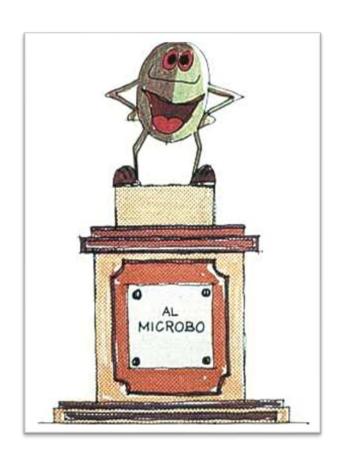
## Microbiology per non microbiologists

Brief microbiological training for production and Public Health people

(Pharma, Agro-Food, Dairy, Beverage, Catering, Cosmetic, Public Health, Biotechnology, HVAC, Laboratory, Pharmacy, Hospital)

Roberto Ligugnana

## LIGU *Bacteria*Family Monument



Why a monument for the microorganisms?

You will know the reason at the end of slides presentation

#### **Terminology**

- Bio-aerosol
- Controlled environment contamination
- Bacteria
- Calibration
- Colony (CFU)
- Cross Contamination
- Disinfection

- Germ
- Laminar flow
- Microorganism
- Mold & Yeast
- Sterile
- Sterilization
- Streptococci
- Unidirectional flow

#### The big "handicap"

You do not see the microorganisms in «real time» with your eyes and therefore the humans are not easily involved.

#### The Staff

The personell should be therefore trained to understand the presence of germs around us.

It is not easy to convince the staff to follow the correct hygiene procedures.

#### The microorganisms

The germs are not visible with niked eyes and the results of their negative activity are visible only after several hours or days.

It is therefore difficult to connect a «disaster» (contamintion) with a specific wrong activity.

#### Germs visible to naked eye

To be sure the staff understand the reasons of a correct hygiene, we should demonstrate that the microorganisms are present.

A practical example is more effective that 1000 words!

#### Germs visible to naked eye



# Practical suggestions to know the basis of microbiology

Few simple activities:

- A. Basic simple microbiology with the help of "comics"
- B. Basic simple Microscopy
- C. Growth of micro-organisms on agar (Demo, A, B, C, D, E, F)
- D. Grafic to evidentiate the results

## Basic Microbiology by «comics»

The basic microbiology should be presented in simple way to be sure that it is very well understood.

The slides should be presented by the Production Manager with the adaptation to the local production conditions.

#### LIGU Bacteria Family

LIGU Bacteria family consists of:

- Bacterium robyophylum
- Bacterium liguophylum
- Bacterium triobasophylum
- Bacterium Christmasophylum



The life of LIGU Bacteria family will help us to discover the worldof micro-organisms



My name is Bacterium robyophylum



#### My identification card

- DOMAIN: Bacteria
- KINGDOM: Eubacteria
- PHYLUM: Proteobacteria
- CLASS: Hygienebacteria
- ORDER: Ligubacteriales
- FAMILY: Ligubacteriaceae
- GENUS: Ligu
- NAME: Bacterium robyophylum



My name is Bacterium liguophylum



#### My identification card

- DOMAIN: Bacteria
- KINGDOM: Eubacteria
- PHYLUM: Proteobacteria
- CLASS: Hygienebacteria
- ORDER: Ligubacteriales
- FAMILY: Ligubacteriaceae
- GENUS: Ligu
- NAME: Bacterium liguophylum



My name is Bacterium triobasophylum



#### My identification card

- DOMAIN: Bacteria
- KINGDOM: Eubacteria
- PHYLUM: Proteobacteria
- CLASS: Hygienebacteria
- ORDER: Ligubacteriales
- FAMILY: Ligubacteriaceae
- GENUS: Ligu
- NAME: Bacterium triobasophylum



My name is Bacterium christmasophylum



#### My identification card

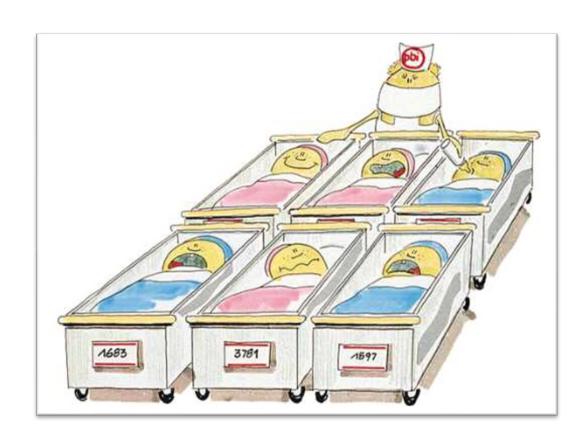
- DOMAIN: Bacteria
- KINGDOM: Eubacteria
- PHYLUM: Proteobacteria
- CLASS: Hygienbacteria
- ORDER: Ligubacteriales
- FAMILY: Ligubacteriaceae
- GENUS: Ligu
- NAME: Bacterium christmasophylum

