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MagMAX™ DNA Multi-Sample Ultra Kit

Frequently asked questions about the buccal swab and 50–µL whole blood workflows

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WARNING! Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves. Safety Data Sheets (SDSs) are available from **www.lifetechnologies.com/support**.

General questions about the pharmocogenetics workflow

What are the most commonly used samples with the workflow?

The workflow is the most commonly used on the following samples:

- Buccal swabs
- 50 µL of whole blood collected in EDTA or sodium citrate

How long is the workflow?

The duration of the workflow depends on the type of sample and on any necessary upfront processing or concentration. On average, the time required to complete the workflow is:

- Total time (including incubations and hands-on time): ~2 hours
- Hands-on time: ~20–30 minutes

What are the downstream applications?

The isolated genomic DNA is suitable for a wide range of downstream applications, including OpenArray® analysis, standard qPCR, genotyping, and copy number variant assays.

What plates are recommended for processing samples?

MagMAX™ Express-96 Deep Well plates (Life Technologies, Cat. no. 4388476) are required for the up-front lysis, binding, and washing on the instrument. MagMAX™ Express-96 Standard plates (Life Technologies, Cat. no. 4388475) are required for the DNA elution.

What plate covers are recommended?

Use adhesive films such as MicroAmp® Clear Adhesive Film (Life Technologies, Cat. no. 4306311) to seal the plates, to prevent cross-contamination during agitations and incubations. Adhesive films are easier to peel off than foil, which is more susceptible to puncture.

What quantification methods are recommended?

Standard curve analysis is the most accurate quantitation method, whereas UV absorbance measurements can be used to assess both the concentration and the quality of the isolated DNA.

- **Standard curve analysis.** Use the TaqMan® RNase P Copy Number Reference Assay (Cat. no. 4403326) for human genomic DNA and the TaqMan® DNA Template Reagents (Cat. no. 401970) to create a standard curve. Refer to *Creating Standard Curves with Genomic DNA or Plasmid DNA Templates for Use in Quantitative PCR* (Pub. no. 4371090).
- **UV absorbance measurements.** Use a NanoDrop® or other comparable instrument. Pure genomic DNA should have an A_{260}/A_{280} ratio of approximately 1.6–2.0.

What samples besides 50–µL whole blood and buccal swabs can be used?

Workflows have been developed and optimized for an array of sample types, including:

- Other blood samples:
  - Larger volumes of whole blood (up to 350 µL) collected in EDTA or sodium citrate (only for use on KingFisher™ Flex-24 instrument)
  - Leukocytes (concentrated from up to 200 µL of whole blood)
  - Blood cards (Whatman FTA)
- Oral rinse samples collected in water, saline, or commercially available mouthwash
- Saliva
- Urine

Questions about the buccal swabs workflow

What buccal swabs are recommended?

Use one of the following types of buccal swabs with foam tips:

- Puritan™ PurFlock™ Ultra Flocked Swabs (Fisher Scientific, Cat. no. 22-025-192)
- Puritan™ HydraFlock® Swabs, standard tip (Puritan, Cat. no. 25–3306–H)
- Sterile Foam Tipped Swabs (Puritan, Cat. no. 25-1506 1PF)
- 4N6FLOQSwabs™, regular tip (Life Technologies, Cat. no. 4473979)

Do not use cotton swabs, as they may contain PCR inhibitors.

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A Thermo Fisher Scientific Brand

Can buccal swabs be shipped after sample collection?

Buccal swabs can be shipped at 25°C or below. Exposure of the swabs to high temperatures (over 30°C) during shipment has been associated with poor performance in the workflow.

To prevent degradation, ensure that buccal swabs are completely dry before shipment. Place the swabs in a paper envelope for storage after collection—paper envelopes are permeable and allows the swabs to dry effectively.

What is the best way to remove the buccal swabs from the lysate?

- Option 1: Transfer the lysate to a new MagMAX™ Express-96 Deep Well Plate (Life Technologies, Cat. no. 4388476).

  This option eliminates contamination risks and saves time. To transfer lysates, set a multi-channel micropipetor to ~300 µL and transfer one row at the time. Each well should contain 200–250 µL after transfer.

- Option 2: Remove the swabs from the plate using forceps.

  Rinse the forceps in 70% ethanol between samples, to prevent cross-contamination. Press the swabs against the side of the well when removing them, to prevent sample loss.

Why is there no RNase treatment?

RNase A treatment is not required in this workflow. RNA that might be present in the samples is likely to be degraded in the course of the experiment and should not interfere with downstream applications.

Questions about the 50–µL whole blood workflow

Can heparin be used to collect samples?

Heparin is not recommended as an anti-coagulant since it can cause inhibition of PCR reactions. We strongly recommend collecting the blood samples in EDTA or sodium citrate, instead.

What type of incubator can be used for the Proteinase K incubation?

Use an incubator with open slats, because it allows adequate flow around the plate wells, to ensure that samples quickly reach and maintain the incubation temperature. Flat heat blocks are not recommended because they do not provide adequate contact with the heating area and the cone-shaped bottom of the wells.

We also recommend checking the internal temperature in the plate: fill a well with 200 µL of PK Mix, place a thermometer or a probe in the well, and monitor the temperature over time.

Will condensation at the top of the wells cause cross-contamination?

You can avoid cross-contamination by taking the following precautions:

- Seal the plates with a plate sealer, making sure to apply pressure around all the wells.
- Let the plates cool down for 2–3 seconds before removing the cover. That will allow to the condensation to recede and will prevent sample splashing.
- Briefly centrifuge the plates for 1–2 minutes at 1500 × *g*.

Why do samples appear cloudy?

On rare occasions, the sample may turn cloudy after adding the Multi-Sample DNA Lysis Buffer. The sample will clear up when isopropanol is added. This will not have any impact on the yield or quality of the DNA.

Revision history

Revision	Date	Description
A.0	November 2014	New document

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