



|            | 121  | 131 | 141 | 151 | 161 | 171 | 180 |
|------------|--|-----|-----|-----|-----|-----|-----|
| OKSK090075 | ALTGGLELTRDPTTPEHASLVGLLLSLYRLSKISGQNTGNYKTNIADRIEQIFETAPFVK |     |     |     |     |     |     |
| OKSK090076 | .....IS.....   |     |     |     |     |     |     |
| MOSK090043 | .....IS.....   |     |     |     |     |     |     |
| ARSK090058 | .....IS.....   |     |     |     |     |     |     |
| KSSK090055 | .....I.....  |     |     |     |     |     |     |
| NEFE090019 | .....IQ.....   |     |     |     |     |     |     |
| V212TXSK   | .....D.....I.....  |     |     |     |     |     |     |
| 5422SCSK   | .....IS.....L.....S.....M.....                               |     |     |     |     |     |     |
| ARDG090042 | .....M.....IS.....   |     |     |     |     |     |     |
| MNSK090088 | .....V.....VS.....   |     |     |     |     |     |     |
| L000160WSK | .....V.....VS.....   |     |     |     |     |     |     |
| 3789SK     | .....M.P.....VS.....   |     |     |     |     |     |     |
| V657CASK   | .....M.M.....VS.....   |     |     |     |     |     |     |
| V644CASK   | .....M.....VS.....   |     |     |     |     |     |     |
| V648CASK   | .....M.....VS.....   |     |     |     |     |     |     |
| V684SBC    | .....M.....VS.....   |     |     |     |     |     |     |
| V854SLP    | .....S.....  |     |     |     |     |     |     |
| FLdg1994   | .....M.....IS.....   |     |     |     |     |     |     |
| TX692007CY | .....M.....VS.....   |     |     |     |     |     |     |
| FLRC090148 | .....M.....I.....S.....                                      |     |     |     |     |     |     |
| 79L1020LB  | .....M.....V.....  |     |     |     |     |     |     |
| 4995AZSK   | .....M.....VS.....   |     |     |     |     |     |     |
| 1563CL     | .....M.....IS.....   |     |     |     |     |     |     |
| 906BB      | .....M.....VS.....   |     |     |     |     |     |     |

|            | 181   | 190 | 200 | 210 | 220 | 230 | 240 |
|------------|---|-----|-----|-----|-----|-----|-----|
| OKSK090075 | IVEHHTLMTTHKMCANWSTIPNFRFLAGTYDMFFSRIEHLYSAIRVGTVVVTAYEDCSGLV |     |     |     |     |     |     |
| OKSK090076 | .....   |     |     |     |     |     |     |
| MOSK090043 | .I.....   |     |     |     |     |     |     |
| ARSK090058 | .I.....   |     |     |     |     |     |     |
| KSSK090055 | .I.....A.....   |     |     |     |     |     |     |
| NEFE090019 | .I.....A.....   |     |     |     |     |     |     |
| V212TXSK   | .I.....   |     |     |     |     |     |     |
| 5422SCSK   | .I.....   |     |     |     |     |     |     |
| ARDG090042 | .....   |     |     |     |     |     |     |
| MNSK090088 | .....   |     |     |     |     |     |     |
| L000160WSK | .....   |     |     |     |     |     |     |
| 3789SK     | V.....  |     |     |     |     |     |     |
| V657CASK   | .....   |     |     |     |     |     |     |
| V644CASK   | .....   |     |     |     |     |     |     |
| V648CASK   | .....S.....   |     |     |     |     |     |     |
| V684SBC    | .....   |     |     |     |     |     |     |
| V854SLP    | V.....  |     |     |     |     |     |     |
| FLdg1994   | .....N.....   |     |     |     |     |     |     |
| TX692007CY | .....   |     |     |     |     |     |     |
| FLRC090148 | .....   |     |     |     |     |     |     |
| 79L1020LB  | .....   |     |     |     |     |     |     |
| 4995AZSK   | .....   |     |     |     |     |     |     |
| 1563CL     | .....   |     |     |     |     |     |     |
| 906BB      | .....A.....   |     |     |     |     |     |     |

|            |     |     |     |     |     |     |
|------------|-----|-----|-----|-----|-----|-----|
|            | 241 | 250 | 260 | 270 | 280 | 290 |
| 300        |     |     |     |     |     |     |
| OKSK090075 | S   | F   | T   | G   | F   | I   |
| OKSK090076 | .   | .   | .   | .   | .   | .   |
| MOSK090043 | .   | .   | .   | .   | .   | .   |
| ARSK090058 | .   | .   | .   | .   | .   | .   |
| KSSK090055 | .   | .   | .   | .   | .   | .   |
| NEFE090019 | .   | .   | .   | .   | .   | .   |
| V212TXSK   | .   | .   | .   | .   | .   | .   |
| 5422SCSK   | .   | .   | .   | .   | .   | .   |
| ARDG090042 | .   | .   | .   | .   | .   | .   |
| MNSK090088 | .   | .   | .   | .   | .   | .   |
| L000160WSK | .   | .   | .   | .   | .   | .   |
| 3789SK     | .   | .   | .   | .   | .   | .   |
| V657CASK   | .   | .   | .   | .   | .   | .   |
| V644CASK   | .   | .   | .   | .   | .   | .   |
| V648CASK   | .   | .   | .   | .   | .   | .   |
| V684SBC    | .   | .   | .   | .   | .   | .   |
| V854SLP    | .   | .   | .   | .   | .   | .   |
| FLdg1994   | .   | .   | .   | .   | .   | .   |
| TX692007CY | .   | .   | .   | .   | .   | .   |
| FLRC090148 | .   | .   | .   | .   | .   | .   |
| 79L1020LB  | .   | .   | .   | .   | .   | .   |
| 4995AZSK   | .   | .   | .   | .   | .   | .   |
| 1563CL     | .   | .   | .   | .   | .   | .   |
| 906BB      | .   | .   | .   | .   | .   | .   |

|            |     |     |     |     |     |     |     |
|------------|-----|-----|-----|-----|-----|-----|-----|
|            | 301 | 310 | 320 | 330 | 340 | 350 | 360 |
| OKSK090075 | S   | S   | N   | A   | V   | G   | H   |
| OKSK090076 | .   | .   | .   | .   | .   | .   | .   |
| MOSK090043 | .   | .   | .   | .   | .   | .   | .   |
| ARSK090058 | .   | .   | .   | .   | .   | .   | .   |
| KSSK090055 | .   | .   | .   | .   | .   | .   | .   |
| NEFE090019 | .   | .   | .   | .   | .   | .   | .   |
| V212TXSK   | .   | .   | .   | .   | .   | .   | .   |
| 5422SCSK   | .   | .   | .   | .   | .   | .   | .   |
| ARDG090042 | .   | .   | .   | .   | .   | .   | .   |
| MNSK090088 | .   | .   | .   | .   | .   | .   | .   |
| L000160WSK | .   | .   | .   | .   | .   | .   | .   |
| 3789SK     | .   | .   | .   | .   | .   | .   | .   |
| V657CASK   | .   | .   | .   | .   | .   | .   | .   |
| V644CASK   | .   | .   | .   | .   | .   | .   | .   |
| V648CASK   | .   | .   | .   | .   | .   | .   | .   |
| V684SBC    | .   | .   | .   | .   | .   | .   | .   |
| V854SLP    | .   | .   | .   | .   | .   | .   | .   |
| FLdg1994   | .   | .   | .   | .   | .   | .   | .   |
| TX692007CY | .   | .   | .   | .   | .   | .   | .   |
| FLRC090148 | .   | .   | .   | .   | .   | .   | .   |
| 79L1020LB  | .   | .   | .   | .   | .   | .   | .   |
| 4995AZSK   | .   | .   | .   | .   | .   | .   | .   |
| 1563CL     | .   | .   | .   | .   | .   | .   | .   |
| 906BB      | .   | .   | .   | .   | .   | .   | .   |

