MagMAX™ DNA Multi-Sample Ultra 2.0 Kit

With MagMAX™ DNA Cell and Tissue Extraction Buffer for high-throughput isolation of DNA from tissues

Catalog Number A45721

Pub. No. MAN0018807 **Rev.** A.0

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**.

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. This protocol describes automated isolation of DNA from tissue using the KingFisher™ Flex and the KingFisher™ Duo Prime instruments.

Contents and storage

Reagents provided in the kit are sufficient for 100 reactions using ≤20 mg of tissue per reaction.

Table 1 MagMAX™ DNA Multi-Sample Ultra 2.0 Kit

Component	Amount	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[1]	12 mL	

[1] The amount of Elution Solution provided in the kit is sufficient for preparing 50 samples. If needed, additional Elution Solution can be purchased separately (Cat. No. A36582).

Table 2 MagMAX™ DNA Cell and Tissue Extraction Buffer

Component	Amount[1]	Storage
MagMAX™ DNA Cell and Tissue Extraction Buffer (Extraction Buffer)	60 mL	15–30°C

[1] Also available separately (Cat. No. A45469).

For 1,000 reactions use Cat. No. A36578 (Proteinase K), A36579 (DNA Binding Beads), A36580 (Wash I Solution), A36581 (Lysis/Binding Solution), 2 × A36582 (Elution Solution), A36583 (Enhancer Solution), and A45470 (MagMAX™ DNA Cell and Tissue Extraction Buffer).

Required materials not supplied

Unless otherwise indicated, all materials are available through **thermofisher.com**. MLS: Fisher Scientific (**fisherscientific.com**) or other major laboratory supplier.

Item	Source
Instrument	
Magnetic particle processor (one of the following, depending on the quantity/volume of sample to be processed): • KingFisher™ Duo Prime Purification System • *For ≤20 mg of tissue*: KingFisher™ Flex Purification System with 96 Deep-Well Head • *For 20–100 mg of tissue*: KingFisher™ Flex Purification System with 24 Deep-Well Head	5400110 5400630 5400640
Consumables	
Deep-well plates: • *For ≤20 mg of tissue*: KingFisher™ Deepwell 96 Plate • *For 20–100 mg of tissue*: KingFisher™ Flex 24 Deep-well plate	95040450 95040470
96-well standard plates (for use with KingFisher™ Flex only; tip comb placement and eluate storage): KingFisher™ 96 KF microplates (200 µL)	97002540
Tip comb, compatible with the magnetic particle processor used: • KingFisher™ Duo Prime 12-tip comb (for use with KingFisher™ Deepwell 96 Plate) • KingFisher™ Duo Prime 6-tip comb (for use with KingFisher™ Flex 24 Deep-well plate) • KingFisher™ 96 tip comb for DW magnets (KingFisher™ Flex protocol only) • KingFisher™ Flex 24 Deep Well Tip Comb and plate (KingFisher™ Flex protocol only)	97003500 97003510 97002534 97002610
Elution strip (KingFisher™ Duo Prime protocol only; elution step): • Elution strip • KingFisher™ Duo Cap for elution strip	97003520 97003540
Equipment	
Adjustable micropipette	MLS
Multichannel micropipette	MLS

Item	Source
Reagents	
PureLink™ RNase A (20 mg/mL) (RNase A)	12091021
Ethanol, 96–100% (molecular biology grade)	MLS
Nuclease-Free Water	AM9932
Materials	
MicroAmp™ Clear Adhesive Film	4306311

Table 3 Additional materials required for tissue homogenization

Item	Source
Equipment	
One of the following homogenization instruments, depending on the procedure used: • Bead-beating instrument for 96-well plates • 10-speed mechanical homogenizer • Vortex with adaptor capable of holding 2-mL bead-beating tubes	MLS
Hybridization oven[1]	MLS
Consumables	
Bead-beating plates or tubes, one of the following[2]: • MagMAX™ Microbiome Bead Plate • MagMAX™ Microbiome Bead Tubes	A42331 A42351
Foil seals[3]	Fisher Scientific™ 14-222-342

[1] Required for Proteinase K digestion methods only.
[2] Required for bead-beating methods only.
[3] Required if you are using a bead-beating plate.

