Studying ACE2 orthologs in various species to determine binding interactions to 2019-nCoV

Madeleine King, Maya Paniagua, Karina Vescio, and Lizzy Urbina

BIOL368/S20
Department of Biology
Loyola Marymount University
30 April 2020

Outline

- Wan et al. (2020) used predictive framework to develop a model for 2019-nCoV.
- Wan et al. (2020) found five important amino acid residues in ACE2.
- Can 2019-nCoV bind to certain species ACE2 receptor?
- Species that have ACE2 receptor were obtained from Uniprot.
- Phylogenetic tree was made to determine relatedness between species.
- iCn3D viewer was used to find ACE2 binding sequence.
- Related species were compared to ACE2 α helix of human and mouse.
- Different residues were analyzed for structure-function relationships.
- We predict that bat, orangutan, and horse should all be able to bind to 2019-nCoV due to residue analysis.
- Future research is needed to determine sequence-structure-function relationship of ACE2 orthologs to 2019-nCoV.

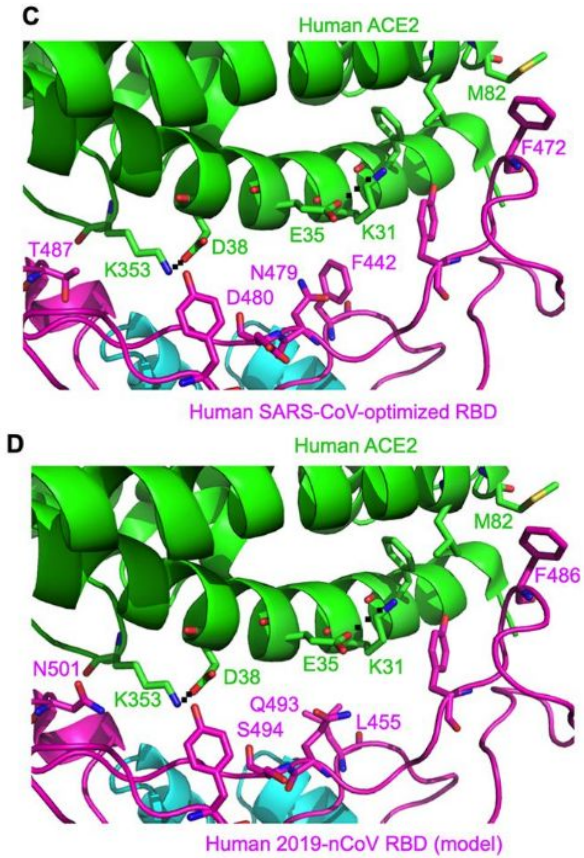
Outline

- **Wan et al. (2020) used predictive framework to develop a model for 2019-nCoV.**
- **Wan et al. (2020) found five important amino acid residues in ACE2.**
- **Can 2019-nCoV bind to certain species ACE2 receptor?**
- **Species that have ACE2 receptor were obtained from Uniprot.**
- **Phylogenetic tree was made to determine relatedness between species.**
- **iCn3D viewer was used to find ACE2 binding sequence.**
- **Related species were compared to ACE2 α helix of human and mouse.**
- **Different residues were analyzed for structure-function relationships.**
- **We predict that bat, orangutan, and horse should all be able to bind to 2019-nCoV due to residue analysis.**
- **Future research is needed to determine sequence-structure-function relationship of ACE2 orthologs to 2019-nCoV.**

Wan et al. (2020) used predictive framework to develop a model for 2019-nCoV

- **SARS-CoV and 2019-nCoV share 77% sequence similarities, which suggests they may bind to the same receptor, ACE2.**

- **SARS-CoV and 2019-nCoV are similar in structure and binding.**



Wan et al. (2020) found five important amino acid residues in ACE2 receptor orthologs

- **Changes in these residues can either enhance or inhibit 2019-nCoV binding to ACE2.**

- **Mouse and Rat are the two predicted species that 2019-nCoV could not bind to.**

A

ACE2	31	35	38	82	353
Human	K	E	D	M	K
Civet	T	E	E	T	K
Bat	K	K	D	N	K
Mouse	N	E	D	S	H
Rat	K	E	D	N	H
Pig	K	E	D	T	K
Ferret	K	E	E	T	K
Cat	K	E	E	T	K
Orangutan	K	E	D	M	K
Monkey	K	E	D	M	K

The Main Question

Can 2019-nCoV bind to certain species ACE2 receptor?

