

# MagMAX™ DNA Multi-Sample Ultra 2.0 Kit

High throughput isolation of DNA from buffy coat

Catalog Number A36570

Pub. No. MAN0017323 Rev. E



**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 [thermofisher.com/support](http://thermofisher.com/support).

## Product description

The Applied Biosystems™ MagMAX™ DNA Multi-Sample Ultra 2.0 Kit is developed for scalable, rapid purification of high-quality DNA from a variety of sample matrices. DNA purified with this kit can be used in a broad range of molecular biology downstream applications, such as sequencing, genotyping, and qPCR. One kit is sufficient to process 40–60 buffy coat samples at 50–200 µL input volume. This protocol guides through automated isolation of DNA from buffy coat using the KingFisher™ Flex, KingFisher™ Apex, and the KingFisher™ Duo Prime.

## Contents and storage

Reagents provided in the kit are sufficient for 100 reactions.

**Table 1** MagMAX™ DNA Multi-Sample Ultra 2.0 Kit (Cat. No. A36570)

Component	Quantity	Storage
Enhancer Solution	4.5 mL	15–30°C
Proteinase K	4.5 mL	
Binding Solution	45 mL	
DNA Binding Beads	4.5 mL	
Wash I Solution	110 mL	
Elution Solution	12 mL	

For 1000 reaction volume use Cat. No. A36578 (Proteinase K), A36579 (DNA Binding Beads), A36580 (Wash I Solution), A36581 (Lysis/binding Solution), A36582 (Elution Solution), and A36583 (Enhancer Solution).

## Required materials not supplied

Unless otherwise indicated, all materials are available through [thermofisher.com](http://thermofisher.com). "MLS" indicates that the material is available from [fisherscientific.com](http://fisherscientific.com) or another major laboratory supplier.

Item	Source
<b>Instrument</b>	
<b>Magnetic particle processor (one of the following, depending on quantity/volume of sample to be processed):</b>	
KingFisher™ Duo Prime Magnetic Particle Processor	5400110
KingFisher™ Flex Magnetic Particle Processor 96DW with 96 deep-well head	5400630
KingFisher™ Apex with 96 deep-well head	5400930
<b>Consumables</b>	
<b>Deep-well plates:</b>	
KingFisher™ deep-well 96 plate	95040450
<b>96-well standard plates (for use with KingFisher™ Flex and KingFisher™ Apex only; Tip comb placement):</b>	
KingFisher™ 96 KF plate	97002540
<b>Tip comb, compatible with the magnetic particle processor used:</b>	
KingFisher™ Duo Prime 12-tip comb, for use with KingFisher™ deep-well 96 plate	97003500
KingFisher™ 96 tip comb for deep-well magnets, KingFisher™ Flex and KingFisher™ Apex protocols only	97002534
<b>Elution strip (for use with KingFisher™ Duo Prime only):</b>	
KingFisher Duo elution strip	97003520
KingFisher Duo cap for elution strip	97003540
<b>Equipment</b>	
Adjustable micropipettors	MLS
Multi-channel micropipettors	MLS
<b>Reagents</b>	
Ethanol, 96–100% (molecular biology grade)	MLS
Nuclease-Free Water	AM9932
(Optional) MagMAX™ DNA multi-sample Ultra 2.0 Elution Solution	A36582

Item	Source
<b>Materials</b>	
MicroAmp™ Clear Adhesive Film	4306311

## General guidelines

- Perform all steps at room temperature (20–25°C) unless otherwise noted.
- Precipitates and high viscosity can occur if Enhancer Solution and Binding Solution are stored when room temperature is too cold. If this occurs, warm them at 37°C and gently mix to dissolve precipitates and reduce viscosity. Avoid creating bubbles.
- Yellowing of the Binding and Wash I Solution is normal and will not affect buffer performance.
- Per-plate volumes for reagent mixes are sufficient for one plate plus overage. To calculate volumes for other sample numbers, refer to the per-well volume and add 10% overage.
- *(Optional)* To prevent evaporation and contamination, cover the prepared processing plates with paraffin film or MicroAmp™ Clear Adhesive Film until they are loaded into the instrument.
- The kit is compatible with samples that are collected in K<sub>2</sub>EDTA, K<sub>3</sub>EDTA, Streck DNA, Streck RNA, and Sodium Citrate Blood Collection Tubes (BCT).

