SwabUp™ Lab Monitoring

For the regular monitoring of lab work area and detection of target and amplicon DNA contaminations

INSTRUCTIONS FOR USE

FOR USE IN RESEARCH AND QUALITY CONTROL

Symbols

Lot No.

Order No.

Expiry date

Storage temperature

Number of extractions

Manufacturer

INDICATION

Small amounts of amplicon- or target DNA contaminations could lead to PCR artifacts and false positive results in the highly sensitive PCR technique. Originating from aerosolized fragments in centrifuges, pipettes and other lab equipment or from small splatters during working with open reaction tubes, contaminant DNA is very hard to remove and can lead to cross contaminations between samples. A single DNA molecule can be detected in the amplification process leading to widespread problems throughout the testing procedure and interpretation of results. Unfortunately, DNA contaminations can occur occasionally even in experienced labs and will go unnoticed unless detected in PCR.

The purpose of SwabUp™ Lab Monitoring kits is the tracing of DNA contamination hot-spots in molecular biology labs in order to efficiently eliminate them and prevent future occurrence. Regular monitoring and cleaning of lab and work area can help the early detection and avoidance of DNA contaminations. SwabUp™ Lab Monitoring kits combine components for sample collection, DNA extraction and PCR amplification and therefore exhibit a competent system for environment-monitoring.

SwabUp™ Lab Monitoring kit is very easy to use. Collection swab applicators are packaged individually in sealed plastic peel-pouches. The shaft of the applicator is made of plastic and the top-end (tip) is made of flocked nylon fibers, exhibiting excellent absorption ability. The collection swab applicators have a molded breakpoint in the shaft of the applicator, which facilitates easy breakage of the swab applicator after collecting the sample and transport into the tube containing the Collection Buffer. After extensive testing, these swabs were specifically selected for the purpose of this kit, as they have proven to be especially suitable for the procedure. The DNA extraction system was optimized for the efficient detection of the smallest amounts of contaminant DNA. In addition, a contamination-free ready-to-use PCR system is added in the SwabUp™ Lab Monitoring Plus kit, to exclude DNA cross-contaminations which are caused by user‘s own potentially contaminated PCR reagents and buffers. This PCR system includes a lyophilized hot start Taq polymerase containing PCR mix, compatible with both conventional- and qPCR.

PRINCIPLE OF THE METHOD

The method is simple and consists of five general steps: (1) Collection of samples using the provided swab applicators, (2) selective binding of DNA to spin columns, (3) removal of residual contaminants and inhibitors, and (4) elution of purified DNA.

The DNA extraction procedure is necessary in order to avoid PCR inhibition through inhibiting substances such as fabrics, tissues, dust or a high protein content of the collected sample. It should therefore be performed prior to the analysis of collected samples through PCR amplification. The procedure does not require phenol/chloroform extraction and needs minimal hands-on time (approx. 30 minutes), providing DNA ready-to-use for PCR.

REAGENTS AND COMPONENTS

Each kit contains reagents and components for 10 or 50 samples. The expiry date of the unopened package is marked on the package label. Components of DNA extraction system must be stored at room temperature. Collection Buffer tubes must be stored at 2 – 8 °C immediately after delivery. Swabs can be stored at 2 – 30 °C.

Kit Component	10 Swab-Samples (Cat. no. 181-0010)	50 Swab-Samples (Cat. no. 181-0050)
Swabs	10 units	50 units
Collection Buffer tubes	10 units	50 units
Spin columns	10 units	50 units
Collection tubes	10 units	50 units
Starting Buffer	5 ml	15 ml
Binding Buffer	10 ml	25 ml
Buffer SW1	3 ml (add 3 ml ethanol, abs., before first use)	15 ml (add 15 ml ethanol, abs., before first use)
Buffer SW2	4 ml (add 16 ml ethanol, abs., before first use)	12 ml (add 48 ml ethanol, abs., before first use)
Elution Buffer	2 ml	2 × 2 ml

The lot-specific QC certificate (Certificate of Analysis) can be downloaded from our website (www.minerva-biolabs.com).

