

VIASURE MULTIPLEX

Rhinovirus + Enterovirus Real Time PCR Detection Kit

Patógeno. Descripción

Human rhinoviruses (HRVs) and human enteroviruses (HEVs) are the most common cause of infections in people worldwide. They are members of the *Enterovirus* genus of the virus family *Picornaviridae*. They are small viruses (30nm diameter) with a single-stranded RNA genome of 7,000 to 7,500 nucleotides enclosed in an icosahedral capsid. HRVs include 153 currently known types divided into three species (A, B and C), while HEVs consist of 104 types belonging to four species (A, B, C and D). Traditionally, human enteroviruses are categorized into polioviruses and nonpolio enteroviruses (coxsackieviruses, echoviruses, and numbered enteroviruses).

HRVs are the usual cause of common cold but are also frequently found in otitis media, sinusitis, bronchitis, pneumonia, and asthma exacerbations. Therefore, due to they are restricted to the respiratory tract the mode of transmission are mostly the via aerosols of respiratory droplets and from fomites (contaminated surfaces), including direct person-to-person contact. Currently there is no specific antiviral treatment for rhinovirus infection.

In contrast to HRVs, replication of HEVs is not restricted to the respiratory tract but also can take place in the small intestine and spread to various target organs. They are readily transmitted from person to person through an air and/or via a fecal-oral route, or even through contaminated objects. Most HEV infections are asymptomatic or manifest common cold-like symptoms. However, HEV infections can be more severe, causing poliomyelitis, meningitis, encephalitis, myocarditis, exanthema, acute hemorrhagic conjunctivitis, and severe generalized infections in newborns. Therefore, sample collection for

HEVs diagnosis should be performed according to clinical manifestations. Cerebrospinal fluid (CSF), blood, respiratory samples and stool samples are commonly used.

Differential diagnosis of HRV and HEV infections is epidemiologically important. Specific identification of these viruses already has implications for the supportive management of patients and will become more significant when specific antiviral drugs become available. Nucleic acid amplification techniques have replaced the isolation of viruses in cell cultures as the method of choice for the detection of picornaviruses, partly due to the outstanding sensitivity, specificity, and rapidity of such techniques. The recently identified species C HRVs cannot be cultivated in standard cell lines but can be amplified by reverse transcription (RT)-PCR. Both HRVs and HEVs have conserved 5' noncoding regions (NCRs) and a few nearly identical sequence motifs, allowing the design of universal primers for their amplification in RT-qPCR. RT-qPCR has been shown to be far more sensitive than cell culture for detection of these viruses.

VIASURE *Rhinovirus + Enterovirus* Real Time PCR Detection Kit is designed for the diagnosis of Rhinovirus and/or Enterovirus in respiratory samples. The detection is done in one step real time RT format where the reverse transcription and the subsequent amplification of specific target sequence occur in the same reaction well. The isolated RNA target is transcribed generating complementary DNA by reverse transcriptase which is followed by the amplification of a conserved sequence of 5'UTR region using specific primers and a fluorescent-labelled probe.



Analytical sensitivity

VIASURE Rhinovirus + Enterovirus Real Time PCR Detection Kit has a detection limit of ≥ 10 RNA copies per reaction for Rhinovirus and Enterovirus (figures 1 and 2).

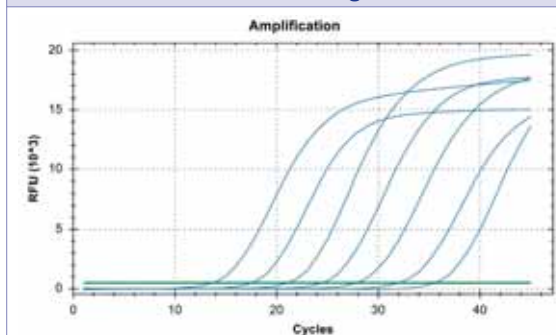


Figure 1. Dilution series of Rhinovirus (10^2 – 10^1 copies/rxn) template run on the Bio-Rad CFX96 Touch™ Real-Time PCR Detection System (FAM channel).

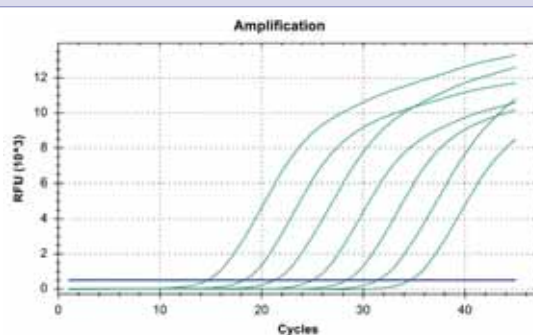


Figure 2. Dilution series of Enterovirus (10^2 – 10^1 copies/rxn) template run on the Bio-Rad CFX96 Touch™ Real-Time PCR Detection System (HEX channel).

Components

Reagent/Material	Description	Colour	Quantity
Rhinovirus + Enterovirus 8-well strips	A mix of enzymes, primers-probes, buffer, dNTPs, stabilizers and Internal control in stabilized format	White	6/12 X 8-well strip
Rehydration Buffer	Solution to reconstitute the stabilized product	Blue	1 vial x 1,8 mL
Rhinovirus + Enterovirus Positive Control	Non-infectious synthetic lyophilized cDNA	Red	1 vial
Negative Control	Non template control	Violet	1 vial x 1 mL
Water RNase/DNase free	Water RNase/DNase free	White	1 vial x 1 mL
Tear-off 8-cap strips	Optical caps for sealing Wells during thermal cycling	Transparent	6/12 x 8-cap strip

Kit References

Reference	Description
VS-RHE106L	Viasure Rhinovirus + Enterovirus Real Time PCR Detection Kit 6 x 8-well strips, low profile
VS-RHE106H	Viasure Rhinovirus + Enterovirus Real Time PCR Detection Kit 6 x 8-well strips, high profile
VS-RHE112L	Viasure Rhinovirus + Enterovirus Real Time PCR Detection Kit 12 x 8-well strips, low profile
VS-RHE112H	Viasure Rhinovirus + Enterovirus Real Time PCR Detection Kit 12 x 8-well strips, high profile
VS-RHE113L	Viasure Rhinovirus + Enterovirus Real Time PCR Detection Kit 96-well plate, low profile
VS-RHE113H	Viasure Rhinovirus + Enterovirus Real Time PCR Detection Kit 96-well plate, high profile

Work Flow

One-step rehydration of wells and add your extracted DNA



STEP 1

Add 15 μ l of rehydration buffer into each well



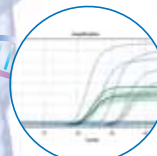
STEP 2

Add 5 μ l of DNA sample / positive control / negative control



STEP 3

Load the strips into the thermocycler and run the specified protocol



STEP 4

Interpretate results