Small Animal Model ACE2 and Antibody Response Comparison

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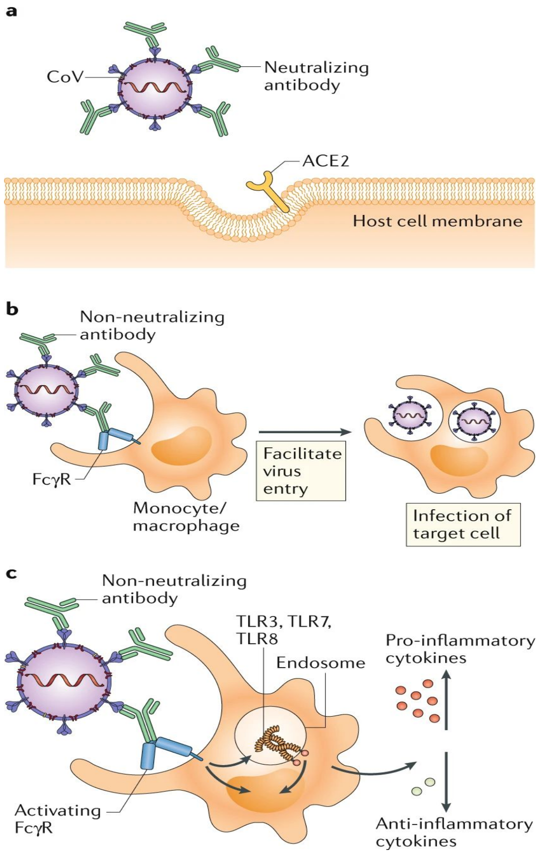
Outline

- Efficacy of RBD nAb in vivo in Syrian hamsters shows promise for human studies
- Syrian hamsters as a model for studying respiratory diseases in humans
- Small animal model ACE2 sequences showed varied relationships
- Comparison of ACE2 sequences in humans vs. small animal models
- SARS-CoV-2 Antibody Neutralizing Potencies
- Future neutralizing antibody work using small animal models

Neutralizing antibodies to the cause of SARS-CoV-2 can help guide vaccine design

- Enrolled recovered SARS-CoV-2 patients (Rogers et al, 2020)
- Isolated potent neutralizing antibodies (nAbs)
  - Two epitopes on the RBD and non-RBD epitopes on the spike protein
- Found an nAb to SARS-CoV-2 and demonstrated its efficacy in small animals
  - Likely useful in medical interventions in humans



Mechanism of neutralizing antibodies (Iwasaki and Yang, 2020)

Efficacy of RBD nAb in vivo in Syrian hamsters shows promise for human studies

- The most potent Abs target the RBD-A epitope, which overlaps with the ACE2 binding site (Rogers et al, 2020)
- RBD-A nAbs were able to compete with ACE2
- Vaccine development should focus on the RBD, as there are strong nAb responses visible (Rogers et al, 2020)
- Multiple small animal model studies on SARS-CoV-2 found promising nAbs

How similar are the ACE2 receptors for SARS-CoV-2 within small-animal models compared to humans with reference to neutralizing antibody activity?

- Small animal models used for biomedical research to test therapies
  - Done prior to human clinical trials
- Most commonly used small animal models:
  - Syrian hamsters, mice, rats, guinea pigs, monkeys, rabbits, ferrets

Syrian hamster in the lab (Cohen, 2020)

