</sup> analyzer, please contact Antech's In-House Diagnostics Technical Support team at VetSupport@heska.com or call 800.464.3752, option 3. Our technical support team is available by phone 24 hours a day.





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