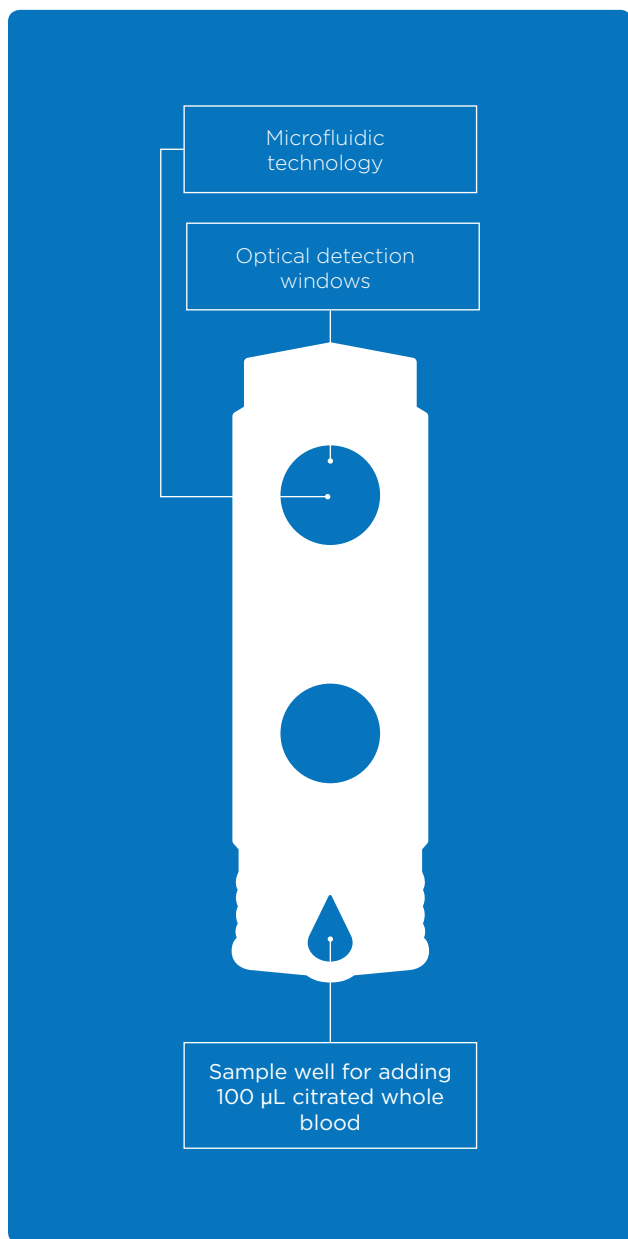


VETSCAN[®] VSpro

Utilization Guide





VSpro PT/aPTT Combination Test



Coagulation testing includes the evaluation of both prothrombin time (PT) and activated partial thromboplastin time (aPTT). Testing determines if a significant coagulation factor deficiency exists, and if so, which factor(s) are affected. PT is used to evaluate the extrinsic and common pathways, while aPTT is used to evaluate the intrinsic and common pathways.

The VETSCAN VSpro makes point-of-care coagulation testing easy, fast, and affordable. Immediate PT and aPTT test results offer numerous clinical benefits for veterinary patients and financial benefits for the veterinary practice.



Quick Reference Guide

The VETSCAN VSpro PT and aPTT Combination Test is used in conjunction with the VSpro Analyzer for fast, convenient, and cost-effective point-of-care coagulation testing. This test cartridge provides quantitative determination of PT and aPTT simultaneously from 100 µL of citrated canine or feline whole blood.

Cartridge Storage: Refrigerate at 2°C to 8°C (39°F to 46°F)

Cartridge Stability: Do not expose cartridges in the foil pouch to direct sunlight, and do not leave them at room temperature (15-30°C, 59-86°F) for more than 3 hours.

Use cartridges within 10 minutes of removing from pouch.