|            | 361 | 370 | 380 | 390 | 400 | 410 | 420 |
|------------|-----|-----|-----|-----|-----|-----|-----|
| OKSK090075 | R   | D   | E   | K   | E   | L   | Q   |
| OKSK090076 | .   | .   | .   | .   | .   | .   | .   |
| MOSK090043 | .   | .   | .   | .   | .   | .   | .   |
| ARSK090058 | .   | .   | .   | .   | .   | .   | .   |
| KSSK090055 | .   | .   | .   | .   | .   | .   | .   |
| NEFE090019 | .   | .   | .   | .   | .   | .   | .   |
| V212TXSK   | .   | H   | .   | .   | .   | .   | .   |
| 5422SCSK   | .   | .   | .   | .   | .   | .   | .   |
| ARDG090042 | .   | .   | D   | .   | .   | I   | .   |
| MNSK090088 | .   | .   | D   | .   | .   | .   | .   |
| L000160WSK | .   | .   | D   | .   | .   | .   | .   |
| 3789SK     | .   | .   | D   | .   | .   | .   | .   |
| V657CASK   | .   | .   | ID  | .   | .   | I   | .   |
| V644CASK   | .   | H   | D   | .   | .   | .   | .   |
| V648CASK   | .   | R   | H   | ID  | .   | .   | .   |
| V684SBC    | .   | .   | AD  | .   | .   | A   | .   |
| V854SLP    | .   | .   | A   | D   | .   | .   | .   |
| FLdg1994   | .   | .   | D   | .   | .   | .   | .   |
| TX692007CY | .   | .   | D   | .   | .   | .   | .   |
| FLRC090148 | .   | D   | .   | .   | .   | I   | .   |
| 79L1020LB  | .   | .   | .   | .   | S   | .   | .   |
| 4995AZSK   | .   | S   | .   | .   | S   | .   | .   |
| 1563CL     | .   | .   | .   | .   | .   | .   | .   |
| 906BB      | .   | S   | D   | .   | .   | I   | .   |

|            | 421                            | 430 | 440 | 450                  |
|------------|--------------------------------|-----|-----|----------------------|
| OKSK090075 | IRRYVSVSSNHQARPNSFAEFLNKTYSNDS |     |     | (SCSK I)             |
| OKSK090076 | .                              | .   | .   | (SCSK I)             |
| MOSK090043 | .                              | .   | .   | (SCSK IIA)           |
| ARSK090058 | G                              | .   | .   | (SCSK IIB)           |
| KSSK090055 | .                              | .   | .   | (SCSK IIIA)          |
| NEFE090019 | .                              | .   | .   | (SCSK IIIB)          |
| V212TXSK   | .                              | .   | .   | (SCSK)               |
| 5422SCSK   | .                              | .   | .   | (SCSK)               |
| ARDG090042 | .                              | .   | S   | (NCSK)               |
| MNSK090088 | .                              | T   | S   | (NCSK)               |
| L000160WSK | .                              | T   | S   | (NCSK)               |
| 3789SK     | .                              | .   | S   | (NCSK)               |
| V657CASK   | .                              | .   | S   | (CASK a)             |
| V644CASK   | .                              | .   | S   | (CASK b)             |
| V648CASK   | .                              | .   | S   | (CASK c)             |
| V684SBC    | Q                              | .   | S   | (SBC SK)             |
| V854SLP    | .                              | .   | S   | (CENTRAL MEXICAN SK) |
| FLdg1994   | T                              | .   | S   | (TX COYOTE)          |
| TX692007CY | .                              | .   | S   | (TX GRAY FOX)        |
| FLRC090148 | .                              | .   | P   | (FL RACCOON)         |
| 79L1020LB  | .                              | .   | S   | (BAT VARIANT)        |
| 4995AZSK   | .                              | .   | S   | (AZ SK VARIANT)      |
| 1563CL     | .                              | .   | S   | (BAT VARIANT)        |
| 906BB      | .                              | .   | S   | (BAT VARIANT)        |

Figure S1. An alignment of nucleoprotein sequences of representative skunk-associated rabies viruses examined in this study with comparison to the sequences of other viral variants that circulate in sympatric hosts of the US mid-west. A member of the SCSK type I group is used as the reference isolate. The (!) symbol identifies residues having notable substitutions in some variants.

|            | -19  | -10 | 1                     | 10 | 20                             | 30                   | 40                 |
|------------|--|-----|-----------------------|----|--------------------------------|----------------------|--------------------|
| OKSK090075 | MVLQALLFAPFLISSLCLGKFPITYTIPDKLGPWSPIDIHHLSCPNNLVIEDEGCNSLSG |     |                       |    |                                |                      |                    |
| OKSK090076 | .....  |     |                       |    |                                |                      | V.....             |
| MOSK090043 | . . . K . . . . . M . . . . .                                |     | .....                 |    |                                |                      | V.....             |
| ARSK090058 | .....  |     |                       |    |                                |                      | V.....             |
| KSSK090051 | . . . . . F . . . . .  |     | .....                 |    |                                |                      | V.....             |
| NEFE090019 | . . . . . M . . . . .  |     |                       |    | .....                          |                      | V . . . . N . . .  |
| KSSK090012 | .....  |     |                       |    |                                |                      | V.....             |
| ARDG090042 | . . P . . . . VV . L . GF . . . F . . . . .                  |     | .....                 |    |                                | V . . . . TN . . .   |                    |
| MNSK090088 | . . P . . . . VV . L . GF . . . F . . . . .                  |     | .....                 |    |                                | V . . . . TN . . .   |                    |
| L920867WSK | . . P . . . . VV . L . GF . . . F . . . . .                  |     | .....                 |    |                                | V . . . . TN . . .   |                    |
| CA982      | . FP . . . . . VSL . . . . . F . . . . .                     |     | .....                 |    |                                | V . . . . TN . . .   |                    |
| CASK2      | . . S . . . . . VSP . . L . . S . E . . . . .                |     | .....                 |    |                                | V . . . . TN . . .   |                    |
| MEXSK3636  | . . P . . . . . V . L . VF . . . F . . . . .                 |     | .....                 |    |                                | V . . . . TN . . .   |                    |
| MEXSK3644  | . . P . . . . . V . L . . PPV . . . . .                      |     | .....                 |    |                                | V . . . . SN . . .   |                    |
| A10-3020   | . . P . V . . . . V . L . VF . . . F . . . . .               |     | .....                 |    |                                | V . . . . TN . . .   |                    |
| Fox-8007   | . . P . V . . . . V . L . VF . . . F . . . . .               |     | . . . . . L . . . . . |    | .....                          |                      | V . . . . TN . . . |
| A10-0515   | . . P . . . . . V . L . VFPS . . . . .                       |     | .....                 |    |                                | V . . . . N . . .    |                    |
| A10-0511   | . . . . . V . L . VFPS . F . . . . .                         |     | .....                 |    |                                | V . . . . N . . .    |                    |
| A10-3932   | . . P . . . . . V . I . VF . . . F . . . . .                 |     | .....                 |    |                                | V . . . . TN . . .   |                    |
| FLRC090148 | . . PW . . . . . V . . . . . F . . . . .                     |     | .....                 |    |                                | V . . . . .          |                    |
| SM5596     | . IP . . . . . YV . L . . . . . F . G . . . . .              |     | .....                 |    |                                | V . . . . T . . . .  |                    |
| SM4871     | . IP . . . . . YV . L . . P . . . . F . G . . . . .          |     | .....                 |    |                                | V . . . . T . . . .  |                    |
| SM3844     | . IP . . . . . YV . L . . . . . F . G . . . . .              |     | .....                 |    |                                | V . . . . T . . . .  |                    |
| SHBRV      | . IP . . . . Q . V . L . . P . . . . F . . . . .             |     | .....                 |    |                                | A . . . . T . . . .  |                    |
| TN310      | . IP . . . . . VSL . . P . . . . . E . . . . .               |     | . . . . . T . . . . . |    | . . . . . A . ND . . T . . . . |                      |                    |
| TN209      | . IP . . . . . V . L . . P . . . . . E . . . . .             |     | .....                 |    |                                | A . ND . . T . . . . |                    |

