Materials List for:

Light-Induced Molecular Adsorption of Proteins Using the PRIMO System for Micro-Patterning to Study Cell Responses to Extracellular Matrix Proteins

Cristina Melero*[1], Aljona Kolmogorova*[1], Paul Atherton[1], Brian Derby[2], Adam Reid[3,4], Karin Jansen[5], Christoph Ballestrem[1]

[1]Faculty of Biology, Medicine and Health. Division of Cell Matrix, Biology and Regenerative Medicine, Wellcome Trust Centre for Cell-Matrix Research, The University of Manchester

[2]School of Materials, The University of Manchester

[3]Blond McIndoe Laboratories, School of Biological Sciences, Faculty of Biology, Medicine and Health, Division of Cell Matrix Biology and Regenerative Medicine, The University of Manchester, Manchester Academic Health Science Centre

[4]Department of Plastic Surgery & Burns, Wythenshawe Hospital, Manchester University NHS Foundation Trust, Manchester Academic Health Science Centre

[5]Department of Pathology, UMC Utrecht

*These authors contributed equally

Correspondence to: Karin Jansen at kjansen6@umcutrecht.nl, Christoph Ballestrem at christoph.ballestrem@manchester.ac.uk

Materials

Name	Company	Catalog Number	Comments
Alexa 488 protein labeling kit	Invitrogen	A10235	Working concentration: N.A.
Alexa 647 protein labeling kit	Invitrogen	A20173	Working concentration: N.A.
CAD cells	ECACC	8100805	Working concentration: N.A.
Conjugated fibrinogen-488	Molecular Probes	F13191	Working concentration: 10 µg/ml
DMEM culture medium	Gibco	11320033	Working concentration: N.A.
Epifluorescence Microscope**	Nikon	Eclipse Ti inverted	Working concentration: N.A.
Fibronectin	Sigma	F4759	Working concentration: 10 µg/ml (after labelling with Alexa 488 protein labeling kit, see above) (diluted in PBS)
Fiji-Image J	www.imagej.nih.gov	Version 2.0.0-rc-54/1.51f	Working concentration: N.A.
Fluorescent highlighter	Stabilo	Stabilo Boss Original	Working concentration: N.A.
HEPES	Gibco	15630080	Working concentration: 1M
Inkscape software	Inkscape	Check last update	Working concentration: N.A.
Laminin-red fluorescent rhodamine	Cytoskeleton, Inc.	LMN01	Working concentration: 10 µg/ml (diluted in PBS)
Leonardo software	Alvéole	version 4.11	Working concentration: N.A.
L-Glutamine	Sigma	G7513	Working concentration: 1%
Micro-manager software	Open imaging	Check last update	Working concentration: N.A.
Motorized x/y stage	PRIOR Scientific	Proscan II	Working concentration: N.A.
NIS Elements Software	Nikon	NIS Elements AR 4.60.00 64-bit (With Nikon jobs)	Working concentration: N.A.
PBS (without Ca^{2+}, Mg^{2+})	Sigma	D8537	Working concentration: 1X
PDMS Stencils	Alvéole	visit www.alveolelab.com	Working concentration: N.A.
PEG-SVA	Laysan bio, Inc.	MPEG-SVA-5000-1g	Working concentration: 50 mg/ml
Phalloidin 405	Abcam	ab176752	Working concentration: 1:1000
Photo-initiator (PLPP)	Alvéole	Classic PLPP	Working concentration: 14.5 mg/ml
Photo-initiator (PLPP gel)	Alvéole	PLPP gel	Working concentration: 4.76% diluted in ethanol
Plasma cleaner	Harrick Plasma	PDC-32G (115V)	PDC-32G-2 (230V)	Working concentration: N.A.

PLL-PEG	SuSoS (also distributed by Alvéole)	www.alveolelab.com	Working concentration: 0.1 mg/ml (diluted in PBS)
Poly-L-Lysine	Sigma	P4707	Working concentration: 0.01%
Primo equipment	Alvéole	www.alveolelab.com	Working concentration: N.A.
Pen/Strep	Thermo Fisher	15140122	Working concentration: 1%
Tubulin anti-alpha antibody	Abcam	DM1A	Working concentration: 1:1000 CAD cells
Tubulin anti-beta 3 antibody	Sigma	T8660	Working concentration: 1:500 DRG neurons
UV adhesive	Norland Products	NOA81	Working concentration: N.A.
1 well glass bottom dish	Cellvis	D35-20-1.5-N	Working concentration: N.A.
6 well glass bottom dish	Cellvis	P06-20-1.5-N	Working concentration: N.A.
20x objective**	Nikon	no phase ring (check updated catalogue)	Working concentration: N.A. **Epifluorescence microscope: images were acquired and patterns were generated on an Eclipse Ti inverted microscope (Nikon), coupled to PRIMO micro-patterning equipment (Alvéole), using a 20x objective (0.75 S Plan Fluor (nophasering, Nikon). Nikon specific filter sets for GFP, mCherry and Cy5 were used and fluorescent light source was LED (Lumencor) although other fluorescence sources and filter sets can be used. The microscope has an automated x/y stage (PRIOR Scientific) for the printing of multi-field patterning and Nikon Perfect Focus to prevent focus drift. The images were collected using a Retiga R6 (Q-Imaging) camera.