For general laboratory use.

MagNA Pure FFPET Buffer Set

Version: 01

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Buffer set for deparaffinization and lysis of formalin-fixed, paraffin-embedded tissue (FFPET) samples in automated and manual nucleic acid isolation workflows.

Cat. No. 08 447 144 001	1 kit 2 sets To process up to 48 samples in automated workflows or up to 200 samples in manual pretreatment workflows

Store the set at +15 to +25°C.

1. General Information

1.1. Contents

Vial / Bottle	Cap	Label	Function / Description	Content
1	black	MagNA Pure FFPET Buffer Set, Deparaffinization Reagent	Deparaffinizes FFPET samples.	1 glass bottle, 50 mL
		MagNA Pure FFPET Buffer Set, Deparaffinization Reagent	▪ Barcoded empty bottles. ▪ To transfer deparaffinization reagent for automated workflows or for aliquoting.	2 reagent bottles, for 25 mL each
2	black	MagNA Pure FFPET Buffer Set, Lysis Buffer	Lyses deparaffinized tissue.	2 bottles, 22 mL each
3	black	MagNA Pure FFPET Buffer Set, Isopropanol	Enhance binding of nucleic acids to MGP.	4 bottles, 22 mL each

1.2. Storage and Stability

Storage Conditions (Product)

When stored at +15 to +25°C, the set is stable through the expiry date printed on the label.

Vial / Bottle	Cap	Label	Storage
1	black	Deparaffinization Reagent	Store at +15 to +25°C. ⚠ ***Store the kit and all bottles in an upright position, including the filled, used 25 mL bottles of Deparaffinization Reagent. Do not shake, tilt, rotate, or drop any of the bottles.*** ⚠ ***Keep protected from light.***
2	black	Lysis Buffer	
3	black	Isopropanol	

1.3. Additional Equipment and Reagent required

Standard laboratory equipment and reagents

- Serological pipettes
- Bottles or tubes to prepare the lysis solution
- 1.5 mL safe-lock reaction tubes
- Pipettes with disposable, positive-displacement tips
- Proteinase K*

1.4. Application

The MagNA Pure FFPET Buffer Set is intended for the:

- Deparaffinization and lysis of formalin-fixed, paraffin-embedded tissue samples in automated and manual nucleic acid isolation workflows.

  With an optimized chemistry and protocol, the set eliminates the need to incubate FFPET samples overnight or to use xylene.

1.5. Preparation Time

Assay Time

For manual pretreatment

Hands-on time	Approximately 10 minutes for 8 samples
Total processing time	Approximately 105 minutes for 8 samples

2. How to Use this Product

2.1. Before you Begin

Sample Materials

Use only freshly cut FFPET sections. For one reaction, use up to 6 FFPET sections of 4 or 5 µm using the MagNA Pure 24 Instrument. The overall amount of tissue in one FFPET sample must not exceed 30% of the sample block surface area.

⚠ ***Do not use more than the specified amount of FFPET sample, otherwise the performance of the subsequent nucleic acid purification process may be negatively affected. The yield and quality of the isolated nucleic acids are strongly related to type of tissue, age of sample, and fixation protocol used. Using FFPET sections with high tissue content may significantly lower the nucleic acid recovery. For FFPET samples with high tissue content, or samples with no additional information about their nature, reduce the number of FFPET sections to fit to the requirements of the downstream application.***

Control Reactions

⚠ ***Always run appropriate controls.***

Safety Information

Precautions

⚠ ***Read all instructions carefully before using the MagNA Pure FFPET Buffer Set.***

The reagents in the set contain dangerous or hazardous substances.

- Do not allow the reagents to touch the skin, eyes, or mucous membranes. If contact does occur, wash the affected area immediately with large amounts of water.
- If the reagents are spilled, dilute the spill with water before wiping it up.
- Guanidine hydrochloride contained in the Lysis Buffer is an irritant. Always wear gloves and follow standard safety precautions to minimize contact when handling.

  ⚠ ***Do not mix sodium hypochlorite (found in commercial bleach solutions) or acidic solutions directly with the Lysis Buffer or sample preparation waste. This mixture can produce a highly toxic gas.***
- Good laboratory practice is essential to the proper performance of this reagent set.
- Avoid microbial and nuclease contamination of reagents when removing aliquots from reagent bottles; use sterile disposable pipette tips.
- Closely follow procedures and guidelines provided to ensure proper performance. Any deviation from the procedures and guidelines may affect optimal performance.
- False positive results may occur if carryover of samples is not adequately controlled during sample handling and processing.
- Immediately after use, cap all reagent bottles with their dedicated caps for reuse on the same instrument and store them according to the Instructions for Use.

Laboratory handling

All human sourced material and all resulting waste should be considered potentially infectious and should be handled with universal precautions.

- Thoroughly clean and disinfect all work surfaces with disinfectants recommended by the local authorities.
- Do not eat, drink, or smoke in the designated work area.
- Do not pipette by mouth.
- Wear gloves, coats, and eye protection when handling samples and reagents. Gloves must be changed between handling samples and the MagNA Pure FFPET Buffer Set reagents to prevent contamination when the reagents are used for manual pretreatment of samples. Avoid contaminating gloves when handling samples and controls.
- Wash hands thoroughly after handling samples and FFPET Buffer Set reagents, and after removing the gloves.

Waste handling

- Discard unused reagents and waste in accordance with country, federal, state, and local regulations.

Working Solution

Preparation of lysis solution

1. In an appropriate bottle or tube, premix the Lysis Buffer with Proteinase K.
   – All volumes are for one sample.
   – The required volume depends on the subsequent nucleic acid purification method. According to the number of samples processed, prepare the Lysis solution in bulk. Always add volume for one extra sample, and mix gently by inversion.

