

Intracellular ATP Kit HS

Measurement of ATP in cells and micro-organisms after elimination of extracellular ATP



- Fast assay: Results within minutes
- Highly sensitive: 10^{-17} mol ATP (~ 5 bacteria)
- Reliable: ATP standard in liquid form
- Flexible: Choice of μ -plate & cuvette methods

Leader in luminescent
ATP-assays

BioThema
LUMINESCENT ASSAYS

A graphic element of the BioThema logo, consisting of a stylized yellow and blue shape that resembles a test tube or a flame, positioned to the right of the text.

Intracellular ATP Kit HS

Intended use

The kit is intended for determination of intracellular ATP (adenosine triphosphate) in living cells. Extracellular ATP in the samples is enzymatically degraded. All living cells contain ATP where it plays the role of energy currency between different cellular processes. The intracellular concentration of ATP is carefully regulated to similar levels in all types of cells. ATP is therefore a good estimate of the total intracellular volume. Most bacterial cells contain approx. 2×10^{-18} mol ATP per cell, while most eukaryotic cells, as a result of their larger size, contain 10^{-15} mol ATP or more. The detection limit of the reagent is 10×10^{-18} mol ATP with a sensitive luminometer.

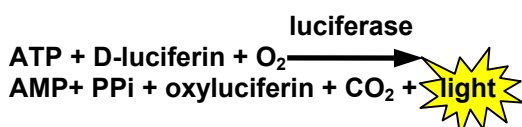
Applications

The sensitivity of this kit allows determination of low ATP levels in a variety of circumstances provided it is not important to degrade ATP in somatic cells:

1. Bacteriological control of liquids (e. g. drinking water, process water, beverages and beer)
2. Pharmaceuticals and cosmetics
3. Cell adhesion to surfaces
4. Quality assurance of laundry
5. Bacterial growth
6. Antibiotic effects on bacteria

Assay principles

Before the assay extracellular ATP is enzymatically degraded. Subsequently intracellular ATP is released from the cell using Extractant B/S also inactivating the ATP degrading enzyme system. Finally ATP is assayed using ATP reagent HS.



The intensity of the light is proportional to the amount of ATP and is measured in a luminometer. The light emission is measured before and after addition of a known amount of ATP standard. This makes it possible to calculate the amount of ATP in unknown samples expressing the result in pmol (10^{-12} mol).

Instruments

Any tube or microplate luminometer can be used. If a microplate luminometer is used it should have minimum two dispensers and should be able to mix the microplate.

Kit contents

The kit contains reagents for 150-375 assays (each vial of ATP Reagent HS is sufficient for 25 assays in a tube luminometer and 62.5 assays in a microplate luminometer).

1. ATP Reagent HS (6 vials). Lyophilised reagent containing luciferase and luciferin. The luciferase activity in the reconstituted reagent consumes ATP at a rate of approx. 6%/min
2. Diluent B 10 mL (6 vials). Buffer used to reconstitute ATP Reagent HS
3. Extractant B/S 10 mL (1 vial)
4. ATP standard 5 mL (1 vial; 10^{-7} mol/L ATP)
5. ATP Eliminating Reagent (1 vial)
6. Diluent C 10 mL (1 vial). Used to reconstitute ATP Eliminating Reagent

Assay procedure

- 1 Mix sample and ATP Eliminating Reagent and incubate 10 min at room temperature
- 2 Add Extractant B/S
- 3 Add ATP Reagent HS and measure light I_{smp}
- 4 Add 10 μL 100 nmol/L ATP Standard, i.e. 1 pmol ATP, and measure light emission, $I_{\text{smp+std}}$

Calculations:

Calculate amount of ATP (pmol) in the sample by the equation:

$$\text{ATP}_{\text{smp}} = I_{\text{smp}} / (I_{\text{smp+std}} - I_{\text{smp}})$$

Product characteristics

Detection limit: 10^{-17} mol ATP

ATP consumption (decay rate of light): 6 %/min

No. of determinations (tubes): 150

No. of determinations (microplate): 375

Ordering info

Article No: 266-111

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