## 10. LIGU Bacteria Family Sexual life



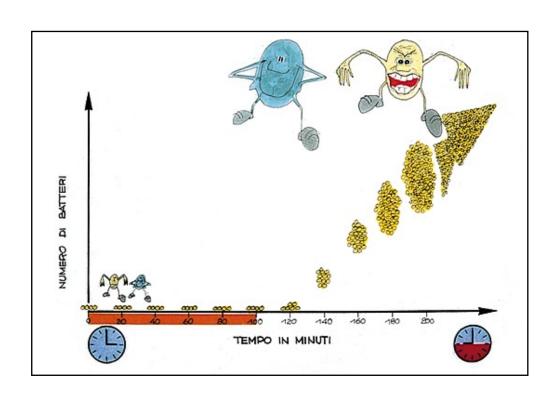
They are very sensible to the "sex appeal"

# 11. LIGU *Bacteria family* Reproduction



They are very prolific

#### 12. LIGU Bacteria Family



Their names are in the Guiness book: a new generation in about 20 minutes!

## 13. LIGU Bacteria Family Social life



The family is very important

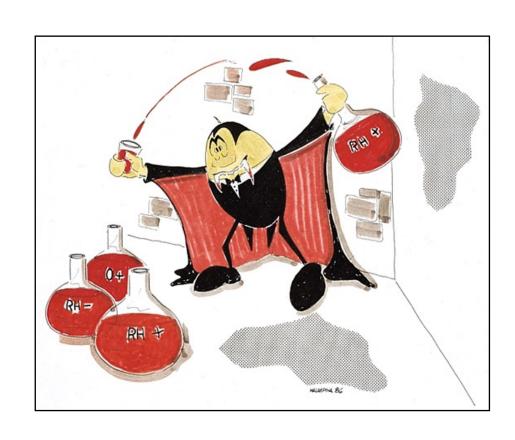
#### 14. LIGU Bacteria Family Food and drinks



The food quality is very important for his health.

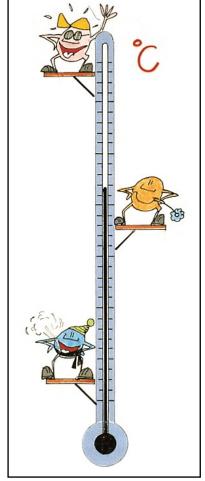
He prefers the international kitchen.

### 15. LIGU Bacteria Family Drinks



He prefers fresh blood

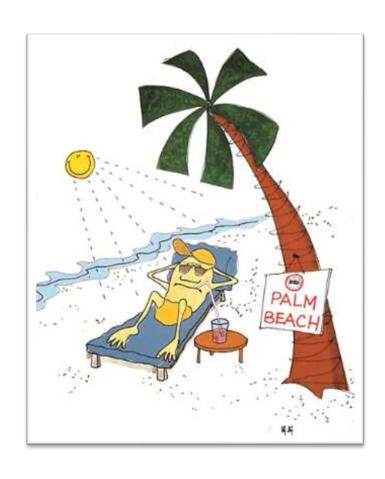
## 16. LIGU *Bacteria Family*Temperature



He is able to survive at different environmental temperatures (from -0°C + 70°C)



He sometimes prefers cold, dry environments



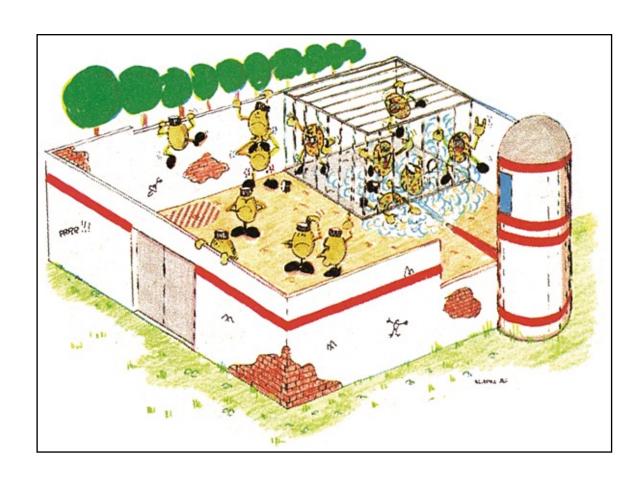
He sometimes prefers warm, wet environments



