

sbeadex[®] forensic kit and KingFisher 96 instrument

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Description

The sbeadex[®] forensic kit (Cat. No. 41501 or 41510) has been developed to extract genomic DNA from a wide variety of forensic materials. The magnetic particle based DNA extraction protocol can be easily automated using a KingFisher 96 (Thermo Fisher Scientific) magnetic particle manipulator. Using magnetic rods, which are protected against contamination by a tip comb, the magnetic beads are transferred from one buffer plate to the next during the extraction process. This instrument can process up to 96 samples per run. In addition, the KingFisher 96 instrument is compatible with liquid handling systems and sample handling devices, thus making a hands-free medium to high throughput system a reality.

sbeadex[®] coated magnetic particles bind DNA using a novel two-step binding mechanism in the presence of detergents and salts. After binding and washing steps, the purified DNA is released in the elution buffer. The sbeadex[®] forensic kit is supplied with ready-to-use buffers. The processing time on the KingFisher 96 is approximately 30 minutes.

The method described here is a universal forensic protocol which can be used for a wide range of forensic material without adaptation. Whenever necessary, customisation of the protocol is possible using the software provided with the instrument.

The kit has been developed using the following sample materials:

- Buccal swabs
- Dried blood spots
- Saliva spots on cigarette ends, stamps, bottle necks and drink containers
- Chewing gum
- Hair roots
- Finger nails
- Touched items
- Semen
- Small amounts of fresh blood.

Notes

- A copy of the instrument protocol is available on request (email: extraction@lgcgenomics.com)
- The instrument protocol is compatible with the KingFisher software version 2.6.22
- For tips and advice on how to adapt the instrument protocol for the BindIt[™] software of the KingFisher Flex instrument please email extraction@lgcgenomics.com
- See the sbeadex[®] forensic kit protocol for further information about the kit, limitations of product use, safety information etc.

Equipment and reagents

Product description	Cat. No.	Labware required per run
sbeadex [®] forensic kit (96 tests)	41501	-
sbeadex [®] forensic kit (960 tests)	41510	-
KingFisher 96 magnetic particle processor	5400500*	-
KingFisher 96 DW magnet	24073430*	-
KingFisher 96 tip comb for DW magnets	97002534*	1
KingFisher 96 plate 200 µL	97002540*	1
DW 96 plate, V-bottom, polypropylene	95040450*	4
Ultrapure water (not part of the kit)	User supplied	-
Bottom-pierced deep well plates for 'back pack' centrifugation	Available on request from LGC Genomics	1

Table 1: Equipment and reagents required for DNA extraction using sbeadex[®] forensic kit on KingFisher 96.

* supplied by Thermo Fisher Scientific

Importing instrument protocol

To save the instrument protocol to your computer:

1. Open KingFisher software
2. Select **Cancel** in the Startup window
3. Select **Protocol** → **Import/Export data**.
4. Click **Read file** on the left side of the 'Import/Export protocols' window. An 'Open' window appears
5. Select the protocol you want to import ('**sbx_forensic_KF96.kf2**') and click **Open**
6. The protocol appears in the 'Protocols in file' list
7. Select protocol '**sbx_forensic_KF96.kf2**' in the 'Protocols in file' list and click **Import**
8. A message will appear that the update of the database was successful
9. Now you can start the protocol directly from the software or transfer it to the KingFisher 96 instrument
10. Select **Instrument** → **Send protocol to Instrument**
11. Select the protocol ('**sbx_forensic_KF96.kf2**') from the list 'Protocols for selected instrument' and click **Send protocol**
12. After the transfer of the protocol to the KingFisher 96 instrument a message will appear indicating the successful transfer

Instrument procedure sbeadex® forensic kit

1. Fill the following deep well/ KingFisher plates with sbeadex® forensic kit reagents as specified in table 2:

- Plate 'Binding_Pos1' (Binding buffer FN and sbeadex® particle suspension FN only. Ensure the magnetic particles are thoroughly re-suspended before dispensing.)
 - Plate 'Wash1_Pos2'
 - Plate 'Wash2_Pos3'
 - Plate 'Wash3_Pos4'
 - Plate 'Elution_Pos5'
2. Note protocol **sbx_forensic_KF96** is designed to purify genomic DNA from different forensic materials. Depending on the sample type, the volume of Lysis buffer FN to be added to the sample may differ
 3. Add 115 µL of Lysis buffer FN and 5 µL Protease to each sample and mix thoroughly. Alternatively you can prepare a premix of Lysis buffer FN and Protease for the corresponding sample number and add 120 µL of the premix to each sample. (Note: use the premix immediately after preparation to prevent any decrease / loss in protease activity)
 4. Incubate for 10 minutes at 55°C (recommended incubation time for hair roots: 2 hours at 55°C)
 5. Remove 100 µL of the lysate and add to the prepared plate 'Binding_Pos1', see table 2. (In case of buffer absorbing sample materials, e.g. swabs, removal of the lysate can be simplified by back pack centrifugation - contact extraction@lgcgenomics.com for details)
 6. Select the **sbx_forensic_KF96** protocol on the KingFisher 96 instrument
 7. Load the prepared plates as prompted by the software and start the instrument
 8. After approximately 30 min the protocol will be finished and the genomic DNA is ready for downstream analysis.

Plate name in protocol	Plate type	Well content	Volume
Comb_Pos6	KingFisher 96 KF plate	Tip comb	-
Binding_Pos1	DW 96 plate, V-bottom	Lysate	100 µL
		Binding buffer FN	200 µL
		Particle suspension FN	10 µL
Wash1_Pos2	DW 96 plate, V-bottom	Wash buffer FN 1	300 µL
Wash2_Pos3	DW 96 plate, V-bottom	Wash buffer FN 2	300 µL
Wash3_Pos4	DW 96 plate, V-bottom	Ultrapure water	300 µL
Elution_Pos5	KingFisher 96 KF plate	Elution buffer FN	100 µL

Table 2: Plate filling instructions for KingFisher 96 and sbx_forensic_KF96 protocol.



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