USER-SUPPLIED CONSUMABLES AND EQUIPMENT

SwabUp™ Lab Monitoring kit contains reagents and components for collection of samples and extraction of DNA. Additional consumables and equipment are supplied by the user:

- Ethanol > 96 % abs.
- DNase-free reaction tubes (1.5 ml or 2 ml)
- Microcentrifuge and heat block for 1.5 ml (or 2 ml) reaction tubes
- Pipettes with corresponding DNase-free filter tips (100 and 1000 µl)
- Components and equipment for PCR amplification (recommended: MB Taq DNA Polymerase, ConviFlex™ DNAmp Mix, or SwabUp™ Lab Monitoring Plus kit which additionally contains a PCR amplification system. See Related Products for ordering information).

SPECIMEN

Different surfaces like desktops and lab work area, as well as equipment in molecular biological labs (e.g. centrifuge, pipettes, reaction tube racks etc.) are easily exposed to target and amplicon DNA contaminations. By touching doorknobs, paper, computer keyboard and -mouse before and after doing lab work, lab operators unintentionally carry over and spread DNA contaminations regardless of how experienced and careful they are. In addition, applying certain PCR techniques such as two-step qPCR or nested conventional PCR increases the risk of causing amplicon DNA contaminations by carrying over DNA contaminants from one PCR to the next through pipetting.

Samples should therefore be collected from surfaces and/or equipment which are easily exposed to target and amplicon DNA contaminations, e.g., centrifuge, pipettes, reaction tube racks, door-knobs, lab books, computer keyboard, computer mouse, touchpad, desktops and any surface of a molecular lab work area.

Each sample should be collected by using the swab top-end and thoroughly swabbing a different 10×10 cm surface.

RECOMMENDATIONS

SwabUp™ Lab Monitoring kit is recommended for the regular monitoring of the lab work area and detection of target or amplicon DNA contaminations. Detection of DNA contamination hot-spots will help maintain a clean work area and avoid PCR artifacts and inaccurate data. Therefore, we recommend performing this test in regular time intervals. It is also recommended to set the PCR amplification with the most frequently used primer sets, or those with the most frequent reoccurrence of irregularities or unspecific results.

For your assistance we provide you with instructions for lab monitoring, which were set up after years of experience and extensive testing, a table for the documentation of the lab-monitoring process, as well as initial guidlines for the measures which should be taken in case of a DNA contamination (s. Appendix I). By following these instructions and guidlines you will be able to track the source of DNA contaminations in your lab and the route on which they are carried over, and take necessary measures to eliminate these contaminations and prevent future occurrence.

SwabUp™ Lab Monitoring kit is for research use only. It is not recommended for clinical and diagnostic applications or for the detection of RNA contaminations.

PRECAUTIONS

SwabUp™ Lab Monitoring kit should be used by trained laboratory staff only. All samples should be handled with all due care and attention. Always wear a suitable lab coat, goggles and disposable gloves.

The sample preparation waste contains Binding Buffer and Buffer SW1, which may form highly reactive compounds when combined with bleaching agents. DO NOT add bleaching agents or acidic solutions directly to the sample preparation waste. Clean with suitable laboratory detergent and water, if any liquid is spilt.

Binding Buffer contains propan-2-ol and polyethylene glycol octylphenol ether and is therefore flammable, harmful and irritant. Buffer SW1 contains guanidinium thiocyanate and is therefore harmful and irritant. In case of skin or eye contact wash thoroughly with running water and seek medical attention immediately.

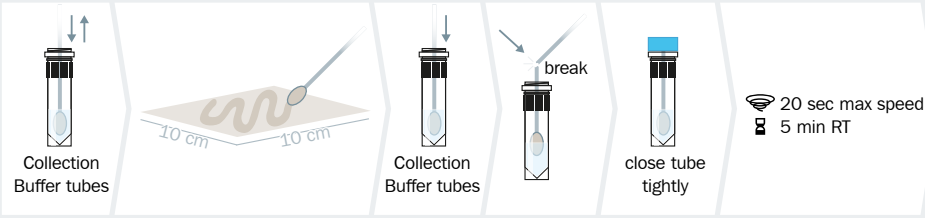
For more information please read safety data sheets (SDS) on our website: www.minerva-biolabs.com.