|            | 41   | 50 | 60                    | 70                            | 80                  | 90                  | 100       |
|------------|--|----|-----------------------|-------------------------------|---------------------|---------------------|-----------|
| OKSK090075 | FSYMELKVGYISAIKVNNGFTCTGVVTEAETYTNFVGYVTTTTFKRKHFPRMPDACRSAYNW |    |                       |                               |                     |                     |           |
| OKSK090076 | .....  |    |                       |                               |                     |                     |           |
| MOSK090043 | .....  |    |                       |                               |                     |                     |           |
| ARSK090058 | .....  |    |                       |                               |                     |                     |           |
| KSSK090051 | .....  |    |                       |                               |                     |                     |           |
| NEFE090019 | .....  |    |                       |                               |                     |                     |           |
| KSSK090012 | .....  |    |                       |                               |                     |                     |           |
| ARDG090042 | .....  |    |                       |                               |                     | T . . . . A . . . . |           |
| MNSK090088 | .....  |    |                       |                               |                     | T . E . . A . . . . |           |
| L920867WSK | .....  |    |                       |                               |                     | T . E . . A . . . . |           |
| CA982      | .....  |    |                       | I . . . . V . . . . S . . . . | T . . . . A . . . . |                     |           |
| CASK2      | .....  |    |                       |                               |                     | T . . . . A . . . . |           |
| MEXSK3636  | .....  |    |                       |                               |                     | I . . . . A . . . . |           |
| MEXSK3644  | . . . . . D . . . . .  |    | . . . . . I . . . . . |                               | .....               |                     |           |
| A10-3020   | .....  |    |                       |                               |                     | T . . . . A . . . . |           |
| Fox-8007   | . . . . . S . . . . .  |    |                       | . . . . . R . . . . .         |                     | T . . . . A . . . . |           |
| A10-0515   | .....  |    |                       |                               |                     | T . . . . A . . . . |           |
| A10-0511   | .....  |    |                       |                               |                     | T . . . . A . . . . |           |
| A10-3932   | .....  |    |                       | . . . . . R . . . . .         |                     | T . . . . A . . . . |           |
| FLRC090148 | .....  |    |                       |                               |                     |                     |           |
| SM5596     | .....  |    |                       |                               |                     |                     | A . . . . |
| SM4871     | .....  |    |                       |                               |                     |                     | A . . . . |
| SM3844     | .....  |    |                       |                               |                     |                     | A . . . . |
| SHBRV      | .....  |    |                       |                               |                     |                     | A . HD .  |
| TN310      | .....  |    |                       |                               |                     |                     | A . HD .  |
| TN209      | .....  |    |                       |                               |                     | T . . . . A . HD .  |           |

|            | 101  | 110 | 120 | 130 | 140 | 150 | 160 |
|------------|--|-----|-----|-----|-----|-----|-----|
| OKSK090075 | KLAGDPRYEESLHNPYPEYHWLRTVKTTKESLIVISPSVADLDPYDKSLHSRIFPSGKCS |     |     |     |     |     |     |
| OKSK090076 | .....L   |     |     |     |     |     |     |
| MOSK090043 | .....D.....G....   |     |     |     |     |     |     |
| ARSK090058 | .....D.....G....   |     |     |     |     |     |     |
| KSSK090051 | .....D.....G....   |     |     |     |     |     |     |
| NEFE090019 | .....D.....L...G....   |     |     |     |     |     |     |
| KSSK090012 | .....D.....G....   |     |     |     |     |     |     |
| ARDG090042 | .M.....D.....VI.....V.....                                   |     |     |     |     |     |     |
| MNSK090088 | .M.....D.....VI.....V.....                                   |     |     |     |     |     |     |
| L920867WSK | .M.....D.....VI.....V.....                                   |     |     |     |     |     |     |
| CA982      | .M.....D.....VI.....V.....                                   |     |     |     |     |     |     |
| CASK2      | .M.....D.....VI.....V.....                                   |     |     |     |     |     |     |
| MEXSK3636  | .M.....D.....VI.....V..G....                                 |     |     |     |     |     |     |
| MEXSK3644  | .I.....T..D.....VI.....MV..N....                             |     |     |     |     |     |     |
| A10-3020   | .M.....D.....VI.....T.....V..G....                           |     |     |     |     |     |     |
| Fox-8007   | .M.....D.....VI.....T.....V..G....                           |     |     |     |     |     |     |
| A10-0515   | .M.....T..D.....VI.....V.....                                |     |     |     |     |     |     |
| A10-0511   | .M.....D.....VI.....V.....                                   |     |     |     |     |     |     |
| A10-3932   | .M.....D.....R.....VI.....V.....                             |     |     |     |     |     |     |
| FLRC090148 | .M.....D.....I.....G...T                                     |     |     |     |     |     |     |
| SM5596     | .M.....Q...D.S.....I.....V..G...L                            |     |     |     |     |     |     |
| SM4871     | .V.....Q...D.S.....I.....V..G...L                            |     |     |     |     |     |     |
| SM3844     | .M.....Q...D.S.....I.....V..G...L                            |     |     |     |     |     |     |
| SHBRV      | .M.....D..Q...D.....VI.....V.....L                           |     |     |     |     |     |     |
| TN310      | .M.....D..Q...D.....VI.....V.....L                           |     |     |     |     |     |     |
| TN209      | .M.....A..Q...D.....VI.....V.....L                           |     |     |     |     |     |     |

|            | 161  | 170 | 180 | 190 | 200 | 210 | 220 |
|------------|--|-----|-----|-----|-----|-----|-----|
| OKSK090075 | MTVSSTYCSTNHEYTIWMPEKVLGTSCDIFTTSKGGKASKGGKTCGFVDERGLYKSLK |     |     |     |     |     |     |
| OKSK090076 | .....R.....  |     |     |     |     |     |     |
| MOSK090043 | .....P.....AS.....R.....                                   |     |     |     |     |     |     |
| ARSK090058 | .....A.....R.....  |     |     |     |     |     |     |
| KSSK090051 | .....A.....T.....  |     |     |     |     |     |     |
| NEFE090019 | .....A.....T.....  |     |     |     |     |     |     |
| KSSK090012 | .....A.....T.....  |     |     |     |     |     |     |
| ARDG090042 | I..A.....D.....NPR.....N.R.....S.....I                     |     |     |     |     |     |     |
| MNSK090088 | I..F.....D.....NPR.....N.R.....S.....                      |     |     |     |     |     |     |
| L920867WSK | I.....D.....NPR.....N.R.....S.....                         |     |     |     |     |     |     |
| CA982      | I.....L.....NLR.....N.R.....R.S.....                       |     |     |     |     |     |     |
| CASK2      | I.....L..D.....NLR.....N.R.....R.S.....                    |     |     |     |     |     |     |
| MEXSK3636  | ...F.....D.....NPR.....AN.R.....R.S.....                   |     |     |     |     |     |     |
| MEXSK3644  | I..Y.....D.....A.....N.R.....R.....R                       |     |     |     |     |     |     |
| A10-3020   | I.....D.....NPR.....N.R.....S.....                         |     |     |     |     |     |     |
| Fox-8007   | I.....D.....SPR.....N.R.....S.....                         |     |     |     |     |     |     |
| A10-0515   | .....C..D.....NPR.....N.R.....S.....                       |     |     |     |     |     |     |
| A10-0511   | .....C..D.....NPR.....N.R.....S.....                       |     |     |     |     |     |     |
| A10-3932   | .I.....D.....NPR.....IN.R.....S.....                       |     |     |     |     |     |     |
| FLRC090148 | .L.....D.....A.....N.....                                  |     |     |     |     |     |     |
| SM5596     | .I.I.....D.....EAR.....N.R.....                            |     |     |     |     |     |     |
| SM4871     | .I.I.....D.....EAR.....N.R.....                            |     |     |     |     |     |     |
| SM3844     | .I.I.....D.....EAR.....N.R.....                            |     |     |     |     |     |     |
| SHBRV      | .I.....D.....VEAR.....N..K...R.W.....                      |     |     |     |     |     |     |
| TN310      | .I.....D.....VEPR.....N.R.....RV.....                      |     |     |     |     |     |     |
| TN209      | .I.....D.....VEPR.....N.R.....RV.....                      |     |     |     |     |     |     |

