



Surface Sampling

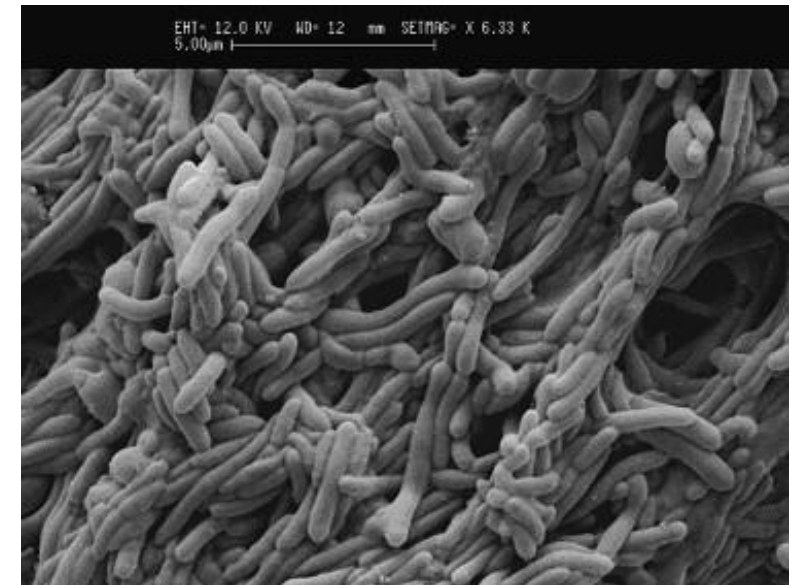
Simple Methods to Check Cleaning and Disinfection

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Microbes in Surfaces Often Hide in *Biofilms*

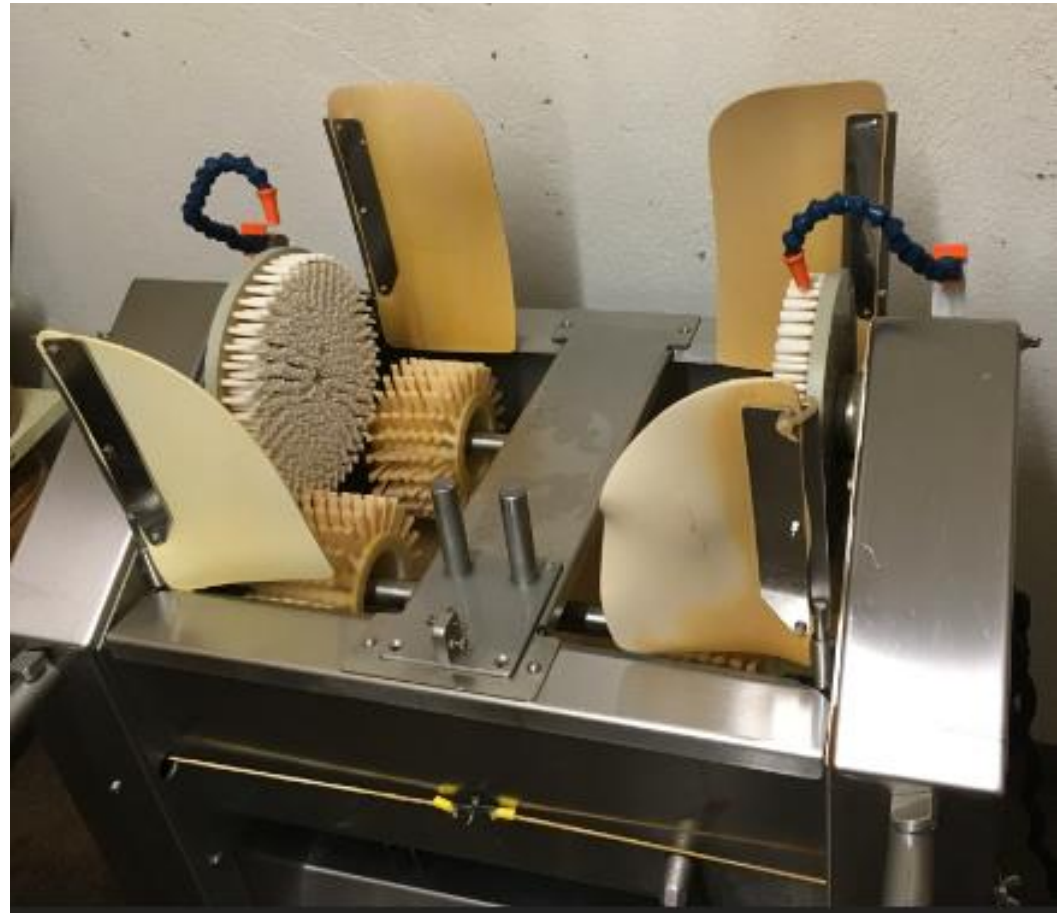
- Microbes can adhere to surfaces and *form biofilms*
 - Biofilms may be source of contamination and pose a hazard to health
 - Many *Listeria monocytogenes* and *Bacillus cereus* strains are known to have a strong adhesion ability to surfaces
 - Biofilms are formed to all wet surfaces with traces of nutrients
 - Biofilms protect microbes from cleaning and disinfection treatments
- *Biofilms are often difficult to eradicate*



