

## J.T.Baker® Endonuclease Shipping Stability

### OBJECTIVE

J.T.Baker® Endonuclease is a bioengineered enzyme that effectively degrades both single stranded and double stranded DNA and RNA in various applications. The recommended storage condition for J.T.Baker® Endonuclease is -20°C to prevent loss of activity and maintain a 24-month expiry, however, J.T.Baker® Endonuclease enzyme demonstrated stability at 25 °C up to 24-weeks. Refer to Avantor Lab Report JTB.00005.00, *Stability of J.T. Baker® Endonuclease*.

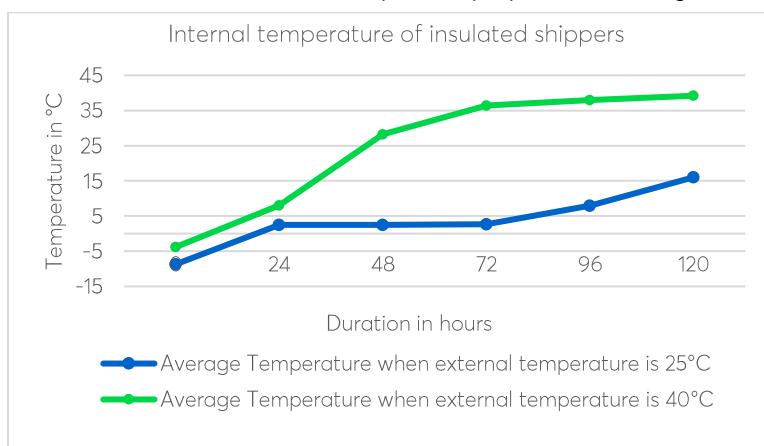
J.T.Baker® Endonuclease Biotech Reagent part numbers BAKRGT20-50, BAKRGT20-62, and BAKRGT20-80 are shipped in Credo Cube™ Series 20 M rated temperature-controlled shippers, monitored using cold chain logistics. This specialized packaging ensures the reagents maintain stable temperature conditions ranging from -15°C to -25°C and are thus out of scope for this study. J.T.Baker® Endonuclease Ultrapure Bioreagent and J.T.Baker® Endonuclease Biotech Reagent less than 5,000 KU are shipped in VWR insulated Sonoco ThermoSafe®2 shippers using icepacks. To evaluate the stability of enzyme shipped under these conditions, a shipping study was conducted evaluating shipping temperatures up to 40°C.

### METHOD

Five VWR insulated Sonoco ThermoSafe® shippers were prepared for the study per packaging instructions described in Packaging Qualification Shipping Study, PKG-QUAL-0018. Duplicate samples of J.T.Baker® Endonuclease were packaged in secondary packaging and placed in each shipper containing icepacks and a Sensitech TempTale®3 ultra-temperature data logger. Once sealed, a TempTale® data logger was placed on the outside of each shipper to monitor the external temperature conditions. Three shippers were placed at 25°C and the remaining shippers were placed at 40°C. After five days, the duplicate samples were removed from their respective storage conditions and stability assessed by evaluating the enzyme activity. The activity of endonuclease was determined using a validated analytical method based on sonicated salmon sperm DNA as a substrate where one unit of endonuclease is defined as the amount of enzyme that will produce acid-soluble oligonucleotides equivalent to  $\Delta A_{260}$  of 1.0 in 30 minutes 37°C.

### RESULTS

The internal temperature results for the duration of the study are displayed below in Figure 1.



