ATP BIOLUMINESCENCE

APPLICATIONS AND MEASUREMENTS OF BIO-CONTAMINATION IN THE RESTORATION INDUSTRY

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GOALS OF DISCUSSION

- What is ATP?
- Background on ATP Technology
- Science vs. Myths
- Recent Studies and Verification of Technology
- The Ideal Condition for Restoration
- ATP & Restoration Related Applications
- Advantages and Disadvantages or Limitations
- The Present and Future of ATP in Restoration

WHAT IS ATP?

- ATP is: Adenosine Triphosphate
- It's the energy molecule within living cells
- For ease of understanding ATP is the "contaminant" or the biological marker being collected and measured.
- The "contaminant" could be bacteria, fungi, somatic cells (tissue, blood, etc.), plant cells or other organic residues that are living.
- Literally found in mitochondria of cell
- Not viral however.

- ATP assay detection 1980's
- Primary application was to detect microbes and food residues for process and manufacturing purposes.
- Lumac was the initial pioneer of technology
- Purpose of testing was for surface hygiene monitoring within plant.
- Extracted enzyme was used for creating the biochemical reaction (luciferin).

- When ATP (contamination) is mixed with luciferin a bio-chemical reaction occurs
- This reaction is bioluminescence (light is emitted)
- The unique quality of this reaction is ATP and luciferin produce a linear amount of light.
- 1 femtomole of ATP will produce 1 Relative Light Unit (RLU). - "Fire Fly Reaction"
- This reaction is measured in a luminometer.

- The past 30 years have seen many improvements
- Stability of reagents are much better (1-2 days to upwards of a year)
- Some are liquid stable and provide efficient reactions.
- Luminometers are smaller, more sensitive and cost effective (bench top to hand held)
- Person running the test had to have skills similar to lab tech - now anyone can do it.

TYPICAL ATP SYSTEM



TYPICAL ATP SYSTEM



- When living organisms eat they are providing 'fuel' for their cells to function. This fuel, in a cellular form, is called ATP (found in Krebs Cycle)
- The presence of ATP indicates either:
- (1) life (anything from microbes to man)
- (2) nutrient source that can support life (food)

No ATP = lack of any life or food source

No ATP means the surface must be BIOLOGICALLY CLEAN

- ADENOSINE TRI-PHOSPHATE
- Fire Fly enzyme use ATP to produce LIGHT
- The enzyme used is LUCIFERIN LUCIFERASE



The reaction is called BIOLUMINESCENCE and is measured in RELATIVE LIGHT UNITS (RLU)

TECHNOLOGY AND BACKGROUND

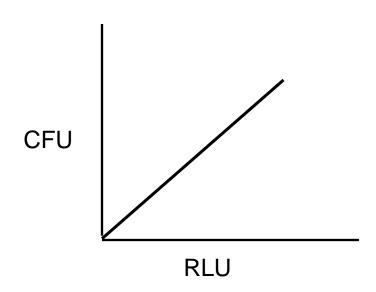
- Bioluminescence testing allows for the following:
 - Easy, rapid method of assessing the hygiene level (cleanliness) of a surface
 - Results available in under 1 minute allowing immediate corrective action (Real time)
 - Reduces need for culture testing (long turnaround time avoided and <10% cost)
 - Tests are easy to perform (no specific skills or formal training are required)

TECHNOLOGY AND BACKGROUND

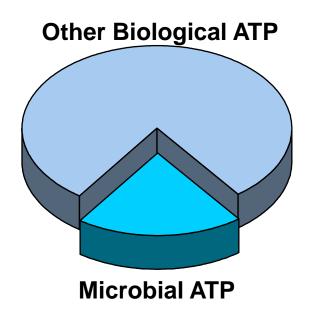
- Using (ATP bioluminescence) as a bio "Marker" or means of total bio-burden measurement (hyphae-fungi, bacteria, bio-films, etc.).
- Adenosine Tri-Phosphate (ATP) is present in <u>ALL living</u> organic material or cells.
- Converts biological contamination or presence into numerical values for interpretation.

TECHNOLOGY AND BACKGROUND

CFU / ATP correlation



With pure culture of microorganisms, without other residues, RLU correlate well with CFU (ie: bacteria)



With these typical proportions of ATP on a swab, RLU does not correlate with CFU but with "cleanliness" or potential for pathogens on unhygienic surfaces (typical environment)

SCIENCE VS. MYTHS WITH ATP

- Science: Measures the intercellular ATP of living cells (biocontamination)
- Myth: Detects dead cells (as cells die or become damaged the ATP molecule is altered or damaged and cannot be detected
- Science: Designed for surface or liquid testing
- Myth: Detects contamination in the air

SCIENCE VS. MYTHS WITH ATP

- Myth: Organic building materials (paper and wood) give false positives.
- Science: These materials do not have available ATP that is extractable from the material. The porosity of these materials typically traps organic residues, which can cause a positive presence. This is typically very low overall if the materials are clean

SCIENCE VS. MYTHS WITH ATP

- Myth: I applied a disinfectant and the level of ATP increased instead of decreasing. Stupid machine is not working!
- Science: The intercellular ATP was released onto the surface and into solution so the detection of "free ATP" is efficient. Give the chemical time to dwell, rinse or dry then collect samples a few hours later. ATP molecule deterioration will occur.

