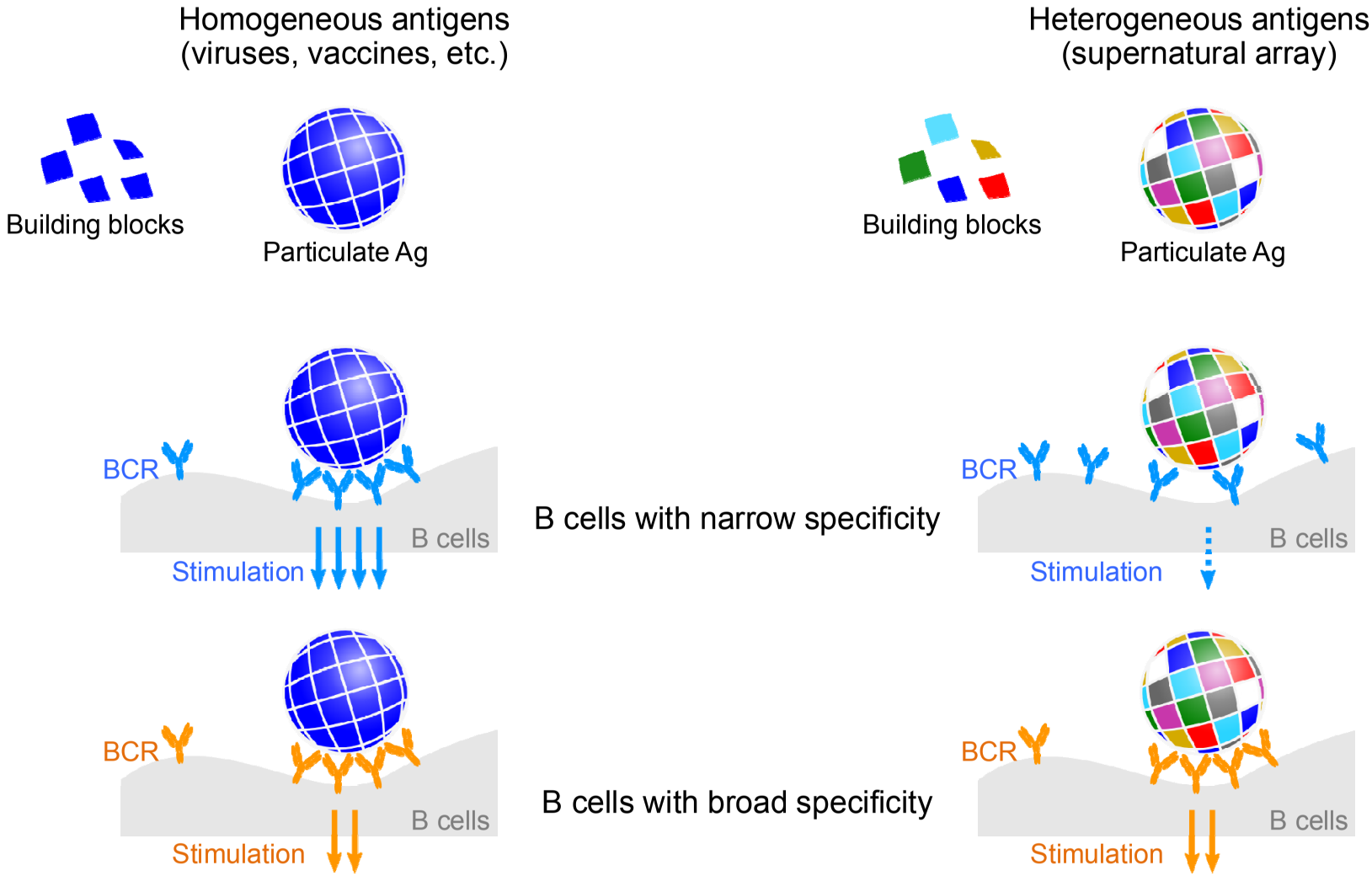

Homogeneous antigens
(viruses, vaccines, etc.)
Building blocks
Particulate Ag
Heterogeneous antigens
(supernatural array)
Building blocks
Particulate Ag
BCR
B cells
Stimulation
B cells with narrow specificity
BCR
B cells
Stimulation
BCR
B cells
Stimulation
B cells with broad specificity
BCR
B cells
Stimulation