Outline

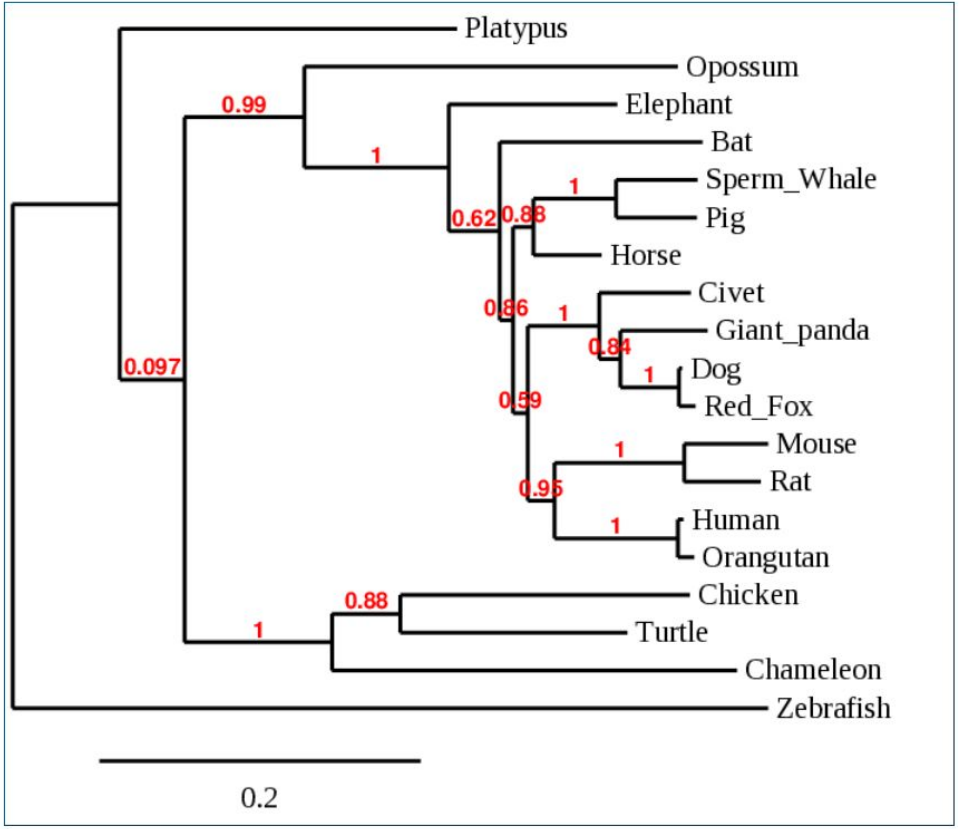
- Wan et al. (2020) used predictive framework to develop a model for 2019-nCoV.
- Wan et al. (2020) found five important amino acid residues in ACE2.
- Can 2019-nCoV bind to certain species ACE2 receptor?
- **Species that have ACE2 receptor were obtained from Uniprot.**
- **Phylogenetic tree was made to determine relatedness between species.**
- **iCn3D viewer was used to find ACE2 binding sequence.**
- **Related species were compared to ACE2 α helix of human and mouse.**
- Different residues were analyzed for structure-function relationships.
- We predict that bat, orangutan, and horse should all be able to bind to 2019-nCoV due to residue analysis.
- Future research is needed to determine sequence-structure-function relationship of ACE2 orthologs to 2019-nCoV.

Species with ACE2 receptor were discovered using Uniprot

- ***ACE2*** **was entered into Uniprot to find orthologs in different animal species.**
- **19 species were obtained to compare conservation of ACE2**
  - **Includes mammals, reptiles, birds, fish, etc.**

	Angiotensin-converting enzyme 2	**ACE2**, UNQ868/PRO1885	Homo sapiens (Human)	805
	Angiotensin-converting enzyme 2	**Ace2**	Mus musculus (Mouse)	805
	Angiotensin-converting enzyme 2	**Ace2**	Rattus norvegicus (Rat)	805
	Angiotensin-converting enzyme 2	**ACE2**	Paguma larvata (Masked palm civet)	805

No pattern was observed in phylogeny using the entire sequence of ACE2 receptor



iCn3D was used to find the sequence of the ACE2 α helix that 2019-nCoV binds to

- **Wan et al. (2020) PDB ID was inputted into iCn3D viewer to determine where 2019-nCoV binds to ACE2.**

