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# Applying Real – Time Biological Testing to Monitor Drying Efforts of Category 1 Water Loss

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#### - Background -

The constant challenge for the indoor environmental specialist is determining the degree of microbial contamination that may be present on a building material that has been impacted by Category 1 water. The industry trend is to attempt to dry, restore and eliminate the potential for bacteria and mold growth on many finished building materials such as gypsum board, carpeting, carpet padding, etc. per the IICRC S500 Standard. Currently the techniques being used to measure and monitor the drying or restoration and remediation process is to collect environmental information (temperature, relative humidity, dew point, grains per pound, etc.) as well as moisture content measurements of the wet materials to evaluate the restoration process and determine the condition of the material over a period of drying time.

The primary objectives of any water loss event is to assess the extent of water damage present, remove the excess water from the impacted area of the building and perform drying of the restorable building materials and structure to eliminate the potential for growth of bacteria and fungi on the water impacted building materials. The generally benign nature of Category 1 water loss is often underestimated with respect to the potential for microbial amplification within the water impacted area of a building. The IICRC S500 defines Category 1 water "Clean Water" as, "Clean water originates from a source that does not pose substantial harm to humans. Examples of clean water sources may include, but are not necessarily limited to, broken supply water lines, tub or sink overflows with no contaminants, appliance malfunctions involving supply water lines, melting ice or snow, falling rainwater, broken toilet tanks and toilet bowls that do not contain contaminants or additives." The IICRC S500 Standard further states the water extraction and drying process should begin within 48 hours of the initial loss event to reduce the potential for microbial growth amplification.

A new technology being introduced into the restoration, remediation and environmental testing industries is the use of real-time microbial detection for testing the level of biological contamination of building material surfaces that may have been compromised due to water damage events, high humidity conditions, etc. where Category 1 water is the primary source of the water incursion. Real-time microbial detection is made possible by measuring the concentration of adenosine triphosphate (ATP) found within each living cell of the biological contamination that may be present, including fungi, bacteria and biofilms.

## - Study Objectives -

The intent of this study is to answer the following questions using real-time biological testing:

- 1. Can the finished building materials impacted by a Category 1 water loss be restored prior to microbial contamination growth if the loss is addressed within 24 hours of the event?
- 2. Should attempts to dry or restore the finished building materials be implemented after 24 hours of a water loss discovery?
- 3. If visual mold growth is not present on the finished building materials, should drying of these materials continue for longer than 72 hours of the event?
- 4. At what point in time during the restoration work should the decision be made to switch from restoration into remediation of the water impacted building materials?
- 5. Can biocides be appropriately introduced into the drying process of a Category 1 water loss that was discovered after 24 hours of the loss to allow for the drying process to continue beyond the 72 hour threshold?

- Real-Time Biological Testing – Background and Science Involved -In the 1980's, the detection of ATP bioluminescence was applied to the detection of microbes in foods as well as measuring the hygiene status of process surfaces. Adenosine triphosphate (ATP) is the chemical compound found in all organic matter including fungi, bacteria, somatic cells, plant cells, etc. ATP is known biologically as the "universal energy carrier" within living cells and is a significant biochemical component of the Krebs Cycle. In the ATP-luminometric test, the firefly enzyme (luciferase) in the presence of its substrate, luciferin, oxygen and magnesium ions catalyzes conversion of chemical energy of ATP into light through oxidation-reduction reaction (Figure 1).

Figure 1:

luciferase				
$ATP + D$ -luciferin $+ O_2$	$\rightarrow$	$AMP + oxyluciferin + CO_2 + phosphate$	+ Light	

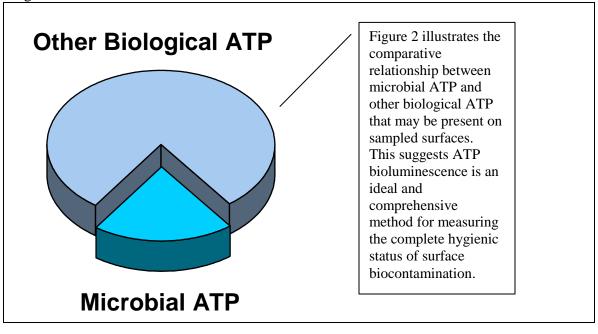
The quantity of light generated is directly proportional to the amount of biological ATP present, thus, the light units can be measured to estimate the biomass of cells in a sample. With state of the art equipment, and highly purified reagents, it is possible to detect trace amounts of microbial ATP corresponding to approximately  $10^2 - 10^3$  in concentration. Quantification of intracellular microbial ATP can be conveniently accomplished using rapid and simplified extraction and assay procedures. The light emitted by this process can be monitored by a variety of luminometers. Supplying companies provide customers with test kits with all necessary reagents. The reagents are injected into the instruments and readout is reported in relative light units (RLUs). By knowing the number of

microorganisms responsible for generating known RLUs, one can estimate the number of microorganisms in the collected sample. This correlation between surface cleanliness and microbial plate counts has made ATP bioluminescence a widely accepted method for the food, healthcare, industrial manufacturing and pharmaceutical industries.

This version of the ATP bioluminescence method based on detecting all ATP on a surface involves collecting samples by swabbing the surface. Reading of the bioluminometers may be assessed numerically or as "acceptable" or "unacceptable". The procedure can be easily performed by almost anyone, with little training, in less than one minute. Portable luminometer reading units test swabs with pre-packaged reagents. The user swabs the surface to be tested, activates the swab by placing it into the solution of reagents then inserts it into the chamber of the luminometer to obtain the measurement.

