## **SUPPLEMENTARY FIGURE 1**

HA trimer.

All of the reference free 2D class averages contain the C-terminal GFP at the base of the









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4		

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14	4

## 1 SUPPLEMENTARY FIGURE 2.

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In previous work, a 3D structure of an α2-6 biantennary N-glycan with three lacNAc
repeats was produced in a bidentate co-complex with HA (Upper Panel). This was

1 modeled by first superimposing the terminal disaccharide onto a disaccharide in a crystal 2 structure co-complex. Then the glycosidic linkages were adjusted within their low energy 3 ranges and dihedral angles were selected that brought the second, unbound motif towards 4 a second binding site. The binding motif was pulled into position during an all-atom 5 molecular dynamics simulation and finally, the restraints were released to determine if the structure was stable. This method produced a bidentate co-complex for  $\alpha$ 2-6 biantennary 6 7 N-glycan with three, four or five lacNAc repeats. In contrast, this method did not find 8 bidentate N-glycans with α2-3 linkages. Although N-glycans containing α2-3 linkages and 9 five lacNAc repeats could span both sites (Lower panel), the  $\alpha$ 2-3 linkage places the motif 10 in the opposite orientation required by the binding site.

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