Microbiological (CFU) measurements - in theory and practice

Human IC:

Temperature Controlled Airflow
CFU measurements

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Let me introduce myself briefly

- Henrik Skredsvik
- Architect
- Accidentely slipped into ventilation for OR:s 16 years ago
- Principal Architect at Avidicare
- Work with system design, R&D and measurements

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Topics Covered Today

Measuring Colony Forming Units (CFU)

-Why
-Where
-How

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Why measure bioburden (CFU)?

- Technical check of installations
- Establish hygiene level (SSI)
- Control compliance to a standard
- Infection tracing

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Surgical Site Infections (SSIs)

- Surgical site infections (SSIs) are a major global issue, causing significant suffering, fatalities, and skyrocketing healthcare costs.
- A single infection can cost anywhere between $10,000 and $100,000 per case.
- Despite preventive measures like sterile protocols, prophylactic antibiotics, and ventilation, 2–10% of surgeries still result in infection depending on geography.
- The world's best hospitals achieve infection rates below 1%, proving there is massive potential for improvement.

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What is bioburden?

- Bacteria
f
- Fungi (spores)

What is a Colony Forming Unit?

A colony-forming unit (CFU) is **a unit of measurement used in microbiology to estimate the number of viable bacteria or fungal cells in a sample**. It represents the number of individual cells or groups of cells that can divide and multiply to form a visible colony on a culture medium.

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Where do the CFUs come from?

- Bacteria come mainly from people when in a protected environment. We shed skin scales and aerosols from nose and mouth.

- Fungi (spores) are generally considered a building problem but can enter, for example, via cardboard boxes and people.

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Where can CFU measurements be useful?

- Clean rooms (pharmaceutical)
- Food industry (processing and packaging)
- **Hospitals**

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Hospitals

- Operating rooms
- Instrument preparation rooms
- Hybrid rooms
- Sterile processing departements

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Ways to measure CFU

- Passive sampling
- Active sampling
- Contact sampling

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Passive sampling

Settling plates placed in relevant places exposed for a specified amount of time.

Gives a CFU/h value per area unit

CFU per volume unit can be calculated assuming a sedimentation rate (0.3m/min skin scales)

Should not be used in rooms with high airflows

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Passive sampling

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Active sampling

Agar plates or filters placed in an air extracting device running for a specified amount of time with a specified airflow (1m³/10min)

Gives a CFU value per volume unit (m^3)

Real time trackers (Biotrak)

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Active sampling

Slit sampler

Sieve Sampler

Sartorius

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Slit sampler

Draws air at a controlled flow (e.g., 100 L/min) through a narrow **slit**, impacting particles directly onto a rotating agar plate (0.1rpm)

Can be fitted with a sterile silicon hose to reach the wound area

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Sieve sampler

Draws air at a controlled flow rate through a perforated **sieve**, impacting particles directly onto a static agar plate.

Can’t be fitted with a hose, hard to reach the wound area

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Sartorius sampler

Draws air at a controlled flow rate through a gelatine filter. This filter must then be moved with sterile tweezers to an agar plate in order to be incubated.

Doesn’t need a hose to reach the wound area. Needs attending by “scrub” nurse

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Real time (Biotrak)

Draws air at a controlled flow rate through a flourecent light. Counts number of viable particles probably by measuring reflection level.

Purpose is constant monitoring, not CFU measurements.

Gives high readings compared to agar plates

Expensive

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Avoid samplers that expose the plates for fast moving air longer than 10 min

The air can dehydrate bacteria and media which may result in a false low reading.

