SwiftX™ DNA

Handbook

Content

Storage

SwiftX™ DNA **must be stored at 2-8°C** and can be used until the expiry date indicated on the labels. SwiftX™ DNA can be stored **intermediately at up to 25°C** for up to 8 weeks.

Product Use

For Research Use Only.

SwiftX™ DNA is designed for rapid extraction of genomic and mitochondrial DNA from human or animal samples as well as DNA from bacteria, parasites (protozoa as well as helminth eggs), fungi and viruses.

SwiftX DNA can be applied to a **wide range of specimens**. Sample types applicable to the direct extraction protocol without cell concentration include swabs (e.g. buccal, nasal, pharyngeal, rectal), pre-concentrated cells, dried blood spots, blood cards, tissue samples (e.g. skin, muscle, liver, brain, bone marrow), Fine-needle aspirates, cerebrospinal fluid, hair follicles, fingernails.

Sample types applicable to the extraction protocol utilizing the cell capturing step include whole blood, urine, saliva, throat washes, cerebrospinal fluid, mammalian and bacterial cell cultures, homogenized tissue suspensions. Swab samples in guanidine-free transport media or alcohol-containing transport media (e.g. liquid-based cytology media).

The procedure can be performed manually as well as in an automated fashion. Automation of the nucleic acid extraction protocol is possible with a variety of robotic pipetting and handling systems due to its minimal number of working steps. Any adaptation must be performed and validated by the user.

Safety information

SwiftX™ DNA comprises of two buffers (Buffer EN and DL) and paramagnetic particles (Beads A). All components of the kit are free of hazardous substances. The safety data sheets (SDS) for SwiftX™ DNA components are available upon request.

Take caution when working with biological samples and always treat them as potentially infectious. Users are advised to always wear appropriate personal protective equipment.

Quality control

Each batch of SwiftX™ DNA is tested against defined specifications to ensure consistent product quality. A Certificate of Analysis can be provided upon request.

Equipment to be provided by the user

- Appropriate personal protective equipment
- Pipets and disposable pipet tips (aerosol barriers recommended)
- 1.5mL and 2mL microcentrifuge tubes (safe-lock-caps or screw-caps recommended) or a deep-well plate
- Magnetic stand
- Vortexer
- Heating device (water bath, heating block, or thermo shaker)
- optional: Proteinase K (20mg/mL)

Purchase information for kit and accessories

Item	Description	Cat.no.
SwiftX™ DNA, 25 extractions	Kit for DNA extraction from various specimen types and organisms	SXD-25
Mini heat block	Compact dry heat block for incubation of 15x 1.5mL microtubes	ACC-12
Magnetic separation rack	Rack with Neodymium magnets for 12x 1.5mL or 2mL microtubes	MAG-12
Proteinase K, 1 mL	Proteinase K (20 mg/mL) for improved lysis	PRK-1mL
Buffer EN, 25 mL	Enrichment buffer for SwiftX™ DNA	BEN-25mL

Principles

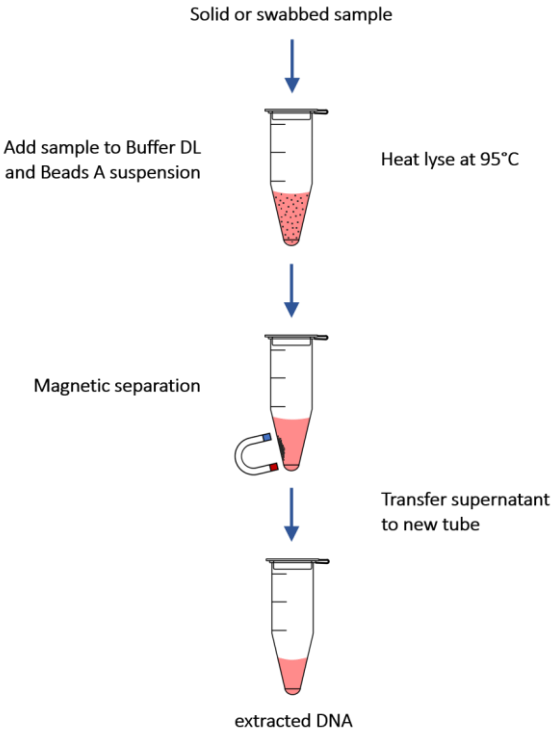
SwiftX™ DNA is designed for rapid extraction of DNA from a wide range of biological samples. The different components of SwiftX™ DNA have specific functionalities within the workflow of DNA extraction.

Buffer EN stabilizes biological cells such as bacteria, epithelial cells, white blood cells, protozoa during cell capturing and enables efficient binding of biological cells to the paramagnetic particles.