He sometimes prefers environments with river water

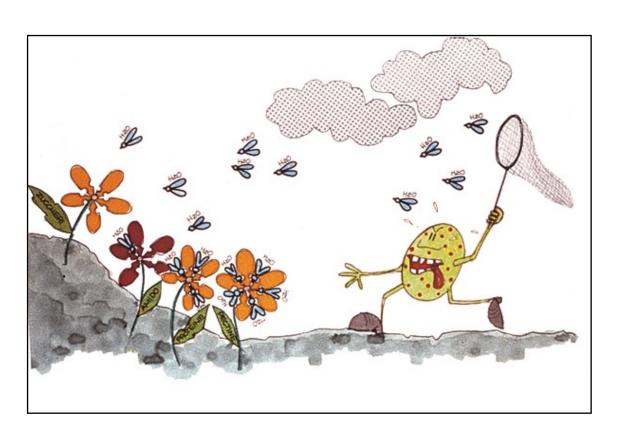


He doesn't like confined spaces



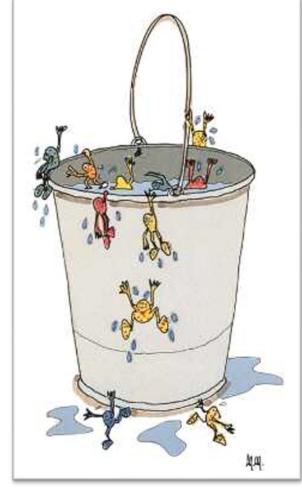
If he is confined, he tries to escape

# 22. LIGU *Bacteria Family*Dry Environment



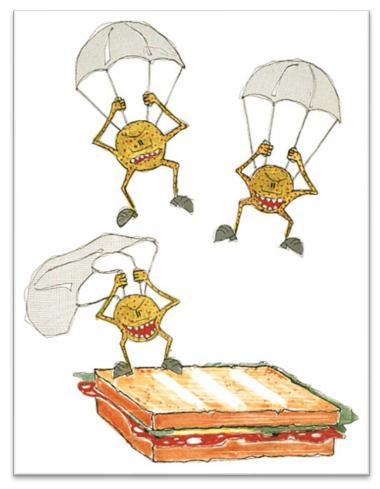
He is looking for water molecules in dry environments (Aw = Water Activity)

## 23. LIGU *Bacteria Family*Spreading



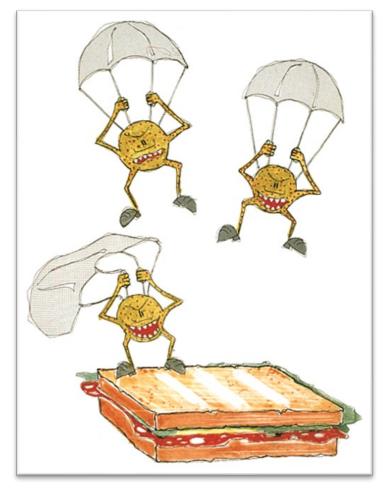
He is always ready to spread everywhere

### 24. LIGU *Bacteria Family* Diffusion



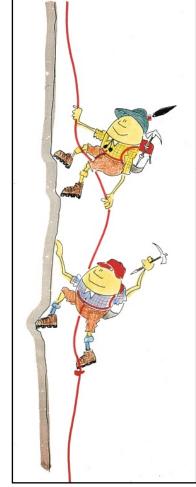
He uses water, air, humans, animals, food, for his spreading in the world around him

#### 25. LIGU *Bacteria Family* Diffusion



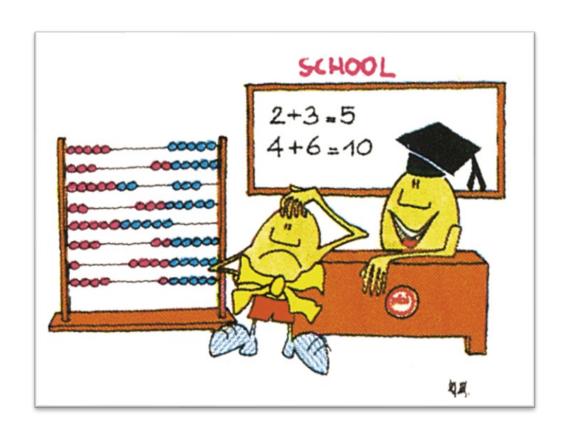
Several germs are connected together to form a colony (Colony Forming Unit)

### 26. LIGU *Bacteria Family*— Diffusion



He is able to survive in difficult situations on any typeof surfaces

### 27. LIGU *Bacteria Family* Education



He is an ideal student: he quickly and easily learns any type of activity

# 28. LIGU *Bacteria Family*Arbiter elegantiarum



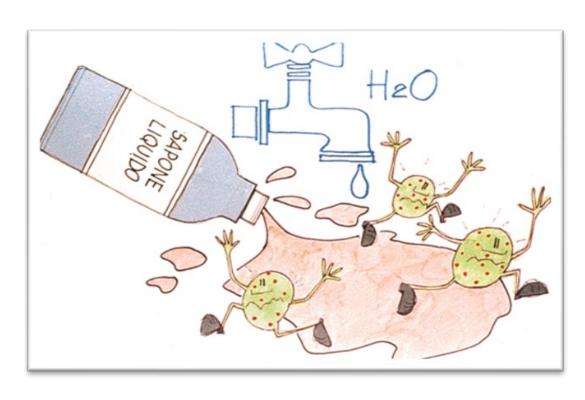
He can dress different colours and parfums

### 29. LIGU *Bacteria Family* Policeman



The humans uses him as «hygiene indicator» to control the Public Health in food, drink, water, etc.