**Figure 1:** Sonoco ThermoSafe® internal temperature results obtained from TempTale® data loggers

The results of each duplicate sample analysis for enzyme activity at each temperature condition are displayed below in Table 1. Overall, there is no significant change in activity observed over the duration of the study. The activity remained within 12% of the control sample and met a specification of  $\geq 250$  Units/ $\mu$ L after 120 hours for both temperature conditions evaluated.

| Sample                   | Shipping temperature | Activity U/ $\mu$ L | Average activity U/ $\mu$ L | Result $\geq 250$ Units / $\mu$ L |
|--------------------------|----------------------|---------------------|-----------------------------|-----------------------------------|
| Container 1, replicate 1 | 25°C                 | 421.81              | 423.83                      | Pass                              |
| Container 1, replicate 2 | 25°C                 | 425.85              |                             |                                   |
| Container 2, replicate 1 | 25°C                 | 469.95              | 460.08                      | Pass                              |
| Container 2, replicate 2 | 25°C                 | 450.21              |                             |                                   |
| Container 3, replicate 1 | 25°C                 | 482.30              | 476.95                      | Pass                              |
| Container 3, replicate 2 | 25°C                 | 471.59              |                             |                                   |
| Container 4, replicate 1 | 40°C                 | 465.02              | 446.24                      | Pass                              |
| Container 4, replicate 2 | 40°C                 | 427.46              |                             |                                   |
| Container 5, replicate 1 | 40°C                 | 452.55              | 445.40                      | Pass                              |
| Container 5, replicate 2 | 40°C                 | 438.24              |                             |                                   |
| Control, replicate 1     | -20°C                | 467.43              | 479.88                      | Pass                              |
| Control, replicate 2     | -20°C                | 492.33              |                             |                                   |

**Table 1:** Enzyme activity of J.T.Baker® Endonuclease stored in insulated Sonoco ThermoSafe® shipping containers and placed in elevated external temperatures to evaluate shipping conditions. Activity was measured at the end of Day 5.

## CONCLUSION

The study was conducted by monitoring the internal temperature of insulated Sonoco ThermoSafe® shipping containers that held duplicate samples of J.T.Baker® Endonuclease. These shippers were exposed to both standard (ambient) and elevated temperatures to simulate different transportation scenarios. After 120 hours, equivalent to five days, enzyme activity was evaluated to assess product stability. Throughout the study, the highest temperature observed was 39.2°C at the 120-hour timepoint under elevated shipping conditions. Findings demonstrate that the enzyme's activity for all samples stayed within 12% of the control sample and met a specification of  $\geq 250$  Units/ $\mu$ L after 120 hours for both temperature conditions evaluated. The study confirms that J.T.Baker® Endonuclease remains stable during shipment when transported in an insulated Sonoco ThermoSafe® shipping container and received within a timeframe of a standard shipping cycle of approximately two days. Longer durations of up to five days may be possible based on the results of this study, however, this information is intended to provide evidence the product is stable for longer periods at elevated temperatures and not a guarantee of product performance. Further testing is recommended if shipping duration exceeds two days. Activity was not assessed when material at elevated temperatures was cycled back to -20C and thawed for use.

## PRODUCT INFORMATION

J.T.Baker® Endonuclease is formulated using 50% Glycerin to prevent freezing, storage at -80°C is not advised. In AAV vector bioprocessing, J.T. Baker® Endonuclease has been shown to retain enzymatic activity in the presence of J.T. Baker® Cell Lysis Solution, part number BAKRG193, enabling robust and efficient cell lysis and DNA/RNA degradation in a single step.



| Part Number | Unit of measure | Quality                    |
|-------------|-----------------|----------------------------|
| BAKR7108-10 | 100 KU          | Ultrapure<br>Bioreagent    |
| BAKR7108-40 | 1,250 KU        |                            |
| BAKR7108-50 | 5,000 KU        |                            |
| BAKRGT20-10 | 100 KU          | Biotech<br>Reagent<br>cGMP |
| BAKRGT20-25 | 500 KU          |                            |
| BAKRGT20-40 | 1,250 KU        |                            |
| BAKRGT20-50 | 5,000 KU        |                            |
| BAKRGT20-62 | 12,500 KU       |                            |
| BAKRGT20-80 | 25,000 KU       |                            |

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