Materials List for:

## A Protein Microarray Assay for Serological Determination of Antigen-specific Antibody Responses Following *Clostridium difficile* Infection

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URL: https://www.jove.com/video/57399

DOI: doi:10.3791/57399

## **Materials**

Name	Company	Catalog Number	Comments
BioRobotics MicroGrid II arrayer	Digilab, Malborough, MA, USA	N/A	Contact arrayer used to automated spotting of the antigens onto the slides.
Scanner InnoScan 710.	Innopsys	N/A	A fluorescent microarray slide scanner with a red (Cy5) laser to read the reaction.
MAPIX software version 7.2.0	Innopsys	N/A	Measure signal intensities of the spots.
Silicon contact pin	Parallel Synthesis Technologies	SMT-P75	Print the samples onto the slides.
Thermo Scientific Nalgene Desiccator	Thermo Scientific	41102426	To store the new and printed slides.
384-well plate	Genetix	X7022UN	To prepare the antigens.
Plate cover	Sigma Aldrich, UK	CLS6570-100EA	To reduce evaporation of the samples.
Aminosilane slides	Schott, Germany	1064875	The slide of choice for printing the antigens.
Slide holders	GraceBio Labs, USA	204862	Divide the slides into identical 16 subarrays. These holders are reusable, removable, leak-proof wells.
Candida albicans lysate	NIBSC	PR-BA117-S	Positive control
Tetanus Toxoid	Athens Research and Technology	04/150	Positive control
Immunoglobulin G (IgG), Normal Human Plasma	Athens research and technology	16-16-090707	Purified Native Human Immunoglobulin G IgG, Human Plasma.
Immunoglobulin G1 (IgG1), Normal Human Plasma	Athens research and technology	16-16-090707-1	Purified Native Human Immunoglobulin G1 IgG1, Human Plasma.
Immunoglobulin G2 (IgG2), Normal Human Plasma	Athens research and technology	16-16-090707-2	Purified Native Human Immunoglobulin G2 IgG2, Human Plasma.
Immunoglobulin G3 (IgG3), Normal Human Plasma	Athens research and technology	16-16-090707-3	Purified Native Human Immunoglobulin G3 IgG3, Human Plasma.
Immunoglobulin G4 (IgG4), Normal Human Plasma	Athens research and technology	16-16-090707-4	Purified Native Human Immunoglobulin G4 IgG4, Human Plasma.

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Immunoglobulin A (IgA), Human Plasma	Athens research and technology	16-16-090701	Purified Native Human Immunoglobulin A (IgA), Human Plasma.
Immunoglobulin A1 (IgA1), Human Myeloma Plasma	Athens research and technology	16-16-090701-1M	Purified Native Human Immunoglobulin A1 (IgA1), Human Plasma.
Immunoglobulin A2 (IgA2), Human Myeloma Plasma	Athens research and technology	16-16-090701-2M	Purified Native Human Immunoglobulin A2 (IgA2), Human Plasma.
Immunoglobulin M (IgM), Human Plasma	Athens research and technology	16-16-090713	Purified Native Human Immunoglobulin M (IgM), Human Plasma.
Biotinylated Goat Anti-Human IgG Antibody, gamma chain specific	Vector Labs	BA-3080	Goat anti- human IgG (γ-chain specific)-biotin antibody reacts specifically with human IgG but not with other immunoglobulins.
Mouse Anti-Human IgG1 Hinge-BIOT	Southern Biotec	9052-08	Goat anti- human IgG1 biotin antibody reacts specifically with human IgG1 but not with other immunoglobulins.
Mouse Anti-Human IgG2 Fc-BIOT	Southern Biotec	9060-08	Goat anti- human IgG2 -biotin antibody reacts specifically with human IgG 2but not with other immunoglobulins.
Mouse Anti-Human IgG3 Hinge-BIOT	Southern Biotec	9210-08	Goat anti- human IgG3-biotin antibody reacts specifically with human IgG3 but not with other immunoglobulins.
Mouse Anti-Human IgG4 pFc'- BIOT	Southern Biotec	9190-08	Goat anti- human IgG-biotin antibody reacts specifically with human IgG but not with other immunoglobulins.
Anti-Human IgA, alpha chain specific, made in goat - Biotinylated	Vector Labs	BA-3030	Goat anti- human IgG -biotin antibody reacts specifically with human IgG but not with other immunoglobulins.
Mouse Anti-Human IgA1-BIOT	Southern Biotec	9130-08	Goat anti- human IgG -biotin antibody reacts specifically with human IgG but not with other immunoglobulins.
Mouse Anti-Human IgA2-BIOT	Southern Biotec	9140-08	Goat anti- human IgG -biotin antibody reacts specifically with human IgG but not with other immunoglobulins.
Mouse Anti-Human IgM-BIOT	Southern Biotec	9020-08	Goat anti- human IgG-biotin antibody reacts specifically with human IgG but not with other immunoglobulins.
0.2 mm syringe filter	Thermo scientific	723-2520	Filter the 5% BSA.
Bovine Serum Albumin (BSA)	Sigma Aldrich, UK	A7284	Use 5% BSA for blocking the slides.
Antibody diluent	Dako, UK	S3022	To dilute the serum and the secondary antibody.
Streptavidin Cy5	eBioscience	SA1011	Detection of the immune reaction.
Purified whole C. difficile toxins A and B (toxinotype 0, strain VPI 10463, ribotype 087)	Toxins Group, Public Health England	NA	
Purified whole C. difficile toxin B (CCUG 20309 toxin B only expressing strain)	Toxins Group, Public Health England	NA	

Precursor form of B fragment of binary toxin, pCDTb	University of Bath	NA	Produced in E. Coli from wholly synthetic recombinant gene construct. Amino acid sequence based on published sequence from 027 ribotype (http://www.uniprot.org/uniprot/A8DS70)
Purified native whole ribotype- specific (001, 002, 027) surface layer proteins	Dublin City University	NA	
Vigam (IVIg preparation 1)	Nottingham University Hospitals NHS Trust	N/A	
Privigen (IVIg preparation 2)	Nottingham University Hospitals NHS Trust	N/A	
Intratect (IVIg preparation 3)	Nottingham University Hospitals NHS Trust	N/A	