## 30. LIGU Bacteria Family Homo sapiens



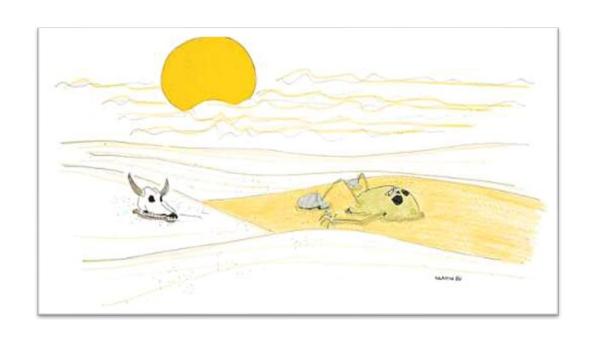
The humans use water and soap to neutralize the bad bacteria

### 31. LIGU *Bacteria Family* Steam



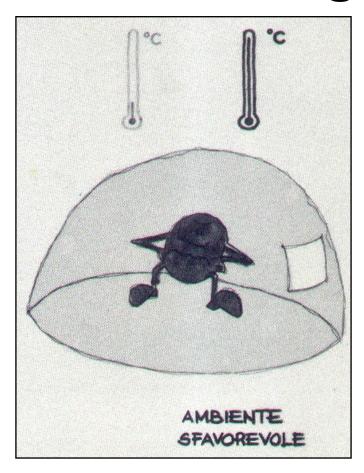
The humans fight bad bacteria with different methods (e.g. steam sterilization)

# 32. LIGU *Bacteria Family* dry



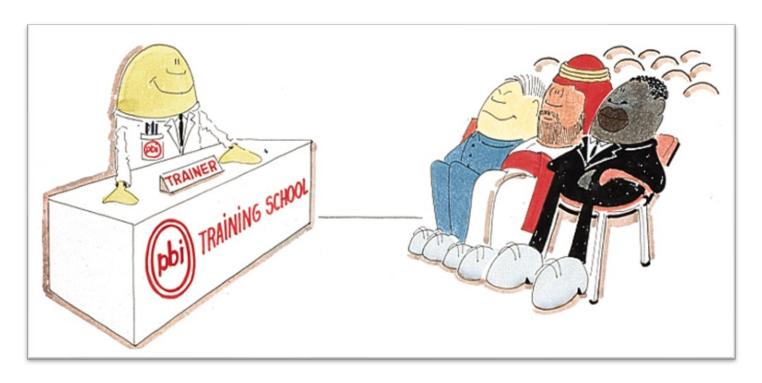
The humans fight bad bacteria with different methods (e.g. dry sterilization)

# 33. LIGU *Bacteria Family* Surviving in adverse condition



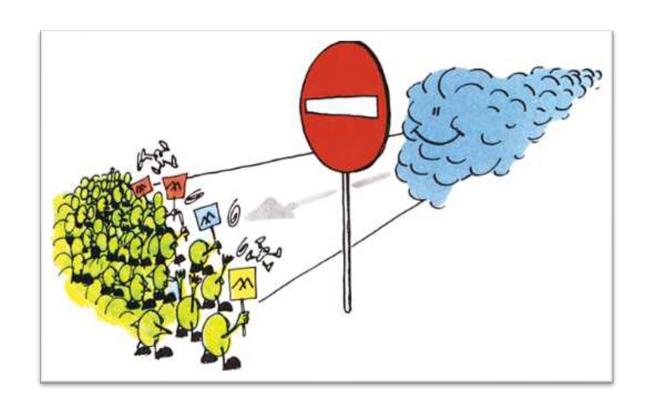
In adverse environmental conditions he protects himself by an envelope (spore) for several years

### 34. LIGU *Bacteria Family* Race



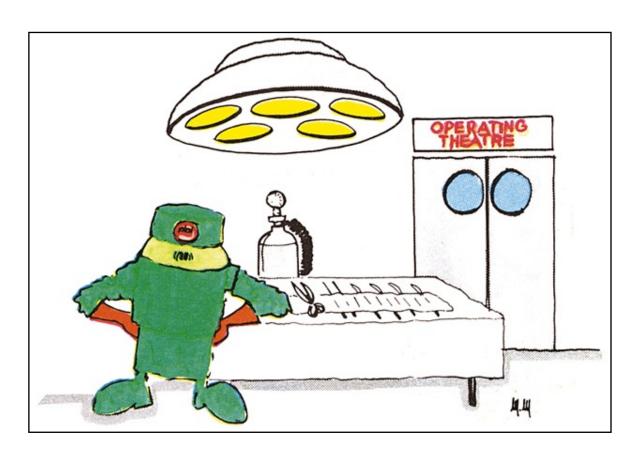
The microorganisms have differen origins