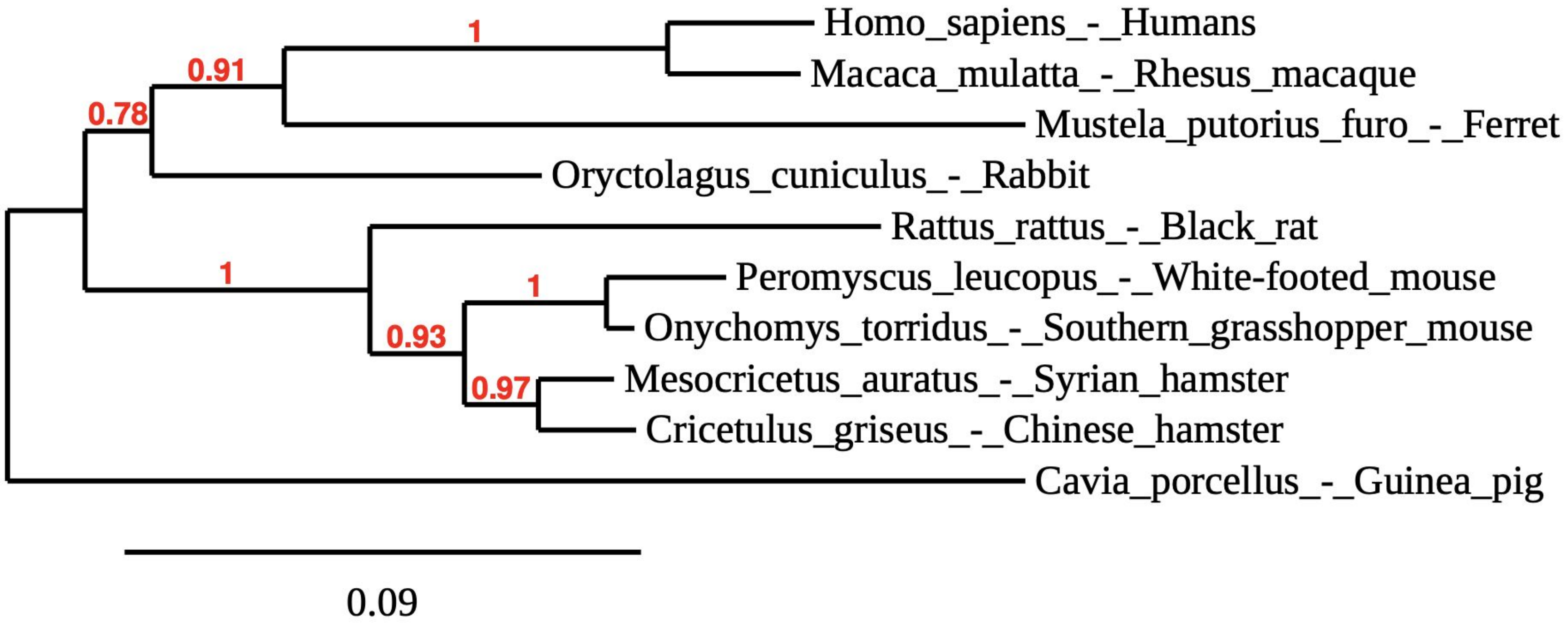
Syrian hamsters as a model for studying diseases in humans

- Our species have similar immune responses to infectious pathogens
- This makes them ideal for studying
  - Pathogenesis of parasitic, viral, and bacterial infections
  - The efficacy and interactions of medications and vaccines
- Comparing our ACE2 sequences is aimed at examining this similarity

Female S. hamster named Vince (Jennison, 2006)

Syrian hamsters as a model... (Maio et al. 2019)

Small animal model ACE2 sequences showed varied relationships

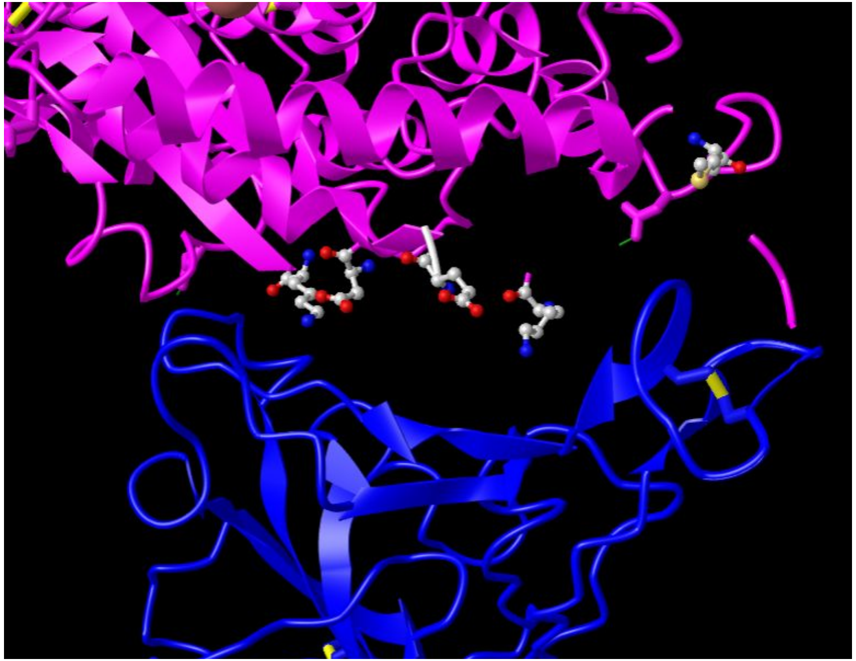


Visualization of Human Critical Residues was Made Using iCn3D Viewer

Here the human ACE2 is in pink and the SARS-CoV-2 spike protein in blue.

