

## Sf9 Spinner Culture Media Recipe

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-CLW

Media to be use with Sf9/Sf21/HiFive insect spinner flask culture. Can also be used for plate cultures, though Pluronic Acid, Gentamicin, and Fungizone may inhibit cell growth/viability and decrease protein expression/secretion. Try using protein free (serum free) media when optimizing protein expression levels.

Preferable, begin with GIBCO Sf-900 II media serum-free media. Sf-900 media is shipped in 500ml or 1000ml volumes. The recipe below is calculated for a single 500ml Sf-900 II media bottle to give the following final concentrations:

10% (v/v)	Fetal Calf Serum (FCS)
10 µg/ml	Gentamicin (antibiotic)
1.0% (v/v)	Fungizone (fungicide)
0.1% (v/v)	Pluronic-F68 (detergent to decrease sheer forces in culture)

### Spinner Culture Media Recipe (500ml Bottle):

1. Begin with 500ml bottle of Sf-900 II serum-free media  
(GIBCO, cat no. 10902-096)
2. Add 50ml of FCS, 100% v/v.  
(freezer stocks are in 25ml aliquots)
3. Add 0.1ml of Gentamicin Stock, 50mg/ml.  
(GIBCO, cat no. 24040-016)
4. Add 5ml of Fungizone Stock, 100% v/v.  
(Irvine Scientific, cat no 9352)
5. Add 0.5ml of Pluronic-F68 Stock, 100% v/v.  
(GIBCO, cat no. 15750-011)

Store media at 4°C, prewarm to 27°C (Room Temperature) when using in Tissue Culture