

## **Supplementary material**

### **Characterization of avian influenza virus attachment patterns to human and pig tissues**

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**Table S1.** Glycan structures in the glycan array with normalized attachment (%) of each virus. The attachment signals were normalized towards the glycan with the highest virus attachment for each tested virus. Only normalized values with a value >3 times larger than baseline are shown. HH3N2 – human H3N2, MH3N2 – mallard H3N2, MH6N1 – mallard H6N1, TH12N5 – ruddy turnstone H12N5, and GH16N3 – black-headed gull H16N3 influenza viruses.

|           | Glycan number | Glycan structure                            | Abbreviation <sup>1</sup> | HH3N2 | MH3N2 | MH6N1 | TH12N5 | GH16N3 |
|-----------|---------------|---|---------------------------|-------|-------|-------|--------|--------|
| No SA     | 1             | GalNAc <sub>asp</sub>                       | Tn                        |       |       |       |        |        |
|           | 2             | Galβ1-3GalNAc <sub>asp</sub>                | TF                        |       |       |       |        |        |
|           | 3             | Galβ1-4Glcβ <sub>sp</sub>                   | Lac                       |       |       |       |        |        |
|           | 4             | Galβ1-4GlcNAcβ <sub>sp</sub>                | LN                        |       |       |       |        |        |
|           | 5             | Galβ1-4GlcNAc-Bi-antennary N-glycan         | LN bi-antennary N-glycan  |       |       |       |        |        |
| 3 Su      | 6             | [3Su]Galβ1-4GlcNAcβ <sub>sp</sub>           | 3Su-LN                    |       |       |       |        |        |
|           | 7             | [3Su]Galβ1-3GalNAcβ <sub>sp</sub>           | 3Su-TF                    |       |       |       |        |        |
|           | 8             | [3Su]Galβ1-3GlcNAcβ <sub>sp</sub>           | 3Su-Le <sup>c</sup>       |       |       |       |        |        |
| 6 Su      | 9             | [6Su]Galβ1-4GlcNAcβ <sub>sp</sub>           | 6Su-LN                    |       |       |       |        |        |
|           | 10            | [6Su]Galβ1-3GlcNAcβ <sub>sp</sub>           | 6Su-Le <sup>c</sup>       |       |       |       |        |        |
|           | 11            | [6Su]Galβ1-4[6Su]GlcNAcβ <sub>sp</sub>      | di-6Su-LN                 |       |       |       |        |        |
|           | 12            | [6Su]Galβ1-3[6Su]GlcNAcβ <sub>sp</sub>      | di-6Su-Le <sup>c</sup>    |       |       |       |        |        |
|           | 13            | [6Su]Galβ1-3GalNAc <sub>asp</sub>           | 6Su-TF                    |       |       |       |        |        |
| Di/tri Su | 14            | [3Su]Galβ1-4[6Su]GlcNAcβ <sub>sp</sub>      | 3,6Su-LN                  |       |       |       |        |        |
|           | 15            | [3,6diSu]Galβ1-4GlcNAcβ <sub>sp</sub>       | 3,6diSu-LN                |       |       |       |        |        |
|           | 16            | [4,6diSu]Galβ1-4GlcNAcβ <sub>sp</sub>       | 4,6diSu-LN                |       |       |       |        |        |
|           | 17            | [4,6diSu]Galβ1-4GlcNAcβ <sub>sp</sub> 3     | 4,6diSu-LN                |       |       |       |        |        |
|           | 18            | [3,6diSu]Galβ1-4[6Su]GlcNAcβ <sub>sp</sub>  | 3,6triSu-LN               |       |       |       |        |        |
| SA        | 19            | Neu5Ac <sub>asp</sub>                       | Neu5Ac                    |       |       |       |        |        |
|           | 20            | Neu5Gc <sub>asp</sub>                       | Neu5Gc                    |       |       |       |        |        |
|           | 21            | [9OAc]-Neu5Ac <sub>asp</sub>                | 9OAc-Neu5Ac               |       |       |       |        |        |
| α2,3 SA   | 22            | Neu5Acα2-3Galβ <sub>sp</sub>                | “avian receptor”          |       | 90    | 31    | 76     |        |
|           | 23            | Neu5Acα2-3Galβ1,4Glcβ <sub>sp</sub>         | 3'SL                      |       | 99    | 77    | 75     |        |
|           | 24            | Neu5Acα2-3Galβ1,4GlcNAcβ <sub>sp</sub>      | 3'SLN                     |       | 61    | 86    | 73     |        |
|           | 25            | Neu5Acα2-3Galβ1,3GalNAc <sub>asp</sub>      | 3'STF                     |       | 77    | 65    | 83     |        |
|           | 26            | Neu5Acα2-3[6Su]Galβ1,4GlcNAcβ <sub>sp</sub> | 6Su-3'SLN                 |       | 75    | 6     | 48     |        |

