

Technical Bulletin

Dialyzed Fetal Bovine Serum and the DHFR⁻ System

The dihydrofolate reductase negative (DHFR⁻) system is a common and powerful selection method. In order to be compatible with DHFR⁻ applications, all cell culture media and supplements must not contain hypoxanthine or thymidine. Even low levels of these 2 nucleotide precursors can cause interference and destroy the effectiveness of the DHFR⁻ system. Dialyzed Fetal Bovine Serum (DFBS) (Catalog No. 12117) lacks hypoxanthine and thymidine while providing the nutrients necessary for healthy cultures.

To remove low molecular weight components such as hypoxanthine and thymidine, Fetal Bovine Serum is dialyzed against 0.85% saline through a 10 kDa cutoff membrane. The extent of the dialysis process directly affects the final levels of hypoxanthine and thymidine and must be closely monitored and controlled. Because in process monitoring of hypoxanthine and thymidine concentrations is costly and time consuming, the level of serum glucose, which can be assessed quickly and easily, is used as a marker.

SAFC Biosciences performed controlled experiments to track the levels of hypoxanthine, thymidine and glucose in serum at various points during the dialysis process resulting in 2 important pieces of information. First, there is a high correlation ($r=0.956$) between the levels of the nucleotide precursors and glucose remaining in serum. This means that the concentration of glucose can be used to directly and accurately determine the concentrations of hypoxanthine and thymidine. Second, the studies showed that when serum glucose levels drop to 5 mg/dL (0.05 g/L), corresponding hypoxanthine and thymidine levels are undetectable by high pressure liquid chromatography (HPLC) analysis.

All DFBS manufactured by SAFC Biosciences is processed until serum glucose levels are at or below 5 mg/dL. This provides our customers with confidence that they have a consistent product for their cell culture applications such as the DHFR⁻ system.

For more information about this subject or other SAFC Biosciences' products and services, please contact our Technical Services department.

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Issued December 2005 XT003
0103

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