Food Industry:

- FDA and USDA ATP is named directly in regulatory documents as a valid method for monitoring hygiene of food contact surfaces
- FSIS Listeria Control Guideline (Sept 2012) -Outlined as a control point method for determining surface hygiene and safety monitoring for Listeria control in production plants.

• Homeland Security:

- US Army- ATP is studied as a rapid detection method for biological contamination due to biowarfare threats
- Conclusion: Proved as a valid method of detection for microbes and was effective in determining cleanup verification.

• Pharmaceutical Industry:

 FDA - ATP is approved as a valid testing method for determining raw and finished product contamination through "Process Analytical Technologies" (PAT).

- Restoration Related:
 - Wonder Makers and Esporta- ATP was used to test sewage related contamination on contents before and after cleaning.
 - Conclusion: Strong correlation was present between lab data and ATP results.
- "appeared that ATP monitoring is an effective tool in field verification of the effectiveness of sewage contamination removal in items laundered with the Esporta Wash System"

• Restoration Related:

- ISSA and CIRI- ATP was used to test cleaning effectiveness in schools (called the Clean Standard: K-12) - Draft published in July 2013 - also in JOEH in June 2013.
- Conclusion: The research validated ATP as a

"simple, rapid and affordable measure of the level of biologically sourced contamination on the interior surfaces of schools.

• Restoration Related:

- ISSA and CIRI- ATP was used to test cleaning effectiveness in schools (called the Clean Standard: K-12) Draft published in July 2013 also in JOEH in June 2013.
- Conclusion: Further the research concluded that ATP is an "...excellent candidate marker for the monitoring of biologically derived soiling/cleanliness.

- Restoration Related:
 - Conclusion:

Research has validated ATP meters as an effective tool that may be used to measure whether various surfaces in schools (i.e., student desks, cafeteria tables, restroom sinks and stall doors) have been effectively cleaned specifically as it pertains to biological contamination..

- Restoration Related:
 - Conclusion:

In addition, the research has produced reasonable ATP range values for three different ATP meters for each high touch point surface tested, and that these ranges "...may be used in a standardized and routine approach to the monitoring of cleaning effectiveness in school buildings based on detection and quantification of biologically derived soiling

SO...WHAT DOES THIS ALL MEAN FOR RESTORATION??

Water Loss, Mold Remediation & Trauma

 ATP appears to be an effective "tool" to have in the restoration tool box

 The applications for initial detection, progress monitoring and post cleaning verification appear to be a valid and verifiable method for bioburden testing

THE IDEAL CONDITION - SURFACE HYGIENE

<u>Hygiene</u>

- A condition promoting sanitary practices
- The science concerned with the prevention of illness and maintenance of health
- What it practically means...CLEANING

(Gross debris removal + detailed cleaning + application of disinfectant/sanitizer = hygienic surface)

THE IDEAL CONDITION - CONTINUED

 After cleaning, surfaces appear visibly clean but <u>organic residue</u> may still remain at measurable concentrations

 Organic residue = anything living or once living (fungi, bacteria, bio-film, somatic cells, etc.)





- Water Loss Applications Including sewage
 - Effective as a monitoring tool for cleaning progress and effectiveness.
 - Effective as a PRV test or pre-PRV test (ATP vs E. coli). **Remember...can't be used to measure specific organism types unless you use the appropriate test for detecting those organisms**.

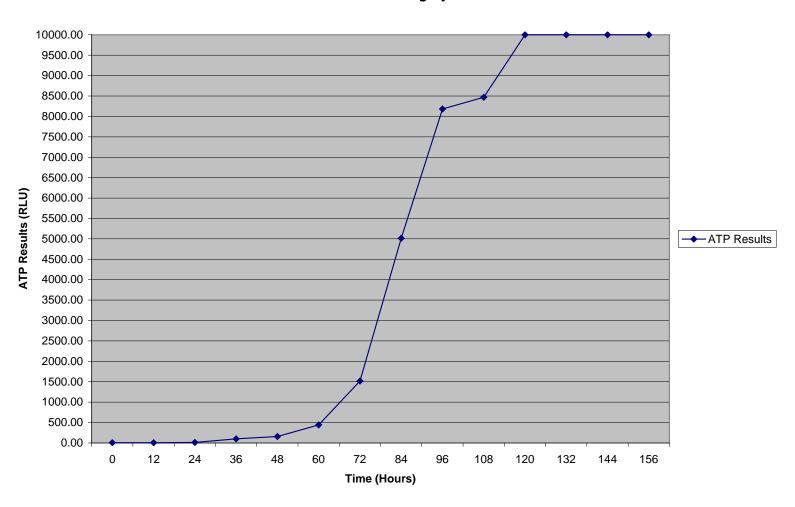
BIO-REVEAL INTERPRETATION

Surface Condition	Moisture Content (%)	ATP Results (RLU)	IICRC S520 Condition
No visible growth	<15	1 – 150 * *housekeeping dependent	1
No visible growth – w/in areas of visible growth	<15	100 – 500	2
Visible growth	>15	> 500	3

• Mold Remediation Applications:

- ATP bioluminescence can detect the presence and growth of mold (lots of ATP in hyphae) on building materials, contents, equipment, etc.
- Can provide contractor a defined scope as to what materials are compromised with biocontamination.
- Can assist in determining if the drying methods are effective on building materials.