Kuva : Scanning –electron microscope (SEM) image on biofilm formed by *Bacillus cereus* –(scale 5 µm). Source: Simões, M., Simoes, L. C., & Vieira, M. J. (2010). [A review of current and emergent biofilm control strategies](#). *LWT-Food Science and Technology*, 43(4), 573-583.



Biofilm is Easily Formed on Moist Surfaces and Splash Areas





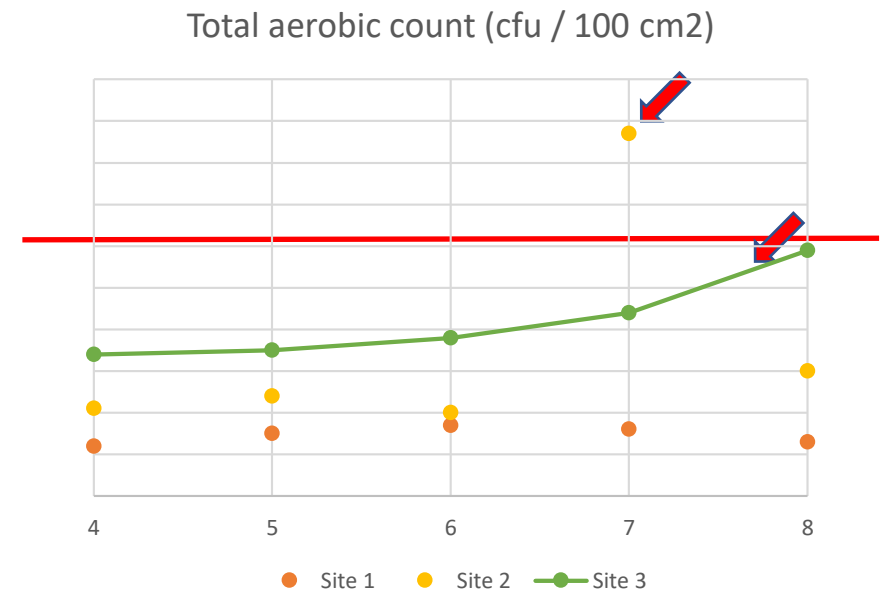
Surface Sampling is Obligatory if a Product poses a risk of Growth of *Listeria monocytogenes*

- The aim of these compulsory samples is **to detect the eventual presence of *L. monocytogenes*** in the processing areas and equipment (Reg. No 2073/2005, art.5.2)
- These samples shall be taken **during production**