Figure 1

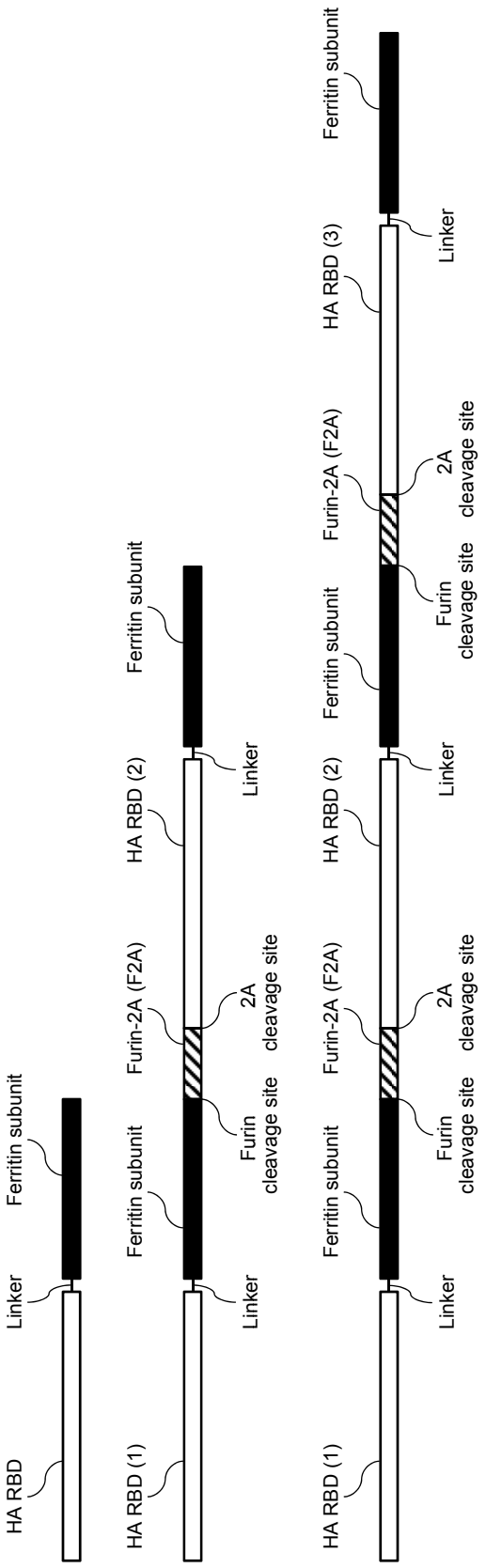

HA RBD
Linker
Ferritin subunit
HA RBD (1)
Linker
Ferritin subunit
Furin
cleavage site
2A
cleavage site
Furin-2A (F2A)
HA RBD (2)
Linker
Ferritin subunit
HA RBD (1)
Linker
Ferritin subunit
Furin
cleavage site
2A
cleavage site
Furin-2A (F2A)
HA RBD (2)
Linker
Ferritin subunit
HA RBD (3)
Linker
Ferritin subunit

