SENSITIVITY AND SPECIFICITY OF RAPIDE - DISTEMPER - ADENOVIRUS AG TEST KIT

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1. Abstract / Résumé

Rapide - Distemper - Adenovirus Ag Test Kit is a new *in-vitro* immunochromatographic one step assay for detection of Distemper and Adenovirus antigens in canine conjunctiva and nasal cavity epithelial cells. Five (5) studies were conducted to check the sensitivity and specificity of this new diagnostic test:
In the first study, **Rapide - Distemper - Adenovirus Ag Test Kit** was compared to Peroxidase Linked assay (PLA) for CDV antigen detection. Results showed a sensitivity of 98.5% (65/66).
In the second study, **Rapide - Distemper - Adenovirus Ag Test Kit** was compared to PCR for CDV antigen detection. Results showed a sensitivity of 98.8% (85/86) and a specificity of 97.7% (129/132).
In the third study, **Rapide - Distemper - Adenovirus Ag Test Kit** was compared to a competitive product already on the market for CDV antigen detection. Results showed a sensitivity of 100% (88/88) and a specificity of 100% (130/130).
In the third study, **Rapide - Distemper - Adenovirus Ag Test Kit** was compared to PCR for CAV antigen detection. Results showed a sensitivity of 94.4% (17/18) and a specificity of 97.7% (42/43).
In the last study, **Rapide - Distemper - Adenovirus Ag Test Kit** was evaluated for potential cross-reactivity with Canine Parvovirus, Canine Parainfluenza Virus, Porcine Parvovirus, Leptospira interogans serovar icterohaemorragiae, *E. coli* sp., Salmonella sp, Ascaris homogenates, Giardia cyst homogenate & Canine influenza virus. The results showed that **Rapide - Distemper - Adenovirus Ag Test Kit** is highly specific for the canine Distemper and Adenoviris antigens with no cross-reaction with any of the other common canine infectious pathogens.
Data from these studies show that **Rapide - Distemper - Adenovirus Ag Test Kit** is highly sensitive and specific for detection of canine Distemper and Adenovirus antigens in conjunctiva and nasal cavity epithelial cells. Therefore, **Rapide - Distemper - Adenovirus Ag Test Kit** is recommended to be adopted in animal clinics for diagnosis of canine Distemper infection, canine infectious hepatitis or kennel cough.

Rapide - Distemper - Adenovirus Ag Test Kit *est un nouveau test d'immuno-migration in-vitro pour la détection de des antigènes du virus de la maladie de Carré et de l'Adénovirus dans des cellules de la conjonctive et de la muqueuse nasale du chien. Cinq (5) études étaient conduites pour vérifier la sensibilité et la spécificité de ce nouveau test de diagnostic:*
Dans la première étude, ***Rapide - Distemper - Adenovirus Ag Test Kit*** *était comparé au test Peroxidase liée (PLA) pour la détection de l'antigène du virus de la maladie de Carré. Les résultats montrent une sensibilité de 98.5% (65/66).*
Dans la deuxième étude, ***Rapide - Distemper - Adenovirus Ag Test Kit*** *était comparé au test PCR pour la détection de l'antigène du virus de la maladie de Carré. Les résultats montrent une sensibilité de 98.8% (85/86) et une spécificité de 97.7% (129/132).*
Dans la troisième étude, ***Rapide - Distemper - Adenovirus Ag Test Kit*** *était comparé à un test compétiteur pour la détection d'antigènes du virus de la maladie de Carré. Les résultats montrent une sensibilité de 100% (88/88) et une spécificité de 100% (130/130).*

*Dans la quatrième étude, **Rapide - Distemper - Adenovirus Ag Test Kit** était comparé au test PCR pour la détection de l'antigène de l'Adénovirus. Les résultats montrent une sensibilité de 94.4% (17/18) et une spécificité de 97.7% (42/43).*
*Dans la dernière étude, **Rapide - Distemper - Adenovirus Ag Test Kit** était évalué pour des réactions croisées potentielles avec le Parvovirus canin, le virus du parainfluenza canin, le Parvovirus porcin, Leptospira interogans serovar icterohaemorragiae, E. coli sp., Salmonella sp,* homogenates d'Ascaris, *homogenates de Giardia et le Virus canin de l'influenza. Les résultats montrent que **Rapide - Distemper - Adenovirus Ag Test Kit** est extrêmement spécifique pour les antigènes du virus de la maladie de Carré et de l' Adénovirus sans aucune réaction croisée avec d'autres pathogènes infectieux canins.*
*Les données de ces études montrent que **Rapide - Distemper - Adenovirus Ag Test Kit** est extrêmement sensible et spécifique pour la détection des antigènes du virus de la maladie de Carré et de l' Adénovirus dans des cellules de la conjonctive et de la muqueuse nasale du chien. Par conséquent **Rapide - Distemper - Adenovirus Ag Test Kit** est recommandé en clinique vétérinaire pour le diagnostic de l'infection du de la maladie de Carré, de l'hépatite infectieuse canine ou de la toux de chenil.*

2. Introduction

2.1 Introduction – Canine distemper is a highly contagious, systemic, viral disease of dogs. It is clinically characterized by a diphasic fever, leukopenia, respiratory catarrh with mucous and watery secretions discharged from the eyes and noses as well as frequently pneumonic and neurologic complications. Canine adenovirus has two types of virus that can both be harmful to dogs. Canine Adenovirus type 1 or Infectious canine hepatitis (ICH) is a worldwide, contagious disease of dogs with signs that vary from a slight fever and congestion of the mucous membranes to severe depression, marked leukopenia, and prolonged bleeding time. Canine Adenovirus type 2 causes kennel cough that affects respiratory organs.

2.2 Background - The **Rapide - Distemper - Adenovirus Ag Test Kit** is an *in-vitro* immunochromatographic one step assay designed for qualitative determination of Canine Distemper and Adenovirus antigens in conjunctiva and nasal cavity epithelial cells.

2.3 Objectives - The primary objective was to determine the sensitivity and specificity of **Rapide - Distemper - Adenovirus Ag Test Kit**.

3. Study design

3.1 Study 1: This is a parallel comparative study. Sixty-six (66) positive samples from clinical cases were collected and tested by Peroxidase Linked Assay (PLA), a laboratory reference standard test to confirm their positive status for canine Distemper virus. All samples were then tested by **Rapide - Distemper - Adenovirus Ag Test Kit** to evaluate the sensitivity compared to PLA.

3.2 Study 2: This is a parallel comparative study. Two hundred-eighteen (218) samples from clinical cases were collected and tested by RT-PCR, a laboratory reference standard test to confirm their positive and negative status for canine Distemper virus. All samples were then tested by **Rapide - Distemper - Adenovirus Ag Test Kit** to evaluate the sensitivity and specificity compared to RT-PCR.

3.3 Study 3: In this study, **Rapide** - Distemper-Adenovirus Ag Test Kit was compared to a competitive leading brand, for detection of CDV antigens. Two hundred-eighteen (218) samples from clinical cases were collected and tested by RT-PCR for canine Distemper virus. All samples were then tested by both **Rapide - Distemper - Adenovirus Ag Test Kit** and the competitive kit for CDV Ag to evaluate the sensitivity and specificity compared to RT-PCR.

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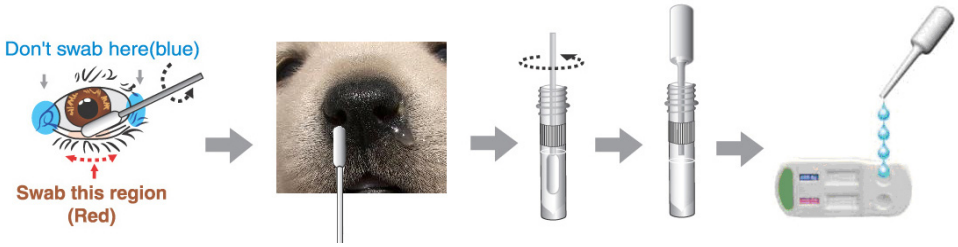
3.4 Study 4: This is a parallel comparative study. sixty-one (61) samples from clinical cases were collected and tested by RT-PCR for canine adenovirus. All samples were then tested by **Rapide - Distemper - Adenovirus Ag Test Kit** to evaluate the sensitivity and specificity compared to RT-PCR.

