PAXgene® Tissue The better the source, the more to explore.



PAXgene® Tissue System Explore more at www.PreAnalytiX.com



Contents

- O3 The PAXgene Tissue System
- O4 An Introduction to the PAXgene Tissue System
- 06 H&E Staining
- 07 Immunohistochemistry
- 08 RNA Purification
- 10 miRNA Purification
- 12 DNA Purification
- 14 The PAXgene Tissue Workflow

The PAXgene Tissue System includes all kits shown in this brochure. For research use only. Not for use in diagnostic procedures.

The PAXgene Tissue System

Traditional tissue fixation methods have been of limited use for molecular analysis. PreAnalytiX now offers a system for simultaneous stabilization of molecular content and preservation of morphology.

- Preserves morphology and nucleic acids
- Stabilizes and purifies RNA, miRNA, and DNA
- Formalin-free

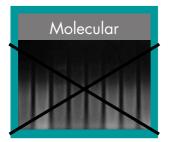
Before PAXgene Tissue

Formalin



Biomolecule stabilization





Molecular

Formalin does not stabilize biomolecules:

Formalin introduces crosslinks between biomolecules, which lead to degradation and chemical modification of nucleic acids.

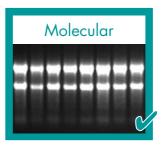
Biomolecule stabilization does not preserve histomorphology:

Stabilization methods for preservation of nucleic acids in tissue, such as RNA*later*® RNA Stabilization Reagent, do not preserve histomorphology.

PAXgene Tissue System

From the same sample





The PAXgene Tissue System stabilizes biomolecules AND preserves histomorphology:

Without crosslinking, the PAXgene Tissue System enables histological analysis and extraction of high-quality RNA, miRNA, and DNA from the same specimen.

An Introduction to the PAXgene Tissue System

- PAXgene Tissue Container for collection, stabilization, storage, and transportation of human tissue specimens
- PAXgene Tissue Kits for purification of total RNA, miRNA, or DNA



Benefits of the PAXgene Tissue System:

- Standardization of tissue collection, fixation, and processing workflow
- Equivalent histological details in H&E staining compared to traditional methods
- Compatible with immunohistochemistry methods
- Preservation of intact RNA without crosslinking for improved molecular results
- Preservation of high-molecular-weight DNA for superior results compared to formalin
- High-quality DNA for sensitive downstream applications, including multiplex or long-range PCR
- Purification of RNA, miRNA, and DNA from one sample

Prefilled dual-chambered container for fixation and stabilization of tissue.

Chamber 1: PAXgene Tissue Fix

- Rapid penetration and fixation
- Fixation in 2-4 hours (up to 12 hours)

 After fixation, tissue is transferred to chamber 2.

Chamber 2: PAXgene Tissue Stabilizer

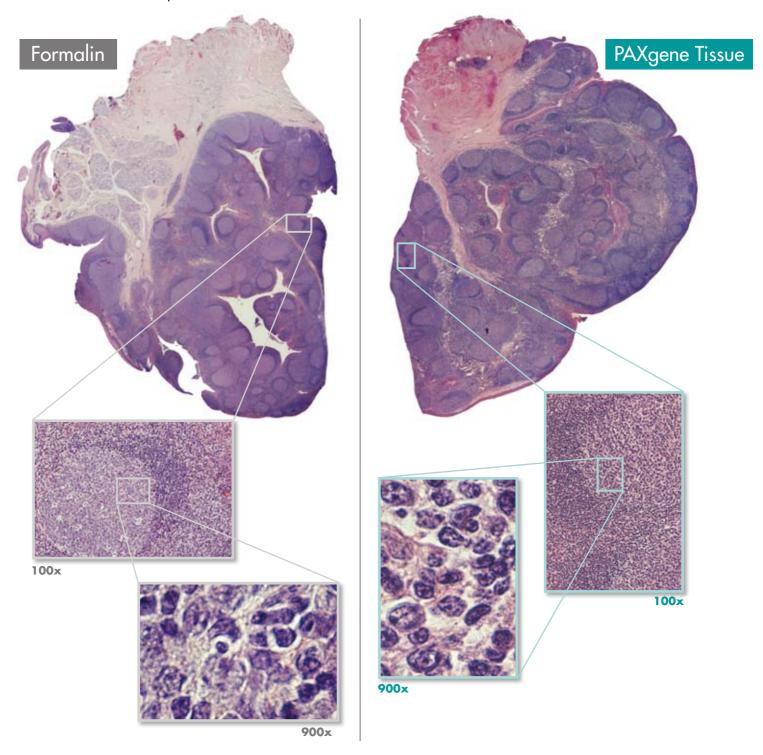
- Stabilization of biomolecules and histomorphology
- Sample can be transported and stored:
 - Up to 7 days at room temperature
 - Up to 4 weeks at 2-8°C
 - For months when frozen at -20°C
 - For years when frozen at -80°C

Stabilized samples can be embedded in paraffin for histological studies. Nucleic acids can be isolated from the stabilized samples using PAXgene Tissue Kits before or after embedding in paraffin.



