**Basofix™ Hematology Reagent System**

**Intended Use**
Basofix is a system of reagents for staining peripheral blood smears with the Wescor Aerospray Hematology Slide Stainer. Its purpose is to fix granules of basophilic leukocytes in the slide.

**INSTRUCTIONS**
Basofix reagents are available ready-to-use (SS-049 series) or as concentrates (SS-135/149 series). Diluted concentrates are interchangeable with ready-to-use reagents. For best results,* replace the normal Red Stain (SS-035C or SS-135C) and Rinse (SS-035A or SS-135A) with Basofix Red Stain (SS-049C or SS-149C) and Basofix Rinse (SS-049A or SS-149A). Refer to your stainer User's Manual for reagent loading instructions.

Use regular Blue Stain (SS-035B or SS-135/149B) and anhydrous, reagent-grade methanol (SS-MeOH or equivalent). The methanol is not used for fixation, but is required for the CLEAN cycle.

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You **must** deactivate the automatic fixation function to prevent the stainer from spraying methanol onto slides you have dipped in Basofix Fixative.* To deactivate fixation, follow the appropriate step for your stainer model:

**For Model 7100A Stainer**
Replace reagent nozzle D with a nozzle plug (Wescor AC-042).

**For Model 7120 Stainer**
Press \[ PROG \]. The display will show:

\[
\text{# OF SLIDES 1-2, 3-4, 5-6, 7+?}
\]

**ALCOHOL FIXATION**

\[
9=ON, 0=OFF
\]

Press \( 0 \) to deactivate alcohol fixation. The display will show:

**MEDIUM 4 FIX OFF**

**7-12 SLIDES**

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* See **DISCUSSION** on page 2
WARNING! These reagents contain moderately toxic chemicals that require care in handling. Always use appropriate safety measures including gloves and eye protection when handling reagents.

Dispense Basofix Pre-dip Fixative into a coplin jar or other convenient vessel.

Basofix is designed to function as a quick-dip fixative. Dip the air dried slide, feathered edge down, into the fixative. Lift the slide and allow it to drain for one to five seconds. Touch any hanging drops off on the jar.

Load the slide into the Aerospray carousel. Repeat steps 3 and 4 for up to 12 slides.

Make certain that slides dipped in Basofix are dry before staining (see DISCUSSION, below).

For Model 7100A Stainer: Run the staining cycle with alcohol fixation deactivated (as described in step 1).

For Model 7120 Stainer: Allow the carousel to stand on the bench for 30 to 60 seconds, or until the methanol component has evaporated from the last slide loaded. Then run the staining cycle with alcohol fixation deactivated (as described in step 1).

DISCUSSION

The effective mechanism of the Basofix system is the deposition of a stain layer after evaporation of the methanol fixative. The quality of granules does not appear to be sensitive to time spent in the pre-dip, but can sometimes be improved by additional dips with a short wait between each. One dip is usually adequate.

Important: If you do not deactivate alcohol fixation, Basofix will be washed from the slide when the stainer sprays methanol for fixation. This will prevent the desired fixation. Deactivating automatic fixation as described in step 1 allows the CLEAN cycle to remain operational (you must keep the methanol connected to the Reagent D line). Model 7120 Aerospray stainers default to FIX ON at power-up. If you are using Basofix with a 7120, be sure to reset FIX OFF after stainer power is cycled.

Slides dipped in Basofix Pre-dip should be dry before staining. Proper drying will occur within 30 to 60 seconds after removing the slide from the pre-dip. With the model 7100A stainer, there is a 90-second delay (while the stainer is attempting automatic fixation) before staining begins. This delay provides ample time for slides to dry, so a carousel of slides dipped in Basofix can be immediately loaded into the 7100A. With the 7120, the fixation delay is eliminated when FIX OFF is set, so staining begins immediately after the cycle is started. For best results, allow a carousel of slides dipped in Basofix to stand on the bench for 30 to 60 seconds before loading the carousel into the 7120.

If you use standard Rinse and Red reagents (SS-035A or SS-135A and SS-035C or SS-135C) with Basofix Fixative, the red cells will be dark red and slides will contain significant debris. To avoid these problems, always use Basofix Rinse (SS-049 or SS-149A) and Red (SS-049C or SS-149C) reagents with Basofix Fixative.

For best results, use freshly prepared slides that have not been prefixed in methanol.

RESULTS

Without Basofix Pre-dip, basophils appear degranulated when stained with the Wescor Aerospray stainer. With Basofix used as directed, most basophils will appear with good to excellent granulation. Their general appearance is similar to those stained with Wright's or Giemsa stains. The basophil population is quite heterogeneous. With any staining system, a fraction of the basophil population will be poorly preserved. The Basofix system delivers good preservation of more than 90 percent of existing basophils.
With continued use, the Basofix Pre-dip Fixative solution will concentrate and finished slides will contain significant debris. Either replace the fixative or dilute it with anhydrous methanol to replace the evaporative loss.

In humid environments the pre-dip fixative may produce refractile artifacts. These artifacts will increase as the Basofix Pre-dip is used and absorbs water from the atmosphere. Under these conditions, mix 1 part Aerofix™ Hematology Fixative (SS-048) with 3 parts Basofix (SS-049P) for the initial reagent and discard the reagent when the artifacts become troublesome. Alternatively, add 5 mL Aerofix Additive (SS-148) to 500 mL Basofix Pre-dip Fixative (SS-049P) to prepare the initial reagent for humid environments.