Figure 2

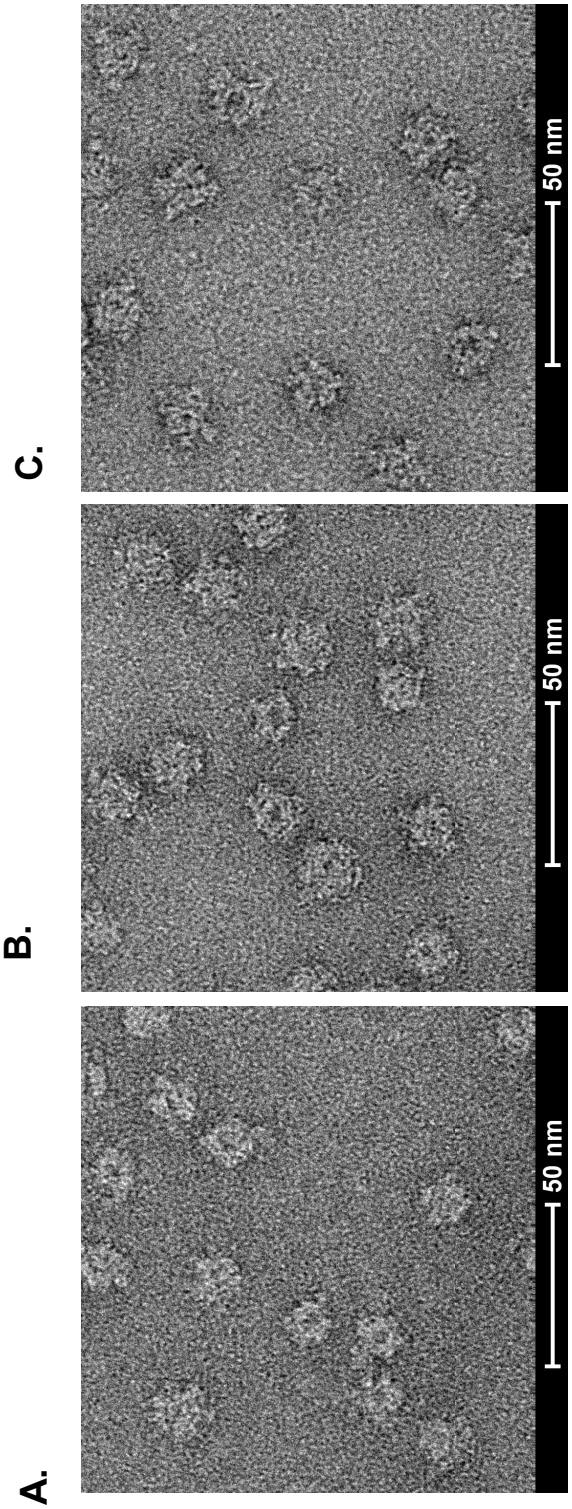


Figure 3

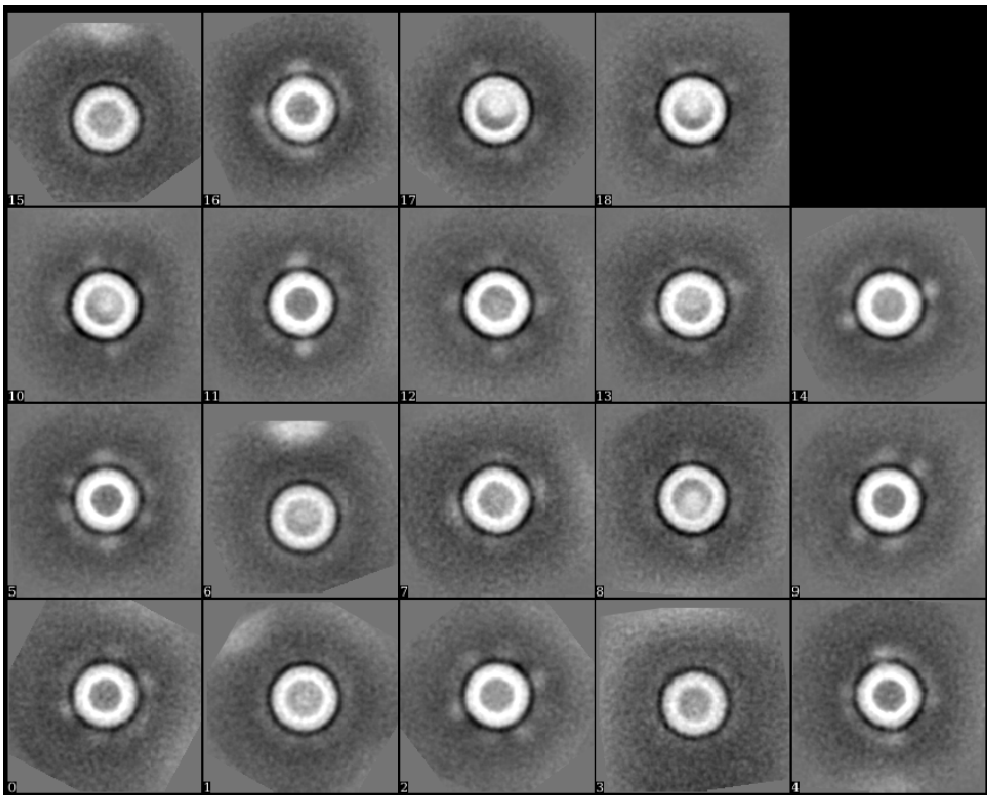


Figure 4

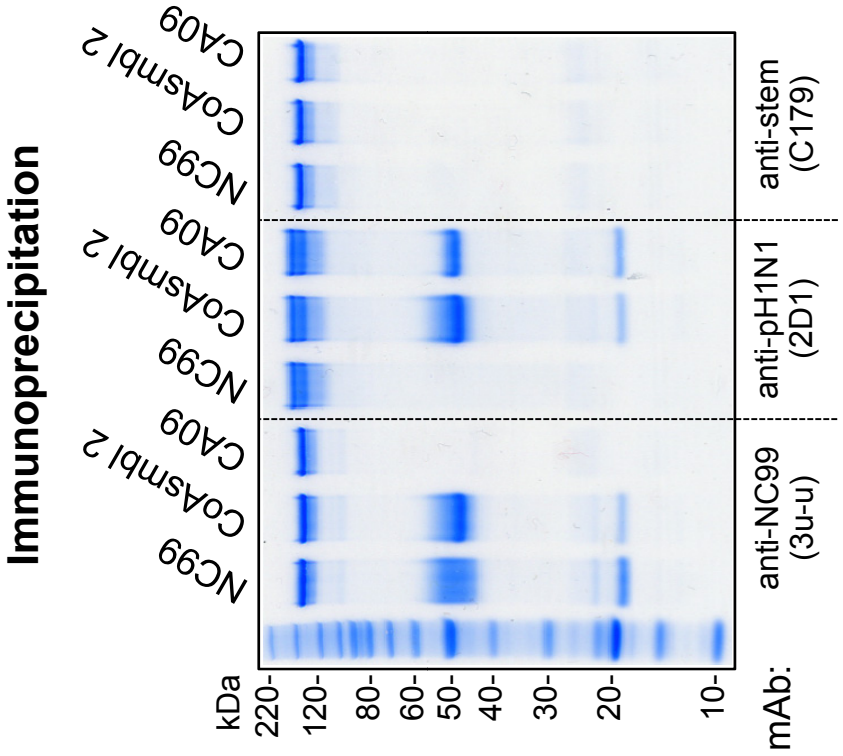

Immunoprecipitation
NC99
CoAsmbl 2
CA09
NC99
CoAsmbl 2
CA09
NC99
CoAsmbl 2
CA09
kDa
220-
120-
80-
60-
50-
40-
30-
20-
10-
mAb:
anti-NC99 (3u-u)
anti-pH1N1 (2D1)
anti-stem (C179)

