

# LINA + qPCR – Bacteria, Resistance, Fungi



**Positive blood culture**  
**2µl**

**BAL**  
**20µl**

LINA

Invert several times or vortex

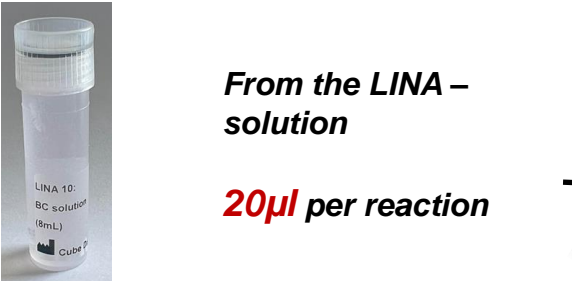


**IPC 20µl (Optional)**

Spin down the tube before use!

LINA

Invert several times or vortex



**From the LINA – solution**

**20µl per reaction**


**“Ready-to-use” PCR- master mix**

Colours:

- Red – Bacteria (16S)
- Green – Fungi (28S)
- Yellow – Resistance
- Blue – IPC

Close PCR-master mixes

**Load PCR - device**



**PCR - protocol:**

94°C 1 min. → Start PCR

**41 Cycles:**

- 94°C 5 sec.
- 56°C 10 sec. +plate read
- 72°C 30 sec.

72°C 1 min.

**Melt (only qPCR):**

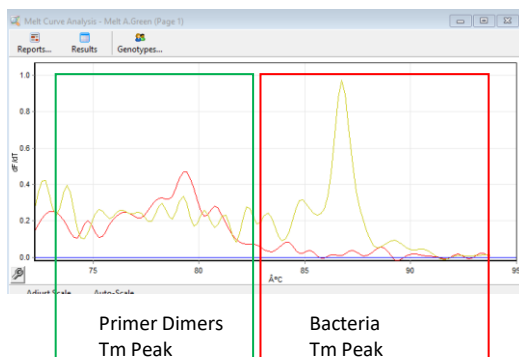
- 75°C to 94°C, increment 0,5°C steps
- 10 sec. +plate read

25°C (hold) Fluor: SYBR Green

**Validated Thermal cyclers**

- Rotor-Gene
- CFX96
- Quantstudio 3/5
- Tpersonal Thermocycler (Biometra)

## PCR interpretation (only qPCR)



1. Optional: Check the Internal Process Control (IPC) - PCR:

- Valid, if Cq < 32
- Discard test results, if IPC is invalid

2. Interpretation of the Cq and melt curves require laboratory expertise and experience in working with blood or BAL samples.

3. Thermal cyclers may differ in their thermal characteristics, therefore the optimization of the temperatures in the protocol may be recommended (for validated devices) if the results are undesirable.