Reference Ranges (seconds):			Dynamic Ranges (seconds): ¹
Test/Species	Canine ¹	Feline ²	
PT	14-19	15-21	11-35
aPTT	75-105	86-137	30-200

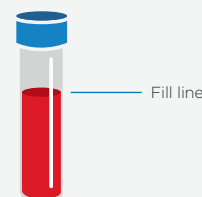


Sample Preparation

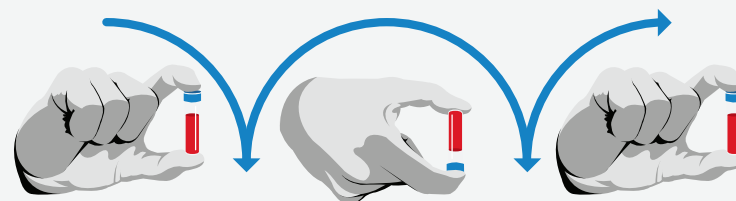
Precautions before blood sample collection:

- As with any blood test, the accuracy is dependent on the quality of the blood sample. Poor sample quality is caused by improper collection and handling techniques.
- Collect blood directly from the largest accessible vein.
- Contamination from thromboplastin, alcohol, and intravenous solutions will interfere with the coagulation assay.^{3,4}
- Hemolyzed samples can lead to testing errors and inconclusive results.
- The sodium citrate tube must be filled completely for accurate results.⁵

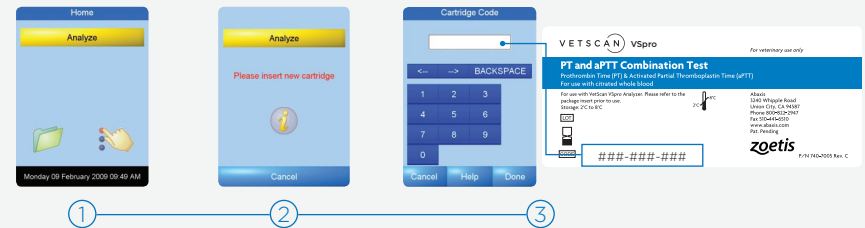
- 1 Fill an evacuated sodium citrate tube to the “**fill line**” with whole blood. The blood is drawn into the tube until the flow stops. For blood collected into a regular syringe, the needle should be removed prior to gently dispensing blood into the tube. If no fill line is seen, fill tube to top of label. **NOTE:** *Under-filled or over-filled citrate tubes may alter results due to the improper anticoagulant to sample ratio, most commonly resulting in a falsely prolonged aPTT.*



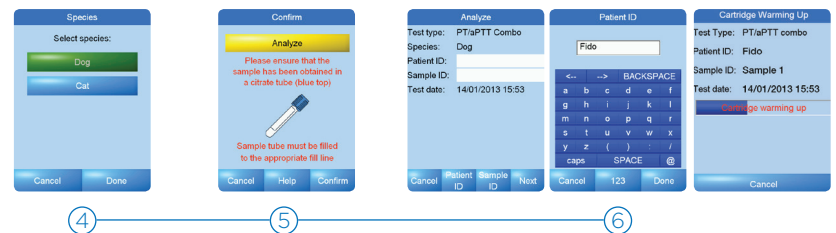
- 2 Gently invert the sodium citrate tube at least 10 times immediately after filling to ensure a good mixture with the anticoagulant. Let the sample sit for 5 minutes before testing to stabilize the mixture of blood and citrate. If the test is delayed, continue to invert every 10 minutes prior to the test. A blood rocker may be used. **NOTE:** *Blood may be tested up to two hours after it has been properly collected without affecting the test result.*



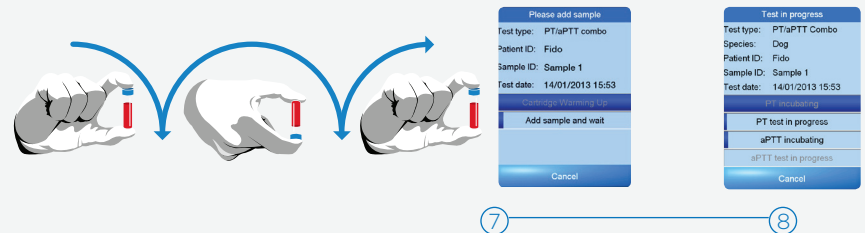
- 1 Remove the PT and aPTT Combination Test Cartridge from the refrigerator and open the pouch. Touch the **"Analyze"** button in the Home menu screen to start the test.
- 2 Insert a new test cartridge when the message **"Please insert new cartridge"** is displayed on the screen.
- 3 Enter the 9-digit **cartridge code** located on the pouch label and touch **"Done"**.



