

Heat Inactivation of Fetal Bovine Serum
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In this procedure, fetal bovine serum (FBS) is heated to 56 °C in a water bath to destroy heat-labile complement proteins prior to use in cell growth medium. This procedure is similar to that recommended by HyClone (HyClone Handling Serum procedure; 1-800 HYCLONE).

Heat Inactivation Procedure

1. Remove 500-ml bottle of FBS from –80 °C freezer and place in refrigerator to thaw overnight. Complete thawing of serum the following day by placing serum in a 37 °C water bath with a water level higher than the serum level in the bottle. Mix by inversion.
2. Once serum is completely thawed, incubate for an additional 15 min to allow serum to equilibrate with the 37 °C bath.
3. Raise the temperature setting of the bath to 56 °C. Use a timer to measure the 35 min needed for the temperature of the bath and serum to come to 56 °C. During this incubation, invert the bottle to mix the serum every 10 min. If necessary, allow an additional 5 min for bath to reach 56 °C.
4. Once bath reaches 56 °C, incubate serum for 30 min. Invert bottle every 10 min.
5. Remove serum from water bath and allow to cool at room temperature for 30 min. Reset water bath to the 37 °C mark.
6. Aliquot 50 ml of treated serum into conical tubes and store at 4 °C or freeze at –20 °C.

Reagents and Materials

Fetal bovine serum (FBS): GIBCO/Invitrogen; catalog no. 26140079

Conical tubes, 50 ml: Greiner; catalog no. 4943

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