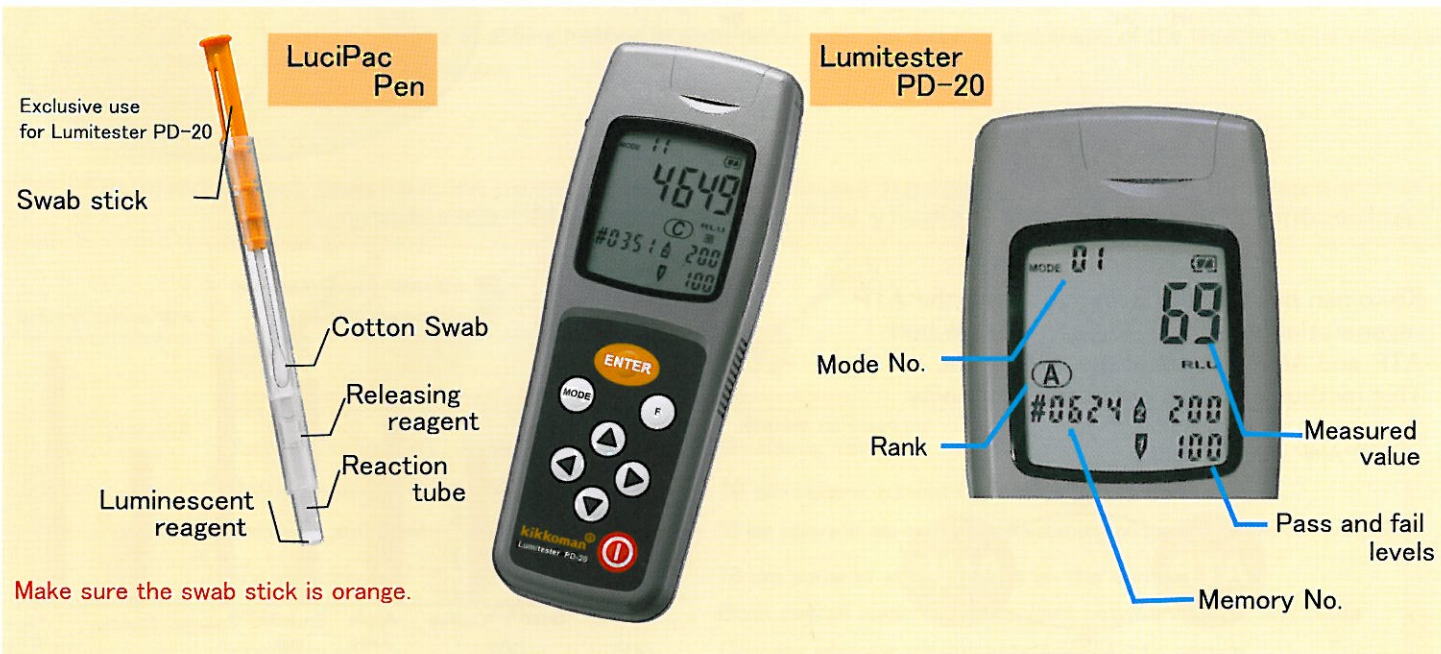


ATP+AMP surface hygiene monitoring using Lumitester PD-20 & LuciPac Pen

A majority of food poisoning incidents are caused by cross-contamination due to ineffective or improper cleaning. ATP+AMP hygiene monitoring is a convenient test method for quickly measuring the degree of cleanliness on site. This provides an excellent tool for hygiene training and cleanliness control to prevent food poisoning.

Reagents and instrument required

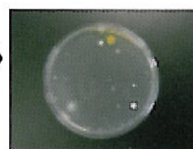


Features of ATP+AMP surface hygiene monitoring

1. Rapid

The results are available immediately!

Conventional Method
(culture method)



Time required
1 to 2 days

ATP+AMP method



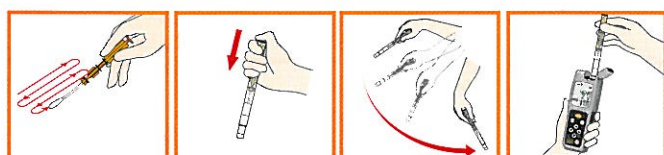
Time required
30 seconds

(including measuring time of 10 seconds)

Occurrence of food poisoning arising from short cleaning in food manufacturing site can be prevented.