|            | 221  | 230 | 240 | 250 | 260 | 270 | 280 |
|------------|--|-----|-----|-----|-----|-----|-----|
| OKSK090075 | GACKLKLKCGVSGLRRLMDGTWVAIQTSDETKWCSPDQLVNLHDLHSDEIEHLVVEELVRKR |     |     |     |     |     |     |
| OKSK090076 | .....  |     |     |     |     |     |     |
| MOSK090043 | .....D.....  |     |     |     |     |     |     |
| ARSK090058 | .....I.....  |     |     |     |     |     |     |
| KSSK090051 | .....G.....  |     |     |     |     |     |     |
| NEFE090019 | .....  |     |     |     |     |     |     |
| KSSK090012 | .....  |     |     |     |     |     |     |
| ARDG090042 | .....L.K.....M.....P.....FR.....K..                            |     |     |     |     |     |     |
| MNSK090088 | .....L.K.....M..E..P.....R.....K..                             |     |     |     |     |     |     |
| L920867WSK | .....L.K.....M.....P.....R.....K..                             |     |     |     |     |     |     |
| CA982      | .....L.....M.....P.....FR.....K..                              |     |     |     |     |     |     |
| CASK2      | .....L.....M.....PS.....FR.....K..                             |     |     |     |     |     |     |
| MEXSK3636  | .....L..V.....M...D...P.....FR...Y.....K..                     |     |     |     |     |     |     |
| MEXSK3644  | .....P.....AT..PE.....F.....D..K..                             |     |     |     |     |     |     |
| A10-3020   | .....L.....M.....P.....FR.....K..                              |     |     |     |     |     |     |
| Fox-8007   | .....L.....M.....L.....FR.....                                 |     |     |     |     |     |     |
| A10-0515   | .....L.....M.I.....P.....FR.....K..                            |     |     |     |     |     |     |
| A10-0511   | .....L.....M...I..P.....FR.....K..                             |     |     |     |     |     |     |
| A10-3932   | .....L.....M..E...P.....FR.....K..                             |     |     |     |     |     |     |
| FLRC090148 | .....L.....F...L.....  |     |     |     |     |     |     |
| SM5596     | .....P.....T.....PS.....F.....K..                              |     |     |     |     |     |     |
| SM4871     | .....P.....T.....PS.....F.....K..                              |     |     |     |     |     |     |
| SM3844     | .....P.....T.....PS.....F.....K..                              |     |     |     |     |     |     |
| SHBRV      | .....P.....S...DI..P.....F.....IK..                            |     |     |     |     |     |     |
| TN310      | .....P.I.....S...EDI..P..R.....F...L.....IKR..                 |     |     |     |     |     |     |
| TN209      | ...R.....P.I..L....S...EDI..P..R.....F.....A..IKR..            |     |     |     |     |     |     |

|            | 281  | 290 | 300 | 310 | 320 | 330 | 340 |
|------------|--|-----|-----|-----|-----|-----|-----|
| OKSK090075 | EECLDALESIMTTKSVSFRRLSHLRKLVPGFGKAYTIFNKTLMEADAHYKSVRTWNEIIP |     |     |     |     |     |     |
| OKSK090076 | .....  |     |     |     |     |     |     |
| MOSK090043 | .....  |     |     |     |     |     |     |
| ARSK090058 | .....  |     |     |     |     |     |     |
| KSSK090051 | .....  |     |     |     |     |     |     |
| NEFE090019 | .....  |     |     |     |     |     |     |
| KSSK090012 | .....  |     |     |     |     |     |     |
| ARDG090042 | .....  |     |     |     |     |     |     |
| MNSK090088 | .....  |     |     |     |     |     |     |
| L920867WSK | .....  |     |     |     |     |     |     |
| CA982      | .....I.....  |     |     |     |     |     |     |
| CASK2      | .....I.....  |     |     |     |     |     |     |
| MEXSK3636  | .....  |     |     |     |     |     |     |
| MEXSK3644  | .....T.....  |     |     |     |     |     |     |
| A10-3020   | .....  |     |     |     |     |     |     |
| Fox-8007   | .....I...V..   |     |     |     |     |     |     |
| A10-0515   | .....R.....V..   |     |     |     |     |     |     |
| A10-0511   | .....R.....V..   |     |     |     |     |     |     |
| A10-3932   | .....  |     |     |     |     |     |     |
| FLRC090148 | .....S.....  |     |     |     |     |     |     |
| SM5596     | .....I.....  |     |     |     |     |     |     |
| SM4871     | .....I.....  |     |     |     |     |     |     |
| SM3844     | .....I.....  |     |     |     |     |     |     |
| SHBRV      | G.....N.....V..  |     |     |     |     |     |     |
| TN310      | N.....K...V..  |     |     |     |     |     |     |
| TN209      | .N.....T..T.....K...V..                                      |     |     |     |     |     |     |



|            | 341   | 350 | 360       | 370 | 380       | 390 | 400   |
|------------|---|-----|-----------|-----|-----------|-----|-------|
| OKSK090075 | SKGCLRGGKCHPHVNGVFFNGIILGPDGHVLIPEMQSSLLQQHIELLESSVIPLMHPLA |     |           |     |           |     |       |
| OKSK090076 | .....   |     |           |     |           |     |       |
| MOSK090043 | .....   |     |           |     |           |     | M     |
| ARSK090058 | .....   |     |           |     |           |     | V     |
| KSSK090051 | .....   |     |           |     |           |     | M     |
| NEFE090019 | .....   |     |           |     |           |     | M     |
| KSSK090012 | .....   |     |           |     |           |     | M     |
| ARDG090042 | .....R.....   |     |           |     |           |     | M     |
| MNSK090088 | ...K...R...   |     | ...V...   |     | .....     |     | M     |
| L920867WSK | ...K.E.R...   |     | ...V...   |     | .....     |     | M     |
| CA982      | .....R.....   |     | ...K..... |     | .....     |     | M...I |
| CASK2      | .....R.....   |     |           |     |           |     | V     |
| MEXSK3636  | .....R.....   |     |           |     |           |     | M     |
| MEXSK3644  | .....R.....   |     |           |     |           |     | V.M   |
| A10-3020   | .....R.....   |     |           |     |           |     | M     |
| Fox-8007   | .....R.....   |     |           |     |           |     | Q     |
| A10-0515   | .....R.....   |     |           |     |           |     | M     |
| A10-0511   | .....R.....   |     |           |     |           |     | M     |
| A10-3932   | .....R.....   |     |           |     |           |     | M     |
| FLRC090148 | .....R.....   |     |           |     |           |     | V     |
| SM5596     | ...K...R...   |     | ...D...   |     | ...N...   |     | V     |
| SM4871     | ...K...R...   |     | ...D...   |     | ...N...   |     | V     |
| SM3844     | ...K...R...   |     | ...D...   |     | ...N...   |     | V     |
| SHBRV      | ...K...R...   |     | ...P...   |     | ...N...   |     | M...T |
| TN310      | ...E.....   |     | ...P..... |     | ...N..... |     | M     |
| TN209      | ...E...R...   |     | .....     |     | ...N..... |     | M     |

