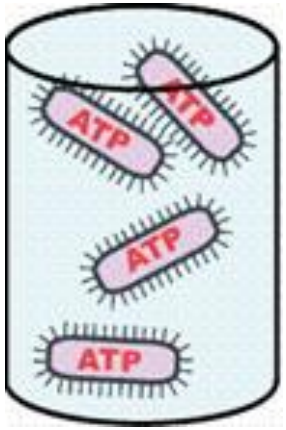
Rapid Microbial Test using ATP assay

Advantage : Shorten testing time → Ship the products quickly

Pre-cultivation



If there is some level of Microorganisms in the product, the pre-cultivation is not needed

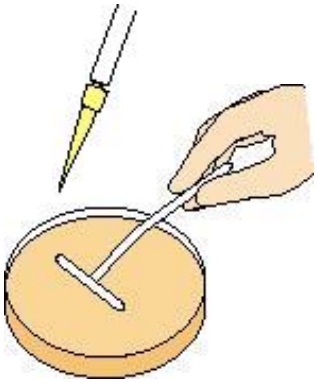
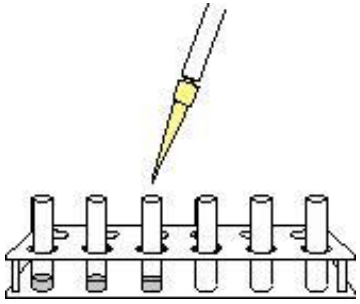


Plate Method

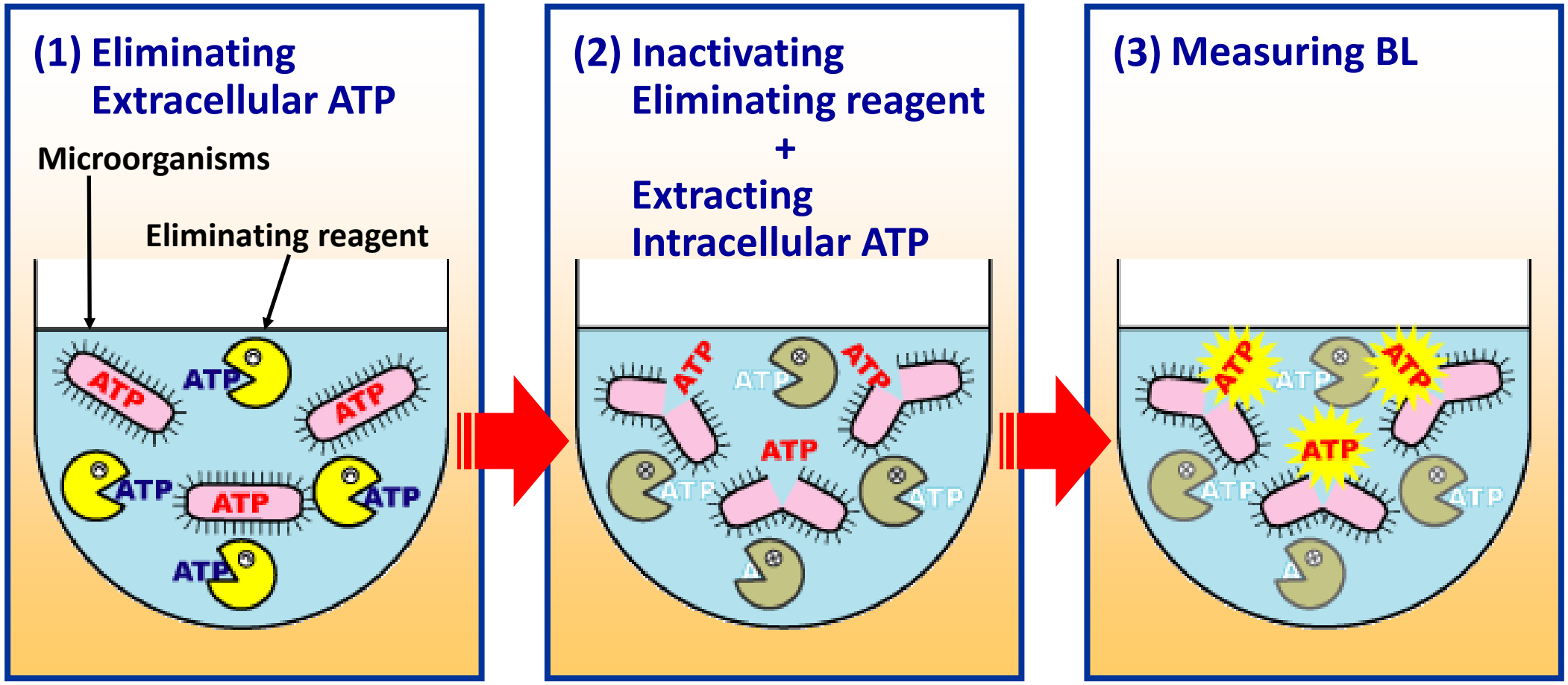
2-3 days



ATP Method

30 min.

