# Reagents area

Purpose	<ul> <li>Preparing DEB™ Extraction Buffer</li> <li>Preparing Complete Master Mix</li> </ul>	
Precautions	No samples or completed PCR reactions	should be brought to this area
Materials	Reagents DEB™ Extraction Buffer PCR Master Mix Primers  Equipment 20-200 µl & 100-1000 µl micropipettes Marker miniRack Microtube rack Trash bin Bottle with bleach solution  Consumables Tissue paper Gloves 200 µl & 1000 µl filter tips PCR tubes 1.5 ml tubes	REPORT  STATE OF THE PORT OF T



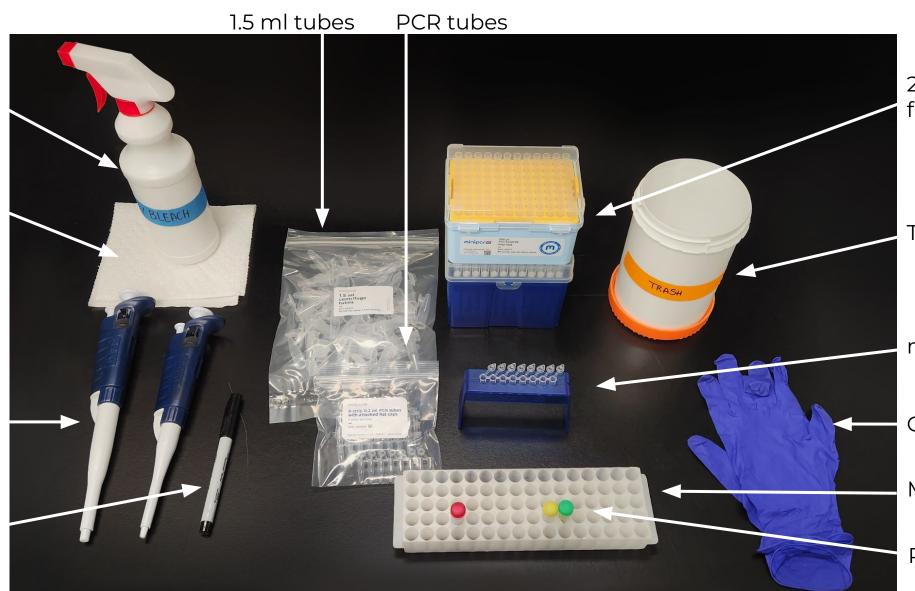
## Reagents area – Materials

Bleach

Tissue paper

100-1000 µl & 20-200 µl micropipettes

Marker



200 μl & 1000 μl filter tips

Trash bin

miniRack

Gloves

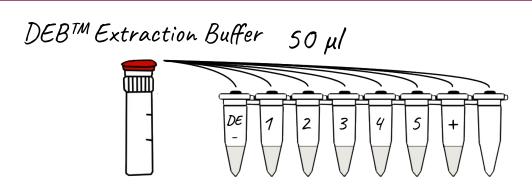
Microtube rack

Reagents

### Reagents area – Setup

#### **DNA** extraction

- Add 50 µl of DEB™ Extraction Buffer to each PCR tube
- One per sample and two for the controls

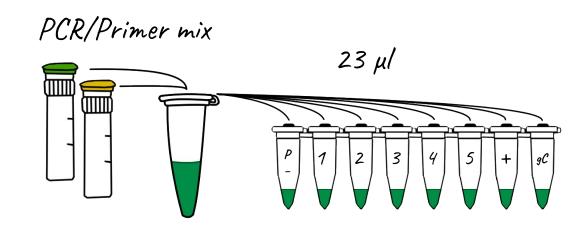


#### **PCR**

- Combine the PCR Master Mix and Primers into a 1.5 ml tube and mix well
- 2. Add 23 µl of the Complete Master Mix to empty PCR tubes and firmly close the caps

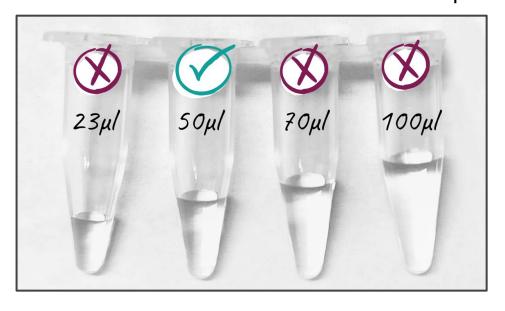
	Number of total tests*		
	1	8	16
PCR Master Mix (µI)	5.5	44	88
Primers (µI)	22	176	352
Total volume (µl)	27.5	220	440

<sup>\*</sup>For other numbers of tests, multiply the volumes required for 1 sample by the total number of tests