|            | 401   | 410         | 420       | 430         | 440           | 450         | 460           |
|------------|---|-------------|-----------|-------------|---------------|-------------|---------------|
| OKSK090075 | DLSTVFKDGDDEAEDFVEVHLPDVHKQISGVDLGLPSWGKYVLMSAGALATLILVISLMKC |             |           |             |               |             |               |
| OKSK090076 | .....   |             |           |             |               |             | F             |
| MOSK090043 | P.....  | D.....      | ...V...   |             | ...F...       |             | MM            |
| ARSK090058 | P.....  | D.....      | ...V...   |             | .....         |             | M             |
| KSSK090051 | P.....  | D.....      | ...V...   |             | ...R...       |             | V             |
| NEFE090019 | P.....  | D.....      | ...V...   |             | ...R...       |             | M             |
| KSSK090012 | P.....  | D.....      | ...V...   |             | ...R...       |             | M             |
| ARDG090042 | P..I.....   | .....       |           | ...M...N... |               | ...SL...    | IA.M.I.L.IT   |
| MNSK090088 | P..I.....   | .....       |           | ...V...N... |               | ...SL...    | IA.M.I.L.IT   |
| L920867WSK | P.....  | .....       |           | ...V...N... |               | ...ISL...   | IA.M.I.L..T   |
| CA982      | P.A.....  | .....       |           | ...V...N... |               | ...I.V...   | IA.M.I.F..T   |
| CASK2      | P.....  | E.....      | ...V...   |             | ...N...A.V... |             | TA.M.IAF..T   |
| MEXSK3636  | P.....  | E.....      | ...V.E... |             | ...N...I.I... |             | IA.M.I.F..T   |
| MEXSK3644  | P.....  | N.....      | ...V.I... |             | ...R.LV.I...  |             | M.T..F..I     |
| A10-3020   | P.....  | .....       |           | ...V...N... |               | ...I...     | IA.M.I.F..T   |
| Fox-8007   | P.....  | E.....      | ...V...   |             | ...N...I...   |             | IA.M.I.F..T   |
| A10-0515   | P.....  | E.....      | ...V...   |             | ...N.....     |             | IA.M.I.F..T   |
| A10-0511   | P.....  | E.....      | ...V...   |             | ...N.....     |             | IA.M.I.F..T   |
| A10-3932   | P.....  | E.....      | ...V...   |             | ...N.....     |             | V.ISRM.L.F..T |
| FLRC090148 | P.A.....  | .....       |           | .....       |               | ...M.I...   | M.M.F.F..T    |
| SM5596     | P.....  | ...I...V... |           | ...N...L... |               | ...M.I.F..A |               |
| SM4871     | P.....  | ...I...V... |           | ...N...L... |               | ...M.T.F..T |               |
| SM3844     | P.....  | ...I...V... |           | ...N...L... |               | ...M.I.F..A |               |
| SHBRV      | P.....  | ...V.EI...  |           | ...L...V... |               | ...A.F.IT   |               |
| TN310      | P.....  | E.....      | ...V.D... |             | ...L...I...   |             | A.V.II        |
| TN209      | P.....  | E.....      | ...V.D... |             | ...L.I...     |             | I..A.V.II     |

|            | 461                | 470                           | 480      | 490      | 500      | 505 |                     |
|------------|--------------------|-------------------------------|----------|----------|----------|-----|---------------------|
| OKSK090075 | CRWVNRTKSI         | QQGPRESERK                    | VSVPQSGK | VISSWESY | KIGGETGL |     | (SCSK I)            |
| OKSK090076 | .....P....         | P.....N.....                  |          |          |          |     | (SCSK I)            |
| MOSK090043 | ..GF.....R..       | SG..G.....N.....              |          |          |          |     | (SCSK IIA)          |
| ARSK090058 | ..GF.....TR.DF.    |                               |          |          |          |     | (SCSK IIB)          |
| KSSK090051 | ..KG.....K..S..... | I..N.....S.....               |          |          |          |     | (SCSK IIIA)         |
| NEFE090019 | ..KG.....R..S..... | I..N.....S.....               |          |          |          |     | (SCSK IIIB)         |
| KSSK090012 | ..KG.....K..S..... | I.....S.....                  |          |          |          |     | (SCSK IIIB)         |
| ARDG090042 | ..R.D.SEPT.RS      | LEGGK.....S.N.....            |          |          |          |     | (NCSK)              |
| MNSK090088 | ..K...PEPT.RS      | LEGGK.....S.....              |          |          |          |     | (NCSK)              |
| L920867WSK | ..K...PEPT.RS      | LEGGK.....S.....              |          |          |          |     | (NCSK)              |
| CA982      | F.GF.LP..T.HSLG    | .T..M.....I.....              |          |          |          |     | (CASK)              |
| CASK2      | ..G..LP..T.RS      | .GGTR....AS.....A..A..S.C..KM |          |          |          |     | (CASK)              |
| MEXSK3636  | ..GR...SE.K.RSL    | GGTG.....S.....S.....         |          |          |          |     | (SBCSK)             |
| MEXSK3644  | ..R..KAG...D.EK.   | KRR.....N.....S.S..R.         |          |          |          |     | (CENTRAL MEXICO SK) |
| A10-3020   | ..R...PE.T.RSL     | GGTG....I.S.....S.....        |          |          |          |     | (TX GRAY FOX)       |
| Fox-8007   | ..R...SE.T.HRL     | .GTG.....S.T.....S.....       |          |          |          |     | (TX GRAY FOX)       |
| A10-0515   | ..R.S.PE.M.RSL     | GGTG....AAS.....S.....        |          |          |          |     | (AZ GRAY FOX)       |
| A10-0511   | ..R...PE.T.RIL     | GGTGK...AAS.....S.....Q.      |          |          |          |     | (AZ GRAY FOX)       |
| A10-3932   | ..R...PE.T.RS      | .GGAG.....S.....S.....        |          |          |          |     | (US COYOTE)         |
| FLRC090148 | ..KR....E..R....   | L.S.....NR.....S....K.        |          |          |          |     | (FL RACCOON)        |
| SM5596     | ..RA...E...RD.G..  | GK...IPS.D.....L..S....R.     |          |          |          |     | (AZ SKUNK VARIANT)  |
| SM4871     | ..RA...E...RD.G..  | GK...I.S.D.....L..S....R.     |          |          |          |     | (BIG BROWN BAT)     |
| SM3844     | ..RA...E...RD.G..  | G....IPS.D.....L..S....R.     |          |          |          |     | (BIG BROWN BAT)     |
| SHBRV      | ..RA...E.T.R.R...  | GG....A..N..I....L..S.S....   |          |          |          |     | (SILVER-HAIRED BAT) |
| TN310      | ..R..K.R.T.R.Q...  | RG.M..A..N..I....L..RES....   |          |          |          |     | (HOARY BAT)         |
| TN209      | ..R..K.G.T.R.H...  | GG.M..V..N..I....L..SES....   |          |          |          |     | (RED BAT)           |

