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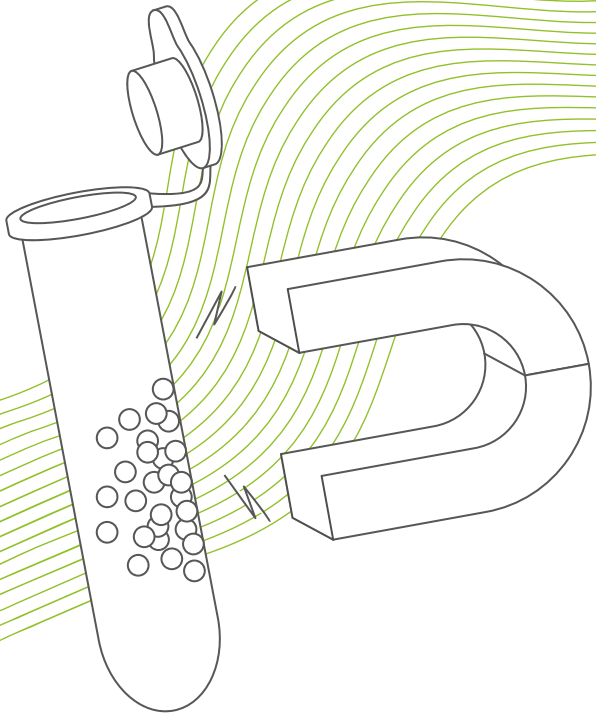

LumiMag® BB, Magnetic Bead DNA Isolation Kit for Blood and Buccal Swab manual

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LumiMag® BB, Magnetic Bead DNA Isolation Kit for Blood and Buccal Swab manual

The kit is designed for rapid and highly-efficient isolation of genomic DNA from whole blood and swabs using magnetic bead technology. It is compatible with KingFisher™ Flex, MagMAX™ Express-96, KingFisher™ Duo Prime, KingFisher™ mL automated platforms and can be used for manual DNA extraction using a magnetic rack.

Extracted DNA is free from protein impurities and can be used for PCR, restriction enzyme digestion, Southern blotting, preparing samples for Sanger sequencing and NGS.

Kit components

Kit component	Count	
	11753 10 minipreps	31753 100 minipreps
B1315, Magnetic Beads (100 mg/mL), 200 uL	1	—
22850, Proteinase K (lyophilized), 10 mg	1	—
B3850, Proteinase K Dilution Buffer, 600 uL	1	—
F4150, Lysis Solution BB, 4 mL	2	—
G6450, Wash Solution MAG A (with GuHCl), 5 mL	1	—
D1315, Magnetic Beads (100 mg/mL), 2 mL	—	1
62850, Proteinase K (lyophilized), 100 mg	—	1
D3850, Proteinase K Dilution Buffer, 1200 uL	—	1
P4150, Lysis Solution BB, 35 mL	—	2
S6450, Wash Solution MAG A (with GuHCl), 50 mL	—	1
K2250, Wash Solution B (Concentrate to be diluted 5x with ethanol 96%), 10.0 mL	1	1

K1350, Elution Buffer (10 mM Tris-HCl, pH 8.5), 10 mL	1	1

Store and transport at room temperature.

Shelf life 12 months.

Required equipment and materials not included in the kit:

- Automated workstation (KingFisher™ Flex, MagMAX™ Express-96, KingFisher™ Duo Prime, KingFisher™ mL)/magnetic rack
- If using a magnetic rack: 1.5 mL Eppendorf™ or analogs tubes (two tubes for DNA isolation from one sample)
- Ethanol 96 %
- Sodium phosphate buffer (PBS) for DNA isolation from epithelium swabs
- Plate shaker (optional)
- Expendable plastics for automated workstation:

KingFisher™ Flex Purification System или MagMAX™ Express-96 Deep Well Magnetic Particle Processor	
95040460	96 Deep-Well Plate Microtiter deep-well
97002540	KingFisher 96 KF Plate 200 µL
97002534	KingFisher 96 tips comb for 96 DW-format
	Adhesive foil for plate sealing

