CELL CULTURE'S MOST 9µm 8um 7_{um} 6µm 1. MYCOPLASMA **3, BACTERIA** 2. YEAST OWING TO THEIR UBIQUITY, SIZE, AND RAPID SIMPLE BACTERIA THAT LACK A CELL WALL, GROWTH RATES, AMONG THE MOST COMMONLY MYCOPLASMA ARE CONSIDERED THE SMALLEST YEAST PARTICLES CAN QUICKLY CONTAMINATE ENCOUNTERED CONTAMINANTS. SELF-REPLICATING ORGANISM. CULTURES AND THEIR RELATIVELY LARGE SIZE **5**µm — MAKES THEM EASY TO IDENTIFY. SIGNS SIGNS SIGNS 1. Due to their very small size they are difficult to detect until they achieve 1. Infected cultures become turbid 1. Infected cultures appear cloudy extremely high densities and cause the culture to deteriorate.

- 2. Chronic infections may manifest with decreased cell proliferation. reduced saturation density, and agglutination
- 3. Can be detected using fluorescent staining, immunostaining, autoradiography, ELISA, or PCR.
- 2. Very little change in pH until contamination becomes heavy, when pH usually increases.
- 3. Under microscopy, yeast appear as oval or spherical particles that may show budding of smaller particles
- 2 May be a thin film on surface
- 3. Often accompanied by sudden drop of pH of the culture medium
- 4. Visible under a low-power microscope as tiny, moving granules. Unde higher magnification, bacteria can display a variety of shapes, including spheres, rods, and spirals,

Anyone who has ever worked in a cell culture lab has experienced it—contamination. Ranging from minor annoyances to major catastrophes, contamination causes loss of time, money, and effort spent in developing cultures and setting up experiments, and can lead to inaccurate results and loss of credibility.

4. MOLD

MOLDS ARE EUKARYOTIC MICROORGANISMS THAT GROW AS MULTICELLULAR FILAMENTS CALLED HYPHAE, SPORES CAN SURVIVE VERY INHOSPITABL CONDITIONS TO BECOME REACTIVATED WHEN SUITABLE CONDITIONS EMERGE.

- 1. Infected cultures remain clear until culture becomes heavily infected and media becomes turbid
- 2. pH remains stable during initial infection, then rapidly increases
- 3. Under a microscope, mycelia appear as thin, wispy filaments, sometimes with a denser cluster of spores.

SIGNS

- 1. Cause cytopathic effects resembling viral damage and can destroy cultures in a matter of days
- 2. Visible under a microscope but slow growth and similarities to cultured cells make detection difficult

5. PROTOZOA

ALTHOUGH RARE, AMOEBA ARE OCCASIONALIY

IDENTIFIED AS CELL CULTURE CONTAMINANTS

 No observable signs apart from cytopathic or other adverse effects on culture

6. VIRUS

NOT PARTICULARLY COMMON, VIRUSES CAN BE

DIFFICULT TO DETECT AND MAY POSE THREATS TO

PRIMATE CELL LINES.

RATORY STAFF WHEN INFECTING HUMAN OR

2. Can be detected by electron microscopy, ELISA, or PCR with appropriate viral primers

PROTECT YOURSELF FROM CELL CULTURE CONTAMINATION

SELECT THE RIGHT EQUIPMENT

- . Choose an incubator with that has design features that will help prevent contamination
- 2. Copper housings and parts can fight contamination-choose them when appropriate
- 3. Internal HEPA filters will reduce or eliminate many airborne particles

USE GOOD ASEPTIC TECHNIQUE

- . Use sealed culture vessels whenever possible
- 2. Avoid pouring media
- 3. Use clean lab coats and restrict them to the cell culture area
- 4. Work with one cell line at a time
- 5. Leave the hood running 24 hrs a day

KEEP YOUR INCUBATOR CLEAN

- 1. Remove humidity pan, shelves, shelf supports, and shields weekly and autoclave all stainless steel parts
- 2. Use disinfectants as appropriate and rinse with fresh distilled water
- 3. Wipe down chamber with disinfectant and allow to dry
- 4. Disinfect all access ports, electrical pass-through, shaft holes etc. and carefully clean around sensors

PRACTICE GOOD HOUSEKEEPING

- Dirty water baths can be a source of contamination and generate aerosols
- 2. Waste containers provide a source of heavily contaminated materials and should not be located near the hood
- 3. Pest control-mice, ants, roaches, flies, and mites, can all be sources of contamination. The presence of food or plants in the lab can attract these undesirable guests

DUTINELY MONITOR FOR CONTAMINATION

- 1. Perhaps the best strategy for reducing contamination is to be proactive and routinely monitor for it
- 2. Supplies, media, work areas, and cultures should be routinely tested for contamination

USE ANTIBIOTICS SPARINGLY

- . Overuse of antibiotics can lead to poor aseptic techniques and resistance
- 2. Use antibiotics only strategically to prevent the loss of critical cultures