Figure S2. An alignment of glycoprotein sequences of representative skunk-associated rabies viruses examined in this study with comparison to the sequences of other viral variants that circulate in sympatric hosts of the US mid-west. A member of the SCSK type I group is used as the reference isolate. The (!) symbol identifies residues which exhibit notable substitutions. Residue number 1 corresponds to the first residue of the mature processed product following removal of the 19 amino acid signal peptide. Certain motifs mentioned in the text are of special note: the linear epitope "WXXXDI" at residues 14-19, a potential glycosylation site within antigenic site II (amino acids 37-39), the core sequence of the linear G5 epitope (residues 261-264), antigenic site III (residues 330-338). The variant type of each sequence is shown at the end of the alignment. The following amino acid replacements are of note: Thr90 is present exclusively in the cosmopolitan lineage except for isolate Tn209; Glu93 is restricted to most NCSK isolates; Glu118 is found in SCSK I only; Pro170 and Ser184 are restricted to SCSK IIA (except ARSK90066); Thr201 is found in SCSK III only; Arg202 is found in CASK and MEX SBC variants only; Ser204 is restricted to the cosmopolitan lineage; Lys234 is observed only in the NCSK clade; Met243 is found only in the cosmopolitan lineage while Ile243 occurs in the American indigenous samples; Ile249 is found in SCSK IIB and in some bat variants; Ile287 is observed only in CASK variants; Met365 and Ile396 are seen only in the Central Mexican skunk variant; Phe441 is in SCSK IIA isolates only; Trp463 is observed in SCSK I only. The lack of location data for the limited number of CASK isolates available precluded their sub-classification as achieved for the N protein analysis.

Table S1. Details of newly characterized rabies viruses examined at the KSU facility in this study.

| <b>Isolate</b> | <b>Host-species</b> | <b>ZIP Code</b> | <b>Date</b> | <b>Accession (N)</b> | <b>Accession (G)</b> |
|----------------|---------------------|-----------------|-------------|----------------------|----------------------|
| KSSK090001     | Striped skunk       | 67516           | 05-Jan-09   | JX855962             | JX856040             |
| KSSK090002     | Striped skunk       | 67560           | 08-Jan-09   | JX855963             | JX856041             |
| KSSK090003     | Striped skunk       | 67560           | 12-Jan-09   | JX855964             | JX856042             |
| NEEQ090004     | Horse               | 68040           | 13-Jan-09   | JX855965             | JX856043             |
| COSK090005     | Striped skunk       | 80836           | 16-Jan-09   | JX855966             | JX856044             |
| KSSK090006     | Striped skunk       | 67520           | 21-Jan-09   | JX855967             | JX856045             |
| KSSK090007     | Striped skunk       | 67147           | 29-Jan-09   | JX855968             | JX856046             |
| NERF090008     | Red fox             | 68416           | 04-Feb-09   | JX855969             | JX856047             |
| KSSK090009     | Striped skunk       | 67653           | 05-Feb-09   | JX855970             | JX856048             |
| KSRC090010     | Raccoon             | 67443           | 11-Feb-09   | JX855971             | JX856049             |
| KSBC090011     | Bobcat              | 67073           | 11-Feb-09   | JX855972             | JX856050             |
| KSSK090012     | Striped skunk       | 67730           | 11-Feb-09   | JX855973             | JX856051             |
| KSSK090013     | Striped skunk       | 67226           | 19-Feb-09   | JX855974             | JX856052             |
| KSSK090014     | Striped skunk       | 67623           | 24-Feb-09   | JX855975             | JX856053             |
| KSDG090015     | Dog                 | 66749           | 24-Feb-09   | JX855976             | JX856054             |
| KSSK090016     | Striped skunk       | 67584           | 25-Feb-09   | JX855977             | JX856055             |
| KSSK090017     | Striped skunk       | 67730           | 27-Feb-09   | JX855978             | JX856056             |
| NEFE090018     | Cat                 | 68701           | 06-Mar-09   | JX855979             | JX856057             |
| NEFE090019     | Cat                 | 69028           | 06-Mar-09   | JX855980             | JX856058             |
| KSSK090020     | Striped skunk       | 67661           | 10-Mar-09   | JX855981             | JX856059             |
| KSSK090021     | Striped skunk       | 67749           | 13-Mar-09   | JX855982             | JX856060             |
| KSRF090022     | Red fox             | 67635           | 16-Mar-09   | JX855983             | JX856061             |
| NESK090023     | Striped skunk       | 68601           | 16-Mar-09   | JX855984             | JX856062             |
| KSSK090025     | Striped skunk       | 67067           | 18-Mar-09   | JX855985             | JX856063             |
| KSSK090027     | Striped skunk       | 66066           | 18-Mar-09   | JX855986             | JX856064             |
| KSSK090028     | Striped skunk       | 67530           | 19-Mar-09   | JX855987             | JX856065             |
| KSSK090030     | Striped skunk       | 66963           | 26-Mar-09   | JX855988             | JX856066             |
| NEFE090033     | Cat                 | 68601           | 31-Mar-09   | JX855989             | JX856067             |

|            |               |       |           |          |          |
|------------|---------------|-------|-----------|----------|----------|
| KSSK090035 | Striped skunk | 67749 | 01-Apr-09 | JX855990 | JX856068 |
| NESK090036 | Striped skunk | 69032 | 01-Apr-09 | JX855991 | JX856069 |
| MOSK090038 | Striped skunk | 65655 | 14-Feb-09 | JX855992 | JX856070 |
| MOSK090041 | Striped skunk | 64491 | 27-Mar-09 | JX855993 | JX856071 |
| ARDG090042 | Dog           | 72444 | 31-Mar-09 | JX855994 | JX856072 |
| MOSK090043 | Striped skunk | 65775 | 31-Mar-09 | JX855995 | JX856073 |
| SDSK090044 | Striped skunk | 57022 | 03-Feb-09 | JX855996 | JX856074 |
| SDSK090045 | Striped skunk | 57225 | 06-Mar-09 | JX855997 | JX856075 |
| SDSK090046 | Striped skunk | 57054 | 31-Mar-09 | JX855998 | JX856076 |
| KSSK090049 | Striped skunk | 67401 | 08-Apr-09 | JX855999 | JX856077 |
| KSSK090050 | Striped skunk | 67861 | 08-Apr-09 | JX856000 | JX856078 |
| KSSK090051 | Striped skunk | 66775 | 09-Apr-09 | JX856001 | JX856079 |
| KSSK090053 | Striped skunk | 67572 | 14-Apr-09 | JX856002 | JX856080 |
| KSSK090054 | Striped skunk | 67730 | 14-Apr-09 | JX856003 | JX856081 |
| KSSK090055 | Striped skunk | 67226 | 15-Apr-09 | JX856004 | JX856082 |
| ARSK090056 | Striped skunk | 72560 | 22-Jan-09 | JX856005 | JX856083 |
| ARSK090057 | Striped skunk | 71920 | 19-Feb-09 | JX856006 | JX856084 |
| ARSK090058 | Striped skunk | 71923 | 03-Mar-09 | JX856007 | JX856085 |
| ARSK090059 | Striped skunk | 71920 | 11-Mar-09 | JX856008 | JX856086 |
| ARSK090060 | Striped skunk | 71920 | 11-Mar-09 | JX856009 | JX856087 |
| ARSK090062 | Striped skunk | 72533 | 26-Mar-09 | JX856010 | JX856088 |
| ARSK090064 | Striped skunk | 71920 | 31-Mar-09 | JX856011 | JX856089 |
| ARSK090065 | Striped skunk | 72653 | 01-Apr-09 | JX856012 | JX856090 |
| ARSK090066 | Striped skunk | 72774 | 03-Apr-09 | JX856013 | JX856091 |
| ARSK090067 | Striped skunk | 75502 | 07-Apr-09 | JX856014 | JX856092 |
| ARSK090068 | Striped skunk | 71920 | 08-Apr-09 | JX856015 | JX856093 |
| NESK090072 | Striped skunk | 65562 | 16-Apr-09 | JX856016 | JX856094 |
| OKBV090073 | Cow           | 73620 | 08-Jan-09 | JX856017 | JX856095 |
| OKSK090075 | Striped skunk | 73096 | 15-Jan-09 | JX856018 | JX856096 |
| OKSK090076 | Striped skunk | 73521 | 26-Feb-09 | JX856019 | JX856097 |
| OKSK090078 | Striped skunk | 73040 | 24-Mar-09 | JX856020 | JX856098 |

