Rockeby Human Influenza A Antigen Test Kit

REF RBL-302 (10 Tests)

REVISION DATE: 05/06
LM074-ENG-0

IVD

INTENDED USE

The **Rockeby Human Influenza A Antigen Test** is a rapid, one step chromatographic *in vitro* immunoassay for the qualitative detection of influenza virus antigen type A in human specimens, using either direct swab or liquid samples.

INTRODUCTION

Procedures currently used to diagnose influenza Type A infection(s) include serological assay, hemagglutination inhibition, polymerase chain reaction, direct specimen immunofluorescence assay (IFA) and culture isolation with a confirmation procedure. The latter is considered the standard method and employs initial viral isolation in cell culture followed by hemadsorption inhibition, immunofluoresence, or neutralization assay to confirm the presence of the Influenza A virus. These methods take more time to get the results, which could be done only in authorized lab centers.

The **Rockeby Human Influenza A Antigen test** is a rapid immunochromatographic assay, easy to use for screening infected patients, only positive samples need be sent to lab centers for confirmation. This rapid test could help in controlling the epidemic of the disease.

PRINCIPLE OF ASSAY

The **Rockeby Human Influenza A Antigen Test** kit is a qualitative, one step chromatographic immunoassay to selectively detect the Influenza A virus with a high degree of sensitivity. In the test procedure, sample is absorbed through an absorbent membrane and allowed to migrate through the membrane. As the sample proceeds through the membrane, the colored conjugate (colloidal gold conjugate), which was pre-dried on the test strip, migrates with the sample. The sample and the conjugate move through the capture region, precoated with immobilised monoclonal antibody to Influenza A virus on the test band region and Protein A on the control band region, and then to the end of the membrane. The bound antibody-antigen complexes are detected by giving a pink-purplish color. The control line contains protein A which binds with the dye conjugate. The control band serves as an indication of proper sample addition and migration plus reagent control.

The format provides a clear read out for positive (two lines) and negative (one line) specimens. The appearance of one line for negative specimens gives an added measure of quality control by demonstrating antibody recognition, assuming that the procedure was done directly and that the reagents are chemically active.

MATERIALS PROVIDED

Rockeby Human Influenza A antigen test device Test devices packed in individual sealed aluminum pouch with desiccant. Store at 2°C ~ 30°C.	10 devices
Assay diluent 5 mls each. Store at 2°C ~ 30°C.	2 bottles
Specimen tubes Plastic tubes, each marked at the 1.0 ml. fill volume.	10 tubes
Sterile cotton swabs Cotton bud wrapped sample collection sticks.	10 pieces
Disposable droppers Ten molded plastic droppers packed in a polybag.	10 pieces
Instruction Sheet	1 copy

PRECAUTIONS

⚠ HEALTH AND SAFETY INFORMATION

1. In case of an accident or contact with eyes, rinse immediately with plenty of water and seek medical advice.
2. Consult a physician immediately in the event that contaminated materials are ingested or come in contact with open lacerations, or other breaks in the skin.
3. Wipe any spills of samples promptly with 1% sodium hypochlorite solution.
4. Autoclave all used and contaminated materials at 121°C, 15 p.s.i. for 30 minutes before disposal. Alternatively, decontaminate materials in 5% sodium hypochlorite solution for 30 - 60 minutes before disposal in biohazard waste-bags.

ANALYTICAL PRECAUTIONS

1. For *in vitro* diagnostic use only.
2. For Professional use only.
3. All specimens should be regarded as potentially infectious.
4. Gloves must be worn.
5. Optimal assay performance requires **STRICT ADHERENCE** to the assay procedure described in this Instruction Sheet. Deviations from the procedure may lead to aberrant results.
6. Do not open or remove test device from their individually sealed pouches until ready for use.
7. Do not use the test kit if the pouch is damaged or the seal is broken.
8. For best results allow all reagents and samples to reach room temperature before use.
9. To ensure proper drop delivery, droppers should be held vertically, while gently dispensing one drop at a time, in quick succesion.
10. Do not reuse test device.
11. Do not interchange reagents between kit lots.
12. Do not use kit components beyond the expiry date printed on the label(s).
13. Handle reagent(s) carefully to avoid spills on work surface. Clean up spills with absorbent paper and water.
14. Reagents supplied should not present a hazard to health if used in accordance to the instructions stated.

