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ProductInformation

Formalin solution

Product Codes **HT50-1-1, HT50-1-2, HT50-1-4, HT50-1-128, HT50-1-320, HT50-1-640**
Store at Room Temperature

Synonyms: 10% Neutral Buffered Formalin; 10% NBF

Product Description

Soon after an organism dies, or a tissue is removed from the body, putrefaction and autolysis begin. Autolysis is retarded by cold, greatly accelerated at temperature of about 30 °C, and almost inhibited by heating to 50 °C. There are very few instances when specimens are examined without fixation or some form of hardening. The aim of fixation is to stop decay, putrefaction and autolysis. Proper tissue fixation is essential for accurate histopathologic evaluation.

If properly preserved, the cell and tissue constituents should appear in as lifelike a manner as possible. In reality each fixative creates a unique set of artifacts. The living cell is fluid, or in a semi-fluid state. Formaldehyde is both a noncoagulant and an additive fixative. Formalin fixation is thought to form cross links between the aldehydes and the proteins, creating a gel, thus retaining cellular constituents in their *in vivo* relationship. Once properly fixed, the tissue should be able to withstand the subsequent stages of tissue processing or staining.

Formaldehyde is known to penetrate tissue quickly, but the fixation process may take a long time to form the various cross links with tissue proteins. Although there is no true universal fixative, 10% Neutral Buffered Formalin is perhaps the most commonly used fixative throughout the world for light microscopy and is a somewhat forgiving fixative. Of the dozens and dozens of available fixatives, it is hard to imagine a fixative more commonly used than 10% NBF. 10% NBF is well suited for large throughput laboratories, and requires a relatively short period of fixation, but can also be used for the long-term storage of tissue.

Reagents

Formaldehyde, 37-40%	100 ml/L
Distilled or deionized water	900 ml/L
Sodium phosphate, monobasic	4.0 g/L
Sodium phosphate, dibasic (anhydrous)	6.5 g/L

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

If shipping tissue fixed in 10% Neutral Buffered Formalin, comply with all state, local or federal laws.

If cells and cellular structures are not properly preserved and stabilized by fixation, any testing performed on the tissue will be of mediocre or possibly unusable quality. Misdiagnosis may result if the tissue is improperly handled or fixed. The end user must determine the proper fixative and the conditions necessary for proper fixation. If the ratio of fixative to tissue is not adequate, tissue may deteriorate or putrefy during long-term storage.

There are tissues and constituents which are not properly preserved by 10% Neutral Buffered Formalin. If there is any doubt as to the proper fixative, consult a standard histology procedural manual[1-6] or contact Technical Service for assistance in choosing an appropriate fixative.

Storage/Stability

Store tightly closed at room temperature. Do not freeze. The expiration dating is printed on the product label.

Preparation Instructions

Ready to use. Dilution may be required for certain specialized applications.

Procedure

1. Fixation should begin as soon as possible.
2. Be sure the tissue is placed in the proper fixative. If the tissue cannot be immediately placed into the fixative, keep the tissue moist and cool. Typically the tissue is kept moist with normal saline or isotonic PBS.

3. The ideal ratio of fixative to tissue should be in the range of 20 to 50 parts of fixative to 1 part tissue. The ratio of fixative to tissue should never be less than 10 to 20 parts of fixative to 1 part tissue. Tissue intended for museum preparation should be placed in a ratio of 100 parts fixative to 1 part tissue and the ratio should never be less than 50 parts fixative to 1 part tissue
4. Whole organs should be injected with fixative as well as immersed in fixative. Large organs can be sliced to allow better penetration of the fixative into the tissue.
5. Hollow organs can be injected with fixative or can be packed with absorbent cotton soaked in fixative before immersion. Some organs such as colon may be opened and pinned to a corkboard before immersion into the fixative.
6. The time needed for fixation can range from just a few hours to several weeks. The time needed will vary upon the tissue type and the size or thickness of the specimen.
7. After fixation has been completed, the fixed tissue should be trimmed to no more than 3 to 5 mm in thickness and placed on a tissue processor for paraffin processing. The first alcohol the tissue contacts should be 70%. Placing formalin fixed tissues into high percentage alcohols can result in the precipitation of the phosphate buffered salts used to prepare the 10% NBF.

References

1. Histotechnology: A Self-Instructional Text, 2nd Edition, Freida L Carson, ASCP Press, Chicago 1997.
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3. An Introduction to Histotechnology, Geoffrey G. Brown, Appleton-Century-Crofts, New York, 1978.
4. Humason's Animal Tissue Techniques, 5th Edition, Edited by Janice K. Presnell and Martin R. Schreibman, The John Hopkins University Press, Baltimore 1997.
5. Theory and Practice of Histological Techniques, Edited by John D Bancroft and Marilyn Gamble, Churchill Livingstone, London, 2002.
6. Cellular Pathology Technique, 4th Edition, Edited by C.F.A. Culling, R.T. Allison and W.T. Barr, Butterworths, London, 1985.

RDK/PHC 04-05-1