# Package Insert



# ViewRNA® Temperature Validation Kit

Cat. No. QV0523

#### **Product Overview**

The ViewRNA Temperature Validation Kit uses a National Institute of Standards and Technology (NIST) calibrated digital thermometer with Type-K beaded probe to assess temperature accuracy. This kit may be used for any ViewRNA Assays to evaluate the temperature of incubators or ThermoBrite systems. For best assay results, we recommend calibrating all incubators including the ThermoBrite with the ViewRNA Temperature Validation Kit before running the assay. It is crucial that all incubators are validated at the assay-specified hybridization temperature.

#### **Intended Use**

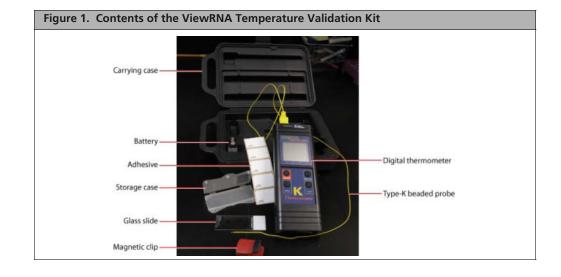
The ViewRNA Temperature Validation Kit is designed to precisely measure the temperature in any incubator used in the ViewRNA Assays. Refer to appropriate ViewRNA Assays user manual for recommended incubators.

#### **Contents and Storage**

The ViewRNA Temperature Validation Kit contains the following components. The kit has a shelf life of 1 year from date of receipt when stored as recommended.

Table 1. ViewRNA Temperature Validation Kit Contents and Storage

| Component           | Description   | Quantity | Storage  |
|---------------------|---|----------|----------|
| Digital Thermometer | NIST calibrated digital thermometer with Type-K beaded probe                    | 1        | 15-30 °C |
| Glass Slide         | 75 mm x 25 mm glass slide   | 1        | 15-30 °C |
| Storage Case        | Storage case for the glass slide  | 1        | 15-30 °C |
| Adhesive            | Secures Type-K beaded probe to the glass slide                                  | 5        | 15-30 °C |
| Magnetic Clip       | Secures Type-K beaded probe while measuring the air temperature (dry incubator) | 1        | 15-30 °C |
| Carrying Case       | Carrying case for the ViewRNA Temperature Validation Kit                        | 1        | 15-30 °C |
| Battery             | 9V battery for the digital thermometer  | 1        | 15-30 °C |



#### **Required Materials Not Provided**

The following materials are required if you are validating the temperature of a ThermoBrite incubator system or humidifying incubator.

Table 2. Required Material Not Provided

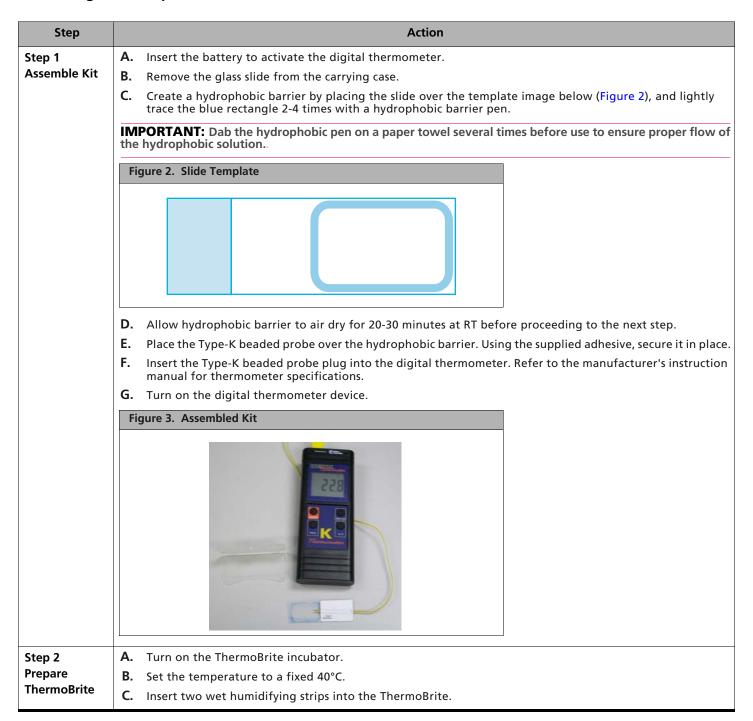
| Item  | Supplier  | Required for validating a                      |
|---|---|--|
| Double-distilled water (ddH <sub>2</sub> O) | Major Laboratory Supplier                               | ThermoBrite Incubator<br>Humidifying Incubator |
| Hydrophobic Barrier Pen                     | Vector Laboratories P/M H4000 or,<br>Affymetrix QVC0500 | ThermoBrite Incubator<br>Humidifying Incubator |
| Aluminum Slide Rack                         | VWR P/N 100493380                                       | Humidifying Incubator                          |

