

Canine Parvovirus Antigen Test Kit

ASSURE®/PARVO

For the Detection of Canine Parvovirus Antigen

DIRECTION INSERT

REFERENCES

Parrish, C.R. et al: Antigenic relationships between canine parvovirus type 2, feline panleukopenia virus and mink enteritis virus using conventional antisera and monoclonal antibodies. *Arch. Virol.* 72:267-278; 1982.

Studdart, M.J. et al: Aspects of the diagnosis, pathogenesis and epidemiology of canine parvovirus. *Aust. Vet. J.* 60:197-200; 1983.

Siegl, G. et al: Characteristics and Taxonomy of Parvoviridae. *Intervirology.* 23:61-73; 1985.

Parrish, C.R.: Emergence and natural history of canine, mink and feline parvoviruses. *Adv. Virus Res.* 38:403-450; 1990.

I. INTRODUCTION

Canine Parvovirus (CPV) is a member of the feline parvovirus subgroup. It is closely related to feline panleukopenia virus and mink enteritis virus. CPV was first recognized as a virus in dogs in 1978 in North America. It has since spread globally and is considered endemic to nearly all populations of domesticated and wild canines.

CPV causes two forms of disease: Myocarditis and enteritis. Due to maternal antibody protection the myocardial form is rare. The enteric form, however, is prevalent and can be fatal to puppies and geriatric dogs. CPV enteritis causes severe, often bloody diarrhea, vomiting, leukopenia and dehydration.

Transmission is fecal-oral and most infections occur from exposure to contaminated feces. CPV is highly contagious and stable under a variety of environmental conditions.

Rapid diagnosis of CPV allows for quarantine and prompt treatment of infected dogs. Diagnosis may be difficult in milder cases. ASSURE®/PARVO is an enzyme

linked immunosorbent assay (ELISA) which detects all strains of Canine Parvovirus shed in the feces. Positive results with the fecal ELISA indicate the presence of canine parvovirus.

II. TEST PRINCIPLES

The bulbous ends of the plastic wands have been coated with antibody to CPV. A second antibody directed against a specific CPV antigen is conjugated to the enzyme horseradish peroxidase (HRP). The fecal sample is incubated simultaneously with both the antibody-coated wand and enzyme-labeled antibodies. If antigen is present, it is captured by the wand. The enzyme-labeled antibodies are in turn captured by the antigen bound to the wand. The unbound enzyme-labeled antibody and feces are removed during the wash step and the wand is placed into a chromogenic substrate.

The development of a distinct blue color in the solution indicates the presence of CPV antigen. In the absence of CPV, no color will develop.

III. CONTENTS OF ASSURE®/PARVO TEST KIT

	10 Test	25 Test
Anti-CPV Antibody		
Coated Wands	10 ea	25 ea
Predispensed HRP-Monoclonal Antibody Conjugate (A Tubes)	10 ea	25 ea
Predispensed Substrate Buffer (B Tubes)	10 ea	25 ea
Bottle C-Chromogen (White Cap)	2.5 mL	3.0 mL
Materials required, but not provided:		
Marking Pen		
Timer		
Distilled or deionized water		
Wash Bottle		

IV. PRECAUTIONS

1. Allow kit to come to room temperature (21°-25° C; 70°-78° F) prior to use; approximately one hour.
2. Do not expose kit to direct sunlight.
3. Do not use expired reagents or mix from different kit serials.
4. Hold reagent vial vertically for proper drop volume.

5. Vaccination with modified live CPV vaccines may cause shedding of viral particles in the feces 4-10 days post-vaccination. This can cause a weak positive result.

6. Dispose of potentially infected specimens appropriately.

7. FOR USE IN TESTING CANINE SPECIMENS ONLY.

V. SAMPLE INFORMATION

Canine fecal material is required. Stool samples may be stored at 2°-7°C (36°-45°F) for 48 hours. If longer storage is required the samples may be stored frozen.

VI. STORAGE AND STABILITY

Store the test kit at 2°-7°C (36°-45°F). Do not freeze. Properly stored reagents are stable until the expiration date.

FOR TECHNICAL ASSISTANCE:
1-800-228-4305

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ASSURE®/PARVO Test Procedure

NOTE: Use canine fecal samples.

Prior to use, allow kit components to come to room temperature (70° to 78° F; 21° to 25° C).

For each sample you will need:	Anti-CPV Antibody Coated Wand. Label with dog's ID.	Predispensed HRP-Monoclonal Antibody Conjugate Tube A.	Predispensed Substrate Buffer Tube B. Label with dog's ID.	Workstation with Reagent C.	Squirt Bottle with Distilled or Deionized Water.
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A. SAMPLE COLLECTION

1A.