STORAGE AND STABILITY

1. Keep the kit at room temperature or refrigerated (2°C ~ 30°C) when not in use. The test device must remain in the sealed pouch until use.
2. The test strip is stable through the expiry date printed on the package label.
3. **DO NOT FREEZE**.
4. Do not store the test kit in direct sunlight.

MATERIALS REQUIRED BUT NOT PROVIDED

1. Timer
2. Tube stand

SPECIMEN COLLECTION AND PREPARATION

Specimens for the direct detection of viral antigen should be taken preferably during the first 3 days after onset of clinical symptoms.

1. Nasopharyngeal, throat or nasal swab should be tested immediately after collection. If not, the swab should be stored in the diluent tube provided with the swab stick broken off. Keep the tubes at 2°C~8°C during transportation. For long term storage, keep the tubes at or below –70 °C.
2. Nasopharyngeal aspirate or nasal wash should be tested immediately after collection. If it cannot be tested within 1 - 2 hours, keep it cool at 2°C~8°C. For long term storage, it should be kept frozen at or below -70°C.
3. For PBS containing allantoic fluid, these should be stored at 2°C~8°C for short term storage (48 - 72 hours). It should be kept frozen for long term storage.

NOTE: The choice for a nasal, nasopharyngeal or throat swab, will depend on whether the patient has discharge that can be collected for testing.

If the patient has a running nose, it is preferable to take a nasal swab.

If the patient has cough with sputum, then take a throat swab.

Nasopharyngeal swabs, nasal aspirate, nasopharyngeal wash,are usually performed by ENT (Ear, Nose and Throat) specialist.

All sample types should be at room temperature before testing.

ASSAY PROCEDURE

IMPORTANT: Strict adherence to the assay procedure will ensure optimal assay performance. Deviations from the procedure may lead to aberrant results.

Allow the kit to warm to room temperature before running the assay. Omit this step if device was stored at 18°C ~ 30°C.

A. Procedure for direct swab sample (nasopharyngeal, throat or nasal swab)

1. Add assay diluent into test tube until 1 ml. marked line.
2. Immerse swab into the test tube containing assay diluent.
3. Mix the swab until the sample has been dissolved into the diluent; remove as much liquid from the swab as possible.
4. Leave the test tube until the large particles have settled down to the bottom of the tube.
5. Remove the test device from the foil pouch, and place it on a flat and dry surface.
6. Using the disposable dropper provided, dispense **eight drops** of the supernatant from extracted sample into the sample well, avoiding adding bubbles.
7. Interpret test results at **10 minutes**.

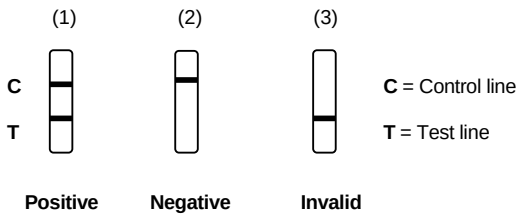
B. Procedure for nasopharyngeal aspirate, nasal wash or allantoic fluid

1. Dispense **three drops** of specimen into the test tube with the disposable dropper provided.
2. Add **six drops** of the assay diluent from the dropper bottle into the tube containing the specimen from above, mix well by pipetting up and down.
3. Dispense **eight drops** of the mixture into the sample well with the same dropper, avoid adding bubbles.
4. Interpret test results at **10 minutes**.

QUALITY CONTROL

1. Positive and negative controls are not included and are optional.
2. No line is visible before running the assay. If the control line at position "C" does not become visible after the assay, the test is considered invalid. Positive samples will have an additional colored line at position "T".

INTERPRETATION OF RESULTS



1. **Positive** for Influenza A antigen if colored bands appear at the Test line (T) and Control line (C) within the viewing window. Any intensity of line should be considered as a positive.
2. **Negative** for Influenza A antigen if only the Control line (C) is visible through the viewing window.
3. **Invalid** if the Control line (C) is absent. If this occurs, the assay should be repeated using a new device.

