PCR*opsis*™ Reagent RVD-E

(Not for Resale)

INTENDED USE (in vitro diagnostic use)

PCR*opsis*™ Reagent RVD-E is intended for extraction-free amplification of RNA and DNA from specimens on swabs, without the need for transport mediums.

PRINCIPLES OF THE PROCEDURE

PCR*opsis*™ Reagent RVD-E is engineered to simultaneously elute material from swabs, bind a variety of reverse transcriptase quantitative polymerase chain reaction (RT-qPCR) / PCR inhibitors found in clinical samples, lyse specimens and stabilize nucleic acids in a manner that's compatible with RT-qPCR / PCR. The product consists of a proprietary mixture of peptides, salts, stabilizers, buffers and sodium azide to achieve this task. Reagent RVD-E allows for extraction-free amplification of nucleic acids without performing extractions, centrifugations or other sample manipulations, which may introduce errors, contaminants and/or skew the representation of RNA fragments.

WARNINGS & PRECAUTIONS

For in vitro Diagnostic Use.

- Observe approved biohazard precautions and aseptic techniques to prevent contamination of the product. To be used only by adequately trained and qualified personnel.
- Pathogenic microorganisms, including hepatitis viruses and Human Immunodeficiency Virus, may be present in clinical specimens. "Standard Precautions"[1-4] and institutional guidelines should be followed in handling all potentially bio-hazardous materials.
- Sterilize all biohazard waste including specimens, containers and mediums after their use.
- Directions should be read and followed carefully.
- Do not re-pack.
- The use of this product in association with a rapid diagnostic kit, diagnostic instrumentation or used in a manner not intended should be validated by the user.
- Do not ingest the reagent.
- Avoid skin contact with reagent since it contains sodium azide to prevent microbial growth.

Storage: This product is ready for use and no further preparation is necessary. The product should be transported and stored in its original container at 4–25˚C until used. Do not overheat. Do not incubate or freeze prior to use. Improper storage will result in a loss of efficacy. Do not use after expiration date, which is clearly printed on the label.

Product Deterioration: PCR*opsis*™ Reagent RVD-E should not be used if (1) there is evidence of damage or contamination to the product, (2) there is evidence of leakage, (3) the color of the reagent has changed from clear-white hazy, (4) the expiration date has passed, or (5) there are other signs of deterioration.

PROCEDURES
Materials Provided: PCR*opsis*™ Reagent RVD-E

Materials Required But Not Provided: Heating device (heating block or thermal cycler), flocked nylon swab, transport tubes, thin walled tube (0.2 ~ 0.6 mL) or 96-well PCR plate, plate sealer, pipette tips

Test Procedure: Collect test specimen using a flocked nylon swab or similar. Proper specimen collection, transport and storage is critical for successful nucleic acid amplification. For specific guidance regarding specimen collection procedures, consult published reference manuals.[5-11] Clinical specimens should be collected as soon as possible after the clinical onset of disease. Highest viral titers are present during the acute illness.

Recommended swabs: synthetic swabs (nylon, rayon, dacron, polyester) with aluminum or plastic shafts

Material to be tested: specimen-containing swab properly transported in a sterile tube

1. Thoroughly mix Reagent RVD-E to ensure homogeneity, but avoid creating bubbles unnecessarily
    1. Reagent RVD-E has a hazy, white color when homogenized and normal settlement occurs if not regularly mixed
2. Elute material from swab:
    1. Add 200 µL of Reagent RVD-E to transport tube with swab
    2. Make sure the swab is at least partially submerged into Reagent RVD-E
    3. Vortex for ~40 seconds to elute sample
3. Specimen lysis & nucleic acid stabilization:
    1. Transfer ~100 µL of eluted sample into a thin-walled PCR tube / plate and then cap tube or apply plate sealer to plate to prevent evaporation
    2. Heat at 95˚C
        1. Mammalian: 5 minutes
        2. Viruses: 10 minutes
        3. Bacteria: 15 minutes
        4. NOTE: heating for a longer period of time does not negatively affect results
    3. Let cool at room temperature for ~10 seconds before continuing
4. Pipette up & down to ensure complete mixing
5. Use 5 - 10 µL of lysed / stabilized sample in your desired RT-qPCR / qPCR procedure
    1. Lysed sample can represent 25% ~ 50% of your final RT-qPCR mixture (i.e., 5 ~ 10 µL sample into a total volume of 20 µL)

Quality Control: All lots of PCR*opsis*™ Reagent RVD-E are tested for microbial contamination and the ability to amplify viral RNA without extraction. If aberrant quality control results are noted, patient results should not be reported.

RESULTS

Results obtained will partially depend on proper and adequate specimen collection, transport and processing in the laboratory. PCR*opsis*™ Reagent RVD-E may result in unreliable results when used beyond the intended use.

LIMITATIONS OF THE PROCEDURE

- Performance characteristics of PCR*opsis*™ Reagent RVD-E were validated using SARS-CoV-2, S. *aureus* and P. *aeruginosa* dried on flocked nylon swabs. The use of alternative microorganisms, swabs, gene targets and / or detection methods may affect the performance of the product.
- Improper transport and storage of test swabs may reduce the detection of desired gene targets.
- Follow recommended guidelines for specimen collection, transport and storage as this may affect the ability to amplify gene targets.

PERFORMANCE CHARACTERISTICS

The performance of PCR*opsis*™ Reagent RVD-E was compared to traditional RNA extraction methods (e.g., Qiagen's QIAamp Viral RNA Kit) from the same samples. These studies used SARS-CoV-2 absorbed and dried onto flocked nylon swabs, spiked samples processed using both methods and RT-qPCR was performed using IDT qPCR probe assay and Promega GoTaq® Probe 1-Step RT-qPCR System. Observed Ct values between both methods are within 5 Ct of each other.

AVAILABILITY – NOT FOR RESALE

Cat. #	Description
7833025	PCR*opsis*™ Reagent RVD-E, 25 mL
7833100	PCR*opsis*™ Reagent RVD-E, 100 mL
7833500	PCR*opsis*™ Reagent RVD-E, 500 mL

MANUFACTURER

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REFERENCES

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Glossary of Symbols Used