⚠ ***Always prepare Lysis solution immediately before each use. Avoid introducing foam or bubbles.***

Lysis Buffer [µL]	Proteinase K [µL]	Lysis solution [µL]
150	15	165
200	20	220

2.2. Protocols

Manual pretreatment of samples

When using a MagNA Pure System, follow the instructions provided in the Instructions for Use for the isolation of nucleic acids from FFPET samples.
For manual pretreatment, follow the steps below. Then continue with a manual or a semi-automated nucleic acid isolation protocol.

⚠ ***Immediately before use, transfer the required Deparaffinization Reagent to one of the empty 25 mL reagent bottles provided with the kit.***

1. For FFPET sample collection:
   ⚠ ***Remove excess paraffin from the FFPET block or FFPET slide prior to collecting FFPET sections when using the MagNA Pure 24 System.***
   – Add up to 6 FFPET sections of 4 to 5 µm to a 1.5 mL tube and cap the tube.
   – Centrifuge the tube at 5,000 × *g* for 30 seconds at +15 to +25°C until samples collect at the bottom of the tubes.
   – Repeat the centrifugation step if necessary.

2. For FFPET deparaffinization, add 300 µL Deparaffinization Reagent directly to the FFPET sample and cap the tube.
   – Incubate while shaking at 2,000 rpm for 5 minutes at +56°C. Alternatively, incubate for 20 minutes at +56°C without shaking.

3. For FFPET lysis incubation, depending on the subsequent nucleic acid purification method, add 200 or 150 µL Lysis solution to 300 µL sample prepared in Step 2.
   – See section, **Working Solution** for information on preparing the Lysis solution.
   ℹ *The Lysis solution migrates to the bottom of the tube and a bilayer forms.*
   – Incubate the capped tubes at +56°C for 60 minutes.
   ⚠ ***Higher tissue input may require additional lysis incubation time.***

4. For FFPET reverse crosslinking, incubate at +80°C for 30 minutes without shaking.

5. For nucleic acid purification, transfer the appropriate FFPET lysate volume, such as 200 µL, or 150 µL, from the bottom of the 1.5 mL tube to a clean sample processing tube, such as a spin column or processing cartridge.

   ℹ *Small amounts of the Deparaffinization Reagent transferred to the sample processing tube or processing cartridge do not affect the purification performance.*

– Immediately proceed with the nucleic acid purification.

3. Troubleshooting

Observation	Possible cause	Recommendation
Inconsistent performance of nucleic acid isolation.	Insufficient FFPET lysis and tissue digestion result in clumping and incomplete transfer of lysate to processing tube or processing cartridge.	Use freshly cut FFPET sections.
		Ensure that the FFPET sample is collected at the bottom of the sample tube. *Always centrifuge the sample tube before the deparaffinization step. If required, repeat the centrifugation step.*
		Additional recommendations for manual workflows: ▪ Avoid transferring too much paraffin when collecting the sample. ▪ During the deparaffinization step, shake the samples while incubating. ▪ Mix the Lysis solution sufficiently before use. ▪ Extend the FFPET lysis incubation when processing FFPET samples with high tissue content.
Slimy bottle of Deparaffinization Reagent.	Long-term storage of Deparaffinization Reagent in the 25 mL reagent bottle.	Transfer the Deparaffinization Reagent from the glass bottle to the empty 25 mL reagent bottle immediately before the first use. ***Do not use Deparaffinization Reagent that was transferred to the 25 mL reagent bottles more than 28 days ago.***
Lack of optimal performance of nucleic acid in downstream applications.	Insufficient sample input or high tissue content per purification sample may result in decreased nucleic acid yield.	For samples with low (<30%) FFPE tissue content, add up to 6 FFPET sections of 5 µm thickness per sample tube.
		For samples with high FFPE tissue content, do not exceed 4 FFPET sections of 5 µm thickness per sample tube.

4. Supplementary Information

4.1. Conventions

To make information consistent and easier to read, the following text conventions and symbols are used in this document to highlight important information:

Text convention and symbols	
Information Note: Additional information about the current topic or procedure.	
Important Note: Information critical to the success of the current procedure or use of the product.	
① ② ③ etc.	Stages in a process that usually occur in the order listed.
❶ ❷ ❸ etc.	Steps in a procedure that must be performed in the order listed.
* (Asterisk)	The Asterisk denotes a product available from Roche Diagnostics.

4.2. Changes to previous version

First version

4.3. Ordering Information

Roche offers a large selection of reagents and systems for life science research. For a full overview of related products and manuals, please visit and bookmark our homepage lifescience.roche.com.

Product	Pack Size	Cat. No.
Reagents, kits		
Proteinase K, recombinant, PCR Grade	1.25 ml, > 50 U/ml	03 115 887 001
	5 ml, > 50 U/ml	03 115 828 001
	25 ml, > 50 U/ml	03 115 844 001

4.4. Trademarks

MAGNA PURE is a trademark of Roche.
All other product names and trademarks are the property of their respective owners.

4.5. License Disclaimer

For patent license limitations for individual products please refer to:
http://technical-support.roche.com.

4.6. Regulatory Disclaimer

For general laboratory use.

4.7. Safety Data Sheet

Please follow the instructions in the Safety Data Sheet (SDS).

4.8. Contact and Support

If you have questions or experience problems with this or any Roche product for Life Science, please contact our Technical Support staff. Our scientists are committed to providing rapid and effective help.
Please also contact us if you have suggestions for enhancing Roche product performance or using our products in new or specialized ways. Such customer information has repeatedly proven invaluable to the research community worldwide.
To ask questions, solve problems, suggest enhancements or report new applications,
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