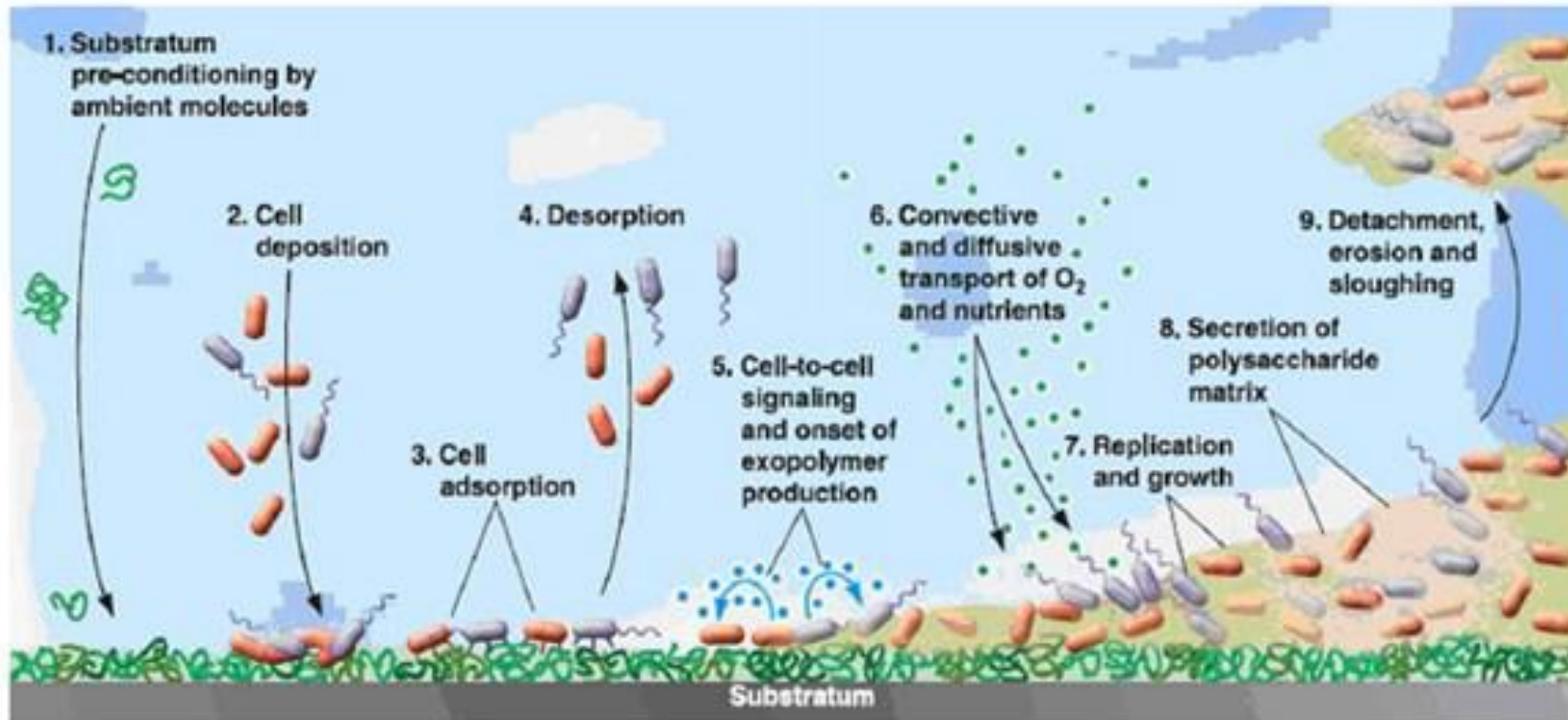
Surface Sampling for Hygiene Indicator Organisms

- **Recommended** and **useful** to evaluate cleaning and disinfection practices
 - monitor **trends in hygiene indicator microbes** at different sites in the dairy





Formation and Detachment of Biofilms on Processing Surfaces



Source: Simões, M., Simoes, L. C., & Vieira, M. J. (2010). [A review of current and emergent biofilm control strategies](#). *LWT-Food Science and Technology*, 43(4), 573-583.

