Formamide Hybridization Buffer

Catalog No.	Size
50-86-10	2 x 120 mL

DESCRIPTION
Formamide Hybridization Buffer is a formamide-based solution for use in the prehybridization and hybridization of biotinylated nucleic acid probes to nucleic acids fixed on membranes. As a prehybridization 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
Formamide Hybridization Buffer consists of 2 x 120 ml of Formamide Hybridization Buffer (Cat. No. 50-86-09). Store at 2-8°C. Stable for a minimum of one year from date of receipt when stored at 2-8°C. 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
Formamide Hybridization Buffer is a formamide-based solution prepared with DNase/RNase free reagents.

APPLICATIONS
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 Herring Sperm DNA (See RELATED PRODUCTS) must be added to a final concentration of 100 μg per ml.

Prehybridization:
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 prehybridization 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 Random Primer Biotinylation Kit (See RELATED PRODUCTS).
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 MSDS for disposal instructions.

RELATED PRODUCTS

Herring Sperm DNA, sheared & denatured	Cat. No. 60-00-14
Hybridization Bags	Cat. No. 60-00-51
Biodyne B Membrane	Cat. No. 60-00-50
20X SSC	Cat. No. 50-86-05
RNADetector™ Northern Hybridization and Detection Kit	Cat. No. 54-30-01
DNADetector™ Southern Hybridization and Detection Kit	Cat. No. 54-30-00
Detector™ Random Primer DNA Biotinylation Kit	Cat. No. 60-01-00

See KPL's catalog for a complete list of nucleic acid labeling and detection systems, antibodies, substrates, and immunohistochemistry products.

The product listed herein is for research use only and is not intended for use in human or clinical diagnosis. Nothing disclosed herein is to be construed as a recommendation to use this product in violation of any patients. However, said information and product are offered without warranty or guarantee since the ultimate conditions of use and the variability of the materials treated are beyond our control. We cannot be responsible for patent infringements or other violations that may occur with the use of this product. No claims beyond replacement of unacceptable material or refund of purchase price shall be allowed.

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