|            |               |       |            |          |          |
|------------|---------------|-------|------------|----------|----------|
| OKSK090079 | Striped skunk | 73456 | 04-Apr-09  | JX856021 | JX856099 |
| OKSK090080 | Striped skunk | 73438 | 15-Apr-09  | JX856022 | JX856100 |
| KSBV090081 | Cow           | 66783 | 17-Apr-09  | JX856023 | JX856101 |
| NESK090082 | Striped skunk | 68745 | 20-Apr-09  | JX856024 | JX856102 |
| KSSK090084 | Striped skunk | 67438 | 21-Apr-09  | JX856025 | JX856103 |
| NESK090085 | Striped skunk | 69101 | 22-Apr-09  | JX856026 | JX856104 |
| MNSK090088 | Striped skunk | 56151 | 07-Apr-09  | JX856027 | JX856105 |
| SDSK090090 | Striped skunk | 57042 | 08-Apr-09  | JX856028 | JX856106 |
| SDSK090093 | Striped skunk | 57054 | 21-Apr-09  | JX856029 | JX856107 |
| SDSK090094 | Striped skunk | 57020 | 21-Apr-09  | JX856030 | JX856108 |
| MNSK090095 | Striped skunk | 56164 | 22-Apr-09  | JX856031 | JX856109 |
| KSSK090097 | Striped skunk | 67749 | 30-Apr-09  | JX856032 | JX856110 |
| MOSK090098 | Striped skunk | 65793 | 21-Apr-09  | JX856033 | JX856111 |
| KSSK090100 | Striped skunk | 67863 | 06-May-09  | JX856034 | JX856112 |
| NESK090101 | Striped skunk | 68822 | 07-May-09  | JX856035 | JX856113 |
| NEBV090102 | Cow           | 68966 | 07-May-09  | JX856036 | JX856114 |
| NESK090103 | Striped skunk | 69101 | 07-May-09  | JX856037 | JX856115 |
| NESK090104 | Striped skunk | 69045 | 11-May-09  | JX856038 | JX856116 |
| FLRC090148 | Raccoon       | 32303 | 19-June-09 | JX856039 | JX856117 |

Table S2. Additional rabies virus isolates employed in this study

| Isolate designation | Host species  | Location                | Year | Strain                | GenBank Accession number<br>N gene | GenBank Accession number<br>G gene |
|---------------------|---------------|-------------------------|------|-----------------------|------------------------------------|------------------------------------|
| 4995AZSK            | Skunk         | Flagstaff, AZ           | 2001 | AZ skunk <sup>1</sup> | AY170402                           |                                    |
| 5470AZSK            | Skunk         | Flagstaff, AZ           | 2001 | AZ skunk <sup>1</sup> | AY170403                           |                                    |
| SM5596              | Striped skunk | AZ, USA                 | 2004 | AZ skunk <sup>1</sup> |                                    | JQ685964                           |
| V684SBCSK           | Skunk         | Baja California, Mexico | 1992 | SBC skunk             | AY854593                           |                                    |
| SKBCS2007           | Spotted skunk | Baja California, Mexico | 2007 | SBC skunk             | FJ228543                           |                                    |
| 3908Mxslpsk         | Spotted skunk | Baja California, Mexico | 2000 | SBC skunk             | FJ228545                           |                                    |
| MEXSK3636           | Spotted skunk | Mexico                  | 2009 | SBC skunk (MexSK-2)   |                                    | JQ685975                           |
| CA982               | Skunk         | USA                     | 1994 | CASK                  |                                    | JQ685894                           |
| CASK2               | Skunk         | USA                     | 1974 | CASK                  |                                    | JQ685970                           |
| V637CASK*           | Skunk         | Glenn Co., CA           | 1994 | CASK                  | JQ513539                           |                                    |
| V638CASK*           | Skunk         | Sutter Co., CA          | 1994 | CASK                  | JQ513540                           |                                    |
| V640CASK*           | Dog           | Sonoma Co., CA          | 1994 | CASK                  | JQ513541                           |                                    |
| V644CASK*           | Mountain lion | Yolo Co., CA            | 1994 | CASK                  | JQ513542                           |                                    |
| V646CASK*           | Skunk         | Glenn Co., CA           | 1994 | CASK                  | JQ513543                           |                                    |
| V648CASK*           | Skunk         | Colusa Co., CA          | 1994 | CASK                  | JQ513544                           |                                    |
| V649CASK*           | Dog           | Mendocino Co., CA       | 1997 | CASK                  | JQ513545                           |                                    |
| V650CASK*           | Skunk         | Trinity Co., CA         | 1997 | CASK                  | JQ513546                           |                                    |
| V651CASK*           | Skunk         | Mariposa Co., CA        | 1997 | CASK                  | JQ513547                           |                                    |
| V652CASK*           | Skunk         | Mariposa Co., CA        | 1997 | CASK                  | JQ513548                           |                                    |
| V654CASK*           | Skunk         | Amador Co., CA          | 1997 | CASK                  | JQ513549                           |                                    |
| V656CASK*           | Skunk         | Glenn Co., CA           | 1997 | CASK                  | JQ513550                           |                                    |
| V657CASK*           | Fox           | Mariposa Co., CA        | 1997 | CASK                  | JQ513551                           |                                    |
| V658CASK*           | Skunk         | Mariposa Co., CA        | 1997 | CASK                  | JQ513552                           |                                    |

