Mold Growth in NIH Cold Rooms

Introduction and Purpose

The NIH Main Campus currently has an inventory of almost 200 walk-in cold rooms. In many of them, mold growth appears to be an existing, and on-going concern. The sources of mold growth inside these areas are varied and are usually not related to issues with the mechanical systems that maintain the conditions inside the units.

The aim of this document is to standardize procedures to assess, prevent, and – if necessary – remediate mold contamination within these spaces. It is possible with a modicum of effort to ensure a mold-free environment inside the cold rooms.

Bottom Line Up Front (BLUF)

Cold rooms are prone to elevated humidity, which may contribute to the growth of mold. Follow these steps to prevent mold growth:

- Minimize personnel activity inside the cold room
- Remove any items/materials that can contribute to mold growth (e.g. wood, particle board, cardboard, paper products, and absorbent materials)
- Upon entering the cold room, perform a visual observation check for mechanical and structural issues (e.g. condensation, broken evaporator units and fans, faulty latches and door seals, clogged sinks, and inconsistent temperatures)
- Implement a schedule for regular cleaning of the cold room

General information about mold

While most think of warm, moist environments as supporting mold growth, colder temperatures with high moisture can also provide conditions amenable to mold proliferation. In order for mold to grow, it needs a substrate (surface) upon which to settle; a nutritional source (food); and available moisture/humidity, especially chronically moist/humid conditions. The ambient temperature of an area provides little influence on the ability of mold to grow.

Mold spores are all around us; in the air, in our hair and clothing, and as “dust” on surfaces; and in the indoor and outdoor environments. As long as evaporation/drying manages to stay ahead of accumulating moisture, then mold proliferation is unlikely to occur. However, when moisture accumulates more rapidly than the natural drying process, the microbial ecology changes and can amplify mold growth to easily visible patterns and blotches that we associate with this phenomenon.

Mold becomes a concern because some people have; or can develop a sensitivity to mold that manifests itself in an allergic reaction. According to the CDC, typical symptoms include itchy, watery eyes; runny nose; cough, wheezing and a scratchy throat. In some people the allergy concerns are more serious. People who have existing strong allergic responses to certain substances may be more susceptible to serious mold allergic reactions. People who are in an immunocompromised state or have asthma or other underlying respiratory conditions may also be at risk for significant (and sometimes severe mold reactions). In a few rare cases, toxins produced from some mold types can cause a life-threatening situation.
General Cold Room information

Refrigerators and walk-in cold rooms are ideal environments for mold to grow due to high moisture/humidity levels, and abundant food sources such as paper, cardboard, cell culture media, etc.

Most cold rooms operate as a “closed” ventilation system. This means that air is constantly recirculating through the room itself. As the diagram at right shows, cold air blows out from the evaporator (the fans near the ceiling in most NIH units) and cycles through the room. As it circulates through the room it warms up (relatively), where it is brought back into the evaporator, and chilled once more before starting the cycle anew. This pattern can allow humidity to build up in the space. Opening the door is usually the only way “fresh” air gets into this space. However, allowing warm fresh air into the room increases the overall ambient temperature, causing the condensing and evaporator units to work harder. This scenario increases the humidity levels. In addition, this influx of warm air allows condensation to form on surfaces, which also increases the risk of mold growth.

Causes of mold growth in cold rooms

The elevated humidity levels in refrigerators and walk-in units are ideal environments to support mold growth. There are several contributing factors that can increase the risk of mold growth inside a cold room. They can be roughly broken down into infrastructural and procedural factors.

