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# TECHNICALLY *Speaking*

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## 10X Tris-EDTA Buffer For Heat Induced Epitope Recovery, pH 9.0

CLSG90448

Lot:

**Intended Use:**

To recover the masked antigens because of the over fixation in the cross linking fixatives like formalin.

**Introduction:**

In order to perform immunostaining, the tissue specimens should be fixed in appropriate fixative. The purpose of such fixative is to conserve the tissue from autolysis, to preserve tissue structures and to immobilize antigens. However, this requires harsh treatment of the antigens. As a result, antigens undergo chemical alteration of their primary, secondary and tertiary structures. Because of changes in the protein containing epitopes or in neighboring proteins, antigenic sites may be masked. In past, enzymatic treatment with proteolytic enzymes i.e. pepsin, trypsin or pronase has been performed to regain the masked antigens. Shi et al. (1991) have reported that the treatment of the tissue section with heavy metal solution in a microwave can regain the masked antigens significantly. However, heavy metals in the solution increase the risk of exposure of lab personals to lead. To solve this problem, we have developed a new antigen unmasking solution in tablet form, which is free from heavy metals. Use of this antigen unmasker can prevent the risk of unnecessary exposure to the lab personal and also resolve the handling and disposal problems.

**Format:**

500 ml. 10X Tris-EDTA Retrieval buffer, pH 9.0.

**Storage:**

Store at room temperature.

**Preparation of Reagent:**

Dilution one part of buffer with nine part of distilled water.

**Procedure:**

1. Deparaffinize and bring tissue section to buffer.
2. Fill the plastic coplin jar with the antigen unmasker solution.
3. Place the jar in the in steamer or water bath.
4. Preheat steamer or water bath containing coplin jars to 95-100°C.
5. Place the deparaffinized slides (1 to 3 slides/jar) in the coplin jar and incubate for 20-40 minutes (optimal incubation time should be determined by the end user).
6. Remove the coplin jars from the water bath and allow the slides to cool down for 20 minutes to reach to room temperature.
7. Wash the slides in de-ionized water and then with wash buffer and proceed for immunostaining.

*Continued...*

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**Reference:**

Shi et al. J Histochem Cytochem 39: 741, 1991.

**Laboratory Reagent For Research**

JV 05/20/08