

# Assured Biotechnology Corporation

November 2008

## Monthly Newsletter

Welcome to the 2nd installment of AssuredBio's monthly newsletter. Clients be sure to contact us to get your New UPS labels. DHL is no longer in service.

-Edward A. Sobek, Ph.D.  
President/Lab Director

[www.assuredbio.com](http://www.assuredbio.com)

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New Mail Carrier: UPS Call For new Labels (865) 813-1700

#### Laboratory Automation

When we first began running Mold-Specific Quantitative PCR samples approximately 18 months ago, we were performing all manipulations by hand. Just for perspective, each ERMI sample that we run requires 37 pipetting steps for DNA and 37 pipetting steps for the detection mixes. These are relatively small volumes that required the complete attention of the person loading the plate. Only 10 samples fit onto a single PCR plate that constitutes a full run, and loading that plate required two people for at least 2½ hours. This is a tremendous investment of time and energy by a laboratory.

The higher volume of samples and demand for rapid turn-around-times has necessitated changes in our laboratory procedures. We purchased a robot from a major manufacturer and began testing its capabilities. After changing its settings slightly, it was obvious that the robot could pipet much more quickly, accurately and precisely than we could. Reactions are much more consistent because of this. Plates of 10 samples can be loaded in under 60 minutes, and this increases the number of samples that can be processed each day without decreasing the quality of the finished product. Also, because of the manner in which the robot picks up liquid from the samples and dispenses it into the plates, there is less wasted DNA and detection mix in the process. The increase in throughput, decrease in time spent on each sample and decrease in reagent waste associated resulted in a process that was less expensive to the laboratory. In turn, these savings were passed on to our clients.

Although the robot is a complex piece of equipment, use of the robot is very simple. In only 10 minutes, all necessary reagents can be loaded



Automated MSQPCR Robot for DNA

onto the deck of the instrument. Tubes are loaded into racks (by hand) in a specific order, and a fresh plate is placed onto a specific location on the deck. The only burden on the user is to place the tubes in the correct order in the blocks because the robot has no way to distinguish them. These racks are solid metal blocks that are chilled between runs. Molecular biology reagents are commonly kept cold to prevent degradation, so these blocks extend the life of detection mixes. After initiating the run, the robot scans across the deck of the robot to ensure that disposable tips, tube racks and plates are in the proper places. Then, the robot begins a sequence of commands that pick up DNA/detection mixes from certain tubes and dispense them into certain areas of the plate. Calibration and maintenance of the robot is performed by the manufacturer, ensuring that the robot is performing correctly. In general, using the robot is a very hands-off process.

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### THE TRACE

#### Notes on Sample Volume

The trace of a spore trap captures a wide array of particulates (e.g. mold spores, pollen, insects, and dust). The proper identification and quantification of airborne particulates is dependent upon sample volume. The sample volume is determined by multiplying the sampling flow rate by the duration of the

sampling period. Expressed in liters; this value allows the laboratory analyst to calculate spores per cubic meter of air. A sample's volume can dictate the quality of lab results. This value impacts the limit of detection (or analytical sensitivity) and the specificity of the lab's identifications. The limit of detection is, in theory, the fewest number of airborne spores that must be present in an environment for them to appear in a collected sample. An increase in sample volume will decrease the limit of detection. This means as the sample volume goes up, fewer spores have to be present for detection.

On the surface, it may appear that by increasing collection time, laboratory counts become more reliable. However, that is not necessarily the case. Collecting too much sample (sample overloading) is problematic. When this occurs, the analyst loses the ability to look at the fine details in the trace. Spores may be misidentified or grouped in to broad classifications instead of identified as *Aspergillus* sp., *Trichoderma* sp., or *Scopulariopsis* sp. Smaller spores may also go undetected due to the overloading of non fungal particulates. These are not desirable results when determining the mold burden of a space.

What to do? Sample wisely and keep sample volume in mind. Always record the sample volume. Be mindful of the sampling environment. Reduce collection times in areas known to have high particulate such as wall cavities, ceiling, and air ducts. Finally, never collect for a period longer than that recommended by the manufacturer of the spore trap. By controlling the sample volume, you can control the quality of your lab results.

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#### Sci-Pubs: Rainfall and Autism

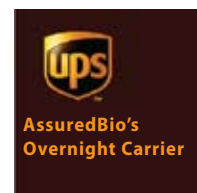
In the past 30 years, autism rates have increased from approximately one in 2,500 to one in 150 children. Some of the increase is likely due to more active monitoring and changes in diagnostic criteria.

A recent study in the scientific journal, *Archives of Pediatric Adolescent Medicine*, found a connection between precipitation and autism in California, Washington and Oregon (Waldman, Nicholson et al. 2008). Children living in counties with higher levels of annual precipitation appear more likely to have higher prevalence rates of autism, according to report. The results raise the possibility that an environmental trigger for autism may be associated with precipitation and may affect genetically vulnerable children.

After reading the article I immediately e-mailed Michael Waldman, the lead scientist on the study. I wanted to know if he had any information concerning water intrusion or flooding insurance claims. You probably know where I'm going with this. In the article they speculate that more rainfall results in children staying indoors and playing more video games, watching TV and perhaps increased exposure to household chemicals. However, not only was average rainfall associated with increased rates of autism, but the volume or inches of rain per rainfall event correlated to greater rates of autism. That means strong storms, heavy downpours, and flooding, correlate with increased rates of autism.

We all know what happens in storms and floods...water intrusion...then mold. Yes, the children are inside more when it rains, but TV and video games aren't the only exposure. They also risk greater exposure to a moldy environment and microbial toxins stemming from all the rain. Unfortunately, Dr. Waldman informed me that he did not have the data I sought and nor was there any data or studies that he knew of that examined the relationship between mold or microbial toxins and autism. Perhaps his publication may lead to future studies. Regardless, the relationship between autism and rainfall is intriguing.

Waldman, M., S. Nicholson, et al. (2008). "Autism Prevalence and Precipitation Rates in California, Oregon, and Washington Counties." *Arch Pediatr Adolesc Med* 162(11): 1026-1034.



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