

Materials List for:

# The *Xenopus* Oocyte Cut-open Vaseline Gap Voltage-clamp Technique With Fluorometry

Michael W. Rudokas<sup>1</sup>, Zoltan Varga<sup>1</sup>, Angela R. Schubert<sup>1</sup>, Alexandra B. Asaro<sup>1</sup>, Jonathan R. Silva<sup>1</sup>

<sup>1</sup>Department of Biomedical Engineering, Washington University in St. Louis

Correspondence to: Jonathan R. Silva at [jonsilva@wustl.edu](mailto:jonsilva@wustl.edu)

URL: <https://www.jove.com/video/51040>

DOI: [doi:10.3791/51040](https://doi.org/10.3791/51040)

## Materials

Name	Company	Catalog Number	Comments
External Solution	Brand	Catalog Number	[Final], weight, or volume
N-methyl-D-glucamine (NMDG)	Sigma-Aldrich	M2004	25mM
MES Sodium Salt	Sigma-Aldrich	M5057	90mM
HEPES	Research Products International	H75030	20mM
Calcium hydroxide	Sigma-Aldrich	239232	2mM
MES Hydrate	Sigma-Aldrich	M8250	variable (pH to 7.4)
Internal Solution			
N-methyl-D-glucamine (NMDG)	Sigma-Aldrich	M2004	105mM
MES Sodium Salt	Sigma-Aldrich	M5057	10mM
HEPES	Research Products International	H75030	20mM
Ethylene glycol-bis(2-aminoethylether)-N,N,N',N'-tetraacetic acid (EGTA)	Sigma-Aldrich	E4378	2mM
MES Hydrate	Sigma-Aldrich	M8250	variable (pH to 7.4)
Depolarizing Solution			
KCl	Sigma-Aldrich	221473	110mM
Magnesium chloride	Sigma-Aldrich	M8266	1.5mM
Calcium Chloride	Caisson	C021	0.8mM
HEPES	Research Products International	H75030	10mM
Pipet Solution			
KCl	Sigma-Aldrich	221473	3M
Saponin Solution			
Saponin	Sigma-Aldrich	47036	0.125g
Internal Solution	See above		50mL
Agar Bridge Solution			
N-methyl-D-glucamine (NMDG)	Sigma-Aldrich	M2004	100ml of 1M
HEPES	Research Products International	H75030	1.2g
MES Hydrate	Sigma-Aldrich	M8250	variable (pH to 7.4)
Granulated Agar	Research Products International	A20250	3%
NMDG Storage Solution			
NMDG, HEPES, MES Hydrate solution	see above		40ml
Water			60ml
Name of Material/ Equipment	Company	Catalog Number	Comments/Description
High Performance Oocyte Clamp	Dagan	CA-1B	

Data Acquisition System	Axon CNS	Digidata 1440A	
Oscilloscope	Tektronix	TDS 210	
Rack Power Filter	APC	G5	
Heating/Cooling Bath Temperature Controller	Dagan	HCC-100A	
PC	Dell	Optiplex 990	
pCLAMP 10.3 Voltage Clamp Software	Molecular Devices, LLC	pCLAMP10.3	
TMC Vibration Control TableTop Platform	TMC	64 SERIES	
TMC Vibration Control Air Table	TMC	<a href="#">20 Series</a>	
V1/I Electrode Data Collector	Dagan	part of CA-1B	
MX10L Micromanipulator	Siskiyou	MX10L	
Bath/Guard (I/V) Headstage (with appropriate connectors)	Dagan	part of CA-1B	
Microscope	Omano	OM2300S-JW11	
Temperature Control Bath	Custom or Dagan	Custom or HE-204C	Custom chamber made from materials from Cool Polymers (D-series). Dagan also provides a prefeabricated stage (HE-204C).
Custom AgCl Pellet Container	Custom	Custom	Custom machined
Ag/AgCl electrode, pellet, 2.0 mm	Warner	E-206	
External Oocyte Bath	Custom or Dagan	Custom or CC-1-T-LB	Custom machined or purchased from Dagan
Internal Oocyte Bath	Custom or Dagan	Custom or CC-TG-ND	Custom machined or purchased from Dagan
Capillaries for Agar Bridges and Pulled Electrodes	Warner	G150T-4	
Rotatable Mounts for the Microscope, Micromanipulator, and Bath	Siskiyou	SD-1280P	
Fiber-Lite	Dolan-Jenner	LMI-600	
Regular Bleach	Clorox	470174-764	
Xenopus laevis Oocytes	Nasco	LM535M (sexually mature females)	
90 Na <sup>+</sup> External Solution	See Solutions sheet		
10 Na <sup>+</sup> Internal Solution	See Solutions sheet		
3 M KCL	See Solutions sheet		
Saponin	Sigma-Aldrich	47036	
NMDG Storage Solution	See Solutions sheet		
5mL transfer pipets	SciMart	GS-52	
Modified KCl electrode injector	BD	309659	Plastic syringe tip melted to allow for injection of solution into electrodes. Alternatively, a Microfil by WPI can be purchased.
Microvacuum	Custom	Custom	
Forceps	VWR	63040-458	
Oocyte Handling Tools (Pipette Pump)	VWR	53502-222	
Deionized Water Squirt Bottle	VWR	16649-911	
Vaseline Petroleum Jelly	Fisher Scientific	19-086-291	

Additional Materials Required for VCF Recordings:			
VCF Microscope	Nikon	Eclipse FN1	
Nikon CFI APO 40XW NIR Objective	Nikon	N40X-NIR	
X-Y Translator System for Fixed-Stage Upright Microscopes	Sutter Instruments	MT500-586	
External VCF Oocyte Bath	Custom		Custom machined. The chamber dimensions are 2.7 x 1.9 x 0.4 cm.
Internal VCF Oocyte Bath	Custom		Custom machined. The chamber dimensions are 1.6 x 1.6 x 0.4 cm.
Modified Temperature Control Bath	Custom		Custom chamber made from materials from Cool Polymers (D-series). The chamber dimensions of the modified temperature controller bath are 2.7 x 1.9 x 0.3 cm for the horizontal chamber, and 1 x 2.5 x 0.5 cm for the vertical chamber.