Infrastructure issues:

- If there is a failure in the evaporator or condenser units, users will likely notice by changes in the monitoring status, or elevated temperatures when entering the unit. With proper maintenance, a breakdown in these systems is likely to be very rare. If addressed right away, significant mold growth is unlikely.
- Other infrastructural issues can include a blockage in the evaporator drip pan. The drip pan is a shallow pan underneath the evaporator unit. There is a drain in one end (you can follow the drainpipe from the pan to its end, either in a sink inside the cold room, or to another wastewater pipe outside the unit). An overflowing drain pan is caused from the drain pan or line being clogged. If the drip pan fills with water, there may be a clog in the drainpipe. If you can reach around inside the drip pan, near the edge, with a finger, or piece of stiff paper, check to see if it is wet when you pull your finger, or the paper out. If using a finger, be cautious of any potentially sharp edges. This is usually an easy fix that can be resolved by clearing the line. There are products available that can be placed in the drain pan to help breakdown and reduce the sediment in the lines.
What kind of casework is inside the space? Some cold rooms (especially older ones) have shelves, cabinets and countertops that are made with exposed particle board, on at least one of the sides. This material can absorb moisture and contribute to increased humidity levels inside the room; as well as support the growth of mold itself. It is recommended that metal cabinetry, shelves and countertops (stainless steel if available) be used, as these items are easier to clean and less likely to contribute to mold growth.

Check the seals/gaskets around the entry door(s). The seals should be clean, and not frayed or deteriorating. Check especially down near the bottom of the door. These can become extremely dirty or worn out over time. A defective door seal can allow for air outside the room to enter the cold room and increase humidity and condensation. Torn/worn door gaskets are fairly inexpensive and easy items to replace. The best way to check the condition is to actually walk inside the cooler with the lights off and close the door. If you can see light coming in, then they might need replacement.

Lastly, ensure that the door closes firmly on its own. It should form a good seal around the frame and take some effort to push back open. The door closure assist on the outside of the door grabs the door and pulls it tightly closed. These assists are usually made up of springs or a small hydraulic cylinder. Both can wear out over time resulting in a door that never completely closes.

If there are problems with any of these infrastructural items, please contact your ORF building facility manager and ask to have them evaluated. Your building manager may be able to assist with replacement of old casework with newer types as well.

**Procedural Issues:**
While it is important to see if there is anything structurally significant with the cold room that can potentially enhance the risk of mold growth, in the vast majority of the cases, there are issues with how the room is used, and what is being stored within the cold room that have a greater likelihood of contributing to mold risk in the space.

- The first question to ask is when was the room last cleaned? Periodic wipe downs of surfaces and materials, as described below, can be a significant contributor to minimizing mold growth.
- In addition, many of these rooms contain abundant mold food sources such as paper, cardboard, growth media, and certain types of caulking.
- And as stated above, mold growth in these types of units is exacerbated by temperature swings resulting from too frequent entries and exits from the space.
- The amount of people in a space, and for how long they – or any individual – is inside the cold room are also major contributors to increased humidity levels inside the room. Human beings exhale large amounts of moisture along with carbon dioxide. In addition to increasing mold growth risk, personnel can experience inhalation exposure risk due to a buildup of carbon dioxide when they are in cold rooms. Some institutions have run studies to survey the buildup of carbon dioxide in a cold room and determined that the OSHA Permissible Exposure Limit (PEL) for carbon dioxide of 5000 ppm is achieved when 4 individuals occupy a cold room continuously for a period of about 55 minutes. Although the OSHA PEL is for periods of 8-hours, most recommend limiting the number of people who can occupy a cold room safely, based on the
carbon dioxide level, to no more than 2 people-hours per day, to provide for variations in the size of the cold rooms.

- Keep doors firmly shut – if left open, water condensation on surfaces increases due to high relative humidity, promoting mold growth.
- Immediately clean up spilled laboratory liquids, e.g., buffers and media. Moisture may lead to rust, corrosion or degradation of environmental room integrity, e.g., shelves.
- Limit the storage of organic materials, e.g., paper products, cardboard, miscellaneous trash, etc., and promptly dispose of wet or damp materials
- Store paper & porous materials, e.g., Kimwipes, cardboard, blotting paper, etc., in closed, air-tight, plastic containers. Do not use cardboard boxes or other absorptive materials as storage containers in environmental rooms.
- Do not use the cold room as a storage closet. The only items and equipment that should be inside the cold room are those things that must be kept at cold temperatures. Manage your inventory, and do not over-purchase supplies.