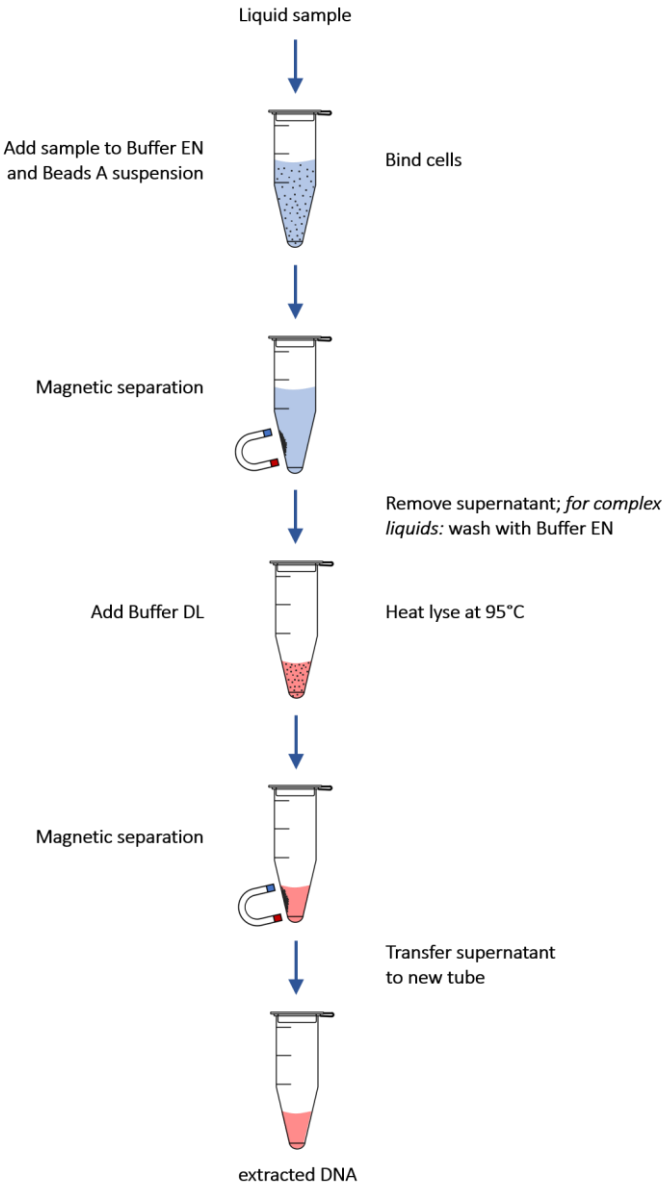
The paramagnetic particles **Beads A** show a broad binding property to cells and proteins. This effect is leveraged in two ways during SwiftX™ DNA-based extraction. Firstly, Beads A enable a species-independent concentration of cells from liquid specimens. Secondly, during and after heat lysis, Beads A are utilized to remove cell debris and other particulate matter from the lysis mixture.

In conjunction with application of heat, **Buffer DL** enables an efficient lysis of viruses, bacteria, protozoa, and animal cells. Buffer DL is fully compatible with a wide range of amplification chemistries.

Direct extraction procedure with reverse purification



Cell capturing with subsequent extraction procedure and reverse purification



Overview of applications and recommended protocols

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<tr><td>Homogenized tissue suspensions</td></tr>
<tr><td>Liquid-based cytology media</td></tr>
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See section "Additional advice and application notes" for use of Proteinase K and further information.

Protocol 1: DNA extraction from swabs and other solid specimens

To do before starting

- ***Read the complete protocol.***
- Heat a water bath, heating block, or thermal shaker to 95°C.
- Heat a second water bath or heating block to 60°C if you intend to perform a Proteinase K incubation step (see separate section on Application Notes).

1. **Depending on the nature of your sample, follow the respective step:**

    a. **In case of swab samples, elute sample by swirling the swab for at least 5 seconds in 100 – 500µL of Buffer DL while pressing the swab towards the tube wall to enhance release of the material.**

    b. **In case of solid samples such as cell pellets, hair follicles or tissue biopsies, add 100 – 500µL of Buffer DL to your sample.**

    Note: The elution volume is dependent on the type and size of the swab or the sample, respectively. Low volumes increase the concentration of the target material, but also the concentration of potential inhibitors.

2. **Shake or vortex Beads A for 30 seconds to ensure homogeneous suspension. Add 30µL of Beads A to the sample mixture. Mix well for 10 seconds by vortexing or inverting the tube.**

3. **Incubate lysis mixture at 95°C for 5 – 15 minutes.**

    Note: You find the recommended lysis time for your application in the table "Overview of applications". If a thermal shaker is used, shake at maximum speed.

