Precipitates in serum products

HYCLONE SERA

There are several types of precipitates that can and do exist in fetal bovine serum (FBS) and other serum products used in cell culture. In addition to fatty ester of cholesterol and some protein precipitates, fibrin and calcium phosphate commonly constitute precipitates in sera. While precipitates can induce initial concern, there are no adverse effects caused by precipitates on the performance of the serum in cell culture applications.

Fibrin

Fibrin appears in serum as larger material (up to 1–2 mm) visible to the naked eye. Because serum is collected and rapidly processed under cold conditions, some fibrinogen (the soluble precursor of the clot-forming protein fibrin) can remain in solution during the production process. After final filtration, the clotting process can be completed, resulting in fibrin ending up in your bottle of serum as a precipitate.

Calcium phosphate

Calcium phosphate is also a common precipitate that will appear as a general cloudiness in the serum. This precipitate appears as small black dots when observed under an inverted microscope. These dots can appear to be moving due to Brownian motion.

This type of precipitate is often mistaken as microbial contamination. Calcium phosphate precipitate increases upon incubation of the serum at 37°C. Hence, we recommend serum not to be tested for sterility by direct incubation of the serum at 37°C, but by plating 1 mL of the suspect serum on a bacterial agar plate (such as a Tryptone soy agar) and then incubating the plate at 37°C and looking for the growth of bacterial colonies. Also, a Gram stain of the material observed under oil (1000 × total magnification) can confirm the presence or absence of contamination.

Cell Growth

Testing and experience indicate that precipitates do not alter the performance of the serum as a supplement for cell culture. This observation has been confirmed by customers and other serum suppliers.

Filtration

If enough precipitate is present in the serum, it can contribute to difficulty in filtering serum. In general, we do not recommend re-filtering serum products for cell culture. HyClone™ sera pass through final filtration using either 100 nm or 40 nm filters and undergo extensive quality testing. In a laboratory setting, there should not be a need to re-filter the serum product. In large-scale manufacturing, it is common for the serum to be filtered with the medium directly into the culture vessel.

Management of precipitates

Precipitation in serum can be difficult to predict and prevent. However, precipitation does not alter the performance of the serum.

Precipitation will increase with:

- Heat inactivation of the serum
- Incubation at 37°C
- Freeze and thaw processes
- Thawing without mixing
- Gamma irradiation
- Long term storage at 2°C to 8°C
- Storage in a self-defrosting freezer

The tendency of serum products to form precipitates varies, and actual causes are not fully understood. We have not determined any trends that would allow us to accurately predict the presence or degree of precipitation. Precipitates can appear in all serum products regardless of the supplier. This can be confirmed by statements from other serum suppliers.

cytiva.com/hyclone

CY15727-13Nov20-DF

cytiva