**Identifying Mold Growth in the Cold Rooms**

Obvious visible mold includes the black/grayish growth that occurs around the sinks, on bench paper, cardboard or other cellulosic materials, and on the walls, ceilings, floors and laboratory equipment. However, the “not so obvious” mold growth includes the white, powdery substances commonly found on all surfaces of the unit as well. Most people assume this is oxidative degradation, but it is in fact a species of mold. The photos below show some examples of cold-room mold growth, and the different types of surfaces on which it can grow.
Preventing mold growth in cold rooms

Unabated mold growth within the cold room may lead to mycological contamination of storage areas and materials, research projects and elevate the risk of potential health problems due to the inhalation of mold spores. Spores can also be tracked out of the room and around the entire floor of the building. Minimizing mold growth requires the control of moisture, e.g., standing/leaking water, limiting the presence of organic materials, minimizing the amount and length of time that personnel spend inside, and practicing good housekeeping principles in the space.

Please strongly consider implementing these practices for any cold rooms under your cognizance. The Appendix for this document contains sample checklists for both routine entry checks, as well as for periodic cleaning of the space.

Minimizing personnel activity:

- Because cold rooms do not have ventilation systems, recommended occupancy of cold rooms is limited to a total of two hours per 24-hour period (1 person for 2 hours, 2 people for 1 hour, etc.).
- Entrants into the cold room should know exactly what they are going to be doing, and what they need prior to entering the space. Have a plan before going through the door.
- Never prop open the door. Ensure that it closes securely behind you when you enter into the space, and when you exit it. Besides introducing extra humidity and condensation, keeping the door open can also introduce additional airborne mold spores into the cold room.

Removing any items/materials that can contribute to mold growth:

- The storage of cellulose containing materials is a leading cause of mold growth. Mold growth can contribute to contamination of research materials. Preventing mold growth in cold rooms is achieved by controlling condensation/moisture and removing materials contributing to mold growth.
- Remove all wood. Wood and particle board shelves and cabinetry can absorb moisture and, because it is composed of cellulose, is an ideal breeding ground for mold. Wood shelves should be replaced with open stainless-steel shelves that permit air flow throughout the storage area.
- Remove all cardboard and paper products. These surfaces act just like wood and promote mold growth. If some paper products (e.g., Kimwipes, some storage boxes) are required, place them in a closed plastic container between uses. Should visible mold be found on a paper product, discard the item immediately.
- If absorbent materials must be stored in the cold room, store them in plastic containers with lids that provide a good seal.
- If unpacking delivery boxes inside the cold room, do not leave the packing materials behind; and do not store the entire box in the cold room. Do not store full, unopened shipping boxes inside the room.

Checking the following, each time you enter the cold room:

- Check for signs of condensation. Make sure that there is no standing water in your walk-in cooler that can contribute to condensation build up. Wipe up any spills or standing water.
- Ensure that the evaporator unit, and fans appear to be working properly. If there is more than one unit, make sure to check all of them, each time.
• Frequent visual observations should become standard procedures for all walk-in cold rooms on campus.
• Report all plumbing leaks, faulty latch/door seal, inconsistent temperatures or lighting issues or electrical problems to your facilities management team.
• If your unit contains a sink, check that there is no standing water in the sink, and report any clogged drains.
• Some units have their condensers above the ceiling of the cold room. These are usually in Permit Required Confined Spaces (PRCS) and are labeled as such. Do not remove ceiling panels; or go into these areas as special procedures are necessary to enter the PRCS.