H&E Staining with the PAXgene Tissue System

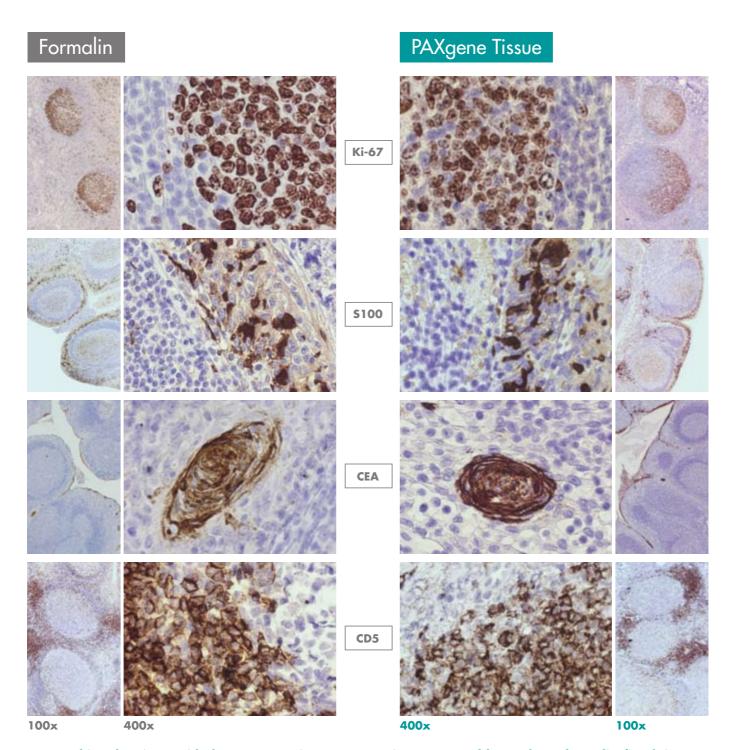
- Intact histomorphology
- Staining pattern comparable to formalin-fixed tissue
- Chromatin structure preserved



H&E staining with the PAXgene Tissue System gives comparable results to formalin-fixed tissue: A cross-section of a human palatine tonsil was divided in half. One half was fixed in the PAXgene Tissue Container, and the other half was fixed in neutral buffered formalin. The fixed tissues were embedded in paraffin, sectioned, and stained with hematoxylin and eosin.

Immunohistochemistry with the PAXgene Tissue System

- Staining intensities comparable to formalin-fixed paraffin-embedded tissue (FFPE)
- Antigen retrieval steps eliminated in 3 out of 4 cases



Immunohistochemistry with the PAXgene Tissue System gives comparable results to formalin-fixed tissue:

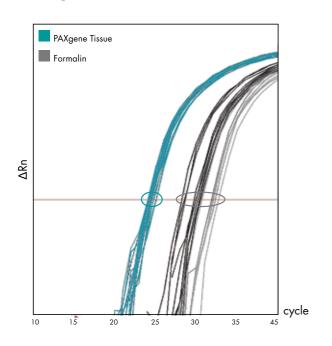
Human palatine tonsil tissue was fixed in PAXgene Tissue Containers or with neutral buffered formalin and embedded in paraffin. Primary antibodies to the indicated antigens were linked to a streptavidin-peroxidase conjugate by a biotinylated secondary antibody (LSAB method). Sections were counterstained with hematoxylin. Antigen retrieval was necessary for all antigens with the formalin-fixed samples. Antigen retrieval was only required with CD5 when using the PAXgene Tissue System.

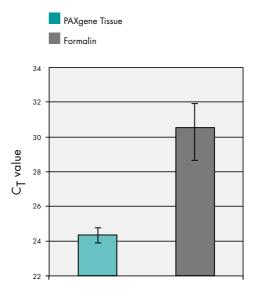
RNA Purification with the PAXgene Tissue System

- Isolation of intact RNA
- No enzyme inhibition
- Reliable quantitative real-time RT-PCR
- High-molecular-weight RNA
- Amplification of RNA up to 1 kb



Improved real-time RT-PCR with the PAXgene Tissue System compared to formalin fixation