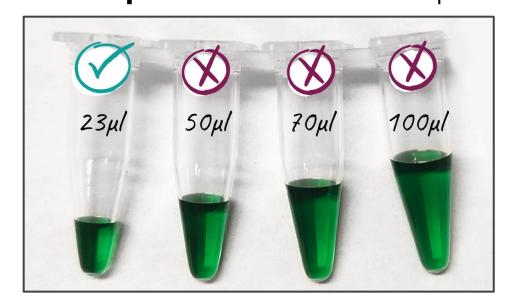


### Reagents area – Volume Reference

### **DEB™ Extraction Buffer:** 50 µl



### Complete Master Mix: 23 µl



# Sampling area

Purpose	<ul> <li>Preparing samples</li> <li>Transferring lysates to the Complete Master Mix</li> <li>Transferring control to the Complete Master Mix</li> </ul>
Precautions	Material exposed to the Sampling area should <u>not be returned to the Reagents area</u>
Materials	Reagents gelPCR™ Control 1  Equipment Microcentrifuge (optional) 1-10 µl micropipette Marker miniRack Trash bin Bottle with bleach solution  Consumables Tissue paper Gloves 10 µl filter tips Parchment paper Sample collection tools (e.g. punches, razor blades, disposable tweezers, etc.)



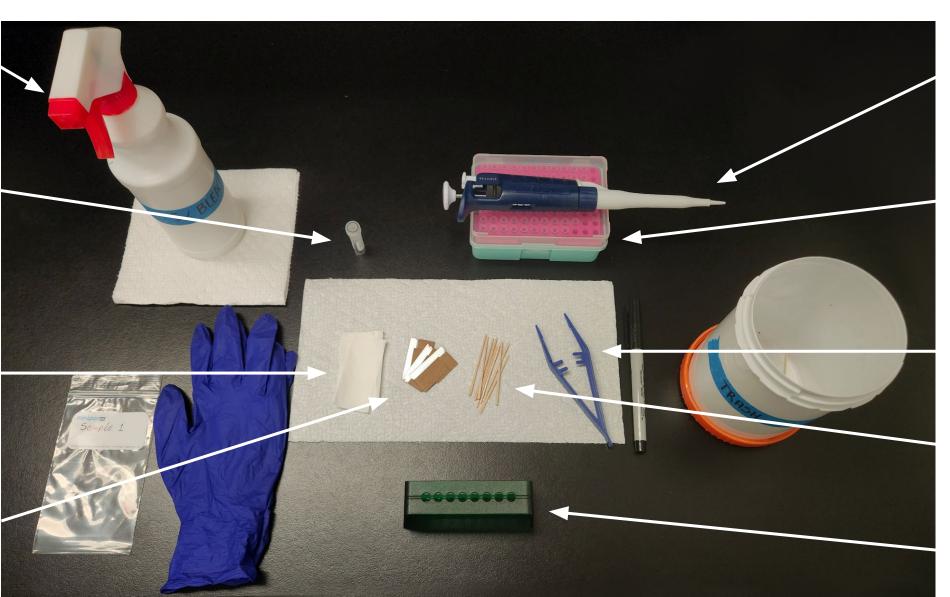
# **Sampling area – Materials**

Bleach

gelPCR™ Control 1

Parchment paper

Razor blade



1-10 µl micropipette

10 µl filter tips

Tweezers

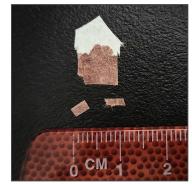
Toothpicks

miniRack

# Sampling area – Lysis

- Add sample to the DEB™ Extraction Buffer (reminder: clean or change gloves in between samples!)
- 2. Close the cap firmly
- 3. Dispose of used collection tools
- 4. Repeat for all samples

#### **Example of sample lysate tube**





#### Move to the PCR area and start the Lysis program

#### Return to the Sampling area once Lysis is complete

- 1. Transfer 2 µl of lysate to the corresponding tube with PCR mix (reminder: change tip between samples!)
- 2. Close the cap firmly
- 3. Repeat for all samples
- 4. Add 2 µl of gelPCR Control 1 to one tube (optional)

Move to the PCR area and start the PCR program

# **PCR** area

Purpose	<ul><li>Running PCR</li><li>Visualizing test results</li></ul>
Precautions	Material exposed to the PCR area should not be returned to the Reagents area
Materials	Reagents SeeGreen™ tabs TBE DNA ladder  Equipment miniPCR® Phone Gel-electrophoresis & visualization system (e.g. blueGel™) 2-20 µl micropipette Trash bin  Consumables 20 µl filter tips Gloves



### **PCR** area – Materials



### PCR area - PCR

- 1. Load tubes into the miniPCR®
- 2. Run the specific PCR protocol







### PCR area – Gel Run

- 1. Prepare gel using SeeGreen™ tabs (reminder: completely dissolve the tab!)
- 2. Prepare 1X TBE buffer
- 3. Set up the gel electrophoresis device (if using blueGel™, add 30 ml buffer to the chamber)
- 4. Load 10 µl of ladder to the first well
- 5. Load 10 µl of each sample (reminder: change tip between samples!)
- 6. Run for 15-30 min
- 7. Take a picture and analyze the results
- Discard used material





