Materials List for:

## High-Throughput Quantitative RT-PCR in Single and Bulk *C. elegans* Samples Using Nanofluidic Technology

Laetitia Chauve\*<sup>1</sup>, Jérémie Le Pen\*<sup>2,3,5</sup>, Francesca Hodge<sup>1</sup>, Pia Todtenhaupt<sup>1</sup>, Laura Biggins<sup>1</sup>, Eric A. Miska<sup>2,3,4</sup>, Simon Andrews<sup>1</sup>, Olivia Casanueva<sup>1</sup>

Correspondence to: Olivia Casanueva at olivia.casanueva@babraham.ac.uk

URL: https://www.jove.com/video/61132

DOI: doi:10.3791/61132

## **Materials**

Name	Company	Catalog Number	Comments
100µM stock primers for genes of interest.			
20X DNA Binding Dye	Fluidigm	100-7609	for 96.96 chip (Single-Array Chip)
2X Assay Loading Reagent	Fluidigm	100-7611	
384 well plates			
8-strip PCR tubes			
96 well ice block			
96 well plates			
96.96 Dynamic Array IFC	Fluidigm	BMK-M-96.96	Referenced throughout the manuscript as "Single-Array Chip"
96-well sealing tape			
BioMark & EP1 Software	Fluidigm	101-6793	Contains: Biomark HD Data Collection software, Real-Time PCR Analysis software, SNP Genotyping Analysis software, Digital PCR Analysis software, Curve Analysis software
BioMark or BioMark HD system	Fluidigm	100-2451 K1	Referenced throughout the manuscript as "nanofluidics thermocycler"
Control Line Fluid Kit for 96.96 and FlexSix IFCs	Fluidigm	89000021	Referenced throughout manuscript as "control line fluid"
DNA Suspension buffer	TEKnova	T0021	
domed PCR caps			
Exonuclease I	New England BioLabs	M0293L	
Exonuclease I reaction buffer	New England BioLabs	B0293S	
FLEXsix DELTAgene Sample Reagent	Fluidigm	100-7673	referenced throughout manuscript as "sample reagent"
FLEXsix Gene Expression IFC	Fluidigm	100-6308	referenced throughout manuscript as "Multi-Array Chip"
Fluidigm Real-Time PCR Analysis User Guide	Fluidigm	68000088	This is the protocol used as a basis for section 2 of the protocol, referenced within the manuscript.

<sup>&</sup>lt;sup>1</sup>Babraham Institute

<sup>&</sup>lt;sup>2</sup>Gurdon Institute, University of Cambridge

<sup>&</sup>lt;sup>3</sup>Department of Genetics, University of Cambridge

<sup>&</sup>lt;sup>4</sup>Wellcome Trust Genome Campus, Wellcome Trust Sanger Institute

<sup>&</sup>lt;sup>5</sup>Laboratory of Virology and Infectious Disease, The Rockefeller University

<sup>\*</sup>These authors contributed equally

IFC Controller MX	Fluidigm	68000112 11	Referenced throughout the manuscript as "nanofluidics PCR priming machine"
IFC Controllers SOFTWEAR	Fluidigm	100-2297	Contains all softwear and scripts required for running the IFC controller MX
liquid nitrogen in dewar			
Microcentrifuge	StarLab	C1301B-230V	Used to spin down PCR tube strips in the protocol.
Nuclease Free Water			
plate spinner	Labnet	K4050725	mini plate spinner, mps 1000. referenced through the protocol as "tabletop plate spinner"
Power SYBR Green Cells-to-Ct kit	invitrogen	4402955	This kit is that which is adapted for use in nematodes in the Wormto-CT protocol. Contense: Store Stop Solution, DNase I and 20X RT Enzyme Mix at -20°C. Store Lysis Solution, 2X SYBR RT Buffer (referenced throughout the manuscript as RT buffer), and Power SYBR®Green PCR Master Mix (refferenced througout the manuscript as PCR Master Mix). This Kit is for 400 reactions, kits also available for 40 and 100 reactions.
PreAmp Master Mix	Fluidigm	100-5580, 100-5581	
Reverse Transcription Master Mix  —480 reactions	Fluidigm	100-6299	One tube also available for 96 reactions (PN100-6298)
Rnase Free water			
SsoFast EvaGreen Supermix with Low ROX	Bio-Rad Laboratories	172-5211	referenced throughout the manuscript as "fluorescent probe supermix"
Standard 96 and 384-well Thermal Cycler			
TE Buffer (10mM Tris, pH8.0, 1.0mM EDTA	TEKnova	T0224	Referenced throughout the manuscript as Tris-EDTA buffer
ThermoMixer C	Eppendorf	5382000031	Referenced throughout the text as "thermal mixer"
Trizol	Thermo-Fisher	15596026	Referenced through the protocol as "guanidium thiocyanate-phenol-chloroform"
Warm water bath			