

SUPPLEMENTARY MATERIAL FOR:

**Median filter algorithm for estimating the threshold of detection on custom protein arrays**David D. Smith<sup>1</sup>, Susan Kovats<sup>2</sup>, Terry D. Lee<sup>1</sup>, and Leticia Cano<sup>3</sup><sup>1</sup>City of Hope National Medical Center and Beckman Research Institute, Duarte, CA, <sup>2</sup>Oklahoma Medical Research Foundation, Oklahoma City, OK, and <sup>3</sup>NHLBI, NIH, Bethesda, MD, USA*BioTechniques* 41:74-78 (July 2006)

Spot	Span=2	Span=6	Span=8	Span=9	Span=10
<b>A1</b>	<b>15</b>	<b>16</b>	<b>16</b>	<b>16</b>	<b>16</b>
<b>A2</b>	<b>10</b>	<b>16</b>	<b>16</b>	<b>16</b>	<b>16</b>
<b>A3</b>	<b>14</b>	<b>16</b>	<b>16</b>	<b>16</b>	<b>16</b>
<b>A4</b>	<b>11</b>	<b>16</b>	<b>16</b>	<b>16</b>	<b>16</b>
<b>A5</b>	<b>8</b>	<b>16</b>	<b>16</b>	<b>16</b>	<b>16</b>
A10	1	0	0	0	0
B1	2	0	0	0	0
B2	3	2	2	2	2
C1	1	0	0	0	0
C10	1	0	0	0	0
C11	1	0	0	0	0
D5	1	1	1	1	1
<b>E1</b>	<b>13</b>	<b>14</b>	<b>15</b>	<b>16</b>	<b>16</b>
<b>E2</b>	<b>10</b>	<b>7</b>	<b>12</b>	<b>11</b>	<b>15</b>
E3	8	8	7	6	8
E4	3	0	0	0	0
<b>F1</b>	<b>16</b>	<b>16</b>	<b>16</b>	<b>16</b>	<b>16</b>
<b>F2</b>	<b>15</b>	<b>16</b>	<b>16</b>	<b>16</b>	<b>16</b>
<b>F3</b>	<b>11</b>	<b>16</b>	<b>16</b>	<b>16</b>	<b>16</b>
<b>F4</b>	<b>11</b>	<b>16</b>	<b>16</b>	<b>16</b>	<b>16</b>
F5	6	3	0	0	0
<b>G1</b>	<b>14</b>	<b>14</b>	<b>14</b>	<b>16</b>	<b>16</b>
<b>G2</b>	<b>15</b>	<b>14</b>	<b>14</b>	<b>16</b>	<b>16</b>
<b>G3</b>	<b>13</b>	<b>11</b>	<b>12</b>	<b>13</b>	<b>16</b>
G4	8	4	5	3	4
G5	3	0	0	0	0
<b>H1</b>	<b>16</b>	<b>15</b>	<b>15</b>	<b>13</b>	<b>15</b>
<b>H2</b>	<b>10</b>	<b>15</b>	<b>16</b>	<b>16</b>	<b>16</b>
H3	11	8	10	11	12
H4	3	5	5	5	3

The threshold value for the median filter was determined by the partition analysis. Wells in the bolded rows were easily detectable by the naked eye.

## Short Technical Reports

	1	2	3	4	5	6	7	8	9	10	11	12	
A	2.5	0.5	0.25	0.05	0.025	0.005	0.0025	0.0005	0.00025	0.00005	0.000025	0.000005	Flu
B													
C													
D													
E	2.5	0.5	0.25	0.05	0.025	0.005	0.0025	0.0005	0.00025	0.00005	0.000025	0.000005	IgG
F	25	5	2.5	0.5	0.25	0.05	0.025	0.005	0.0025	0.0005	0.00025	0.00005	IgG
G	12.5	2.5	1.25	0.25	0.125	0.025	0.0125	0.0025	0.00125	0.00025	0.000125	0.000025	IgG
H	2.5	0.5	0.25	0.05	0.025	0.005	0.0025	0.0005	0.00025	0.00005	0.000025	0.000005	IgG

**Supplementary Figure S1. Template of antigen plate.** Flu vaccine was spotted in row A. Immunoglobulin G (IgG) was spotted in rows E–H. Rows B–D are blank rows containing buffer only. Values are the actual amount of material (in nanograms) spotted in the wells.