**IMPORTANT!** Do not collect blood in Sodium Heparin BCTs because the heparin used as the anticoagulant in these tubes inhibits downstream applications.

- To process up to 200 buffy coat samples, purchase additional Elution Solution (A36582).

## Guidelines for Proteinase K digestion

- Do not pre-mix the Enhancer Solution and Proteinase K.
- Do not change the order of pipetting.

## Guidelines for DNA Binding Bead Mix

- Vortex the DNA Binding Beads thoroughly, combine them with the Binding Solution in a nuclease-free tube, then invert the tube until homogeneous. This mixture can be stored for up to 1 day before aliquoting into the plates.
- Ensure that the beads stay fully mixed within the solution during pipetting.
- Avoid creating bubbles during mixing and aliquoting.

## Before first use of the kit

Prepare Wash II Solution: Make 80% ethanol from 100% absolute ethanol and Nuclease-Free Water.

**IMPORTANT!** The Wash I Solution and Lysis/Binding Solution may develop inert white or brown particulates that float in the solution. Visual particulate is not a cause for concern and does not negatively affect performance.

## Before each use of the kit

Vortex DNA Binding Beads to fully resuspend the beads before each use.

## Perform DNA purification using KingFisher™ Flex (50–200 µL)

Use up to 200 µL of buffy coat sample or up to 2 mL of whole blood equivalent as starting material. Do not exceed 200 µL of buffy coat.

- 1 Set up the instrument**
  1. Ensure that the instrument is set up for processing with the proper magnetic head (96 deep-well) for your application.
  2. Ensure that the proper heat block (96 deep-well, not standard) is installed for your application.
  3. Ensure that the proper program (**MagMAX\_Ultra2\_Buffycoat\_FLEX**) has been downloaded from the product page and loaded onto the instrument.

- 2 Set up the processing plates**

Set up the Wash, Elution, and Tip Comb Plates outside the instrument according to the following table.

Plate ID	Plate position	Plate type	Reagent	Volume per well
Wash I Solution Plate	2	Deep Well	Wash I Solution	1000 µL
Wash II Solution Plate 1	3	Deep Well	Wash II Solution	1000 µL
Wash II Solution Plate 2	4	Deep Well	Wash II Solution	500 µL
Elution Plate	5	Deep Well	Elution Solution	200–300 µL
Tip Comb	6	Place a 96 Deep-well Tip Comb in a Standard Plate		

**Note:** The plates will be loaded onto the instrument immediately after the Sample Plate has been prepared in the following section.

**Note:** To complete all 200 buffy coat sample preparations using the A36750 kit, additional Elution Solution (A36582 ) must be purchased.

### 3 Prepare Sample Plate and digest with Proteinase K

1. Transfer appropriate amount (according to the following table) of Enhancer Solution first, then sample, and lastly Proteinase K, to the appropriate wells of a deep-well plate. This plate will become the Sample Plate.

Enhancer Solution (µL)	Sample Volume (µL)	Proteinase K (µL)
5	50	10
10	100	20
15	150	30
20	200	40

**Note:**

- Do not pre-mix the Enhancer Solution and Proteinase K.
  - Do not change the order of pipetting.
  - Once all components are added, proceed immediately to the instrument processing. There is no need for manual mixing beforehand.
2. Select the program **MagMAX\_Ultra2\_Buffycoat\_FLEX** on the instrument.
  3. Start the run, and load the prepared plates into position when prompted by the instrument. During this on board Proteinase K sample digestion (~20 minutes) prepare the DNA Binding Bead Mix.

### 4 Purify the gDNA

1. Prepare DNA Binding Bead Mix according to the following table.

Component	Volume per well	Volume per plate (96 samples)
Binding Solution	400 µL	42.24 mL
DNA Binding Beads	40 µL	4.22 mL
<b>Total volume</b>	<b>440 µL</b>	<b>46.46 mL</b>

2. When instructed by the instrument (~20 minutes after the run has started), remove the Sample Plate and add 440 µL of DNA Binding Bead Mix to each sample.

**Note:** Remix Binding Bead Mix frequently during pipetting to ensure even distribution of beads to all samples/wells. Mixture is viscous, pipet slowly to ensure that the correct amount is added.

