

**Materials List for:****Observation and Quantification of Telomere and Repetitive Sequences Using Fluorescence *In Situ* Hybridization (FISH) with PNA Probes in *Caenorhabditis elegans***Beomseok Seo<sup>1</sup>, Junho Lee<sup>1,2,3</sup><sup>1</sup>Institute of Molecular Biology and Genetics (IMBG), Seoul National University<sup>2</sup>Department of Biological Sciences, Seoul National University<sup>3</sup>Department of Biophysics and Chemical Biology, Seoul National UniversityCorrespondence to: Junho Lee at [elegans@snu.ac.kr](mailto:elegans@snu.ac.kr)URL: <https://www.jove.com/video/54224>DOI: [doi:10.3791/54224](https://doi.org/10.3791/54224)**Materials**

Name	Company	Catalog Number	Comments
PNA probe	PANAGENE	custom order	
Anti-Digoxigenin-Fluorescein, Fab fragments	Roche	11207741910	use 1:200 diluted in PBST
Digoxigenin-dUTP	Roche	11573152910	
Bovine serum albumin	SIGMA-ALDRICH	A-7906	
Paraformaldehyde	SIGMA-ALDRICH	P-6148	prepare 4% paraformaldehyde by heating in DW with few drops of NaOH. add 0.1 volume of 10x PBS.
Vectashield	Vector Laboratories	H-1200	
Hybridization solution			3x SSC, 50% formamide, 10% (w/v) dextran sulfate, 50 µg/ml heparin, 100 µg/ml yeast tRNA , 100 µg/ml sonicated salmon sperm DNA
Hybridization wash solution			2x SSC, 50% formamide
Formamide	BIONEER	C-9012	toxic
Methanol	Carlo Erba		
Acetone	Carlo Erba		
Heparin	SIGMA-ALDRICH	H3393	make 10 mg/ml for stock solution
Dextran sulfate	SIGMA-ALDRICH	67578	
10x PBS			For 1 L DW : 80 g NaCl, 2.0 g KCl, 27 g Na <sub>2</sub> HPO <sub>4</sub> •7H <sub>2</sub> O, 2.4 g KH <sub>2</sub> PO <sub>4</sub>
PBST			1x PBS, 0.1% tween-20
Polysorbate 20	SIGMA-ALDRICH	P-2287	Commercial name is Tween-20
Poly-L-Lysine solution (0.1% w/v)	SIGMA-ALDRICH	P-8920	prepare fresh 0.01% w/v solution before use
M9			3 g KH <sub>2</sub> PO <sub>4</sub> , 6 g Na <sub>2</sub> HPO <sub>4</sub> , 5 g NaCl, 1 ml 1 M MgSO <sub>4</sub> , H <sub>2</sub> O to 1 L
Bleaching solution			20% sodium hypochlorite, 0.5 M KOH
Antibody buffer			1x PBST, 1 mM EDTA, 0.1% BSA, 0.05% Sodium azide (toxic)
Blocking solution			Antibody buffer with 5% bovine serum albumin (BSA)
Illustra Microspin G-50	GE healthcare	27-53310-01	

20x SSC			To make 1 L, 175.3 g of NaCl, 88.2 g of sodium citrate, H <sub>2</sub> O to 1 L, adjust pH to 7.0
2x SSCT			2x SSC, 0.1% tween-20
10x digoxigenin-dUTP mix			1 mM dATP, 1 mM dGTP, 1 mM dCTP, 0.65 mM dTTP, 0.35 mM DIG-11-dUTP
PCR purification columns	Cosmo genetech	CMR0112	
Glass cleaner / ULTRA CLEAN	Dukssan pure chemicals	8AV721	
Multi-well glass slide	MP biomedicals	96041205	
Nematode growth media			To make 1 L, 3 g of NaCl, 17 g of agar, 2.5 g of peptone, H <sub>2</sub> O to 974 ml. Autoclave and cool the flask. Add 1 ml of 1 M CaCl <sub>2</sub> , 1 ml of 4 mg/ml cholesterol in ethanol, 1 ml of 1 M MgSO <sub>4</sub> , 25 ml of 1 M KPO <sub>4</sub> .
Levamisole	SIGMA-ALDRICH	196142	
Razor	Feather	blade No. 11	
Rnase A	Enzyomics		
BSA	SIGMA-ALDRICH	A7906	
Confocal microscope	Zeiss	LSM 510	EC Plan-Neofluar 100X was used as objective lens.
Humid chamber			Plastic box filled with paper towel soaked in DW
Image Analysis Software	Dr. Peter Landsdorp	TFL-telo	<a href="http://www.flintbox.com/public/project/502">http://www.flintbox.com/public/project/502</a>