### 35. LIGU *Bacteria Family* in the traffic



He doesn't like to be neutralized by HEPA filters

### 36. LIGU *Bacteria Family* CleanRoom



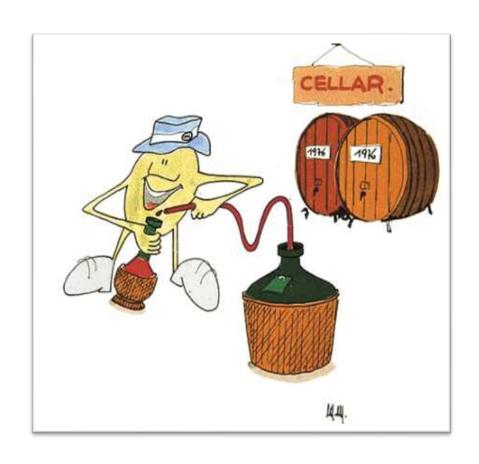
He is considered an enemy in Clean Room and operating theatres in hospitals

## 37. LIGU *Bacteria Family* The capture from the air



The microorganisms can be captured from the air by impact on agar culture using an aspiration

## 38. LIGU *Bacteria Family* The *homo sapiens* best friend



Food, dairy, wine, medicines are produced with his help. Thanks to his activity, several human lifes are saved and fantastic lunches and dinners are served.

# 39. LIGU *Bacteria Family* The *Homo sapiens* best friends



He is our friend!

If homo sapiens is on our planet, it is also his merit

# 40. LIGU *Bacteria Family*Bad Boy



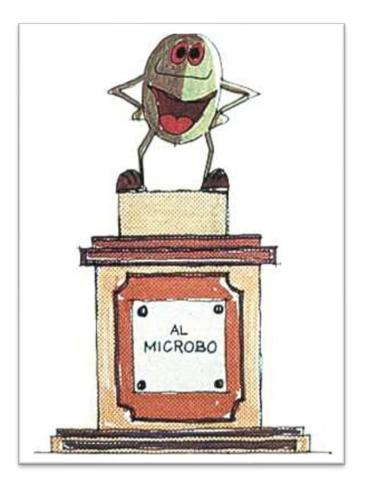
Over 95% of bacteria are «good boy» but a minority are «bad boy».

# 41. LIGU Bacteria Family Guilty



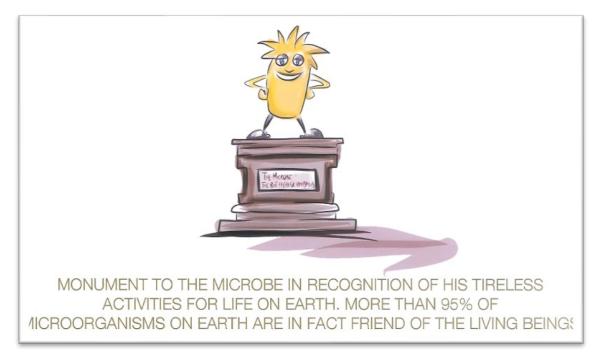
Some microorganisms are «bad boy» and when they produce disasters are death sentenced

### 42. LIGU *Bacteria Family* monument

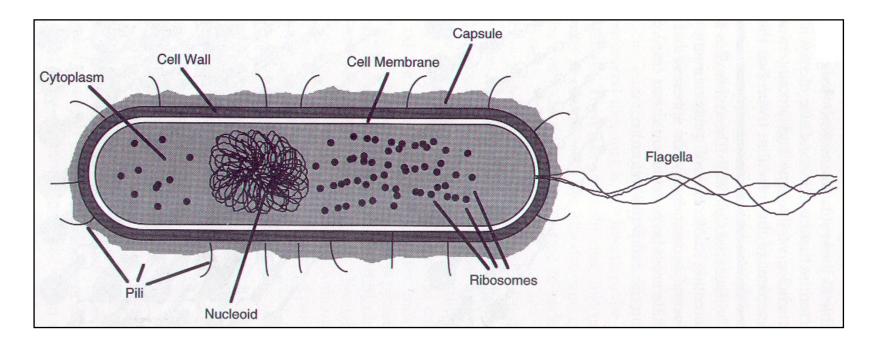


Man can live on earth thanks to the intense activity of microbes and it is therefore properly to raise a monument in their honor

### 43. LIGU *Bacteria Family* monument

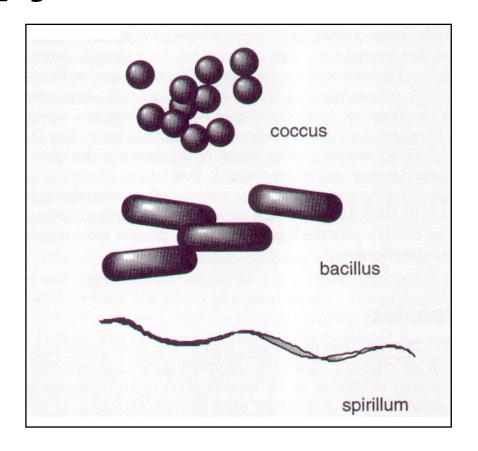


Man can live on earth thanks to the intense activity of microbes and it is therefore properly to raise a monument in their honor

