

## Solution Preparation

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### **2X CTAB Buffer**

100 mM Tris-HCl (pH8.0)  
 1.2 M NaCl  
 20 mM EDTA  
 2% CTAB  
 0.2%  $\beta$ - mercaptoethanol

### **(Per 800 ml)**

80 ml 1 M Tris-HCl pH 8.0  
 192 ml 5 M NaCl  
 32 ml 0.5 M EDTA  
 16.0 g powder  
 add right before use (1/500 vol)

### **EDTA, 0.5 M** (500 ml stock, pH 8.0)

93.0 g EDTA (disodium ethylenediaminetetra-acetate·2H<sub>2</sub>O) in 400 ml of H<sub>2</sub>O  
 Adjust pH to 8.0 (~10 g NaOH pellet), it dissolves when pH is right, add H<sub>2</sub>O to 500 ml

### **Ethidium bromide, 10 mg/ml**

Dissolve 0.2 g ethidium bromide in 20 ml H<sub>2</sub>O. Mix well and store at 4°C in dark  
*CAUTION: Ethidium bromide is a mutagen and must be handled carefully.*

### **KCl, 1 M**

74.6 g KCl  
 H<sub>2</sub>O to 1 liter

### **MgCl<sub>2</sub>, 1 M**

20.3 g MgCl<sub>2</sub>·6H<sub>2</sub>O  
 H<sub>2</sub>O to 100 ml

### **MgSO<sub>4</sub>, 1 M**

24.6 g MgSO<sub>4</sub>·7H<sub>2</sub>O  
 H<sub>2</sub>O to 100 ml

### **NaCl, 5 M**

146.1 g NaCl  
 Add H<sub>2</sub>O to 500ml

### **TBE (Tris/borate/EDTA) electrophoresis buffer 1 liter:**

108 g Tris base (890 mM)  
 55 g boric acid (890 mM)  
 40 ml 0.5 M EDTA, pH 8.0 (see recipe; 20 mM)

### **1x TE buffer**

10 mM Tris-HCl (pH 8.0)  
 1 mM EDTA (pH 8.0)

### **Per 500 ml**

5 ml 1 M Tris-HCl  
 1 ml 0.5 M EDTA  
 add H<sub>2</sub>O to 500 ml

**Tris-HCl [tris(hydroxymethyl)aminomethane], 1 M**

Dissolve 121 g Tris base in 800 ml H<sub>2</sub>O  
Adjust to desired pH with concentrated HCl  
Mix and add H<sub>2</sub>O to 1 liter

*Approximately 70 ml of HCl is needed to achieve a pH 7.4 solution, and approximately 42 ml for a solution that is pH 8.0.*

*NOTE: The pH of Tris buffers changes significantly with temperature, decreasing approximately 0.028 pH units per 1°C. Tris-buffered solutions should be adjusted to the desired pH at the temperature at which they will be used. Because the pK<sub>a</sub> of Tris is 8.08, Tris should not be used as a buffer below pH ~7.2 or above pH ~9.0.*

**dNTP dilution from Boehringer Mannheim**

To make an 800 µl stock dNTP with final concentration 1.25mM of ea. dNTP:  
The concentration for each original dNTP is 10 µmole/100 µl = 0.1M  
Take 10 µl 0.1M dNTP each into 760 µl dH<sub>2</sub>O

**TLE (for RNA extraction)**

0.2M Tris  
0.1M LiCl  
5mM EDTA pH8.2  
Preparation in 1L:  
10ml 0.5M EDTA  
4.25g LiCl  
DEPC treated and autoclave  
--> add 22.88g Tris HCl  
add 6.64g Tris base  
--> bring up to 1000ml with DEPC H<sub>2</sub>O