

## Fixation Methods

**Immediate** fixation of cytology specimens is critical to the preservation of the cellular components. It is important that no air-drying occurs prior to fixation. If a smear is already air-dried it should not be put in alcohol fixative. Please note on the requisition if the slide(s) being submitted are fixed or air-dried. Formalin fixation is not appropriate for cytology specimens. Specimens should not be exposed to formalin or formalin fumes. This alters the cells and interferes with the staining reactions. There are several fixation techniques available, depending on the type and volume of the specimen. See specimen collection techniques and fixation procedures for specific details.

1. Spray Fixative – suitable for specimens that are submitted on a slide(s). This would include specimens such as Pap smears, FNA specimens, and endoscopic brushing specimens.
2. 95% alcohol (usually used within a Coplin jar) - suitable for specimens that are submitted on a slide(s). This would include specimens such as Pap smears, FNA specimens, and endoscopic brushing specimens. The slides should be immersed in the alcohol for a minimum of 15 minutes. Alternatively, the fixative may be pipetted onto a slide until the smear is totally saturated and then allowed to dry.
3. Saccomanno Collection Fluid – a green colored fixative for the collection of sputum.
4. CytoLyt Solution (Sioux Falls clients) - a clear fixative for the collection of fluid specimens. A 50/50 ratio of specimen to fixative is appropriate (if this unavailable use 50% alcohol).
5. 50% Alcohol – (Mankato clients) a clear fixative for the collection of fluid specimens. A 50/50 ratio of specimen to fixative is appropriate.

**Quick Reference Guide to Fixation Techniques**

Specimen Type	Recommended Fixation Technique	Comments
<b>Large Volume Specimens:</b> Abdominal and Pelvic washings Body Cavity Fluids (pleural, peritoneal) Urines Gastric/Esophageal washings	Mix with equal amounts of:  <u>CytoLyt Solution for Sioux Falls clients</u> <u>or</u> <u>50% Alcohol for Mankato clients</u> Fix no more than 50 ml of specimen; submit the remainder of body cavity fluid unfixed.	Refrigerate the unfixed portion of the specimen if possible.
<b>Small Volume Specimens:</b> FNA (fluid – not slides) Breast fluid CSF Cyst fluid Synovial fluid Bronchial washing	Mix with equal amounts of:  <u>CytoLyt Solution for Sioux Falls clients</u> <u>or</u> <u>50% Alcohol for Mankato clients</u> Use 10 ml of fixative if specimen volume is under 10 ml.	With very small amounts of fluid it may be easier to transfer the fixative into the collection device (syringe, suction collection tubes) first. Then into a suitable container to submit the specimen.
<b>Direct Smears:</b> Pap Smears FNA specimens Brushings Nipple Secretions	<u>95% alcohol:</u> The slides should be immersed in the alcohol for a minimum of 15 minutes. Alternatively, the fixative may be pipetted onto a slide until the smear is totally saturated and then allowed to dry. <u>or</u> <u>Spray fix:</u> Hold the bottle of spray fix 3-4 inches from the slide and disperse an even layer of fixative over the slide. Preferred method for Paps.	Pap smears should be completely dry before placing them into cardboard containers. The endoscopic brush may be submitted in 50% alcohol after the slides have been prepared.