KingFisher™ Duo Prime	
97003530	KingFisher Duo Combi pack for Microtiter Deepwell plate (plates, tips combs, andelution strips)
or	
97003520	KingFisher Duo elution strip
97003500	Tips comb (12-channel) for 96 deep-well Microtiter plate

95040460	96 deep-well Microtiter plate

KingFisher™ mL	
97002141	KingFisher mL Combi 240 (strips and tips combs)
or	
97002111	KingFisher mL tips comb
97002121	KingFisher Strip mL

Before you start

1. Dilute *Wash Solution B* concentrate 5-fold with ethanol 96 % (add 4 volumes of ethanol 96 % to the concentrate volume labeled on the bottle), label the cap with a mark that ethanol has been added.
2. Add 1 mL of *Proteinase K Dissolution Buffer* in a tube with lyophilized Proteinase K, carefully mix, discard drops by centrifugation.

   ! Prepared proteinase K solution should be stored in a freezing chamber at -20 °C.
3. If *Lysate Solution BB* or *Wash Solution MAG A* contains a precipitate, warm them up in a thermostat up to 50 °C and wait until completely dissolved.
4. If using a plate shaker, choose optimal conditions for efficient mixing:
   - Make sure that the plate is tightly fixed in the shaker.
   - Add 1 mL of water to plate wells and cover the plate with foil. Determine the maximal shaker rate with which water from the wells does not splash out from the well to the foil.

Preparation of samples

Epithelium swabs:

1. Add 1 mL of PBS in a clean 1.5 mL tube.
2. Carefully wash the stick with the epithelium swab with PBS. Cut off the upper part of the stick and close the tube.
3. Shake the contents of the tube using vortex for 2–3 min.
4. Use 200 µL of the supernatant as a sample.

Whole blood: Carefully mix blood in the tube and use 200 µL as a sample.

DNA isolation with automated workstation KingFisher™ Flex, MagMAX™ Express-96

1. Prepare plates for automated sample treatment using the instrument according to the table.

   ! To prevent contamination and solution evaporation, seal prepared plates with adhesive foil with the solutions before loading to the instrument.

Plate ID	Plate position in the instrument	Plate type	Reagent	Volume per well
Wash Plate 1	2	Deep-well	Wash solution MAG A	500 µL
Wash Plate 2	3	Deep-well	Wash solution B	500 µL
Elution	4	Standard	Elution buffer	90 µL
Tip Comb	5	Standard	Place the tips comb in the plate	

2. Carefully resuspend the magnetic bead suspension using a vortex.
3. In an individual tube, prepare a mixture of magnetic particles with *Proteinase K* solution (30 µL of mixture per 1 sample = 20 µL of *magnetic particle* suspension + 10 µL of *Proteinase K* solution).
4. Carefully mix the mixture of *magnetic particles* and *proteinase K*. Apply 30 µL aliquots of the mixture in deep plate wells.
5. Add 200 µL of the whole blood/epithelium swab sample (see section *Sample preparation*) to the mixture of *magnetic particles* and *proteinase K*.
6. Carefully mix the contents of plate wells:
   - Using the plate shaker for 2 min at the maximal rate (determined in advance, see Section *Before you start*),
     or
   - Pipette, then incubate for 2 min at room temperature.
7. Add 700 µL aliquots of *Lysate Solution BB* in wells.

8. Proceed to automated sample treatment using the instrument. Choose a relevant program and start it.

   Use MagMAX_CORE program:

Instrument name	Program with heating	Program without heating (optional)
KingFisher™ Flex	MagMAX_CORE_Flex.bdz	MagMAX_CORE_Flex_no_heat.bdz
KingFisher™ 96 MagMAX™ Express-96	MagMAX_CORE_KF-96.bdz	MagMAX_CORE_KF-96_no_heat.bdz

9. According to instrument instructions, load prepared plates with the solutions (item 1) and samples (item 7) to the instrument.

Isolated DNA storage: In a freezing chamber (-20 °C), short-term storage at 4 °C.