LIMITATIONS OF PROCEDURE

Optimal assay performance requires strict adherence to the assay procedure and instructions described. Deviation from the procedure(s) may lead to aberrant results. A NEGATIVE result does not exclude the possibility of exposure to or infection with Influenza A. Supplemental test such as PCR (H5, H7 and H1- H15) and other clinical information available may be useful in the discrimination of Influenza A positive samples.

LIMITED EXPRESSED WARRANTY DISCLAIMER

The manufacturer makes no expressed warranty other than that the test kit will function as a veterinary *in vitro* diagnostic assay within the specifications and limitations described in the Instruction Sheet when used in accordance with the instruction contained herein. The manufacturer disclaims any warranty expressed or implied, including such expressed or implied warranty with respect to merchantability, fitness for use or implied utility for any other purpose. The manufacturer's liability is limited to either replacement of the product or refund of the purchase price of the product. The manufacturer shall not be liable to the purchaser or third parties for any damage, injury or economic loss howsoever caused by the product in the use or in the application thereof.

PERFORMANCE

Specificity

Table 1: Results of a specificity study indicating that the Rockeby Human Influenza A Antigen test had no cross reactivity with other viruses found in the respiratory system, which includes parainfluenza virus, adenovirus and measles virus.[7]

Result by IFA and/or culture	*No. of test*	*RT-PCR (H1, H3, H5)*	***Rockeby Human Influenza A*** *Clinical Samples*	
			Negative	**Positive**
Parainfluenza 1	3	Neg	3	0
Parainfluenza 3	1	Neg	1	0
Adenovirus	2	Neg	2	0
* Mixed infection	3	Neg	3	0
Negative for all viruses	13	Neg	13	0
Overall	22	22	22	0

* Adenovirus/parainfluenza type 2, adenovirus, parainfluenza type 3, parainfluenza virus 1 / measles.

Sensitivity

Table 2: Results of a study showing that the Rockeby Human Influenza A Antigen test kit is able to detect H3N2 and H5N1 infections.[7]

Assay	***Clinical samples***		***Virus isolation***	
	#ID39 (H3N2)	*#ID1 (H5N1)*	*#ID39 (H3N2)*	*#ID1 (H5N1)*
IFA (infected cells)	Pos	Pos	Pos	Pos
Kit "A"	Pos	NA*	Pos	Pos
Kit "B"	NA*	NA*	Pos	Pos
Kit "C"	NA*	NA*	Pos	Pos
Rockeby Human Influenza A	Pos	NA*	Pos	Pos

** Not applicable*

Tested by WHO Collaborating Centre for Reference and Research on Influenza, Australia.[7]

BIBLIOGRAPHY

1. Bird Flu fact sheet, Department of Disease Control, Public Health Ministry Thailand
2. WHO Collaborating Centre for reference and research on Influenza. Reagents for Influenza Virus Diagnosis 2003, www.influenzacentre.org
3. OIE. 2000. Highly pathogenic avian influenza In: Manual of standards for diagnostic tests and vaccines, 4th ed.
4. Perkin LEL. and Swayne DE. 2003. Comparative Susceptibility of selected avian and mammalian species to a Hong Kong-origin H5N1 high-pathogenicity avian influenza virus. Avian Disease 47:956-967.
5. Swayne DE. Senne DA. and Beard CW. 1998. Influenza In: Isolation and identification of avian pathogens, 4th ed. AAAP, Pennsylvania, USA, 150-155.
6. Webster RG. 1997. Predictions for future human influenza pandemics. Journal of Infectious Disease 176 (supp. 1): 14-19.
7. Rockeby biomed confidential data on file.

TECHNICAL PROBLEMS / COMPLAINTS

Should there be a technical problem / complaint:

1. Note the kit(s) lot number(s) and the expiry date.
2. Retain the kit(s) and the test device(s).
3. Contact Rockeby biomed or your local distributor.

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The following are graphical symbols used in or found on Rockeby biomed products and packaging. These symbols are the most common ones appearing on diagnostic devices and their packaging. They are explained in more detail in the British and European Standard BS EN 980: 2003.

Use by
Synonym for this :
Expiry Date

In vitro diagnostic medical device

Batch Code
Synonyms for this are:
Lot Number
Batch Number

Catalogue Number

Temperature Limitation

Attention.
See Instruction for Use

Manufacturer

Do not reuse

Contains sufficient for <n> tests

Consult instructions for use