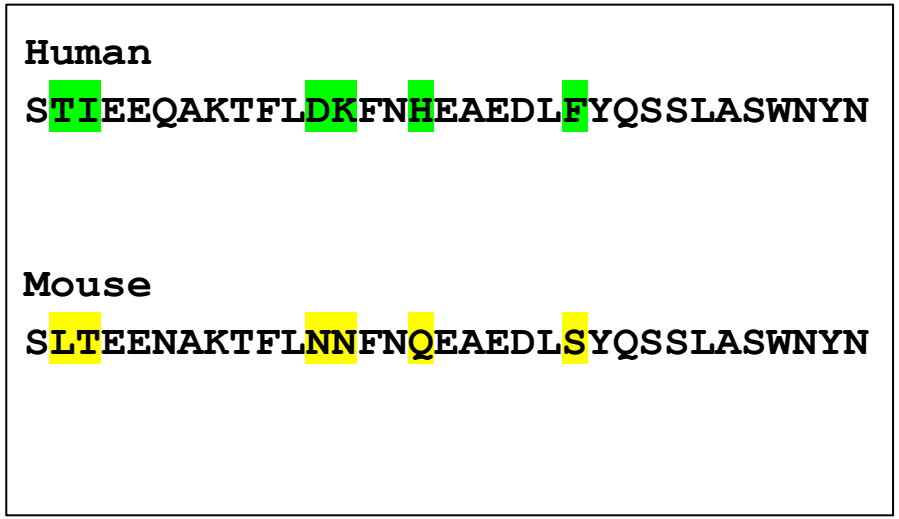
- **α helix sequence:**
  - **STIEEQAKTFLDKFNHEAEDLF YQSSLASWNYN**

Pink: ACE2
Tan: 2019-nCov

Residue differences in Mouse ACE2 α helix could account for differences in binding

- Binding helices of human and mouse species were compared for similarity.

- 6 residues were found that could cause differences in binding.



Outline

- Wan et al. (2020) used predictive framework to develop a model for 2019-nCoV.
- Wan et al. (2020) found five important amino acid residues in ACE2.
- Can 2019-nCoV bind to certain species ACE2 receptor?
- Species that have ACE2 receptor were obtained from Uniprot.
- Phylogenetic tree was made to determine relatedness between species.
- iCn3D viewer was used to find ACE2 binding sequence.
- **Related species were compared to ACE2 α helix of human and mouse.**
- **Different residues were analyzed for structure-function relationships.**
- We predict that bat, orangutan, and horse should all be able to bind to 2019-nCoV due to residue analysis.
- Future research is needed to determine sequence-structure-function relationship of ACE2 orthologs to 2019-nCoV.

Five animals were chosen to compare ACE2 α helix sequence to human

- **Animals with close relation to Wan et al. (2020) species were chosen from phylogenetic tree.**
- **Focused on 6 residues which differed between mouse and human.**
  - **Positions 2, 3, 12, 13, 16, and 22**

Animal	Sequence
Horse	STTEDLAKTFLEKFNSE AEELSHQSSLASWSYN
Platypus	K-PEEEARQFLTQFNKQ AEDLSYQSSLASWEYN
Civet	STTEELAKTFLETFNYE AQELSYQSSVASWNYN
Bat	STTEDEAKMFLDKFNTK AEDLSHQSSLASWDYN
Orangutan	STIEEQAKTFLDKFNHE AEDLFYQSSLASWNYN

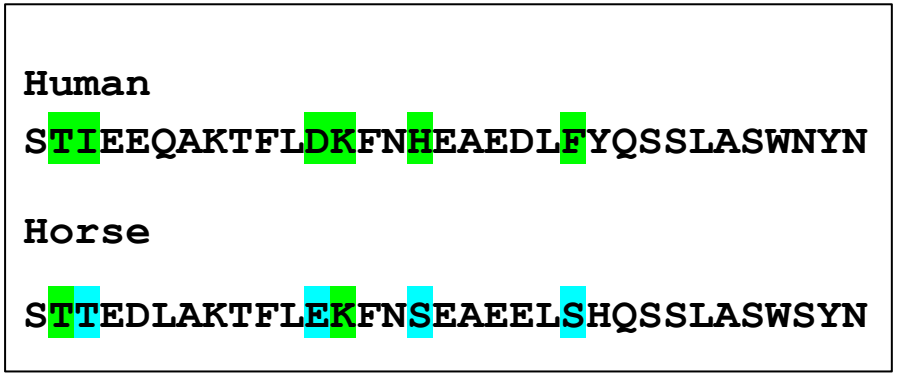
Some residues are conserved in Horse

I→ T= nonpolar to polar

D→ E= both - charged

H→ S= + charged to polar

F→ S= nonpolar to polar



Platypus showed differences in every residue

T→ - = loss of polar A.A.