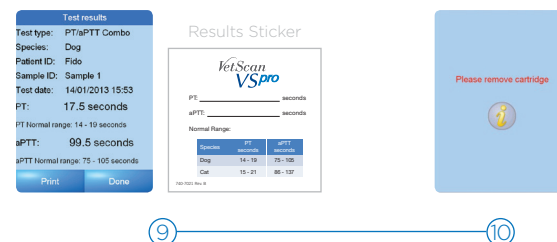
- 4 Select species and press **"Done"**.
- 5 Touch **"Confirm"** to acknowledge that the sample has been obtained in a sodium citrate (blue top) tube.
- 6 Enter **"Patient ID"** and/or **"Sample ID"** (optional). Touch **"Done"** to confirm and then **"Next"** to continue. The **"Cartridge Warming Up"** period will begin. **NOTE: Do not add sample while cartridge is warming up!**



- 7 When the analyzer beeps and flashes the message **"Add sample and wait"**, use the disposable pipette provided to add sample to the well. **NOTE: Prior to adding sample, gently invert sample tube 10X to obtain a uniform mixture for testing.**
- 8 Next screen shows **"Test in progress"**. It lasts approximately 10 minutes from the time the sample is put in to the well.



- 9 When analysis is complete, **"Test Results"** will appear. Use a **"Results Sticker"** (provided in the VSPRO Starter Kit and can be re-ordered: PN 740-7021) to put the test results into a file; print results if printer is attached; or save results to a computer through proper connectivity. (See the VSPRO manual for more information). The VSPRO stores up to 1000 test results. Press **"Done"** to exit the test result.
- 10 **"Please remove cartridge"** that has been used and dispose of it and all test material appropriately, and place the sealing cartridge in the cartridge slot.

