SileksMagNA-G™ Blood DNA Isolation Kit

Catalog No KDBL100

100 DNA isolation reactions from 100 µl blood volume each

Research use only

Shipping: ambient temperature

Storage: +4ºC at least for one year

Kit Contents

Components (all are ready-to-use)	Volume
START Buffer	12 ml
Lysis & Binding Buffer	24 ml
SileksMagNA-G™ Magnetic Particles	1 ml
Wash 1 Buffer	30 ml
Wash 2 Buffer	30 ml
Wash 3 Buffer	30 ml
Final Wash Buffer	30 ml
DNA Elution Buffer	10 ml

Related Equipment and Reagents by Sileks

Cat. No MPD001 SileksMagNA-G™ Magnetic Particles
Cat. No EQRM06 MagRack6 Magnetic Rack
Cat. No EQRM16 MagRack16 Magnetic Rack
Cat. No EQRM40 MagRack40 Magnetic Rack
Cat. No EQMM201 LabMix Mini 201 Mixer

Safety (more information in product material safety data sheet (MSDS))

- When working with chemicals, wear a lab coat, disposable gloves, and protective goggles. Some components may cause health damage if swallowed, breathed, in contact with eyes or skin. Follow MSDS instructions for safety.
- Avoid mixing the kit components with strong acids and alkali.
- In case of contact with eyes, rinse immediately with plenty amount of water. In case of contact with skin, immediately wash the skin with soap and plenty amount of water.

SileksMagNA-G™ Blood DNA Isolation Kit

Advantages

- High DNA yield and consistent results
- High purity (>1.8 OD_{260}/OD_{280}) DNA, excellent for molecular biology applications such as PCR, sequencing, restriction digestion, sequencing on chips, hybridization on chips etc.

Applications

- Rapid and efficient isolation of DNA from whole mammalian blood samples
- DNA isolation from both fresh and frozen blood samples
- The kit can be used for DNA isolation from birds, reptilians or amphibians, however isolation conditions must be optimized

Principle

The SileksMagNA-G™ Blood Isolation Kit is designed for an efficient and rapid (30 min) isolation DNA from whole blood using SileksMagNA-G™ silica-covered magnetic particles. All components of the kit are ready to use and there is no need to supply additional reagents for preparing wash buffers. Provided components are sufficient to isolate high yield DNA from 100 blood samples of 100 µl volume each. Single isolation provides up to 7 µg of pure homogeneous DNA. The kit can be successfully applied for both fresh and frozen mammalian blood samples. The kit can be used for DNA isolation from birds, reptilians or amphibians, however due to different numbers of DNA-containing cells in other organisms, isolation conditions must be optimized individually.

Isolation procedure with SileksMagNA-G™ Blood DNA Isolation Kit consists of a few short and simple steps:

- Sample lysis: blood cells are destroyed and all cell components and nucleic acids are released into the solution
- Binding: nucleic acids bind to magnetic particles
- Washing: residual contaminants are washed away while pure DNA remains bound to magnetic particles
- Drying: incubation at 60°C removes traces of washing buffers
- Elution: purified nucleic acids are eluted from magnetic particles

SileksMagNA-G™ Blood DNA Isolation Protocol

Important Notes

- SileksMagNA-G™ particles are optimized for isolation of genomic DNA and preferably bind long nucleic acids. But it is always obtained some amount of RNA and small/fragmented DNA in your preparation.
- Precipitate may form in some buffers; however, this has no effect on the quality. Warm up the buffer at 50°C to dissolve precipitate if formed. All buffers must be mixed well before use to produce a uniform suspension.
- Frozen blood has to be thawed and mixed well before starting the DNA isolation procedure. Reagents typically used to avoid blood clotting do not interfere with the procedure.
- "*Mix thoroughly*" in the protocol means that the solution must be mixed either by manual pipetting (20 times) or using LabMix mixer for 5 seconds on low/medium speed. "*Mix well*" means that the solution must be shaken 5-10 times.

