KPL Formamide Hybridization Buffer

Catalog No.	Size
5960-0023 (50-86-10)	2 x 120 mL

DESCRIPTION

KPL Formamide Hybridization Buffer is a formamide-based solution for use in the pre-hybridization and hybridization of biotinylated nucleic acid probes to nucleic acids fixed on membranes. As a pre-hybridization solution, the buffer blocks sites on the membrane and prevents nonspecific binding of biotinylated probe. As a hybridization solution, the buffer facilitates binding of biotinylated probes to homologous RNA or DNA on a membrane.

FORM/STORAGE/STABILITY

KPL Formamide Hybridization Buffer consists of 2 x 120 mL Store at 2-8°C. Stable for a minimum of one year from date of receipt when stored at 2-8°C. KPL Formamide Hybridization Buffer will precipitate upon storage at cold temperatures. **For best results**, warm buffer to 37°C until all components are in solution, and aliquot into DNase/RNase free tubes.

CONTENT

KPL Formamide Hybridization Buffer is a formamide-based solution prepared with DNase/RNase free reagents.

APPLICATIONS

KPL Formamide Hybridization Buffer may be used for detection of biotinylated probes on membranes in procedures such as Northern and Southern Blotting, and dot blots.

USE

Hybridization temperature should be optimized between 42°C-68°C depending on the melting temperature of the probe. Prior to use, sheared and denatured KPL Herring Sperm DNA must be added to a final concentration of 100 µg per mL.

Pre-hybridization:

1. Warm at least 0.1 ml of Formamide Hybridization Buffer per cm^2 of membrane. A minimum volume of 3 mL is required when using a hybridization bottle (4 cm diameter x 14 cm long). If using less than 3 mL, a hybridization bag is recommended.
2. Add sheared, denatured herring sperm DNA to the buffer to a final concentration of 100 µg per mL.
3. Place membrane in a hybridization bottle or bag and add the prepared pre-hybridization buffer.
4. Incubate for 1 hour at hybridization temperature (42°C-68°C).

Denaturation and Hybridization:

1. For DNA probes: heat probe to 95°C for 10 minutes and immediately place on ice. For RNA probes: heat probe to 68°C for 10 minutes and immediately place on ice.

For best results, do not denature the probe using alkaline treatment.

2. Add the biotinylated probe directly to the prehybridization solution at a concentration of 50 ng per mL. Be careful to pipet the probe into the solution and not directly onto the blot.

Note: Probe should be quantitated as described in the KPL Detector PCR DNA Biotinylation Kit.

3. Incubate overnight at the appropriate hybridization temperature (42°C-68°C).
4. Stringency washes and detection protocols should be carried out according to standard protocols.

PRODUCT SAFETY AND HANDLING

This product is considered hazardous as defined by The Hazard Communication Standard (29 CFR 1910.1200). Avoid contact with skin and eyes. In case of contact or spillage, clean with copious amounts of water. Consult product SDS for disposal instructions.

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RELATED PRODUCTS	CAT. NO.
KPL Detector™ PCR DNA Biotinylation Kit	5910-0031 (60-01-01)
KPL Detector HRP Chemiluminescent Blotting Kit	5910-0027 (54-30-00)
KPL Herring Sperm DNA, sheared & denatured	5920-0003 (60-00-14)
KPL Biodyne®B Membrane	5960-0025 (60-00-50)
KPL Hybridization Bags	5960-0026 (60-00-51)
KPL 5X Detector Block	5920-0004 (71-83-00)
KPL 5X Phosphatase Wash Solution	5960-0018 (50-63-15)
KPL 10X Phosphatase Assay Buffer	5960-0017 (50-63-14)

The product listed herein is for research use only and is not intended for use in human or clinical diagnosis.