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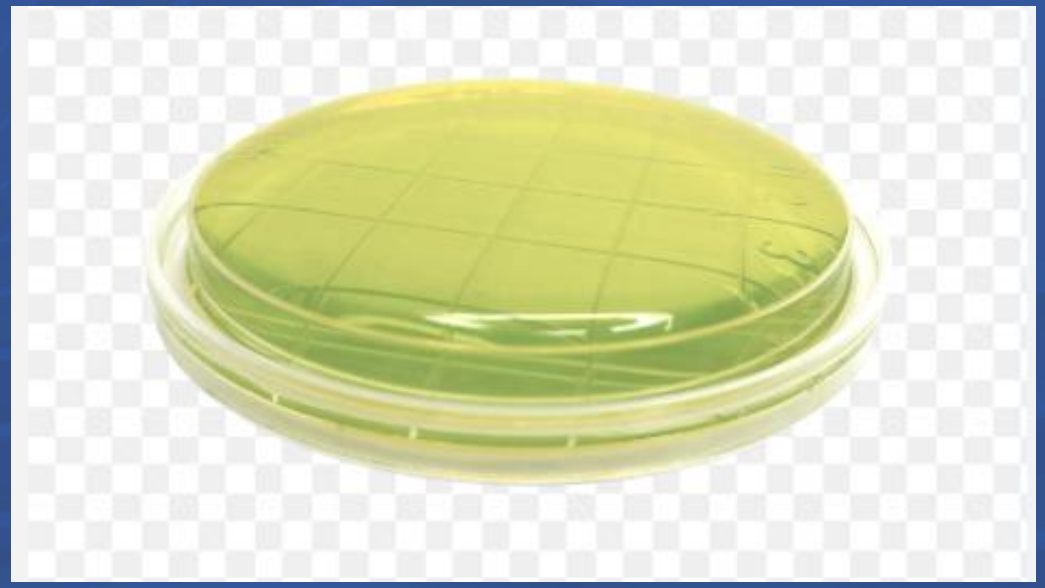
Contact sampling

Special type of agar plate

Diameter 40-60mm

CFU per sample (or per cm^2)



Surfaces, air ducts, Air Handling Units

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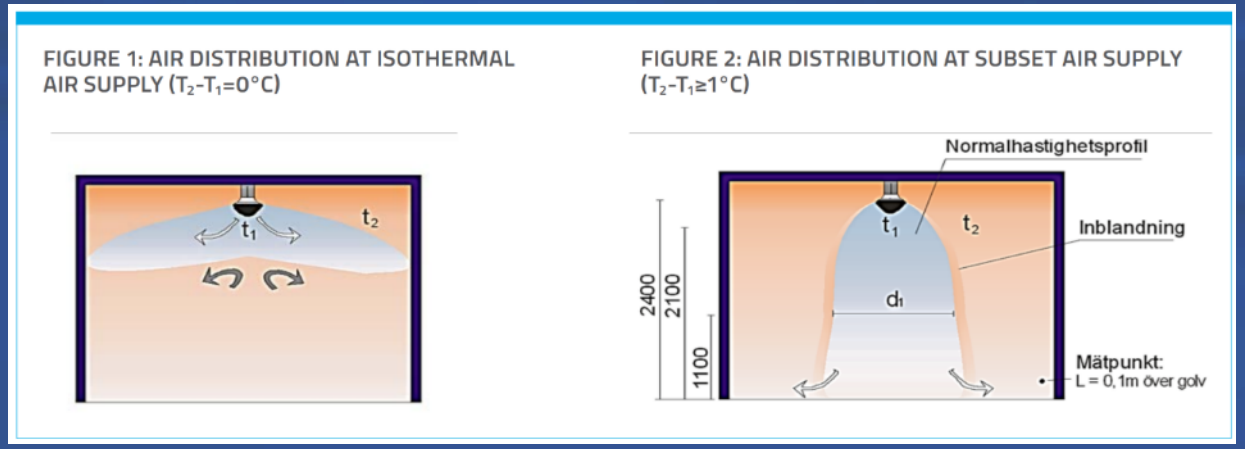
Airshowers

- The Opragon Airshower is a unique supply air diffuser
- It has low turbulence degree (”laminar”) and a half-spheric distribution shape
- The distribution pattern expands the area of air which lower velocity to allow gravity to take over



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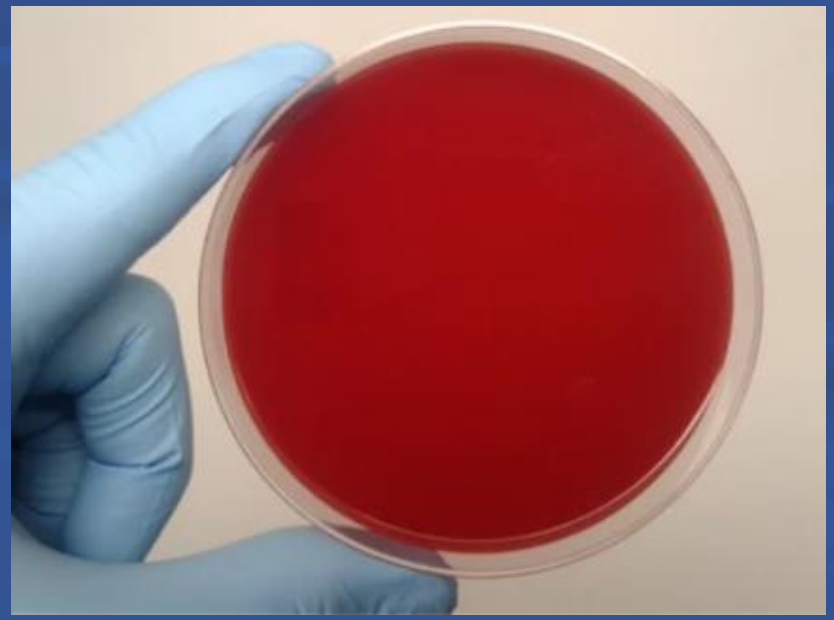
Agar plates

