

## ONCORE Pro FISH Kit

Pretreatment and Post-hybridization Reagents for FISH Procedures on the ONCORE Pro Automated Slide Stainer  
902-OPRR6064K-020822

**Catalog Number:** OPRR6064K T60

**Description:** 60 tests

### Intended Use:

For Research Use Only. Not for use in diagnostic procedures.

### Summary & Explanation:

ONCORE Pro FISH Kit contains reagents used in the pretreatment of formalin-fixed paraffin-embedded tissues (FFPE) to enhance probe accessibility to nucleic acid targets as well as a post-hybridization buffer used to eliminate non-specific binding of the probe.

In FFPE tissues, FISH protocols usually require a combination of acid, heat, and enzymatic pretreatment to achieve proper staining. Hydrochloric acid (HCl, 0.2N) is a weak acid commonly used in FISH protocols. FISH Retrieval is a citrate-based heat pretreatment solution, pH 6.0. It is a non-toxic, non-flammable, odorless, sodium azide and thimerosal-free solution. FISHzyme is a commonly used pepsin digestive enzyme. Combination of all 3 pretreatment reagents provides a synergistic effect on probe accessibility to nucleic acid targets.

Reagent Alcohol, 70% is necessary for tissue dehydration before a probe application.

PF SSC Wash Buffer is a ready-to-use saline sodium citrate buffer (pH 7.0-7.5) that contains 0.3% NP40 and Proclin 950 as a preservative. PF SSC Wash Buffer contains 30 mM citrate, which is commonly identified as 2X SSC. This buffer is used as a post-hybridization wash buffer in FISH procedures.

### Known Applications:

Fluorescent *in situ* hybridization (FFPE tissues)

### Reagents Provided:

ONCORE Pro FISH Kit is comprised of 3 solutions in pre-filled vials, plus empty vials for users to fill with hydrochloric acid (HCl, 0.2N), Reagent Alcohol 70% and DAPI.

One kit is sufficient to perform 60 tests:

FISH Retrieval (OPRR6065 T30 x 2) 14.5 mL

FISHzyme\* (OPRR6066 T60 x 1) 14.5 mL

PF SSC Wash Buffer (OPRR6075 T6 x 10) 10.5 mL

HCl, 0.2N (OPRR6063 T20 x 3)

Reagent Alcohol, 70% (OPRR6067 T20 x 3)

DAPI (OPRR6074 T60 x 1)

\*FISHzyme should be stored at 2°C to 8°C.

Please note that reagent vials are labeled with T counts, which refers to how many slides a full vial can stain. For example, if a vial is labeled with a REF number ending in "T20", then this reagent, when full, will stain 20 slides. The "T60" kit can stain 60 slides and will contain 3 of the "T20" reagent vials to meet staining requirements. When scanning reagents, the ONCORE Pro software will display separate values for the "Number of Tests Required" and "Number of Tests Scanned". The "Number of Tests Required" refers to the numbers of dispenses aka "tests" needed for the run. The "Number of Tests Scanned" refers to the number of dispenses aka "tests" that are remaining in the scanned vial(s), which is retrieved from the RFID tag. Please note that more than one dispense aka "test" may be required to complete each slide. After scanning the reagent vials, the "Number of Tests Scanned" must be

greater than or equal to the "Number of Tests Required" to ensure sufficient reagent to complete the run.

### Reconstitution, Dilution and Mixing:

HCl, 0.2N and Reagent Alcohol, 70% vials should be filled to the neck of the vials with the corresponding solutions before placing on the instrument for use. No other preparation is required.

DAPI working solution should also be filled to the neck of the vial before placing on the instrument.

Prior to mixing with TBS, the DAPI concentrate should be kept at 2°C to 8°C and be given 30 minutes to reach room temperature.

### Materials and Reagents Required but Not Provided:

Reagents and materials, such as FISH probes and ancillary reagents are not provided. See the table provided for a list of reagents required but not provided. Call Technical Support for additional information on reagents and instrument accessories.

### Reagents Required but Not Provided:

	Reagent Name
1.	ONCORE Pro ISH Dewax Kit (Biocare SKU: OPRI6020KT60)
2.	ONCORE Pro FISH Probe (Biocare SKU varies)
3.	ONCORE Pro Wash Buffer (Biocare SKU: OPRI6012MM)
4.	HCl, 0.2N
5.	Reagent Alcohol, 70%
6.	DAPI (120ng/mL)
7.	DI water
8.	Fluoro Care Anti-Fade Mountant (Biocare SKU: FP001)

### Storage and Stability:

Store at room temperature, except FISHzyme. Store FISHzyme at 2°C to 8°C. The product is stable to the expiration date printed on the label, when stored under these conditions. Do not use after expiration date.

### Instructions for Use:

FISH Retrieval, FISHzyme, and PF SSC Wash Buffer are provided in vials ready for use on the ONCORE Pro Automated Slide Stainer. Uncap the vials and place in the ONCORE Pro reagent tray. Fill the labeled empty vials with HCl 0.2N, DAPI and Reagent Alcohol, 70% (as appropriate), and place in the ONCORE Pro reagent tray. The ONCORE Pro Automated Slide Stainer will apply reagent as required in the selected protocol. Refer to the ONCORE Pro Automated Slide Staining System User Manual for detailed instructions on instrument operation and additional protocol options.

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### Health Hazard Irritant

### Technical Support:

Contact Biocare's Technical Support at 1-800-542-2002 for questions regarding this product.

### Review the table provided for recommended pepsin digestion times for FFPE tissues on the ONCORE Pro.

Recommended Pepsin Digestion Times	
Tissue Type	Digestion Time (minutes)
Breast	30 - 35
Prostate	30 - 35
Melanoma	30 - 40
Renal Cell Carcinoma	30 - 40
GIST	35 - 40
Tonsil	35 - 40
Bladder	35 - 45
Lung	35 - 45
Stomach	35 - 45
Cervix	40 - 45
Colon	40 - 45
Lymphoma	40 - 45

### References:

1. Analysis of genes and chromosomes by nonisotopic in situ hybridization. Lichter P, et al. Genet Anal Tech Appl. 1991 Feb;8(1):24-35.
2. Fluorescence in situ Hybridization (FISH). Bayani J, Squire JA. Curr Protoc Cell Biol. 2004.
3. Clinical and Laboratory Standards Institute (CLSI). Protection of Laboratory Workers from Occupationally Acquired Infections; Approved Guideline-Fourth Edition (M29-A4) Wayne, PA 2014.

### Limitations:

This product is provided for Research Use Only (RUO) and is not for use in diagnostic procedures. Suitability for specific applications may vary and it is the responsibility of the end user to determine the appropriate application for its use.

### Precautions:

1. Refer to reagent Safety Data Sheet for precautions.
2. Specimens, before and after fixation, and all materials exposed to them should be handled as if capable of transmitting infection and disposed of with proper precautions. Never pipette reagents by mouth and avoid contacting the skin and mucous membranes with reagents and specimens. If reagents or specimens come in contact with sensitive areas, wash with copious amounts of water. (3)
3. Microbial contamination of reagents may result in an increase in nonspecific staining.
4. Incubation times or temperatures other than those specified may give erroneous results. The user must validate any such change.
5. Do not use reagent after the expiration date printed on the vial.

