

## Temperature Gradient Analysis for Determining Melting Point of Proteins Using the NanoPlus

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The NanoPlus includes a temperature gradient feature for size and zeta potential. This unique feature allows the researcher to further utilize the built-in Peltier thermo-electric controller for temperature studies. The sample cell in the NanoPlus can be held at a specific temperature between 5°C to 90°C or a gradient of increasing or decreasing temperature in 0.1°C increments or greater. This is ideal for determining the melting point, or denaturing point, of proteins using the NanoPlus.

To set up a temperature gradient for protein analysis:

1. Set the “Manual Temperatures Setting” under “Measurement Parameters” to “Gradient” in the Size SOP Designer.
2. Complete the “Gradient Temperature (°C)” table with the start temperature, end temperature, interval, and equilibration

time in seconds. This setting tells the NanoPlus to collect size data at each temperature point and save each point as a separate data file in the “Size Analysis” database.

Once the analysis is complete, the software also automatically generates a “Temperature Gradient Analysis” file that can be opened under “Size Analysis.” The file will have the ending .tgr for temperature gradient and, when opened, will display Size (nm) versus Temperature (°C). The Cumulants value will be used for the size points.

A sample of bovine albumin (BSA) is tested using the temperature gradient function. The test was repeated and in both cases, the melting point was found to be at 68.0°C.