Figure 5

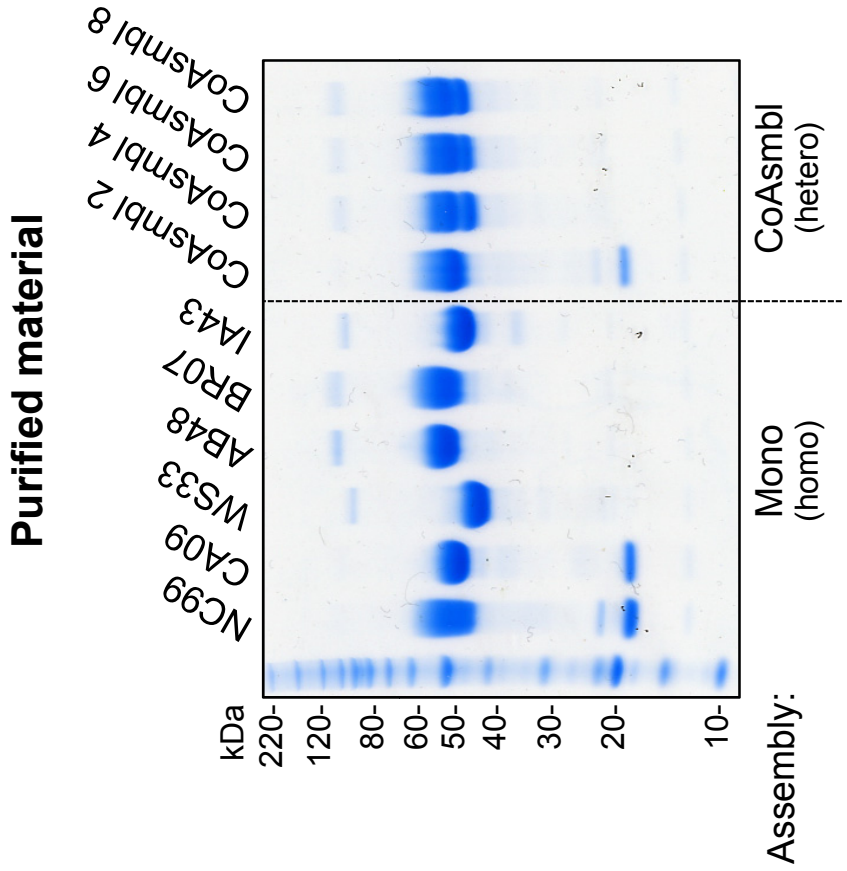

Purified material
CoAsmbl 8
CoAsmbl 6
CoAsmbl 4
CoAsmbl 2
IA43
BR07
AB48
WS33
CA09
NC99
kDa
220-
120-
80-
60-
50-
40-
30-
20-
10-
Assembly:
Mono
(homo)
CoAsmbl
(hetero)

Figure 6

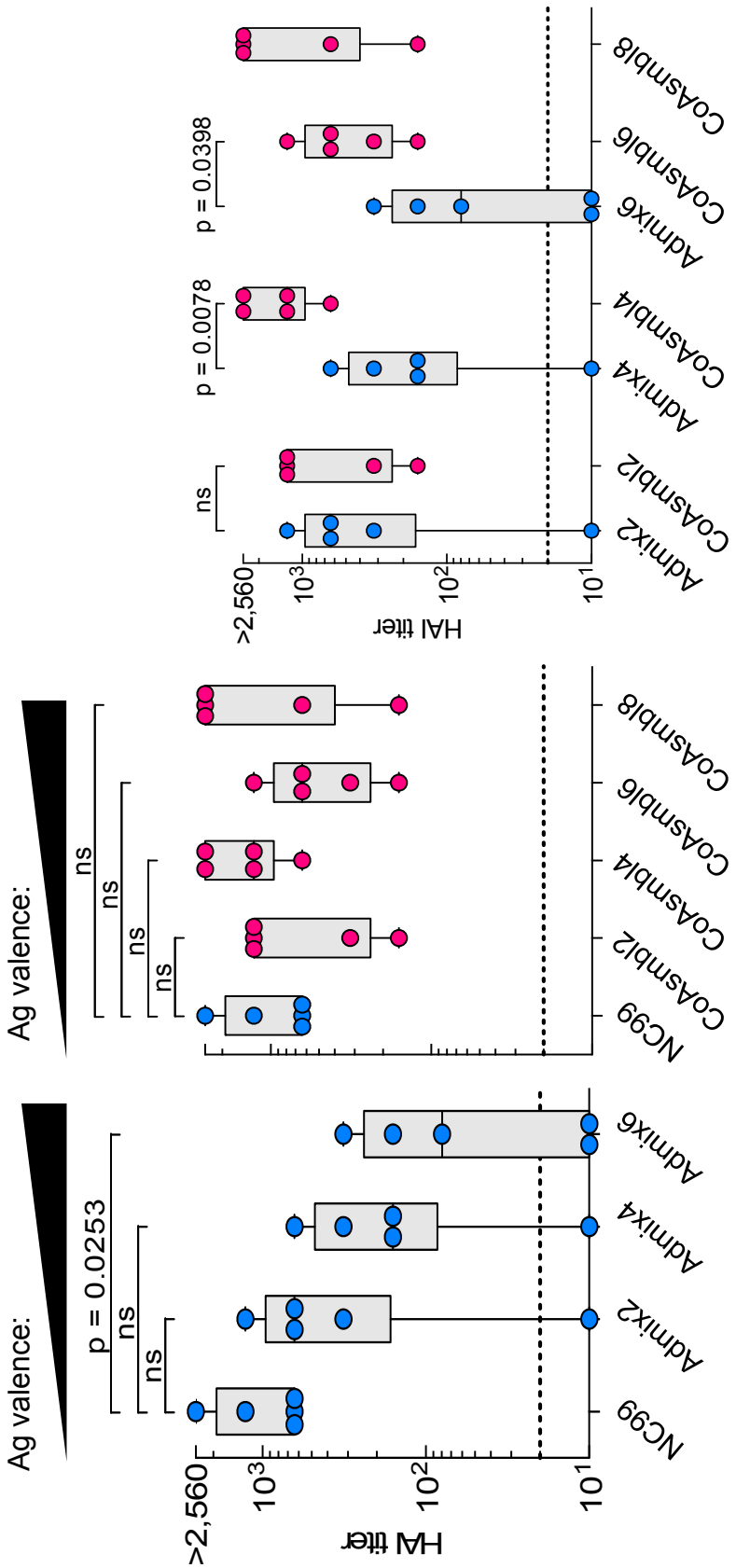

Ag valence:
p = 0.0253
ns
ns
HAI titer
>2,560
10^3
10^2
10^1
NC99
Admix2
Admix4
Admix6
Ag valence:
ns
ns
ns
ns
NC99
CoAsmb12
CoAsmb14
CoAsmb16
CoAsmb18
HAI titer
>2,560
10^3
10^2
10^1
ns
p = 0.0078
p = 0.0398
Admix2
CoAsmb12
Admix4
CoAsmb14
Admix6
CoAsmb16
CoAsmb18

