



LoopDeetect

Listeria monocytogenes

Gene amplification detection solution

Traditional protocols



User manual

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Definitions

DNA (Deoxyribonucleic Acid): A molecule composed of two complementary strands made up of nucleotides (A, T, C, G) that carry genetic information. The composition of the nucleotide sequence is specific to each species but varies among individuals. The DNA molecule forms a stable double helix that enables the replication and transmission of genetic information.

Primers: Short sequences of single-stranded oligonucleotides that are complementary to a target region. They initiate DNA synthesis by providing a starting point for the polymerase. They ensure the specificity of the amplification by specifically targeting the region of interest.

RNA (Ribonucleic Acid): A molecule that is generally single-stranded and composed of nucleotides (A, U, C, G). It plays a role in gene expression (mRNA, tRNA, rRNA) and certain catalytic functions. RNA molecules are generally present in greater quantities than DNA within the cell.

Control interface: LoopiX Cloud software or LoopiX App, a computer or smartphone solution for controlling the LoopiX® automated system

RT-LAMP (Reverse transcription Loop-mediated isothermal amplification): A molecular biology analytical technique for in vitro gene amplification, classified as an isothermal amplification method (commonly referred to as PCR at a constant temperature). It involves a set of 4 to 7 primers ensuring the specific detection of RNA with high sensitivity without thermal cycling. This isothermal amplification technique combines reverse transcription of RNA into cDNA followed by rapid amplification via a primer system and a specific polymerase.

Master Mix: A ready-to-use reaction mixture containing enzymes, dNTPs, buffer, and cofactors. Optimizes reproducibility and simplifies the preparation of amplification reactions.

PCR (Polymerase Chain Reaction): An analytical technique in molecular biology for in vitro gene amplification, the purpose of which is to replicate a portion of the genetic sequence of the target of interest in large numbers. It involves successive and repeated thermal cycles (denaturation, hybridization, and elongation by a DNA polymerase).

Preliminary information

This manual is the property of Loop Dee Science World Wide. Any copying or reproduction of this manual without the prior written permission of Loop Dee Science World Wide is prohibited. All users must read this manual carefully before use to familiarize themselves with all the information provided with the kits.

The kits offered by Loop Dee Science World Wide are molecular biology kits based on:

- Isothermal amplification technology - an innovative DNA and RNA amplification technology that enables the amplification and detection of target genetic material in a short analysis time.
- Magnetic bead technology - a technology that enables the capture and purification of genetic material through various washing buffers, thereby optimizing assay performance.

No claims or representations are intended to provide information for the diagnosis, prevention, or treatment of any disease.

To achieve optimal results, it is necessary to strictly follow the operating procedures and recommendations for use.

The kits in the LoopDeetect Food Safety range are validated and declared compliant by Loop Dee Science World Wide, solely in combination with the products, reagents, and devices recommended and tested by Loop Dee Science World Wide.

Test performance and validation

Performance data for the kit are available upon request.

AOAC PTM Validation: Certificate Number 082803

The LoopDeetect *Listeria monocytogenes* kit has been validated by the AOAC Research Institute under the Performance Tested Methods (PTM) program for the detection of *Listeria monocytogenes* using the following matrices and protocols:

- Stainless steel surface swabs with and without swab enrichment
- Plastic (polystyrene) surface swabs with and without swab enrichment

Validation of the LoopDeetect *Listeria monocytogenes* kits was performed using the Purification Kit Plus manual extraction solution, the Purifix® 1 and Purifix® 10 automated extraction solutions (and related kits), and the LoopiX® amplification instrument. A positive result obtained with the LoopDeetect *Listeria monocytogenes* solution should be considered presumptive. It is recommended to confirm it using a standardized reference method.

Description of symbols

Symbol	Descriptions
	View the user manual
	Manufacturer's name and address
	Date of manufacture
	Product batch number
	Expiration date
	Enough content for n tests
	Storage temperature limit
	Product code
	Does not tolerate moisture

	Store away from direct light and heat
	Single-use
	RUO (Research Use Only) device. Not for use in human diagnostics
	Sterilized by irradiation
	Sterilized with ethylene oxide
	Toxic, irritating, sensitizing, narcotic
	Flammable

Description of kits and packaging of packs

These kits are intended for professionals in the human and animal food industries, collectively referred to as the “food industry”, (producers, processors, distributors, and restaurant operators), as well as for food testing laboratories and hygiene specialists.

They are designed for use in self-monitoring, in accordance with current practices in the food industry. The LoopDetect method applied without enrichment is not intended to serve as a diagnostic method, but rather as a screening and decision-support tool. It is not designed to replace regulatory control methods, but to complement them

The kits are suitable for various types of applications:

- Surface swabbing.
- Confirmation of colonies from a Petri dish culture (*not included in validation study*)
- Analysis of food or environmental samples in culture media (*not included in validation study*)

Before use, it is essential to have read and understood all the instructions for the devices and products used during the protocol.

All instructions are available by following the link: loopdeescience.com/downloads

Sample collection kit

Kit	Kit content
<u>Surface Swab Kit with S1 Collection Tube (pack of 50)</u>	50 swabs 50 S1 sampling tubes
<u>S1 Surface Swab Kit (without swabs) (pack of 50)</u>	50 S1 sampling tubes

Purification kits

Purification kits enable the extraction of bacterial DNA and RNA from samples derived from environmental or food matrices (*not included in validation study*) for analysis using LoopDeetect detection kits.

They are based on an extraction technology that uses functionalized magnetic beads capable of capturing the genetic material present in the sample. The extraction protocol consists of four steps:

- **Lysis:** breaking down bacterial cell walls to release genetic material.
- **Binding:** capture of DNA/RNA by the functionalized beads.
- **Wash(es):** removal of impurities.
- **Elution:** recovery of genetic material.

Kit and description	Kit content				
<u>Purification kit Plus (PFS+) by 50</u> This kit allows for the manual extraction and purification of genetic material from swabs taken from environmental or food samples. <u>Catalog number</u> EXT2 B Ba 50	50 tubes of lysis buffer PFS+ L1 (Lysis buffer) 50 tubes of binding PFS+ B2 (Binding tubes) 50 tubes of wash buffer PFS+ W3 (Wash buffer) 50 tubes of wash buffer PFS+ W4 (Wash buffer) 50 tubes of wash buffer PFS+ W5 (Wash buffer) 50 tubes of elution buffer PFS+ E6 (Elution buffer) Liquids are transferred using two methods: <table border="1"> <tr> <td>Option 1</td><td>3 boxes of P1000 tips</td></tr> <tr> <td>Option 2</td><td>50 3.5-mL transfer pipettes 50 6-mL transfer pipettes 50 cotton swabs</td></tr> </table>	Option 1	3 boxes of P1000 tips	Option 2	50 3.5-mL transfer pipettes 50 6-mL transfer pipettes 50 cotton swabs
Option 1	3 boxes of P1000 tips				
Option 2	50 3.5-mL transfer pipettes 50 6-mL transfer pipettes 50 cotton swabs				
<u>PurifiX® 1 Purification kit Plus by 50</u> This kit enables the extraction and purification of genetic material from environmental or food matrices using the PurifiX® 1 purification system. <u>Catalog number</u> PFX1 P2 B Ba 50	50 tubes of lysis buffer PFS+ L1 (Lysis buffer) 50 PurifiX® 1 purification barrel 50 PurifiX® 1 tips				
<u>PurifiX® 8 Purification kit Plus by 50</u> This kit enables the extraction and purification of genetic material from environmental or food samples using the PurifiX® 8 purification system. <u>Catalog number</u> PFX8 P2 B Ba 50	50 tubes de tampon de lyse PFS+ L1 (Lysis buffer) 50 PurifiX® 8 purifications strips 28 PurifiX® 8 magnetic rods combs				
<u>PurifiX® 10 Purification kit Plus by 50</u> This kit enables the extraction and purification of genetic material from environmental or food samples using the PurifiX® 10 purification system. <u>Catalog number</u> PFX10 P2 B Ba 50	50 tubes de tampon de lyse PFS+ L1 (Lysis buffer) 50 PurifiX® 10 purifications strips 28 PurifiX® 10 magnetic rods combs				

LoopDeetect detection Kits

LoopDeetect kits enable the detection of a specific bacterial target in environmental (food industry surfaces) and food samples. When used with the LoopiX® automated system, they amplify a specific region of DNA or RNA from the target bacterium or bacterial genus via an isothermal reaction.

These kits are intended solely for qualitative analysis.

They come in the form of lyophilized reagents, packaged in a conical tube under a protective atmosphere.

Each tube contains:

- An amplification mastermix
- A set of primers specific to the target of interest
- A reaction protection system

LoopDeetect kits work exclusively with the LoopiX® automated system:

Kit Name	Kit content
LoopDeetect <i>L.monocytogenes</i> (by 50) Catalog number LMO LY 25	- 50 R1 reaction tubes (clear tube / gray pouch)
LoopDeetect <i>L.monocytogenes</i> (by 25) Catalog number LMO LY 50	- 25 R1 reaction tubes (clear tube / gray pouch)

Positive controls

The **positive controls** provided by Loop Dee Science consist of linear or plasmid DNA targeting the same region amplified by the corresponding LoopDeetect kits.

They are intended for use as external quality controls for the entire system (Extraction system manual or automatized, LoopiX® automated system + LoopDeetect kit).

Warning: Positive controls are not standard solutions; they should be treated as purified positive samples during handling.

Instructions for use:

- The positive control is compatible only with the corresponding LoopDeetect detection kit.
- It allows for validation of the entire analytical workflow.

We recommend using this control in the following cases:

- Upon receipt of each new batch of LoopDeetect kits,
- When qualifying a new LoopiX® automated system,
- For the certification of a new operator,
- As a periodic control, according to the frequency of use defined by the user,
- To validate the compatibility of different matrices with the method (Inhibition Control).

Positive control Name	Kit content	Related kit
LoopDeetect <i>Listeria</i> +	- 1 positive control (yellow tube / red bag) - 1 tube (S1)	LoopDeetect <i>L.monocytogenes</i> LoopDeetect <i>Listeria spp.</i>

Materials

Depending on the protocol being followed, certain equipment may be required:

Contact your distributor for information about the various types of equipment.

Application	Product identification
All applications	LoopiX® automated system
All applications	Fixed-volume 25µL pipette (or variable-volume 10–100µL pipette) Variable pipette 100–1000 µL

All applications	100 µL or 1000 µL filter tips (compatible)
Extraction	Magnetic rack
Extraction	PurifiX® 1 automated system
Extraction	PurifiX® 8 automated system
Extraction	PurifiX® 10 automated system

General Safety Precautions

Before handling the samples, please observe the following precautions to ensure reliable sample collection and handling. These precautions are essential to guarantee the quality of the analyses and must be followed at all times.

- Never mix components from different kits.
- Samples may contain potentially infectious biological agents; handle them with care.
- Certain chemicals in the purification kits pose risks. Safety data sheets (SDS) are available upon request. Always wear appropriate personal protective equipment (PPE): gloves, goggles, lab coat, and mask.

It is strongly recommended that you read the entire protocol and ensure that you fully understand it before conducting the tests.

The LoopDeetect *Listeria monocytogenes* kit enables the detection of *Listeria monocytogenes*, based on current knowledge.

Some matrices can inhibit amplification and require specific preparation (list available upon request - Contact us). It is strongly recommended that matrices be tested and validated before routine use.

Health and safety guidelines

- These kits are not intended for human diagnostic use.
- The products and the LoopiX® automated system must be used strictly in accordance with the user manuals. Loop Dee Science Worldwide assumes no liability for misuse or failure to follow the manufacturer's instructions.
- Before use, clean, disinfect, and carefully organize work surfaces.
- Follow laboratory hygiene protocols and implement a one-way workflow to prevent contamination.
- Strictly follow the kit usage instructions: storage conditions, sample type, order of steps, incubation times, and agitation and sampling times.
- Do not use expired kits (expiration date indicated on the label).

Sampling

- If the surface is damp, there is no need to moisten the swab.
- Ensure a large, swabbed area and use the entire swab by rotating it with your fingers while vigorously rubbing the surface to dislodge bacteria.
- After collection, analyse the samples as quickly as possible; otherwise, transport and store the samples in their original tubes at a temperature between +1°C and +5°C.

Lysis and purification

- Lysis buffer **L1** contains chaotropic salts, including guanidine hydrochloride (**toxic and irritating**).
- The lysed sample must be analysed within two hours.
- Homogenize the purified sample by suction-and-discharge before collection.
- Tube **B2** must remain open throughout the procedure, particularly during the drying step.

- **B2** tubes contain chaotropic salts and ethanol (**toxic and flammable**).
- The wash buffers in the purification kits also contain ethanol (**flammable**).
- After extraction, analyse the samples promptly. For long-term storage, collect the eluate and store it in a clean 1.5 mL tube at -20°C.

Preparation of the reaction tube

- Do not open the packet containing the lyophilized powder or positive control until you are ready to use it.
- Check that the lyophilized powder pellet is at the bottom of the tube. If necessary, tap the tube gently.
- Ensure that the tube and its contents are intact:
- Do not use if the pouch is damaged (torn, poorly sealed).
- Do not use if the contents have a pinkish color or are sticky.
- Never store lyophilized or dehydrated tubes outside their original pouch.

Analysis

- Place the 25 µL sample directly onto the pad at the bottom of the tube, ensuring there are no air bubbles. Gently tap the tube if necessary to remove any bubbles.
- During the test, never unplug, open, or interrupt the LoopiX® before the process is complete.
- A reaction tube must never be used for a second analysis, as this may produce a false result. If necessary, repeat the procedure starting from the sample collection.
- After use, never open the reaction tube to avoid contaminating the work environment and the risk of errors in future analyses.
- At the end of the analysis, immediately discard the reaction tube and any used consumables to prevent contamination.
- At the end of the analysis, clean all used items (equipment, work area, devices, etc.) using an alcohol-based solution and an absorbent cloth.

Materials

- Regularly check the condition and volume accuracy of your pipettes. Replace them if they are damaged (due to impact or a fall) or if you have any doubts.
- Never perform an analysis with an empty LoopiX® or an empty tube, as this may damage the device.
- Never perform a purification cycle with a device lacking protective consumables, as this may damage the device's magnetic rods.
- Perform all procedures at a temperature between +15°C and +25°C.
- Reagents are for single use only and must be disposed of after use (see the "Disposal - Waste Management" section).

Protocols

Before you begin, please read the “General Handling Precautions” carefully to ensure proper sample collection and handling.

It is strongly recommended that you read the entire protocol and ensure that you fully understand it before proceeding with the tests. The following table lists the various protocols that can be used depending on the application and the validation area:

	Purification PFS+	Purification: PurifiX® Range
AOAC PTM 082603		
Surface swabbing of stainless steel without enrichment	SWA_01	SWA_02
Surface swabbing of plastic (polystyrene) without enrichment	SWA_01	SWA_02
Surface swabbing of stainless steel with enrichment	ENR_01	ENR_02
Surface swabbing of plastic (polystyrene) with enrichment	ENR_01	ENR_02
Surface swabbing	SWA_01	SWA_02
Food or environmental sample with enrichment*	MIL_01	MIL_02
Colony from a Petri dish culture*	COL_01	COL_02
Negative control	NEG_01	
Positive control	POS_01	
Inhibition control	INH_01	

*(not included in validation study)

SWA_01 - Surface swabbing without enrichment / Manual PFS+ purification

Sampling

- Moisten the swab with buffer S1.
- Collect the surface sample using the swab.
 - In the context of AOAC validation, it is recommended to perform double swabbing of surfaces in order to confirm presumptive positive results.
 - Collect both samples at the same time but store the second sample between 2°C and 8°C.
 - Proceed with confirmation as soon as a positive result is obtained.
- Immerse the swab head in tube **L1** and release the sample contents by rotating and moving the swab back and forth for 15 seconds, then press the swab against the side of the tube to extract the liquid.