2. Simple

3. Numerical



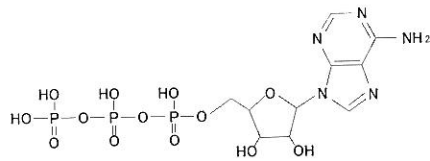
Swab. Push. Shake well. Measure with PD-20 for 10 seconds.

- Results are displayed numerically as RLU (Relative Light Unit).
- Collected data can be transferred to PC easily and used to analyse from various aspects.

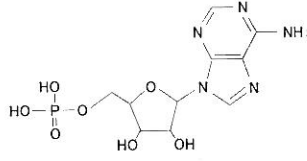
What is ATP? What is AMP?

ATP (adenosine triphosphate) is the primary molecule involved in metabolism in all living organisms. AMP (adenosine monophosphate) is derived from ATP during the processing, such as heat treatment and fermentation.

ATP (adenosine **tr**iphosphate)

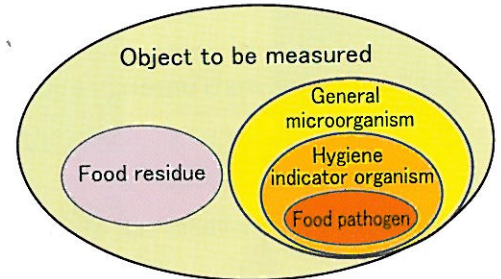


AMP (adenosine **mon**ophosphate)



Object to be measured

In ATP+AMP hygiene monitoring, the degree of contaminant is measured using as total ATP+AMP, which microorganisms and food residues have. ATP+AMP is present in microorganisms, food residues, etc. and as such is an excellent indicator of biological contamination.

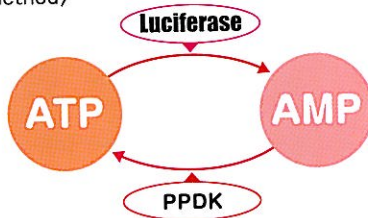


Achievement of super-high sensitivity with both ATP and AMP detection

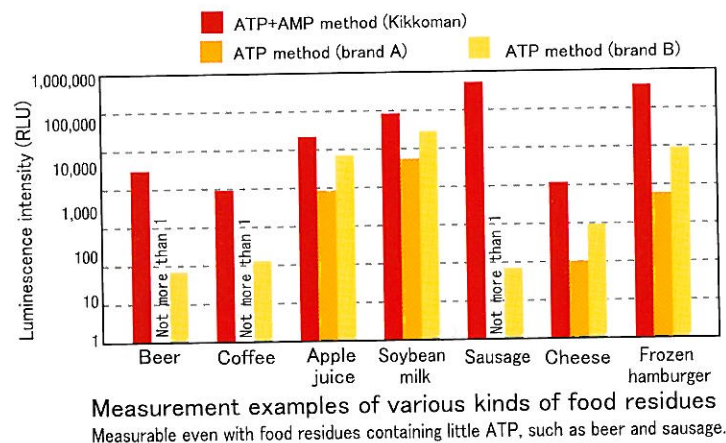
Kikkoman has created a method using the ATP regeneration enzyme PPDk to measure both ATP and AMP as part of the ATP cycle. This method provides enhanced sensitivity.

(US Patent No. 5891659).