Principle of Microbial Biomass Assay



ATP from only microorganisms is detected after ATP in sample is degraded

Comparison Between CheckLite Series

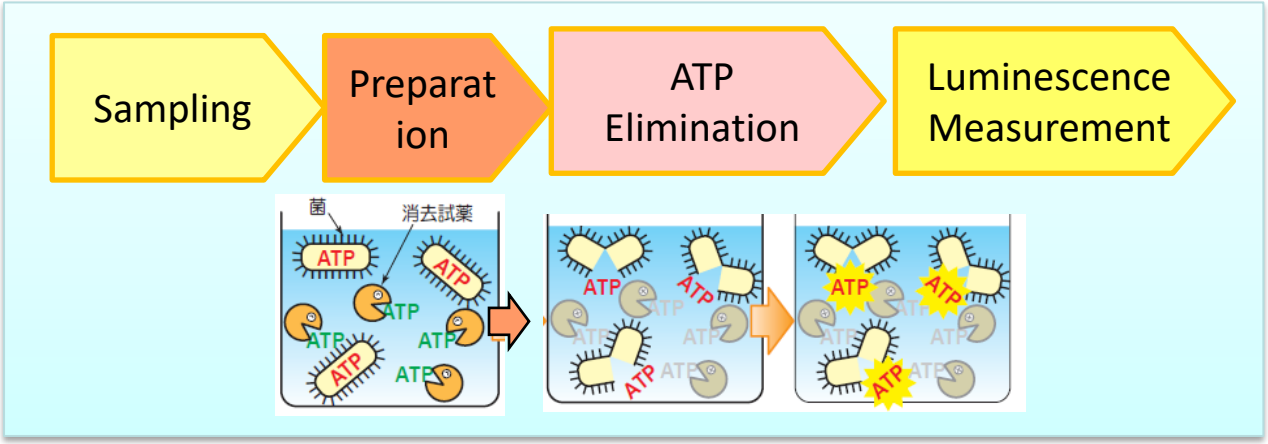
	Assay/kit	Luminescence Reagent	Luminescence Reagent (HS)	ATP Releasing Reagent	ATP Eliminating Reagent	Sample Dilution Buffer	Sample Treatment Buffer
CheckLite 250	250	O					
CheckLite 250 Plus	250	O		O			
CheckLite HS Set	**100**		O	O	O		
CheckLite AT100	**100**		O	O	O	O	O

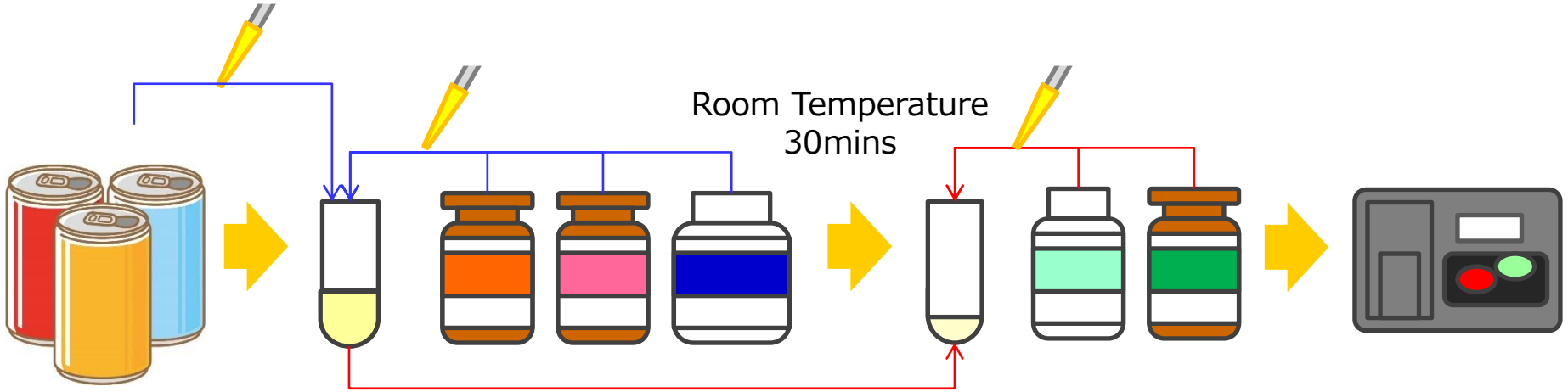
How to do validation

Step	To Do	Check Point
1. Check Background RLU of the Product	Check RLU of product (no dilution, 10 times dilution, etc) of the product	Whether Backgroud RLU is too high or not Inhibition Effect
2. Check necessity of Treatment Buffer	Check RLU of products with or without Treatment Buffer	Choose condition which shows lower RLU
3. Target microbes spike test	Examples of target microbes; • Bacillus subtilis • Staphylococcus aureus • Escherichia coli • Pseudomonas aeruginosa • Lactobacillus plantarum Spike 10 - 100 CFU/ml Check necessary pre-cultivation time 35ºC, 24h, 44h, 72h	Check bacteria count (CFU） and bioluminescence (RLU) Necessary pre-cultivation time. Check RLU per 1 CFU
4. Set Standard RLU value	Set standard value from the background RLU from step 1 and result from microbes spike test of step 3	Normally standard RLU value should be 3-10 times higher value than background RLU