ADDITIONAL NOTES

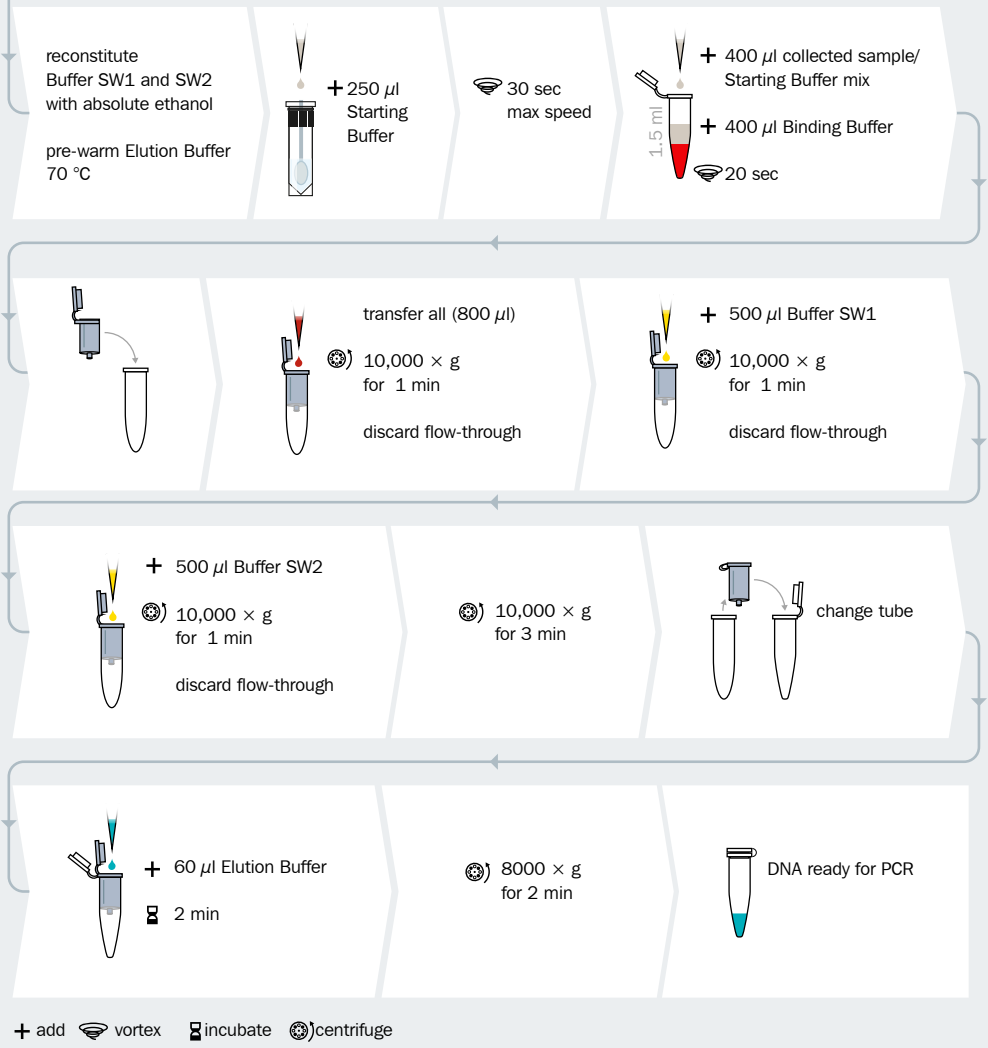
⇨	These instructions must be understood to successfully use the SwabUp™ Lab Minitoring kit. The reagents supplied should not be mixed with reagents from different lots but used as an integral unit. The reagents of the kit must not be used beyond shelf life.
⇨	To avoid DNA cross-contaminations during the process, the test should be performed under sterile and DNA-free conditions.
⇨	DNA extraction should be performed immediately after sample collection to avoid DNA cross-contaminations through storage.
⇨	Follow the exact protocol. Any deviation from the extraction method may affect the results.
⇨	We recommend including control samples on a regular basis to monitor the reliability of your results. It is also advantageous in case of troubleshooting.
⇨	Do not use other alcohols apart from ethanol as it will lead to inconsistent yields.
⇨	Pre-heating of Elution Buffer improves the yield significantly.

