

## Recipes for making recording stock solutions

### Two stock solutions: 10x normal Ringer's and 10x bicarbonate stock

To make 10 x modified Ringer's Stock for 0.5 liter:

KCl	0.93	(in grams)
Glucose	9.01	
NaCl	36.82	
NaH <sub>2</sub> PO <sub>4</sub> - H <sub>2</sub> O	0.86	
1 M solution MgCl <sub>2</sub>	10 mL	
0.5 M solution CaCl <sub>2</sub>	20 mL	

To make 10 x Bicarbonate Stock for 0.5 liter:

NaHCO <sub>3</sub>	10.92	(in grams)
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### To make 1x recording ACSF of 0.5 Liters

**Use a 100ml cylinder to measure 50 ml 10x modified Ringer's stock, add it into a 500 ml volumetric flask**

**Then rinse the cylinder and add about 300 ml double distilled water**

(to dilute Mg<sup>++</sup> and Ca<sup>++</sup> concentration before adding bicarbonate)

**Next, add 50 ml 10x Bicarbonate stock solution**

**Finally, add enough water to get an exact volume of 500 ml**

1x modified Ringer's

2.5 mM	KCl
10 mM	Glucose
126 mM	NaCl
1.25 mM	NaH <sub>2</sub> PO <sub>4</sub> - H <sub>2</sub> O
2 mM	MgCl <sub>2</sub> - 6H <sub>2</sub> O
2 mM	CaCl <sub>2</sub> - 2H <sub>2</sub> O
26 mM	NaHCO <sub>3</sub>

**Original recipe from the Soltesz lab at UCI**

## Xu lab protocol and instructions

# Making Recording Stock Solutions

## Stock solutions: 10x normal Ringer's and 10x Bicarbonate

### 10x modified Ringer's Stock for 0.5 Liter:

KCl	0.93 g
Glucose	9.01 g
NaCl	36.82 g
NaH <sub>2</sub> PO <sub>4</sub> - H <sub>2</sub> O	0.86 g
1M solution MgCl <sub>2</sub>	10 mL
0.5M solution CaCl <sub>2</sub>	20 mL

- 1) Add 300~400mL ddH<sub>2</sub>O into a 500mL beaker
- 2) Add clean stir bar into beaker & place beaker on stir plate
- 3) Turn on stirring (300~600 rpm); DO NOT turn on heat
- 4) Place weighing boat/paper on scale and measure directed amount of an ingredient with spatula [For small amount, please use the fine scale; for anything significantly more than 1 gram, please use the flat top scale]
- 5) Add ingredient into beaker and allow time to dissolve
- 6) Use another clean weighing boat/paper and spatula to measure next ingredient, add it into beaker, and so on
- 7) Get the 5 ml mechanical pipette and use 5 mL pipette tips to extract MgCl<sub>2</sub> and CaCl<sub>2</sub> solutions

**\*Add ingredients IN ORDER to prevent solution becoming murky (undesired reactions)**

- 8) Wait until all ingredients are dissolved, then pour solution into a 500mL volumetric flask and fill solution up to the 500mL mark with ddH<sub>2</sub>O (use pipette tip to add water when close to mark)
- 9) Put stopper on flask & shake/mix solution until it's homogenous
- 10) Pour solution into a 500mL bottle and label it with '10x Ringer's', your initials, and date (use colored tape)
- 11) Store bottle in 4 degrees fridge, clean up area

## Xu lab protocol and instructions

### 10x Bicarbonate stock for 0.5 Liter:

NaHCO<sub>3</sub>                      10.92 g

- 1) Add ~400mL ddH<sub>2</sub>O into a 500mL beaker
- 2) Add clean stir bar into beaker & place beaker on stir plate
- 3) Turn on stirring (300~600 rpm); DO NOT turn on heat
- 4) Place weighing boat/paper on scale and measure 10.92 g of NaHCO<sub>3</sub> with spatula
- 5) Add NaHCO<sub>3</sub> into beaker and allow time to dissolve
- 6) Pour solution into a 500mL volumetric flask and fill solution up to the 500mL mark with ddH<sub>2</sub>O (use pipette tip to add water when close to mark)
- 7) Put stopper on flask & shake/mix solution until it's homogenous
- 8) Pour solution into a 500mL bottle and label it with '10x Bicarbonate', your initials, and date (use colored tape)
- 9) Store bottle in 4 degrees fridge, clean up area

### 1x recording ACSF for 0.5 Liter:

- 1) Use a 100mL graduated cylinder to measure 50mL of 10x Ringer's stock
  - 2) Add 50mL Ringer's stock into a 500mL volumetric flask
  - 3) Rinse cylinder by filling it with ddH<sub>2</sub>O and then adding it into the volumetric flask (add ~300mL ddH<sub>2</sub>O this way)
- \*This dilutes Mg<sup>2+</sup> and Ca<sup>2+</sup> concentrations in solution so they won't react**
- 4) Measure 50mL of 10x Bicarbonate stock in graduated cylinder and add it into the flask
  - 5) Fill solution up to the 500mL mark with ddH<sub>2</sub>O (use pipette tip to add water when close to mark)
  - 6) Put stopper on flask & shake/mix solution until it's homogenous
  - 7) Pour solution into a 500mL bottle and label it with '1x recording', your initials, and date (use colored tape)
  - 8) Store bottle in 4 degrees fridge, clean up area