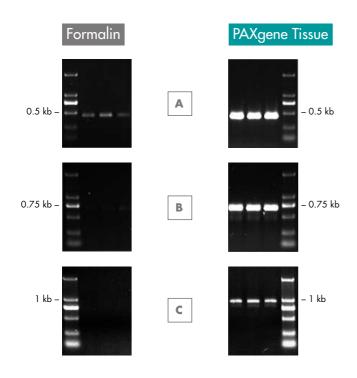




β-actin real-time RT-PCR from human palatine tonsil RNA:

Triplicate RNA extractions per tissue specimen from human palatine tonsils from 6 different patients were amplified. Samples were treated with the PAXgene Tissue System reagents or neutral buffered formalin (NBF). Total RNA was extracted from sections of paraffinized tissue using the PAXgene Tissue RNA Kit or, for the NBF treated tonsils, a commercially available kit for FFPE tissues.

More effective, reliable, and reproducible results in RT-PCR with the PAXgene Tissue System compared to formalin



One-step RT-PCR amplification of human single copy genes of increasing amplicon length:

RNA from human palatine tonsil tissue was treated with neutral buffered formalin or the PAXgene Tissue System.

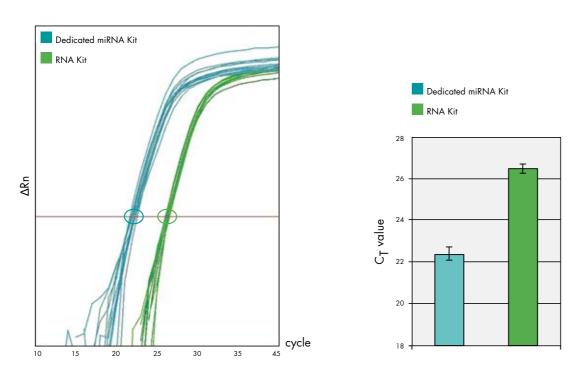
- A: 439 bp of TAFII
- **B:** 690 bp of alpha catenin
- C: 960 bp of hnRNPA2

miRNA Purification with the PAXgene Tissue System

- Optimized protocol for copurification of all RNAs
- Enrichment for small RNAs (≥18 nucleotides)
- No enzyme inhibition
- Reliable quantification of miRNA
- Highly reproducible



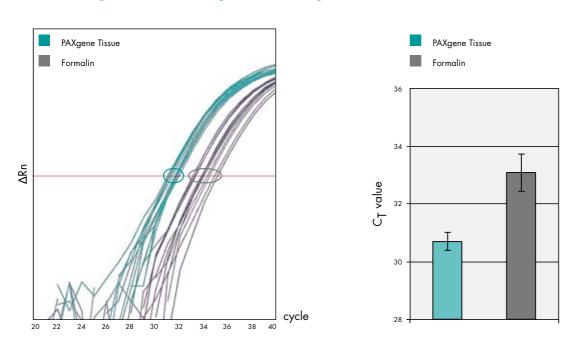
Improved copurification of small RNAs with the dedicated PAXgene Tissue miRNA Kit



Real-time RT-PCR of miR-10a. Lower C_T values indicate higher yields of miRNA:

Amplification was performed with RNA from PAXgene Tissue fixed, paraffin-embedded tissue with or without optimization of small RNA copurification. miRNA was analyzed using the QIAGEN® miScript PCR System.

Improved miRNA quantification in real-time RT-PCR with the PAXgene Tissue System compared to formalin fixation



Real-time RT-PCR of miR-103. Lower C_T values compared to FFPE tissue:

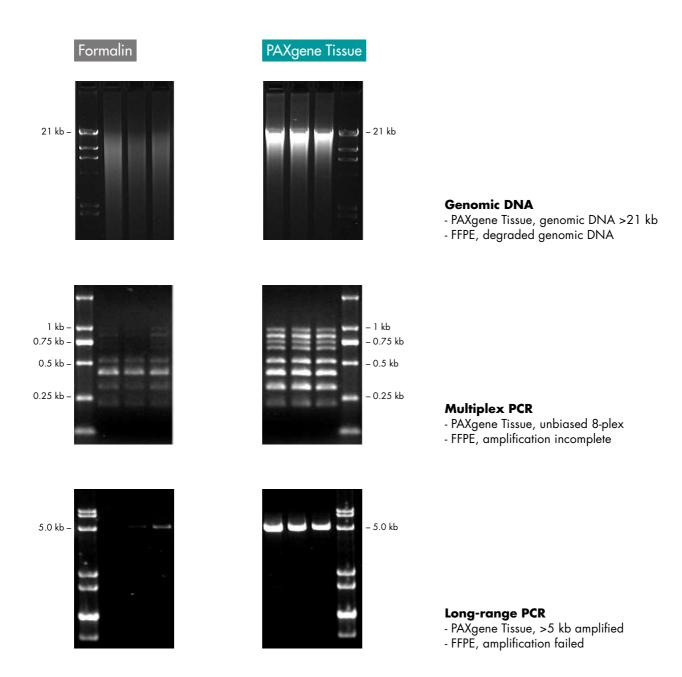
Amplification was performed using 5 ng of total RNA extracted from sections of paraffinized tissue using the PAXgene Tissue miRNA Kit, optimized for copurification of small RNAs or, for the NBF treated tissue, a commercially available miRNA Kit for FFPE samples.

