**Samples answers for student progress check questions included in red in the student worksheet:**

**STRUCTURE OF THE SPIKE PROTEIN:**

How many protein chains do you see? Based on your answer how can you characterize this protein (i.e. what is its quaternary structure?) Hint: Each chain is depicted with a different color.

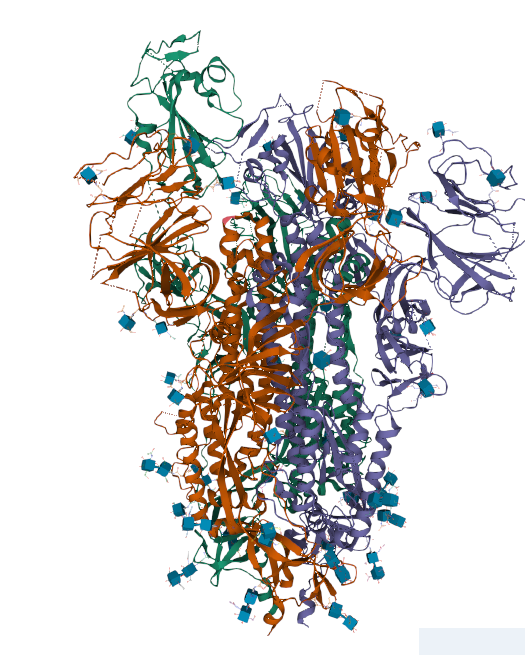
3

What kinds of secondary structures do you see?

Mainly alpha helix

What do the blue cubes represent in the structure? Remember when you hover your mouse over it you can read off the element from the lower right hand of the window.

Various carbohydrate molecules

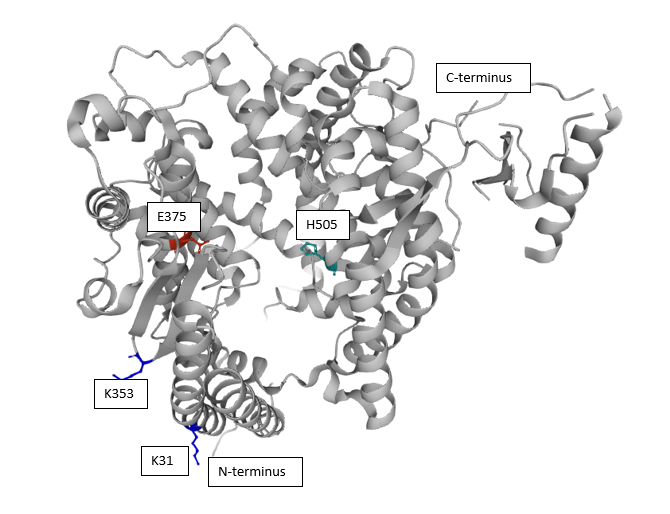
Where are the N- and C-termini of the chains? Orient the structure so that the C-termini of the protein chains are at the bottom of the page.

You should now be seeing only the RBD of the spike protein on your screen.

**STRUCTURE OF THE ACE2 RECEPTOR:**

Open the structure in the 3D Viewer and identify these residues on the ACE2 structure by displaying them in their ball and stick representations on the ACE2 structure. Also note the N- and C- termini. Using the residues for spike protein binding, identify the face of the ACE2 protein that is expected to be in contact with the Spike protein’s receptor binding domain.

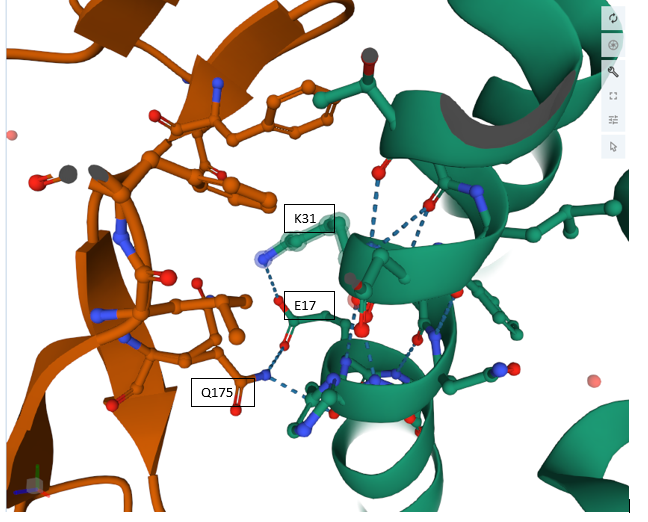
This view presents all the side chains in an accessible way to bind the RBD of the spike protein.



**STRUCTURE OF SARS CoV-2: ACE2 complex**

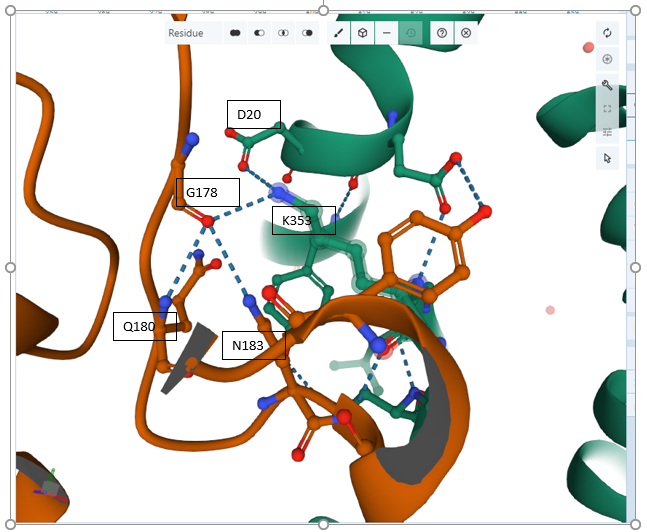
Examine this structure and note down the non-covalent interactions the residues you identified on the ACE2 protein make with residues within a 5A radius. Pay special attention to any interaction at the interface between SARS CoV-2 and ACE2 and any salt bridges that might be functionally important. Note that the two proteins are shown in different colors in the default view.

