

Materials List for:

Direct Gene Knock-out of Axolotl Spinal Cord Neural Stem Cells via Electroporation of CAS9 Protein-gRNA Complexes

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Materials

Name	Company	Catalog Number	Comments
Agarose	Sigma-Aldrich	A9539	
Benzocaine	Sigma-Aldrich	E1501-100G	
Benzocaine 0.03 % (wt/vol)	Mix 500 ml of 10× TBS, 500 ml of 400% (wt/vol) Holtfreter's solution and 30 ml of 10% (wt/vol) benzocaine stock solution. Fill up the volume to 10 L with dH2O. The solution can be stored at room temperature for up to 6 months.		
Benzocaine 10 % (wt/vol)	Mix 50 g of benzocaine in 500 ml of 100% (vol/vol) ethanol. The solution can be stored at room temperature for up to 12 months.		
Borosilicate glass capillaries 1.2 mm O.D., 0.94 mm I.D.	Stutter Instrument	BF120-94-8	
CaCl ₂ ·2H ₂ O	Merck	102382	
CAS9 buffer, 10x	Mix 200 mM HEPES and 1.5 M KCl in RNase-free water. Adjust pH to 7.5. Filter sterilize, aliquot and store at -20 °C for up to 24 months		
CAS9-NLS protein	PNA Bio	CP03	
Cell culture dishes, 10cm	Falcon	351029	
Dumont #5 - Fine Forceps	Fine Scientific Instruments	11254-20	
Electroporator	Nepa Gene	NEPA21	
	BEX	Pulse Generator CUY21EDIT II	
Fast Green FCF	Sigma-Aldrich	F7252-5G	
Fast Green FCF Solution, 5x	Dissolve 12.5 mg of Fast Green FCF powder in 10 mL of 1× PBS.		
Flaming/Brown Micropipette Puller	Stutter Instrument	P-97	
Holtfreter's solution 400% (wt/vol)	Dissolve 11.125 g of MgSO ₄ ·7H ₂ O, 5.36 g of CaCl ₂ ·2H ₂ O, 158.4 g of NaCl and 2.875 g of KCl in 10 L of dH2O. The solution can be stored at room temperature for up to 6 months.		
KCl	Merck	104936	
MgSO ₄ ·7H ₂ O	Merck	105886	

Microloader pipette tips	Eppendorf	5242956003	
Micromanipulator	Narishige	MN-153	
NaCl	Merck	106404	
Pneumatic PicoPump	World Precision Instruments	SYS-PV830	
Ring Forceps	Fine Scientific Instruments	11103-09	
Stereomicroscope	Olympus	SZX10	
Tris base	Sigma-Aldrich	T6066	
Tris-buffered saline, 10x	Dissolve 24.2 g of Tris base and 90 g of NaCl in 990 ml of dH ₂ O. Adjust pH to 8.0 by adding 10 ml of 37% (vol/vol) HCl. The solution can be stored at room temperature for up to 6 months.		
Tweezers w/Variable Gap 2 Round Platinum Plate Electrode, 10mm diameter	Nepa Gene	CUY650P10	