Lysis

- Close tube **L1** and place it in the LoopiX®.
- Using the control interface, start a “Lysis and Amplification” run. The indicator light flashes red throughout the lysis process.
- At the end of lysis, the indicator light flashes blue. Remove tube **L1** from the LoopiX®.
- Using a pipette with a tip or a 6 mL transfer pipette, aspirate the entire contents of tube **L1**.
- Fermer le tube **L1** et le placer dans le LoopiX®.

Purification using the PFS+ kit

- Transfer the lysate to tube **B2** using a 6-mL pipette or a pipette fitted with a tip.
- Homogenize with 15 up-and-down pipetting strokes, ensuring that the beads are evenly distributed without clumping.
- Wait 1 minute, then place tube **B2** on the magnetic stand to capture the beads.
- Discard the supernatant without touching the beads, leaving **B2** on the stand.
- Add buffer **W3** and homogenize again.
- Place tube **B2** on the magnetic stand to capture the beads.
- Discard the supernatant without touching the beads; tube **B2** remains on the stand.
- Add buffer **W4** and repeat the steps for homogenization, bead capture, and supernatant removal.
- Add buffer **W5** and repeat the same steps for homogenization, bead capture, and supernatant removal.
- Dry the bottom of the tube with a Lemoine swab or a pipette fitted with a new tip (nothing should remain at the bottom of the tube), then discard the swab or tip.
- Let air dry for 5 minutes with tube **B2** open.
- Using a 3.5 mL pipette or a pipette fitted with a new tip, add buffer **E6** and homogenize. Wait 1 minute.
- Place tube **B2** on the magnetic stand to capture the beads.
- Using a pipette fitted with a new tip, withdraw 25 µL from tube **B2** without touching the beads.
 - To confirm positive results, it is recommended that you transfer the entire sample to a clean, properly labeled 1.5-mL tube.
 - Store this sample between 2°C and 8°C for use within the same day. Store the sample at -18°C for long-term storage.

Analysis

- Transfer the 25 µL to reaction tube **R1**.
- Close tube **R1** by pressing down with your finger until you hear a “click” indicating that it is securely closed, then place it in the LoopiX®.
- Start the analysis by pressing the button on the front panel of the LoopiX® or via the control interface.
- At the end of the analysis, read the result directly from the status LED and the control interface:
 - **Red LED**: positive result – *Listeria monocytogenes* detected
 - **Green LED**: negative result – *Listeria monocytogenes* not detected

Confirmation of positive results

In the context of AOAC validation, a positive LoopDeetect *Listeria monocytogenes* result is considered a presumptive positive. It is recommended to confirm the result using an appropriate reference method. Alternatively, the result can be confirmed directly from the extract using an appropriately validated PCR method. Before using a validated method (e.g. : iq-check *Listeria monocytogenes*, ez-check *Listeria monocytogenes*, *Listeria monocytogenes*), please concentrate the DNA using, for example, a MWCO-type membrane.

- Load the DNA eluate into the filter tube
- Centrifuge at 14,000 × g for 5 minutes
- Discard the filtrate.
- Add 50 µL of the desired TE buffer to the filter and re-centrifuge for 5 minutes
- Recovery: invert the filter unit into a clean tube and centrifuge briefly (2-3 min, 6,000 g) to collect the concentrated retentate.

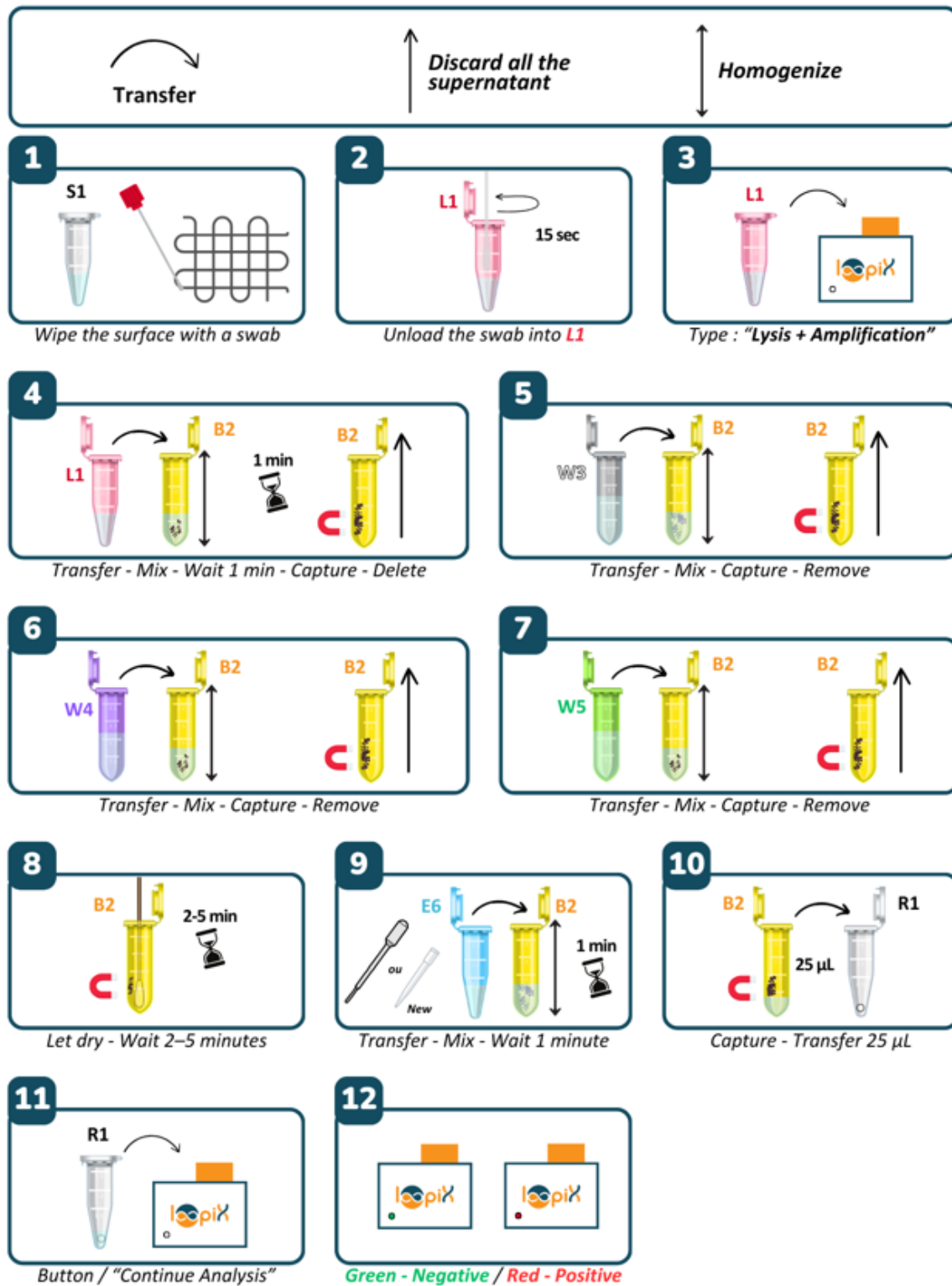
Follow the amplification instructions of the chosen manufacturer.

End of analysis

- Discard the **R1** reaction tube and **NEVER** open it.
- Discard all consumables used for the analysis.
- Using an alcohol-based solution and an absorbent cloth, clean the work area, tools (pipettes, stands, etc.), and devices (LoopiX®, PurifiX®, etc.).
- Perform a 30-minute UV sterilization cycle for devices equipped with this feature.

SWA_01

Surface swabbing without enrichment / Manual PFS+ purification+



SWA_02 - Surface swabbing without enrichment / Purifix® Line

Sampling

- Moisten the swab with buffer S1.
- Collect the surface sample using the swab.
 - In the context of AOAC validation, it is recommended to perform double swabbing of surfaces in order to confirm presumptive positive results.
 - Collect both samples at the same time but store the second sample between 2°C and 8°C.
 - Proceed with confirmation as soon as a positive result is obtained.
- Immerse the swab head in tube **L1** and release the sample contents by rotating and moving the swab back and forth for 15 seconds, then press the swab against the side of the tube to extract the liquid.

Lysis

Lysis with LoopiX :

- Close tube **L1** and place it in the LoopiX®.
- Using the control interface, start a “Lysis and Amplification” run. The indicator light flashes red throughout the lysis process.
- At the end of lysis, the indicator light flashes blue. Remove tube **L1** from the LoopiX®.
- Using a pipette with a tip or a 6 mL transfer pipette, aspirate the entire contents of tube **L1**.
- Fermer le tube **L1** et le placer dans le LoopiX®.

Lysis with dry bath:

The Purifix® 10 comes with a built-in heating element that is ready for immediate use. For other machines in the Purifix® series, a separate heating element is required.

- Seal tube **L1** and place it in the heating block preheated to **95°C**.
- Start the lysis for **5 minutes**.
- At the end of the lysis, remove tube **L1** from the heating block and wait 2 minutes for it to cool.
- Using a pipette with a tip or a 6-mL transfer pipette, aspirate the entire contents of tube **L1**.

Purification using a PurifiX® series automated system

- Transfer the lysate to **well 1** of the PurifiX® strip using a 6-mL transfer pipette or a pipette fitted with a tip.
- Place the purification strip into the PurifiX®:
 - PurifiX® 1: Place the cartridge into the cup, then place the cup into the PurifiX® 1.
 - PurifiX® 8: Place the strip on the rack, then place the rack in the PurifiX® 8.
 - PurifiX® 10: Place the purification strip directly into the PurifiX® 10.
- Place the comb(s) into the device, making sure to push them all the way in.
- Close the device lid and start the run using the control interface.
- At the end of purification, discard the comb(s) and remove the purification strip.
- Using a pipette fitted with a new tip, aspirate **25 µL** from the last well of the strip (PurifiX® and PurifiX® 10: **well 8** / PurifiX® 8: **well 6**) and proceed with the analysis.
 - To confirm positive results, it is recommended to transfer the entire extract to a clean, properly labeled 1.5-mL tube.

- Store this extract between 2°C and 8°C for use within the same day. Store the extract at -18°C for longer-term use.

Analysis

- Transfer the 25 µL to reaction tube **R1**.
- Close tube **R1** by pressing down with your finger until you hear a “click” indicating that it is securely closed, then place it in the LoopiX®.
- Start the analysis by pressing the button on the front panel of the LoopiX® or via the control interface.
- At the end of the analysis, read the result directly from the status LED and the control interface:
 - **Red LED:** positive result – *Listeria monocytogenes* detected
 - **Green LED:** negative result – *Listeria monocytogenes* not detected

Confirmation of positive results

In the context of AOAC validation, a positive LoopDeetect *Listeria monocytogenes* result is considered a presumptive positive. It is recommended to confirm the result using an appropriate reference method. Alternatively, the result can be confirmed directly from the extract using an appropriately validated PCR method. Before using a validated method (e.g. : iq-check *Listeria monocytogenes*, ez-check *Listeria monocytogenes*, *Listeria monocytogenes*), please concentrate the DNA using, for example, a MWCO-type membrane.

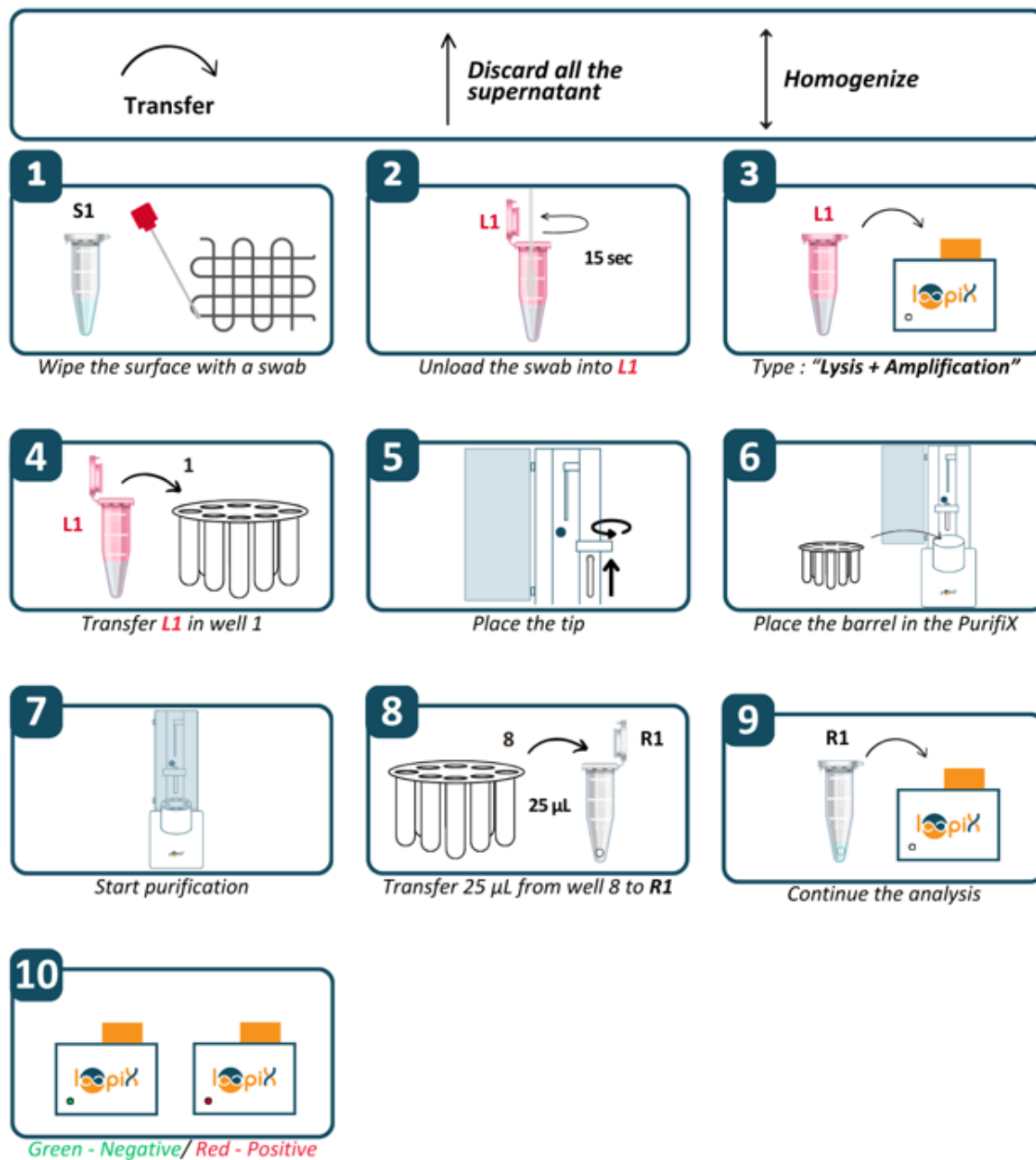
- Load the DNA eluate into the filter tube
- Centrifuge at 14,000 × g for 5 minutes
- Discard the filtrate.
- Add 50 µL of the desired TE buffer to the filter and re-centrifuge for 5 minutes
- Recovery: invert the filter unit into a clean tube and centrifuge briefly (2-3 min, 6,000 g) to collect the concentrated retentate.