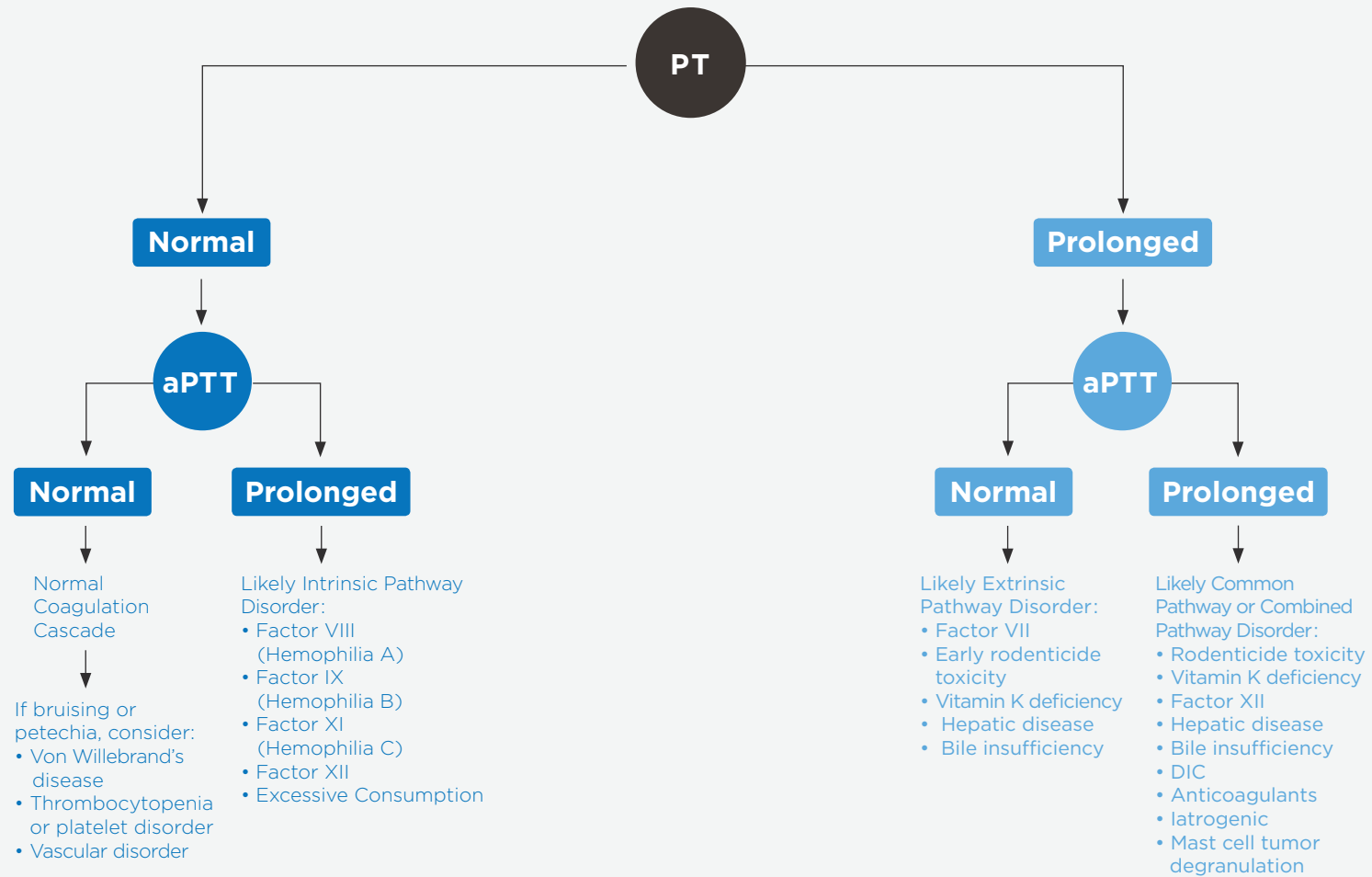


PT (Prothrombin Time):

Measures the Extrinsic and Common Pathways

aPTT (Activated Partial Thromboplastin Time):

Measures the Intrinsic and Common Pathways





Suggested Test Utilization

Pre-surgical testing

Pre-surgical testing should be considered for any patient, regardless of age. Inherited or congenital hemophilia cannot be determined through any other testing methods or on physical examination. Many of these deficiencies cause mild prolongation of clotting times without clinical signs.

Hemophilia A, or factor VIII deficiency, is the most common inherited coagulopathy of animals. Hemophilia A is most prevalent in German Shepard dogs. Hemophilia B, factor IX, deficiency affects cats and dogs. Other less common hereditary coagulation deficiencies have been reported in animals as well.⁷

Preventive care

Baseline values are as important for coagulation testing as they are for any other analyte. Stress, illness, injury, medications and surgery can all affect coagulation test results, so baseline values are vital for interpretation.

Hepatic disease⁸

Any patient with increased liver enzymes, possible hepatic dysfunction, or confirmed hepatopathy will benefit from coagulation testing. This becomes imperative should the patient require invasive surgery or biopsy/aspirate of an internal organ.

Liver disease can affect the coagulation cascade in multiple ways, as the liver produces most of the coagulation factors. Consider that:

- Many of the clotting factors are synthesized and cleared by the liver.
- Vitamin K is fat soluble, so its absorption depends on adequate bile production and flow.

Any disease state that affects the liver can lead to a coagulation abnormality including:

- Inflammation (hepatitis, cholangiohepatitis)
- Neoplasia
- Biliary stasis
- Use of chronic medications (NSAIDs, anesthetics, chemotherapeutics, etc.)