|                 |    |   |                                  |     |     |     |    |     |
|-----------------|----|---|----------------------------------|-----|-----|-----|----|-----|
|                 | 27 | KDN $\alpha$ 2-3Gal $\beta$ 1-4GlcNAc $\beta$ sp                                | Kdn-LN                           |     |     |     |    |     |
|                 | 28 | Neu5Aca2-3Gal $\beta$ 1-4[6Su]GlcNAc $\beta$ sp                                 | 6Su-3'SLN                        |     | 97  | 28  | 90 | 100 |
|                 | 29 | GalNAc $\beta$ 1-4[Neu5Aca2-3]Gal $\beta$ 1-4Glc $\beta$ sp                     |                                  |     |     |     |    |     |
|                 | 30 | Neu5Aca2-8Neu5Aca2-3Gal $\beta$ 1-4Glc $\beta$ sp                               | 3'diSL                           |     |     |     |    |     |
|                 | 31 | GalNAc $\beta$ 1-4[Neu5Aca2-8Neu5Aca2-3]Gal $\beta$ 1-4Glc $\beta$ sp           |                                  |     |     |     |    |     |
|                 | 32 | Neu5Aca2-8Neu5Aca2-8Neu5Aca2-3Gal $\beta$ 1-4Glc $\beta$ sp                     | 3'triSL                          |     |     |     |    |     |
|                 | 33 | GalNAc $\beta$ 1-4[Neu5Aca2-8Neu5Aca2-8Neu5Aca2-3]Gal $\beta$ 1-4Glc $\beta$ sp |                                  |     |     |     |    |     |
|                 | 34 | Neu5Aca2-3Gal $\beta$ 1-4GlcNAc-Bi-antennary N-glycan                           | 3'SLN bi-antennary N-glycan      |     | 100 | 80  | 82 |     |
|                 | 35 | Neu5Aca2-3Gal $\beta$ 1-4GlcNAc-Tri(I)-antennary N-glycan                       | 3'SLN tri(I)-antennary N-glycan  |     | 100 | 100 | 98 |     |
|                 | 36 | Neu5Aca2-3Gal $\beta$ 1-4GlcNAc-Tri(II)-antennary N-glycan                      | 3'SLN tri(II)-antennary N-glycan |     | 100 | 89  | 97 |     |
|                 | 37 | Neu5Aca2-3Gal $\beta$ 1-4GlcNAc-Tetra-antennary N-glycan                        | 3'SLN tetra-antennary N-glycan   |     | 92  | 15  | 75 |     |
| $\alpha$ 2,6 SA | 38 | Neu5Aca2-6GalNAc $\alpha$ sp  | "human receptor"                 |     |     |     |    |     |
|                 | 39 | Neu5Aca2-6Gal $\beta$ sp  | "human receptor"                 |     |     |     |    |     |
|                 | 40 | Neu5Aca2-6Gal $\beta$ 1-4Glc $\beta$ sp   | 6'SL                             |     |     |     |    |     |
|                 | 41 | Neu5Aca2-6Gal $\beta$ 1-4GlcNAc $\beta$ sp                                      | 6'SLN                            | 31  |     |     |    |     |
|                 | 42 | Neu5Aca2-6Gal $\beta$ 1-4GlcNAc $\beta$ 1-3Gal $\beta$ 1-4GlcNAc $\beta$ sp     | 6'SdiLN                          | 100 | 5   |     |    |     |
|                 | 43 | Neu5Aca2-6Gal $\beta$ 1-3GalNAc $\alpha$ sp                                     | 6'STF                            |     |     |     |    |     |
|                 | 44 | Neu5Gca2-6Gal $\beta$ 1-4GlcNAc $\beta$ sp                                      | 6'Neu5Gc-LN                      |     |     |     |    |     |
|                 | 45 | [9OAc]Neu5Aca2-6Gal $\beta$ 1-4GlcNAc $\beta$ sp                                | 9OAc-6'SLN                       |     |     |     |    |     |
|                 | 46 | [6Su]Gal $\beta$ 1-3[Neu5Aca2-6]GalNAc $\alpha$ sp                              |                                  |     | 86  | 47  | 85 |     |
|                 | 47 | Neu5Aca2-6Gal $\beta$ 1-4[6Su]GlcNAc $\beta$ sp                                 | 6Su-6'SLN                        | 38  |     |     |    |     |
|                 | 48 | Gal $\beta$ 1-3[NeuAca2-6]GlcNAc $\beta$ 1-3Gal $\beta$ 1-4Glc $\beta$ sp       |                                  |     |     |     |    |     |
|                 | 49 | Neu5Aca2-6Gal $\beta$ 1-4GlcNAc-Bi-antennary N-glycan                           | 6'SLN bi-antennary N-glycan      |     |     |     |    |     |
|                 | 50 | Neu5Aca2-6Gal $\beta$ 1-4GlcNAc-Tri(I)-antennary N-glycan                       | 6'SLN tri(I)-antennary N-glycan  |     | 5   |     |    |     |