Follow the amplification instructions of the chosen manufacturer.

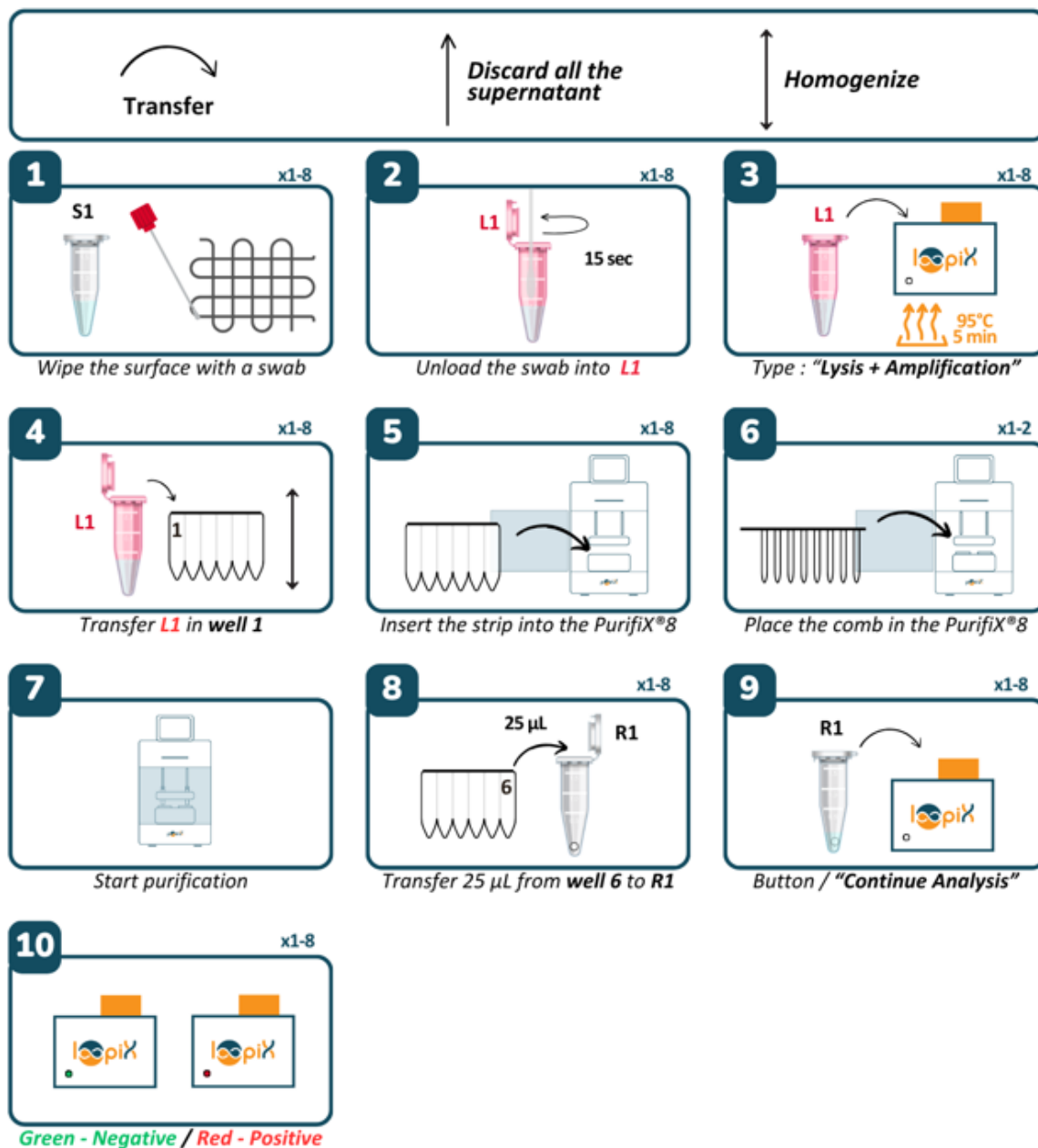
End of analysis

- Discard the **R1** reaction tube and **NEVER** open it.
- Discard all consumables used for the analysis.
- Using an alcohol-based solution and an absorbent cloth, clean the work area, tools (pipettes, stands, etc.), and devices (LoopiX®, PurifiX®, etc.).
- Perform a 30-minute UV sterilization cycle for devices equipped with this feature.

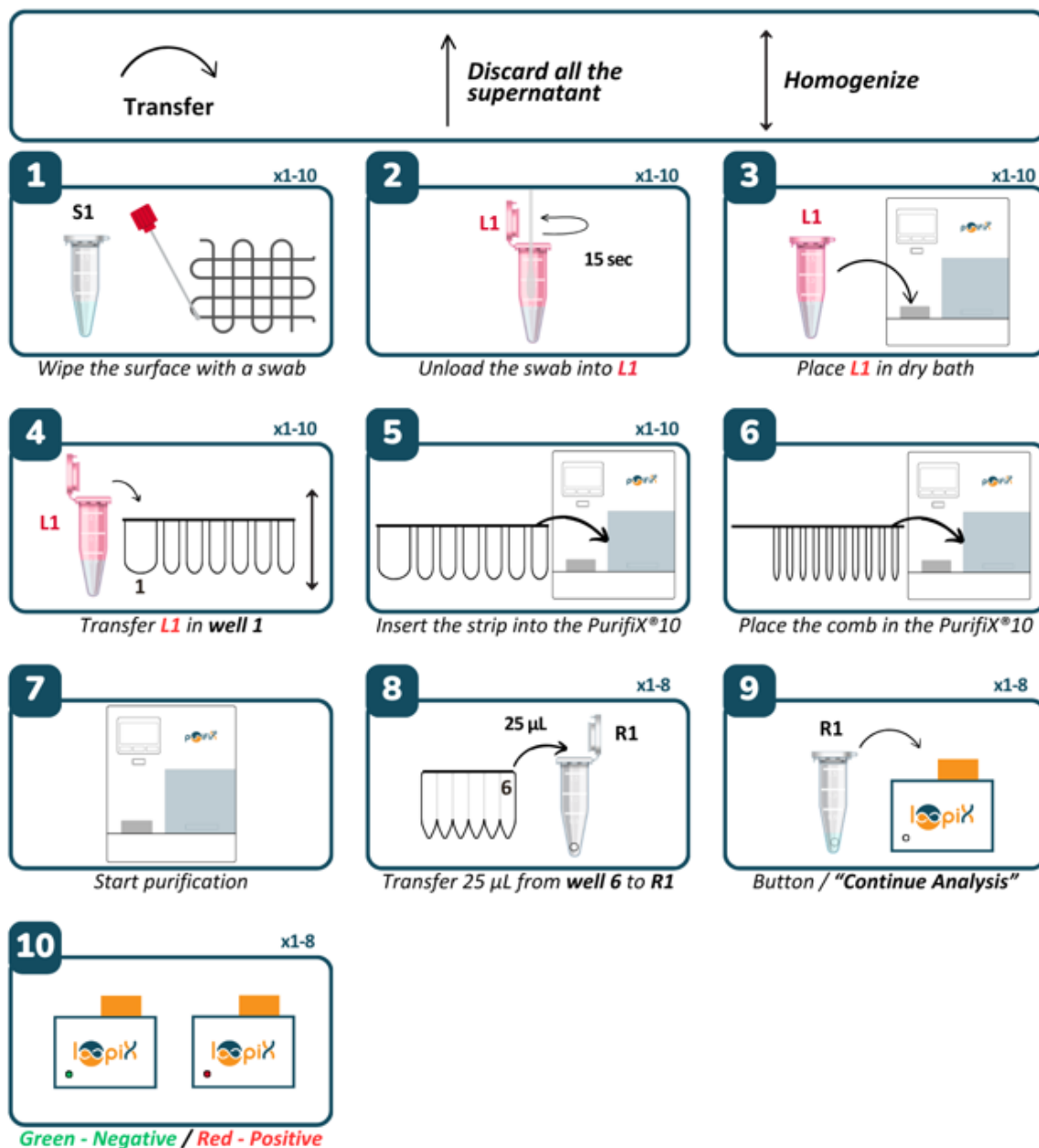
SWA_02 Surface swabbing without enrichment / PurifiX®1



SWA_02 Surface swabbing without enrichment / PurifiX®8



SWA_02 Surface swabbing without enrichment / PurifiX®10



ENR_01 - Surface swabbing after enrichment / Manual PFS+ purification

Sampling

- Moisten the swab with buffer **S1**.
- Collect the surface sample using the swab.
- Place the swab in a 10-mL Fraser ½ tube.
- Incubate at 30+/- 1°C for 25 hours (+/- 1 hour)
- Transfer 100 µL to a 10 mL Fraser tube.
- Incubate at 37 +/- 1°C for 24 hours (+/- 1 hour).
- Homogenize the sample, then transfer **200 µL** to the **L1** lysis tube.

Lysis

- Close tube **L1** and place it in the LoopiX®.
- Using the control interface, start a “Lysis and Amplification” run. The indicator light flashes red throughout the lysis process.
- At the end of lysis, the indicator light flashes blue. Remove tube **L1** from the LoopiX®.
- Using a pipette with a tip or a 6 mL transfer pipette, aspirate the entire contents of tube **L1**
- Fermer le tube **L1** et le placer dans le LoopiX®.

Purification using the PFS+ kit

- Transfer the lysate to tube **B2** using a 6-mL pipette or a pipette fitted with a tip.
- Homogenize with 15 up-and-down pipetting strokes, ensuring that the beads are evenly distributed without clumping.
- Wait 1 minute, then place tube **B2** on the magnetic stand to capture the beads.
- Discard the supernatant without touching the beads, leaving **B2** on the stand.
- Add buffer **W3** and homogenize again.
- Place tube **B2** on the magnetic stand to capture the beads.
- Discard the supernatant without touching the beads; tube **B2** remains on the stand.
- Add buffer **W4** and repeat the steps for homogenization, bead capture, and supernatant removal.
- Add buffer **W5** and repeat the same steps for homogenization, bead capture, and supernatant removal.
- Dry the bottom of the tube with a Lemoine swab or a pipette fitted with a new tip (nothing should remain at the bottom of the tube), then discard the swab or tip.
- Let air dry for 5 minutes with tube **B2** open.
- Using a 3.5 mL pipette or a pipette fitted with a new tip, add buffer **E6** and homogenize. Wait 1 minute.
- Place tube **B2** on the magnetic stand to capture the beads.
- Using a pipette fitted with a new tip, withdraw 25 µL from tube **B2** without touching the beads.
 - To confirm positive results, it is recommended that you transfer the entire sample to a clean, properly labeled 1.5-mL tube.
 - Store this sample between 2°C and 8°C for use within the same day. Store the sample at -18°C for long-term storage.

Analysis

- Transfer the 25 µL to reaction tube **R1**.
- Close tube **R1** by pressing down with your finger until you hear a “click” indicating that it is securely closed, then place it in the LoopiX®.
- Start the analysis by pressing the button on the front panel of the LoopiX® or via the control interface.
- At the end of the analysis, read the result directly from the status LED and the control interface:
 - **Red LED**: positive result – *Listeria monocytogenes* detected
 - **Green LED**: negative result – *Listeria monocytogenes* not detected

Confirmation of positive results

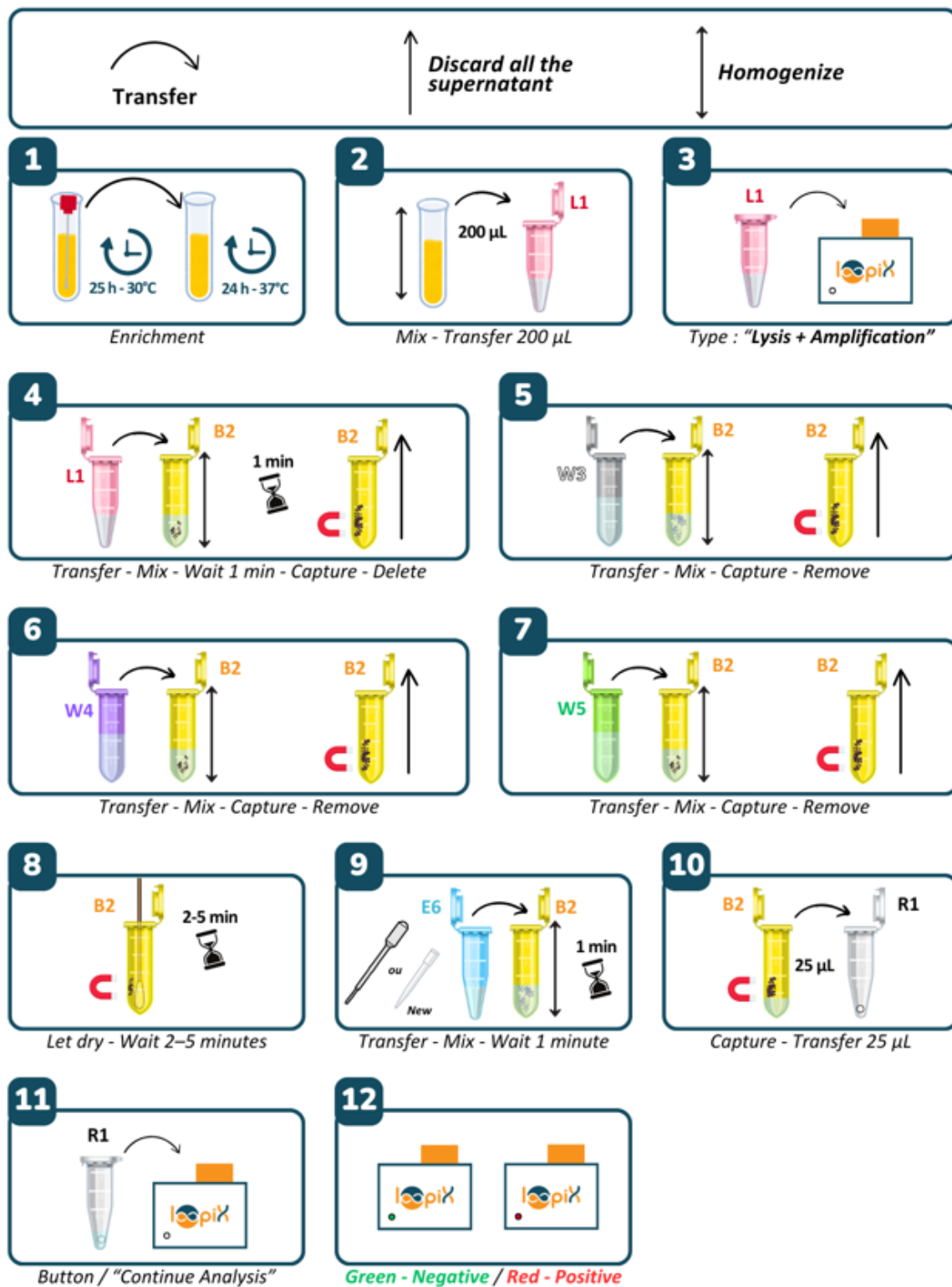
In the context of AOAC validation, a positive LoopDeetect *Listeria monocytogenes* result is considered a presumptive positive. It is recommended to confirm the result using an appropriate reference method. Alternatively, the result can be confirmed directly from the extract using an appropriately validated PCR method. Before using a validated method (e.g. : iq-check *Listeria monocytogenes*, ez-check *Listeria monocytogenes*, *Listeria monocytogenes*), please concentrate the DNA using, for example, a MWCO-type membrane.