Test Procedure - CheckLite AT-100 -

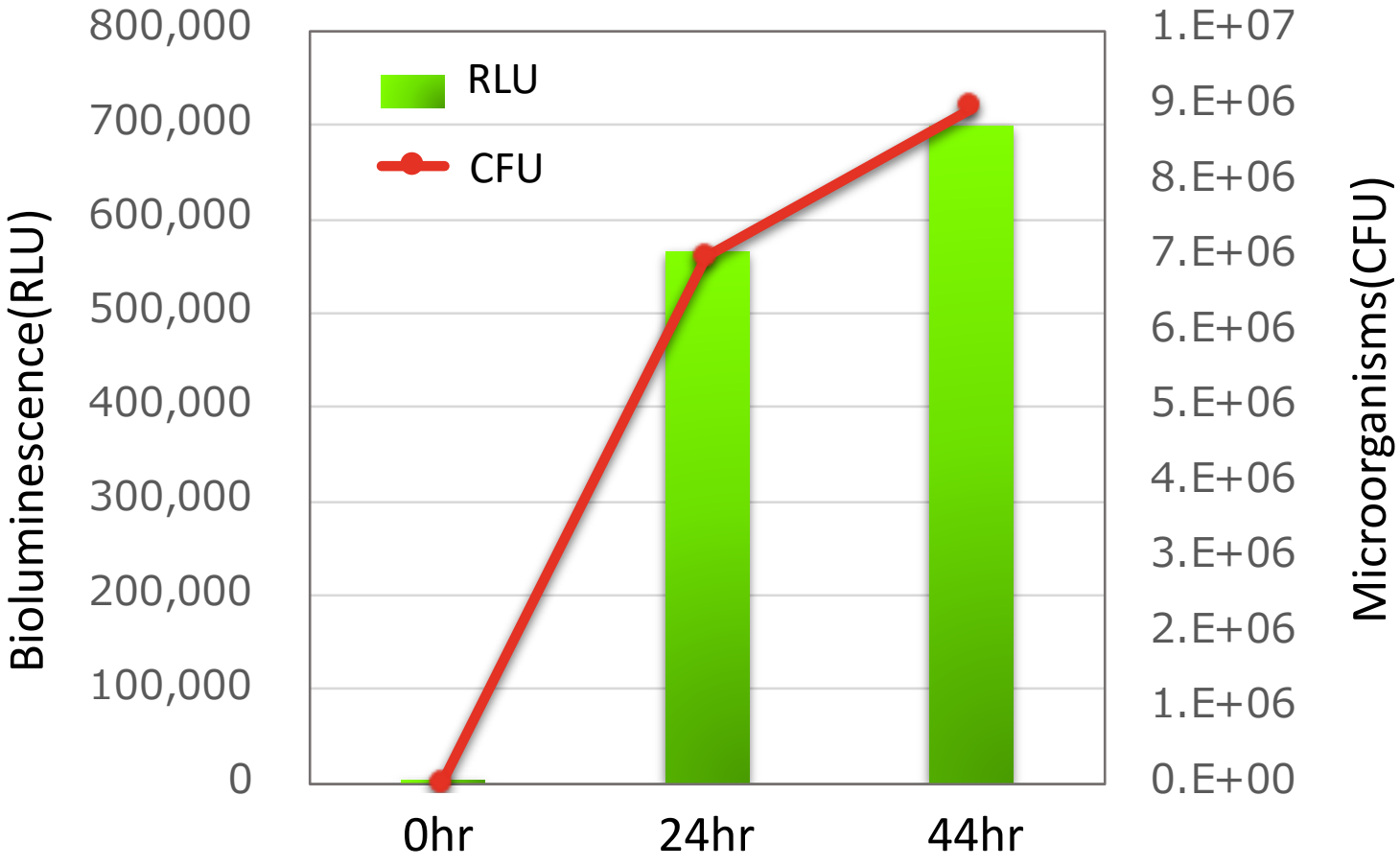
100µL Sample
100µL Treatment Buffer
700µL Dilution Buffer
100µL ATP Eliminating Reagent
↓Room Temperature 30mins
100µL Sample solution
100µL ATP Releasing Reagent
100µL Luminescence Reagent (HS)
↓
Measure RLU using Lumitester C-110





Pre-incubation | Sampling | Add Treatment buffer Dilution Buffer Eliminating Reagent | Mix sample solution ATP Releasing Reagent Luminescence Reagent | Measurement

Application Examples - Soymilk -



Strain: *Bacillus subtilis* (spike 20 CFU)

After 24hrs pre-incubation, *Bacillus subtilis* is detectable in Soymilk by CheckLite