PROTOCOL

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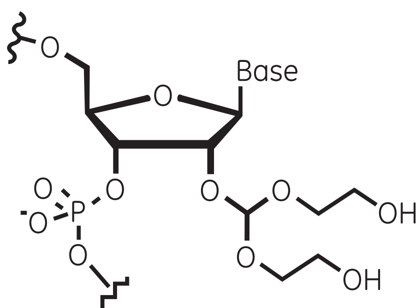
Receiving RNA

This protocol is for deprotection of RNA oligonucleotides.

Introduction

All custom synthesized RNA oligos are delivered 2'-ACE protected unless deprotection services were specified at the time of the order. All other protecting groups have been cleaved from the RNA. Unless otherwise requested, all internucleotidic linkages are phosphates and the 5'- and 3'-ends are both hydroxyl groups. The general structure of 2'-ACE protected RNA is as follows:



The 2'-protecting groups confer resistance to nuclease degradation and help to minimize secondary structure. Using anion exchange HPLC, a 0.5% aliquot of every 2'-protected oligo is analyzed prior to shipping. The remaining 99.5% aliquot of the crude oligo is dried down in vacuum, packaged and shipped in the stable 2'-protected form to best ensure consistent quality. The 2'-protecting groups are then easily removed in 30 minutes at pH 3.8 using the enclosed aqueous buffer. The method for preparing additional deprotection buffer is as follows:

Materials for 100 mL 2' deprotection buffer

Consumables

1. 100 mL sterile bottle or flask.

Chemicals

1. RNase-free water.
2. Acetic Acid (glacial 17.5 M).
3. TEMED (Tetramethylethylenediamine).

Procedure

2'- Deprotection buffer is dilute (100 mM) acetic acid adjusted to a pH of 3.4-3.8 using TEMED.

1. Mix 571 µL glacial acetic acid with 99.4 mL RNase-free water to make 100 mL of 100 mM acetic acid.
2. Adjust the pH of the 100 mM acetic acid to 3.4–3.8 using TEMED.

When the RNA oligos are received, they must be stored at –20 °C until ready for use. The specification sheet which accompanies each individual oligo can be removed from the packaging and placed in a notebook for reference.

If you have any questions, contact

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