|          |    |   |                                  |  |  |    |     |  |
|----------|----|---|----------------------------------|--|--|----|-----|--|
|          | 51 | Neu5Ac $\alpha$ 2-6Gal $\beta$ 1-4GlcNAc-Tri(II)-antennary N-glycan | 6'SLN tri(II)-antennary N-glycan |  |  | 3  |     |  |
|          | 52 | Neu5Ac $\alpha$ 2-6Gal $\beta$ 1-4GlcNAc-Tetra-antennary N-glycan   | 6'SLN tetra-antennary N-glycan   |  |  |    |     |  |
| oligo SA | 53 | Neu5Ac $\alpha$ 2-3Gal $\beta$ 1-3[Neu5Ac $\alpha$ 2-6]GalNAcasp    | 3'S[Neu5Ac $\alpha$ 2-6]TF       |  |  | 84 | 100 |  |
|          | 54 | Neu5Ac $\alpha$ 2-8Neu5Acasp  | diNeu5Ac                         |  |  |    |     |  |
|          | 55 | Neu5Ac $\alpha$ 2-8Neu5Ac $\alpha$ 2-8Neu5Acasp                     | triNeu5Ac                        |  |  |    |     |  |

1. Abbreviations where applicable.

**Table S2.** Number of virus stained human individuals in full section vs. TMA format.

N/A – not applicable.

| <b>Tissue</b>      | <b>Human H3N2</b>   |            | <b>Mallard H3N2</b> |            | <b>Turnstone H12N5</b> |            |
|--------------------|---------------------|------------|---------------------|------------|------------------------|------------|
|                    | <b>Full section</b> | <b>TMA</b> | <b>Full section</b> | <b>TMA</b> | <b>Full section</b>    | <b>TMA</b> |
| <b>Eye</b>         | N/A                 | N/A        | 3                   | 1          | 3                      | N/A        |
| <b>Nasopharynx</b> | 4                   | N/A        | 4                   | 5          | 4                      | 5          |
| <b>Bronchus</b>    | 4                   | N/A        | 4                   | 1          | 4                      | 1          |
| <b>Alveoli</b>     | 4                   | N/A        | 4                   | 6          | 4                      | 6          |
| <b>Colon</b>       | N/A                 | N/A        | N/A                 | 7          | N/A                    | 7          |

**Table S3.** Number of lectin stained human individuals in full section vs. TMA

format. N/A – not applicable.

| <b>Tissue</b>      | <b>MAA-II</b>       |            | <b>SNA</b>          |            |
|--------------------|---------------------|------------|---------------------|------------|
|                    | <b>Full section</b> | <b>TMA</b> | <b>Full section</b> | <b>TMA</b> |
| <b>Eye</b>         | 1 <sup>1</sup>      | N/A        | 1 <sup>1</sup>      | N/A        |
| <b>Nasopharynx</b> | 2                   | 7          | 2                   | 6          |
| <b>Bronchus</b>    | 1                   | 1          | 1                   | 1          |
| <b>Alveoli</b>     | 1                   | 6          | 1                   | 6          |
| <b>Colon</b>       | N/A                 | 6          | N/A                 | 6          |

1. Repeated staining with the same individual performed.

**Table S4.** Number of viral stained pig individuals in full section vs. TMA format. N/A – not applicable.

| Tissue     | Human H3N2   |     | Mallard H3N2 |     | Mallard H6N1 |     | Turnstone H12N5 |     | Gull H16N3   |     |
|------------|--------------|-----|--------------|-----|--------------|-----|-----------------|-----|--------------|-----|
|            | Full section | TMA | Full section | TMA | Full section | TMA | Full section    | TMA | Full section | TMA |
| Trachea    | N/A          | 5   | N/A          | 5   | N/A          | 5   | N/A             | 5   | N/A          | 5   |
| Bronchiole | 2            | N/A | 4            | N/A | 4            | N/A | 2               | N/A | 4            | N/A |
| Alveoli    | 2            | N/A | 4            | N/A | 4            | N/A | 2               | N/A | 4            | N/A |

**Table S5.** Number of lectin stained pig individuals in full section vs. TMA format. N/A – not applicable.

| Tissue     | MAA-II         |     | SNA          |     |
|------------|----------------|-----|--------------|-----|
|            | Full section   | TMA | Full section | TMA |
| Trachea    | 1              | N/A | 1            | N/A |
| Bronchiole | 2 <sup>1</sup> | N/A | 2            | N/A |
| Alveoli    | 2 <sup>1</sup> | N/A | 2            | N/A |

1. Repeated staining with the same individuals performed.