



January 2009

## Monthly Newsletter

This is the third installment of our newsletter. Today we introduce coliforms, how to handle ERMI dust samples and other spore-trap structures that you may not be aware of.

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**Coliform Testing** by  
-E.A. Sobek, Ph.D.

**Ensuring the Integrity of Your Dust Sample.**  
-Merissa McGraw



**The Trace**  
- Lyn Pope

### Coliform Testing

Coliforms are fecal-associated bacteria and have the potential to cause disease. The most common is *E. coli*. Indoor contamination by coliforms is a primary concern when black water contamination is suspected. There are three types of water that you should be aware of:

**Potable Water-** Drinking or tap water; water that is safe for human consumptions.

**Grey Water-** Water from domestic household use such as dishwashing, bathing, and laundry.

**Black Water-** water associated with sewage, and contains fecal material.

Black water contamination is most often associated with sewage backup. The toilet gets clogged and overflows or the city is working on the sewer lines and inadvertently, causes blowback, whereby sewer sludge is reversed and forcible ejected back into a home or business.

There is an episode of "Dirty Jobs" whereby, Mike Rowe, the show's star and narrator, has to clean out a woman's bathroom in New York city that was encrusted with sewage from blowback). Believe me this can happen. I know firsthand! Three years ago, the city of Oak Ridge was clearing sewer lines outside the lab. I was working at my lab bench and heard this gurgling sound coming from the bathrooms. The gurgling sound turned into a horrid gushing howl and the floor began to vibrate.

Then came a most noxious, foul odor, second only to a rotting carcass that has been in the sun for a week. I walked to the men's restroom and there was a nasty liquid seeping from under the door (black water). When I opened the door (very slowly mind you), the stench about knocked me over and there was sewage all over the walls. The blowback was so strong that the toilet lid was blown clear off and lying in the sink. Needless to say, I was extremely irritated with the City's lack of safeguards to prevent blowback. However, they did come in and clean it up and even-

tually passed my clearance test. One city worker suggested that I, "put a cinder block on the toilet lid the next time we clean out the lines", which would presumably prevent sewage from blowing all over the bathroom walls.

If you are called in as an IEP (indoor environmental professional), and black water is readily identified as the moisture source, you'll probably recommend an EPA registered biocide treatment alongside standard remediation methods. In addition to any mold clearance testing, you'll want to recommend a coliform test to ensure that all the harmful bacteria have been killed. I recommend swab samples. Depending on the size of the area and sanitation level required (medical examination room vs. home bathroom) one or more swabs samples should be collected. In a medical situation more samples are recommended with a zero tolerance test protocol. Start by dividing the room into quads and swabbing multiple areas from each quad. Hence for quad one, use a single swab to sub-sample multi-

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ple areas. Don't just swab the floor, remember bacteria can readily become aerosolized and contaminate walls and ceilings. Repeat the procedure in each quad. Get your swabs from the lab. They will have sterile swabs in a buffer solution (maintains coliform viability). If any samples come back positive recommend a second biocide treatment of that particular quad(s).

The other situation where a coliform test is often requested involves potable well water. Municipalities test drinking water daily for coliforms, so inspectors really have no cause to test tap water that is supplied by a municipality. However, well water is another story. Wells can become contaminated by coliforms due to run off, nearby farms and septic systems. In a CDC study (1999-2000), it was found that contaminated private well water caused 26% of the drinking water outbreaks that made people sick. Home owners who drink well water should have the wells tested annually. The lab provides a special sterile collecting bottle. Call your lab for the protocol. Often home inspectors end up testing well water. The test requires a 48 hour incubation period and results are delivered in three days. The results may indicate only the presence or absence of coliforms or a coliform count per milliliter of water, depending on the client's needs. If the test is positive the home inspector will direct the home owner to have the well "shocked". The shock

treatment is a chemical that effectively kills all the coliforms in the well.

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### Ensuring the Integrity of Your Dust Sample.