Many types of growth medium

Most common blood agar and soy agar (soy can be sterilized)

Different sizes 70mm to 140mm



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Incubation

Normally 48 to 72 hours
in 35-37°C

There are other temperatures and times depending on type of agar and what species of bacteria/fungi you are looking for

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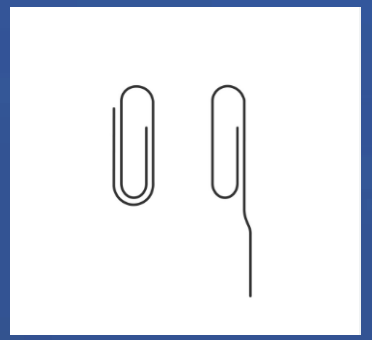
Counting CFU

If many, use pin or a paperclip to mark the ones counted

Take a picture of each plate
Number visible (optional)

To determine species let a biolab incubate and count



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Useful knowledge

- Number each plate (water proof pen) Not the lid!
- Transport, store and incubate plates upside down
- When exposing, don’t place lid upside down
- Only touch outside of plate and lid
- Let the plates reach room temp before measuring
- After exposure secure lid with sticky tape
- Start incubation **a.s.a.p.**(keep at room temp until)

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Measuring CFU in an OR (SIS-TS39:2015)

- Active sampling during live surgery (1m³/10min)
- <50cm from wound (about 1.2m above floor level) a sterile hose may be needed, on the instrument table or in the periphery
- 50 min or more of " knife time" is prefered
- Have a protocol ready to note time, plate no, number of people, door openings etc
- Take note of how the team is dressed
- Save one unexposed agar plate as negative control

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Mätning Hybridsal, Operation (stent, bukaorta)

Antal	Personal	Special-arbetsdräkt	Bomulls-klädsel	Hjälm	Mössa	Sterilrock	Munskydd	Handskar	
4	Operationsteam	4		4		4	4	4	
4	Övriga	4		4			4		

Händelseförlopp:

Händelse	Dörr öppnas	Kl	Antal pers.	Platta nr:	Mät-punkt	CFU/m3	CFU platta
Ingreppet startar, DÖ 9:19 och 9:24	2	9:17	8-9	1	1	3	3
	0	9:17		2	2	6	6
	0	9:27	8	3	1	1	1
	0	9:27		4	2	4	4
	0	9:37	8	5	1	<1	0
	0	9:37		6	2	3	3
	0	9:47	8	7	1	<1	0
	0	9:47		8	2	4	4
DÖ 9:53 -1 och 10:06 +1	2	9:57	7-8	9	1	<1	0
	0	9:57		10	2	5	5
DÖ 10:12 -1	1	10:08	8	11	1	<1	0
	0	10:08		12	2	1	1
Kontroll-agar				0		n/a	0