Vitamin K deficiency or antagonism

Vitamin K is an essential cofactor for coagulation factors II, VII, IX and X. Factor VII has the shortest half-life and will deplete the earliest, therefore, PT is often prolonged first.⁹

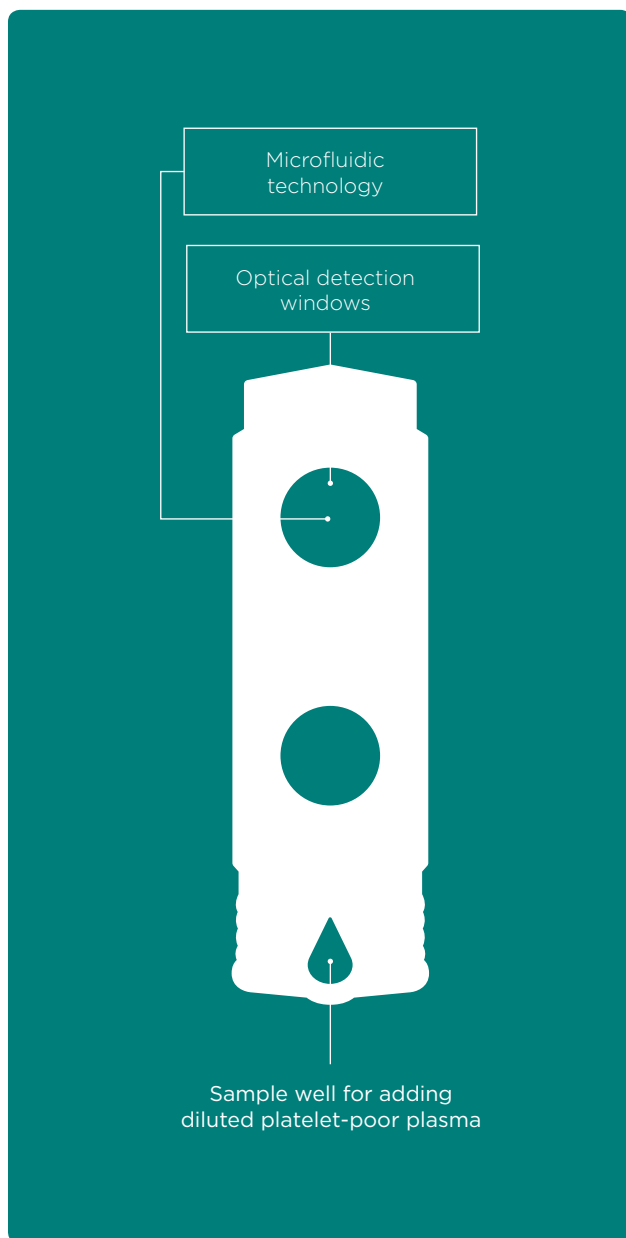
Some causes of Vitamin K deficiency are:¹⁰

- Rodenticide toxicity
- Cholestatic liver disease (reduced bile flow reduces absorption)
- Liver Failure
- Malabsorption disorders
- Medications

Other disease states where coagulation testing is indicated:

- Any patient with unexplained bleeding, bruising or petechial hemorrhage
- Snake bite
- Infectious disease
- Immune-mediated disease
- Shock or severe systemic disease; potential for DIC (disseminated intravascular coagulopathy)
- Actively bleeding patients





VSpro Fibrinogen Test



Quick Reference Guide

The VETSCAN VSpro Fibrinogen Test cartridge is used in conjunction with the VSpro Analyzer to evaluate the fibrinogen level in horses, a sensitive and specific marker for inflammation. Early recognition of systemic inflammation is essential for diagnosis of disease and formulating a treatment plan. Quantitative serial testing for fibrinogen can help effectively monitor treatment and the response to therapy.

Cartridge Storage: Refrigerate at 2°C to 8°C (36°F to 46°F)

Cartridge Stability: Do not expose cartridges in the foil pouch to direct sunlight, and do not leave them at room temperature (15-30°C, 59-86°F) for more than 3 hours.

Use cartridges within 10 minutes of removing from pouch.