#### Overview

Here procedures are provided for the temperature validation of the following incubators:

- ThermoBrite incubator
- Humidifying incubator (tissue culture incubator without CO2)
- Dry incubator

#### Validating the Temperature of a ThermoBrite Incubator



**Temperatures** 

the +1°C deviation.

ensure accuracy.

### Action Step Place the assembled glass slide onto the ThermoBrite with the hydrophobic barrier facing up. Make sure Step 3 the Type-K beaded probe is securely attached to the slide. **Set Up Glass** Slide IMPORTANT: The glass slide must remain in contact with the hot plate at all times to ensure an accurate reading (see Figure 4). Figure 4. Complete Setup Add 400 µL of ddH<sub>2</sub>O onto the slide. Make sure the ddH<sub>2</sub>O stays within the hydrophobic barrier and is in contact with the Type-K beaded probe (see Figure 5). Figure 5. Glass Slide on the Hot Plate with ddH<sub>2</sub>O Added Affymetrix Close the lid making sure there is sufficient slack in the wiring to prevent the slide from lifting off the hot Step 4 plate surface. **Equilibrate the ThermoBrite** Wait 10 minutes for the temperature to equilibrate. Allow an additional 20 minutes equilibration time before recording the temperature. Repeat Step 3 - Step 4 to measure the temperature at four different regions of the hot plate. Step 5 Measure and If necessary, adjust the temperature setting so that the thermometer displays the assay-specified **Adjust** temperature. For example, if the digital thermometer indicates the temperature is 39°C when the ThermoBrite is set at 40°C, change the setting temperature of the ThermoBrite to 41°C to accurately reflect

IMPORTANT: The fixed temperature of the ThermoBrite and the actual temperature measured by the digital thermometer may differ by  $\pm 2^{\circ}$ C. We recommend calibrating the ThermoBrite at least once a month to

#### Validating the Temperature of a Humidifying Incubator

## Step **Action** Insert the battery to activate the digital thermometer. Step 1 **Assemble Kit** Remove the glass slide from the carrying case. Create a hydrophobic barrier by placing the slide over the template image below (Figure 6), and lightly trace the blue rectangle 2-4 times with a hydrophobic barrier pen. **IMPORTANT:** Dab the hydrophobic pen on a paper towel several times before use to ensure proper flow of the hydrophobic solution. Figure 6. Slide Template D. Allow hydrophobic barrier to air dry for 20-30 minutes at RT before proceeding to the next step. Place the Type-K beaded probe over the hydrophobic barrier. Using the supplied adhesive, secure it in place. Insert the Type-K beaded probe plug into the digital thermometer. Refer to the manufacturer's instruction manual for thermometer specifications. Turn on the digital thermometer device. Figure 7. Assembled Kit Step 2 Turn on the humidifying incubator. **Prepare** To maintain the humidity level at more than 65% (without CO<sub>2</sub>) for the hybridization condition, fill bottom Humidifying tray nearly full with ddH<sub>2</sub>O. Incubator C. Set the temperature to 40°C. D. Allow the incubator to equilibrate for 30 minutes before starting the calibration.

#### Step 3 Set Up Glass Slide

Step

#### Action

**A.** Place the assembled glass slide onto an aluminum rack with the hydrophobic barrier facing up. Make sure the Type-K beaded probe is securely attached to the slide.

