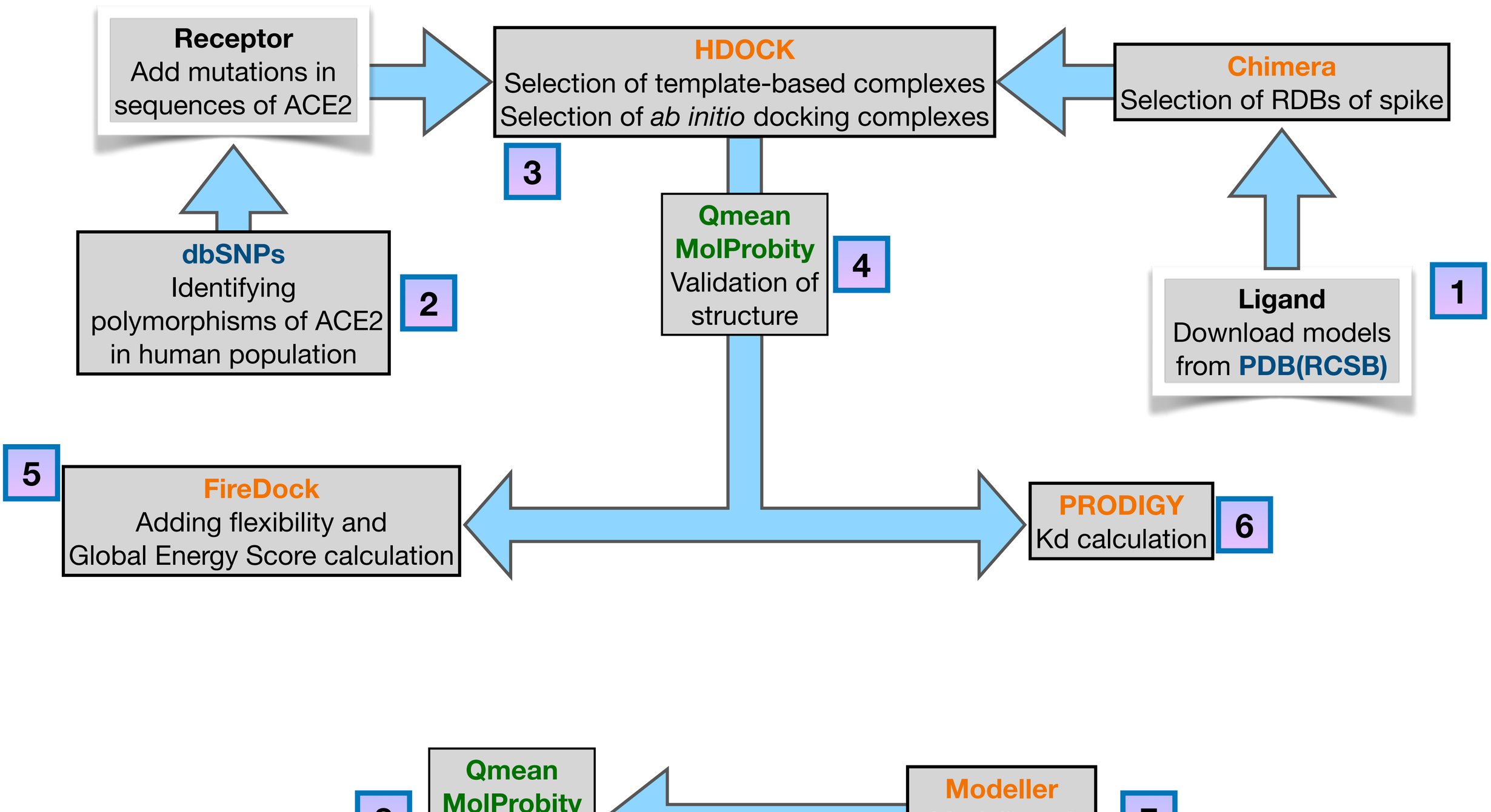

ACE2 missense variant - protocol

1. 6LZG, 6M0J and 6M17 models were downloaded from PDB (RCSB). RDBs were selected with Chimera and saved in a separate .pdb file.
2. The amino acid residues of the wild type ACE2 sequence were replaced with those of the missense variant from dbSNP database
3. ACE2/RDB complexes were assembled by HDOCK.
4. Qmean and MolProbity were used to validated HDOCK complexes.
5. Complexes were then analyzed by FireDock for adding flexibility and global energy score calculation.
6. PRODIGY was used to calculate the dissociation constants of ACE2/RBD complexes.
7. MODELLER 9.25 was used in parallel to confirm HDOCK models.
8. Qmean and MolProbity were used to validated MODELLER 9.25 complexes.
9. SwarmDock was used to perform a docking (receptor models of step 7; ligand the pdb files of step 1).

Step 3, 4 and 5 were repeated in order to calculate the binding affinity between ACE2 and RBD variants. ACE2 sequences were used as receptor while and asRDB variants sequences were used as ligands.