DNA isolation using automated workstation KingFisher™ Duo Prime, KingFisher™ mL

1. Carefully resuspend the magnetic bead suspension using a vortex.
2. In an individual tube, prepare a mixture of *magnetic particles* with *Proteinase K* solution (30 µL of mixture per 1 sample = 20 µL of *magnetic particle suspension* + 10 µL of *Proteinase K* solution).
3. Carefully mix the mixture of *magnetic particles* and *proteinase K*. Apply 30 µL aliquots of the mixture in plate/strip wells.
4. Add 200 µL of the whole blood/epithelium swab sample (see section *Sample preparation*) to the mixture of *magnetic particles* and *proteinase K*.
5. Carefully mix the contents of plate/strip wells:
   - Using the plate shaker for 2 min at the maximal rate (determined in advance, see *Section Before you start*),
     or
   - If using KingFisher™ mL: pipette, then incubate for 2 min at room temperature.
6. Add 700 µL aliquots of *Lysate Solution BB* in wells.
7. Apply *Wash Solution MAG A*, *Wash Solution B*, *Elution Buffer*, and samples to corresponding plate/strip wells according to the table below.

   Load tips combs and plates/strips prepared according to the table at the same time.

KingFisher™ Duo Prime				
Row ID	Row in plate	Plate type	Reagent	Volume per well
Sample	A	Deep-well	Sample (prepared in item 6)	930 µL
Wash 1	B		Wash solution MAG A	500 µL
Wash 2	C		Wash solution B	500 µL

Elution	Individual strip	Elution strip	Elution buffer	90 µL
Tip Comb	H	Deep-well	Place the tips comb in the plate	

KingFisher™ mL				
Position ID	Strip position in the instrument	Well	Reagent	Volume per well
Sample	1	Standard	Sample (prepared in item 6)	930 µL
Wash 1	2		Wash solution MAG A	500 µL
Wash 2	3		Wash solution B	500 µL
Elution	4		Elution buffer	90 µL
Tip Comb	—	—	Place the comb in the holder.	

8. Proceed to automated sample treatment using the instrument. Choose the relevant program and start it.

Use MagMAX_CORE program:

Instrument name	Program with heating	Program without heating
KingFisher™ Duo Prime	MagMAX_CORE_DUO.bdz	MagMAX_CORE_DUO_no_heat.bdz
KingFisher™ mL	—	MagMAX_CORE_mL_no_heat.bdz

Isolated DNA storage: In a freezing chamber (-20 °C), short-term storage at 4 °C.

DNA isolation using a magnetic rack

1. Carefully resuspend the magnetic bead suspension using a vortex.
2. In an individual tube, prepare a mixture of *magnetic particles* with *Proteinase K* solution (30 µL of mixture per 1 sample = 20 µL of *magnetic particles* + 10

µL of *Proteinase K* solution).

3. Carefully mix the mixture of *magnetic particles* and *proteinase K*. Introduce 30 µL of the mixture in individual tubes compatible with the magnetic rack.
4. Add 200 µL of the whole blood/epithelium swab sample (see section *Sample preparation*) in the tube with the mixture of *magnetic particles* and *proteinase K*.
5. Carefully mix the tube contents using a vortex. Incubate for 2 min at room temperature.
6. Add 700 µL aliquots of *Lysate Solution BB* in each tube. Carefully mix using a vortex.
7. Place the tubes in the magnetic rack. Wait until *magnetic particles* completely come into the sediment. Carefully discard the supernatant (for example, with a pipette).
8. Add 500 µL of *Wash Solution MAG A*, carefully mix using vortex or by pipetting. Place the tubes in the magnetic rack, remove the supernatant after beads sedimentation.
9. Add 500 µL of *Wash Solution B*, carefully mix using vortex or by pipetting. Place the tubes in the magnetic rack, remove the supernatant after beads sedimentation.
10. Add 90 µL of *Elution buffer*, carefully mix using vortex or by pipetting. Incubate for 5 min at room temperature. Place the tubes in the magnetic rack. The supernatant contains isolated genomic DNA. Transfer the supernatant to a new clean tube.

Isolated DNA storage: In a freezing chamber (-20 °C), short-term storage at 4 °C.

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