4. **Remove sample tube from heating device and mix well for 5 seconds. Remove condensate from the lid before opening by shaking down or tapping the tube on the work bench.**

5. **Place sample tube into a magnetic stand at room temperature for 1 minute to let the magnetic particles separate.**

6. **Open the lid while the tube remains in the magnetic stand and transfer the supernatant into a new tube for storage or use in downstream applications.**

DNA extracts can be stored at -20°C if samples shall be processed later.

Protocol 2: DNA extraction from liquid specimens

This protocol describes the workflow of extracting DNA from up to 1mL liquid samples, such as urine, cell cultures, CSF, throat washes, saliva, or transport media such as Liquid Amies or saline. This protocol includes a cell binding step before DNA extraction.

To do before starting

- ***Read the complete protocol.***
- Heat a water bath, heating block, or thermal shaker to 95°C.
- Heat a second water bath or heating block to 60°C if you intend to perform a Proteinase K incubation step (see separate section on Application Notes).

- Shake or vortex Beads A for 30 seconds to ensure homogeneous suspension.
- Prepare enough **"binding mix"** of Buffer EN and Beads A for your extractions as follows:
    - Determine the volume of sample you want to process (200µL – 1mL)
    - Pipette an equal volume of Buffer EN into a tube, e.g. 500µL of Buffer EN if you want to process 500µL of liquid sample)
    - Add 30µL of Beads A to the Buffer EN
      Note: do not modify the volume of Beads A, it must always be 30µL independent of the volume of Buffer EN
    - Mix well for 5 seconds by vortexing or inverting the tube.

1. **Add the prepared "binding mix" (see above) to a microcentrifuge tube.**

2. **Add the desired volume of liquid sample (200µL – 1mL) to the "binding mix". Mix well by pipetting up and down or by vortexing or inverting the tube for 10 seconds.**

   Note: If highly viscous samples are used, it is recommended to vortex them for 30 seconds at high speed to reduce the viscosity.

3. **Incubate sample tubes at room temperature for 3 minutes.**

4. **Place sample tube into a magnetic stand and let the magnetic particles separate at room temperature for 1 minute.**

5. **Open the lid while the tube remains in the magnetic stand. Remove the supernatant by pipetting. Discard the supernatant.**

6. **Add 50 – 200µL of Buffer DL to the tube and remove the tube from the magnetic stand. Resuspend the magnetic particles by vortexing or shaking for 10 seconds.**

7. **Incubate lysis mixture at 95°C for 5 – 15 minutes.**

   Note: You find the recommended lysis time for your application in the table "Overview of applications". If a thermal shaker is used, shake at maximum speed.

8. **Remove sample tube from the heating device and mix well for 5 seconds. Remove condensate from the lid before opening by shaking down or tapping the tube on the work bench.**

9. **Place sample tube into a magnetic stand and let the magnetic particles separate at room temperature for 1 minute.**

10. **Open lid while the tube remains in the magnetic stand and transfer the supernatant into a new tube for storage or use in downstream applications.**

DNA extracts can be stored at -20°C if samples shall be processed later.

Protocol 3: DNA extraction from complex liquid specimens

This protocol describes the workflow of extracting DNA from up to 200µL of complex liquid samples, such as whole blood and homogenized tissue suspensions, or liquid-based cytology media. This protocol includes a cell binding step and a subsequent wash step before DNA extraction.

To do before starting

- ***Read the complete protocol.***
- Heat a water bath, heating block, or thermal shaker to 95°C.
- Heat a second water bath or heating block to 60°C if you intend to perform a Proteinase K incubation step (see separate section on Application Notes).

- Shake or vortex Beads A for 30 seconds to ensure homogeneous suspension.
- Prepare enough **"binding mix"** of Buffer EN and Beads A for your extractions as follows:
    - Determine the volume of sample you want to process (50 – 200µL)
    - Pipette two volumes of Buffer EN into a tube, e.g. 400µL of Buffer EN if you want to process 200µL of complex liquid sample)
    - Add 30µL of Beads A to the Buffer EN
      Note: do not modify the volume of Beads A, it must always be 30µL independent of the volume of Buffer EN
    - Mix well for 5 seconds by vortexing or inverting the tube.

1. **Add the prepared "binding mix" (see above) to a 1.5mL microcentrifuge tube.**

2. **Add the desired volume of complex liquid sample (50µL – 200µL) to the "binding mix". Mix well by pipetting up and down or by vortexing or inverting the tube for 10 seconds.**

3. **Incubate sample tubes at room temperature for 3 minutes.**

4. **Place sample tube into a magnetic stand and let the magnetic particles separate at room temperature for 2 minutes.**

5. **Open the lid while the tube remains in the magnetic stand. Remove the supernatant by pipetting. Discard the supernatant.**

6. **Add 500µL of Buffer EN to the tube and remove the tube from the magnetic stand. Resuspend the magnetic particles by vortexing or shaking for 10 seconds.**

7. **Place sample tube into a magnetic stand and let the magnetic particles separate at room temperature for 1 minute.**

8. **Open the lid while the tube remains in the magnetic stand. Remove the supernatant by pipetting. Discard the supernatant.**

9. **Add 50 – 200µL of Buffer DL to the tube and remove the tube from the magnetic stand. Resuspend the magnetic particles by vortexing or shaking for 10 seconds.**

10. **Incubate lysis mixture at 95°C for 5 – 15 minutes.**

    Note: You find the recommended lysis time for your application in the table "Overview of applications". If a thermal shaker is used, shake at maximum speed.