- Load the DNA eluate into the filter tube
- Centrifuge at 14,000 × g for 5 minutes
- Discard the filtrate.
- Add 50 µL of the desired TE buffer to the filter and re-centrifuge for 5 minutes
- Recovery: invert the filter unit into a clean tube and centrifuge briefly (2-3 min, 6,000 g) to collect the concentrated retentate.

Follow the amplification instructions of the chosen manufacturer.

End of analysis

- Discard the **R1** reaction tube and **NEVER** open it.
- Discard all consumables used for the analysis.
- Using an alcohol-based solution and an absorbent cloth, clean the work area, tools (pipettes, stands, etc.), and devices (LoopiX®, PurifiX®, etc.).
- Perform a 30-minute UV sterilization cycle for devices equipped with this feature.



ENR_02 - Surface swabbing after enrichment / Purifix® Line

Sampling

- Moisten the swab with buffer **S1**.
- Collect the surface sample using the swab.
- Place the swab in a 10-mL Fraser ½ tube.
- Incubate at 30+/- 1°C for 25 hours (+/- 1 hour)
- Transfer 100 µL to a 10 mL Fraser tube.
- Incubate at 37+/- 1°C for 24 hours (+/- 1 hour).
- Homogenize the sample, then transfer **200 µL** to the **L1** lysis tube.

Lysis

Lysis with LoopiX :

- Close tube **L1** and place it in the LoopiX®.
- Using the control interface, start a “Lysis and Amplification” run. The indicator light flashes red throughout the lysis process.
- At the end of lysis, the indicator light flashes blue. Remove tube **L1** from the LoopiX®.
- Using a pipette with a tip or a 6 mL transfer pipette, aspirate the entire contents of tube **L1**
- Fermer le tube **L1** et le placer dans le LoopiX®.

Lysis with dry bath:

The Purifix® 10 comes with a built-in heating element that is ready for immediate use. For other machines in the Purifix® series, a separate heating element is required.

- Seal tube **L1** and place it in the heating block preheated to **95°C**.
- Start the lysis for **5 minutes**.
- At the end of the lysis, remove tube **L1** from the heating block and wait 2 minutes for it to cool.
- Using a pipette with a tip or a 6-mL transfer pipette, aspirate the entire contents of tube **L1**

Purification using a PurifiX® series automated system

- Transfer the lysate to **well 1** of the PurifiX® strip using a 6-mL transfer pipette or a pipette fitted with a tip.
- Place the purification strip into the PurifiX®:
 - PurifiX® 1: Place the cartridge into the cup, then place the cup into the PurifiX® 1.
 - PurifiX® 8: Place the strip on the rack, then place the rack in the PurifiX® 8.
 - PurifiX® 10: Place the purification strip directly into the PurifiX® 10.
- Place the comb(s) into the device, making sure to push them all the way in.
- Close the device lid and start the run using the control interface.
- At the end of purification, discard the comb(s) and remove the purification strip.
- Using a pipette fitted with a new tip, aspirate **25 µL** from the last well of the strip (PurifiX® and PurifiX® 10: **well 8** / PurifiX® 8: **well 6**) and proceed with the analysis.
 - To confirm positive results, it is recommended to transfer the entire extract to a clean, properly labeled 1.5-mL tube.
 - Store this extract between 2°C and 8°C for use within the same day. Store the extract at -18°C for longer-term use.

Analysis

- Transfer the 25 µL to reaction tube **R1**.
- Close tube **R1** by pressing down with your finger until you hear a “click” indicating that it is securely closed, then place it in the LoopiX®.
- Start the analysis by pressing the button on the front panel of the LoopiX® or via the control interface.
- At the end of the analysis, read the result directly from the status LED and the control interface:
 - **Red LED**: positive result – *Listeria monocytogenes* detected
 - **Green LED**: negative result – *Listeria monocytogenes* not detected

Confirmation of positive results

In the context of AOAC validation, a positive LoopDeetect *Listeria monocytogenes* result is considered a presumptive positive. It is recommended to confirm the result using an appropriate reference method. Alternatively, the result can be confirmed directly from the extract using an appropriately validated PCR method. Before using a validated method (e.g. : iq-check *Listeria monocytogenes*, ez-check *Listeria monocytogenes*, *Listeria monocytogenes*), please concentrate the DNA using, for example, a MWCO-type membrane.

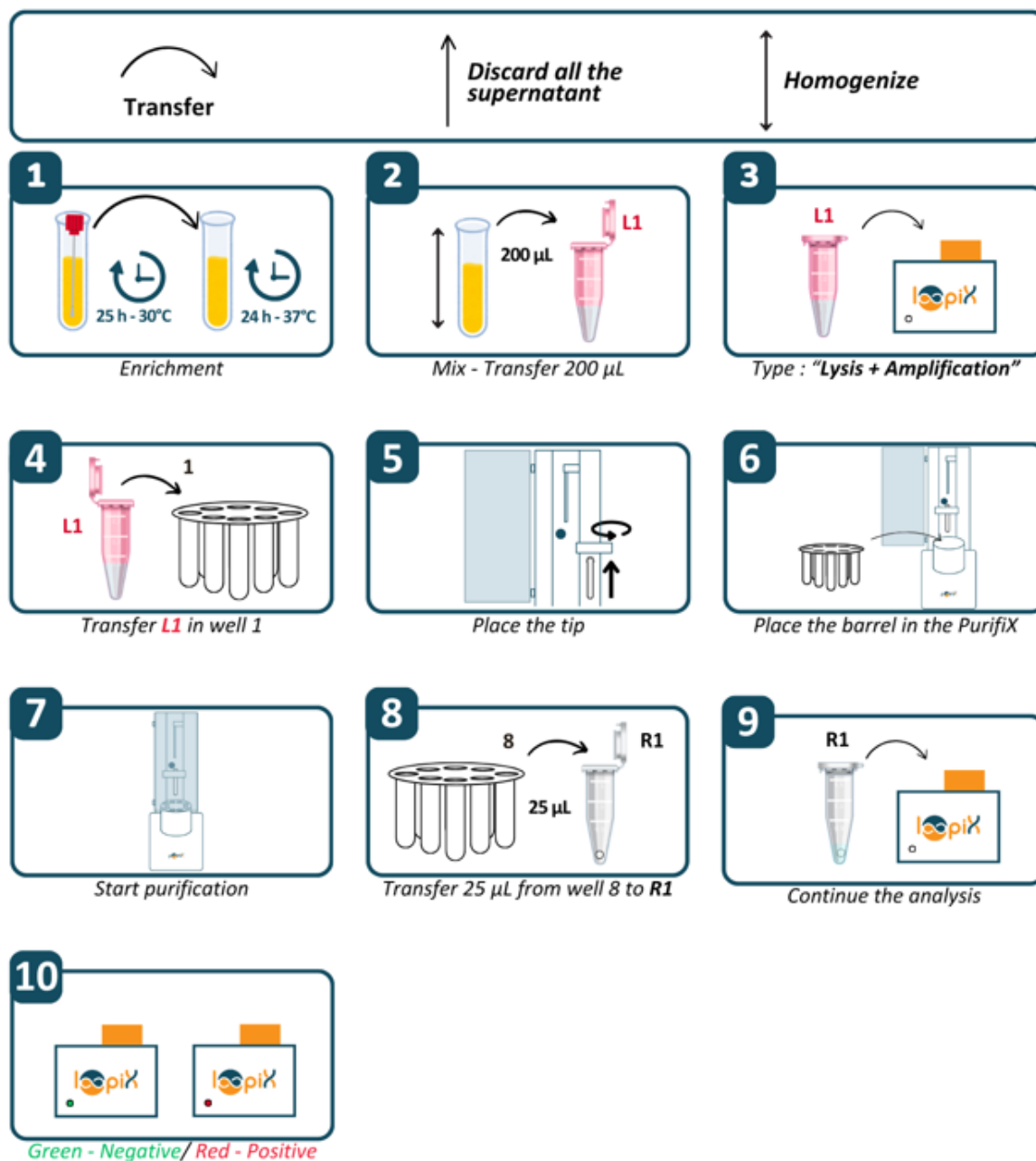
- Load the DNA eluate into the filter tube
- Centrifuge at 14,000 × g for 5 minutes
- Discard the filtrate.
- Add 50 µL of the desired TE buffer to the filter and re-centrifuge for 5 minutes
- Recovery: invert the filter unit into a clean tube and centrifuge briefly (2-3 min, 6,000 g) to collect the concentrated retentate.

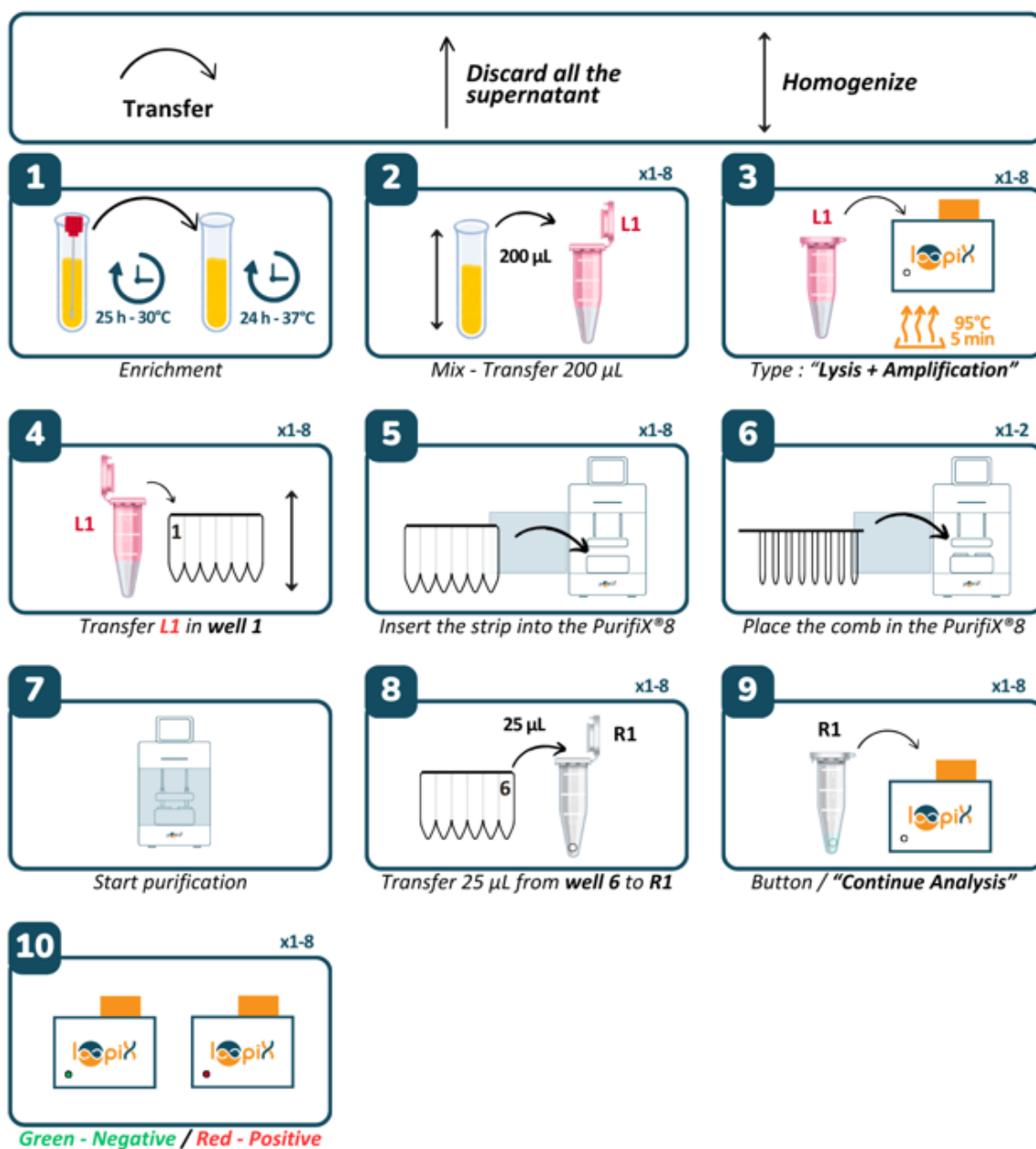
Follow the amplification instructions of the chosen manufacturer.

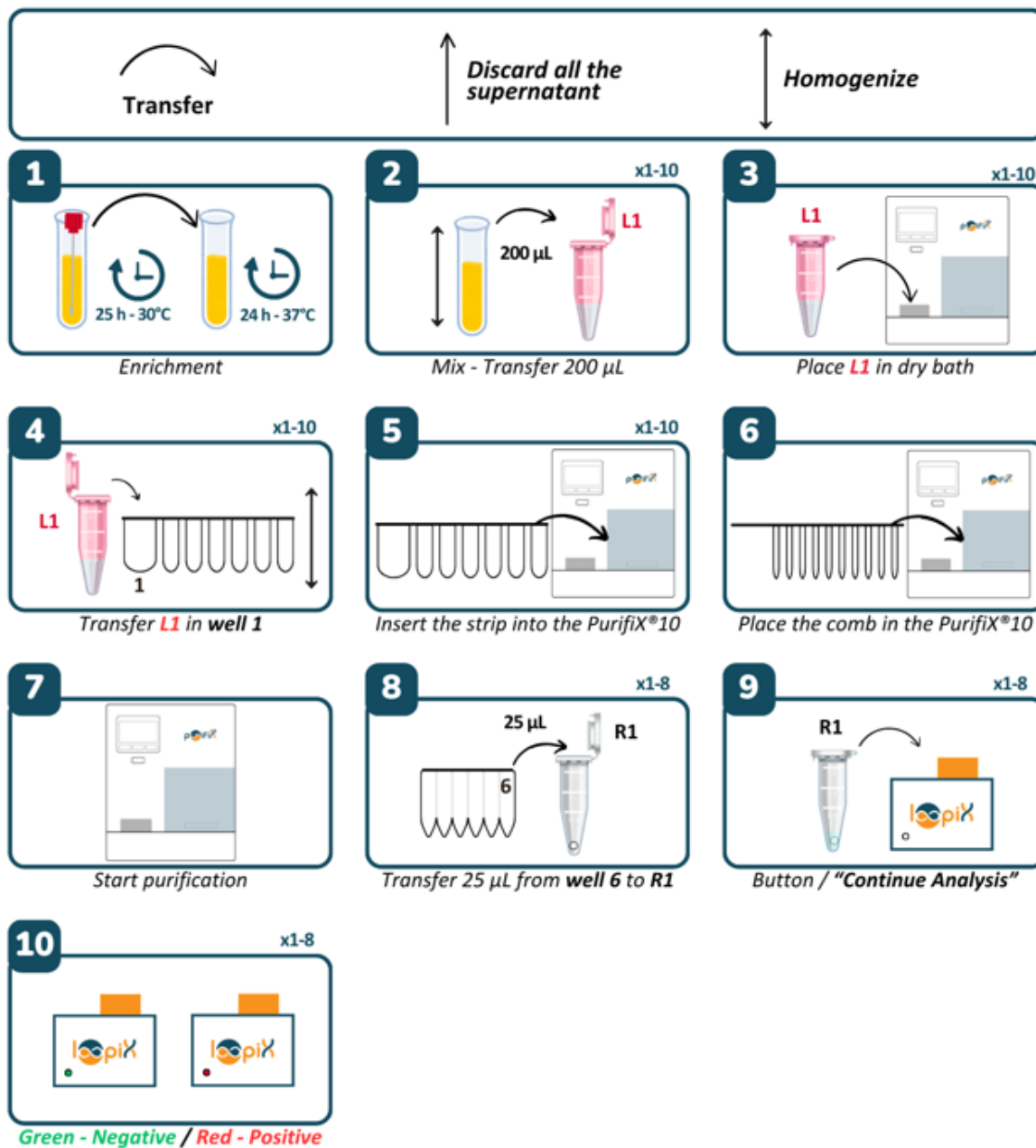
End of analysis

- Discard the **R1** reaction tube and **NEVER** open it.
- Discard all consumables used for the analysis.
- Using an alcohol-based solution and an absorbent cloth, clean the work area, tools (pipettes, stands, etc.), and devices (LoopiX®, PurifiX®, etc.).
- Perform a 30-minute UV sterilization cycle for devices equipped with this feature.