Reference Range: ^{11, 12}			Dynamic Range: ^{11, 12}	
	g/L	mg/dL	g/L	mg/dL
Equine	1.5 - 4.0	150 - 400	0 - 20	0 - 2000



Sample Preparation

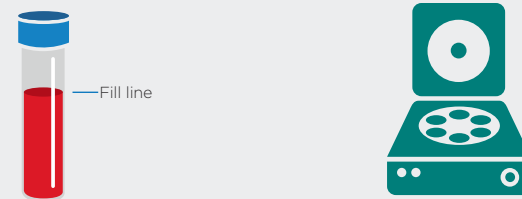
Precautions before blood sample collection:

- As with any blood test, the accuracy is dependent on the quality of the blood sample. Poor sample quality can be caused by improper collection and handling techniques.
- Collect blood directly from the largest accessible vein.
- Contamination from thromboplastin, alcohol and intravenous solutions will interfere with the fibrinogen assay.
- Use of hemolyzed samples may result in lower than expected values.
- Ensure precision when mixing platelet-poor plasma and diluent, and only use the pre-filled microtube supplied.
- The platelet-poor plasma and diluent solution must be mixed gently. Do not use a vortex mixer, and do not shake the sample
- The tube must be filled completely for accurate results.

- 1 Fill an evacuated sodium citrate tube to the “**fill line**” with whole blood. The blood is drawn into the tube until the flow stops. For blood collected into a regular syringe, the needle should be removed prior to placing blood into the sodium citrate tube. Do not place sample on a rocker.

NOTE: Under-filled or over-filled citrate tubes may alter results due to the improper anticoagulant-to-sample ratio.

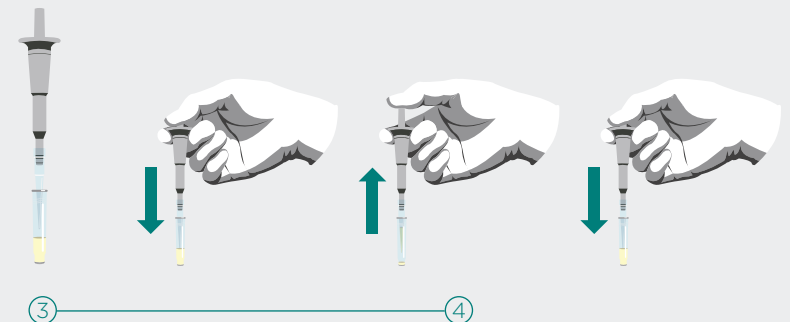
- 2 Spin the citrated whole blood sample to platelet-poor plasma using a centrifuge. Refer to package insert for centrifuge requirements.



① ————— ②

- 3 Use a pipette and one of the supplied pipette tips to extract 100 μ L of platelet-poor plasma from the centrifuged sample tube. Deposit the 100 μ L of platelet-poor plasma into the pre-filled diluent tube by piercing the foil lid with the pipette tip.

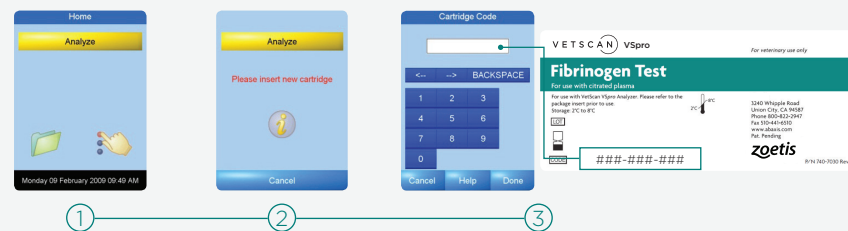
- 4 Gently mix the diluent and sample by extracting and depositing the mixture using the pipette. The sample is now ready for testing.



