

TD-P Revision 3.0

Protocol

Creation Date: 5/25/2016 Revision Date: 9/7/2018

Preparation of Mucoid Samples Utilizing ThinPrep Non-Gyn (From Hologic[®]) and DTT

Introduction

The proper collection and preparation of mucoid specimens are necessary to perform an accurate analysis and obtain reliable results. Often, the mucus in these samples can have deleterious effects because it causes cells to clump up upon collection, affecting the test's accuracy. Dithiothreitol (DTT) is a mucolytic agent and can reduce the amount of mucus in samples from various tissues, including respiratory samples. DTT can be used in conjunction with ThinPrep Non-Gyn, which has been used in laboratories around the world for more than 15 years. ThinPrep Non-Gyn offers a standardized specimen preparation method for a variety of cytologic specimens. The slides are easier and less time-consuming to screen and interpret, because the cells are limited to a defined area (20 mm diameter) on a clear background with excellent cellular presentation. ThinPrep Non-Gyn offers one single, well-preserved slide per case and decreased screening time per slide. Here, we describe a general protocol for the processing of mucoid specimens with ThinPrep Non-Gyn and DTT.

Materials

- ThinPrep Non-Gyn kit
- Dithiothreitol (GoldBio Catalog # DTT)
- ThinPrep Processor

Storage and Handling

- Store desiccated at -20°C.
- Prepare solutions fresh, daily.
- DTT may be shipped on blue ice and should be stored immediately upon arrival at -20°C.

Method

Procedure for processing mucoid specimens including respiratory and gastrointestinal samples

1. Collect sample directly into CytoLyt solution OR add 30 ml of CytoLyt solution to the fresh specimen immediately after collection.



Note: Large specimens (greater than 20 ml) should be concentrated before the addition of CytoLyt solution to the sample.

Gold Biotechnology/ FM-000008 Preparation of Mucoid Specimens utilizing DTT

Note: If DTT is being used with mucoid samples, add stock before agitation. To use DTT with the ThinPrep system, prepare a stock solution by adding 2.5 g DTT to 30 ml of CytoLyt solution. This solution is suitable for use up to 1 week when stored at room temperature (15-30°C). Add 1 ml of this stock solution to the sample.

TD-P Revision 3.0

TD-P Date: 9/7/2018

- 2. Vortex for a minimum of 5 minutes in a "hands-free" vortex.
- 3. Concentrate the sample by centrifugation at 600 g for 10 minutes. A pellet should form and be visible at the end of centrifugation.
- 4. Pour off the supernatant and resuspend the cell pellet. Resuspension can be done on a vortex or may be achieved by pipetting the pellet back and forth with a plastic pipette.
- 5. Confirm the cell pellet has been resuspended and is in liquid form. If the cell pellet is not in liquid form, add 30 ml of CytoLyt solution and repeat steps 2 and 4.

6. Add the specimen to PreservCyt Solution Vial.

05









Gold Biotechnology/ FM-000008 Preparation of Mucoid Specimens utilizing DTT

7. Allow to stand in PreservCyt Solution for 15 minutes.

8. Fix, stain, and evaluate by running on the ThinPrep Processor using Sequence 3 (Mucoid). Refer to the ThinPrep processor operator's manual for detailed instrument and specimen preparation instructions.

Associated Products

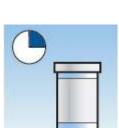
Dithiothreitol (DTT) (GoldBio Catalog # DTT)

References

Tang C.S., et al. (1994). Dithiothreitol homogenization of prefixed sputum for lung cancer detection. *Diagnostic Cytopathology*. 1994 10(1):76-81.

Tockman M.S., et al. (1995). Safe separation of sputum cells from mucoid glycoprotein. Acta Cytologica. 1995 39(6):1128-36.







TD-P Revision 3.0

TD-P Date: 9/7/2018