

HistoGene LCM Frozen Section Staining Kit

Prepare frozen tissue for laser capture microdissection while maintaining high-quality RNA



Benefits

- Simplified tissue staining and dehydration, typically in less than 15 minutes
- Preserves RNA quality
- Validated formula provides good contrast

The Applied Biosystems™ HistoGene™ LCM Frozen Section Staining Kit comes complete with all the reagents and supplies needed for preparing frozen tissue sections for laser capture microdissection (LCM). Dehydration and staining solutions, slide jars— even specially coated glass slides are included, along with a detailed, step-by-step protocol (Figure 1). LCM certified, the HistoGene kit permits consistent preparation of quality samples ready for laser capture.

Provide superior staining while preserving RNA

The Applied Biosystems™ HistoGene™ Stain is a special solution designed to stain tissues for LCM that are subsequently used as sources of RNA. It is a fast-penetrating stain that provides good contrast by

Figure 1. The HistoGene LCM Frozen Section Staining Kit.

differential staining of nuclei (purple) and cytoplasm (light pink) (Figure 2). By minimizing the exposure of tissues to water, where nucleases may be activated, the HistoGene process helps to preserve RNA integrity that may be otherwise compromised when using longer staining protocols.

Retain low-abundance mRNA

RT-PCR analysis of specific genes from cells captured from tissues processed using the HistoGene kit shows retention of both low-abundance mRNA and higher-abundance species

(Figure 3). The mRNA profile of samples prepared with the HistoGene kit appears free of degradation.

Maintain RNA integrity

Tissues prepared for LCM using HistoGene kit reagents, supplies, and instructions yield high-quality RNA. Data from electrophoretic analysis of total RNA, and RT-PCR analysis of specific genes confirm retention of RNA integrity.

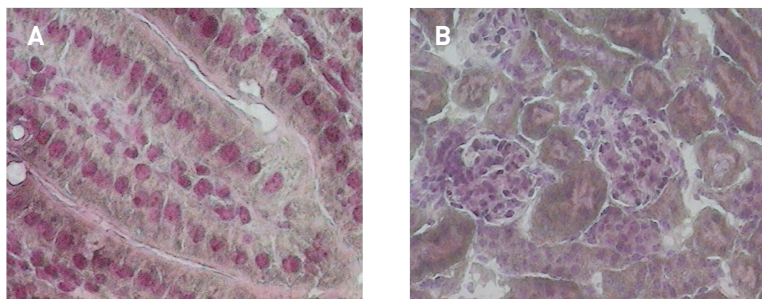


Figure 2. Examples of tissues prepared with the HistoGene LCM Frozen Section Staining Kit. (A) Mouse small intestine. (B) Mouse kidney.

Produce superior microarrays

The HistoGene LCM Frozen Section Staining Kit is one member of a growing family of Applied Biosystems products designed to seamlessly work together to produce high-quality microarray expression results (Figure 4). Using the Applied Biosystems™ PicoPure™ RNA Isolation Kit, RNA can be recovered with high yield in a minimal volume from as few as 10 cells. Picogram quantities of RNA in samples prepared using the kit can be amplified to microgram amounts using the Applied Biosystems™ RiboAmp™ RNA Amplification Kit, providing RNA ready for labeling and hybridization to microarrays. When used in combination, Applied Biosystems instruments and kits help increase the reliability and reproducibility of gene expression studies performed on microarrays.

Obtain high-quality RNA from many tissue types

The HistoGene kit has been validated by examining RNA integrity on an expanding list of tissue types. All tissues tested to date using the HistoGene kit have yielded high-quality RNA (Table 1).

Ordering information

Product	Quantity	Cat. No.
HistoGene LCM Frozen Section Staining Kit	72 slides	KIT0401
HistoGene Refill Kit (includes dehydration chemicals and stain only)	72 tissue preps	KIT0419
HistoGene LCM Immunofluorescence Staining Kit	32 slides	KIT0420
HistoGene Staining Solution	8 mL	KIT0415

Table 1. Tissues validated with the HistoGene kit.

Human	Mouse	
Foreskin	Kidney	Salivary gland
Ileum	Liver	Thymus
Jejunum	Brain	Small intestine

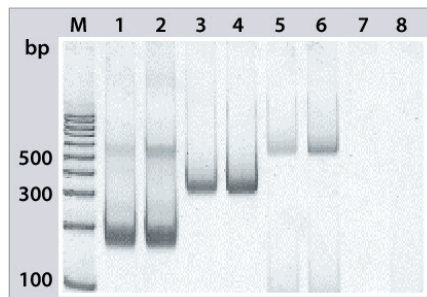


Figure 3. Detection of low-abundance mRNA in samples prepared with the HistoGene kit.

RT-PCR was performed on RNA from 500 cells captured from mouse liver and kidney. Equal quantities of cDNA were analyzed with three primer sets: elongation factor 1- α (EF-1 α , high-abundance gene, ~3,000 copies/cell, 187 bp), glyceraldehyde-3-phosphate dehydrogenase (GAPDH, medium-abundance gene, 300–3,000 copies/cell, 357 bp), and protein phosphatase 1 (PP1, low-abundance gene, <300 copies/cell, 498 bp). Samples were run on a 6% polyacrylamide gel and stained using Invitrogen™ SYBR™ Gold Nucleic Acid Gel Stain. M: molecular weight markers. Lane 1: kidney EF-1 α . Lane 2: liver EF-1 α . Lane 3: kidney GAPDH. Lane 4: liver GAPDH. Lane 5: kidney PP1. Lane 6: liver PP1. Lane 7: liver EF-1 α no-RT control. Lane 8: no-RT template control.

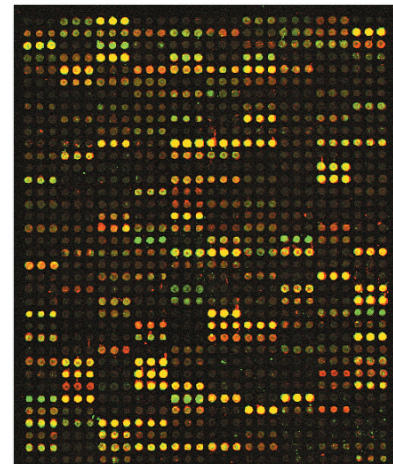


Figure 4. Expression microarray of RNA from LCM samples.

Mouse kidney sections were prepared using the HistoGene kit and cells captured from the collecting ducts and glomeruli using the Applied Biosystems™ ArcturusXT™ LCM System and Applied Biosystems™ CapSure™ HS LCM Caps. RNA was isolated using the PicoPure RNA Isolation Kit and amplified using the RiboAmp RNA Amplification Kit. Amplified RNA was converted to Applied Biosystems™ Cy⁵ dye-labeled cDNA and hybridized onto separate microarrays composed of 3,000 mouse ESTs and scanned. Spot images were converted to pseudocolor and overlaid. Red spots show genes expressed in duct cells only, green spots show genes expressed in glomeruli only, and yellow spots show genes expressed in both structure types.

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