

## Product Information

### Calf Serum

Product Number **C 9676**

Storage Temperature -20 °C

#### Product Description

This item has been deleted and is being replaced with Bovine Serum (Product No. B 9433), due to availability. Product No. B 9433 is obtained from animals with the following profile:

healthy animals obtained from an abattoir

16-18 month steers and heifers

1,100-1,300 pounds in weight

serum total protein of 6.5 - 8.5 g%.

Calf Serum is derived from clotted whole blood or plasma, from a donor source or an abattoir, and the animals are less than 10 months old. The product may exhibit some precipitation due to lipid content, which varies from lot to lot. This tends to occur in all serum products and the performance of the product is not affected. Upon microscopic examination, these lipids often appear as large round bodies, even as doublets. Lipid aggregates are readily dissolved when exposed to lipase. Brownian motion may be mistaken for motility. However, these particles will not Gram stain or show any growth in culture on aerobic, anaerobic, or fungal media.

Historically, bovine serum has been the most widely used growth supplement for cell culture media because of its high content of growth promoting factors. When used at appropriate concentrations, it supplies many defined and undefined components that have been shown to satisfy specific metabolic requirements for the culture of cells *in vitro*. Serum is an extremely complex mixture of many small and large biomolecules with different, physiologically balanced, growth-promoting and growth-inhibiting activities. Some of the major functions of serum are to provide growth factors, hormones, attachment and spreading factors, binding proteins, lipids, and minerals.<sup>1</sup>

The quality, type, and concentration of serum can all affect the growth of cells. It can also affect cloning efficiency, plating efficiency, and the cellular morphology. For use in cell culture media, supplementation with calf serum ranges from 5-20%. The optimal concentration must be determined for each cell line and application.

#### Precautions and Disclaimer

For Laboratory Use Only. Not for drug, household or other uses.

#### Storage/Stability

This serum is stable when stored at the recommended storage temperature of -10 to -40 °C, until the date indicated on the label. Serum should not be kept in a "frost-free" freezer unit. The repeated heating cycles of the freezer unit during self-defrosting can be harmful to serum.

#### Procedure

##### Thawing

1. Remove the serum from the freezer and allow it to sit at room temperature for 10 to 20 minutes.
2. Place serum bottles in a 30 to 37 °C water bath. It is very important that the temperature of the water bath not exceed 37 °C. Higher temperatures may denature the proteins, potentially gelling the serum.
3. Gently agitate each serum bottle every 10 minutes until completely thawed. Do not vigorously shake the bottles. Extreme agitation can cause foaming. It is very important that the serum be mixed thoroughly throughout the thawing process. Failure to agitate during the thawing process can cause the serum lipids and proteins to precipitate, resulting in turbidity or flocculence.
4. It is important to remove the bottles from the water bath when completely thawed. Do not allow the bottles to remain in the water bath for an extended period of time. The serum should never become hot or very warm.
5. When the serum is completely thawed, refrigerate promptly at 2 to 8 °C. For best results, aliquot serum into smaller, single-use sizes and return aliquots not intended for immediate use to the freezer.

6. It is not uncommon for trace amounts of cryoprecipitate, fibrin, or flocculent matter to remain. This will not affect the performance of the serum. However, if you wish to remove this, return the serum to the 37 °C water bath and gently swirl or mix the serum for several minutes. This will usually allow the proteins or lipids to go back into solution. This may also disappear when the serum is added to the culture media and incubated at 37 °C.

#### Heat Inactivation

1. Prior to heat inactivation, thaw the serum according to previous instructions. Allow containers to come to room temperature. Swirl bottles of serum immediately before adding to the water bath.
2. A water-filled control bottle (T1) should be placed in a 56 °C water bath. Place the serum bottles in the water bath, so the water level matches the serum level in the bottle. Do not completely submerge the containers. When the temperature of T1 reaches 56 °C, start the timer set for 30 minutes. Do not allow temperatures to exceed 56 °C. Higher temperatures will denature the proteins, gel the serum, or alter the performance of the serum.

3. Gently swirl the bottles every 5 to 10 minutes while in the water bath and check the temperature in the control bottle (T1). Plastic containers may bow slightly after heat treatment. This is not harmful to the serum.
4. At the end of 30 minutes remove the serum bottles from the water bath. Allow the bottles to return to room temperature before returning to the freezer. Failure to allow the serum to cool before freezing can cause the bottles to crack. If desirable, aliquot the serum into smaller volumes and then freeze. Repeated thaw-freeze cycles are not recommended.

#### **References**

1. Freshney, R. I., Culture of Animal Cells: A Manual of Basic Technique, 3rd ed., (John Wiley & Sons, Inc., 1994) pp. 90-91.
2. Barile, M. F. and Kern, J., Isolation of *Mycoplasma arginini* from commercial bovine sera and its implication in contaminated cell cultures. Proc. Soc. Exp. Biol. Med., **138(2)**, 432-437 (1971).

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