Regularly cleaning the cold room:
• Cold rooms need to be cleaned on a regular basis. If the coolers are shared among several groups, then establish a Standard Operating Procedure and schedule for cleaning responsibility. Although cleaning sounds like a huge undertaking, it is quite simple. Wet cleanup activities are recommended (dry sweeping, dusting, or brushing can release mold into the air and may cause inhalation exposures and spread potential contamination).
• There is no set policy on how often these rooms should be cleaned. Frequency is usually dependent on several different variables, including how often it is used; what is stored within the space, etc. A good rule of thumb is to start cleaning on a quarterly basis and increase or decrease frequency as needed.
• Consider implementing a cleaning history record for cold rooms.
• The typical cleaning process:
  o Remove all unnecessary paper products from the walk-in unit, or place in sealed plastic bins.
  o Use a slightly diluted (similar dilution as when washing dishes) general dish detergent; and wipe all surfaces down with a cloth towel. Wipe up (or dry off) any excess water to prevent additional moisture from being present in the cooler.
  o Other options for cleaning solutions -particularly for addressing mold growth - include a 1:10 dilution mixture of household bleach. However, never use bleach on metal surfaces (bleach on metal can result in pitting of the material). You can wipe down metal surfaces using a 70% ethanol solution to avoid pitting on the metal.
  o Recommended personal protective equipment would be splash goggles, lab coat and disposable gloves; standard laboratory PPE. Unless wiping down mold growth, respiratory protection in the form of a surgical mask, or N-95 respirator is usually not required. Even then an N-95 respirator is not usually warranted. Most mild to moderate mold growth inside the cold room can be effectively and safely addressed by lab personnel with standard laboratory PPE.
    If there is mold growth in the space (including minor amounts), personnel who have a known sensitivity to mold/other allergic conditions; or, other respiratory conditions should not engage in cleaning the cold room until the moldy materials and/or surfaces have been addressed.
  o Surfaces to be cleaned include:
    • Interior and exterior surfaces of cabinetry
- Countertops, work benches, and table surfaces
- Shelves
- Inside of door(s)
- Door gaskets
- Lab-owned equipment and associated parts
- Light switches
- Sinks
- Walls, including white, powdery substance that can be mistaken for oxidation
- Floors
- Window (if applicable)
- DO NOT WET WIPE ELECTRICAL FIXTURES/OUTLETS. UNPLUG ALL ELECTRICAL EQUIPMENT PRIOR TO WIPING DOWN.

- When cleaning/disinfecting any moldy areas – even with a bleach solution – the mold may leave behind stained surfaces that are not removed no matter how vigorous the scrubbing. These do not present any elevated risk, nor are these areas more likely to flare up again if regrowth occurs.
- Mold is not considered a hazardous waste product. Cleaning materials, or moldy items do not need any special waste handling (unless the materials are designated as such to begin with). They should be bagged up in plastic bags, tied closed, and disposed of in the regular waste stream.
- Be prepared to show documentation of cleaning to DOHS personnel when laboratory inspections are conducted.

If the cold room has been allowed to have rampant, unchecked, and heavy mold growth, the research team(s) may elect to have a professional remediation team engaged to provide this initial clean-up. If this is the case, please contact the DOHS office for a consultation to determine the best course of action, and for information to assist you on initiating contact with a professional cleaning service.

**Long-term shutdowns of the cold room**

If the cold room is going to be shut down for an extended period (greater than 24-hours), please follow these steps:

- Empty the cold room of anything that is going to be stored elsewhere. To be safe, wipe down any items to be relocated with a disinfecting solution to minimize the risk of introduction of contaminants to the new location.
- Discard any items no longer needed through the proper waste stream(s).
- Thoroughly clean the unit using the prescribed cleaning steps outlined above.
- Keep the doors open to allow for complete drying, and do not close the doors until the unit is ready to be re-entered into service.
- Let the empty unit run for about 24 hours before returning materials back into the cold room.

**References**