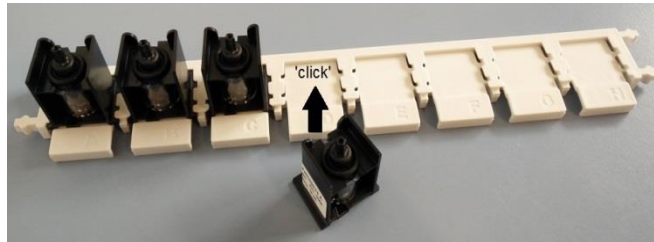
**Storage:** -25° to -18°C

# hybcell – Bacteria, Fungi or Pathogens

Unpack hybcell



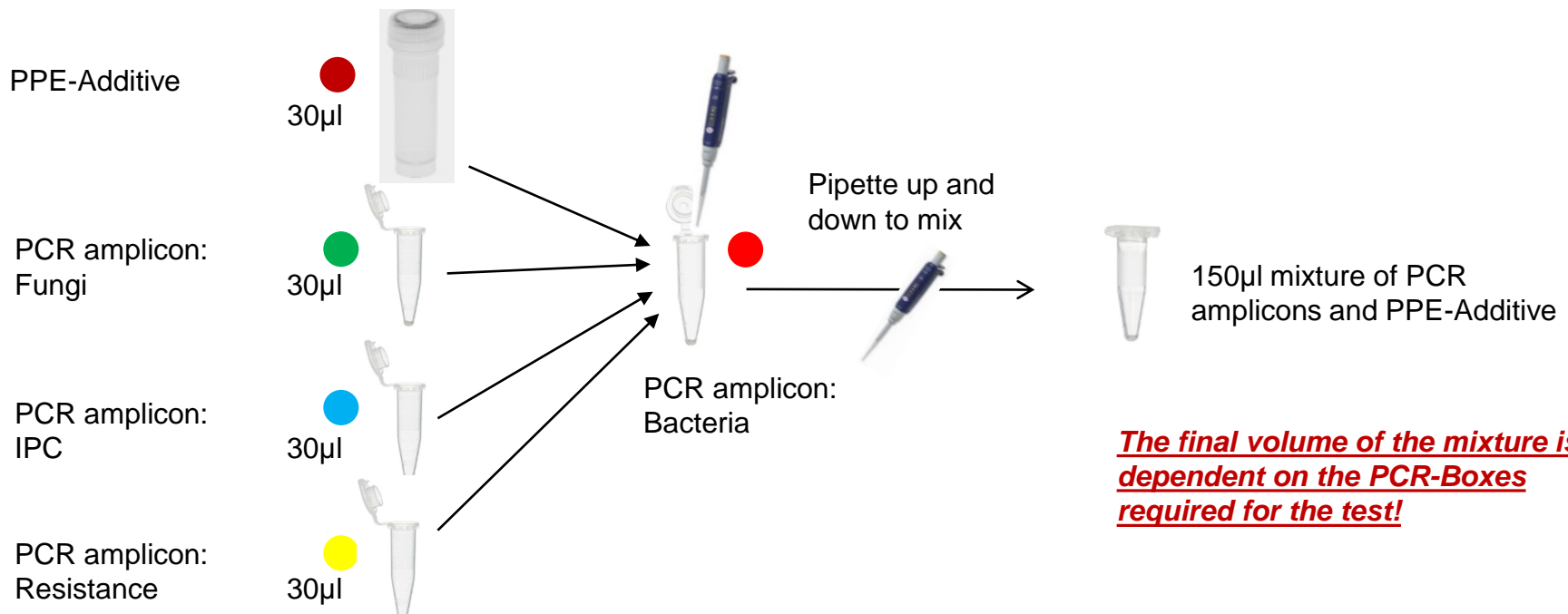
Insert into rack: "click"



	●	●	●	●
	Bac. PCR	Res. PCR	Fun. PCR	IPC PCR
hybcell Bacteria	●	●		●
hybcell Fungi			●	●
hybcell Pathogens	●	●	●	●



**Combine all the desired amplicons (30µl each) into any one of the amplicon tubes. Pipette 30µl of the PPE-Additive into the mixing tube.**