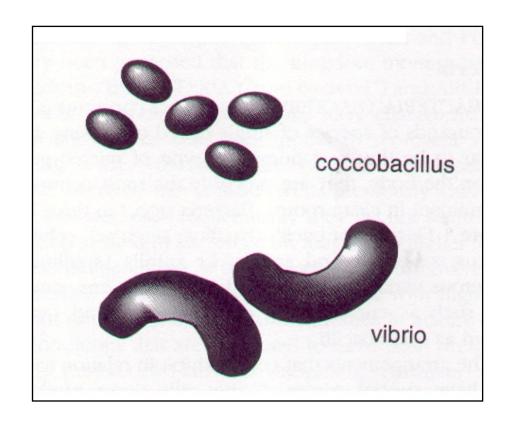


The microbial cell

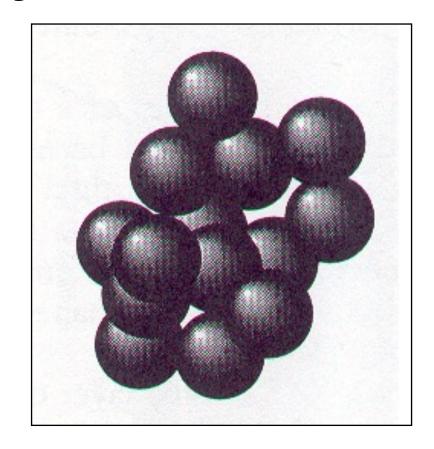
The bacteria are classified in 3 groups depending from their looking: coccus, bacillus, spirillum

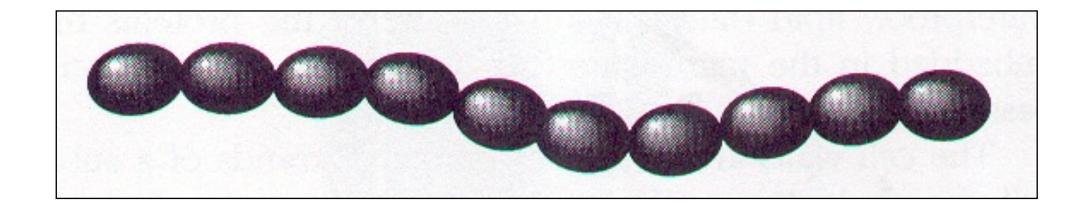


Some bacteria have different looking: coccobacillus, vibrio



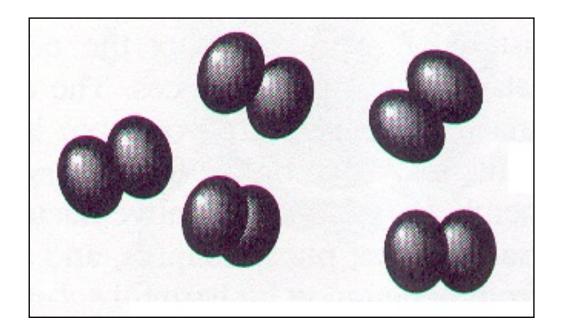
For the spherical shape and union these bacteria are called staphylococcus





The name Streptococcus is used for the description of coccus that quite often are found in chains

Two united coccus are called diplococcus



#### Demo A

<u>Purpose</u>. To show why the surface shoud be cleaned and washed before disinfection.

#### Action.

- The operator press a Contact Plate on the cleaned and disinfected surface.
- 2. The operator repeats the same action on a dirty surface.

The colonies (CFU) will appear on agar surface after 24-48 hours of incubation at 32-37°C.



#### Demo A

#### Comment.

At the end of incubation time, the not cleaned dirty surface will show a higher number of colonies.

This will be the demonstration how the correct sanification is important.

#### <u>Demo B</u>

<u>Purpose</u>. To show why utensils and surfaces in contact with food must be cleaned

Action. The operator press the Contact plates on the surface of blames, mixers, working table before and after the cleaning procedures

#### **Demo B**

#### Comment.

At the end of incubation time, the higher number of colonies on the Contact plate used before cleaning protocol will show the importance of a correct procedure.

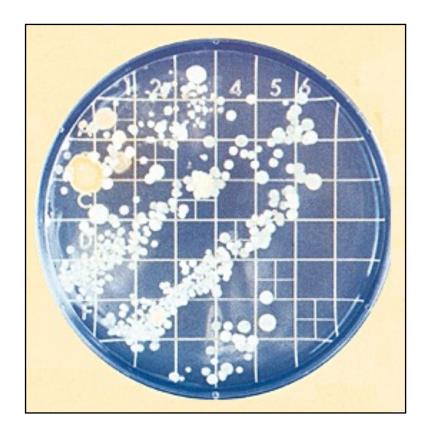
#### **Demo C**

Purpose. To show the importance of hands washing after the use of toilet.

