

Neutral Buffered Formalin

For In-Vitro Diagnostics For Professional Use only

Neutral Buffered Formalin is one of the most widely used fixatives to inhibit putrefaction, autolysis and other forms of tissue degradation. The concentrations range from 10-20% (4-8% formaldehyde) and are combined with phosphate buffering salts to give a pH of around 7.

Sample Preparation

Tissue specimens should be placed in 10% Neutral Buffered Formalin immediately after removal from the body.

Specimen size to be fixed in 10% Neutral Buffered Formalin may vary from a whole organ to gross sections due to its rapid penetration rate.

This product is designed to be used as a primary fixative at its full strength and does not require any further dilution or additive. The solution must be changed daily.

The concentration and the temperature at which the NBF is used influence fixation. Lille states that 10% formalin fixes adequately in 48 hours at room temperature (20°C to 25°C); 10% formalin fixes in 24 hours at 35°C; and 20% formalin hardens in 3 hours at 55°C. As autolysis is hastened by higher temperatures, it is better to fix tissues for a longer time at room temperature.¹

Mode of Action

Neutral Buffered Formalin contains formaldehyde which reacts with tissue groups, predominantly with groups found in amino acids containing reactive hydrogen.

Formalin fixation has three modes of action: penetration, cross-linking and binding that occur simultaneously but at different rates; binding being the slowest.

The major site of reaction occur at the amino group in amino acid side chains where methylene bridges form and link protein chains together. This creates a very important positive group helpful in attracting eosin dye during H&E staining.

10% Neutral Buffered Formalin may not be suitable for prolonged storage of lipids and carbohydrate fixation. NBF is a non-coagulant fixative. DNA and RNA are reduced by 10-35% by formalin fixation

¹ Sheehan, pg 44

and tissue for the demonstration of nucleic acids should be fixed in Carnoy's fluid.

Warnings and Precautions

Use 10% Neutral Buffered Formalin in a well ventilated area, and wearing protective gloves and clothing when handling the product.

Limited evidence of carcinogenic effect.

Please consult the SDS and packaging labels before use.

Ingredients

| Chemical | CAS | Conc |
|------------------------------|-----------|------------|
| Formaldehyde | 50-00-0 | 3.7 – 8.0% |
| Sodium di-hydrogen Phosphate | 7558-80-7 | <1% |
| Di-sodium hydrogen phosphate | 7558-79-4 | <1% |
| Water | 7732-18-5 | Balance |

Specifications

| | |
|----------------|--------------------------|
| Appearance | Clear, colourless liquid |
| Assay | 3.7-4.2% |
| pH @ 25°C | 6.9-7.1 |
| Solids | 1.0-1.5% |
| Phosphate | 0.7-0.8% |
| Density @ 20°C | 1.000 – 1.040g/mL |

Stability

Neutral Buffered Formalin is stable for 3 years when stored in a sealed container at room temperature away from heat and light.

Technical Procedure

1. Prepare a container 15-20 times the volume of the tissue specimen to be fixed, and fill the container with an adequate amount of Neutral Buffered Formalin.
2. Immerse the specimen into the container making sure of a complete immersion and let it soak for an appropriate time depending on the specimen size. Generally 1 hour immersion for each of the first two stations of a conventional tissue processor is employed.

For a complete fixation, immersion time may take sometime between 1 or 2 weeks.

Increased temperature may allow a more rapid penetration process, but it is highly advised to consult with literature the effect of temperature on the tissue specimen.

3. When a post-treatment with a dehydrant is required, ensure that the solution contains less than 70% ethanol as to prevent phosphate precipitates from forming.



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References

1. Sheehan, D.C. and Hrapchak, B.B.: *Theory and Practice of Histotechnology*, 2nd Edition; 1987, Battelle Press

Ordering Information

| Code | Product Name | Size |
|--------------|--|-------|
| FNNJJ010 | Formalin Neutral Buffered 10% Green Dye | 500mL |
| FNNJJ018 | Formalin Neutral Buffered 10% Solution | 5L |
| FNNJJ01810 | Formalin Neutral Buffered 10% | 10L |
| FNNJJ01810B | Formalin Neutral Buffered 10% Blue Dye | 10L |
| FNNJJ0185 | Formalin Neutral Buffered 10% | 2.5L |
| FNNJJ0185B | Formalin Neutral Buffered 10% Blue Dye | 2.5L |
| FNNJJ0186 | Formalin Neutral Buffered 10% | 1L |
| FNNJJ018B | Formalin Neutral Buffered 10% Blue Dye | 5L |
| FNNJJ018G | Neutral Buffered Formalin 10% Green Dye | 5L |
| FNNJJ019 | Formalin Neutral Buffered 10% | 20L |
| FNNNBFB-20L | Neutral Buffered Formalin 10% Blue | 20L |
| FNNJJ016 | Formalin Buffered Concentrate | 5L |
| FNNJJ0160 | Formalin Buffered Concentrate | 10L |
| FNNJJ0160A | Formalin Buffered Concentrate Blue Indicator | 5L |
| FNNJJ016A2.5 | Formalin Buffered Concentrate x8 Blue Dye | 2.5L |
| FNNJJ017 | Formalin Buffered Concentrate | 20L |
| FNNJJ017NF | Formalin Buffered Concentrate Modified Low | 20L |