<ATP+AMP method>

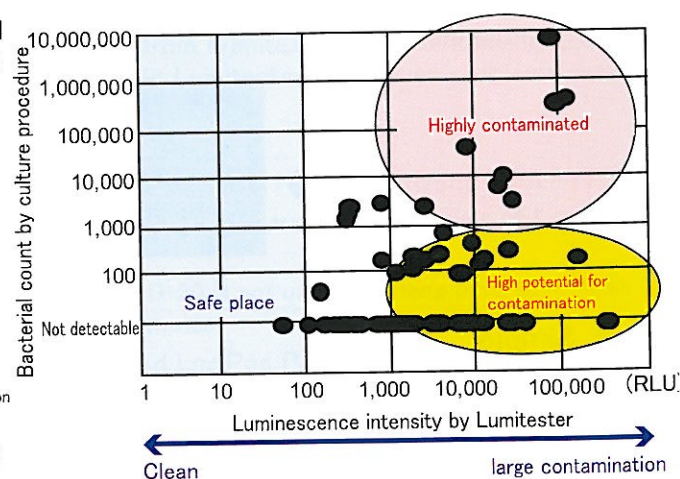
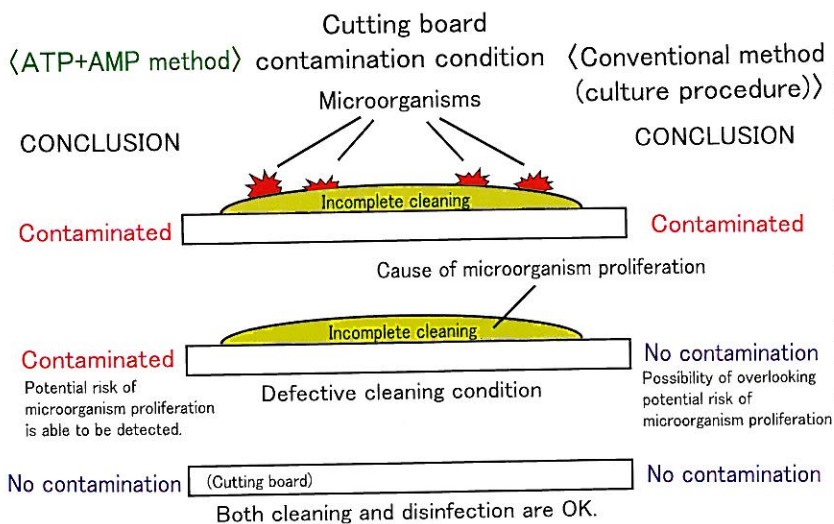


* PPDk...Pyruvate orthophosphate dikinase



What is the clean state?

That is in the state where the microorganisms and food residues are sufficiently clean. Let's achieve the clean state by ATP+AMP method.



When food residue is present, there is a possibility for microorganisms to proliferate quickly. The conventional culture procedure can detect microorganisms but the ATP+AMP method also detects food residues. Consequently, ineffective cleaning can be accurately determined.

Operation example of ATP+AMP hygiene monitoring

Establish test locations

Test locations should be established at the following points:

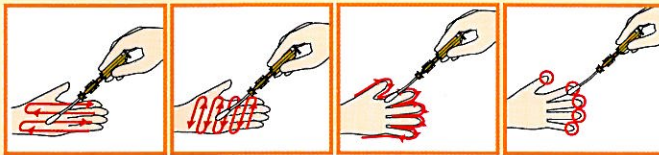
- Areas difficult to wash and easy for contaminants to remain.
- Areas where not only cleaning but also disinfection and sterilization are conducted.
- Areas in contact with ready to eat foods.
- Areas at risk of cross-contamination, such as hands and fingers of employees. etc.

Establish control targets

- Set the control target to 200 RLU for flat and smooth surfaces (metal, glass, etc.) and 500 RLU for things with surface irregularities and susceptible to scratches (plastic products, etc.).**
- These recommended values are not always applicable to any locations. The goal is to set reasonable targets that can be met with rigorous testing and proper cleaning.
 - Decide a swabbing method in accordance with the material and shape of the location to be examined and implement.

Example

〈Hands and fingers〉 Swab every direction of the palm, between fingers, fingertips, etc. The pass and fail levels are 1500 RLU and 3000 RLU, respectively.



〈Kitchen〉

| Portions for the test | Pass (RLU) | Fail (RLU) | Swabbing method |
|-----------------------|------------|------------|---|
| Cutting board | 500 | 1000 | 10 cm square around the center |
| Colander and bowl | 200 | 400 | 10 cm square of the center bottom portion and top end portion of the inside |
| Kitchen counter | 200 | 400 | 10 cm squares at five points on the surface |
| Knife | 200 | 400 | Both overall blade surfaces, joint between handle and blade |
| Stainless vat | 200 | 400 | Corners where contaminants are likely to remain. |
| Round pot | 200 | 400 | Three inside areas (bottom, middle stand, upper stand) |
| Refrigerator (handle) | 200 | 400 | Inside and outside of the overall handle |
| Refrigerator (inside) | 500 | 1000 | All directions of 10 cm square at the shelf center |

〈Manufacturing line〉 Valve portions and joints where contaminants are likely to remain.