PROCEDURE - OVERVIEW

1. Sample Collection



2. DNA Extraction



+ add vortex incubate centrifuge

This procedure overview is not a substitute for the detailed manual.

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PROCEDURE - STEP BY STEP

1. Sample Collection

1.	Take out a swab applicator from the plastic peel-pouch by peeling the shaft-end of the pouch open. Note: you should always open the pouch at the shaft-end. Do not touch the tip of the swab during sampling.
2.	Open the Collection Buffer tube and dip the tip of the swab applicator into the ***Collection Buffer*** until it is completely soaked.
3.	Take the soaked swab out of the tube carefully and wipe the surface you wish to test thoroughly. A surface of 10 × 10 cm is recommended for optimal results.
4.	Transfer the swab applicator into the Collection Buffer tube. Use the molded breakpoint in the shaft of the swab applicator to break the shaft so that the top-end of the swab is left inside the tube.
5.	Close the tube tightly and vortex for 20 sec at maximum speed.
6.	Incubate samples at room temperature for 5 min. Samples are now ready for DNA extraction.

2. DNA Extraction

⇨ Before first use reconstitute Buffer SW1 and Buffer SW2 with absolute ethanol.
⇨ Set the heat block to 70 °C and equilibrate required amount of Elution Buffer to 70 °C.

1.	Add ***250 µl of Starting Buffer*** to the collected sample and vortex at maximum speed for at least 30 sec.
2.	Transfer 400 µl to a DNase-free 1.5 ml reaction tube and add ***400 µl of Binding Buffer*** to the sample. Vortex immediately and thoroughly in order to prevent any precipitation of nucleic acids. Do not centrifuge the sample and proceed immediately with step 3.
3.	Place a spin column in a collection tube. Transfer the Binding Buffer/sample - mix (approx. 800 µl) into the spin column. Note: be careful not to moisten the rim of the spin column.
4.	Centrifuge the spin column at ≥ 10,000 × g for 1 min. Discard the flow-through from the collection tube and reassemble spin column and collection tube.
5.	Add ***500 µl of Buffer SW1***. Centrifuge the spin column at ≥ 10,000 × g for 1 min, discard the flow-through and reassemble the spin column and collection tube.
6.	Add ***500 µl of Buffer SW2***. Centrifuge the spin column at ≥ 10,000 × g for 1 min, discard the flow-through and reassemble the spin column and collection tube.
7.	Centrifuge at full speed for 3 min in order to remove residual Buffer SW2.
8.	Discard the collection tube and place the spin column into a new DNase-free 1.5 ml reaction tube.

9.	Pipette 60 µl of pre-heated ***Elution Buffer*** (70 °C) into the spin column directly onto the center of the silica membrane. Be careful not to damage the membrane in the process. The membrane's surface should be covered with Elution Buffer.
10.	Incubate at room temperature for 2 min, then centrifuge at 8,000 × g for 2 min.
11.	The eluates can be used directly for PCR. If not analyzed immediately, eluates can be stored at 2 to 8 °C for a week or at ≤ -18 °C for long-term storage.

APPENDIX I

1. Lab Monitoring and tracking of contamination hot-spots

In order to supervise the efficiency of cleaning procedures in molecular biology labs, a comprehensive lab- and environment monitoring should be carried out in 3-month intervals.

Date	Lab operator/s	Lab	Room/s No.