ENR_02 Surface swabbing after enrichment / PurifiX®1







MIL_01 - Sample after enrichment (excluding swabs) / Manual PFS+ purification

Sampling

- Prepare the sample for analysis in accordance with applicable standards or the company's internal procedures (enrichment or revitalization if necessary), using filter bags to retain solid particles whenever possible.
- Homogenize the sample, then transfer **200 µL** to lysis tube **L1**

Lysis

- Close tube **L1** and place it in the LoopiX®.
- Using the control interface, start a "Lysis and Amplification" run. The indicator light flashes red throughout the lysis process.
- At the end of lysis, the indicator light flashes blue. Remove tube **L1** from the LoopiX®.
- Using a pipette with a tip or a 6 mL transfer pipette, aspirate the entire contents of tube **L1**
- Fermer le tube **L1** et le placer dans le LoopiX®.

Purification using the PFS+ kit

- Transfer the lysate to tube **B2** using a 6-mL pipette or a pipette fitted with a tip.
- Homogenize with 15 up-and-down pipetting strokes, ensuring that the beads are evenly distributed without clumping.
- Wait 1 minute, then place tube **B2** on the magnetic stand to capture the beads.
- Discard the supernatant without touching the beads, leaving **B2** on the stand.
- Add buffer **W3** and homogenize again.
- Place tube **B2** on the magnetic stand to capture the beads.
- Discard the supernatant without touching the beads; tube **B2** remains on the stand.
- Add buffer **W4** and repeat the steps for homogenization, bead capture, and supernatant removal.
- Add buffer **W5** and repeat the same steps for homogenization, bead capture, and supernatant removal.
- Dry the bottom of the tube with a Lemoine swab or a pipette fitted with a new tip (nothing should remain at the bottom of the tube), then discard the swab or tip.
- Let air dry for 5 minutes with tube **B2** open.
- Using a 3.5 mL pipette or a pipette fitted with a new tip, add buffer **E6** and homogenize. Wait 1 minute.
- Place tube **B2** on the magnetic stand to capture the beads.
- Using a pipette fitted with a new tip, withdraw 25 µL from tube **B2** without touching the beads.
 - To confirm positive results, it is recommended that you transfer the entire sample to a clean, properly labeled 1.5-mL tube.
 - Store this sample between 2°C and 8°C for use within the same day. Store the sample at -18°C for long-term storage.

Analysis

- Transfer the 25 µL to reaction tube **R1**.
- Close tube **R1** by pressing down with your finger until you hear a "click" indicating that it is securely closed, then place it in the LoopiX®.

- Start the analysis by pressing the button on the front panel of the LoopiX® or via the control interface.
- At the end of the analysis, read the result directly from the status LED and the control interface:
 - **Red LED:** positive result – *Listeria monocytogenes* detected
 - **Green LED:** negative result – *Listeria monocytogenes* not detected

Confirmation of positive results

In the context of AOAC validation, a positive LoopDeetect *Listeria monocytogenes* result is considered a presumptive positive. It is recommended to confirm the result using an appropriate reference method. Alternatively, the result can be confirmed directly from the extract using an appropriately validated PCR method. Before using a validated method (e.g. : iq-check *Listeria monocytogenes*, ez-check *Listeria monocytogenes*, *Listeria monocytogenes*), please concentrate the DNA using, for example, a MWCO-type membrane.

- Load the DNA eluate into the filter tube
- Centrifuge at 14,000 × g for 5 minutes
- Discard the filtrate.
- Add 50 µL of the desired TE buffer to the filter and re-centrifuge for 5 minutes
- Recovery: invert the filter unit into a clean tube and centrifuge briefly (2-3 min, 6,000 g) to collect the concentrated retentate.

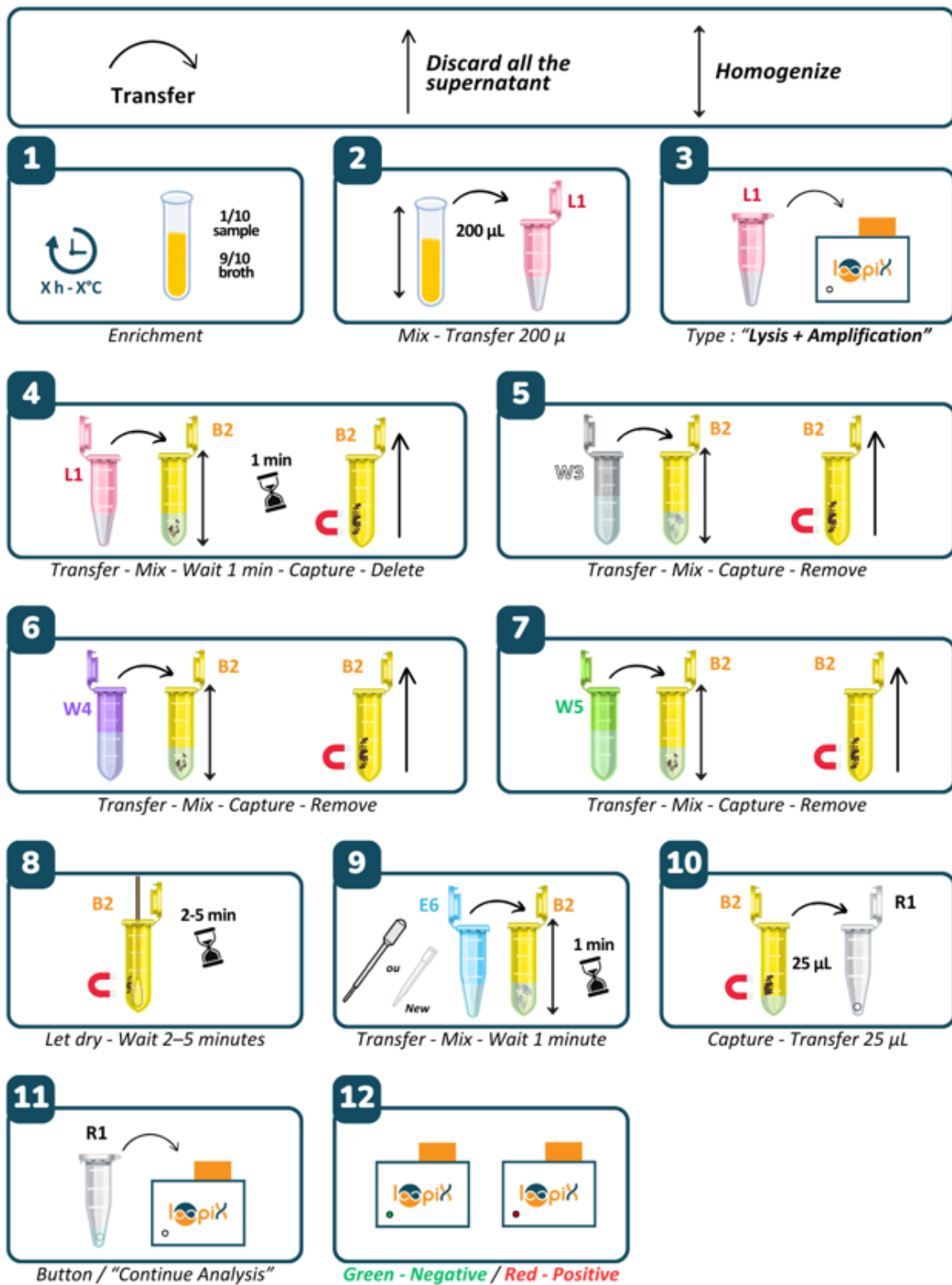
Follow the amplification instructions of the chosen manufacturer.

End of analysis

- Discard the **R1** reaction tube and **NEVER** open it.
- Discard all consumables used for the analysis.
- Using an alcohol-based solution and an absorbent cloth, clean the work area, tools (pipettes, stands, etc.), and devices (LoopiX®, PurifiX®, etc.).
- Perform a 30-minute UV sterilization cycle for devices equipped with this feature.

MIL_01

Sample after enrichment (excluding swabs) / Manual PFS+ purification



MIL_02 - Sample after enrichment (excluding swabs) / Purification: Purifix® Line

Sampling

- Prepare the sample for analysis in accordance with applicable standards or the company's internal procedures (enrichment or revitalization if necessary), using filter bags to retain solid particles whenever possible.
- Homogenize the sample, then transfer **200 µL** to lysis tube **L1**

Lysis

Lysis with LoopiX :

- Close tube **L1** and place it in the LoopiX®.
- Using the control interface, start a "Lysis and Amplification" run. The indicator light flashes red throughout the lysis process.
- At the end of lysis, the indicator light flashes blue. Remove tube **L1** from the LoopiX®.
- Using a pipette with a tip or a 6 mL transfer pipette, aspirate the entire contents of tube **L1**
- Fermer le tube **L1** et le placer dans le LoopiX®.

Lysis with dry bath:

The Purifix® 10 comes with a built-in heating element that is ready for immediate use. For other machines in the Purifix® series, a separate heating element is required.

- Seal tube **L1** and place it in the heating block preheated to **95°C**.
- Start the lysis for **5 minutes**.
- At the end of the lysis, remove tube **L1** from the heating block and wait 2 minutes for it to cool.
- Using a pipette with a tip or a 6-mL transfer pipette, aspirate the entire contents of tube **L1**

Purification using a PurifiX® series automated system

- Transfer the lysate to **well 1** of the PurifiX® strip using a 6-mL transfer pipette or a pipette fitted with a tip.
- Place the purification strip into the PurifiX®:
 - PurifiX® 1: Place the cartridge into the cup, then place the cup into the PurifiX® 1.
 - PurifiX® 8: Place the strip on the rack, then place the rack in the PurifiX® 8.
 - PurifiX® 10: Place the purification strip directly into the PurifiX® 10.
- Place the comb(s) into the device, making sure to push them all the way in.
- Close the device lid and start the run using the control interface.
- At the end of purification, discard the comb(s) and remove the purification strip.
- Using a pipette fitted with a new tip, aspirate **25 µL** from the last well of the strip (PurifiX® and PurifiX® 10: **well 8** / PurifiX® 8: **well 6**) and proceed with the analysis.
 - To confirm positive results, it is recommended to transfer the entire extract to a clean, properly labeled 1.5-mL tube.
 - Store this extract between 2°C and 8°C for use within the same day. Store the extract at -18°C for longer-term use.

Analysis

- Transfer the 25 µL to reaction tube **R1**.

- Close tube **R1** by pressing down with your finger until you hear a “click” indicating that it is securely closed, then place it in the LoopiX®.
- Start the analysis by pressing the button on the front panel of the LoopiX® or via the control interface.
- At the end of the analysis, read the result directly from the status LED and the control interface:
 - **Red LED:** positive result – *Listeria monocytogenes* detected
 - **Green LED:** negative result – *Listeria monocytogenes* not detected

Confirmation of positive results

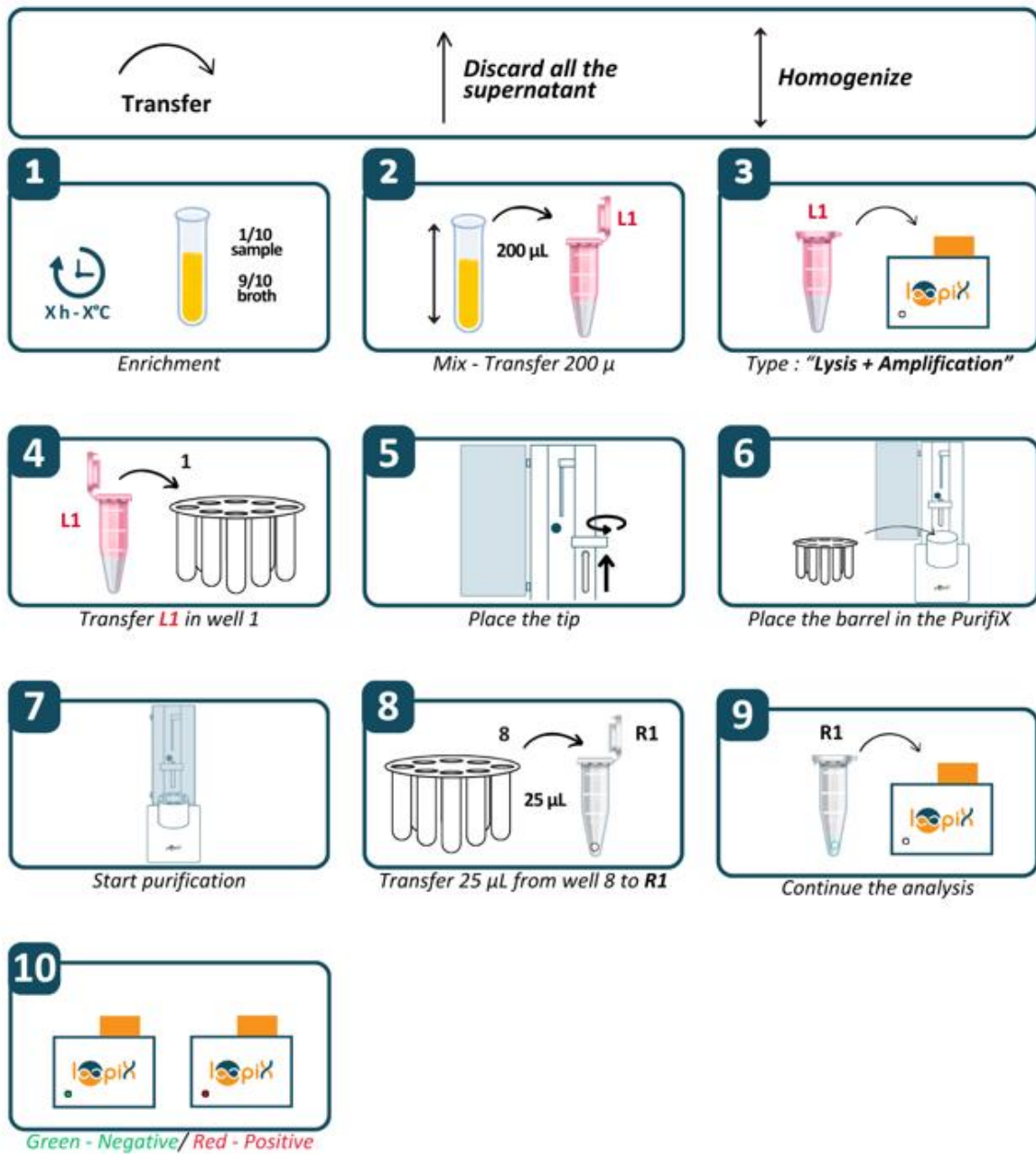
In the context of AOAC validation, a positive LoopDeetect *Listeria monocytogenes* result is considered a presumptive positive. It is recommended to confirm the result using an appropriate reference method. Alternatively, the result can be confirmed directly from the extract using an appropriately validated PCR method. Before using a validated method (e.g. : iq-check *Listeria monocytogenes*, ez-check *Listeria monocytogenes*, *Listeria monocytogenes*), please concentrate the DNA using, for example, a MWCO-type membrane.

