**Spike protein of SARS-CoV-2: Impact of single amino acid mutation and effect of drug binding to the variant-*insilico* analysis.**

Supplementary data



Figure 1.1: Sequence alignment of NTD- S1-domain of S-protein of diverse members. Conserved domain analysis was carried for the following species: SARS-CoV (PDB ID: 5X4S\_A, 6ACC\_A), HKU1 (PDB ID: 6ACC\_A), MERES (PDB ID: 5X4R\_A, 6NB3\_A), Bat Coronavirus HKU9-2 (PDB ID: ABN10919), Murine hepatitis virus (PDB ID: 3JCL\_A), Human coronavirus OC43 (PDB ID: 6NZK\_A). Amino acids in red indicates highly conserved region and blue indicated moderately conserved region. The dashed line indicates variation in the sequence length after alignment and lower cases of amino acid indicates unaligned amino acids. The # symbol represents the amino acids involved in the catalytic or binding activity.



Figure 1.2: Sequence alignment of CTD of S1-domain of S-protein. Conserved domain analysis was carried out for the following species: SARS-CoV-2 (PDB ID: 6M0J\_E), HCoV-OC43 (PDB ID: 6NZK\_A), MERS (PDB ID: 4ZS6\_A, 6NB3\_A), SARS (PDB ID: 3SCI\_E, 6VSB\_C, 6ACC\_A), Murine Hepatitis Virus (PDB ID: 3JCL\_A), HKU1 (PDB ID: 5GNB\_A, 5I08\_A), HKU4 (PDB ID: 4QZV\_B), 2cEMC (PDB ID: 4KR0\_B).



Figure 1.3: Sequence alignment of S2-domain of S-protein. Feline infectious peritonitis virus (PDB ID: P10033), Human coronavirus 229E (PDB ID: P15423), Avian infectious bronchitis virus (PDB ID: P11223), SARS coronavirus (PDB ID: P59594).

Table 1: List of S-protein sequence Accession number obtained from NCBI viruses with the Country name.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| So. No. | Accession Number | Country | So.No. | Accession Number | Country |
| 1. | NC\_045512 | Wuhan | 41. | MT607244 | India |
| 2. | MT627325 | China | 42. | MT576034 | India |
| 3. | MT622319 | China | 43. | MT576042 | India |
| 4. | MT568634 | China | 44. | MT509669 | USA |
| 5. | MT568635 | China | 45. | MT246488 | USA |
| 6. | MT568637 | China | 46. | MT252690 | USA |
| 7. | MT568640 | China | 47. | MT345820 | USA |
| 8. | LC553263 | Japan | 48. | MT259262 | USA |
| 9. | LC553257 | Japan | 49. | MT641485 | USA |
| 10. | LC553269 | Japan | 50. | MT269282 | USA |
| 11. | MT590598 | Taiwan | 51. | MT246461 | USA |
| 12. | MT499173.1 | USA | 52. | MT293177 | USA |
| 13. | MT460139 | USA | 53. | MT293208 | USA |
| 14. | MT506885 | USA | 54. | MT263403 | USA |
| 15. | MT499201.1 | USA | 55. | MT263412 | USA |
| 16. | MT444608 | USA | 56. | MT263418 | USA |
| 17. | MT444612 | USA | 57. | MT259236 | USA |
| 18. | MT439311 | USA | 58. | MT262915 | USA |
| 19. | MT461603 | USA | 59. | MT263457 | USA |
| 20. | MT499205 | USA | 60. | MT412200 | USA |
| 21. | MT499200 | USA | 61. | MT385426 | USA |
| 22. | MT506888 | USA | 62. | MT385440 | USA |
| 23. | MT506701 | USA | 63. | MT385458 | USA |
| 24. | MT500122 | Pakistan | 64. | MT509958 | Bangladesh |
| 25. | MT270101 | Germany | 65. | MT622321 | Italy |
| 26. | MT507271 | Jamaica | 66. | MT527178 | Italy |
| 27. | MT499208 | Poland | 67. | MT528235 | Italy |
| 28. | MT470102 | France | 68. | MT528237 | Italy |
| 29. | MT450935 | Australia | 69. | MT077125 | Italy |
| 30. | MT451139 | Australia | 70. | MT510690 | Africa |
| 31. | MT439595 | India | 71. | MT510691 | Africa |
| 32. | MT576044 | India | 72. | MT510692 | Africa |
| 33. | MT635269 | India | 73. | MT510693 | Africa |
| 34. | MT635270 | India | 74. | MT510696 | Africa |
| 35. | MT635407 | India | 75. | MT510697 | Africa |
| 36. | MT635410 | India | 76. | MT510700 | Africa |
| 37. | MT635856 | India | 77. | MT510703 | Africa |
| 38. | MT509651 | India | 78. | MT510694 | Africa |
| 39. | MT509657 | India | 79. | MT510695 | Africa |
| 40. | MT509504 | India | 80. | MT510698 | Africa |