Protocol

1. Add **120 µl** of well mixed **START Buffer** to 100 µl of blood sample and mix thoroughly.
2. Incubate at room temperature for **5 minutes**. Incubation up to 15 minutes increases DNA yield.
3. In a separate tube mix the following components: **240 µl** of well mixed **Lysis & Binding Buffer** and **7 µl** of well mixed **SileksMagNA-G™ Magnetic Particles**. Mix thoroughly.
4. Add the prepared suspension of magnetic particles to the tube, containing prepared sample. Mix thoroughly. Incubate for **5 minutes** at room temperature, mix once or twice during incubation.
5. To collect magnetic particles with bound DNA, place the tube in a **magnetic rack** for **1 minute**. Discard the supernatant. Be careful not to disturb magnetic particles collected at the tube wall.
6. Place the tube in **a non-magnetic rack**. Add **300 µl** of well mixed **Wash 1 Buffer** and mix thoroughly until a homogeneous suspension is obtained.
7. Place the tube in a **magnetic rack** to collect the particles. Discard the supernatant.
8. Place the tube in a **non-magnetic rack**. Add **300 µl** of well mixed **Wash 2 Buffer** and mix thoroughly until a homogeneous suspension is obtained.
9. Place the tube in a **magnetic rack** to collect the particles. Discard the supernatant.
10. Place the tube in a **non-magnetic rack**. Add **300 µl** of **Wash 3 Buffer** and mix thoroughly until a homogeneous suspension is obtained.
11. Place the tube in a **magnetic rack** to collect the particles. Discard the supernatant.
12. Place the tube in a **non-magnetic rack**. Add **300 µl** of **Final Wash Buffer** and mix thoroughly.

To store DNA safely for a very long time, you can terminate the procedure at this step. Long-term storage of nucleic acids bound to magnetic particles in Final Wash Buffer is the safest option, excellent for DNA conservation or transportation purposes. To use isolated DNA after years of storage at -20ºC the procedures listed below must be carried out to obtain pure DNA.

13. Place the tube in a **magnetic rack** to collect the particles. Discard the supernatant.
14. Incubate the tube in a **thermal block at 60°C for 5 minutes** to dry the pellet of magnetic particles.
15. Add **50 µl** of **DNA Elution Buffer**. Thoroughly resuspend particles until a homogeneous suspension is obtained. If you wish to have higher DNA concentration, use 25 µl of DNA Elution Buffer.
16. Incubate in a **thermal block at 60°C for 5 minutes**.
17. Place the tube in a **magnetic rack** to collect the particles. Transfer the supernatant containing DNA to a fresh tube. To get rid of all particles, repeat the same with the eluate: place in a **magnetic rack** and transfer supernatant into a new tube.
18. The isolated DNA can be stored at -20°C or used directly. DNA is stable and suitable for a long storage. If required, treat it with RNase to remove RNA and use it for subsequent applications. Check the quality of DNA on appropriate agarose gel if needed.

Troubleshooting

Problem	Possible causes	Solution
Low DNA yield	The sample contains too little cells that have DNA or the sample is old.	Take 2x bigger sample volume for isolation, and double all following reagent volumes accordingly. Try to perform elution in smaller volume.
	Too much starting material	Use recommended amounts of samples and all buffers. Too much initial material reduces DNA yield due to suboptimal lysis, binding and wash.
	Incomplete drying of particles before adding DNA Elution Buffer.	Increase the drying time in thermal block after removing Final Wash buffer. Check if all liquids are evaporated before elution.
	Incomplete lysis	Mix the sample more thoroughly after adding START Buffer.
	Too big volume of DNA Elution Buffer used	Adjust optimal amount of DNA Elution Buffer to provide required DNA concentration.
OD_{260}/OD_{280} ratio is too low	Protein contamination	Mix all buffers and all mixes during the procedure like recommended in each step. If too high DNA concentration is suspected, dilute it and measure purity once again.