How many times have you received a package from your parcel carrier that was not in its ideal state? We have all fell victim to the crumpled, dented, and ripped package. As the parcel business grew the need for automation also grew. This means as your package is on its way to Assured Bio, it is placed on multiple conveyor belts and trucks that may drop, crush, and bend your sample. As a result of these harsh travel conditions, the opportunity for your dust collector to break open is increased.

Many problems can occur if a dust collector breaks open. The most obvious is loss of dust. Although only 5 mg of dust is needed by an analyst to run your ERMI sample, you do not want to cut the sampling capabilities in half by offering only a fraction of the dust you collected. Statistically you will have better results with a full volume of dust recovered from the dust collection device. For example, you would not survey a group of people, throw away half of the surveys, and expect to get a correct average of answers. This same idea holds true for your ERMI sample. You do not want to throw away half of your mold answers before you even get started.

Another problem can occur if dust is lost. What if your sample is a clearance test? There may be very little dust to begin with which means every bit of dust is precious. There have been times when an analyst was unable to process a sample due to there not being enough dust to run DNA analysis. This not only inconveniences you but your client because now another sample needs to be collected. Why can the dust from the packaging not be recovered and used for analysis? This is because there are many spores associated with packaging that may not have been part of your original sample. Unfortunately there is not a means of deciphering dust from your collection device and dust from your envelope or box. This is why a new sample would be in the best interest of your project.

What can you do to help prevent dust collectors from breaking open? Each dust collector comes with tight fitting caps for both ends of your collection device. Do not throw these away. Once the sample has been collected make sure both caps are tightly twisted onto the collection device. If you try to pull these caps straight off they should remain intact. If a cap is misplaced, make sure the cap that remains is placed at the top of your device where dust flows in and out of the filter. Finally, choose your packaging wisely. You do not want to place a dust collector in a large box where it

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has the capacity to bounce and move during shipping.

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## The Trace

When performing a direct examination on a swab, tape lift, or bulk material, analyst often have the ability to identify fungal structures instead of just spores alone. This additional information can give an inspector additional insight as to the severity of the fungal growth and contamination. In order to take advantage of this, one must be able to understand the implication of the various fungal structures.

So what are these additional structures that can be identified when collecting a sample as a tapelift, swab, or bulk?

**Hyphae** can be describe as a long filamentous cell of a fungus or individual filament. It is the main method of growth of a fungus and is the vesicle through which nutrients are absorbed. When found as a grouping of cells, it is also known as mycelium. When this structure is found present in an indoor air quality sample, it is usually considered as an indicator of active mold growth.

**Chlamydo spores** are thick walled asexual spores that are formed from a hyphal cell. They have the ability to act as a resting spore. Chlamydo spores are capable of remaining dormant for long periods of time under extreme conditions. The implication of their pres-

ence is the ability of the fungus to resume growth under its ideal environmental conditions.

A **Perithecia** is a ball shaped structure found in several genera of Ascomycetes. It acts as a sack containing multiple ascospores. The perithecia will release spores one by one upon their maturity. Their presence in a sample means that the fungal concentration can multiply rapidly.

**Sclerotia** is a hardened, dense mass of hyphae which is stored with reserved food material. It can become detached and remain dormant until growth conditions become favorable. The presence of sclerotia indicates that with favorable conditions, this fungus can begin to grow.

There are many other fungal structures that can be identified using direct examination techniques however this list of four are the most commonly identified structures. Please know that the lab will gladly report additional findings and be available to discuss them.

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## Announcements

AssuredBio has just purchased two brand new Roche LC480-2 Quantitative PCR Systems. They arrived this week and are up and running. We use the LC480 to analyze samples for MSQPCR and ERMI. Our old system was an LC480-1. Two systems will allow us to meet the needs of our clients as our ERMI/

MSQPCR assays volume continues to rapidly expand.



Two Roche LC480-2's

Also, the newsletter has been expanded to more than one page. I tried to squeeze down to one page, but we just have to much information to share with you.

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