Figure 2: Ramachandran plot of the modelled protein. A) Wild type S-protein of SARS-CoV-2 B) D614G mutant of S-protein C) G476S mutant of S-protein D) V483A mutant of S-protein

Table 2:

The interacting amino acids of wild type S-protein and mutants are tabulated along with the human ACE2 receptor amino acids.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Wild type | G476S | V483A | D614G | ACE2 |
| SER459 | ARG346THR345 | ASP482LEU517HIS519LEU518 | LYS458SER459ASN460 | TYR65 |
| TRP353PRO426LEU425LYS424ARG355 | THR478PRO479SER477 | TYR351ALA352 | TRP353ASN422TYR423LYS424 | ASN46 |
| LYS462PHE464CYS488TYR489SER459ARG457ASN487PRO491PRO463PHE490 | TYR451ALA348ASN450SER349LEU452ASN354PHE347ARG346 | LYS424 | PRO463TYR489ASN460LYS462LEU461PHE464SER459 | LYS13 |
| VAL483GLU484GLN474GLY482ASN481GLY485ASN487 | LEU492SER469PRO491TYR351ARG454PHE490 | ARG454 | GLY485ASN481PHE486GLY482ASN487GLN474VAL483GLU484 | ASP20 |
| THR478PRO479 | ILE472TYR473 | THR478GLU484GLY476PRO479SER477 | PRO479THR478 | ASN312 |
| ARG454GLY485GLU484 | LEU452PHE490LEU492 | PHE490ARG454 | ARG457ARG454GLU484 | LEU21 |
| GLU484VAL483GLY482GLN474 | ALA475PRO491ILE468ARG454SER476ILE472PHE490SER469 | LYS458TYR489PHE490ARG454PRO491ASN481 | ALA474GLY476GLN474GLU484 | GLN24 |
| ASN481GLY482CYS480PRO479GLN474 | THR470SER469ILE468ILE472 | CYS480THR478 | GLN474ASN481GLY482PRO479CYS480 | TYR23 |
| PRO479THR478ALA475SER477CYS480GLY476GLN474 | GLN474TYR473SER476ALA475ILE472 | CYS480TYR489ASN481THR478GLY482 | CYS480ALA475THR478SER477GLN474GLY476 | LEU27 |
| ARG454TYR421ARG457 | LEU452ASN450TYR451 | TRP353LYS424 | TYR421ARG454ARG457 | PHE54 |
| ASN487ARG457ARG454TYR489PRO491PHE490 | SER349TYR351LEU452ASN450ALA352LEU492TYR351 | LYS424 | TYR489ASN487PHE490PRO491ARG454ARG457 | GLU17 |
| PHE490PRO463PRO491ASN487ARG457SER459TYR489CYS488PHE464LYS462 | ARG346PHE347ASN354LEU452SER349ASN450ALA348TYR451 | LYS424 | SER459SER464LEU461LYS462ASN460TYR489PRO463 | LYS13 |
| ARG457TYR421LEU455ARG454PHE456 | ASN448GLY446ASN450TYR449GLY447 | TYR423LEU425LYS424ARG355TRP353PRO426SER514ASP398 | PHE456LEU455TYR421ARG454ARG457 | GLU57 |
| PRO479THR478 | GLN474TYR473ILE472 | SER377THR478 | THR478 | ARG339 |
| SER477THR478ALA475GLY476 | VAL483CYS480ALA475SER476GLN474 | TYR489PHE486GLU484GLY485THR478ALA483SER477 | GLY476SER477ALA475 | ASN31 |
| ALA475TRP353ARG355 | CYS480SER476PRO479THR478VAL483SER477 | TYR489ASN487PHE490LEU492CYS488 | TRP353ALA475ARG355 | ASN43 |
| PRO479ASN481 | ILE472 | CYS480THR478 | ASN481 | ASP337 |
| GLU516ASN394ARG357TYR396 | CYS480PRO479ASN481 | TYR489ASN487 | HIS519TYR396GLU516ARG357 | GLU39 |
| ASP420LYS424TYR351TYR421ASN422GLU484 | LEU492SER494 | ARG454TRP353TYR351ALA352TYR421ASN422VAL350 | GLY485TRP353TYR421ARG454ASP420TYR351GLU484ASN422 | LYS50 |
| SER459ASN460 | ARG346THR345 | PRO426 | SER459ASN460 | PHE10 |
| ARG457PHE456 | ASN450ARG346 | PHE429PHE515ASP428PRO426 | SER459ARG457PHE456LYS458 | LEU61 |
| TYR421ASP420LYS424 | TYR449 | ARG355TRP353 | TYR421LYS424ASP420 | ALA53 |
| ARG457 | ASN450 | LYS424ARG355PRO426 | ARG457LYS458SER459 | GLN58 |