

Recipes for Physiological Recording

Sucrose slicing solution

Reagents	MM	MW	Gram/L	Gram/4L
KCl	2.5	74.56	0.186	0.746
NaH ₂ PO ₄	1.25	137.99	0.173	0.692
MgCl ₂	7	203.3	7ml stock	28ml stock
CaCl ₂	0.5	147.02	0.5ml stock	2ml stock
NaHCO ₃	25	81.01	2.1	8.4
Glucose	11	198.17	2.18	8.72
Sucrose	234	342.3	80.1	320.4

K-gluconate intracellular solution

Reagents	Concentration (mM)	Molecular weight	To make 100ml (mg)	To make 20ml (mg)
KCl	20	74.56	149	
K-gluconate	120	234.56	2810	
MgCl ₂	2	203.3	40	
EGTA	0.2	380.4	7.6	
HEPES	10	238.3	238.3	
*Na ₂ -ATP	2	551.1	112.2	

pH 7.3 mit KOH, ~305mOsm

Potassium Intracellular solution for 100ml

1. Potassium Gluconate 120mM 2,810.5mg
2. Potassium Chloride 20 150
3. GTP 0.3
4. EGTA 0.2 (or 0.02) 0.8 (0.08)
5. HEPES 10 24
6. Buffer pH to 7.25 with KOH, and filtered with 0.2µm filter, then add ATP
7. ATP-Mg 4 200
8. Aliquot by 1.5ml

Cesium Intracellular solution for 100ml (243mM, modified from Weliky and Katz, J. Neurosci. 1994, 14:7291-7305) @

Reagents	Concentration (mM)	Molecular weight	To make 100ml (mg)	To make 20ml (mg)
D-gluconic acid (free acid, 45-50% w/w)	110	196.2	3.5ml solution	0.7ml
CsOH	110	167.9	1,847	369.4
CsCl ₂	10	168.4	168.4	33.7
EGTA	1	380.4	38	7.6
CaCl ₂	1	147.02	14.7	2.94
HEPES	10	238.3	238.3	47.7
*ATP-Mg	1	507.2	51	10.14

*Filtered with 0.2µm filter before adding ATP-Mg.

***Take 3.0ml above solution, then + 9mg biocytin (0.3%) + 5.2mg QX-314 (lidocaine, 5mM) as final recording solution**

*QX-314 (lidocaine) (5.2mg to make 3.0ml)	5mM	343.3	171.7mg	34.3mg
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***the gluconic acid is calculated based on the formula density (1.25)=Mass/volume.**

135 KCl for IPSC (285mosm) for 100ml from Zhou and Hablitz 1999 (J. Neurophysiol. 81:976) and Kraushaar and Jonas 2000 (J. Neurosci 20:5594)

1. KCl	135mM	1,0125mg
2. HEPES	10	24
3. Na-ATP	2	100
4. Na-GTP	0.2	10
5. MgCl ₂	2	41
6. EGTA	0.1	0.4
7. PH to 7.4 with KOH		

0.3% Biocytin solution for 2ml

Biocytin	27	6 (8.12mM)
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Aliquot by 50µl each

0.3% Lucifer Yellow

Lucifer Yellow	6.5	6 (142mM)
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Aliquot by 50µl each

Alexa

Intracellular solution	250µl
Alexa	25µl (0.365% sol)

Aliquot by 5 µl each

KREBS-RINGER SOLUTION FOR 5.5L OF WATER (172mM)

Prepare in this sequence:

1. Sodium Chloride	124	39,860mg
2. Potassium Chloride	2.5	1,025 use fine scale
3. Sodium Phosphate, monobasic 1.25		949 use fine scale
4. Sodium bicarbonate	26	12,015

Following two solutions need to prepare separately 100ml of stock 1M Calcium Chloride and 100ml of stock 1M Magnesium Sulfate

1. Calcium Chloride	14,700mg
2. Magnesium Sulfate with 7H ₂ O	24,650mg

Before preparing the solutions:

- Clean the scale surfaces with wet and then dry kimwipes
- Rinse weighing spatulas in deionized water and dry out with kimwipes
- Take weighing dishes only from closed boxes in the draw

- Put weighing dishes and spatulas only on the clean paper towel
- Wipe out the spatulas with kimwipes after weighing each chemicals
- Tightly close the bottles and put them in the cold room right away after the chemicals dissolved

1.5L of final solution + 3.0ml stock Calcium Chloride (2mM) + 1.5ml of stock Magnesium Sulfate (1.0) + 2.0 scoop of dextrose (15mM) or 1.5 scoop of dextrose (10mM)

Yousheng Shu intracellular solution

	MW	mM	gm/100ml	
K-gluconate	234.25	145	3.397	
KCl	74.56	3	0.022	0.3ml 1M stock
MgCl ₂	203.3	2	0.041	0.2ml 1M stock
Na ₂ ATP	551.1	2	0.110	
HEPES	238.3	10	0.238	
EGTA	380.4	0.2	0.008	0.2ml 100mM stock

pH 7.25 280-290 mOsm

Alexa Fluor 488 biocytin 0.975 mg/ml=1 mM
Final concentration will be 100uM alexa dye plus 0.1% biocytin

Or add biocytin to final concentration 0.2%
Alexa Fluor 488 0.57mg/ml= 1 mM

Reyes A and Sakmann B (1999) solution for both interneurons and pyramidal cells

K-gluconate intracellular solution (current-clamp recordings in pairs) contained

(~305mOsm)

Material	Concentration (mM)	MW	Mg/50ml	Mg/100ml
K-gluconate	120	234.25	1410	
KCl	20	74.55	74.55	
Na ₂ ATP	4	551.1	112.22	
Na ₂ GTP	0.3	565.12	8.48	
Na ₂ phosphocreatine	5	255.1	63.78	
EGTA	0.1	380.4	1.902	
HEPES	10	238.3	119.15	

FORMULA(for making 50ml): AMOUNT(mg)=MWxCmM/20

MW: molecular weight

CmM: concentration of mM

From Reyes A and Sakmann B (1999) Developmental switch in the short-term modification of unitary EPSPs evoked in layer 2/3 and layer 5 pyramidal neurons of rat neocortex. J. Neurosci. 19:3827-35

6 mM KCl, modified from Reyes A and Sakmann B (1999, JN) solution

K-gluconate intracellular solution (current-clamp recordings in pairs) contained (~305mOsm)

Material	Concentration (mM)	MW	Mg/50ml	Mg/100ml
K-gluconate	120	234.25	1410	2810
KCl	6	74.55	22.5	45
Na ₂ ATP	4	551.1	112.22	112.2
Na ₂ GTP	0.3	565.12	8.48	17

Na2 phosphocreatine	5	255.1	63.78	127.6
EGTA	0.2	380.4	3.8	7.6
HEPES	10	238.3	119.15	238.3

FORMULA(for making 50ml): $AMOUNT(mg) = MW \times CmM / 20$

MW: molecular weight

CmM: concentration of mM

From Reyes A and Sakmann B (1999) Developmental switch in the short-term modification of unitary EPSPs evoked in layer 2/3 and layer 5 pyramidal neurons of rat neocortex. J. Neurosci. 19:3827-35