DNA Purification with the PAXgene Tissue System

- High-molecular-weight DNA
- Unbiased amplification in multiplex PCR
- Superior performance in long-range PCR



Intact genomic DNA and improved performance in PCR with the PAXgene Tissue System compared to formalin fixation



Agarose gel electrophoresis. Genomic DNA, multiplex, and long-range PCR:

Human palatine tonsil specimens were treated with the PAXgene Tissue System or with neutral buffered formalin (NBF). Total DNA was extracted from sections of paraffinized tissue using the PAXgene Tissue DNA Kit or, in the case of the NBF treated tonsils, a commercially available DNA kit for FFPE tissues.

The PAXgene Tissue Workflow

Resection tissue and place into a standard tissue cassette (tissue cassette to be supplied by user). Remove screw cap/rack assembly. Snap tissue cassette into the rack. Submerge the rack holding the tissue cassette into chamber 1 containing PAXgene Tissue Fix. Fix for 2 to 4 hours* (maximum fixation 18 hours). After fixation, remove the rack with the tissue cassette from chamber 1 and transfer it into chamber 2 containing PAXgene Tissue Stabilizer. Screw the cap back into place. The container is ready for storage or transport with morphology and nucleic acids preserved. After processing and paraffin embedding, the block of tissue is ready for sectioning. Sections can be used for anatomical pathology or for purification of RNA, miRNA, or DNA.

^{*} Optimal results achieved within 2 to 4 hours. Longer fixation times may compromise RNA quality.

For up-to-date licensing information and product-specific disclaimers about QIAGEN products, see the respective QIAGEN kit handbook or user manual. QIAGEN kit handbooks and user manuals are available at www.qiagen.com or can be requested from QIAGEN Technical Services or your local distributor.

Trademarks:

PAXgene[®], PreAnalytiX[®] (PreAnalytiX GmbH); QIAGEN[®], MinElute[®] (QIAGEN Group).

RNA*later*® is a trademark of AMBION, Inc., Austin, Texas and is covered by various U.S. and foreign patents.

1055346 05/2009 © 2009 PreAnalytiX, all rights reserved.

PreAnalytiX GmbH
Feldbachstraße
CH-8634 Hombrechtikon

www.preanalytix.com



Ordering Information

For more ordering information and online ordering go to: www.qiagen.com

Product	Contents	Cat. no.
PAXgene Tissue Containers — acid stabilization of human tissue PAXgene Tissue Containers (10)	for collection, fixation, and nucleic issues For collection, fixation, and stabilization of 10 samples: 10 Prefilled Reagent Containers, containing PAXgene Tissue Fix and PAXgene Tissue Stabilizer	765112
PAXgene Tissue RNA Kit — for purification of total RNA from tissues fixed and stabilized in PAXgene Tissue Containers PAXgene Tissue RNA Kit (50) For 50 RNA preps: PAXgene RNA MinElute®		765134
	Spin Columns, PAXgene Shredder Spin Columns, Processing Tubes, Microcentrifuge Tubes, Carrier RNA, RNase-Free DNase, and RNase-Free Buffers; to be used in conjunction with PAXgene Tissue Containers	
PAXgene Tissue miRNA Kit — for purification of microRNA and total RNA from tissues fixed and stabilized in PAXgene Tissue Containers		
PAXgene Tissue miRNA Kit (50)	For 50 RNA preps: PAXgene RNA MinElute Spin Columns, PAXgene Shredder Spin Columns, Processing Tubes, Microcentrifuge Tubes, Carrier RNA, RNase-Free DNase, and RNase-Free Buffers; to be used in conjunction with PAXgene Tissue Containers	766134
PAXgene Tissue DNA Kit — for purification of DNA from tissues fixed and stabilized in PAXgene Tissue Containers		
PAXgene Tissue DNA Kit (50)	For 50 DNA preps: PAXgene DNA Mini Spin Columns, Processing Tubes, Microcentrifuge Tubes, Carrier RNA, and Buffers; to be used in conjunction with PAXgene Tissue Containers	767134

Discover more about the PAXgene Tissue System at: www.preanalytix.com