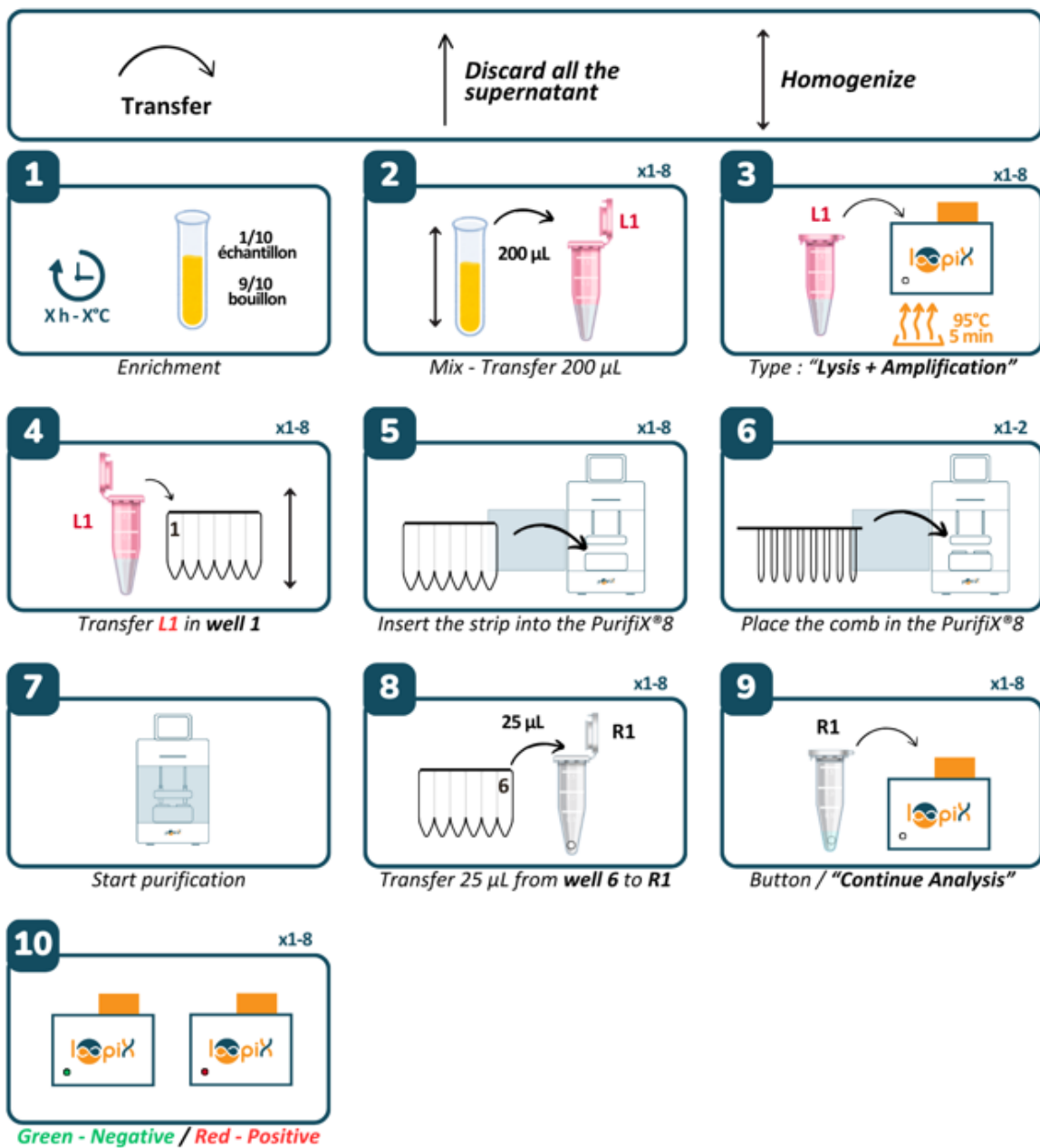
- Load the DNA eluate into the filter tube
- Centrifuge at 14,000 × g for 5 minutes
- Discard the filtrate.
- Add 50 µL of the desired TE buffer to the filter and re-centrifuge for 5 minutes
- Recovery: invert the filter unit into a clean tube and centrifuge briefly (2-3 min, 6,000 g) to collect the concentrated retentate.

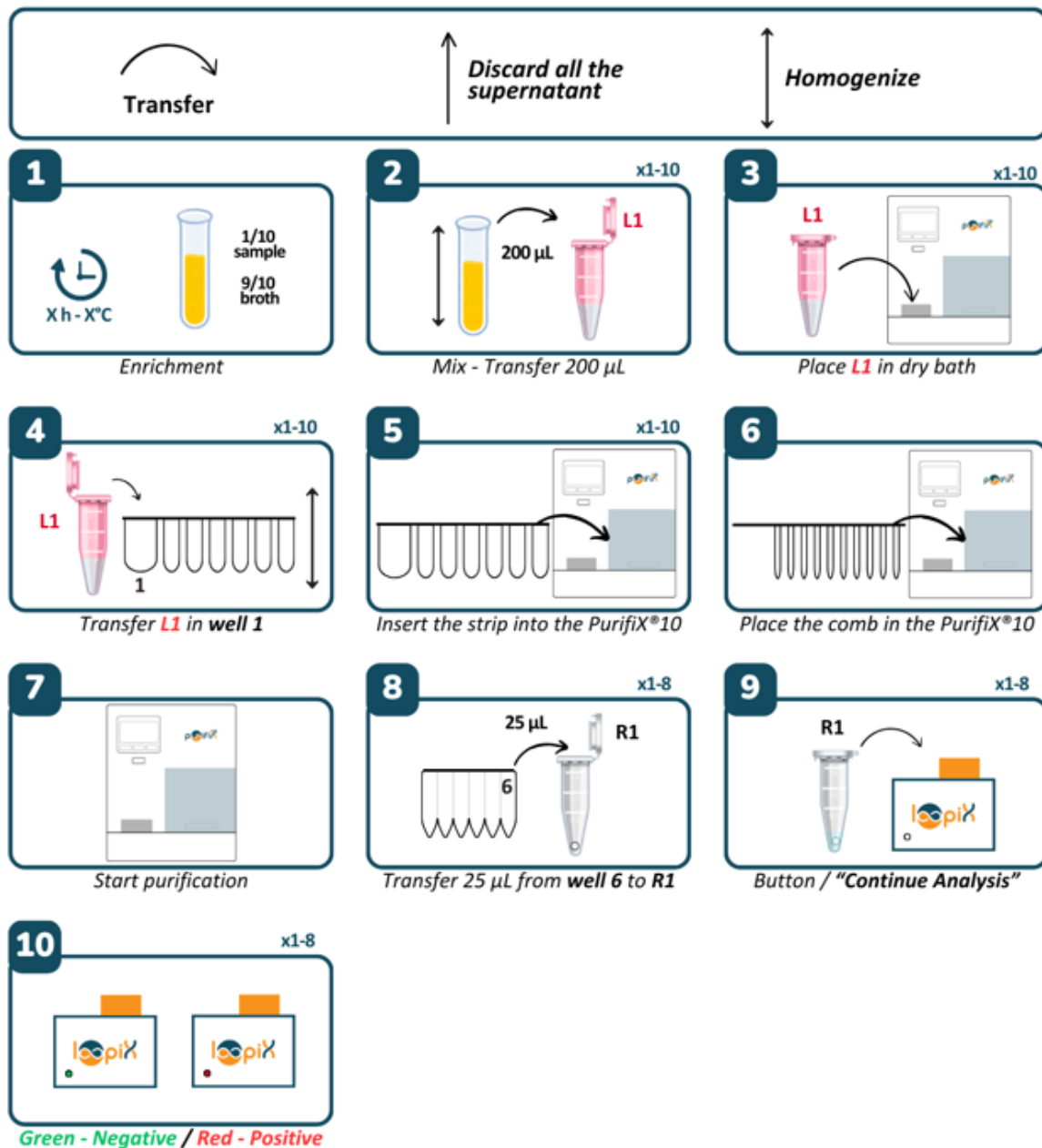
Follow the amplification instructions of the chosen manufacturer.

End of analysis

- Discard the **R1** reaction tube and **NEVER** open it.
- Discard all consumables used for the analysis.
- Using an alcohol-based solution and an absorbent cloth, clean the work area, tools (pipettes, stands, etc.), and devices (LoopiX®, PurifiX®, etc.).
- Perform a 30-minute UV sterilization cycle for devices equipped with this feature.







COL_01 - Colony isolated from a Petri dish culture / Manual PFS+ purification

Sampling

- Using a loop, pick up a colony and resuspend it in lysis tube **L1**

Lysis

- Close tube **L1** and place it in the LoopiX®.
- Using the control interface, start a “Lysis and Amplification” run. The indicator light flashes red throughout the lysis process.
- At the end of lysis, the indicator light flashes blue. Remove tube **L1** from the LoopiX®.
- Using a pipette with a tip or a 6 mL transfer pipette, aspirate the entire contents of tube **L1**
- Fermer le tube **L1** et le placer dans le LoopiX®.

Purification using the PFS+ kit

- Transfer the lysate to tube **B2** using a 6-mL pipette or a pipette fitted with a tip.
- Homogenize with 15 up-and-down pipetting strokes, ensuring that the beads are evenly distributed without clumping.
- Wait 1 minute, then place tube **B2** on the magnetic stand to capture the beads.
- Discard the supernatant without touching the beads, leaving **B2** on the stand.
- Add buffer **W3** and homogenize again.
- Place tube **B2** on the magnetic stand to capture the beads.
- Discard the supernatant without touching the beads; tube **B2** remains on the stand.
- Add buffer **W4** and repeat the steps for homogenization, bead capture, and supernatant removal.
- Add buffer **W5** and repeat the same steps for homogenization, bead capture, and supernatant removal.
- Dry the bottom of the tube with a Lemoine swab or a pipette fitted with a new tip (nothing should remain at the bottom of the tube), then discard the swab or tip.
- Let air dry for 5 minutes with tube **B2** open.
- Using a 3.5 mL pipette or a pipette fitted with a new tip, add buffer **E6** and homogenize. Wait 1 minute.
- Place tube **B2** on the magnetic stand to capture the beads.
- Using a pipette fitted with a new tip, withdraw 25 µL from tube **B2** without touching the beads.
 - To confirm positive results, it is recommended that you transfer the entire sample to a clean, properly labeled 1.5-mL tube.
 - Store this sample between 2°C and 8°C for use within the same day. Store the sample at -18°C for long-term storage.

Analysis

- Transfer the 25 µL to reaction tube **R1**.
- Close tube **R1** by pressing down with your finger until you hear a “click” indicating that it is securely closed, then place it in the LoopiX®.
- Start the analysis by pressing the button on the front panel of the LoopiX® or via the control interface.
- At the end of the analysis, read the result directly from the status LED and the control interface:

- **Red LED:** positive result – *Listeria monocytogenes* detected
- **Green LED:** negative result – *Listeria monocytogenes* not detected

Confirmation of positive results

In the context of AOAC validation, a positive LoopDeetect *Listeria monocytogenes* result is considered a presumptive positive. It is recommended to confirm the result using an appropriate reference method. Alternatively, the result can be confirmed directly from the extract using an appropriately validated PCR method. Before using a validated method (e.g. : iq-check *Listeria monocytogenes*, ez-check *Listeria monocytogenes*, *Listeria monocytogenes*), please concentrate the DNA using, for example, a MWCO-type membrane.

- Load the DNA eluate into the filter tube
- Centrifuge at 14,000 × g for 5 minutes
- Discard the filtrate.
- Add 50 µL of the desired TE buffer to the filter and re-centrifuge for 5 minutes
- Recovery: invert the filter unit into a clean tube and centrifuge briefly (2-3 min, 6,000 g) to collect the concentrated retentate.

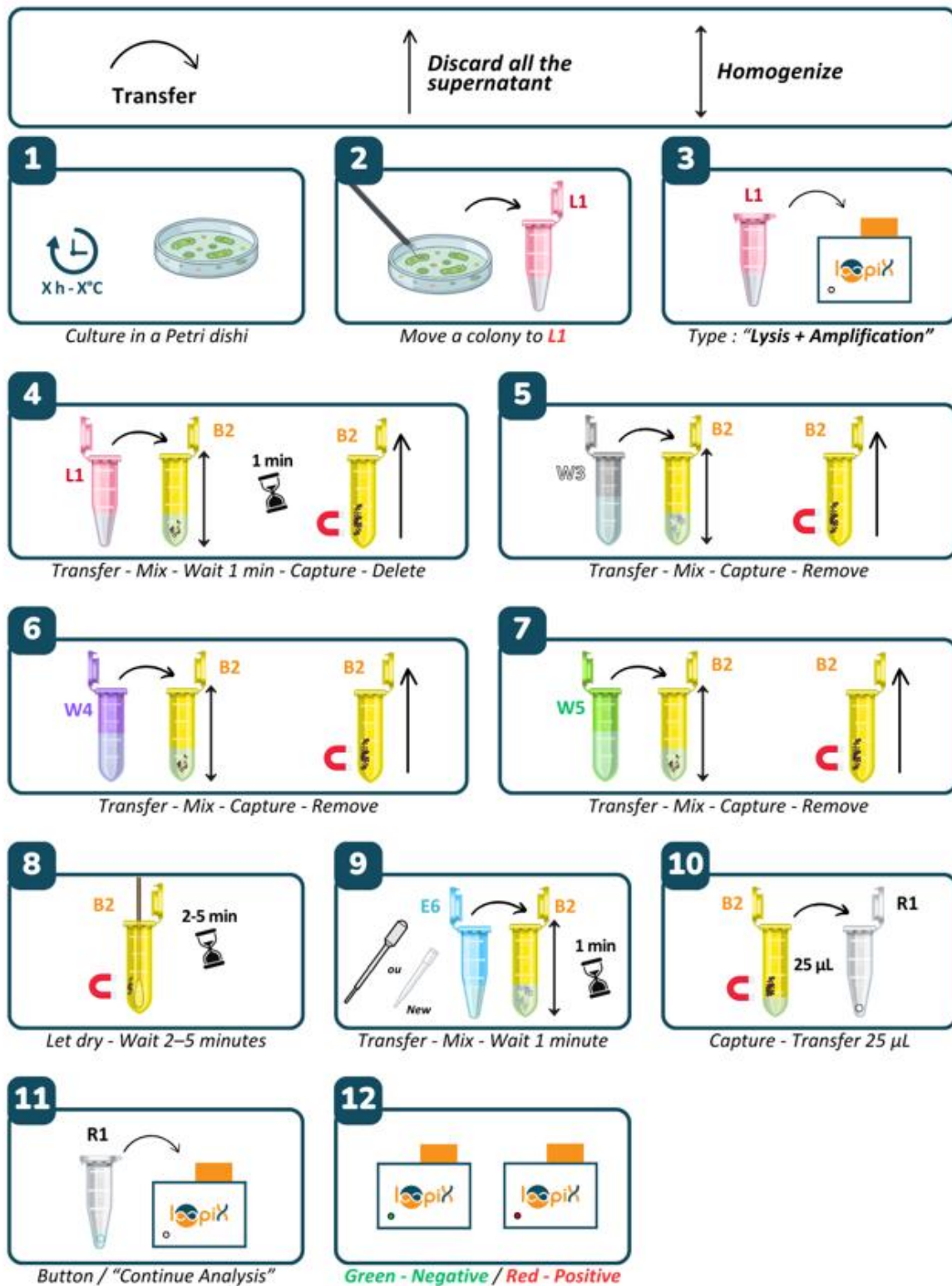
Follow the amplification instructions of the chosen manufacturer.