**IMPORTANT:** The glass slide must remain flat on the aluminum rack at all times to ensure an accurate reading.

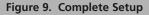
**B.** Add 400 µL of ddH<sub>2</sub>O onto the slide. Make sure the ddH<sub>2</sub>O stays within the hydrophobic barrier and is in contact with the Type-K beaded probe (see Figure 8).

Figure 8. Glass Slide on the Aluminum Rack with ddH<sub>2</sub>O Added



# Step 4 Equilibrate Humidifying Incubator

- A. Place the assembled glass slide, on the aluminum rack, into the humidifying incubator.
- **B.** Close the door making sure there is sufficient slack in the wiring to prevent the slide from lifting off the aluminum rack.
- **C.** Wait 10 minutes for the temperature to equilibrate.
- **D.** Allow an additional 20 minutes equilibration time before recording the temperature (see Figure 9).





#### Step 5 Measure and Adjust Temperatures

- A. Repeat Step 3 -Step 4 to measure the temperature at four different regions of the humidifying incubator.
- **B.** If necessary, adjust the temperature setting so that the thermometer displays the assay-specified temperature. For example, if the digital thermometer indicates the temperature is 39°C when the humidifying incubator is set at 40°C, change the setting temperature of the incubator to 41°C to accurately reflect the +1°C deviation.

**IMPORTANT:** The fixed temperature of the humidifying incubator and the actual temperature measured by the digital thermometer may differ by  $\pm$  2°C. We recommend calibrating the humidifying incubator at least once a month to ensure accuracy.

## Validating the Temperature of a Dry Incubator

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| Step                     | Action   |  |
|--------------------------|--|--|
| Step 1<br>Assemble Kit   | A. Insert the battery to activate the digital thermometer.   |  |
|                          | Slide the Type-K beaded probe through the magnetic clip as shown in Figure 10. Make sure the Type-K beaded probe is pointing upward so that it is not in contact with any metal surface when put inside the incubator. |  |
|                          | C. Insert the Type-K beaded probe into the digital thermometer. Please refer to the manufacturer's instruction manual for thermometer specifications.  |  |
|                          | D. Turn on the digital thermometer device.   |  |
|                          | Figure 10. Assembled Kit   |  |
|                          | Digital thermometer Type-K beaded probe  Magnetic clip   |  |
| Step 2                   | A. Turn on the dry incubator.  |  |
| Prepare Dry<br>Incubator | <ul><li>B. Set the temperature to 40°C.</li><li>C. Allow the dry incubator to equilibrate.</li></ul>   |  |

# Step Step 3 Measure and Adjust Dry Incubator Temperature

#### Action

- A. Place the assembled Type K beaded probe inside the incubator (see Figure 11).
- **B.** Close the door making sure there is sufficient slack in the wiring.

#### Figure 11 Complete Setup for Dry Incubator



- **C.** Wait 10 minutes for temperature to equilibrate.
- **D.** Record the temperature.
- E. If necessary, adjust the dry incubator temperature settings so that the digital thermometer reads 40°C.

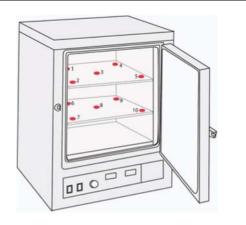
**IMPORTANT:** The fixed temperature of the dry incubator and the actual temperature measured by the digital thermometer might differ by  $\pm$  2°C. We recommend calibrating the dry incubator at least once every 6 months to ensure accuracy.

# Step 4 Assess Dry Incubator Temperature Uniformity

A. Repeat Step 2 - Step 3 to measure the temperature at multiple regions in the incubator to determine temperature uniformity (see Figure 12 for an example).

**IMPORTANT:** Make sure the temperature equilibrates before recording the measurements.

Figure 12. Dry Incubator Uniformity Test Positions



**IMPORTANT:** The temperature for all positions should be  $40 \pm 1^{\circ}$ C.

#### Step 5 Assess Ramp-Up Time

- **A.** Open the incubator door for 1 minute then close the door and measure the time needed for the temperature to return to 40°C.
- **B.** Repeat the previous step two more times.

**IMPORTANT:** Do not use the incubator for the assay if it takes more than 5 minutes to return to 40°C or if it overshoots by more than 2°C during recovery.

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