# **Product Information**

### **ΔTm** - Protein Cryoprotection Screening Kit

Item No. 600950

### **About This Assay**

Cayman Chemical has partnered with ΔTm Technologies LLC to provide a rapid, sensitive, and economical method for assessing both protein stability and protein-ligand interactions using a Thermal Stability Assay (TSA). These proprietary formulations ensure accurate and reproducible data while minimizing the deleterious effects of increasing temperature and its impact on the experimental solutions. This technology can be used to optimize experimental conditions before proceeding to more extensive studies such as ITC, SPR, or crystallography.

By using the Cryoprotection Screening Kit, TSA will generate novel protein freezing conditions by determining which conditions give the largest change in T<sub>m</sub> values. This positive change occurs as the protein, in its native state, is unfolded and intercalated with dye to give a new melting temperature value (see Figure 1). ΔTm Technologies has narrowed the range of conditions to 96 unique conditions that help with cryoprotection formulation, eliminating any guesswork, and providing you with answers.

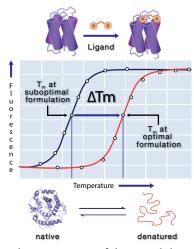


Figure 1. Graphical representation of the typical thermal stability assay.

### Kit Protocol

### Materials Needed But Not Supplied

- 1. Minimally 500 μl of ~0.5 mg/ml (~250 μg) protein
- A real-time PCR (RT-PCR) instrument
- 96-well format plate with adhesive plate seal that works for your RT-PCR

### **Kit Contents**

| Item Number | Item  | Amount Provided    | Storage |
|-------------|---|--------------------|---------|
| 600951      | ΔTm - Cryoprotection 96-Well Plate with lid | 25 μl in each well | -20°C   |
| 600942      | Protein Binding Dye                         | 25 μΙ              | -20°C   |

WARNING: This product is for laboratory research only: not for administration to humans. Not for human or veterinary DIAGNOSTIC OR THERAPEUTIC USE.

This material should be considered hazardous until information to the contrary becomes available. Do not ingest, swallow, or inhale. Do not get in eyes, on skin, or on clothing. Wash thoroughly after handling. This information contains some, but not all, of the information required for the safe and proper use of this material. Before use, the user must review the complete Safety Data Sheet, which has been sent via email to your institution.

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at the time of delivery.

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## **Product Information**

# Cayman

### General Information

- Incubate the kit at 37°C for 30 minutes prior to use.
- Pre-chill the RT-PCR plate at 4°C for 15-30 minutes prior to addition of reagents to the wells.
- The dye vial and kit plates should be stored at -20°C and away from light until the time of use.
- To guarantee accurate results, all solutions must be pipetted accurately.

### Assay Set Up

- 1. Add 5 µl of dye to 500 µl of protein sample. NOTE: The dye to protein ratio can be modified for your protein. Smaller proteins or proteins with a small hydrophobic core may require increased dye concentrations.
- 2. Transfer 5 µl of each solution from the Cryoprotection Assay Plate to a corresponding well of the RT-PCR plate.
  - Care must be taken to make sure the Thermal Stability Solutions are placed in the correct well (i.e., A1 formulation well gets put into the A1 well of your RT-PCR plate, etc.). NOTE: Well H12 is reserved for your protein in its current state. Please make sure to add 5 µl of the protein/dye mixture and 5 µl of your original protein buffer to well H12.
- 3. Add  $5 \mu l$  of the protein/dye mixture to each well of the plate.

| Component                         | Volume |
|-----------------------------------|--------|
| Protein Sample + Dye (>0.5 mg/ml) | 5 μΙ   |
| Kit Solution                      | 5 μΙ   |
| Total Well Volume                 | 10 μΙ  |

- 4. Mix the solutions by pipetting up and down (5 times).
  - Gently placing all solutions at the bottom of the well will eliminate the need to centrifuge the plate.
  - b. Make sure not to introduce air bubbles into the well.
- 5. Seal the RT-PCR plate with a seal that is compatible with your plate.

### Running the Thermal Stability Assay

Immediately load the plate into the instrument and begin the analysis. TSA heating schedules vary depending on your instrument. A typical TSA run will involve the following:

- 1. Heat the RT-PCR plate to 20°C
- 2. Ramp the temperature to 90°C in 0.5°C increments
- 3. Ramp the temperature to 4°C with a two minute schedule

NOTE: Total time for a TSA is 62 minutes using these parameters.

### Analyzing the Results

Once the run is finished, upload your data file (accepted format: .csv) to  $\underline{www.caymanchem.com/deltatm}$  and follow the instructions on the page. Once you have entered your data, the conditions with the largest change in  $T_m$  will be displayed on the screen as well as emailed to you at the address you entered.

### **Related Products**

For a list of related products please visit: www.caymanchem.com/catalog/600950

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