3.5 Study 5: In this study, **Rapide - Distemper - Adenovirus Ag Test Kit** was evaluated for potential cross-reactivity with other canine infectious pathogens which are commonly present in fecal materials: Canine Parvovirus, Canine Parainfluenza Virus, Porcine Parvovirus, Leptospira interogans serovar icterohaemorragiae, *E. coli* sp., Salmonella sp, Ascaris homogenates, Giardia cyst homogenate & Canine influenza virus.

4. Materials & methods:

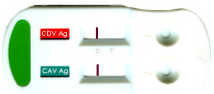
4.1 The **Rapide - Distemper - Adenovirus Ag Test Kit** is an *in-vitro* immunochromatographic one step assay designed for qualitative determination of Canine Distemper and Adenovirus antiges in conjunctiva and nasal cavity epithelial cells. The study investigators were instructed to process the samples as indicated in the figure below:

[Figure for test procedures]

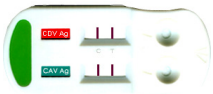


4.2 The Interpretation of the test results is conducted as follows:

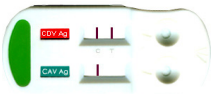
Negative Result:



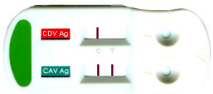
Simultaneous CDV and CAV Positive



CDV Positive



CAV Positive



5. Analysis

5.1 Sensitivity and specificity will be determined by standard calculation:

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Sensitivity(%) = 100 X $\frac{\text{No. of specimens with positive results by Rapide - Distemper - Adenovirus Ag Test Kit}}{\text{No. of positive specimens confirmed by PLA or PCR}}$

Specificity(%) = 100 X $\frac{\text{No. of specimens with negative results by Rapide - Distemper - Adenovirus Ag Test Kit}}{\text{No. of negative specimens confirmed by PCR}}$

6. Results:

6.1 Study 1:

Sensitivity for CDV Ag		Gold Standard: PLA	
		Positive	Negative
Rapide - Distemper - Adenovirus Ag Test Kit	Positive	65	0
	Negative	1*	0
Sensitivity	98.5% (65/66)		

6.2 Study 2:

Sensitivity and Specificity for CDV Ag		Gold Standard: PCR	
		Positive	Negative
Rapide - Distemper - Adenovirus Ag Test Kit	Positive	85	3
	Negative	1	129
Sensitivity	98.8% (85/86)		
Specificity	97.7% (129/132)		

6.3 Study 3:

Sensitivity and Specificity for CDV Ag		Competitive Kit	
		Positive	Negative
Rapide - Distemper - Adenovirus Ag Test Kit	Positive	88	0
	Negative	0	130
Sensitivity	100% (88/88)		
Specificity	100% (130/130)		

6.4 Study 4:

Sensitivity and Specificity for CAV Ag		Gold Standard: PCR	
		Positive	Negative
Rapide - Distemper - Adenovirus Ag Test Kit	Positive	18	1
	Negative	1	42
Sensitivity	94.4% (17/18)		
Specificity	97.7% (42/43)		

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6.5 Study 5:

Pathogens	Titer	Results
Canine parvovirus	$10^{4.0}TCID_{50}$/ml	Negative
Canine parainfluenza virus	$10^{4.0}TCID_{50}$/ml	Negative
Porcine parvovirus	$10^{6.0}TCID_{50}$/ml	Negative
Leptospira icterohaemorrhagiae	OD 1.0	Negative
E. coli spp.	$10^{8.0}$ CFU/ml	Negative
Salmonella spp.	$10^{8.0}$ CFU/ml	Negative
20% *Ascaris* homogenates	10%	Negative
Giardia cyst suspension	10%	Negative
Canine Influenza virus	2^{7} HI	Negative

7. Discussion and conclusion:

These studies showed that **Rapide - Distemper - Adenovirus Ag Test Kit** is highly sensitive and specific for the qualitative detection of Distemper and Adenovirus antigen in conjunctiva and nasal cavity epithelial cells. Using **Rapide - Distemper - Adenovirus Ag Test Kit** is very simple, quick and it does not require any special equipments. Therefore, **Rapide - Distemper - Adenovirus Ag Test Kit** is recommended to be adopted in animal clinics for diagnosis of canine Distemper infection, canine infectious hepatitis and kennel cough.