End of analysis

- Discard the **R1** reaction tube and **NEVER** open it.
- Discard all consumables used for the analysis.
- Using an alcohol-based solution and an absorbent cloth, clean the work area, tools (pipettes, stands, etc.), and devices (LoopiX®, PurifiX®, etc.).
- Perform a 30-minute UV sterilization cycle for devices equipped with this feature.

COL_01

Colony from Petri dish culture / Manual PFS+ purification



COL_02 - Colony grown in a Petri dish / Purification: Purifix® Line

Sampling

- Using a loop, pick up a colony and resuspend it in lysis tube **L1**

Lysis

Lysis with LoopiX :

- Close tube **L1** and place it in the LoopiX®.
- Using the control interface, start a “Lysis and Amplification” run. The indicator light flashes red throughout the lysis process.
- At the end of lysis, the indicator light flashes blue. Remove tube **L1** from the LoopiX®.
- Using a pipette with a tip or a 6 mL transfer pipette, aspirate the entire contents of tube **L1**
- Fermer le tube **L1** et le placer dans le LoopiX®.

Lysis with dry bath:

The Purifix® 10 comes with a built-in heating element that is ready for immediate use. For other machines in the Purifix® series, a separate heating element is required.

- Seal tube **L1** and place it in the heating block preheated to **95°C**.
- Start the lysis for **5 minutes**.
- At the end of the lysis, remove tube **L1** from the heating block and wait 2 minutes for it to cool.
- Using a pipette with a tip or a 6-mL transfer pipette, aspirate the entire contents of tube **L1**

Purification using a PurifiX® series automated system

- Transfer the lysate to **well 1** of the PurifiX® strip using a 6-mL transfer pipette or a pipette fitted with a tip.
- Place the purification strip into the PurifiX®:
 - PurifiX® 1: Place the cartridge into the cup, then place the cup into the PurifiX® 1.
 - PurifiX® 8: Place the strip on the rack, then place the rack in the PurifiX® 8.
 - PurifiX® 10: Place the purification strip directly into the PurifiX® 10.
- Place the comb(s) into the device, making sure to push them all the way in.
- Close the device lid and start the run using the control interface.
- At the end of purification, discard the comb(s) and remove the purification strip.
- Using a pipette fitted with a new tip, aspirate **25 µL** from the last well of the strip (PurifiX® and PurifiX® 10: **well 8** / PurifiX® 8: **well 6**) and proceed with the analysis.
 - To confirm positive results, it is recommended to transfer the entire extract to a clean, properly labeled 1.5-mL tube.
 - Store this extract between 2°C and 8°C for use within the same day. Store the extract at -18°C for longer-term use.

Analysis

- Transfer the 25 µL to reaction tube **R1**.
- Close tube **R1** by pressing down with your finger until you hear a “click” indicating that it is securely closed, then place it in the LoopiX®.

- Start the analysis by pressing the button on the front panel of the LoopiX® or via the control interface.
- At the end of the analysis, read the result directly from the status LED and the control interface:
 - **Red LED:** positive result – *Listeria monocytogenes* detected
 - **Green LED:** negative result – *Listeria monocytogenes* not detected

Confirmation of positive results

In the context of AOAC validation, a positive LoopDeetect *Listeria monocytogenes* result is considered a presumptive positive. It is recommended to confirm the result using an appropriate reference method. Alternatively, the result can be confirmed directly from the extract using an appropriately validated PCR method. Before using a validated method (e.g. : iq-check *Listeria monocytogenes*, ez-check *Listeria monocytogenes*, *Listeria monocytogenes*), please concentrate the DNA using, for example, a MWCO-type membrane.

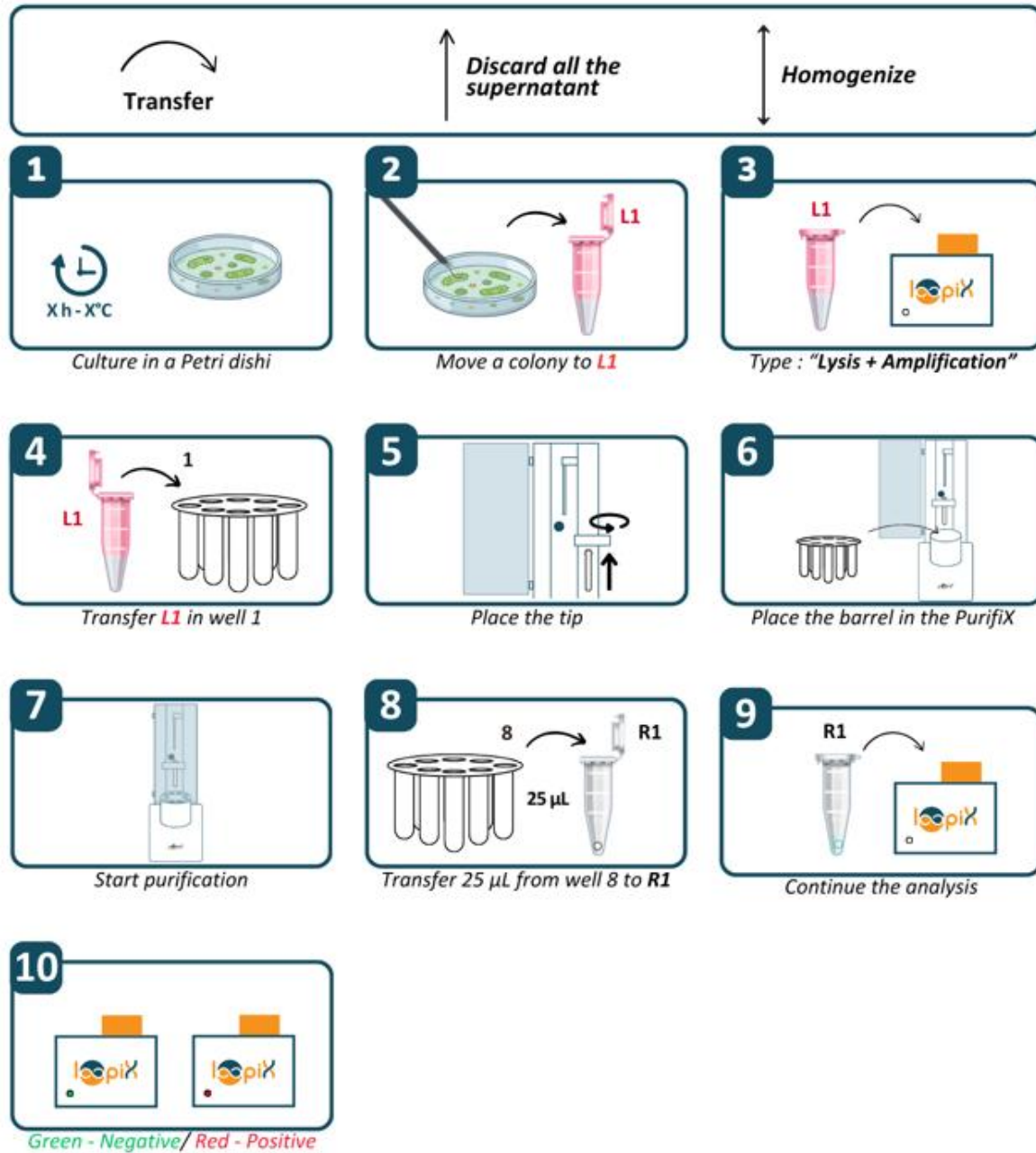
- Load the DNA eluate into the filter tube
- Centrifuge at 14,000 × g for 5 minutes
- Discard the filtrate.
- Add 50 µL of the desired TE buffer to the filter and re-centrifuge for 5 minutes
- Recovery: invert the filter unit into a clean tube and centrifuge briefly (2-3 min, 6,000 g) to collect the concentrated retentate.

Follow the amplification instructions of the chosen manufacturer.

End of analysis

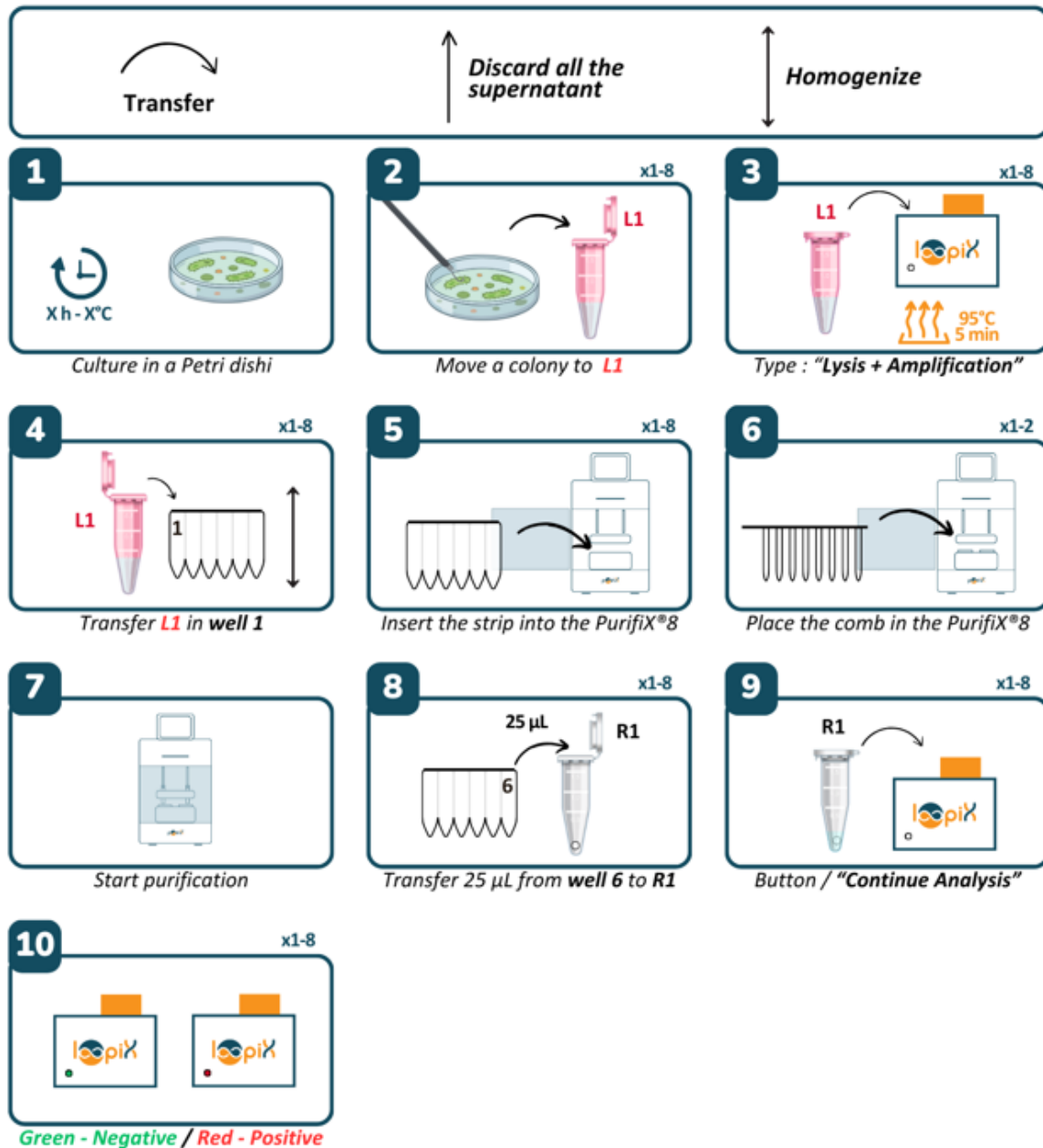
- Discard the **R1** reaction tube and **NEVER** open it.
- Discard all consumables used for the analysis.
- Using an alcohol-based solution and an absorbent cloth, clean the work area, tools (pipettes, stands, etc.), and devices (LoopiX®, PurifiX®, etc.).
- Perform a 30-minute UV sterilization cycle for devices equipped with this feature.

COL_02 Colony grown in a Petri dish / PurifiX®1



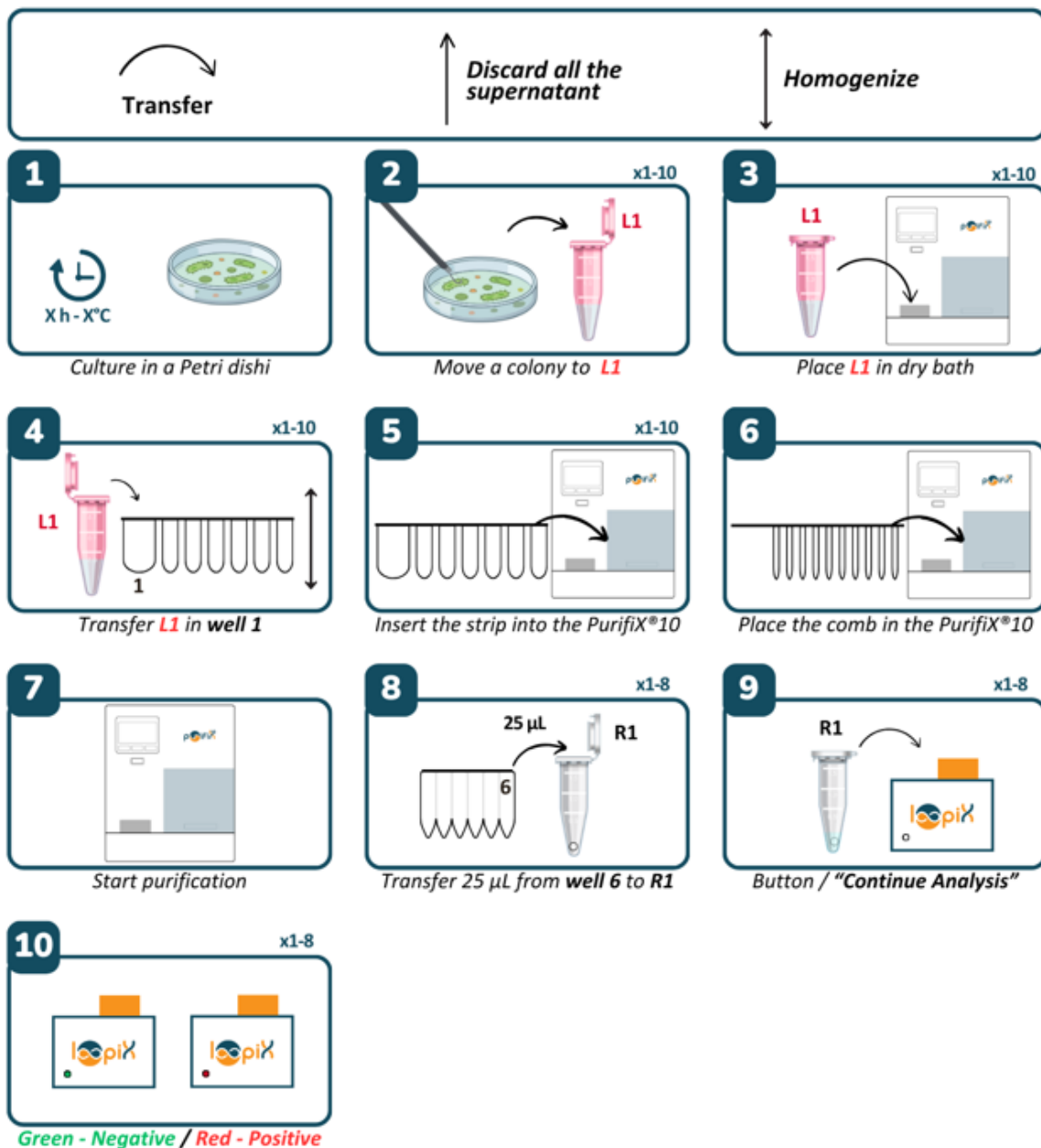
COL_02

Colony grown on a Petri dish / PurifiX®8



COL_02

Colony grown on a Petri dish / PurifiX®10



NEG_01 – Negative control

Preparation

- Using a pipette with a tip, withdraw **25 µL** from tube **S1**.

