

Materials List for:

# Generation of Chimeric Axolotls with Mutant Haploid Limbs Through Embryonic Grafting

Lucas D. Sanor<sup>1</sup>, G. Parker Flowers<sup>1</sup>, Craig M. Crews<sup>1</sup>

<sup>1</sup>Department of Molecular, Cellular, and Developmental Biology, Yale University

Correspondence to: Craig M. Crews at [craig.crews@yale.edu](mailto:craig.crews@yale.edu)

URL: <https://www.jove.com/video/60156>

DOI: [doi:10.3791/60156](https://doi.org/10.3791/60156)

## Materials

Name	Company	Catalog Number	Comments
#55 Dumont Forceps	Fine Science Tools	11295-1	Only use Dumostar material (can be autoclaved)
Amphotericin B	Sigma Aldrich	A2942-20ML	20 mL
Antibiotic-Antimycotic 100x	Thermo Fisher	15240062	
Ciprofloxacin	Sigma Aldrich	17850-5G-F	
Ficoll 400 (polysucrose 400)	bioworld	40600032-3	Ficoll 400
Gentamicin	Sigma Aldrich	G1914-250MG	
Heating/Cooling Incubator	RevSci	RS-IF-233	
Human Chorionic Gonadotropin	Merk		Chorulon
Megascript T7 Transcription Kit	Thermo Fisher	AM1334	40 reactions
Miroscope Cooling Stage	Brook Industries	Custom	Custom
NLS Cas9 Protein	PNABio	CP01-200	4 vials of 50 µg protein each
Plasmocin	Invivogen	ant-mpt-1	Treatment level
<b>Recipes</b>			
1.0x Marc's modified Ringer's solution (MMR)			0.1 M NaCl, 2 mM KCl, 1 mM MgSO <sub>4</sub> , 2 mM CaCl <sub>2</sub> , 0.1 mM EDTA, 5 mM HEPES (pH 7.8), pH 7.4
40% Holtfreter's solution			20 mM NaCl, 0.2 mM KCl, 0.8 mM NaHCO <sub>3</sub> , 0.2 mM CaCl <sub>2</sub> , 4 mM MgSO <sub>4</sub> , pH to 7.4