

S4 Table. Identification of minority variants. Minority variant analysis of genetically closely related low pathogenic avian influenza (LPAI) viruses isolated from potential between-farm transmission cases. All nucleotide positions that varied between viruses at consensus level are shown. Minority variants consist of shared minority variants (i.e. minority variants that are present in both viruses) and minority variants at consensus-level variant sites (i.e. minority variants at sites that varied between viruses at consensus level). Minority variants were detected using a minimum frequency of 2.0% and a minimum coverage of 1,000 reads. All viruses were detected as part of the national avian influenza (AI) surveillance program in the Netherlands between January 2006 and September 2016. Detailed information on the virus sequences is provided in S1 Table.

| Potential case | Gene segment † | Nucleotide position | | | Virus 1 | | Virus 2 | | Virus 3 | |
|----------------|----------------|---------------------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|
| | | | Variant A | Variant B | Variant A | Variant B | Variant A | Variant B | Variant A | Variant B |
| H1N5-2007 | PB2 | 2049 | T | C | 100% | 0% | 15% | 85% | | |
| | PA | 885 | A | C | 67% | 33% | 0% | 100% | | |
| | HA | 523 | G | A | 61% | 39% | 0% | 100% | | |
| | NS | 521 | C | T | 65% | 35% | 0% | 100% | | |
| H10N7-2009 | PB2 | 2061 | A | G | 100% | 0% | 26% | 74% | | |
| | PB2 | 2065 | G | A | 100% | 0% | 0% | 100% | | |
| | PB1 | 472 | G | A | 100% | 0% | 0% | 100% | | |
| | PB1 | 1930 | G | A | 100% | 0% | 0% | 100% | | |
| | PB1 | 2142 | A | G | 100% | 0% | 0% | 100% | | |
| | PA | 2059 | A | C | 100% | 0% | 0% | 100% | | |
| | HA | 766 | C | T | 100% | 0% | 11% | 89% | | |
| | HA | 1017 | T | A | 97% | 3% | 97% | 3% | | |
| | NP | 337 | G | A | 100% | 0% | 0% | 100% | | |
| | NS | 30 | G | A | 100% | 0% | 0% | 100% | | |
| | NS | 284 | A | G | 100% | 0% | 0% | 100% | | |
| | H6N1-2010 | PB2 | 646 | C | A | 100% | 0% | 0% | 100% | |
| PB2 | | 2187 | G | A | 100% | 0% | 0% | 100% | | |
| PB1 | | 202 | T | C | 100% | 0% | 0% | 100% | | |
| PA | | 73 | A | G | 100% | 0% | 0% | 100% | | |
| PA | | 1053 | T | C | 100% | 0% | 0% | 100% | | |
| PA | | 1071 | A | T | 100% | 0% | 0% | 100% | | |
| PA | | 1133 | T | G | 100% | 0% | 0% | 100% | | |
| PA | | 1892 | G | T | 100% | 0% | 0% | 100% | | |
| NP | | 287 | C | T | 100% | 0% | 0% | 100% | | |
| NA | | 284 | C | T | 100% | 0% | 2% | 98% | | |
| NA | | 444 | A | C | 55% | 45% | 0% | 100% | | |
| NA | | 710 | G | A | 57% | 43% | 0% | 100% | | |
| NA | | 1297 | G | A | 100% | 0% | 0% | 100% | | |
| NS | | 214 | G | A | 100% | 0% | 4% | 96% | | |

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|------------|-----------|------|-----|------|------|------|------|------|------|------|
| H7N7-2011 | PB2 | 232 | A | C | 100% | 0% | 16% | 84% | | |
| | PB2 | 513 | C | T | 100% | 0% | 16% | 84% | | |
| | PB2 | 1084 | C | T | 100% | 0% | 16% | 84% | | |
| | PB2 | 1359 | C | T | 100% | 0% | 16% | 84% | | |
| | PB1 | 1011 | T | C | 100% | 0% | 15% | 85% | | |
| | PA | 497 | T | C | 73% | 27% | 10% | 90% | | |
| | PA | 1842 | G | A | 100% | 0% | 15% | 85% | | |
| | HA | 445 | G | A | 100% | 0% | 32% | 68% | | |
| | HA | 629 | A | G | 72% | 28% | 8% | 92% | | |
| | HA | 1238 | C | A | 100% | 0% | 35% | 65% | | |
| | NA | 631 | T | C | 100% | 0% | 12% | 88% | | |
| | NA | 926 | G | A | 100% | 0% | 12% | 88% | | |
| | H5N3-2013 | PB2 | 260 | G | A | 100% | 0% | 0% | 100% | |
| PB2 | | 702 | A | G | 100% | 0% | 0% | 100% | | |
| PB2 | | 741 | A | C | 100% | 0% | 0% | 100% | | |
| PB1 | | 213 | C | T | 100% | 0% | 0% | 100% | | |
| PB1 | | 456 | A | G | 100% | 0% | 0% | 100% | | |
| PB1 | | 578 | A | G | 100% | 0% | 0% | 100% | | |
| PB1 | | 1158 | C | T | 100% | 0% | 12% | 88% | | |
| PB1 | | 1611 | C | T | 100% | 0% | 0% | 100% | | |
| PB1 | | 1654 | A | C | 100% | 0% | 0% | 100% | | |
| PB1 | | 1869 | T | C | 100% | 0% | 0% | 100% | | |
| PA | | 294 | T | C | 100% | 0% | 0% | 100% | | |
| PA | | 1158 | G | T | 100% | 0% | 0% | 100% | | |
| HA | | 460 | T | - | 100% | 0% | 0% | 100% | | |
| HA | | 461 | A | - | 100% | 0% | 0% | 100% | | |
| HA | | 462 | T | - | 100% | 0% | 0% | 100% | | |
| HA | | 464 | A | T | 100% | 0% | 0% | 100% | | |
| HA | | 606 | A | G | 100% | 0% | 0% | 100% | | |
| HA | | 617 | A | T | 100% | 0% | 0% | 100% | | |
| HA | | 859 | G | A | 100% | 0% | 0% | 100% | | |
| HA | | 1077 | C | T | 100% | 0% | 0% | 100% | | |
| NP | | 249 | A | G | 100% | 0% | 0% | 100% | | |
| NP | 260 | A | G | 100% | 0% | 0% | 100% | | | |
| NA | 1143 | A | G | 100% | 0% | 0% | 100% | | | |
| MP | 932 | T | G | 95% | 5% | 0% | 100% | | | |
| H10N9-2012 | PB2 | 590 | A | C | 100% | 0% | 32% | 68% | 100% | 0% |
| | PB2 | 1048 | C | T | 100% | 0% | 32% | 68% | 100% | 0% |
| | PB2 | 1960 | G | A | 100% | 0% | 0% | 100% | 3% | 97% |
| | PB2 | 2300 | T | G | 100% | 0% | 0% | 100% | 0% | 100% |
| | PB1 | 318 | C | A | 100% | 0% | 100% | 0% | 4% | 96% |
| | PB1 | 1383 | G | A | 100% | 0% | 0% | 100% | 7% | 93% |