ATP Results for Category 1 Water



BIO-REVEAL INTERPRETATION

Surface Condition	Moisture Content (%)	ATP Results (RLU)	IICRC S520 Condition
No visible growth	<15	1 – 150 * *housekeeping dependent	1
No visible growth – w/in areas of visible growth	<15	50 – 150	2
Visible growth	<15	> 150	3

BIO-REVEAL INTERPRETATION

Surface Condition	Moisture Content (%)	ATP Results (RLU)	IICRC S520 Condition
No visible growth	<15	1 – 150 * *housekeeping dependent	1
No visible growth – w/in areas of visible growth	<15	100 – 500	2
Visible growth	>15	> 500	3

• Carpet Cleaning Applications:

- ATP bioluminescence can detect the presence of bioburden and microbial growth of bacteria and mold on building materials, contents, equipment, etc.
- Can provide contractor a defined scope as to what materials are compromised with biocontamination.
- Can assist in determining if the drying methods are effective on building materials.

Sampled Surface Condition (Biological Contamination)	Bio-reveal Surface Sampling Result (RLU)*	Interpretation Result	Sampled Surface Condition (Biological Contamination)
Relatively Clean or Uncompromised Carpet Materials	<u><</u> 100	PASS (ACCEPTABLE)	Relatively Clean or Uncompromised Carpet Materials
Moderately Soiled or Potentially Contaminated Carpet Materials	≥ 101 and < 300	CAUTION (PASSING BUT DIRTY)	Moderately Soiled or Potentially Contaminated Carpet Materials
Heavily Soiled or Likely Compromised Carpet Materials	≥ 300	FAIL (NOT ACCEPTABLE)	Heavily Soiled or Likely Compromised Carpet Materials

• HVAC Cleaning Applications:

- ATP bioluminescence can detect the presence of bioburden and microbial growth of bacteria and mold in ductwork and on interior equipment, etc.
- Can provide contractor a defined scope as to what materials are compromised with biocontamination.

Sampled Surface Condition (Biological Contamination)	Bio-reveal Surface Sampling Result (RLU)*	Interpretation Result	Sampled Surface Condition (Biological Contamination)
Relatively Clean or	≤ 50	PASS (ACCEPTABLE)	Relatively Clean or
Uncompromised HVAC System Materials			Uncompromised HVAC System Materials
Moderately Soiled or Potentially Contaminated HVAC System Materials	<u>></u> 51 and < 150	CAUTION (PASSING BUT DIRTY)	Moderately Soiled or Potentially Contaminated HVAC System Materials
Heavily Soiled or Likely Compromised HVAC System Materials	<u>></u> 150	FAIL (NOT ACCEPTABLE)	Heavily Soiled or Likely Compromised HVAC System Materials

• Trauma Cleaning Applications:

- ATP bioluminescence can detect the presence of somatic cell and related bioburden on building materials, contents, equipment, etc.
- Can provide contractor a defined scope as to what materials are compromised with biocontamination.

Sampled Surface Condition (Biological Contamination)	Bio-reveal Surface Sampling Result (RLU)*	Interpretation Result	Sampled Surface Condition (Biological Contamination)
Ideally Clean or Decontaminated Surfaces / Materials	<u><</u> 15	PASS (ACCEPTABLE)	Ideally Clean or Decontaminated Surfaces / Materials
Moderately Contaminated Surfaces / Materials	≥ 16 and < 50	CAUTION (PASSING BUT RECLEANING MAY BE NECESSARY)	Moderately Contaminated Surfaces / Materials
Biohazard Contaminated Surfaces / Materials	<u>></u> 50	FAIL (NOT ACCEPTABLE)	Biohazard Contaminated Surfaces / Materials

THE PRESENT AND FUTURE OF ATP

- Presently, ATP testing is widely used in other industries and has been for decades.
- The luminometers are portable, low cost (\$1,000 \$3,000) and durable.
- The swab reagents are very stable, easy to use and cost effective (<\$3 per test)
- The use of ATP technology in other industries is paving the way for future uses and validation.

THE FUTURE....

- Real time and Rapid methods will gain more ground in the industry.
- Other similar types of technology exist presently and are being utilized in the IAQ market.
- Time is money and these technologies allow for savings of both.

THE END

Questions ??????