Table S1. Primers used for full-genome sequencing of A/South Africa/3626/2013 virus genome.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Gene** | **Fragment** | **Primers for RT-PCR** | **Sequence 5’→3’** | **Primers for reaction with Big Dye kit** | **Sequence 5’→3’** |
| PB2 | PB2-1 | PB2 Uni F1 | GATCGCTCTTCAGGGAGCGAAGCAGGTC [Hoffmann DOI: 10.1007/s007050170002] | PB2 Uni F1 | GATC GCTCTTCAGGGAGCGAAAGCAGGTC [Hoffmann DOI: 10.1007/s007050170002] |
| PB2 R1322 | TGCATGGGGTTCAGTCTCTG | PB2 F759 | CACTCCAGGAGGAGAAGTGAG |
|  |  | PB2 R1140 | TGTTGCTCTTCTCCCAACCATTGT |
| PB2-2 | PB2 F1063 | ACGGGCAATCTCCAAACA | PB2 F1063 | ACGGGCAATCTCCAAACA |
| PB2 Uni R | ACTG GCTCTTCTATT AGTAGAAACAAGG TCGTTT  [Hoffmann DOI: 10.1007/s007050170002] | PB2 R1477 | CCATCTTGCTGACTCTTA |
|  |  | PB2 Uni R | ACTG GCTCTTCTATT AGTAGAAACAAGG TCGTTT [Hoffmann DOI: 10.1007/s007050170002] |
| PB1 | PB1-1 | PB1 F1 | GAGCGAAAGCAGGCAAACCA | PB1 R431 | TTTCTGTTTAATGTCCAATCA |
| PB1 R1071 | TGC CAT TTT RTT TGA GAA CAT TAT [Matyushenko et al doi.org/10.1371/journal.pone.0180497] | PB1 F175 | GAAAAGGGGAAGTGGACAACAA |
|  |  | PB1 R835 | GCCCAGACTGTTCAAG |
| PB1-2 | PB1 F594 | AGT AAG RGA CAA CAT GAC CAA GAA [[Matyushenko et al doi.org/10.1371/journal.pone.0180497] | PB1 F594 | AGT AAG RGA CAA CAT GAC CAA GAA [[Matyushenko et al doi.org/10.1371/journal.pone.0180497] |
| PB1 Uni R | ACTG GCTCTTCTATT AGTAGAAACAAGG CATTTT [Hoffmann DOI: 10.1007/s007050170002] | PB1 R1536 | ATTAGCCACAAATCCATAGCG |
|  |  | PB1 R1071 | TGC CAT TTT RTT TGA GAA CAT TAT [Matyushenko et al doi.org/10.1371/journal.pone.0180497] |
|  |  | PB1 Uni R | ACTG GCTCTTCTATT AGTAGAAACAAGG CATTTT [Hoffmann DOI: 10.1007/s007050170002] |
|  |  | PB1 R2273 | ATGGTGGAACAGATCTTCATGATCTC [Deng 2011 DOI: 10.1371/journal.pone.0023400] |
| PA | PA-1 | PA Uni F1 | GATCGCTCTTCAGGGAGCGAAAGCAGGTAC [Hoffmann DOI: 10.1007/s007050170002] | PA F62 | TCGTCGAGCTTGCGGAAAA |
| PA R1709 | AACATGGGCCTCGACACTTG | PA F593 | CCTTTCGTCAGTCCGAAAGAGG  [Deng 2011 DOI: 10.1371/journal.pone.0023400] |
|  |  | PA R619 | CTT CGC CTC TTT CGG ACT GAC G [Matyushenko et al doi.org/10.1371/journal.pone.0180497] |
|  |  | PA R1709 | AACATGGGCCTCGACACTTG |
| PA-2 | PA F1623 | GCCACACAAATGGGAAAAATAC | PA F1270 | GAATTGACTGATTCAAGCTGG |
| PA Uni R | ACTG GCTCTTCTATTAGTAGAAACAAGGTACTT [Hoffmann DOI: 10.1007/s007050170002] | PA F1623 | GCCACACAAATGGGAAAAATAC |
|  |  | PA F2002 | GCTGAATCAAGAAAATTGC |
|  |  | PA Uni R | ACTG GCTCTTCTATTAGTAGAAACAAGGTACTT  [Hoffmann DOI: 10.1007/s007050170002] |
| HA | HA-1 | HA Uni F1 | GATCGCTCTTCAGGGAGCAAAAGCAGGGG [Hoffmann DOI: 10.1007/s007050170002] | HA Uni F1 | GATCGCTCTTCAGGGAGCAAAAGCAGGGG [Hoffmann DOI: 10.1007/s007050170002] |
| HA R903 | CGTGGACTGGTGTATCTGAAATGA | HA F347 | AACGTGTTACCCAGGAGATTTCA |
|  |  | HA R438 | TGGGGAATATCTCAAACCTT |
|  |  | HA R903 | CGTGGACTGGTGTATCTGAAATGA |
|  | HA F776 | GACACTAGTAGAGCCGGGAGACAA | HA Uni R | ACTGGCTCTTCTATT AGTAGAAACAAGG GTGTTTT [Hoffmann DOI: 10.1007/s007050170002] |
| HA Uni R | ACTGGCTCTTCTATT AGTAGAAACAAGG GTGTTTT  [Hoffmann DOI: 10.1007/s007050170002] | HA F776 | GACACTAGTAGAGCCGGGAGACAA |