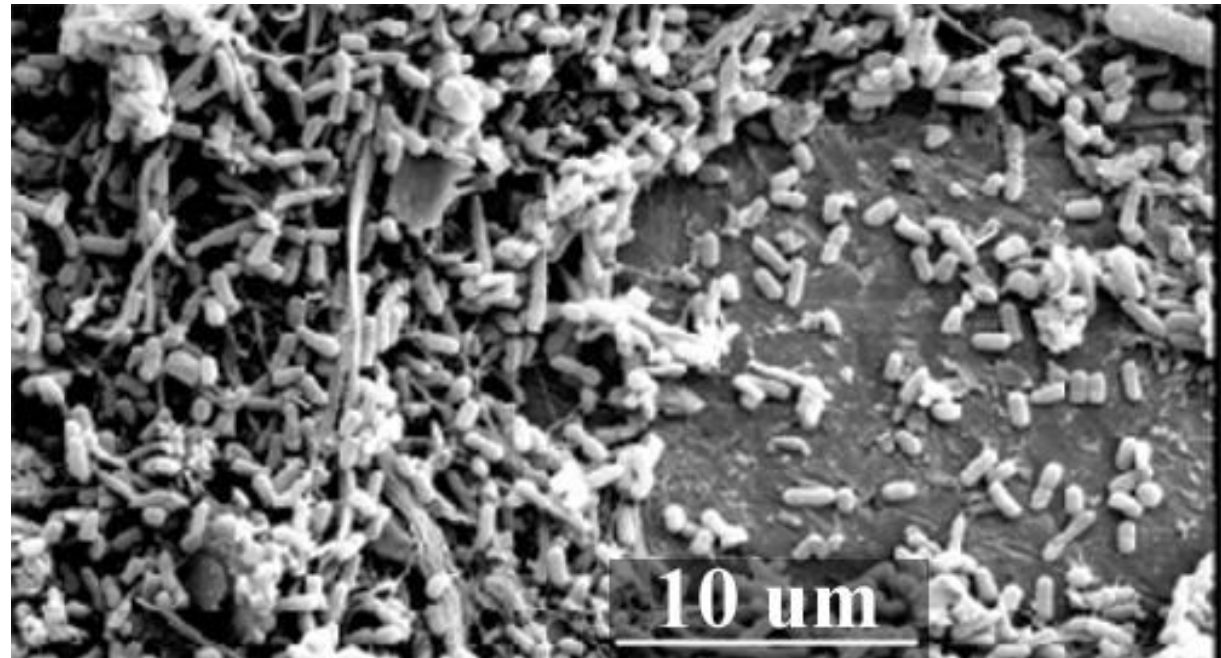
[Biofilm Formation Animation](#) (McGraw-Hill Animations 2017)

[A Review](#): Chmielewski, R. A. N., & Frank, J. F. (2003). Biofilm formation and control in food processing facilities. *Comprehensive reviews in food science and food safety*, 2(1), 22-32.



Importance of Quality of the Surface Material on Hygiene

- Stainless steel is available in various grades and finishes
→ various physico-chemical properties



→ This has an effect on bacterial adhesion

See examples and scanning electron microscope images: Jullien, C., Bénézech, T., Carpentier, B., Lebret, V., & Faille, C. (2003). Identification of surface characteristics relevant to the hygienic status of stainless steel for the food industry. *Journal of Food Engineering*, 56(1), 77-87. Retrieved from [https://doi.org/10.1016/S0260-8774\(02\)00150-4](https://doi.org/10.1016/S0260-8774(02)00150-4)



Where, When and How to Take Environmental Samples?

- As described in the the sampling plan of the premise
- Depending on the aim of the sampling, samples shall be taken:
 - a) From surfaces after proper disinfection. If surfaces or equipment look dirty → clean them again!
 - b) While being used or immediately before it (to detect *Listeria monocytogenes*)
 - c) From hands of workers (to check handwashing)



Food contact surfaces



Non-food contact surfaces



Examples of Surface Sampling and Simple Hygienic Analysis Methods



Surface swabbing
(see videos [QuickMedical](#) and [Quantiswab](#))