Action. The operator press a Contact plate on the finger before and after the use of toilet (with and without hands washing).

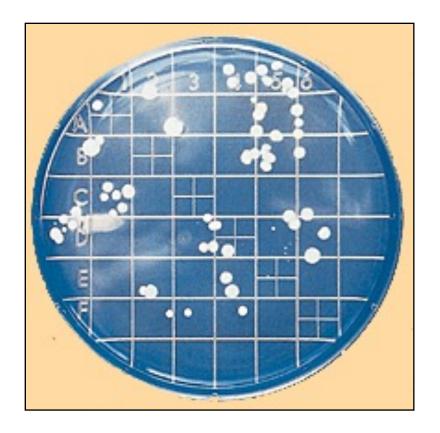
# Microbiological fingers imprint

Fingers Imprint before washing



# Microbiological fingers imprint

Fingers Imprint after cleaning and washing



#### **Demo C**

#### Comment.

At the end of incubation time, the higher number of colonies on the Contact plate used before cleaning will show the importance of a correct procedure at the end of use of toilet

# Surface Example of Microorganisms growth

ufc/24 cm <sup>2</sup> Surfair Plate	Punteggio	Classificaz.
0	+	No
		contaminazione
1-9	++	Bassa
		contaminazione
10-20	+++	Alta
		contaminazione
21-100	++++	Livello di
		Allerta
101- confluenti	+++++	Livello di
		Allarme

#### <u>Demo D</u>

<u>Purpose</u>. To show how the dry cleaning of the floor is a high source of contamination in comparison with wet cleaning

Action. Air sampling with an air sampler before and after dry and wet cleaning cycles

# Microbial air sampling

Microbial air sampler TRIO.BAS MONO



# Microbial air sampling

Microbial air sampler TRIO.BAS DUO



# Microbial air sampling

Microbial air sampler TRIO.BAS TRIO



## Microbial air sampling

### **Protocol**

- A. Insert a Contact plate
- B. Aspirate 250 Its of air
- C. Transfer the plate to incubator
- D. Count the colonies



### <u>Demo D</u>

#### Comment.

The difference in number of colonies on the Contact plates with dry and wet cleaning procedures will show the importance of a correct cleaning protocol

### <u>Demo E</u>

<u>Purpose.</u> To show the efficacy of aerosol disinfectant in a closed environment (e.g. refrigerator, ripening room, etc.)

Action. Air sampling with microbial air sampler before and after disinfection treatment

### <u>Demo E</u>

### Comment.

The lowest number of colonies on the plate after incubation will show the efficacy of the disinfection treatment.

### <u>Demo F</u>

Purpose. To show how a crowded room is more contaminated that an empty room

Action. Air sampling with microbial air sampler «At Rest» (before activity starting) and «In Operation»

# «At Rest» ( e.g.: Low contamination in food factory)



# «In Operation» ( e.g.: High contamination in food factory)



### <u>Demo F</u>

### Comment.

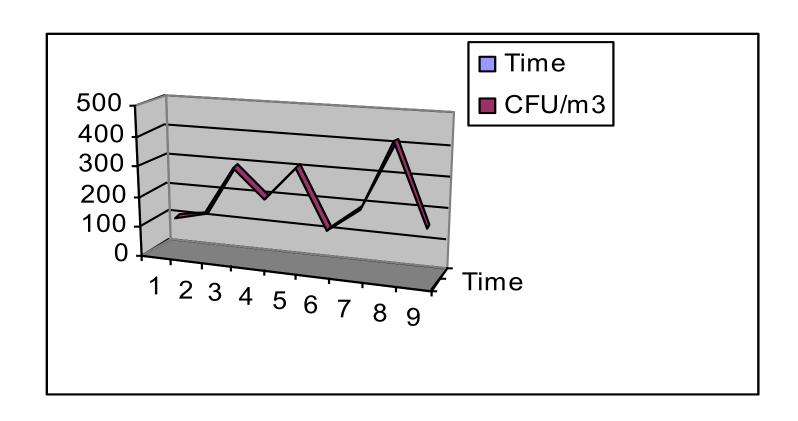
The lower number of colonies on the plate of «At Rest» after incubation will show how the presence of people brings contamination.