Analysis

- Transfer the 25 µL to reaction tube **R1**.
- Close tube **R1** by pressing down with your finger until you hear a “click” indicating that it is securely closed, then place it in the LoopiX®.
- Start the analysis by pressing the button on the front panel of the LoopiX® or via the control interface.
- At the end of the analysis, read the result directly from the status LED and the control interface:
 - **Red LED**: positive result – *Listeria monocytogenes* detected
 - **Green LED**: negative result – *Listeria monocytogenes* not detected

Interpretation of results

The expected result is a negative result (**green LED**).

A positive result (**red LED**) is not acceptable. Thoroughly clean the equipment and the work surface, then repeat the test, carefully following these instructions. If the result is positive again, contact the distributor.

End of analysis

- Discard the **R1** reaction tube and **NEVER** open it.
- Discard all consumables used for the analysis.
- Using an alcohol-based solution and an absorbent cloth, clean the work area, tools (pipettes, stands, etc.), and devices (LoopiX®, PurifiX®, etc.).
- Perform a 30-minute UV sterilization cycle for devices equipped with this feature.

POS_01 – Positive control - LoopDeetect

Materials

WARNING: You must use the specific positive control included in the LoopDeetect kit being tested:

Positive control Name	Kit content	Related kit
LoopDeetect <i>Listeria</i> +	- 1 positive control (yellow tube / red bag) - 1 tube (S1)	LoopDeetect <i>L.monocytogenes</i> LoopDeetect <i>Listeria spp.</i>

- Open the pouch containing the T+ positive control (**yellow tube**). Check for the presence of the tablet (blue coloration at the bottom of the tube).

Preparation

- Using a pipette fitted with a tip, aspirate **50 µL** from tube **S1**.
- Transfer the sample to the blue spot on the **T+** tube of the target.
- Mix thoroughly by repeatedly aspirating and dispensing the sample.
- Using the pipette, aspirate **25 µL** from the **T+** tube.

Analysis

- Transfer the 25 µL to reaction tube **R1**.
- Close tube **R1** by pressing down with your finger until you hear a “click” indicating that it is securely closed, then place it in the LoopiX®.
- Start the analysis by pressing the button on the front panel of the LoopiX® or via the control interface.
- At the end of the analysis, read the result directly from the status LED and the control interface:
 - **Red LED**: positive result – *Listeria monocytogenes* detected
 - **Green LED**: negative result – *Listeria monocytogenes* not detected

Interpretation of results:

The expected result is a positive result (**red LED**).

A negative result (**green LED**) is not acceptable. Repeat the test using a new positive control, following these instructions carefully. If the result is negative again, contact your distributor.

End of analysis

- Discard the **R1** reaction tube and **NEVER** open it.
- Discard all consumables used for the analysis.
- Using an alcohol-based solution and an absorbent cloth, clean the work area, tools (pipettes, stands, etc.), and devices (LoopiX®, PurifiX®, etc.).
- Perform a 30-minute UV sterilization cycle for devices equipped with this feature.

POS_02 – Positive control – Extraction (PFS+ / PurifiX® line

Materials

WARNING: You must use the specific positive control included in the LoopDeetect kit being tested:

Positive control Name	Kit content	Related kit
LoopDeetect <i>Listeria</i> +	- 1 positive control (yellow tube / red bag) - 1 tube (S1)	LoopDeetect <i>L.monocytogenes</i> LoopDeetect <i>Listeria spp.</i>

- Open the pouch containing the T+ positive control (**yellow tube**). Check for the presence of the tablet (blue coloration at the bottom of the tube).

Preparation

- Using a pipette fitted with a tip, aspirate **50 µL** from tube **S1**.
- Transfer the sample to the blue spot on the **T+** tube of the target.
- Mix thoroughly by repeatedly aspirating and dispensing the sample.

Lysis

- Perform the **lysis** step according to the protocol for the method being used.
- **DO NOT ADD AT T+ THIS STEP. Risk of degradation.**

Purification

- Using the pipette, aspirate **all the volume (50µl)** from the **T+** tube.
- Add **T+** to the L1 tube. **Proceed immediately to the next step.**
- Perform the **purification** step according to the protocol for the method being used.

Analysis

- Transfer the 25 µL to reaction tube **R1**.
- Close tube **R1** by pressing down with your finger until you hear a “click” indicating that it is securely closed, then place it in the LoopiX®.
- Start the analysis by pressing the button on the front panel of the LoopiX® or via the control interface.
- At the end of the analysis, read the result directly from the status LED and the control interface:
 - **Red LED:** positive result – *Listeria monocytogenes* detected
 - **Green LED:** negative result – *Listeria monocytogenes* not detected

Interpretation of results:

The expected result is a positive result (**red LED**).

A negative result (**green LED**) is not acceptable. Repeat the test using a new positive control, following these instructions carefully. If the result is negative again, contact your distributor.

End of analysis

- Discard the **R1** reaction tube and **NEVER** open it.
- Discard all consumables used for the analysis.
- Using an alcohol-based solution and an absorbent cloth, clean the work area, tools (pipettes, stands, etc.), and devices (LoopiX®, PurifiX®, etc.).
- Perform a 30-minute UV sterilization cycle for devices equipped with this feature.

INH_01 – Inhibition control

The expected result is a positive result (red LED). This protocol allows you to verify the compatibility of the matrix with the LoopDeetect method. The principle involves adding the target to the sample after purification to confirm that amplification is successful and that the sample does not contain any inhibitory substances that may not have been removed during the purification step.

This protocol can only be performed using the purification kits provided by Loop Dee Science.

Materials

WARNING : You must use the specific positive control included in the LoopDeetect kit being tested:

Positive control Name	Kit content	Related kit
LoopDeetect <i>Listeria</i> +	- 1 positive control (yellow tube / red bag) - 1 tube (S1)	LoopDeetect <i>L.monocytogenes</i> LoopDeetect <i>Listeria</i> spp.

Open the pouch containing the T+ positive control (yellow tube). Check for the presence of the tablet (blue coloration at the bottom of the tube).

Sampling

- Perform the **sampling step** according to the protocol for the method being used.

Lysis

- Perform the **lysis** step according to the protocol for the method being used.

Purification

- Perform the **purification** step according to the protocol for the method being used.

At the end of purification

- Using a pipette with a tip or a transfer pipette, withdraw the entire purified eluate.
- Transfer the sample to the **T+** tube of the target, onto the blue pellet.
- Homogenize by performing a series of up-and-down pipetting motions.
- Using a pipette with a tip, withdraw **25 µL** from the target's **T+** tube

Analysis

- Transfer the 25 µL to reaction tube **R1**.
- Close tube **R1** by pressing down with your finger until you hear a “click” indicating that it is securely closed, then place it in the LoopiX®.
- Start the analysis by pressing the button on the front panel of the LoopiX® or via the control interface.
- At the end of the analysis, read the result directly from the status LED and the control interface:
 - **Red LED**: positive result – *Listeria monocytogenes* detected

- **Green LED:** negative result – *Listeria monocytogenes* not detected

Interpretation of results:

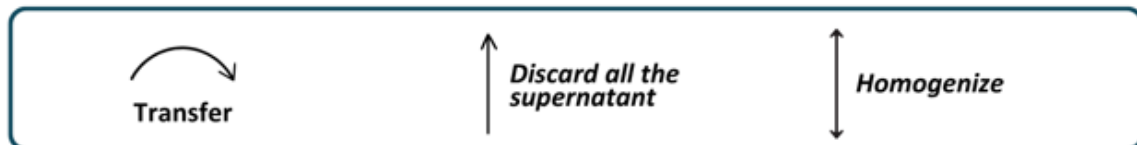
A positive result (**red LED**) indicates the absence of matrix-related inhibition. It is recommended that the test be repeated to confirm the result.

A negative result (**green LED**) indicates the presence of matrix-related inhibition. It is recommended to repeat the test to confirm the result. If the result is negative again, contact the distributor to discuss how to resolve this inhibition.

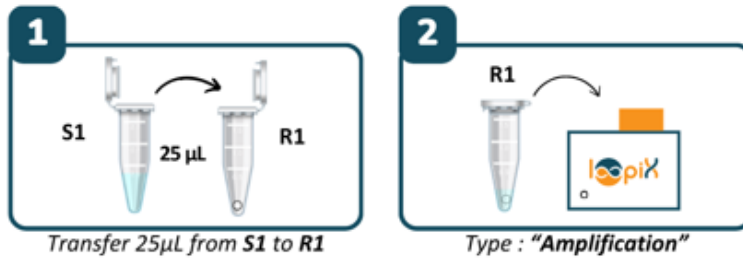
End of analysis

- Discard the **R1** reaction tube and **NEVER** open it.
- Discard all consumables used for the analysis.
- Using an alcohol-based solution and an absorbent cloth, clean the work area, tools (pipettes, stands, etc.), and devices (LoopiX®, PurifiX®, etc.).
- Perform a 30-minute UV sterilization cycle for devices equipped with this feature.

TEM_01 Controls - Negative / Positive / Inhibition

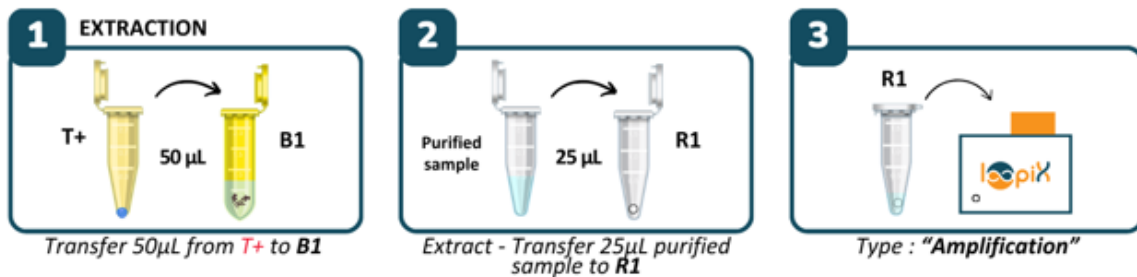
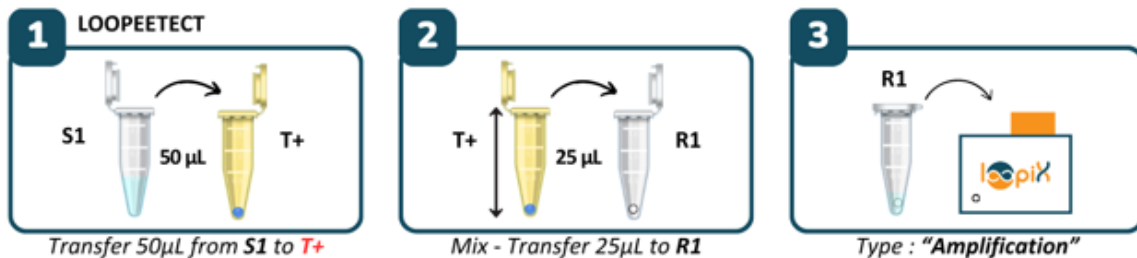


Negative control



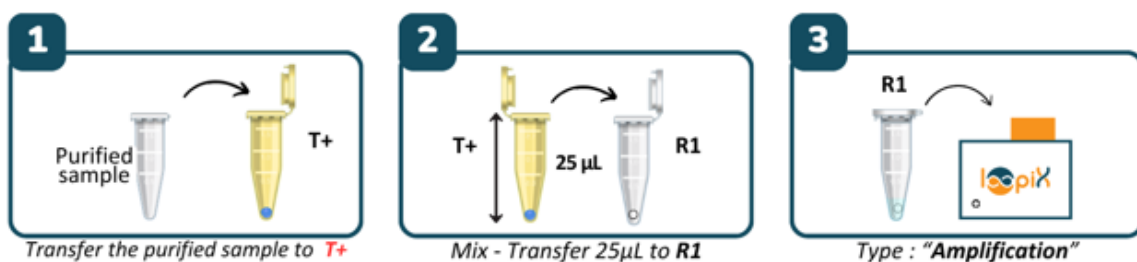
EXPECTED RESULT **GREEN - NEGATIVE**

Positive control



EXPECTED RESULT **RED - POSITIVE**

Inhibition control



INHIBITION: **GREEN - NEGATIVE** // NO INHIBITION : **RED - POSITIVE**

Quality controls

All manufactured products are produced in accordance with our quality system, from the receipt of raw materials through to the sale of the finished product. Each batch undergoes quality control testing and is released for sale only if it meets the acceptance criteria established by Loop Dee Science Worldwide.

Storage and stability

- All kits are usable for 12 months from their date of manufacture and may be used until the expiration date indicated on each kit.
- All reagents included in the kits are for single use only.
- For LoopDeetect kits, be sure to store all reagents in a cool, dry place at a temperature between 2 and 25°C.
- Keep the reagents away from any heat sources (radiators, windows exposed to sunlight, etc.).
- If the room temperature cannot be controlled, store the kits in the refrigerator.
- The reaction tube (R1) has a very short shelf life once removed from its pouch. It is therefore recommended to open the pouch just before use.
- For Purification Kits, be sure to store all reagents in a cool, dry place at a room temperature between 15 and 25°C. Keep the reagents away from any heat sources (radiators, windows exposed to sunlight, etc...)

Disposal – Waste Management

- Hazardous laboratory, chemical, or biological waste must be handled and disposed of in accordance with all local, regional, and national regulations.
- Aluminium pouches can be recycled.

Technical support

Loop Dee Science Worldwide provides customer support for all technical and service-related questions and issues concerning the LoopDeetect product range. For assistance, please contact our support team:

Adress :

Support technique
Loop Dee Science
100 rue Philippe LIVRY-LEVEL
14760 Bretteville-sur-Odon (France)

Phone : +33 231 730 791 (9:00 à 17:00 / CET)

Mail : support@loopdeescience.com

Web site : <https://www.loopdeescience.com>

Document History / Version Tracking

Revision	Date	Description
0.0	Mars-26	Initial public review
1.0	August 26	Addition of the AoAC Performance
1.1	August 26	Addition of the method's objective
1.2	August 26	Addition Modification for the positive result confirmation
1.3	Septembre	Addition of the catalog number and AOAC PTM number