PREVENTING CELL CULTURE CONTAMINATION

Introduction

Cell culture is increasingly prevalent and important in academic research, and along with improved proficiency of cell culture comes a better understanding of the risks and consequences of cell culture contamination. Testing methods and preventive tools are continuously improving, and an awareness of the effects of contamination requires researchers to be conscientious and vigilant. Undetected contamination can lead to widespread downstream effects. Therefore, it is important to understand how contamination can occur and what methods are available to prevent it.

Biological contamination includes bacteria, fungi, Mycoplasmas, viruses, and cross-contamination from other cells. Bacteria and fungi are the most common contaminants, because they are ubiquitous and able to colonize quickly. They are also the easiest to detect. Mycoplasmas can be the most difficult to detect and are resistant to antibiotics. Once detected, contamination can be contained and eliminated, but continuous testing is required.

Even a tiny amount of liquid can cause contamination - one drop of media left on a bottle thread can have a detrimental effect on your resulting data.

Common Sources of Contamination in Labs

**Laboratory Practices**

- Aerosolization of particles
  - Sneezing, coughing, or stirring up dust while moving about the lab
  - Pipetting, vortexing, centrifuging without bio-containment vessels
- Working with multiple cell lines concurrently
- Continuous culturing of working banks
- Overcrowding of items in the autoclave
- Mixing solutions and supplies

**Laboratory Equipment**

- Reservoirs for microbes and fungi
  - Improperly cleaned incubators
  - Water baths, refrigerators, microscopes, cold storage rooms
- Use of feeder cells
- Culture media, bovine sera, reagents, and plasticware
- Use of non-sterile supplies, media, or solutions
- Materials not certified for cell culture use
- Contaminated clothing
Preventing Cell Line Contamination

A contaminated cell line can jeopardize resulting data, create health hazards to lab workers, damage researcher credibility, and waste resources.

What Can You Do To Prevent Contamination?

Effective training and supervision by principal investigators is critical. Regular use of antibiotics is discouraged, because it can mask habitual mistakes or poor techniques. The best prevention against cell contamination is ensuring good laboratory practices among all personnel.

- **Documentation of all cell culture work**
  - Each passage, manipulation, and general cell appearance
- **Ensure proficiency of lab personnel**
- **Proper aseptic techniques**
- **Aliquot stock solutions/reagents**
- **Use one reservoir of medium per cell line**
- **Work with one cell line at a time**
- **Work in a biological safety cabinet**
- **Manageable work load**
- **Use seed stock**

- **Label flasks legibly**
  - Include name of cell line, passage number, date of transfer
  - Use barcoded flasks when possible
- **Allow >5 minutes for air to be filtered between working with each cell line**
- **Quarantine “dirty” cell lines from “clean” cell lines**
- **Clean laboratory regularly**
- **Periodically review and approve lab notebooks**

Further Reading: