

Preparation of deionized glyoxal

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Abstract

A brief protocol for the preparation of glyoxal used in glyoxal/borate and RNase T1-based detection of inosine residues in RNAs.

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Materials

Glyoxal (40%) 50649 by <u>Sigma Aldrich</u> Mixed Bed Resin M8032 by <u>Sigma Aldrich</u>

Protocol

Step 1.

Mix 25 ml of 40% glyoxal to be deionized with 2.5 g of mixed bed resin.

Step 2.

Mix the solution by magnetic stirrer for 30 minutes at room temperature.

• The resin color will change from blue to amber, when its capacity is reached.

Step 3.

Separate the deionized material from the resin by decanting or (sterile) filtration.

Step 4.

Repeat the deionization process (steps 2. and 3.) until the color of the resin does not change anymore.

• Approximately half of the resin particles remain blue and half become amber after a 30-minute incubation.

Step 5.

Check the pH of the glyoxal solution by mixing 200 μ l of glyoxal with 2 μ l of a 10 mg/ml aqueous solution of bromocresol green and observing the color change. Bromocresol green is yellow at pH <4.8 and blue-green at pH >5.2. Alternatively, use pH indicator strips instead of a bromocresol green solution.

Step 6.

Repeat the deionization process (steps 2. and 3.) until the pH of the glyoxal is >5.5.

Step 7.

Final resin removal should be done through a small pore-size filter (e.g. 0.22- $0.45~\mu m$, gamma sterilized) to avoid carry-over of the resin.

Step 8.

Deionized glyoxal can be stored indefinitely at -20 °C (or -80 °C) under nitrogen in tightly sealed small aliquots (e.g. microcentrifuge tubes).

• Use each aliquot only once, then discard.

Warnings

Glyoxal can cause oral, dermal, and respiratory difficulties. Work in gloves and use the fume hood.