- Pre-wet bulbous end of Wand with deionized or distilled water for 3-5 seconds.



1B.

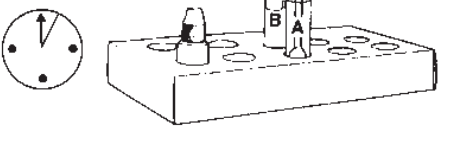
- Swirl Wand in fecal material 3-5 seconds to cover bulbous end with a thin coat of feces.

NOTE: A specimen may be obtained rectally by inserting bulbous end of wand into rectum and **GENTLY** swirling 3-5 seconds. **Do not use a lubricant.**

B. CONJUGATE INCUBATION

2.

- Place bulbous end of Wand in Tube A.
- Twirl Wand **vigorously** until fecal material is suspended in the liquid.
- WAIT 5 MINUTES.**



C. PREPARE B TUBE

3.

During waiting period:

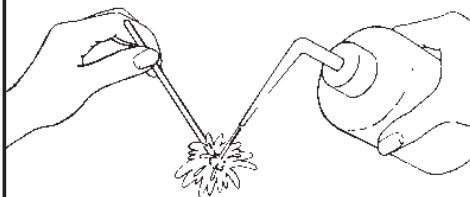
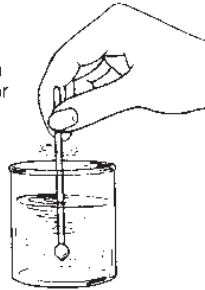
- Remove stopper from Tube B.
- Add 3 drops Bottle C (White Cap) to Tube B.
- Tap to mix.
- Set tube aside for use in Step 5.



D. WASH STEP.

4.

- Remove wand from Tube A.
- Wash bulbous end and tip of wand by swirling/swishing **vigorously** in a cup containing at least 250 mL of deionized or distilled water for a **minimum of 15 seconds.**
- After swirling in cup, continue to wash wand by directing a **forceful stream** of deionized or distilled water against bulbous end and tip of wand and work up handle. Wash until all fecal material is removed from the wand.
- Shake off excess water.
- Repeat washing with a forceful stream **5-7 times.** This will ensure removal of unbound proteins which may contribute to non-specific color development.
- Replace liquid in cup between wands.



NOTE: Bloody samples may require saline rinse.

E. COLOR DEVELOPMENT

5.

- Place washed Wand in Tube B.
- Twirl 1-3 seconds to mix.
- WAIT 5 MINUTES.**



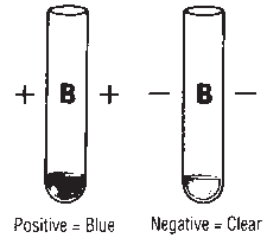
- Remove Wand.
- READ RESULTS.**



F. INTERPRETATION OF RESULTS

6.

- Observe solution against workstation window or a white background for blue color.



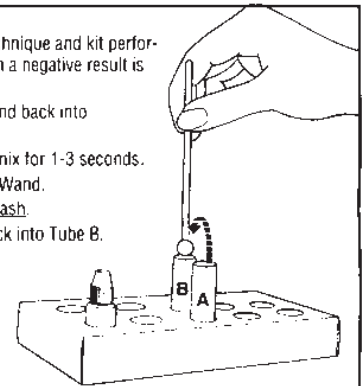
Positive = Blue Negative = Clear

NOTE: Color intensity will vary with level of CPV present.

OPTIONAL PROCEDURAL CONTROL

To verify technique and kit performance when a negative result is obtained:

- Place wand back into Tube A.
- Twirl to mix for 1-3 seconds.
- Remove Wand.
- Do not wash.**
- Place back into Tube B.



Blue color will develop within 1 minute indicating reagents were added correctly and kit is performing properly. If color does not develop, repeat the test. (This is a procedure and reagent check only. CPV antigen is not present.)

GOOD TECHNIQUES = ACCURATE RESULTS

- A thin coat of fecal material on the bulbous end is required. Swirl bulbous end of wand in material for 3-5 seconds.
- In Step 2, feces should be removed from the Wand and suspended in the liquid.
- Washing is the most important step. Wands cannot be overwashed.** Underwashing will result in non-specific color development in the Tube B.
- Read results at 5 minutes. If no color is seen at 5 minutes, the sample is negative.
- Do not use the test kit past the expiration date and do not intermix components from different serial numbers.
- Store kit at 2° to 7° C (36° to 45° F). Allow kit to come to room temperature before use.
- Failure to change wash solution in Step 4 can lead to false color development.

FOR TECHNICAL ASSISTANCE: 1-800-228-4305