〈Environmental inspection〉 High-frequency contact locations such as telephone sets, door knobs, keyboards and mice of personal computers, etc.

Establish analysis schedule

Conduct the test after cleaning and before disinfection and sterilization.

(Conducting the test while foods are being handled will not result in correct conclusion.)

Establish operational guidelines

The table below provides an example of operational guidelines for hygiene control at each location.

| Tested places | Pass/Fail levels (RLU) | | First measurement | Improvement measures | Second measurement |
|---------------------|------------------------|-------|-------------------|----------------------|--------------------|
| | Pass | Fail | | | |
| Hands and fingers | 1,500 | 3,000 | 2,412 B (Caution) | Re-cleaning | 1,323 A |
| Cutting board | 500 | 1,000 | 760 B (Caution) | Re-cleaning | 349 A |
| Bowl | 200 | 400 | 174 A (Passed) | | |
| Kitchen counter | 200 | 400 | 130 A (Passed) | | |
| Vat | 200 | 400 | 44 A (Passed) | | |
| Refrigerator handle | 200 | 400 | 820 C (Failed) | Re-cleaning | 101 A |

Setting of pass/fail criteria

Not more than pass level: A (passed)

More than fail level: C (failed)

Between pass and fail levels: B (caution)

Procedures for swab test

〈Preparation〉


LuciPac Pen

Remove LuciPac Pen from a refrigerator (2–8°C) 20 minutes before starting the measurement, and allow it to reach room temperature.

***Measuring with the LuciPac Pen while it is still cold does not produce correct measurement results.**

Lumitester PD-20



Press “POWER ” key to turn ON. After counting down 8 seconds, Lumitester PD-20 is ready to measure.

Use two AA alkaline batteries or two AA nickel metal-hydride batteries. When battery indicator displayed exhaustion sign, turn off the power and replace batteries.

〈Swabbing〉

1. Pull out a cotton swab stick (orange) and moisten a swab with tap water.

In the event that the surface to be swabbed is wet with water, the swab does not need to be pre-moistened.

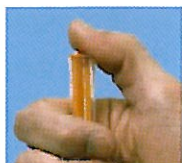


2. Swab the object to be tested.
See page 3 for the swabbing method.



***In the event that alcohol or other disinfectants remain on the surface to be swabbed, results may not be accurate. It is recommended to carry out the test after washing and before using disinfectants. If the test is conducted after using disinfectants, rinse the surface with water or wipe with paper towel, and conduct the test.**

3. Put the swab stick back into the main body and push it through all the way.

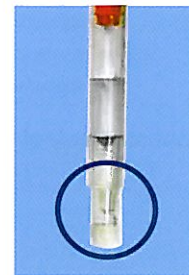


When pushing it to the main body, take care not to get your finger caught in.

Pressing the bottom by hand or against a table helps you push it in more easily.

4. Shake off LuciPac Pen to drop the releasing reagent to the bottom of the reaction tube and dissolve the luminescent reagent.

***Make sure all liquid in the capsule falls into the reaction tube.**



〈Measurement〉

5. Insert LuciPac Pen to the measurement chamber of Lumitester PD-20.

6. Press the “ENTER ” key of Lumitester PD-20 to start measurement. In 10 seconds, the measurement result is displayed.




***Use the accessory stand if possible. If the accessory stand is not used, do not incline the device more than 60 degrees.**



7. Upon completion of measurement, be sure to remove LuciPac Pen from Lumitester PD-20. Leaving LuciPac Pen inserted in Lumitester may cause problems resulting from liquid leakage, etc.

〈After measurement〉

- Press “POWER ” key to turn off the power of Lumitester PD-20.

If Lumitester PD-20 is not used for a long time, save data and remove batteries.

- Dispose of used LuciPac Pen in conformity to regulations of local government.

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