|               |               |                                       |      |  |            |          |
|---------------|---------------|---------------------------------------|------|--|------------|----------|
| 421Kydg07     | Dog           | KY                                    | 2007 | NCSK   | GU991842   |          |
| 1061AZhr08    | Horse         | AZ                                    | 2008 | NCSK   | GU991843   |          |
| L921741WSK    | Striped skunk | Western Canada                        | 1992 | NCSK   | AF344304   |          |
| L920867WSK    | Striped skunk | Western Canada                        | 1992 | NCSK   | AF344306   | AF344307 |
| L000160WSK*   | Striped skunk | MB, Canada                            | 2000 | NCSK   | JQ513557   |          |
| L000416WSK*   | Striped skunk | MB, Canada                            | 2000 | NCSK   | JQ513558   |          |
| L000481WSK*   | Striped skunk | SK, Canada                            | 2000 | NCSK   | JQ513559   |          |
| L000504WSK*   | Striped skunk | MB, Canada                            | 2000 | NCSK   | JQ513560   |          |
| L041025WSK*   | Striped skunk | MB, Canada                            | 2004 | NCSK   | JQ513554   |          |
| L050063WSK*   | Striped skunk | SK, Canada                            | 2005 | NCSK   | JQ513555   |          |
| L050085WSK*   | Striped skunk | MB, Canada                            | 2005 | NCSK   | JQ513556   |          |
| 3789SK        | Skunk         | Chippewa, WI                          | 1998 | NCSK   | AF461045   |          |
| A10-6931      | Fox           | USA                                   | 2010 | NCSK   |            | JQ685987 |
| 2311Mxzacs01  | Spotted skunk | Zacatecas, Mexico<br>San Luis Potosi, | 2001 | Central Mexico skunk<br>Central Mexico skunk | FJ228483   |          |
| 2818Mxslpsk02 | Spotted skunk | Mexico                                | 2002 |  | FJ228484   |          |
| Mxchihbv1999  | Bovine        | Chihuahua, Mexico<br>San Luis Potosi, | 1999 | Central Mexico skunk<br>Central Mexico skunk | FJ228485   |          |
| V854SLPSK*    | Skunk         | Mexico                                | 2002 | Central Mexico skunk                         | JQ513553   | JQ685929 |
| MEXSK3644     | Spotted skunk | Mexico                                | 2009 | (MexSK-1)                                    |            |          |
| PA62RC        | Raccoon       | Washington Co., PA                    | 2003 | Raccoon                                      | EF508144.1 |          |
| 5422SCSK      | Skunk         | Sierra Vista, AZ                      | 2001 | SCSK   | AF483524.1 |          |
| NMsk2007      | Skunk         | NM                                    | 2007 | SCSK   | GU991838.1 |          |
| TXdg1968      | Dog           | TX                                    | 1968 | SCSK   | GU991837.1 |          |
| V211TXSK      | Skunk         | TX                                    | 1994 | SCSK   | EU345002.1 |          |
| V212TXSK      | Skunk         | TX                                    | 1994 | SCSK   | EU345003.1 |          |
| V216TXSK      | Skunk         | TX                                    | 1994 | SCSK   | EU345004.1 |          |
| A10-0511      | Gray fox      | USA                                   | 2009 | AZ fox                                       |            | JQ685943 |
| A10-0515      | Gray fox      | USA                                   | 2009 | AZ fox                                       |            | JQ685899 |
| FLDG1994      | Dog           | FL                                    | 1994 | TX coyote                                    | FJ228528   |          |

|            |                  |                     |      |                                |            |          |
|------------|------------------|---------------------|------|--------------------------------|------------|----------|
| H2USTXCY98 | Coyote           | TX                  | 1998 | TX coyote                      | FJ228530   |          |
| TX692007CY | Coyote           | TX                  | 2007 | TX gray fox                    | FJ228536   |          |
| A10-3932   | Gray fox         | USA                 | 2010 | Coyote                         |            | JQ685985 |
| TX672007GF | Gray fox         | TX                  | 2007 | TX gray fox                    | FJ228538   |          |
| A10-3020   | Gray fox         | USA                 | 2008 | TX gray fox                    |            | JQ685984 |
| Fox-8007   | Gray fox         | USA                 | 2008 | TX gray fox                    |            | JQ685995 |
| 132BB      | Big brown bat    | Adams Co., PA       | 1984 | <i>E. fuscus</i> bat           | AY039229.1 |          |
| 906BB      | Big brown bat    | WA                  | 1987 | <i>E. fuscus</i> bat           | AY039227.1 |          |
| 2049BB     | Big brown bat    | CO                  | 1985 | <i>E. fuscus</i> bat           | AY039228.1 |          |
| 4886BB     | Big brown bat    | Flagstaff, AZ       | 1999 | <i>E. fuscus</i> bat           | AY170400.1 |          |
| 5442BB     | Big brown bat    | Flagstaff, AZ       | 2001 | <i>E. fuscus</i> bat           | AY170412.1 |          |
| SM4871     | Big brown bat    | USA                 | 1999 | <i>E. fuscus</i> bat           |            | JQ685923 |
| SM3844     | Big brown bat    | USA                 | 1995 | <i>E. fuscus</i> bat           |            | JQ685974 |
| 3850MY     | Cave myotis bat  | Flagstaff, AZ       | 1996 | <i>E. fuscus</i> bat           | AY170414.1 |          |
|            | Southeastern     |                     |      |                                |            |          |
| 876MY      | myotis bat       | Highlands Co., FL   | 1988 | <i>Southern Myotis</i> bat     | AY039225.1 |          |
| 1563CL     | California bat   | Shasta Co., CA      | 1986 | <i>Myotis</i> bat              | AF394873.1 |          |
| 1261MY     | Long-eared bat   | Napa Co., CA        | 1990 | <i>Myotis</i> bat              | AF394874.1 |          |
| 872YB      | Yellow bat       | Sarasota, FL        | 1988 | <i>L. intermedius</i> bat      | AF394878.1 |          |
| 79L1020LB  | Little brown bat | AB, Canada          | 1979 | Silver-haired bat              | AF351837.1 |          |
|            | Silver-haired    |                     |      |                                |            |          |
| 2152SH     | bat              | Westchester Co., NY | 1984 | Silver-haired bat              | AF394880.1 |          |
| SHBRV      |                  |                     |      | Silver-haired bat              |            | AY705373 |
|            | Eastern          |                     |      |                                |            |          |
| 1435EP     | pipistrelle      | Houston Co., AR     | 1991 | Silver-haired bat <sup>2</sup> | AF394881.1 |          |
| 814HR      | Hoary bat        | Tehema Co., CA      | 1986 | <i>Lasiurius</i> bat           | AF394883.1 |          |
| 93N6220HR  | Hoary bat        | ON, Canada          | 1993 | <i>Lasiurius</i> bat           | AF351846.1 |          |
| TN310      | Hoary bat        | USA                 | 2004 | <i>Lasiurius</i> bat           |            | JQ685947 |
| 1428RD     | Red bat          | Little Rock, AR     | 1991 | <i>Lasiurius</i> bat           | AY039224.1 |          |
| 2085RD     | Red bat          | Walker Co., TX      | 1986 | <i>Lasiurius</i> bat           | AF394886.1 |          |
| TN209      | Red bat          | USA                 | 2005 | <i>Lasiurius</i> bat           |            | JQ685902 |



RV1333

Human

United Kingdom

2002

EBLV-2

NC\_009528.1

<sup>1</sup> Skunk-adapted strain originating from big brown bat

<sup>2</sup> Also believed to be harbored by the eastern pipistrelle bat

\* Specimens characterized during the course of these studies

**Table S3. Primers used for RT-PCR at KSU.**

| Primer name | Sequence (5' – 3')         | Direction | Position* |
|-------------|----------------------------|-----------|-----------|
| 001         | ACGCTTAACGAMAAA            | forward   | 1-15      |
| 550F        | ATGTGYGCTAAYTGGAGYAC       | forward   | 647-666   |
| 1066NB      | TCCCTGAAGAATCTTCTCTC       | reverse   | 1155-1136 |
| 304         | TTGACGAAGATCTTGCTCAT       | reverse   | 1533-1514 |
| 998NC       | AGGAGCACATGTGGCGATA        | reverse   | 1075-1057 |
| 3259GF      | TTTGACCCTCTTGGATGTGA       | forward   | 3259-3278 |
| 4081GF      | GGATGGAACATGGGTCGCG        | forward   | 4081-4099 |
| 4181GR      | GCTCAATCTCGTCTGAGCGAACGTGC | reverse   | 4181-4153 |
| SYR         | CAAAGGAGAGTTGAGATTGTAGTC   | reverse   | 5542-5519 |
| L1deg       | GAGTTNAGGTTGTAGTCAGAG      | reverse   | 5535-5515 |
| 989GRdeg    | CTKAGACGTCTRAARCTYAC       | reverse   | 4278-4259 |

\* Location based on ERA strain sequence (GenBank accession number EF206707.1). The primer combinations used routinely for RT-PCR were as follows: N gene, 001 / 1066NB and 550F / 304; G gene 3259GF / 4181GR and 4081GF / SYR. The additional alternative primers listed were occasionally used when the standard primers failed to amplify the product.