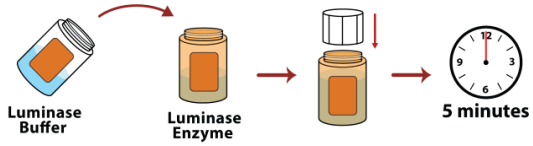
LUMINULTRA®
microbial monitoring

Test Kit Instructions
Deposit & Surface Analysis (DSA)

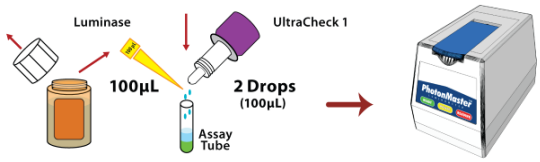
REHYDRATING LUMINASE

- Gently mix the buffer and **Luminase** enzyme.
- Wait 5 minutes for solution to dissolve.



1. ULTRACHECK CALIBRATION (RLU_{ATP1})

- Hold the UltraCheck1 bottle vertical, add 2 drops (100µL) of **UltraCheck1** to a 12x55mm test tube.
- Pipet 100µL of **Luminase** into the test tube**.**
- Swirl tube and take reading within 10 seconds.



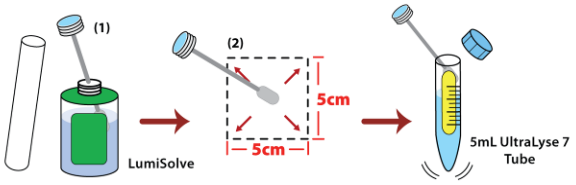
***** If RLU_{ATP1} ≤ 5,000 rehydrate a new bottle of Luminase.

2. SAMPLE PREPARATION

A. **Surface Swab** – A measured area of the surface is swabbed to collect biofilm. ATP is then extracted from the swab.
B. **Measured Deposit** – A deposit is collected and measured. ATP is extracted from the deposit.
C. **Biofilm Collector** – ATP is extracted directly off a biofilm collection device (e.g. corrosion coupon).

2.A SURFACE SWAB

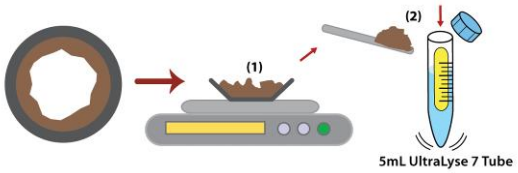
- Obtain a new Sterile Swab and wet with **LumiSolve.** Swab a surface area of approximately 5x5cm (2x2in).
- Insert swab in a **5mL UltraLyse 7 (Extraction) Tube**. Cap and mix the contents of the tube.



TIP: To increase analysis sensitivity, increase the swabbed surface area.

2.B MEASURED DEPOSIT

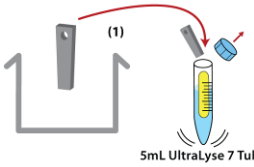
- Obtain a portion of the deposit and weigh 1g of sample.
- Add this to a **5mL UltraLyse 7 (Extraction) Tube**.
- Cap and mix the contents of the tube vigorously to disperse the deposit throughout the fluid.



TIP: A measured volume of deposit (e.g. 1mL) can also be used instead of a weighed amount.

2.C BIOFILM COLLECTOR

- Obtain a biofilm collection device from the process and shake gently to remove excess fluid.
- Note the area of all biofilm-containing surfaces on the device and place it into a **5mL UltraLyse 7 (Extraction) Tube**.
- Cap and mix the contents of the tube vigorously to disperse the deposit throughout the fluid.

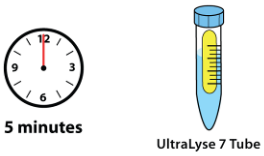


TIP: Attempt to test the biofilm collection device as quickly as possible following removal from process fluid.