General guidelines

- Perform all steps at room temperature (20–25°C) unless otherwise noted.
- Precipitates and high viscosity can occur if Enhancer Solution, Wash I Solution, and Binding Solution are stored when room temperature is too cold. If this occurs, warm them at 37°C, then gently mix to dissolve precipitates. Avoid creating bubbles.
- Yellowing of the Binding Solution 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.

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.

Guidelines for tissue input

- Tissue samples >20 mg must be homogenized with a 10-speed homogenizer (see "Prepare the tissue samples" on page 4).
- If you are using stabilized samples, such as samples stored in RNA*later*™ Stabilization Solution, we recommend using half of the amount of tissue recommended for fresh or frozen samples.
- Use the appropriate amount of tissue sample according to one of the following tables:

Table 4 Tissue input per well: up to 20 mg

Tissue type	Tissue input per well (96 deep-well format)
Fresh or frozen samples	
Small-to-moderate DNA yields (such as brain or liver)	⩽20 mg
High DNA yields (such as spleen or thymus)	⩽10 mg
Stabilized samples[1]	
Small-to-moderate DNA yields (such as brain or liver)	⩽10 mg
High DNA yields (such as spleen or thymus)	⩽5 mg

[1] For example, samples stored in RNA*later*™ Stabilization Solution.

Table 5 Tissue input per well: up to 100 mg

Tissue type	Tissue input per well (24 deep-well format)
Fresh or frozen samples	
Small-to-moderate DNA yields (such as brain or liver)	20–100 mg
High DNA yields (such as spleen or thymus)	10–50 mg
Stabilized samples[1]	
Small-to-moderate DNA yields (such as brain or liver)	10–50 mg
High DNA yields (such as spleen or thymus)	5–25 mg

[1] For example, samples stored in RNA*later*™ Stabilization Solution.

Before first use of the kit

Note: RNase A is recommended for this protocol and must be purchased separately from the kit. See "Required materials not supplied" on page 1.

Prepare Wash II Solution: Make 80% ethanol from 100% absolute ethanol and nuclease-free water.

Before each use of the kit

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

Prepare the tissue samples

Prepare the tissue samples using one of the following methods.

