

# Discovery of Metastatic Regulators using a Rapid and Quantitative Intravital Chick Chorioallantoic Membrane Model

 Konstantin Stoletov<sup>1</sup>, Lian Willetts<sup>2</sup>, Perrin H. Beatty<sup>1</sup>, John D. Lewis<sup>1</sup>
<sup>1</sup>Department of Oncology, University of Alberta, Edmonton, Alberta, Canada <sup>2</sup>Department of Cell Biology & Anatomy, Cumming School of Medicine, University of Calgary

## Corresponding Author

 John D. Lewis  
 jdlewis@ualberta.ca

## Citation

 Stoletov, K., Willetts, L., Beatty, P.H., Lewis, J.D. Discovery of Metastatic Regulators using a Rapid and Quantitative Intravital Chick Chorioallantoic Membrane Model. *J. Vis. Exp.* (), e62077, doi:10.3791/62077 (2021).

## Date Published

February 3, 2021

## DOI

10.3791/62077

## URL

jove.com/video/62077

## Materials

Name	Company	Catalog Number	Comments
1 mL disposable syringes	BD	BD309659	
15 mL conical centrifuge tubes.	Corning	CLS430791-500EA	
18 gauge x 1 1/2 BD precision needle	BD	BD305196	we use 1.2mm x 40mm, it is possible to use shorter needles if preferred
2.5% Trypsin solution	many sources are available		
4T1 mouse breast cancer	ATCC	CRL-2539	
B16F10 mouse melanoma cell line	ATCC	CRL-6475	
Benchtop centrifuge.	many sources are available		Any TC compatible centrifuge that can be used to spin down the cells is suitable
Circular coverslips, 22 mm.	Fisher Scientific	12-545-101	
Collagenase	Sigma	C0130-100MG	
Confocal microscope			We use Nikon A1r
cotton swabs	many sources are available		must be sterilized before use
Culture media appropriate for the cell lines used	many sources are available		We grow HT1080, HEp3 and b16 cell lines in DMEM, 10% FBS media
Egg incubator	many sources are available		An exact model that is necessary depends on the scale of the screen. Available sources are MGF Company Inc., Savannah, GA, or Lyon Electric Company Inc., Chula Vista, CA
eppendor tubes , 1.5ml	Sigma	T4816-250EA	
Fertilized White Leghorn eggs	any local supplier		
fine forceps	many sources are available		must be sterilized before use
Hemocytometer	Millipore-Sigma	MDH-4N1-50PK	
HT1080 human fibrosarcoma cell line	ATCC	CCL-121	
Image analysis software			We use Nikon Elements

Lectin <i>Lens Culinary</i> Agglutinin (LCA) conjugated with Fluorescein or Rhodamine	Vector Laboratories	RL-1042, FL-1041	Dilute stock (5mg/ml) 50-100x depending on the microscope sensitivity. Must be a different color from the color of cell line used for screening
MDA-MB-468 human breast cancer	ATCC	HTB-132	
PBS (1x)	many sources are available		
Plastic weighting dishes	Simport	CA11006-614	dimensions are 78x78x25mm; many other sources are available
small surgical scissors	many sources are available		must be sterilized before use
Sodium borosilicate glass capillary tubes, outer diameter 1.0 mm, inner diameter 0.58 mm, 10 cm length	Sutter Instrument	BF100-58-10	
Square petri dishes (used as lids for the weighting dishes).	VWR	CA25378-115	dimensions are 100x100x15mm; many other sources are available
Stereo fluorescent microscope			We use Zeiss Lumar v12
Tygon R-3603 laboratory tubing	Cole-Parmer	AAC00001	1/32 in inner diameter, 3/32 in. outer diameter, 1/32 in. wall thickness
U-118 MG human glioblastoma	ATCC	HTB-15	
U-87 MG human glioblastoma	ATCC	HTB-14	
Vertical pipette puller	many sources are available		we use David Kopf Instruments, Tujunga, CA; Model 720