The formulation of lyophilized protein-excipient mixtures strives to develop a product with good shelf life and that allows efficient dissolution by the end user with minimal change to the properties of the drug. One of the key properties used in the design of the formulations is the glass transition temperature, Tg, which must be sufficiently above the storage temperature to limit any degradation properties from occurring at significant rates. Several techniques can be used to measure this change in instruments like the PerkinElmer double furnace DSC, shown in Figure 1.

The most common approach to measuring the Tg of lyophilized formulations is the traditional heat-cool-heat run in the DSC. The initial heat measures the formulations Tg as it is received and the transition is often very noisy. Subsequent cooling in a controlled manner allows the material to be measured on the second heat in a more relaxed state, giving a cleaner and less noisy Tg. This

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Figure 1. PerkinElmer’s top of the line power compensated DSC, which is capable of both HyperDSC and StepScan, is shown in the autosampler configuration.
Finally, the use of Thermomechanical Analyzers (TMA) is well known as a method to measure transitions. TMA is less commonly used on powders but has been successfully used to measure the Tg of lyophilized formulations, excipients, and polymers used in drug delivery (such as hydrogels). Scanning at rates of 5-10 °C/minute, the TMA shown in Figure 5 gives slightly higher values for the Tg as would be expected.

Figure 5. TMA runs at 5 degrees under nitrogen purge with the powder loaded into the base cup of a quartz dilatometer. 50 mN of force was applied with a 3 mm diameter probe in compression. As the material softens at the Tg, a clear deflection is seen.

While the general trend in the pharmaceutical industry is to heat at 5-10 degrees a minute, data can be collected at much faster rates as shown here.

Another option for obtaining a cleaner Tg on the first scan is HyperDSC, the use of high ramping rate techniques to both reduce time and minimize the kinetic effects in a sample. The unique design of a power compensation DSC and its small furnaces allow heating rates of up to 500 °C/minute. Rapid heating allows one to trap the kinetic reactions in a sample by scanning faster than they can occur. Applied to lyophilized formulations, a cleaner Tg is obtained as shown in Figure 4. Note also that the Tg shows little to no shift during the second heat.

Figure 4. Hyper DSC runs on the same lyophilized samples as shown above. A very small amount (1.2 mg) of sample was heated, cooled and reheated at 100 °C/min under nitrogen purge. The Tg is cleanly seen in both heats.

Figure 3. StepScan DSC applies a series of micro-steps of a 2 degree temperature jump at a 5 °C/minute rate followed by a 30 second isotherm to the sucrose-BSA mixture shown in Figure 2, resulting in the heat flow seen above. On processing, the signal gives a thermodynamic Cp curve, which contains the thermodynamic data (in this case a clean Tg), and a kinetic IsoK baseline curve. The resulting Tg is sharp and clean of all kinetic interferences.

Figure 2. As seen in this DSC scan of a sucrose BSA formulation, a DSC scan at 20 °C/minute in nitrogen purge gives a Tg on the first heating. Cooling the sample at a controlled rate and reheating at the same 20 °C/minute gives a cleaner Tg that is slightly shifted to higher temperatures.
Conclusions

Several methods are available for measuring the Tg of lyophilized protein-excipient formulations. The first three require only one instrument, the double furnace DSC. This flexibility makes the double furnace DSC, a powerful and cost effective tool in the pharmaceutical laboratory. The PerkinElmer TMA, with its dynamic and immersion capabilities, represents an underused option for formulation research. The method actually used depends on the specific problems of the particular formulation and the flexibility of these instruments is a strong advantage.