If you are using a...	Do this...
(Recommended) 10-speed homogenizer	**Note:** See "Guidelines for tissue input" on page 2. 1. Add Extraction Buffer to a fresh tube as indicated. • For 0–20 mg of tissue—add 500 µL of Extraction Buffer. • For >20 mg of tissue—add 500 µL of Extraction Buffer per 20 mg of tissue. 2. Add the tissue sample to the tube containing Extraction Buffer. 3. Homogenize the tissue using a 10-speed mechanical homogenizer, such as a polytron, set at speed 3 or 4. • For ⩽20 mg of tissue—homogenize for 10–20 seconds, or until the tissue is no longer visible. • For >20 mg of tissue—homogenize for 30–40 seconds, or until the tissue is no longer visible. **IMPORTANT!** Avoid homogenization at high speeds, which can cause splashing and foaming. 4. Transfer each lysate to the appropriate well of a deep-well plate (Sample Row/Sample Plate). • For 96 deep-well formats—transfer 500 µL of lysate. • For 24 deep-well formats—transfer up to 1.25 mL of lysate (for 20–50 mg of tissue) or up to 2.5 mL of lysate (for 50–100 mg of tissue). 5. Proceed to the DNA purification procedure. *(Optional)* Store the lysate at –20°C for later use.
High-throughput bead-beating plate (Cat. No. A42331)	**Note:** • Do not exceed 20 mg of tissue per isolation. • For guidelines for tissue input, see Table 4 (page 2). 1. Centrifuge the 96-well bead plate at 3,700–4,000 rpm for 1 minute to collect the beads on the bottom of the plate. Do not remove the seal before the plate is centrifuged. 2. Remove the seal, then add 600 µL of Extraction Buffer to the required wells in the plate. 3. Add the tissue sample to the tube containing Extraction Buffer. **Note:** To enhance DNA yield, mince the tissue before it is added to the tube. 4. Wipe down the top of the wells and edges of the plate with a lab wipe tissue to remove any sample or bead material. 5. Cover the plate with adhesive film. Press down around each well and the edges of the plate with your thumb or adhesive film applicator. **Note:** For detailed instructions on plate sealing, see *MagMAX™ Bead beating plate guidelines Quick Reference* (MAN0018558). 6. Apply two layers of foil seals on top of the adhesive film. Ensure all of the edges and wells of the plate are sealed properly. 7. Clamp the plate onto the bead mill homogenizer, then disrupt (bead-beat) the samples. • For the Omni Bead Ruptor 96—set at 30 Hz for 10 minutes. • For the Mini Bead Beater 96—set for 10 minutes. 8. Remove the plate from the instrument, then centrifuge at 3,700 rpm for 3 minutes. 9. Transfer up to 500 µL of supernatant to the appropriate well of a deep-well plate (Sample Row/Sample Plate). 10. Proceed to the DNA purification procedure *(Optional)* Store the lysate at –20°C for later use.
Bead-beating tube (Cat. No. A42351)	**Note:** • Do not exceed 20 mg of tissue per isolation. • The amount of Extraction Buffer provided in the kit is sufficient for preparing 75 samples, using this procedure. If needed, additional Extraction Buffer can be purchased separately (Cat. No. A45469 or A45470). • For guidelines for tissue input, see Table 4 (page 2). 1. Set up the vortex with the tube adaptor. 2. Add 800 µL of Extraction Buffer to a bead-beating tube. 3. Add the tissue sample to the tube containing Extraction Buffer. 4. Cap the tube, then vortex upside down for 10 seconds to mix the sample with the buffer. 5. Place the tubes on the adaptor, then vortex at 2,500 rpm for 15 minutes.

If you are using a...	Do this...
	6. Remove the tubes from the vortex, then centrifuge at 14,000 × *g* for 2 minutes. 7. Transfer 500 µL of supernatant to the appropriate well of a deep-well plate (Sample Row/Sample Plate). 8. Proceed to the DNA purification procedure. *(Optional)* Store the lysate at –20°C for later use.
2- to 4-hour Proteinase K digestion	**Note:** • Do not exceed 20 mg of tissue (10 mg spleen or thymus) per isolation. If additional tissue is required, divide the sample into 2 or more tubes, then process separately. • For guidelines for tissue input, see Table 4 (page 2). • If you are using frozen tissue, keep the tissue frozen until it is used in step 3. • If you are using fresh tissue, keep the tissue on ice until it is used in step 3. 1. Prepare the Enhancer/Extraction/PK Mix. For each sample, combine the following components for the required number of samples plus 10% overage. • Enhancer Solution—20 µL • Extraction Buffer—500 µL • Proteinase K—40 µL 2. Add 560 µL of Enhancer/Extraction/PK Mix to a 1.5-mL tube. 3. Add the tissue sample to the Enhancer/Extraction/PK Mix, then incubate using one of the following methods. **Note:** To enhance DNA yield, mince the tissue before it is added to the tube. • **Using a thermal mixer—**set at 65°C, then shake at full speed (1,400 rpm) for 30–60 minutes or until the tissue pieces are dissolved. **Note:** To enhance DNA yield and decrease residual RNA, extend the incubation time up to 4 hours. • **Using a hybridization oven—**incubate at 65°C for 2–4 hours. • **Using a shaking water bath, heat block, or tube rotator placed inside of an incubator—**incubate until the tissue pieces are dissolved, vortexing the samples every 5–15 minutes. 4. Centrifuge briefly, then transfer the entire volume of supernatant to the appropriate well of a deep-well plate (Sample Row/Sample Plate). 5. Proceed to the DNA purification procedure. *(Optional)* Store the supernatant at –20°C for later use.
Overnight Proteinase K digestion (for mouse tail tips or ear notch samples only)	**Note:** This procedure is recommended for mouse tail tips or ear notch samples only. 1. Prepare the Enhancer/Extraction/PK Mix. For each sample, combine the following components for the required number of samples plus 10% overage. • Enhancer Solution—20 µL • Extraction Buffer—500 µL • Proteinase K—40 µL 2. Add 560 µL of Enhancer/Extraction/PK Mix to a 1.5-mL tube. 3. Add the sample to the Enhancer/Extraction/PK Mix, then incubate in a hybridization oven at 65°C for 16–18 hours (overnight). **Note:** Ensure the entire sample is submerged in the Enhancer/Extraction/PK Mix. • For mouse tail tips—add one 2- to 3-mm mouse tail tip. • For ear notch samples—add one 2- to 3-mm ear notch. 4. Centrifuge briefly, then transfer the entire volume of supernatant to the appropriate well of a deep-well plate (Sample Row/Sample Plate). 5. Proceed to the DNA purification procedure. *(Optional)* Store the supernatant at –20°C for later use.