Focusing on K31 (at 5A radius)



Focusing on K353 (at 5A radius)

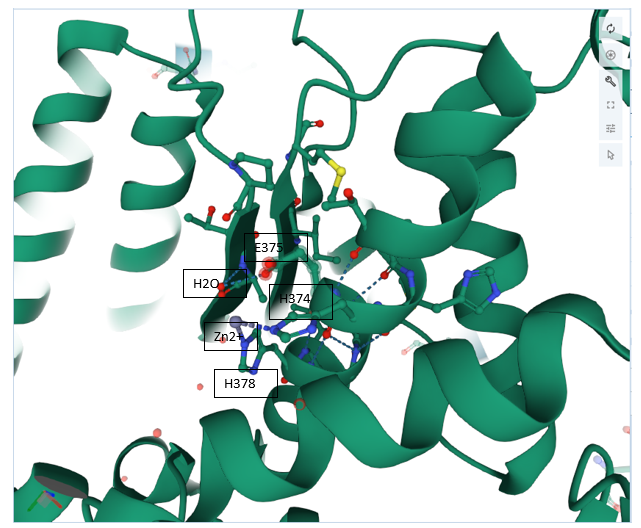
Q175



Q175

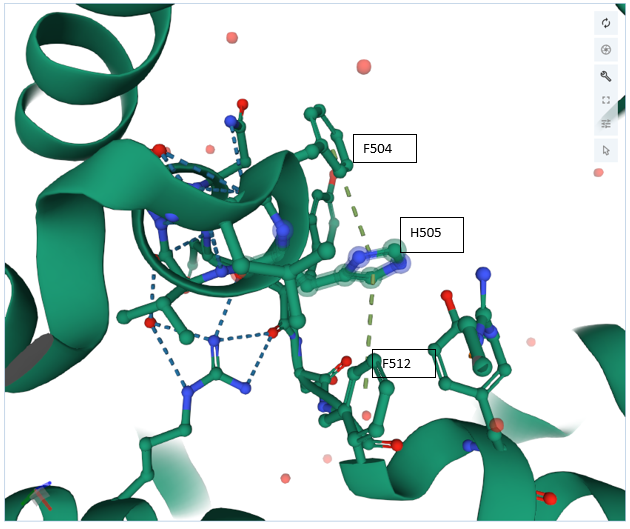
Focusing on E375 (at 5A radius)

Note that based on the structural analysis this active site residue is NOT located at the binding interface and there seems to be a critical network of non-covalent interactions that are facilitated by small ligands (water and a zinc ion) between E375, H374 and H378.



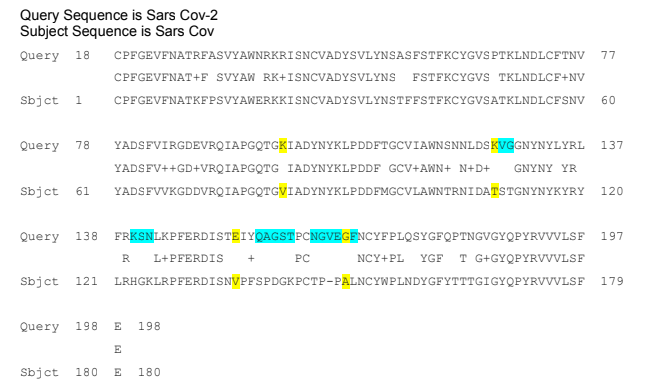
Focusing on H505 (at 5A radius)

Note the less common pi-stacking interactions you observe for this residue in the active site



**How novel is SARS CoV-2?**

Examine the results page and click on the alignment tab.



## CONNECTING GENETIC VARIATION AND PROTEIN STRUCTURE

## Make a table that lists the key amino acid mutations for each of these four variants of concern and the reason for concern.

|  |  |  |  |
| --- | --- | --- | --- |
| Variant Name | Phenotypic Findings | Identified aa mutations | Effect of mutation |
| Alpha | 30-50% more infectious, more deadly, but vaccines still work well | N501Y | helps the virus latch on more tightly to human cells |
| P681H | help infected cells create new spike proteins more efficiently. |
| **H69–V70** | alter the shape of the spike and may help it evade some antibodies |
| **Y144/145** deletions |
| Beta | clinical trials of vaccines are showing that they offer less protection against this strain | N501Y | helps the virus latch on more tightly to human cells |
| **K417N** | helps the virus bind more tightly to human cells. |
| **E484K** | help the virus evade some kinds of antibodies. |
| Gamma | It may be able to overcome the immunity developed after infection by other variants. | N501Y | helps the virus latch on more tightly to human cells |
| **K417T** | helps the virus bind more tightly to human cells. |
| **E484K** | help the virus evade some kinds of antibodies. |
| Delta/Kappa | carries more than a dozen mutations, but is sometimes called a “double mutant” because of two prominent mutations   It has emerged as a fast-growing virus, outpacing other variants of concern. | **E484Q** | help the virus evade some kinds of antibodies. |
| **L452R** |  |

Then locate these residues on the SARS-Cov2 Spike: ACE2 complex you explored above (PDB ID 6M0J). You can also look for these residues using PDB ID 6M17 or on binding of the variant spike protein to one specific neutralizing antibody shown in the PDB ID 7K9Z. Based on the location and characteristics of these mutations make a prediction about their possible functional effect and compare your predictions with the concerns you have listed in your table.