**IMPORTANT!** Avoid creating bubbles during mixing and aliquoting.

3. Immediately place the plate back onto the KingFisher™ Flex and follow the prompts on the instrument to allow the sample processing to proceed.
4. At the end of the run, immediately remove the plates from the instrument and transfer the eluate to the final tube/plate of choice for final storage.

The purified DNA is ready for immediate use. Alternatively, store the plate at -20°C for long-term storage.

## Perform DNA purification using KingFisher™ Apex (50–200 µL)

Use up to 200 µL of buffy coat sample or up to 2 mL of whole blood equivalent as starting material. Do not exceed 200 µL of buffy coat.

- 1 **Set up the instrument**
  1. Ensure that the instrument is set up for processing with the proper magnetic head (96 deep-well) for your application.
  2. Ensure that the proper heat block (96 deep-well, not standard) is installed for your application.
  3. Ensure that the proper program (**MagMAX\_Ultra2\_Buffycoat**) has been downloaded from the product page and loaded onto the instrument.

- 2 **Set up the processing plates** Set up the Wash, Elution, and Tip Comb Plates outside the instrument according to the following table.

Plate ID	Plate position	Plate type	Reagent	Volume per well
Wash I Solution Plate	3	Deep Well	Wash I Solution	1000 µL
Wash II Solution Plate 1	4	Deep Well	Wash II Solution	1000 µL
Wash II Solution Plate 2	5	Deep Well	Wash II Solution	500 µL
Elution Plate	6	Deep Well	Elution Solution	200–300 µL
Tip Comb	1	Place a 96 Deep-well Tip Comb in a Standard Plate		

**Note:** The plates will be loaded onto the instrument immediately after the Sample Plate has been prepared in the following section.

**Note:** To complete all 200 buffy coat sample preparations using the A36750 kit, additional Elution Solution (A36582 ) must be purchased.

- 3 **Prepare Sample Plate and digest with Proteinase K**
  1. Transfer appropriate amount (according to the following table) of Enhancer Solution first, then sample, and lastly Proteinase K, to the appropriate wells of a deep-well plate. This plate will become the Sample Plate.

Enhancer Solution (µL)	Sample Volume (µL)	Proteinase K (µL)
5	50	10
10	100	20
15	150	30
20	200	40

**Note:**

- Do not pre-mix the Enhancer Solution and Proteinase K.
- Do not change the order of pipetting.
- Once all components are added, proceed immediately to the instrument processing. There is no need for manual mixing beforehand.

2. Select the program **MagMAX\_Ultra2\_Buffycoat** on the instrument.
3. Start the run, and load the prepared plates into position when prompted by the instrument. During this on board Proteinase K sample digestion (~20 minutes) prepare the DNA Binding Bead Mix.

- 4 **Purify the gDNA**
  1. Prepare DNA Binding Bead Mix according to the following table.

Component	Volume per well	Volume per plate (96 samples)
Binding Solution	400 µL	42.24 mL
DNA Binding Beads	40 µL	4.22 mL
<b>Total volume</b>	<b>440 µL</b>	<b>46.46 mL</b>

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4 (continued)

2. When instructed by the instrument (~20 minutes after the run has started), remove the Sample Plate and add 440  $\mu$ L of DNA Binding Bead Mix to each sample.

**Note:** Remix Binding Bead Mix frequently during pipetting to ensure even distribution of beads to all samples/wells. Mixture is viscous, pipet slowly to ensure that the correct amount is added.

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**IMPORTANT!** Avoid creating bubbles during mixing and aliquoting.

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3. Immediately place the plate back onto the KingFisher™ Apex instrument and follow the prompts on the instrument to allow the sample processing to proceed.
4. At the end of the run, immediately remove the plates from the instrument and transfer the eluate to the final tube/plate of choice for final storage.

The purified DNA is ready for immediate use. Alternatively, store the plate at  $-20^{\circ}\text{C}$  for long-term storage.

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## Perform DNA purification using KingFisher™ Duo Prime (50–200 µL)

Use up to 200 µL of buffy coat sample or up to 2 mL of whole blood equivalent as starting material. Do not exceed 200 µL of buffy coat.

- 1 **Set up the instrument**
  1. Ensure that the instrument is set up for processing with the proper magnetic head (12 pin) and heat block for your application.
  2. Ensure that the proper program (**MagMAX\_Ultra2\_Buffycoat\_DUO**) has been downloaded from the product page and loaded onto the instrument.