Figure 7

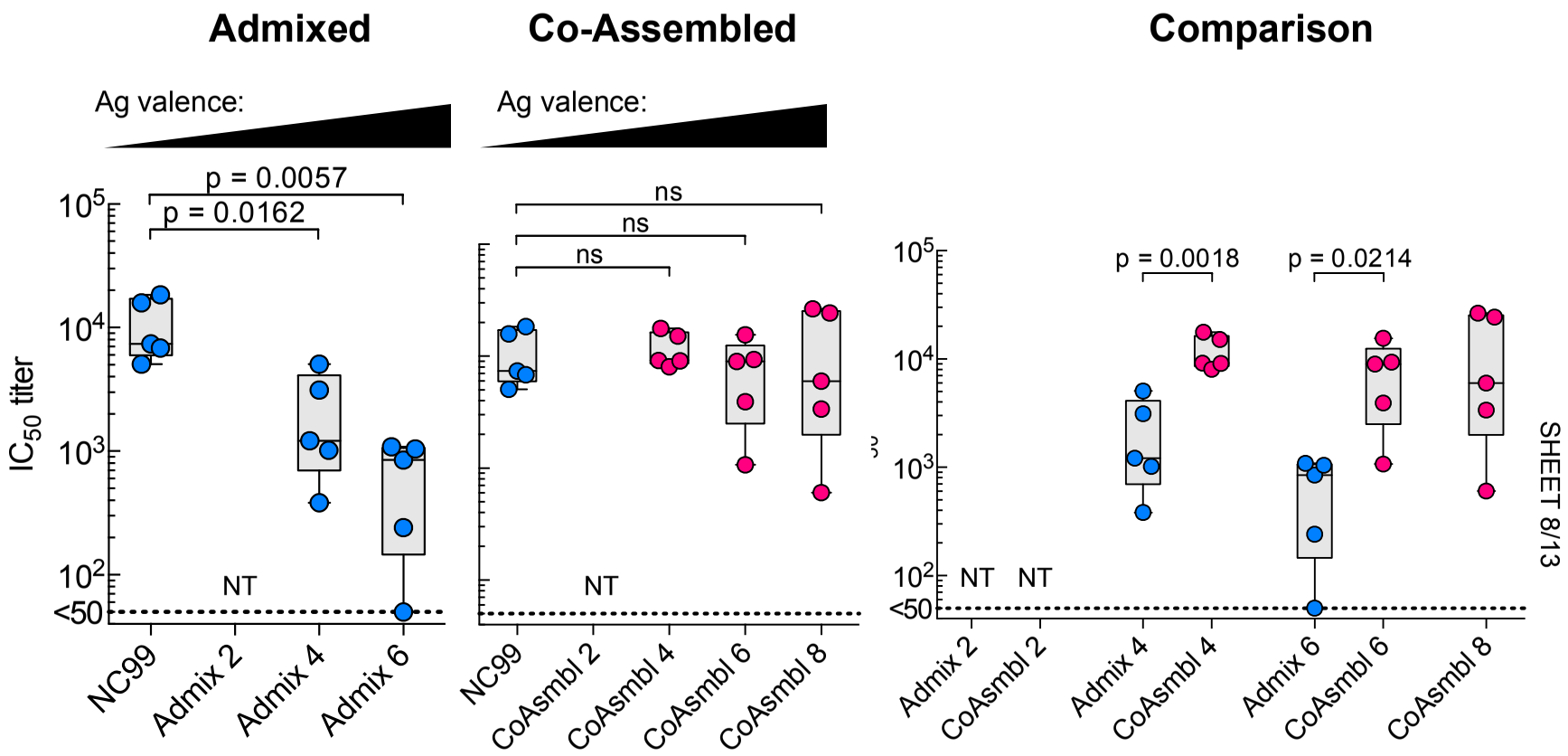

Admixed
Co-Assembled
Comparison
Ag valence:
Ag valence:
p = 0.0057
p = 0.0162
ns
ns
ns
p = 0.0018
p = 0.0214
IC50 titer
10^5
10^4
10^3
10^2
<50
NT
NT
NT
NT
NC99
Admix 2
Admix 4
Admix 6
NC99
CoAsmbl 2
CoAsmbl 4
CoAsmbl 6
CoAsmbl 8
Admix 2
CoAsmbl 2
Admix 4
CoAsmbl 4
Admix 6
CoAsmbl 6
CoAsmbl 8
SHEET 8/13

