

# 10 mM Tris-HCl buffer pH 7.4

This protocol is used to make resuspension buffer for synthetic guide RNAs

Buffered solutions help protect resuspended RNA and DNA against degradation caused by pH changes that occur during freeze-thaw cycles. The following protocol provides instruction for making a buffered 10 mM Tris-HCl solution appropriate for resuspension of nucleic acids, such as Dharmacon™ Edit-R™ synthetic guide RNAs.

## Materials needed:

- Tris base, C<sub>4</sub>H<sub>11</sub>NO<sub>3</sub> (molecular weight: 121.14 g/mol), or 1 M Tris-HCl pH 7.5 can be purchased from Fisher Scientific Cat #BP1757-100
- Concentrated HCl
- Nuclease-free water (Dharmacon Cat # B-003000-WB-100)
- 0.22 µm sterile filter



**All materials should be molecular biology grade and/or nuclease-free.**

## Protocol:

1. For a 1 M solution, dissolve 12.1 g of Tris base in 80 mL of nuclease-free water.
2. Adjust the pH to 7.4 value by slowly adding approximately 6-7 mL concentrated HCl. Adding concentrated HCl to the Tris buffer will increase the temperature of the solution, which affects the pH. Allow the solution to cool to room temperature before making final adjustments to the pH (using more HCl if necessary).



**Always add an acid to an aqueous solution; never add an aqueous solution to an acid.**

3. Adjust the volume of the solution to 100 mL with water.
4. Filter Tris-HCl with 0.22 µm sterile filter.
5. To obtain a 10 mM Tris-HCl pH 7.4 solution, dilute 1 M Tris-HCl pH 7.4 1:100 with nuclease-free water. For example, add 1 mL of 1 M Tris-HCl pH 7.4 to 99 mL of nuclease-free water.



**It is recommended to make small aliquots to avoid contamination.**

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