I → P = both hydrophobic, change of shape

D→T = - charged to uncharged

K→ Q = + charged to uncharged

H→ K = both + charged, change of shape

F→ S = hydrophobic to polar

Only one residue was preserved in Civet

I→ T = nonpolar to polar

D→ E = both - charged

K→ T = + charge to polar

H→ Y = + charge to polar

F→ S = nonpolar to polar

Half of the residues are conserved in Bat

I→ T = nonpolar to polar

H→ T = + charged to polar

F→ S = nonpolar to polar

Entire α helix is conserved in Orangutan

ACE2 orthologs are both conserved and varied

ACE2	2	3	12	13	16	22
Human	T	I	D	K	H	F
Horse	T	T	E	K	S	S
Platypus	-	P	T	Q	K	S
Civet	T	T	E	T	Y	S
Bat	T	T	D	K	T	S
Orangutan	T	I	D	K	H	F

Outline

- Wan et al. (2020) used predictive framework to develop a model for 2019-nCoV.
- Wan et al. (2020) found five important amino acid residues in ACE2.
- Can 2019-nCoV bind to certain species ACE2 receptor?
- Species that have ACE2 receptor were obtained from Uniprot.
- Phylogenetic tree was made to determine relatedness between species.
- iCn3D viewer was used to find ACE2 binding sequence.
- Related species were compared to ACE2 α helix of human and mouse.
- Different residues were analyzed for structure-function relationships.
- **We predict that bat, orangutan, and horse should all be able to bind to 2019-nCoV due to residue analysis.**
- **Future research is needed to determine sequence-structure-function relationship of ACE2 orthologs to 2019-nCoV.**

Can 2019-nCoV bind to these species?

- **Our established criteria assumed that 3 or more conserved residues most likely allows 2019-nCoV to bind to the ACE2 receptor.**

Species	Conserved Residues	Able to bind to receptor
Horse	3	Yes
Platypus	None	No
Civet	1	No
Bat	3	Yes
Orangutan	All	Yes

Position 13 is important for binding of SARS-CoV

- **Wan et al. (2020) found that position 13 is a hotspot for binding interactions**
  - **Lys forms a salt bridge with a neighboring Glu ⇒ leads to favorable interactions**

- **Residue differences in position 13 agree with our predictions**

ACE2	13
Human	K
Horse	K
Platypus	Q
Civet	T
Bat	K
Orangutan	K

Discussion

- **Horses can be infected with Equine Coronavirus which also support our predictions.**
- **SARS-CoV-1 was able to bind to civets' ACE2 receptor.**
  - **However, Wan et al. predicted that civet receptor does not have favorable interactions with 2019-nCoV.**
  - **Cases of other cat species infected with the novel virus are known.**
- **Orangutans and Humans share 97% of DNA with humans.**
- **Predictions could change due to adaptations of the virus to bind to host.**

Future Research

- **Study how certain species can block SARS-CoV-2 using their ACE2 receptor.**
  - **Could be useful in developing a mechanism to treat species that can be infected.**

- **Analyze why SARS-CoV-1 was able to mutate and bind to the ACE2 receptor in civets, but not SARS-CoV-2.**

- **Analyze whether residues in α helix are important hotspots for 2019-nCoV binding.**

Summary

- Wan et al. (2020) used predictive framework to develop a model for 2019-nCoV.
- Wan et al. (2020) found five important amino acid residues in ACE2.
- Can 2019-nCoV bind to certain species ACE2 receptor?
- Species that have ACE2 receptor were obtained from Uniprot.
- Phylogenetic tree was made to determine relatedness between species.
- iCn3D viewer was used to find ACE2 binding sequence.
- Related species were compared to ACE2 α helix of human and mouse.
- Different residues were analyzed for structure-function relationships.
- We predict that bat, orangutan, and horse should all be able to bind to 2019-nCoV due to residue analysis.
- Future research is needed to determine sequence-structure-function relationship of ACE2 orthologs to 2019-nCoV.

Acknowledgements

We would like to thank:

Wan et al. (2020)

Dr. Dahlquist

BIOL368/S20

LMU Biology Department

References

*Home - ORFfinder - NCBI. (n.d.). Retrieved April 22, 2020, from https://www.ncbi.nlm.nih.gov/orffinder/

*NCBI Protein Domains and Macromolecular Structures. (n.d.). Retrieved April 27, 2020, from https://www.ncbi.nlm.nih.gov/Structure/index.shtml

*Phylogeny.fr: Home. (n.d.). Retrieved April 27, 2020, from http://www.phylogeny.fr/

*OpenWetWare. (2020). BIOL368/S20:Week 14. Retrieved April 27 2020, from https://openwetware.org/wiki/BIOL368/S20:Week_14

*RCSB Protein Data Bank. (n.d.). Homepage. Retrieved April 27, 2020, from https://www.rcsb.org/

*UniProt Consortium European Bioinformatics Institute Protein Information Resource SIB Swiss Institute of Bioinformatics. (n.d.). UniProt Consortium. Retrieved April 27, 2020, from http://www.uniprot.org/

*Wan, Y., Shang, J., Graham, R., Baric, R. S., & Li, F. (2020). Receptor recognition by the novel coronavirus from Wuhan: an analysis based on decade-long structural studies of SARS coronavirus. Journal of virology, 94(7). DOI: 10.1128/JVI.00127-20.