11. **Remove sample tube from heating device and mix well for 5 seconds. Remove condensate from the lid before opening by shaking down or tapping the tube on the work bench.**

12. **Place sample tube into a magnetic stand and let the magnetic particles separate at room temperature for 1 minute.**

13. **Open the lid while the tube remains in the magnetic stand. Transfer the supernatant into a new tube for storage or use in downstream applications.**

DNA extracts can be stored at -20°C if samples shall be processed later.

Additional advice and application notes

Processing of tissue, fingernails, hair follicles, and swabs from mucous specimens

To increase the yield of extracted DNA from these sample types, a Proteinase K treatment before the heat lysis step is recommended. For that, perform the following step before the incubation at 95°C: add 5µl of Proteinase K (20mg/mL) to the lysis mixture, mix again, and incubate at 60°C for 10 minutes. If the tissue is not completely dissolved by then, you may incubate another 10 minutes at 60°C. Afterwards, continue as advised in the protocol.

Extraction of DNA from fungi, dermatophytes, and bacteria

The yield of DNA extracted from these cell types can benefit from the use of a Proteinase K treatment before the heat lysis step. For that, perform the following step before the incubation at 95°C: add 5µl of Proteinase K (20mg/mL) to the lysis mixture, mix again, and incubate at 60°C for 10 minutes. Continue as advised in the protocol.

Processing of increased volumes of liquid sample

You can increase the sample volume being processed with Protocols 2 and 3 beyond 1mL. For that, the volume of Buffer EN applied must be increased as well to keep the ratio between sample and Buffer EN as instructed in the respective Protocol. The amount of Beads A is always kept at 30µL. Additional Buffer EN can be ordered separately.

Processing of dirty and complex samples

If a higher purity of the lysate is required, the bound cells can be washed multiple times. For that, repeat Steps 6 to 8 of Protocol 3. Additional Buffer EN can be ordered separately.

It has been reported that an increase in time for the capturing of cells to up to 10 minutes can lead to a higher sensitivity of the DNA extraction procedure.

Always give enough time for magnetic separation of Beads A to allow for optimal cell capture and reverse purification, respectively.

Capturing of intracellular parasites residing in red-blood cells

Red-blood cells (RBC) do not bind to Beads A and can therefore not be concentrated. If intracellular parasites in RBC, e.g. *Plasmodium falciparum*, are intended to be extracted, then the RBC need to be lysed before performing the extraction protocol by adding an appropriate RBC lysis buffer. The hemolyzed sample can then be subjected to DNA extraction using SwiftX DNA Protocol 3.

Limitations

Specimens stored in inactivating media

The cell capturing function of SwiftX™ DNA (see Protocols 2 and 3) cannot be utilized if the cells in the specimen have already been lysed. This is the case if the sample has been transported or stored in an inactivating buffer with high concentration of chaotropic salt, because they tend to lyse biological cells and may reduce the number of intact cells available for capturing. Examples of such transport media are eNAT, PrimeStore MTM, and DNA/RNA Shield. In such case, you need to test whether the target cells are still intact, so they can be captured with Beads A.

Photometric determination of DNA concentrations, e.g. using NanoDrop

Although the reverse purification mechanism actively removes impurities and inhibitors, DNA extracted with SwiftX™ DNA is not as pure as nucleic acids extracted with silica-based purification kits. Thus, photometric determination of A260/A280 ratios and quantification by A260 measurement is of limited use. This is due to the lower limit of quantification of the Nanodrop method, which leads to wrong analysis data especially in samples with low cell count or DNA concentration. Furthermore, Buffer DL absorbs light at 280nm, which generally may lead to misinterpretation of the extraction efficiency. If you still wish to perform a photometric analysis, do not forget to blank your measurement with Buffer DL instead of water or other buffers.

Analysis of extracted DNA using digital PCR

Buffer DL contains low concentrations of detergents. It was observed that DNA samples extracted with SwiftX™ DNA should only be applied to digital PCR analysis if the dPCR system does not require droplet generation, because Buffer DL interferes with droplet stability. If you still wish to use SwiftX™ DNA extracts for digital PCR, we recommend diluting the extracted DNA with water or another detergent-free buffer.

Contacts and disclaimer

This product is owned by:

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