Figure 8

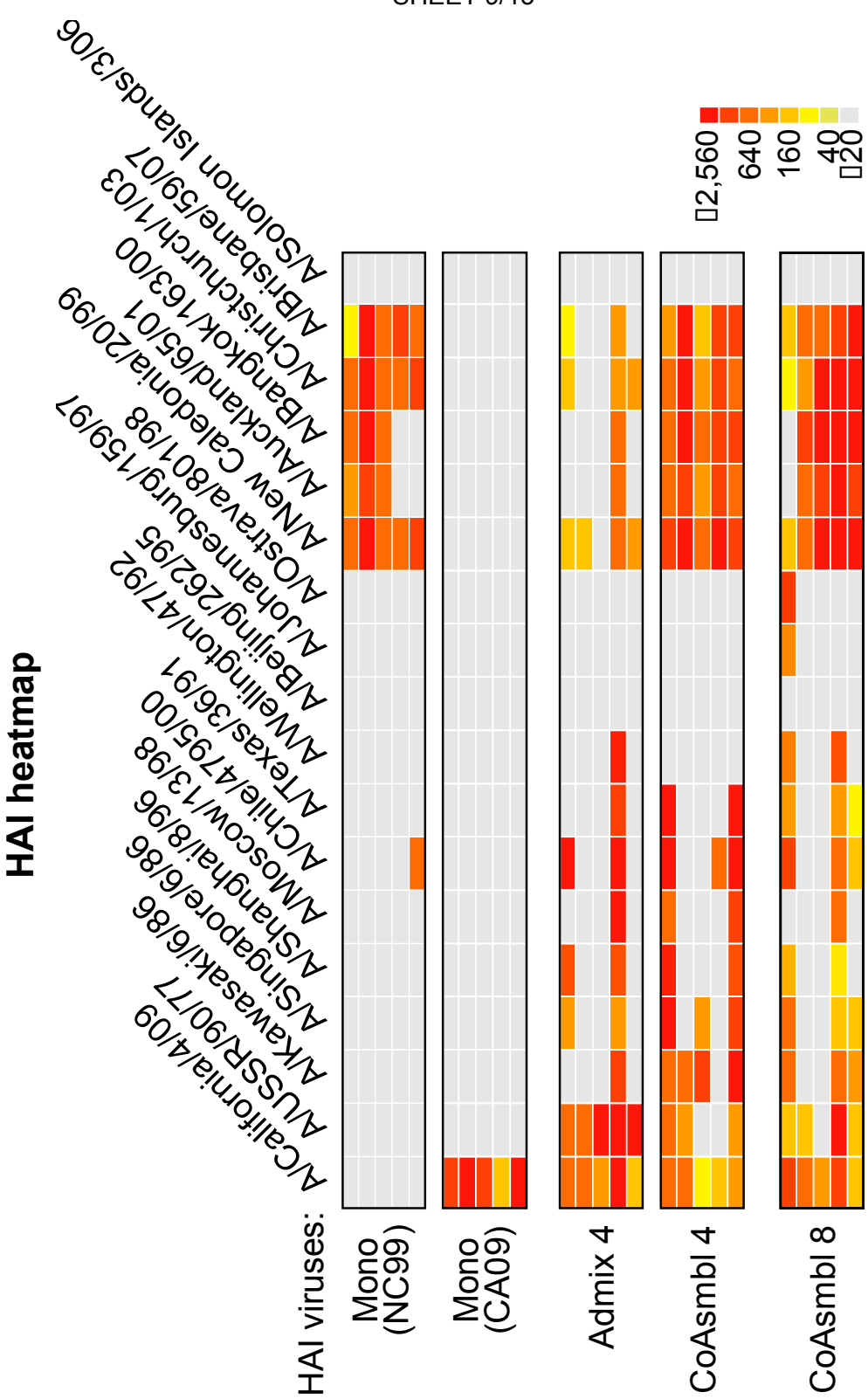

HAI heatmap
HAI viruses:
A/California/4/09
A/USSR/90/77
A/Kawasaki/6/86
A/Singapore/6/86
A/Shangahai/8/96
A/Moscow/13/98
A/Chile/4795/00
A/Texas/36/91
A/Wellington/47/92
A/Beijing/262/95
A/Johannesburg/159/97
A/Ostrava/801/98
A/New Caledonia/20/99
A/Auckland/65/01
A/Bangkok/163/00
A/Christchurch/1/03
A/Brisbane/59/07
A/Solomon Islands/3/06
Mono (NC99)
Mono (CA09)
Admix 4
CoAsmbl 4
CoAsmbl 8
2,560
640
160
40
20