Sample No.	Sample-label	Collection spot/area	Ct-value 1 (sample)	Ct-value 2 (internal control**)	Band (conventional PCR)	Result
1						
2						
3						
4						
5						
6						
7						
8						
9						
10	DNA extraction control*					
DNA amplification controls	Positive control					
	Negative control					
Evaluation and measures						

* DNA extraction control is optional but we recommend including it in the testing for the verification of the extraction procedure.

** Internal control is optional and can be used for the validation of PCR amplification.

2. Measures for elimination and prevention of DNA contaminations

If a DNA contamination was detected in your lab, you should proceed as follows:

1. If a DNA contamination was detected within an area, which is part of the regular cleaning procedure of your lab, you should immediately repeat the procedure using a sodium hypochlorite-containing surface cleaner (you should seek an adequate alternative for sensitive surfaces).

2. You should revise and take measures to improve the cleaning procedure in place.

3. If a DNA contamination was detected within an area, which is not part of the regular cleaning procedure of your lab, you should immediately include this area in your lab cleaning routine. Inform all lab operators and perform a proper training.

4. Repeat PCR amplification after cleaning until no contamination can be detected anymore.

5. Make sure all operators at your lab are well aware of these measures, and trained accordingly.

The above-provided table is available for download on our homepage: www.minerva-biolabs.com

APPENDIX II

Limited Product Warranty
This warranty limits our liability for replacement of this product. No warranties of any kind, express or implied, including, without limitation, implied warranties of merchantability or fitness for a particular purpose, are provided. Minerva Biolabs shall have no liability for any direct, indirect, consequential, or incidental damages arising from the use, the results of use, or the inability to use this product.

Trademarks
SwabUp, ConviFlex, ExtractNow, PCR Clean, Lab Clean, WaterShield and PCR Cycler Check are trademarks of Minerva Biolabs GmbH, Germany.

Related Products

ConviFlex™ DNAmp Mix

191-025/100/250	PCR Mix with Taq polymerase for conventional and qPCR	25/100/250 reactions

SwabUp™ Lab Monitoring Plus

182-0010/0050	Sample collection, DNA extraction and PCR system	10/50 samples

Nucleic Acid Extraction

601-1010/-1050	ExtractNow™ DNA Mini Kit	10/50 extractions
602-1010/-1050	ExtractNow™ Blood DNA Mini Kit	10/50 extractions
603-1010/-1050	ExtractNow™ RNA Mini Kit	10/50 extractions
604-1010/-1050	ExtractNow™ CleanUp Kit	10/50 extractions
605-1010/-1050	ExtractNow™ Plasmid Mini Kit	10/50 extractions
606-1010/-1050	ExtractNow™ Virus DNA/RNA Kit	10/50 extractions

Lab Clean™

15-4100	Molecular microbiology lab cleaner, bottled	1 Liter

PCR Clean™

15-2025/2200	DNA Decontamination Reagent, spray bottle/refill bottles	250 ml/4x 500 ml
15-2201	Wipes	120 wipes in a dispenser box
15-2202	Wipes, refill packs	5 x 120 wipes in a bag
15-2203	Wipes, single wrapped	30 sachets

WaterShield™

15-3015/3020/3050	Water Disinfection Additive for incubators and water baths, 200x concentrate	30 x 5 ml/3 x 50 ml/500 ml

MB Taq DNA Polymerase

53-0050/0100/0200/0250	MB Taq DNA Polymerase (5 U/μl)	50/100/200/250 units
53-1050/1100/1200/1250	MB Taq DNA Polymerase (1 U/μl)	50/100/200/250 units

PCR Cycler Validation

57-2102	PCR Cycler Check™ Advance	6 strips, 8 vials each
57-2103	PCR Cycler Check™ OneStep	100 reactions
57-2202	qPCR Cycler Check™	100 reactions

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Made in Germany

Minerva Biolabs GmbH develops and manufactures products in accordance with DIN EN ISO 9001:2008 and DIN EN ISO 13485:2012 quality system requirement. Reg.No. SY 60096693 0001 & SX 60096692 0001

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