|  |  | HA F1312 | AAGTTGATGATGGATTCCTG |
|  |  | HA R1462 | ATTTCCTTGGCATTGTTTT |
| NP | NP-1 | NP F1 | GGAGCAAAAGCAGGGTAGATAATC | NP F1 | GGAGCAAAAGCAGGGTAGATAATC |
| NP R1195 | TGCTTCTTAGTTCCAGGGTAT | NP R789 | TGGGTTTCGACTTTCTCTTACTTG |
|  |  | NP R1195 | TGCTTCTTAGTTCCAGGGTAT |
| NP-2 | NP F1020 | GAGTCAATTGGTATGGATGGC | NP F1091 | GAAAGAAAGTGATCCCAAGAG |
| NP Uni R | ACTGGCTCTTCTATT AGTAGAAACAAGG GTATTTT [Hoffmann DOI: 10.1007/s007050170002] | NP Uni R | ACTGGCTCTTCTATTAGTAGAAACAAGGGTATTTT [Hoffmann DOI: 10.1007/s007050170002] |
| NA | NA-1 | NA Uni F1 | GATCGCTCTTCAGGG AGCAAAAGCAGG AGT [Hoffmann DOI: 10.1007/s007050170002] | NA F290 | TCTCTGCCCTGTTAGTGGATG |
| NA R880 | TCACTAGAATCAGGATAACAGGA | NA R377 | TTCCCTTATGACAAACACA |
|  |  | NA R880 | TCACTAGAATCAGGATAACAGGA |
| NA-2 | NA F517 | TTCCCTCTCCATACAACTCAA | NA F517 | TTCCCTCTCCATACAACTCAA |
| NA Uni R | ACTG GCTCTTCTATT AGTAGAAACAAGGAGTTTTTT [Hoffmann DOI: 10.1007/s007050170002] | NA F1020 | AGTTGTGGTCCAGTATCGTCTAAT |
|  |  | NA R1219 | GACCACTCATTTATCCCTACGA |
| M |  | M Uni F1 | GAT CGC TCT TCA GGG AGC AAA AGC AGG TAG [Hoffmann DOI: 10.1007/s007050170002] | M F260 | TTTATCCAAAATGCCCTAAATG |
|  | M Uni R | ACTGGCTCTTCTATTAGTAG AAACAAGGTAGTTTTT [Hoffmann DOI: 10.1007/s007050170002] | M R702 | CTGGAGCTAGGATGAGTCCCA |
| NS |  | NS Uni F1 | GATCGCTCTTCAGGGAGCAAAAGCAGGGTG [Hoffmann DOI: 10.1007/s007050170002] | NS R559 | ATTGCATTTTTGACATCCT  [Matyushenko et al doi.org/10.1371/journal.pone.0180497] |
|  | NS Uni R | ACTGGCTCTTCTATTAGTAGAAACAAGGGTGTTTTT [Hoffmann DOI: 10.1007/s007050170002] | NS F108 | GGTGATGCCCCATTCCTTG |

Table S2: Two-way hemagglutination inhibition test

|  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Against SA-M5 | | | | | | | | | | | | |
| Sera SA-M5 | m1 | m2 | m3 | m4 | m5 | m6 | m7 | m8 | m9 | m10 | Average | SD |
| -2 | 1280 | 320 | 640 | 640 | 160 |  |  |  |  |  | 608 | 429.3251 |
| -3 | 640 | 640 | 160 | 160 | 160 | 320 | 320 | 640 | 160 |  | 355.5556 | 223.1093 |
| -4 | 160 | 40 | 320 | 80 | 320 | 320 | 80 | 160 | 40 | 640 | 216 | 187.806 |
| Sera SA-WT | m1 | m2 | m3 | m4 | m5 | m6 | m7 | m8 | m9 | m10 |  |  |
| -2 | 160 | 320 | 160 | 160 | 80 | 640 | 80 | 160 | 160 |  | 213.3333 | 174.356 |
| -3 | 40 | 160 | 320 | 80 | 80 | 160 | 80 | 80 | 320 | 320 | 164 | 113.842 |
| -4 | 10 | 5 | 20 | 10 | 80 |  |  |  |  |  | 25 | 31.22499 |
| Against SA-WT | | | | | | | | | | | | |
| Sera SA-M5 | m1 | m2 | m3 | m4 | m5 | m6 | m7 | m8 | m9 | m10 | Average | SD |
| -2 | 1280 | 320 | 640 | 640 | 160 |  |  |  |  |  | 608 | 429.3251 |
| -3 | 640 | 640 | 160 | 160 | 160 | 320 | 320 | 640 | 160 |  | 355.5556 | 223.1093 |
| -4 | 160 | 40 | 320 | 80 | 320 | 320 | 80 | 160 | 40 | 640 | 216 | 187.806 |
| Sera SA-WT | m1 | m2 | m3 | m4 | m5 | m6 | m7 | m8 | m9 | m10 |  |  |
| -2 | 160 | 320 | 160 | 160 | 80 | 640 | 80 | 160 | 160 |  | 213.3333 | 174.356 |
| -3 | 40 | 160 | 320 | 80 | 80 | 160 | 80 | 80 | 320 | 320 | 164 | 113.842 |
| -4 | 10 | 5 | 20 | 10 | 80 |  |  |  |  |  | 25 | 31.22499 |