Perform DNA purification using KingFisher™ Flex (96 deep-well format)

This method is recommended for ≤20 mg of tissue input.

1 Set up the instrument

a. Ensure that the instrument is set up for processing with the proper magnetic head (96 deep-well) for your application.

b. Ensure that the proper heat block (96 deep-well, not standard) is installed for your application.

c. Ensure that the proper program (**MMX_Ultra2_Cell_Tissue_96_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 of the instrument according to the following table.

Plate ID	Plate position	Plate type	Reagent	Volume per well
≤20 mg of tissue input				
Wash I Solution Plate	2	Deep Well	Wash I Solution	1,000 µL
Wash II Solution Plate 1	3	Deep Well	Wash II Solution	1,000 µL
Wash II Solution Plate 2	4	Deep Well	Wash II Solution	500 µL
Elution Plate	5	Deep Well	Elution Solution	200 µL
Tip Comb	6	Place a 96 Deep-well Tip Comb in a Standard Plate		

Note: Load the plates on the instrument immediately after the Sample Plate is prepared.

3 Prepare DNA Binding Bead Mix

Prepare DNA Binding Bead Mix according to the following table.

Component	Volume per well
Binding Solution	400 µL
DNA Binding Beads	40 µL
Total DNA Binding Bead Mix	**440 µL**

4 Prepare the Sample Plate

a. If needed, transfer 500 µL of each lysate to the appropriate well of a deep-well plate (Sample Plate).

b. (Recommended) Add 10 µL of RNase A to each sample, seal the plate with adhesive film, then mix using a plate shaker set at moderate speed for at least 5 minutes at room temperature.

Alternatively, if a plate shaker is not available, pipet up and down 10 times to mix, then incubate for at least 5 minutes at room temperature.

c. Invert the tube of DNA Binding Bead Mix several times to resuspend the beads, then add 440 µL of DNA Binding Bead Mix to each sample.

Note:
- Remix DNA Binding Bead Mix frequently during pipetting to ensure that the beads are evenly distributed among the samples.
- The mixture is viscous. Pipet slowly to ensure that the correct amount is added to each sample.

d. Immediately proceed to process samples on the instrument (next section).

5 Process samples on the instrument

a. Select the **MMX_Ultra2_Cell_Tissue_96_Flex** program on the instrument.

b. Start the run, then load the prepared plates in their positions when prompted by the instrument.

c. At the end of the run, immediately remove the plate from the instrument, then transfer the eluate to a tube or plate for storage.

Store purified DNA on ice for immediate use, at –20°C for up to 1 month, or at –80°C for long-term storage.