Additionally, significant interest has been generated in using ATP estimation not only for total viable cell counts but also non-culturable cell presence on a surface, which allows the user of ATP bioluminescence technology to evaluate the total bio-burden of the water impacted finished building materials and not just the culturable fungi or bacteria that may be present (Figure 2).

Figure 2:



## - Test Method Description -

The purpose of using ATP bioluminescence to evaluate the biological conditions of Category 1 water impacted building materials is to provide some numerical value of biological contamination that may be present as a result of the water loss. This information can be used to evaluate the initial biological condition of the finished building materials as well as allow for monitoring of the drying process of the water impacted building materials. The test method used to evaluate the potential for using ATP bioluminescence as a tool for determining the drying effectiveness for Category 1 water loss is as follows.

- 1. A small wall section was constructed with two by four inch studs and 5/8" painted, gypsum board fastened to each side of the framed wall section.
- 2. The small wall section was then lowered into a clean, plastic container filled with three to four inches of clean, Category 1 water (filled from a typical sink faucet). The water in the container was allowed to wick up the gypsum board wall section and saturate the materials.
- 3. Swab samples for ATP bioluminescence were collected over a four square inch area and averaged from four representative locations on the exterior surface of the gypsum board prior to being exposed to the "water loss" as well as at 12 hour intervals after exposure. The ATP testing was performed with the *Bio-reveal* system manufactured by Hygiena. The sample results were collected from the water impacted or wicked zone of the gypsum board (lower 12 inches on the wall section) logged and documented throughout the exposure process.
- 4. Samples were collected over a seven day period or until visual fungal growth was identified on the surface of the gypsum board.

### - Summary of Findings -

The results of the ATP testing of the Category 1 water impacted gypsum board wall material are outlined in Figure 3 below.

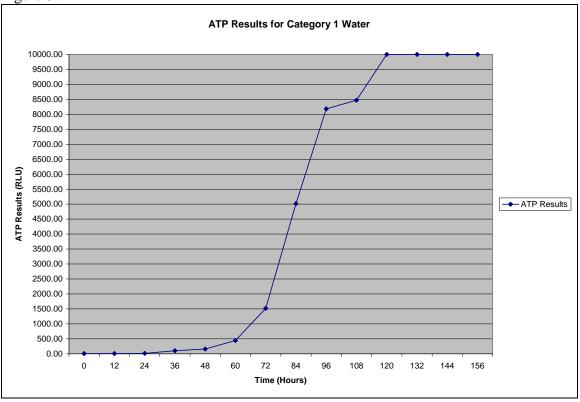


Figure 3:

#### - Conclusions -

Based on the results of the ATP bioluminescence testing of the Category 1 water impacted building materials the following conclusions can be made to answer the initial questions posed at the beginning of the study.

- The gypsum board material sampled at time interval 0 did not appear to have any significant background biological contamination present prior to the introduction of the water loss to the study.
- 1. Can the finished building materials impacted by a Category 1 water loss be restored prior to microbial contamination growth if the loss is addressed within 24 hours of the event?
  - Based on the study findings, the Category 1 water impacted building materials could be restored prior to microbial contamination when the loss is addressed within 24 hours of the event if adequate removal of the excess water is performed and specific drying of the impacted materials is implemented successfully between 36 and 48 hours.

- 2. Should attempts to dry or restore the finished building materials be implemented after 24 hours of a water loss event?
  - The results of the study findings suggest Category 1 water impacted building materials could be restored after 24 hours of the initial event if aggressive restoration and drying measures are implemented to reduce the time involved with water removal and drying. In this situation the restoration work related to drying would have to be completed within one day of on-site arrival to limit microbial growth amplification. If the company performing the restoration work is not well prepared or is delayed for whatever reason once arriving on-site then the decision to attempt to restore the finished building materials may be inappropriate and a wasted effort.
- 3. If visual mold growth is not present on the finished building materials, should drying of these materials continue for longer than 72 hours of the event?
  - Attempting to perform drying of the water impacted, finished building materials after 72 hours does not appear to be a worthwhile exercise even if visual mold growth is not yet present. The bioamplification on the surface of the gypsum board begins to accelerate significantly at this time threshold. Additionally, the use of drying equipment and methods such as fans and air moving equipment could lead to unnecessary distribution of biological growth throughout the impacted area and within the air of the impacted area.
- 4. At what point in time during the restoration work should the decision be made to switch from restoration into remediation of the water impacted building materials?
  - The ATP sample results suggest the appropriate decision to switch from restoration to remediation of the water impacted finished building materials is approximately 60 hours after the initial water loss event. After 60 hours and approaching 72 hours and beyond the bioburden of the impacted material is significant and restoration drying of the finished materials would likely not improve the biological status of those materials.
- 5. Can biocides be appropriately introduced into the drying process of a Category 1 water loss that was discovered after 24 hours of the event to allow for the drying process to continue beyond the 72 hour threshold?
  - ➤ The ATP sample results suggest intervention of a biocide that has the ability to destroy biological growth could and in some situations should be used to assist the restoration and drying efforts. The ideal time to introduce the biocide would be between 36 and 54 hours after the initial event. This will slow the microbial amplification on the impacted material to allow for additional time for further drying of the finished material. The application of a biocide after 60 or 72 hours may not have as an effective result if the bioamplification has become substantial and established.

Overall, the implementation of ATP testing to evaluate the initial condition of the building materials impacted by Category 1 water as well as monitoring the restoration drying efforts of impacted materials appears to be a good method in understanding the potential for bioamplification after a water loss event. The ability to measure the bioburden of the impacted materials along with moisture mapping allows the restoration work to proceed along a logical path to success. Furthermore, measurement of the bioburden on the impacted building material at various stages of the restoration process from initial response to final drying efforts will allow for appropriate decisions to be made to best serve the customer, insurance provider and restorer.

#### References

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