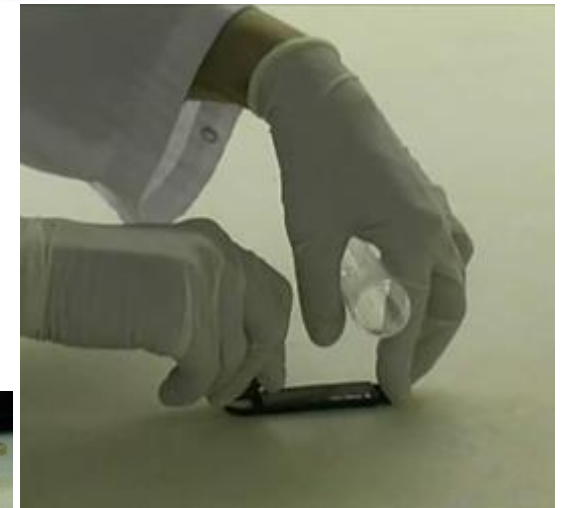
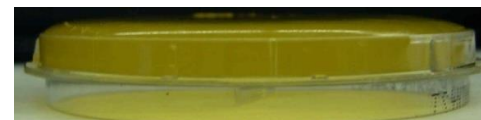


Petrifilm™ (3M) and related products

ATP



Dip slides and Contact plates





Many Commercial Products are Available for Surface Sampling





Contact Slides

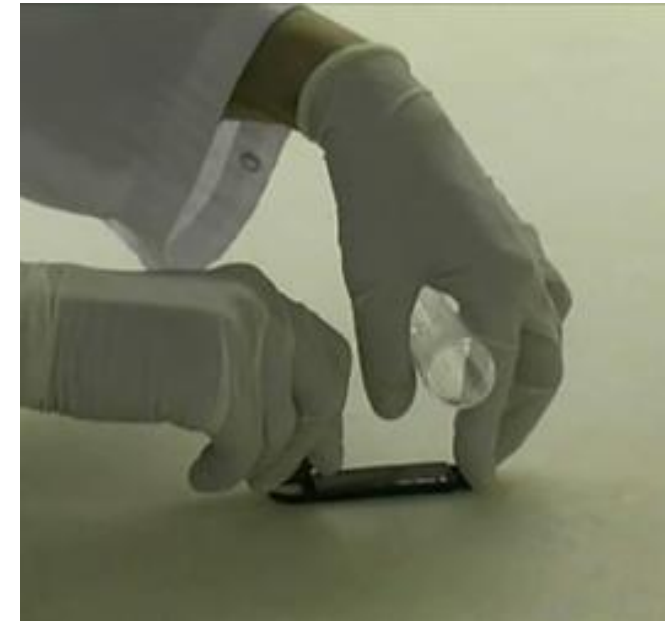
Growth medium for microbes on both side of the slide; for example for total aerobic bacteria, enterobacteria, coliforms, yeasts and moulds etc.

Sampling → Incubation → Interpretation of the results

An example of an evaluation table ([Microcount®](#))

Evaluation table

| mikrocount® | | TPC | | | |
|-------------------------|------------------------|------------------------|------------------------|------------------------|---------------------------|
| | | Total plate count | | | |
| 10 ² CFU/ml | 10 ³ CFU/ml | 10 ⁴ CFU/ml | 10 ⁵ CFU/ml | 10 ⁶ CFU/ml | 10 ⁷ CFU/ml |
| | | | | | |
| < 1 CFU/cm ² | 1 CFU/cm ² | 5 CFU/cm ² | 45 CFU/cm ² | 80 CFU/cm ² | > 100 CFU/cm ² |

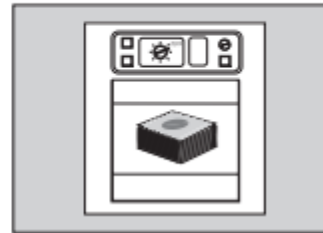
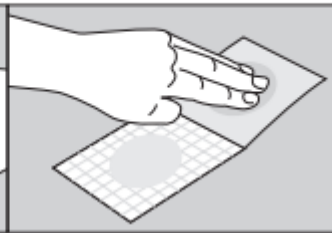
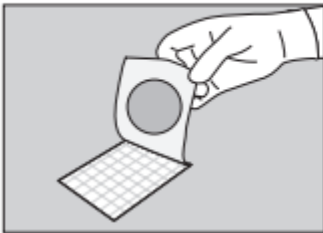


Several providers and products (Envirocheck®, Hygicult®, Hycheck®, Easicult®, Microcount® ...)