3. TOTAL ATP (tATP) ANALYSIS

3.1 EXTRACTION

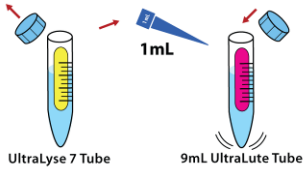
- Allow at least 5 minutes for ATP extraction in the **UltraLyse 7 (Extraction) Tube**.



TIP: When using the biofilm collector method, ensure the device is submerged in the UltraLyse 7 during incubation.

3.2 DILUTION

- Transfer 1mL from the **UltraLyse 7 (Extraction) Tube** to a **9mL UltraLute (Dilution) Tube**.
- Cap and invert three times to mix.



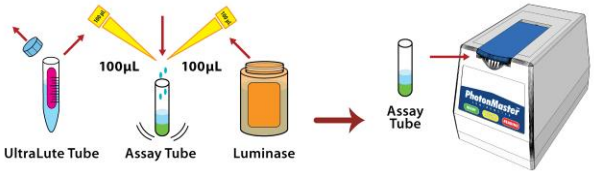
3.3 BACKGROUND MEASUREMENT

- Pipet 100µL of the **Luminase** into a new, clean 12x55mm test tube.
- Put the test tube with **Luminase** into your luminometer and take a reading.
- Record the reading as *Background RLU*. This reading should be below 20 RLU. This tube can then be used for your sample assay (Step 3.4).

TIP: If the results are above 20 RLU, this indicates that some external factors (light, contamination) may be influencing your results. LuminUltra offers cleaning kits for your luminometer. Please contact us to discuss

3.4 ASSAY

- Pipet 100µL of the **UltraLute (Dilution)** solution into a 12x55mm test tube with **Luminase** from Step 3.3.
- Swirl the tube and take reading within 10 seconds. Record the reading as RLU_{tATP}.



CALCULATIONS

The Total ATP (**tATP**) analysis measures all ATP within the deposit, including ATP from living cells as well as ATP released from dead cells.

A – Surface Swab (Default A_{Sample} = 25cm²):

$$tATP\ (pgATP/cm^2) = \frac{RLU_{tATP} - Background\ RLU}{RLU_{ATP1}} \times \frac{50{,}000\ (pgATP)}{A_{Sample}\ (cm^2)}$$

B – Measured Deposit (Default m_{Sample} = 1g):

$$tATP\ (pgATP/g) = \frac{RLU_{tATP} - Background\ RLU}{RLU_{ATP1}} \times \frac{50{,}000\ (pgATP)}{m_{Sample}\ (g)}$$

C – Biofilm Collector:

$$tATP\ (pgATP/cm^2) = \frac{RLU_{tATP} - Background\ RLU}{RLU_{ATP1}} \times \frac{50{,}000\ (pgATP)}{A_{Collector}\ (cm^2)}$$

TIP: You may also divide the result by the number of days the biofilm has had to evolve to obtain a growth rate.

Interpretation Guidelines

ATP-based measurements are extremely sensitive to changes in total microbial quantity. In general, processes will have the best microbial control when **tATP is minimized.**

It is recommend to compare surface/deposit results to bulk fluid results. Good control of biofilm is generally achieved when the biofilm/fluid ratio is <10x, and corrective action is required at levels of 100x or above:

Application	Good Control (pg tATP/cm^2 or g)	Preventive Action (pg tATP/cm^2 or g)	Corrective Action (pg tATP/cm^2 or g)
Potable & Sanitary Water	<10	10 to 1,000	>1,000
Raw, Cooling & Process Water (Oxidizing Biocide)	<100	100 to 10,000	>10,000
Cooling, Process, Bottom & Oilfield Water (Non-Oxidizing Biocide)	<1,000	1,000 to 100,000	>100,000
Bulk Fluid-to-Biofilm Ratio	<10x	10x to 100x	>100x
Biological Filter Media	Process Dependant		