

# Modeling Paracrine Noncanonical Wnt Signaling *In Vitro*

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jove.com/video/63247

## Materials

Name	Company	Catalog Number	Comments
2-Mercaptoethanol	Sigma Aldrich	M-7522	
Antifade mounting medium with DAPI	Vector Laboratories	H-1200-10	Stored at 4 °C
Bovine serum albumin	Santa Cruz Biotechnology	sc-2323	Stored at 4 °C
C2C12 murine myoblast cell line	ATCC	CRL-1772	
Cell culture flasks, 75 cm <sup>2</sup>	ThermoFisher Scientific	156499	
Chamber Slide System, 4-well	ThermoFisher Scientific	154526	
Dulbecco's Modified Eagle's Medium (DMEM), high glucose (4.5 g/L), L-glutamine (2 mM)	Corning	10-017-CV	Stored at 4 °C
Falcon conical centrifuge tubes, 15 mL	Fisher Scientific	14-959-53A	
Falcon permeable support for 24-well plate with 0.4 μM transparent PET membrane	Corning	353095	
Fetal bovine serum	Fisher Scientific	W3381E	Stored in 50 mL aliquots at -20 °C
Gelatin solution, 0.1%	ATCC	PCS-999-027	Stored at 4 °C
Graduated and sterile pipette tips, 10 μL	USA Scientific	1111-3810	
Leukemia inhibitory factor (LIF), 106 unit/mL	Millipore Sigma	ESG1106	
L-glutamine 200 mM (100x)	Gibco	25030-081	
Lipofectamine RNAiMAX	Thermo Fisher Scientific	13778-075	
MEM non-essential amino acids (MEM NEAA) 100x	Gibco	11140-050	
Minimum essential medium (MEM)	Corning	10-022-CV	
Mitomycin C	Roche	10107409001	
Non-stick auto-glass coverslips, 24 x 55 mm	Springside Scientific	HRTCG2455	
O9-1 neural crest cell line	Millipore Sigma	SCC049	
Opti-MEM I, 1x	Gibco	31985-070	
Paraformaldehyde solution in PBS, 4%	Santa Cruz Biotechnology	sc-281692	Stored at 4 °C

Penicillin-streptomycin (10,000 U/ mL penicillin and 10,000 µg/mL streptomycin)	Fisher Scientific	W3470H	Stored in 10 mL aliquots at -20 °C
Phalloidin-iFluor 488	Abcam	ab176753	Stored at -20 °C, Keep out of light
Phosphate-buffer saline (PBS), 1x, without calcium and magnesium, pH 7.4	Corning	21-040-CV	Stored at 4 °C
Recombinant human fibroblast growth factor-basic (rhFGF-basic)	R&D Systems	233-FB-025	
Recombinant human/mouse Wnt5a protein	R&D Systems	645-WN-010	
Sodium pyruvate, 100 mM	Gibco	11360-070	
Square Petri dish with grid	Thomas Scientific	1219C98	
STO murine fibroblast feeder cells	ATCC	CRL-1503	
Triton X-100 solution	Sigma Aldrich	X100-100ML	
Trypsin-EDTA, 0.25%	Fisher Scientific	W3513C	Stored at 4 °C
Zeiss Apotome.2 fluorescence microscope	Carl Zeiss AG		
Zeiss inverted Axio Vert.A1 light microscope	Carl Zeiss AG		
Zen lite 2012 microscopy software	Carl Zeiss AG		imaging software