Petrifilms™ and Related Products

3M™ Petrifilm™ Plate Direct Contact Method



1 Using a hydrated 3M Petrifilm Plate, carefully lift top film. Avoid touching circular growth area. Gel will adhere to top film.

2 Allow the circular gel portion of the top film to contact the surface being tested. Gently rub fingers parallel to the surface over the outer film side of the gelled area to ensure good contact with surface. Rejoin the top and bottom films.

OR Touch finger or portion of hand to hydrated gel area. Rejoin the top and bottom films. Wash hands after finger or hand plating. All 3M Petrifilm Plates except Yeast and Mold Count Plates can be used for finger or hand plating.

3 Incubate and enumerate as directed in product instructions. Refer to 3M Petrifilm Plate Interpretation Guide when enumerating results.

3M™ Petrifilm™ Yeast and Mold Count Plates: On occasion, the gel may split (adhering to both the top and bottom films) when the top film is lifted. If this happens, the plate with gel splitting may still be used for air testing, but is not recommended for direct contact use.

Source: [3M. Environmental monitoring](#)

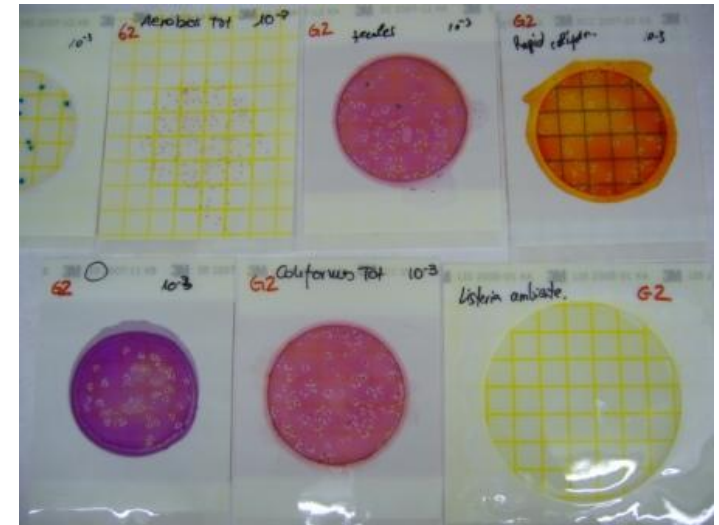
Video: [3M Petrifilm Plates for Environmental Testing](#)

Instructions for

- [Aerobic Count Plates](#)
- [Enterobacteria](#)
- [E. coli / Coliforms](#)

Interpretation guide for

- [Aerobic Count Plates](#)
- [Enterobacteria](#)
- [E. coli / Coliforms](#)

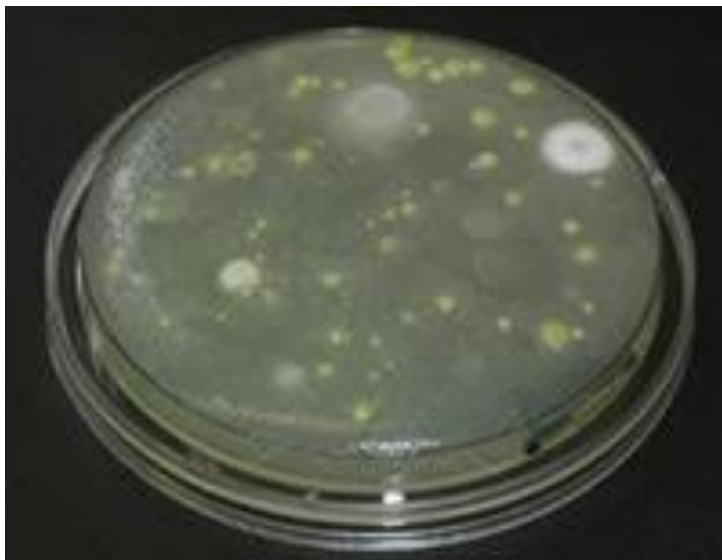
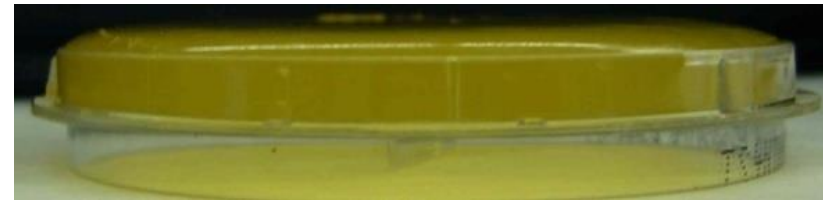


