# ROTA-DIPSTICK

**For in *Vitro* diagnostic use only**

**Rapid test for the detection of Rotavirus in human stool specimen**

## I. INTRODUCTION AND INTENDED USE

Viral gastroenteritis is an infection caused by a variety of viruses that results in vomiting or diarrhea. Many different viruses can cause gastroenteritis, including rotaviruses, adenoviruses. Bacteria like Salmonella or Proteus like Cryptosporidium parvum can cause gastroenteritis.
The main symptoms of viral gastroenteritis are watery diarrhea and vomiting. The affected person may also have headache, fever, and abdominal cramps ("stomach ache"). In general, the symptoms begin 1 to 2 days following infection with a virus that causes gastroenteritis and may last for 5-8 days. Rotavirus is the more frequent cause of acute diarrhea in children under two years of age..
Rota-Dipstick is a screening immunochromatographic assay to detect Rotavirus antigen in stool samples.

## II. PRINCIPLE OF THE TEST

The Rota-dipstick is a qualitative lateral flow immunoassay for the detection of *Rotavirus* antigen in human stool samples. The membrane is pre-coated with monoclonal antibodies against Rotavirus antigens on the test line region. During testing, the sample reacts with the particle coated with anti-Rota antibodies which was pre-dried on the test strip. The mixture moves on the membrane by capillary action. In the case of a positive result the specific antibodies present on the membrane will react with the mixture conjugate and generate a red coloured line in "T" area. The mixture continues to move across the membrane to the immobilized antibody placed in the control band region, a green coloured band always appears. The presence of this green band serves as verification that sufficient volume was added, that proper flow was obtained and as an internal control for the reagents.

## III. REAGENTS AND MATERIALS

Each kit contains all materials needed for 25 tests:
**1.** Dipstick for the immunochromatographic reaction. The strips come in a bottle with a dessicant.
**2.** Bottle containing extraction buffer, with cap dropper (1 x 20 mL).
**3.** Instruction for use
**Required materials (not supplied)**
Specimen collection container - Disposable gloves – Timer.
**Auxiliary reagents (Not supplied with this kit)**
Positive and Negative control (Mascia Brunelli Ref. UD80020)

## IV. PRECAUTIONS

- For in vitro diagnosis only
- Read the instruction for use before performing the test
- Do not use after expiration date
- All the specimens should be considered potentially hazardous and handled in the same manner as an infectious agent. It is suggest to disinfect or autoclave at 121°C for 1 hour
- The tube containing the sensitized strips must be recapped as soon as the necessary number of strips for the operation has been removed, since the strips are sensitive to humidity. Make sure that the desiccant is present.
- The reagents' quality cannot be guaranteed beyond their shelf-life date or if the reagents are stored under inappropriate conditions.
- Do not use the test if pouch is damaged

## V. STORAGE

Store as packaged in the sealed pouch either at refrigerated or room temperature (2-30ºC). The test is stable through the expiration date printed on the sealed pouch. The test must remain in the sealed pouch until use. Do not freeze.

## VI. SAMPLES AND PREPARATION

Collect sufficient quantity of faeces (1-2 g or mL for liquid sample). Stool samples should be collected in clean and dry containers (no preservatives or transport media). The samples can be stored in the refrigerator (2-8ºC) for 1-2 days prior to testing. For longer storage the specimen must be kept frozen at –20ºC. In this case, the sample will be totally thawed, and brought to room temperature before testing. Specimens may be frozen and thawed twice.
**Note**
Stool in transport media, on swabs, or mixed with preservatives is not appropriate for testing. Mix stool as thoroughly as possible prior to pipetting.
**Liquid or Semi-Solid Stools**
Using a separate pipette (included with the kit) for each stool, draw stool of the sample itself. Dispense 6-7 drops of each stool into a separate extraction tube containing 0.7 mL (15 drops) of extraction buffer. Mix carefully.
Care should be taken when pipetting semi-solid stool. The addition of less than indicated of stool may cause a false-negative test. The addition of more than indicated of stool may cause invalid results due to restricted sample flow.
**Formed / Solid Stools**
Collect the stool sample with the tip by dipping in three different places of the same stool specimen.. Verify to transfer a small portion (approximately 6 mm diameter) of stool. Put the collection device back into the plastic test tube, containing 0.7 mL (15 drops) of extraction buffer. Shake the test tube in order to get an homogeneous solution. Wait at least 3 minutes. Repeat the operations just to obtain a dark yellow-brown solution, if necessary.
The transfer of too little stool, or failure to mix and suspend the stool in extraction tube completely may result in a false-negative test results. Care should be taken to transfer no less and no more than the amount indicated. The sample should be thoroughly mixed with a vortex before testing. The addition of excessive amount of stool may cause invalid results due to restricted sample flow.