Figure 9

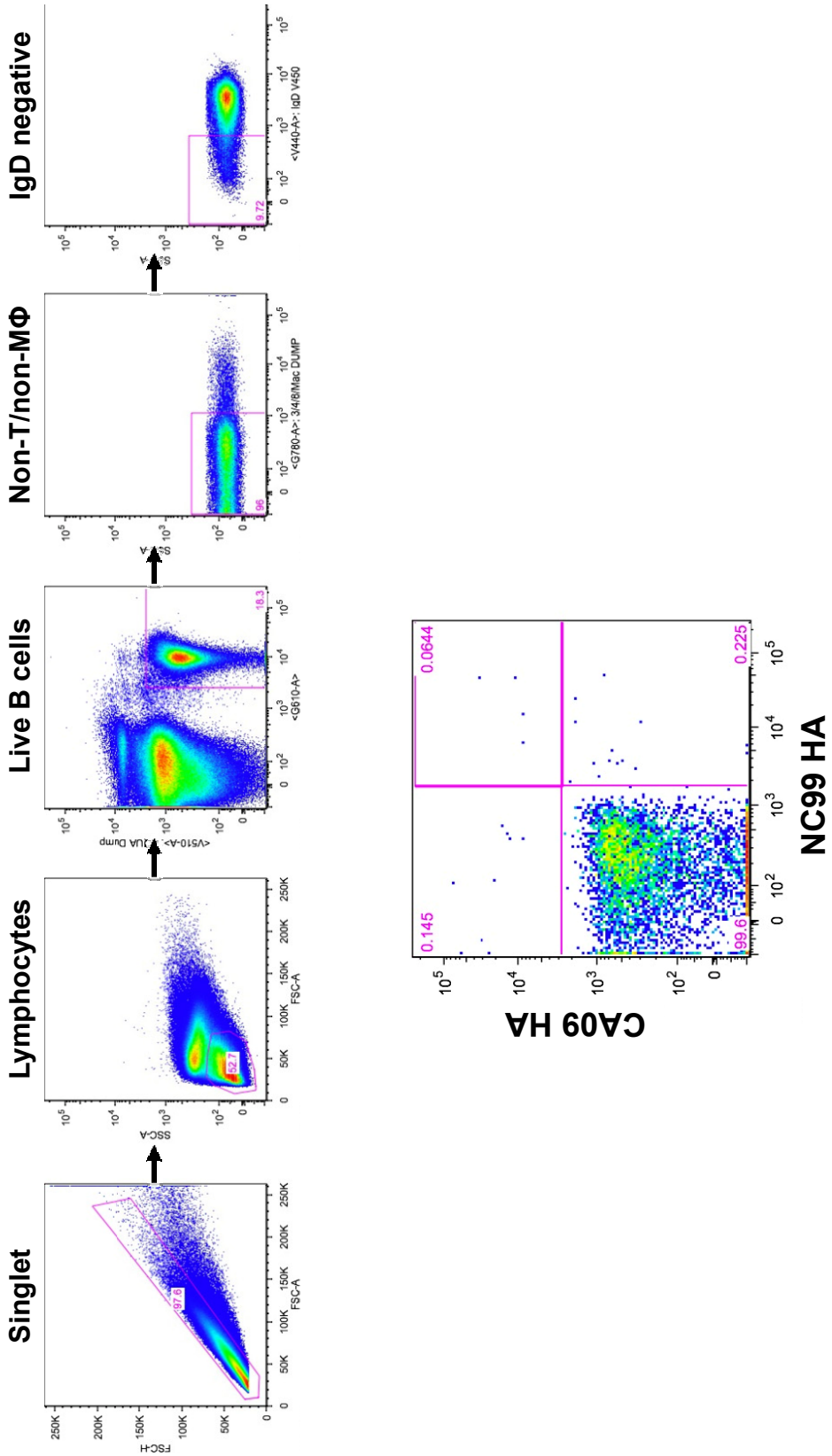

Singlet
Lymphocytes
Live B cells
Non-T/non-MΦ
IgD negative
FSC-H
FSC-A
SSC-A
<V510-A>: 2UA Dump
<G610-A>
<G780-A>: 3/4/8/Mac DUMP
<V440-A>: IgD V450
97.6
52.7
18.3
96
9.72
CA09 HA
NC99 HA
0.145
0.0644
99.6
0.225

Figure 10

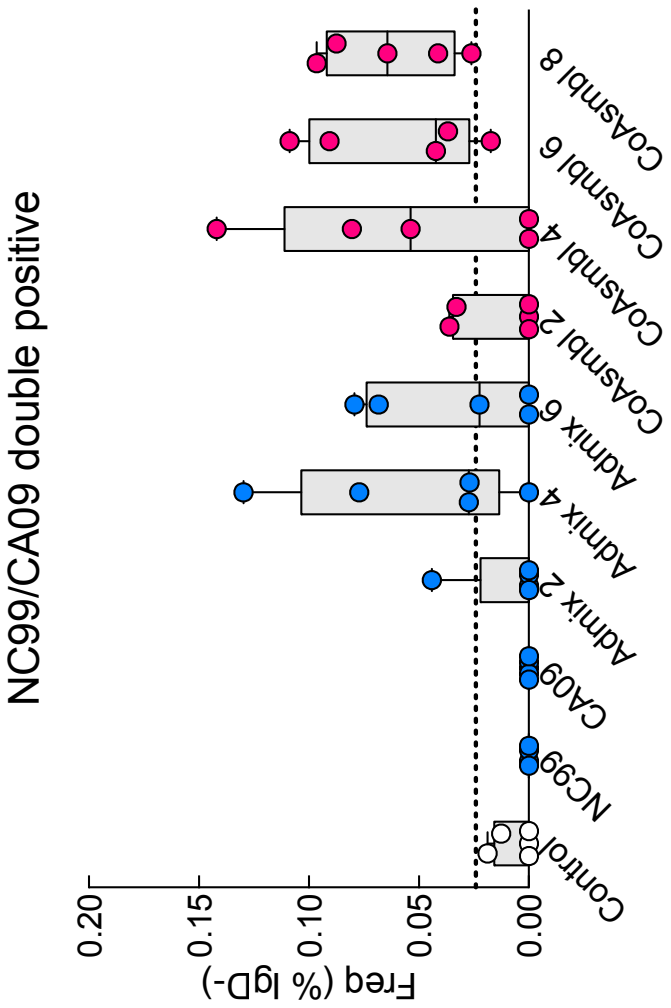

NC99/CA09 double positive
Freq (% IgD-)
0.20
0.15
0.10
0.05
0.00
Control
NC99
CA09
Admix 2
Admix 4
Admix 6
CoAsmbl 2
CoAsmbl 4
CoAsmbl 6
CoAsmbl 8