Perform DNA purification using KingFisher™ Flex (24 deep-well format)

This method is recommended for 20–100 mg tissue input.

1 Set up the instrument

a. Ensure that the instrument is set up for processing with the proper magnetic head (24 deep-well) for your application.

b. Ensure that the proper heat block (24-well) is installed for your application.

c. Ensure that the proper program has been downloaded from the product page and loaded onto the instrument.

- **For 20–50 mg tissue input**—use the **MMX_Ultra2_Cell_Tissue_24M_Flex** program.
- **For 50–100 mg tissue input**—use the **MMX_Ultra2_Cell_Tissue_24L_Flex** program.

2 Set up the processing plates

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

Table 6 Plate setup: 20–50 mg tissue input (24 deep-well)

Plate ID	Plate position	Plate type	Reagent	Volume per well
Wash I Solution Plate	2	Deep Well	Wash I Solution	2 mL
Wash II Solution Plate 1	3	Deep Well	Wash II Solution	2 mL
Wash II Solution Plate 2	4	Deep Well	Wash II Solution	1 mL
Elution Plate	5	Deep Well	Elution Solution	0.5 mL
Tip Comb	6	Place a 24 Deep-well Tip Comb in a Standard Plate		

Table 7 Plate setup: 50–100 mg tissue input (24 deep-well)

Plate ID	Plate position	Plate type	Reagent	Volume per well
Wash I Solution Plate	2	Deep Well	Wash I Solution	5 mL
Wash II Solution Plate 1	3	Deep Well	Wash II Solution	4 mL
Wash II Solution Plate 2	4	Deep Well	Wash II Solution	2 mL
Elution Plate	5	Deep Well	Elution Solution	1 mL
Tip Comb	6	Place a 24 Deep-well Tip Comb in a Standard Plate		

Note: Load the plates on the instrument immediately after the Sample Plate is prepared.

3 Prepare DNA Binding Bead Mix

Prepare DNA Binding Bead Mix according to the following table.

Component	Volume per well	
	For 20–50 mg tissue input	For 50–100 mg tissue input
Binding Solution	1 mL	2 mL
DNA Binding Beads	100 µL	200 µL
Total DNA Binding Bead Mix	**1.1 mL**	**2.2 mL**

4 Prepare the Sample Plate

a. If needed, transfer each lysate to the appropriate well of a deep-well plate (Sample Plate).
   - **For 20–50 mg tissue input**—transfer up to 1.25 mL of lysate to each well.
   - **For 50–100 mg tissue input**—transfer up to 2.5 mL of lysate to each well.

b. (Recommended) Add RNase A to each sample, seal the plate with adhesive film, then mix using a plate shaker set at moderate speed for at least 10 minutes at room temperature.

   Alternatively, if a plate shaker is not available, pipet up and down 10 times to mix, then incubate for at least 10 minutes at room temperature.
   - **For 20–50 mg tissue input**—add 25 µL of RNase A to each sample.
   - **For 50–100 mg tissue input**—add 50 µL of RNase A to each sample.

c. Invert the tube of DNA Binding Bead Mix several times to resuspend the beads, then add DNA Binding Bead Mix to each sample.
   - **For 20–50 mg tissue input**—add 1.1 mL of DNA Binding Bead Mix to each sample.
   - **For 50–100 mg tissue input**—add 2.2 mL of DNA Binding Bead Mix to each sample.

   Note:
   - Remix DNA Binding Bead Mix frequently during pipetting to ensure that the beads are evenly distributed among the samples.
   - The mixture is viscous. Pipet slowly to ensure that the correct amount is added to each sample.

d. Immediately proceed to process samples on the instrument (next section).

5 Process samples on the instrument

a. Select the appropriate program on the instrument.
   - **For 20–50 mg tissue input**—select **MMX_Ultra2_Cell_Tissue_24M_Flex**.
   - **For 50–100 mg tissue input**—select **MMX_Ultra2_Cell_Tissue_24L_Flex**.

b. Start the run, then load the prepared plates in their positions when prompted by the instrument.

c. At the end of the run, immediately remove the plate from the instrument, then transfer the eluate to a tube or plate for storage.

