



Heat-Inactivating Serum

What is heat inactivation?

The practice of heat inactivating serum was originally developed when only serum from adult animals was available for cell culture. Adult serum contains various immune factors, particularly serum complement, which may inhibit or destroy cells under certain conditions. Heating serum is intended to inactivate serum complement. Today, serum is often heat-inactivated without any evidence of beneficial effect, simply because an earlier protocol calls for such action. However, certain applications demand the inactivation of complement; e.g., it is often required to preserve the integrity of immunoassays.

Heat inactivation will increase precipitates and consequent turbidity in the serum.

Gemini will provide heat inactivation for most sera upon request, as a service to our customers. Should you prefer to perform this step in house, we recommend the following protocol:

Recommended Protocol:

Thoroughly thaw serum and swirl to homogenize.

Prepare a control bottle containing water. The control bottle should be stored along with the serum bottles to assure identical initial temperatures. This control bottle will be used to monitor the temperature and should be identical to the serum bottle(s) (e.g., PETG with a cap or stopper).

Place the bottle(s) of serum and the control bottle into a 56°C water bath containing sufficient water to immerse the bottles above the serum level. Suspend a thermometer thermocouple in the water bottle. The thermometer should not touch the sides or bottom of the bottle. Swirl the bottles every 3-5 minutes to ensure uniform heating of the serum. Monitor the temperature of the control bottle closely and begin timing when the temperature reaches 56°C.

After 30 minutes (25 minutes for human/equine serum), remove the serum bottles and cool slowly to room temperature. The serum should be left overnight before aliquotting. If the serum is to be filtered to remove precipitate, first it must be cooled to 4°C overnight, then filtered and aliquotted. Heat-inactivated serum may be re-frozen once, thawed and then used.

Heat-Inactivated serum should be stored at -20°C; otherwise the amount of accumulated precipitate will increase as the serum stands in the thawed state, even if left in a refrigerator. The serum should be stored in small aliquots that can be thawed individually as needed. We do not recommend more than one freeze-thaw cycle after heat-inactivation.