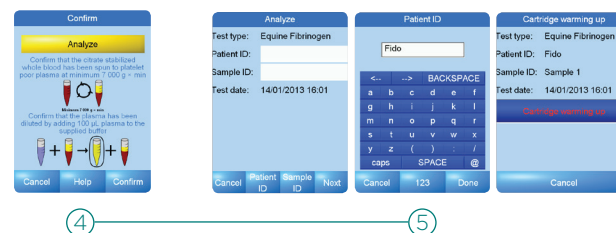


Test Procedure

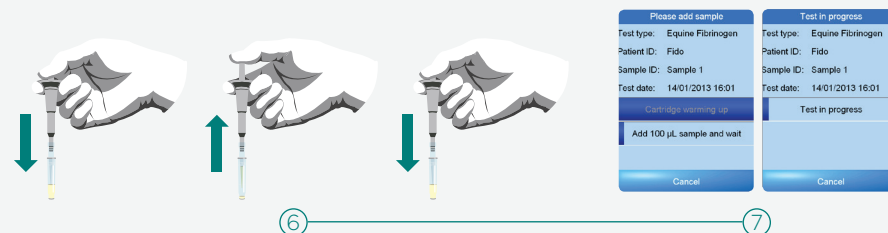
- 1 Remove the Fibrinogen Test Cartridge from refrigeration 10-15 minutes prior to analysis to warm up to room temperature. Touch the **"Analyze"** button in the Home menu screen to start the test.
- 2 Insert a new test cartridge when the message **"Please insert new cartridge"** is displayed on the screen.
- 3 Enter the 9-digit **cartridge code** located on the pouch label and touch **"Done"**.



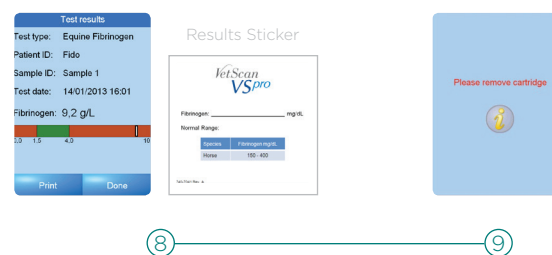
- 4 Touch **"Confirm"** to acknowledge that the sample has been obtained in a sodium citrate tube, spun down to platelet-poor plasma and diluted by adding 100 μ L of the plasma to the supplied pre-filled diluent tube.
- 5 Enter **"Patient ID"** and/or **"Sample ID"**. Touch **"Done"** to confirm and then **"Next"** to continue. The **"Cartridge Warming Up"** period will begin. **NOTE:** Do not add sample while cartridge is warming up!



- 6 When the analyzer beeps and flashes the message **"Add 100 μ L sample and wait"**, use pipette and disposable pipette tip to add 100 μ L of sample to the well. **NOTE:** Prior to adding sample, extract and re-deposit the sample in the tube to obtain a uniform mixture of plasma and diluent for testing. Do not introduce bubbles to the mixture.
- 7 Next screen shows **"Test in progress"**. It lasts approximately 15 minutes from the time the sample is put in to the well.



- 8 When analysis is complete, **"Test Results"** will appear. Use a **"Results Sticker"** (provided in the VSpro Starter Kit and can be re-ordered: PN 740-7041) to put test results in to a file; print results if printer is attached; or save results to a computer through proper connectivity. (See the VSpro manual for more information). The VSpro automatically stores up to 1000 test results. Press **"Done"** to exit the test result



- 9 **"Please remove cartridge"** that has been used and dispose of it and all test material appropriately, and place the sealing cartridge in the cartridge slot.



Suggested Test Utilization

Fibrinogen testing side-by-side with complete blood count (CBC)

Fibrinogen concentrations should be included when performing a CBC in the horse.

- Fibrinogen is an acute phase protein made in the liver that detects inflammation.
- Fibrinogen increases 24 hours after an inflammatory process has started and peaks between 4 and 7 days.
- Fibrinogen may indicate inflammation in the subclinical horse when the leukogram is within normal limits.
- Fibrinogen levels are very sensitive, and even a mild increase should be considered significant.

Systemic inflammatory marker

Early recognition of systemic inflammation using fibrinogen is essential for formulating an effective treatment plan. Inflammation that goes unrecognized can impair a patient's growth and performance.