Store purified DNA on ice for immediate use, at −20°C for up to 1 month, or at −80°C for long-term storage.

Perform DNA purification using KingFisher™ Duo Prime (96 deep-well format)

This method is recommended for ≤20 mg of tissue input.

1 Set up the instrument

a. Ensure that the instrument is set up for processing with the proper magnetic head (12 pin) and heat block for your application.

b. Ensure that the proper program (**MMX_Ultra2_Cell_Tissue_96_Duo**) has been downloaded from the product page and loaded onto the instrument.

2 Set up the processing plates

Prepare the 96 deep-well plate according to the following table.

Row ID	Plate row	Reagent	Volume per well
Plate layout			
Elution Solution	A	Elution Solution	200 µL
Tip Comb	B	Tip Comb	N/A
—	C	Empty	
Wash II Solution	D	Wash II Solution	500 µL
Wash II Solution	E	Wash II Solution	1,000 µL
Wash I Solution	F	Wash I Solution	1,000 µL
—	G	Empty	
Sample	H	Sample[1]	Varies

[1] See "Prepare the tissue samples" on page 4.

Note: Load the plate on the instrument immediately after the Sample Row is prepared.

3 Prepare DNA Binding Bead Mix

Prepare DNA Binding Bead Mix according to the following table.

Component	Volume per well
Binding Solution	400 µL
DNA Binding Beads	40 µL
Total DNA Binding Bead Mix	**440 µL**

4 Prepare the Sample Row

a. If needed, transfer 500 µL of each lysate to Row H (Sample Row) of the prepared deep-well plate.

b. (Recommended) Add 10 µL of RNase A to each sample, pipet up and down 10 times to mix, then incubate for at least 5 minutes at room temperature.

c. Invert the tube of DNA Binding Bead Mix several times to resuspend the beads, then add 440 µL of DNA Binding Bead Mix to each sample in Row H.

Note:
- Remix DNA Binding Bead Mix frequently during pipetting to ensure that the beads are evenly distributed among the samples.
- The mixture is viscous. Pipet slowly to ensure that the correct amount is added to each sample.

d. Immediately proceed to process samples on the instrument (next section).

5 Process samples on the instrument

a. Select the **MMX_Ultra2_Cell_Tissue_96_Duo** program on the instrument.

b. Start the run, then load the prepared plates in their positions when prompted by the instrument.

c. At the end of the run, immediately remove the plate from the instrument, then transfer the eluate to a tube or plate for storage.

5	Process samples on the instrument *(continued)*	Store purified DNA on ice for immediate use, at –20°C for up to 1 month, or at –80°C for long-term storage.

Perform DNA purification using KingFisher™ Duo Prime (24 deep-well format)

This method is recommended for 20–100 mg tissue input.

1 Set up the instrument

a. Ensure that the instrument is set up for processing with the proper magnetic head (6 pin) and heat block for your application.

b. Ensure that the proper program has been downloaded from the product page and loaded onto the instrument.

- **For 20–50 mg tissue input**—use the **MMX_Ultra2_Cell_Tissue_24M_Duo** program.
- **For 50–100 mg tissue input**—use the **MMX_Ultra2_Cell_Tissue_24L_Duo** program.

2 Set up the processing plates

Prepare the 24 deep-well plates according to one of the following tables.

Table 8 Plate setup: 20–50 mg tissue input (24 deep-well)

Row ID	Plate row	Reagent	Volume per well
Plate 1 layout			
Sample	A	Sample[1]	Varies
Wash I Solution	B	Wash I Solution	2 mL
Wash II Solution	C	Wash II Solution	2 mL
Wash II Solution	D	Wash II Solution	1 mL
Plate 2 layout			
Elution Solution	A	Elution Solution	0.5 mL
Tip Comb	B	Tip Comb	N/A
—	C	Empty	
—	D	Empty	

[1] See "Prepare the tissue samples" on page 4.