## VII. PROCEDURES FOR SAMPLES

**Allow the reagents to reach to room temperature prior to testing.**
1. Immerse the sensitized strip in the direction indicated by the arrows.

**2.** Let react for 10 minutes. Results must be read on wet strip after 10 minutes incubation.

## VIII. INTERPRETING THE RESULTS

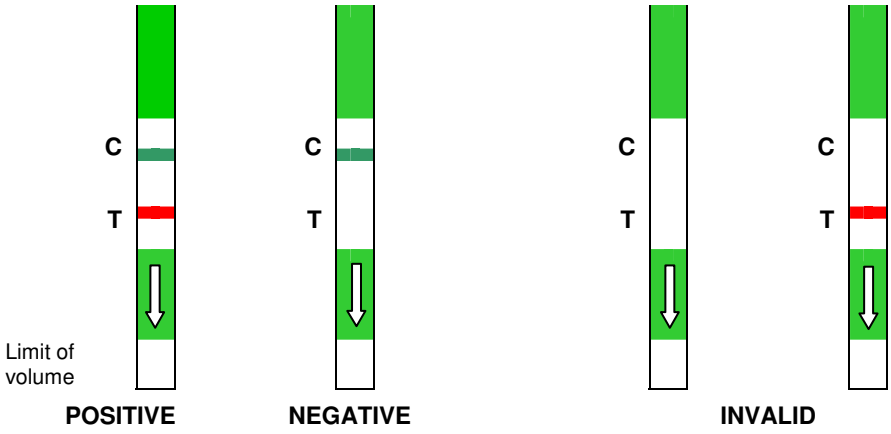
**Negative Test:** In the reading area only one GREEN band appears in the control region "C". This is the control line assuring the correctness of test performing.

**Positive Test:** In addition to the control GREEN band, a clearly distinguishable RED band appears in the test region "T". The intensity of the band colour in the test region is proportionally to the antigen concentration in the sample.

**Invalid**: No band appears in the control region. The test is to be considered as in conclusive and it is recommended to repeat the test.

Insufficient specimen volume, incorrect procedural techniques or deterioration of the reagents are likely the reasons for control line failure. Review the procedure and repeat the tests using a new test.

**Illustration 1**




## IX. PERFORMANCE

**SENSITIVITY and SPECIFICITY**

An evaluation was conducted comparing the results obtained using the Rota-Dipstick to a commercial available Rotavirus ELISA assay. Rota-Dipstick was highly specific (>98%) and also highly sensitive (>99%) compared with the results of that ELISA assay.

**CROSS-REACTIVITY**

It was performed an evaluation to determine the cross reactivity of Rota-Dipstick. There is not cross reactivity with common gastrointestinal pathogens, other organisms and substances occasionally present in faeces: Astrovirus, Adenovirus , Escherichia coli , Campylobacter, Enterovirus and Giardia lamblia.

## X. LIMITS OF THE KIT

- Rota-Dipstick will only indicate the presence of Rotavirus in the specimen (qualitative detection) and should be used for the detection of Rotavirus antigens in faeces specimens only. To confirm the diagnosis additional testing using other clinical methods is recommended
- A positive result does not preclude the possibility of infections by other pathogens.
- Some stool samples can decrease the intensity of the control line.
- This kit is intended for professional use in the laboratory and the test should be performed by qualified personnel with adequate training. Can not be used by the patient at home.

## XI. REFERENCES

1. New Immunochromatographic Method for Rapid Detection of Rotaviruses in Stool Samples Compared with Standard Enzyme Immunoassay and Latex Agglutination Techniques. I. Wilhelmi, J. Colomina, D. Martín-Rodrigo E, Roman, A. Sánchez-Fauquier. European Journal of Clinical Microbiology & Infectious Diseases, October 2001, p. 741-743
2. The Clatterbridge Hospital Study: Comparison of One-Step Assays to the DAKO ELISA. Department of Microbiology, Clatterbridge Hospital, Wirral, England.
3. Rapid Detection of Rotaviruses – Are we underestimating infection in African infants? J Dewar1, M de Beer1, E Elliott2 D Semenya and A Steele. MRC Diarrhoeal Pathogens Research Unit, Medunsa, Pretoria, South Africa. Ampath Laboratories, Johannesburg, South Africa. 2004-08-19

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| IVD | In Vitro Diagnostic Medical Device | | Temperature limitation | LOT | Batch code (EXXX) | | Manufacturer | | Keep dry | NON STERILE | Non-sterile |
| | Consult Instructions for use | | Use by (year/month) | REF | Catalogue number | | Do not reuse | | Fragile, handle with care | | Keep away from heat |

**CONTENT (25 tests)**
Dipstick for immunochromatographic reaction
Extraction buffer
Instruction for use

**REF. VC1001**
25 items
1 x 20 mL
1 item

**EDMA (EDMS) Code 1570909000** CE IVD