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|-----------|-----|------|---|---|------|-----|------|------|------|------|
| | PB1 | 1908 | C | T | 100% | 0% | 0% | 100% | 6% | 94% |
| | PB1 | 2218 | A | G | 100% | 0% | 0% | 100% | 0% | 100% |
| | PA | 392 | T | C | 100% | 0% | 0% | 100% | 3% | 97% |
| | PA | 666 | T | C | 100% | 0% | 32% | 68% | 100% | 0% |
| | PA | 1284 | A | G | 100% | 0% | 0% | 100% | 6% | 94% |
| | PA | 1355 | C | T | 100% | 0% | 100% | 0% | 6% | 94% |
| | PA | 1524 | C | T | 100% | 0% | 0% | 100% | 6% | 94% |
| | PA | 1864 | A | G | 100% | 0% | 0% | 100% | 3% | 97% |
| | HA | 49 | G | A | 100% | 0% | 100% | 0% | 0% | 100% |
| | HA | 544 | A | G | 100% | 0% | 30% | 70% | 100% | 0% |
| | HA | 566 | C | A | 100% | 0% | 0% | 100% | 0% | 100% |
| | HA | 769 | T | C | 100% | 0% | 0% | 100% | 0% | 100% |
| | HA | 1379 | T | C | 100% | 0% | 0% | 100% | 0% | 100% |
| | NP | 681 | G | A | 100% | 0% | 0% | 100% | 3% | 97% |
| | NP | 939 | T | C | 100% | 0% | 0% | 100% | 3% | 97% |
| | NP | 951 | G | A | 100% | 0% | 0% | 100% | 3% | 97% |
| | NP | 1090 | A | G | 100% | 0% | 0% | 100% | 0% | 100% |
| | NP | 1202 | C | G | 100% | 0% | 93% | 7% | 90% | 10% |
| | NA | 45 | T | C | 100% | 0% | 0% | 100% | 0% | 100% |
| | NA | 48 | T | C | 100% | 0% | 0% | 100% | 0% | 100% |
| | NA | 1054 | G | A | 100% | 0% | 100% | 0% | 0% | 100% |
| | MP | 421 | A | G | 100% | 0% | 0% | 100% | 0% | 100% |
| | NS | 209 | A | G | 100% | 0% | 0% | 100% | 0% | 100% |
| | NS | 338 | G | A | 100% | 0% | 0% | 100% | 0% | 100% |
| H6N2-2014 | PB2 | 1785 | A | C | 79% | 21% | 0% | 100% | 0% | 100% |
| | PB1 | 239 | G | A | 64% | 36% | 0% | 100% | 0% | 100% |
| | PB1 | 239 | G | A | 64% | 36% | 0% | 100% | 0% | 100% |
| | PB1 | 354 | T | C | 100% | 0% | 0% | 100% | 0% | 100% |
| | PB1 | 1833 | G | A | 78% | 22% | 0% | 100% | 0% | 100% |
| | PB1 | 1836 | G | A | 100% | 0% | 0% | 100% | 0% | 100% |
| | PA | 318 | A | G | 100% | 0% | 100% | 0% | 0% | 100% |
| | HA | 236 | A | G | 100% | 0% | 100% | 0% | 0% | 100% |
| | HA | 376 | G | T | 100% | 0% | 0% | 100% | 100% | 0% |
| | NP | 46 | A | G | 53% | 47% | 0% | 100% | 0% | 100% |
| | NA | 45 | T | C | 100% | 0% | 100% | 0% | 0% | 100% |
| | NA | 558 | A | G | 100% | 0% | 0% | 100% | 100% | 0% |
| | MP | 604 | A | G | 100% | 0% | 0% | 100% | 0% | 100% |
| | NS | 430 | A | G | 100% | 0% | 100% | 0% | 0% | 100% |

† PB2, polymerase basic protein 2; PB1, polymerase basic protein 1; PA, polymerase acidic protein; NP, nucleoprotein; HA, hemagglutinin; NP, nucleoprotein; NA, neuraminidase; MP, matrix protein; NS, nonstructural protein.