- 2 **Set up the processing plate** Prepare the plate and elution strip according to the following table, preparing the Sample Row in the following section.

Row ID	Plate Row	Reagent	Volume per well
<b>Plate layout</b>			
Sample	A	Sample <sup>[1]</sup>	Varies
Tip Comb	B	Tip Comb	N/A
—	C	Empty	
Wash I Solution	D	Wash I Solution	1000 µL
Wash II Solution	E	Wash II Solution	1000 µL
Wash II Solution	F	Wash II Solution	500 µL
—	G	Empty	
—	H	Empty	
<b>Duo elution strip</b>			
Elution Solution	—	Elution Solution	130 µL

<sup>[1]</sup> See “Prepare Sample Row and digest with Proteinase K” on page 6.

**Note:** The plate will be loaded onto the instrument immediately after the Sample Row has been prepared.

**Note:** To complete all 200 buffy coat sample preparations using the A36750 kit, additional Elution Solution (A36582 ) must be purchased.

- 3 **Prepare Sample Row and digest with Proteinase K**
  1. Transfer appropriate amount (according to the following table) of Enhancer Solution first, then sample, and lastly Proteinase K, to the appropriate wells (Row A) of the deep-well plate that was previously prepared.

Enhancer Solution (µL)	Sample Volume (µL)	Proteinase K (µL)
5	50	10
10	100	20
15	150	30
20	200	40

**Note:**

- Do not pre-mix the Enhancer Solution and Proteinase K.
- Do not change the order of pipetting.
- Once all components are added, proceed immediately to the instrument processing. There is no need for manual mixing beforehand.

### 3 (continued)

2. Select the program **MagMAX\_Ultra2\_Buffycoat\_DUO** on the instrument.
3. Start the run, and load the prepared plate and elution strip into position when prompted by the instrument.

During this on board Proteinase K sample digestion (~20 minutes) prepare the DNA Binding Bead Mix.

### 4 Purify the gDNA

1. Prepare DNA Binding Bead Mix according to the following table.

Component	Volume per well	Volume per plate (12 samples)
Binding Solution	400 µL	5.28 mL
DNA Binding Beads	40 µL	528 µL
<b>Total volume</b>	<b>440 µL</b>	<b>5.81 mL</b>

2. When instructed by the instrument (~20 minutes after the run has started), remove the plate and add 440 µL of DNA Binding Bead Mix to each sample (Row A).

**Note:** Remix Binding Bead Mix frequently during pipetting to ensure even distribution of beads to all samples/wells. Mixture is viscous, pipet slowly to ensure that the correct amount is added.

**IMPORTANT!** Avoid creating bubbles during mixing and aliquoting.

3. Immediately place the plate back onto the KingFisher™ Duo Prime and follow the prompts on the instrument to allow the sample processing to proceed.
4. At the end of the run, immediately remove the plate and elution strip from the instrument.
5. Seal the eluate within the strips using the dedicated caps and store, or transfer the eluate to a final tube/plate of choice for final storage.

The purified DNA is ready for immediate use. Alternatively, store the plate at -20°C for long-term storage.

## Quantitation

To most accurately quantitate gDNA samples isolated from buffy coat, it is recommended to quantitate using a NanoDrop™ spectrophotometer. Another acceptable method is quantitation utilizing qPCR and the Applied Biosystems™ TaqMan™ RNase P Detection Reagents Kit (Cat. No. 4316831).

## Limited product warranty

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**Revision history:** Pub. No. MAN0017323 E

Revision	Date	Description
E	31 July 2024	Important statement added to the user guide under the section "Before first use of the kit".
D.0	11 December 2020	Reduced the buffy coat sample number per kit because more Elution Solution is needed per buffy coat reaction. Support added for KingFisher <sup>™</sup> Apex Purification System.
C.0	15 March 2019	Updated manufacturing address to Vilnius. Added note to address yellowing buffers and viscosity concerns.
B.0	6 December 2017	Addition of quantitation section, and minor edits.
A.0	8 September 2017	New document for MagMAX <sup>™</sup> DNA Multi-Sample Ultra 2.0 Kit.

The information in this guide is subject to change without notice.

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