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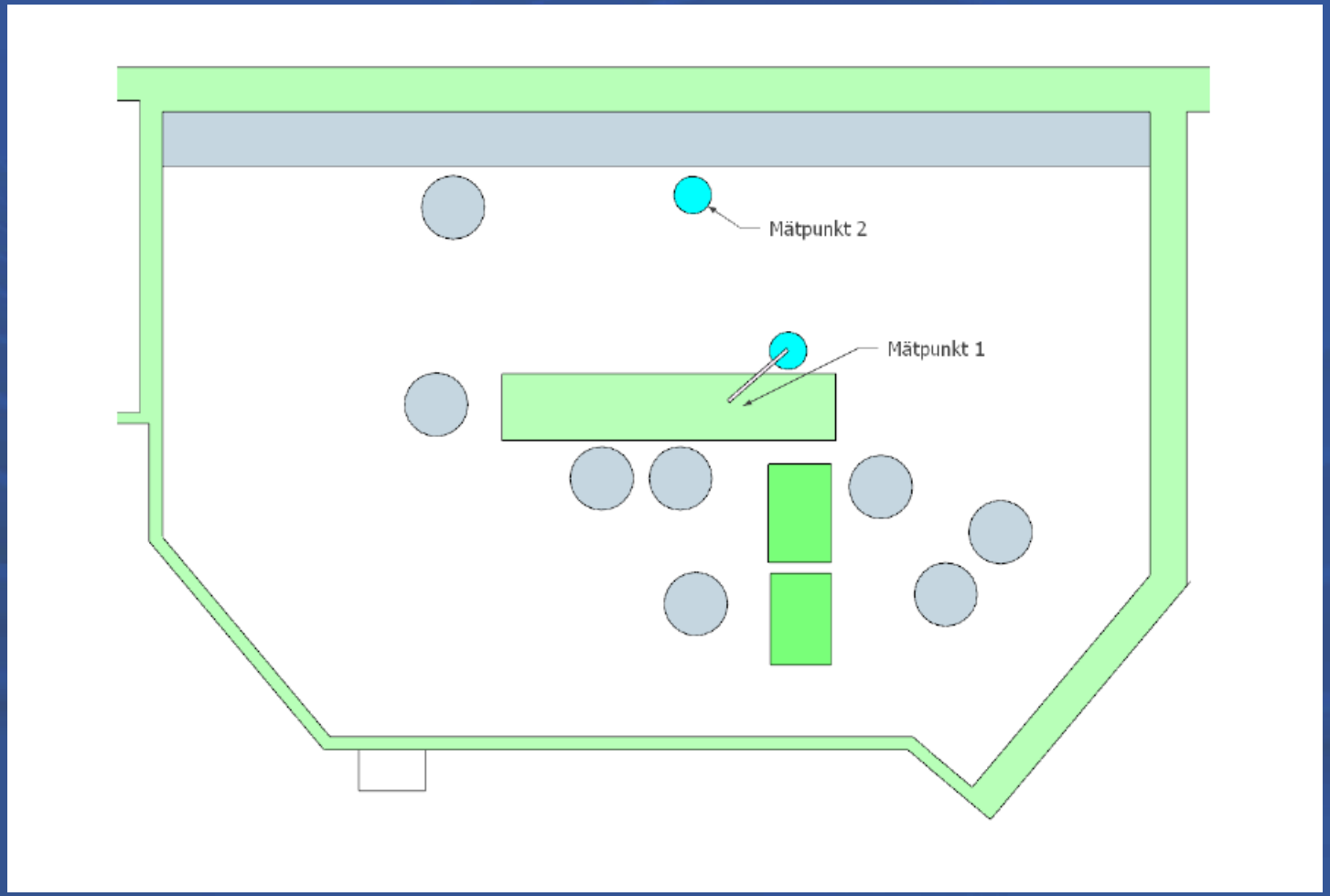
Make a simple sketch of the room

Room layout

Measuring points

Position of people

Position of instrument tables



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Making a Report

- Info about the customer (hospital, contacts etc)
- Who performed the measurements and when
- Equipment used (calibration dates if required)
- Description of how measurements were done
- Which OR and what kind of procedure
- Table with raw data
- Simple sketch of room layout
- Summary and conclusion (often appreciated)

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Why we measure CFU in the OR

- **Purpose of CFU Measurement**
  - Assess microbiological air cleanliness to protect against SSIs (Surgical Site Infections)
  - Validate ventilation performance
  - Provide **trend data** for quality monitoring
- **Regulatory Context**
  - EN 17141: Defines sampling methods and limit values for CFU/m^3
  - Healthcare ORs often align with ISO 5/EN Grade B (≤10 CFU/m^3 at rest) Swedish TS39
  - Supplementary references: ISO 14644, local HTM/HBN or DIN standards

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Useful knowledge (beeing in an OR)

- Patient and patient safety is priority number ONE
- Communicate with staff and get OK for your actions
- Ask where you can position equipment and yourself
- Stay out of sterile field, keep hands behind back
- Don’t bump into staff, instrument tables etc. With one accidental touch hours of preparation can be ruined
- Between plate changes stay close to an exhaust grill
- Don’t leave the OR or open doors without permission

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Take aways from my personal experience

- Don’t do your first measurements on your own
- A good mentor is important
- Attending surgery can be challenging
- Talking to staff and asking for help can be hard (they are focused on surgery)

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Further reading

- **EN 17141: Defines sampling methods and limit values for CFU/m³**
- **Swedish TS39**
- **ISO 14644**
- **HTM/HBN**
- **DIN standards**

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Thank you for listening

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