



Technical Note:

Sf9 Growth Suggestions for using a Bellco Bioreactor Vessel

Introduction

A basic outline is provided below to identify the key operating parameters to be followed to enable the transfer and scale up of suspension Sf9 insect cells from shake flasks to bioreactors. Standard cell culture procedures per the Invitrogen manual should be followed. As this manual highlights only spinner flask use, the following are recommended for scale up.

Scale up Recommended Procedures

- The Sf9 cell line should be passaged from smaller vessels such as shake flask or spinner flask.
 - Seeding of a larger vessel should occur from there.
 - If transferring to a serum-free environment, those conditions should be optimized in smaller volume cultures before moving to larger volume.
- Sf9 cells are optimally grown at 27degree C.
 - At this temperature, the cells should double every 24 to 30 hours.
- An impeller speed should be selected between 80 to 130rpm.
- However, when scaling to a stirred tank bioreactor from a spinner flask, please note that bioreactor impellers are smaller relative to the media volume than spinner flask impellers than so impeller speed will have to be optimized to provide efficient mixing.
- 115 rpm should be the target initial mixing speed for the bioreactor and optimization should occur from there.
- Optimal working volumes should be 30-40% of the vessel volume.
- Samples should be taken once every 24 hours using a 5ml syringe to ensure sterility is maintained.
- Heating blankets are the standard for keeping a larger system at accurate temperature.
- Starting volume: 2.5L for seeding
- Inoculation Density: 0.5×10^6 cells/ml
- Agitation: 115 rpm
- Temperature: 27 degrees C

References

1. Bellco Technical Report TN-2025-001. General Recommendations for Using Bellco Spinner Flasks for Insect Cell Suspension Cultures.
2. Bellco Technical Report TN-2025-002. Key Points to Consider in Spinner Flask Cell Culture.
3. Bellco Technical Report TN-2025-003. Challenges in Scaling a Microcarrier Based Cell Culture from Spinner Flasks to Stirred Tank Bioreactors
4. Invitrogen Life Technologies. Growth and Maintenance of Insect Cell Lines. Rev K (2002). 25-0127

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