- Fibrinogen blood levels are directly related to the severity of the underlying condition.
- Fibrinogen is the preferred inflammatory marker for early detection (subclinical) of *Rhodococcus equi* pneumonia.¹³
- Fibrinogen increases more quickly in synovial fluid than serum amyloid A (SAA) (by several hours) following endotoxin administration into the joint.¹⁴
- When tested serially, fibrinogen provides information regarding treatment efficacy and length, prognosis, especially considering such infectious or inflammatory conditions as: *R. equi* infections, strangles, pigeon fever, pleuropneumonia, abdominal abscess, septic arthritis, endometritis, clostridial myonecrosis, and endocarditis.

Fibrinogen specificity

- Plasma fibrinogen may increase with dehydration and may decrease with severe hepatic disease due to decreased protein production.
- In cases of DIC, fibrinogen may be decreased due to increased utilization (masking the hyperfibrinogenemia associated with the inflammatory process).

Patient monitoring tool

- The degree of hyperfibrinogenemia approximates the severity of disease.
- When used, in conjunction with the clinical findings and other laboratory data, the most effective treatment protocol and prognosis can be monitored closely.
- Fibrinogen levels may be used alone in determining resolution of the inflammatory process when the leukogram is unchanged or has returned to normal.

Plasma Protein:Fibrinogen (PP:F) Ratio

Useful in the interpretation of fibrinogen concentration when hyperproteinemia is present. This ratio is calculated as:

$$\text{PP:F Ratio} = \frac{(\text{plasma TP mg/dl} - \text{fibrinogen mg/dl})}{\text{fibrinogen mg/dl}}$$

Interpretation:¹⁵

<10
Inflammation

10-15
Normal

>15
Dehydration

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¹Data on file, Study No. TI-03294, Zoetis Inc.

²Data on file, Study No. TI-04203, Zoetis Inc.

³Lippi G, Plebani M, Favalaro E. Interference in coagulation testing: focus on spurious hemolysis, icterus, and lipemia. *Seminars In Thrombosis And Hemostasis*. 2013 Apr;39(3):258-266.

⁴Lippi G, Salvagno GL, Montagnana M, Lima-Oliveira G, Guidi GC, Favalaro E. Quality Standards for Sample Collection in Coagulation Testing. *Seminars In Thrombosis And Hemostasis*. 2012;38:565-575.

⁵Monti P, Archer J. Quality assurance and interpretation of laboratory data [Chapter 2]. *BSAVA Manual of Canine and Feline Clinical Pathology*. 2016: 12.

⁶Topper MJ, Welles EG. Hemostasis [Chapter 4]. *Duncan and Prasse's Veterinary Laboratory Medicine: Clinical Pathology*. 2003: 99-135.

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⁸Bain PJ. Liver [Chapter 7]. *Duncan and Prasse's Veterinary Laboratory Medicine: Clinical Pathology*, Fifth Edition. 2011: 224.

⁹Topper MJ, Welles EG. Hemostasis [Chapter 4]. *Duncan and Prasse's Veterinary Laboratory Medicine: Clinical Pathology*, Fifth Edition. 2011: 136.

¹⁰Stokol T. Disorders of haemostasis [Chapter 6]. *BSAVA Manual of Canine and Feline Clinical Pathology*. 2016: 116.

¹¹Data on file, Study No. TI-03300, Zoetis Inc.

¹²Epstein KL, Brainard BM. An evaluation of the Abaxis VSPPro for the measurement of equine plasma fibrinogen concentrations. *Equine vet. J.* 2012; 44(4):449-452.

¹³Passamonti, F et al. *Rhodococcus equi* pneumonia in foals: an assessment of the early diagnostic value of serum amyloid A and plasma fibrinogen concentrations in equine clinical practice. *Vet J.* 2015 Feb;203(2):211-8.

¹⁴Andreassen, M et al. Changes in concentrations of haemostatic and inflammatory biomarkers in synovial fluid after intra-articular injection of lipopolysaccharide in horses. *BMC Vet Res.* 2017 Jun 19;13(1):182.

¹⁵Robinson NE, Sprayberry K. *Current Therapy in Equine Medicine*, Sixth Edition. 2008: 957.