Table 9 Plate setup: 50–100 mg tissue input (24 deep-well)

Row ID	Plate row	Reagent	Volume per well
Plate 1 layout			
Sample	A	Sample[1]	Varies
Wash I Solution	B	Wash I Solution	5 mL
Wash II Solution	C	Wash II Solution	4 mL
Wash II Solution	D	Wash II Solution	2 mL
Plate 2 layout			
Elution Solution	A	Elution Solution	1 mL
Tip Comb	B	Tip Comb	N/A
—	C	Empty	
—	D	Empty	

[1] See "Prepare the tissue samples" on page 4.

Note: Load the plates on the instrument immediately after the Sample Row is prepared.

3 Prepare DNA Binding Bead Mix

Prepare DNA Binding Bead Mix according to the following table.

Component	Volume per well	
	For 20–50 mg tissue input	For 50–100 mg tissue input
Binding Solution	1 mL	2 mL
DNA Binding Beads	100 µL	200 µL
Total DNA Binding Bead Mix	**1.1 mL**	**2.2 mL**

4 Prepare the Sample Row

a. If needed, transfer each lysate to Row A (Sample Row) of the prepared deep-well Plate 1.
   - **For 20–50 mg tissue input**—transfer up to 1.25 mL of lysate to each well.
   - **For 50–100 mg tissue input**—transfer up to 2.5 mL of lysate to each well.

b. (Recommended) Add RNase A to each sample, pipet up and down 10 times to mix, then incubate for at least 10 minutes at room temperature.
   - **For 20–50 mg tissue input**—add 25 µL of RNase A to each sample.
   - **For 50–100 mg tissue input**—add 50 µL of RNase A to each sample.

c. Invert the tube of DNA Binding Bead Mix several times to resuspend the beads, then add DNA Binding Bead Mix to each sample.
   - **For 20–50 mg tissue input**—add 1.1 mL of DNA Binding Bead Mix to each sample.
   - **For 50–100 mg tissue input**—add 2.2 mL of DNA Binding Bead Mix to each sample.

   Note:
   - Remix DNA Binding Bead Mix frequently during pipetting to ensure that the beads are evenly distributed among the samples.
   - The mixture is viscous. Pipet slowly to ensure that the correct amount is added to each sample.

d. Immediately proceed to process samples on the instrument (next section).

5 Process samples on the instrument

a. Select the appropriate program on the instrument.
   - **For 20–50 mg tissue input**—select **MMX_Ultra2_Cell_Tissue_24M_Duo**.
   - **For 50–100 mg tissue input**—select **MMX_Ultra2_Cell_Tissue_24L_Duo**.

b. Start the run, then load the prepared plates in their positions when prompted by the instrument.

c. At the end of the run, immediately remove the plate from the instrument, then transfer the eluate to a tube or plate for storage.

Store purified DNA on ice for immediate use, at –20°C for up to 1 month, or at –80°C for long-term storage.

Quantitate the gDNA samples

To most accurately quantitate gDNA samples that are isolated from tissues, we recommend using a NanoDrop™ spectrophotometer. Quantitation can also be performed using qPCR and the Applied Biosystems™ TaqMan™ RNase P Detection Reagents Kit (Cat. No. 4316831).

Limited product warranty

Life Technologies Corporation and/or its affiliate(s) warrant their products as set forth in the Life Technologies' General Terms and Conditions of Sale at **www.thermofisher.com/us/en/home/global/terms-and-conditions.html**. If you have any questions, please contact Life Technologies at **www.thermofisher.com/support**.

Thermo Fisher Scientific Baltics UAB | V.A. Graiciuno 8, LT-02241 | Vilnius, Lithuania
For descriptions of symbols on product labels or product documents, go to **thermofisher.com/symbols-definition**.

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

Revision history: Pub. No. MAN0018807

Revision	Date	Description
A.0	23 September 2019	New document for new sample type.