Figure 11

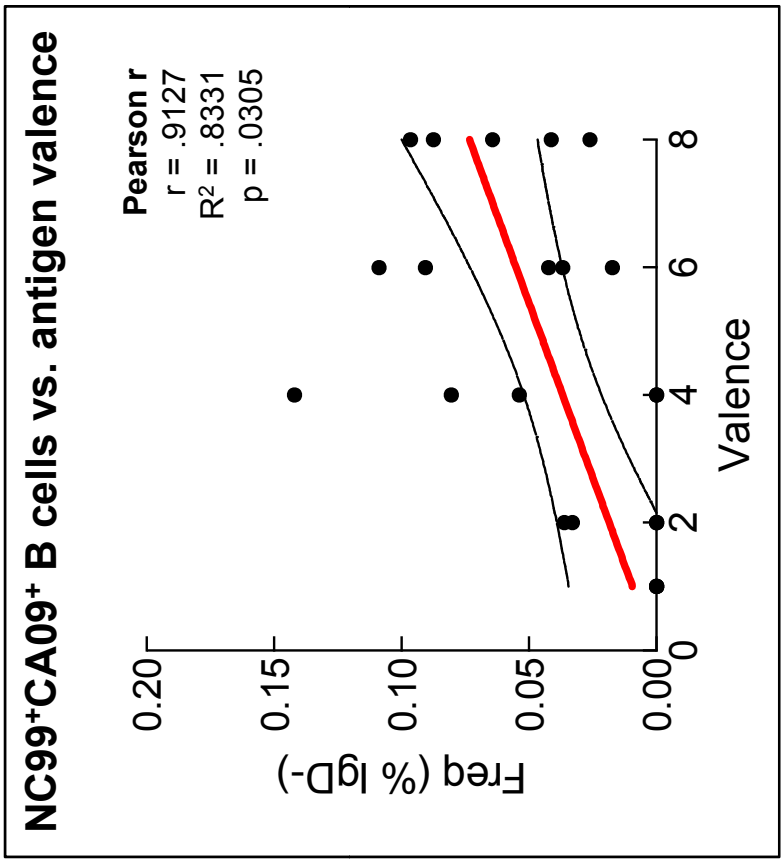

NC99+CA09+ B cells vs. antigen valence
Pearson r
r = .9127
R2 = .8331
p = .0305
Freq (% IgD-)
0.20
0.15
0.10
0.05
0.00
0
2
4
6
8
Valence

Figure 12

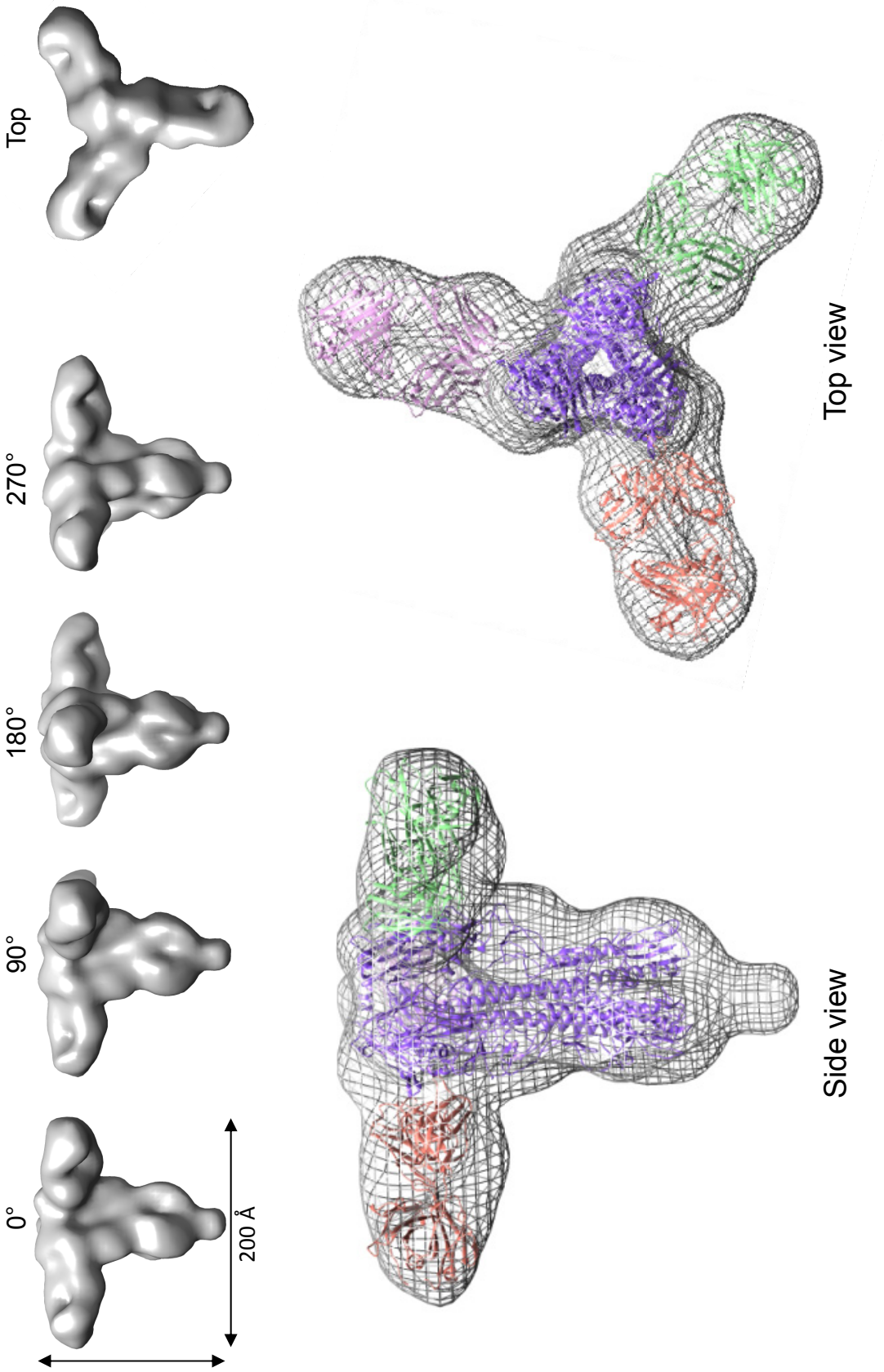


Figure 13