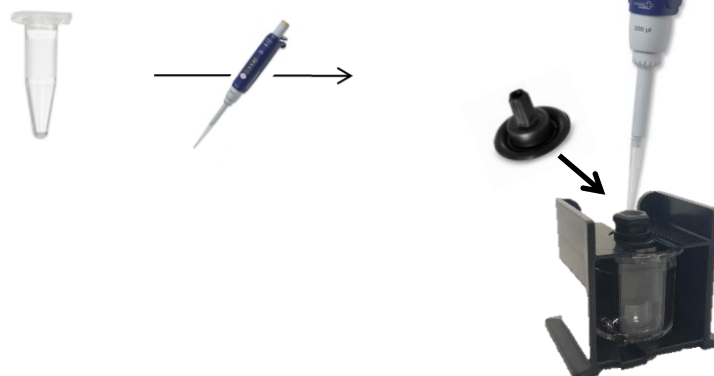


**The final volume of the mixture is dependent on the PCR-Boxes required for the test!**



**Fill the hybcell with the (positive) PCR-amplicon and PPE-Additive mixture**

**Pipette the entire volume gently and at once!**



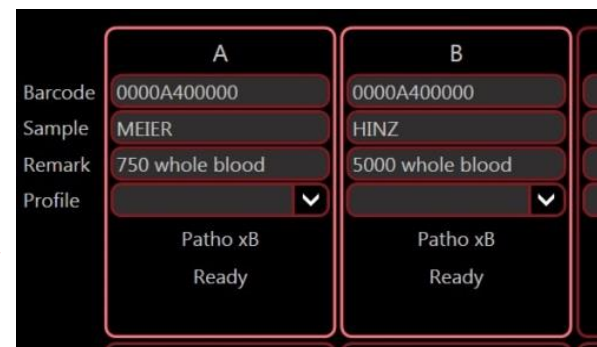
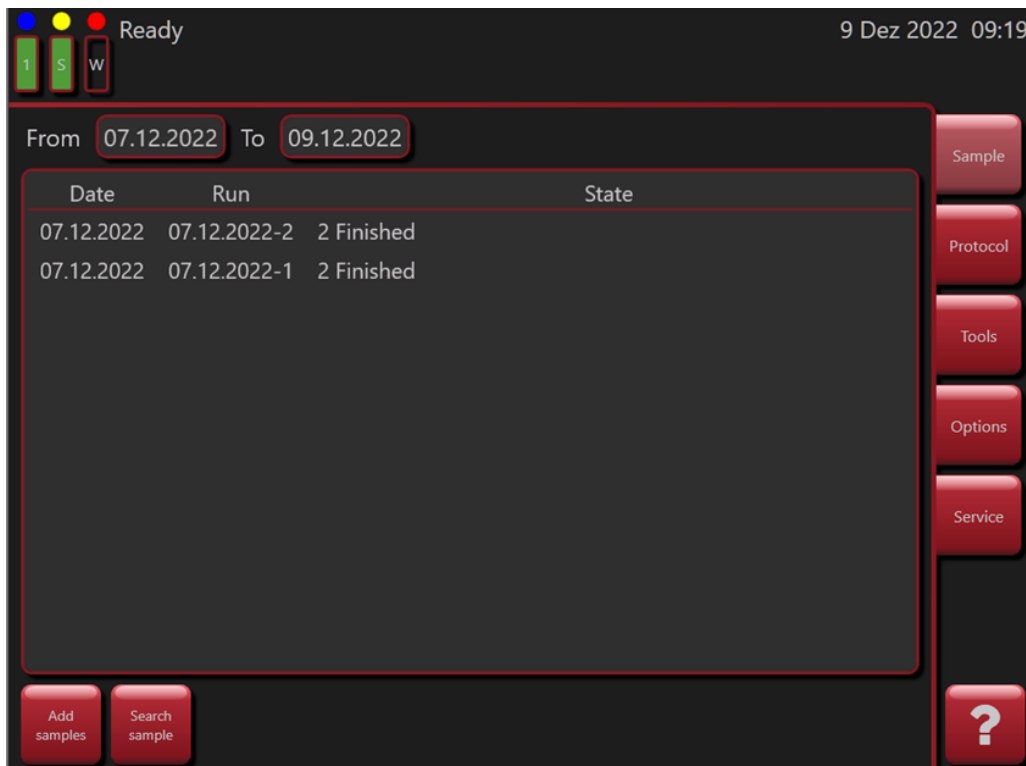
**Insert the tip of the pipette deeply into the hybcell!**

**Try not to wet the hybcell's inside margins!**



# hybcell – Bacteria, Fungi or Pathogens

## Create a run and start



1. Create new sample ("Sample" screen)
2. Insert data (sample ID, hybcell ID)
3. Select samples and start run ("Sample" screen)
4. Insert the rack (Barcode facing inwards) and confirm