# Result presentation on graphic

The reported graphic is an example of what it is possible to produce in Excel

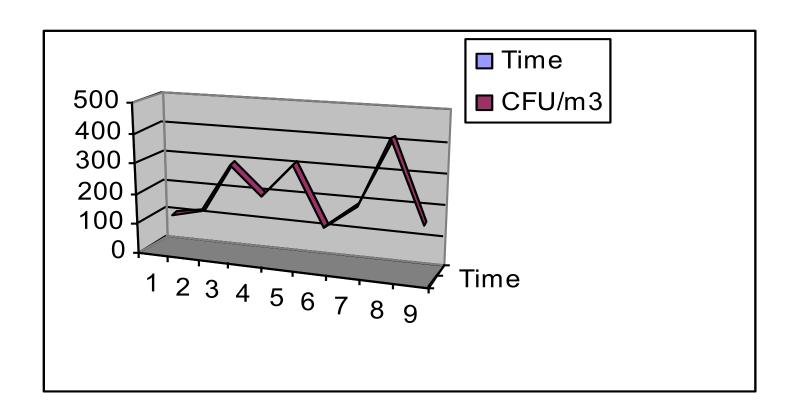
It is important that the graphic is updated and visible for all the personell to give the possibility to everyone to evaluate and comment the results.

### **Graphic example**



#### **Graphic example**

The trend of the graphic (week by week) is the most effective method to involve the operator in the evaluation of operative hygienic conditions





1. Avoid nervous movements such as scratching your head or touching the chin or cheeks.

Such actions contaminate gloves and generate contamination.

- 2. Never open the clothes and reach undergarments when operating.
- 3. The talk should be limited to essentials.

- 4. Everything that falls on the floor should not be reused before being "refurbished". In gathering the fallen objects, protect the gloves or replace them.
- 5. Gloves should be replaced whenever it is believed they are contaminated and in case of damage or breakage.

6. Do not put on the production line spilled products

7. Eating, drinking, chewing are not allowed, not only because the foods and their packages are a source of contamination, but also because the movements of the mouth and face while chewing contribute to increase the particle contamination of the air.

8. Wear clean clothes, light in color, maintaining a high degree of personal cleanliness

- 9. Wash and disinfect the hands carefully with appropriate products, before starting work for any reason.
- 10. Use hand towels and other disposable cleaning cloths.

11. Remove all jewelery and insecure, when you handle the product with your hands; take off your rings.

- 12. If the gloves are used to handle the product, keep them intact, washed, disinfected, replace them frequently.
- 13. Wear hats or other effective means to retain the hair.

- 14. Do not keep personal items or clothing, do not eat or drink, do not smoke in the working areas of the premises, in the areas destined to the cleaning of equipment and utensils.
- 15. Take any precaution to prevent the possible contamination of the product (protective masks, easily disinfected rubber shoes etc.).

## Conclusions for the trainer

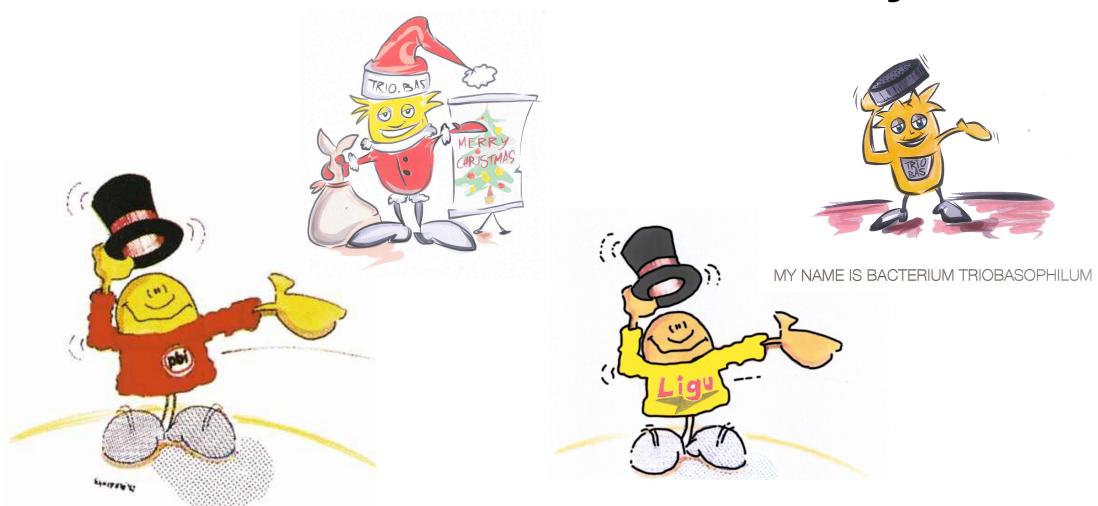
To implement an educational plan, you must follow some basic points:

- 1. Management Involvement
- 2. Availability of material for demonstrations
- 3. Training planned in subsequent sessions in order to enable participants to "absorb" and "digest" the basic knowledge of hygiene

## Conclusions for the trainer

- 4. Persist even in case of initial difficulties and lack of cooperation on the part of some operators
- 5. The practical task must be completed with practical demonstrations largely centered on proper hand washing

### Thanks to the LIGU Bacteria Family actors



## Usus magister est optimus (Latin)

(Cfr.Cicerone, Pro Rabirio Postumo, 4; and e De Orat., 1, 4)

## The exercise and the example are the best teacher