Other products:
[Compact Dry](#), [Rida Count](#)



Contact Plates

- Agar surface is pressed against the surface for a short moment
- → Incubation
- → Counting the colonies
- For example [RODAC](#) Plates





ATP Measurement

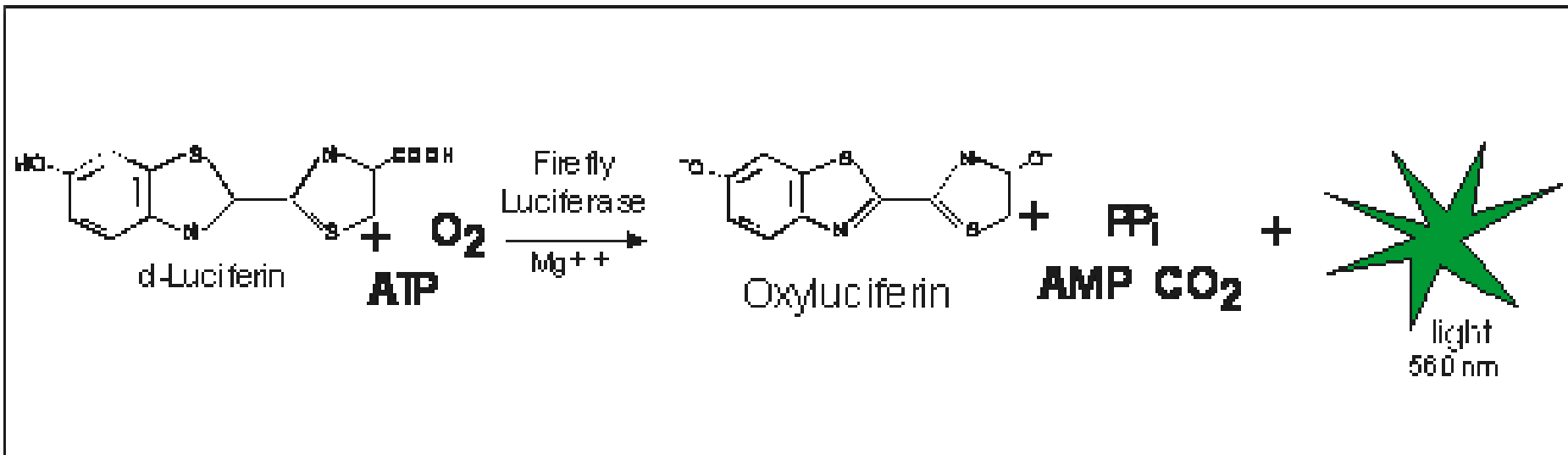
- ATP (adenosine triphosphate) molecules are present in all cells (microbial, plant, animal cells) → good indicator of organic contamination
- In hygiene monitoring, a device called luminometer and test swabs with reagents (luciferin, luciferase enzyme) are used
- Many providers and products ([3M Clean –Trace™](#), [Hygiena](#), [Lumitester...](#))





Principle of ATP Measurement

- ATP reacts with luciferin in the presence of the catalyst (luciferase enzyme) and produces light, which can be recorded by a luminometer



- Rapid method: Result in a couple of minutes
- Suitable tool for checking total surface cleanliness!