This was done by selecting the following residues & displaying them in the ball and stick style with atom colouring.

- K31
- E35
- D38
- M82
- K353



ACE2 percent sequence similarities show wide range of similarity to Syrian hamsters

ACE2 Sequence Similarities	Domestic Guinea Pig	Black Rat	Domestic Ferret	Human	Rhesus monkey	Rabbit	White-Footed Mouse	Southern Grasshopper Mouse	Chinese Hamster
Syrian Hamster	92.4	94.2	94.5	95.8	95.8	96	98.4	98.8	99.4

- Most commonly used small animal models show close similarity
- Humans and rhesus monkeys are the most similar sequences

Comparison of ACE2 critical residues

ACE2 Critical Residue	Domestic Guinea Pig	Domestic Ferret	Human	Rhesus Monkey	Rabbit	Black Rat	White-Footed Mouse	Southern Grasshopper Mouse	Syrian Hamster	Chinese Hamster
31	K	K	K	K	K	E	K	K	K	K
35	K	E	E	E	E	E	E	E	E	E
38	D	E	D	D	D	D	D	D	D	D
82	A	T	M	M	T	N	N	N	N	N
353	K	K	K	K	K	H	K	K	K	K

Comparison of ACE2 sequences in humans vs. small animal models

	Description	Common Name	Max Score	Total Score	Query Cover	E value	Per. Ident	Acc. Len	Accession
☑	angiotensin converting enzyme 2 [Macaca mulatta]	Rhesus monkey	1596	1596	98%	0.0	95.34%	805	ACI04574.1
☑	angiotensin I converting enzyme 2 [Oryctolagus cuniculus]	rabbit	1440	1440	98%	0.0	84.76%	805	QHX39726.1
☑	angiotensin-converting enzyme 2 [Mesocricetus auratus]	golden hamster	1398	1398	98%	0.0	84.26%	805	XP_005074266.1
☑	angiotensin-converting enzyme 2 [Cricetulus griseus]	Chinese hamster	1394	1394	98%	0.0	84.01%	805	XP_027288607.1
☑	angiotensin-converting enzyme 2 [Onychomys torridus]	southern grasshopper mouse	1388	1388	98%	0.0	83.75%	805	XP_036030683.1
☑	angiotensin-converting enzyme 2 [Peromyscus leucopus]	white-footed mouse	1373	1373	98%	0.0	82.87%	805	XP_028743609.1
☑	angiotensin-converting enzyme 2 precursor [Mustela putorius furo]	domestic ferret	1369	1369	97%	0.0	82.74%	805	NP_001297119.1
☑	angiotensin-converting enzyme 2 [Rattus rattus]	black rat	1320	1320	100%	0.0	79.31%	793	XP_032746145.1
☑	LOW QUALITY PROTEIN: angiotensin-converting enzyme 2 [Cavia porcellus]	domestic guinea pig	1285	1285	100%	0.0	77.27%	813	XP_023417808.1

Comparison of ACE2 sequences in humans vs. small animal models

angiotensin-converting enzyme 2 [Macaca nemestrina]

Sequence ID: XP_011733505.1 Length: 805 Number of Matches: 1

See 7 more title(s) See all Identical Proteins(IPG)

Range 1: 12 to 805 GenPept Graphics ▼ Next Match ▲ Previous Match

Score	Expect	Method	Identities	Positives	Gaps
1596 bits(4133)	0.0	Compositional matrix adjust.	757/794(95%)	777/794(97%)	0/794(0%)

```
Query  12   VAVTAAQSTIEEQAKTFLDKFNHEAEDLFYQSSLASWNYNTNITEENVQNMNNAGDKWSA  71
Sbjct  12   .......................................................E....  71

Query  72   FLKEQSTLAQMYPLQEIQNLTVKLQLQALQQNGSSVLSEDKSKRLNTILNTMSTIYSTGK  131
Sbjct  72   ............................................................  131

Query  132  VCNPDNPQECLLLEPGLNEIMANSLDYNERLWAWESWRSEVGKQLRPLYEEYVVLKNEMA  191
Sbjct  132  ....N........D.......EK............G........................  191

Query  192  RANHYEDYGDYWRGDYEVNGVDGYDYSRGQLIEDVEHTFEEIKPLYEHLHAYVRAKLMNA  251
Sbjct  192  .....K....................N.D.......R.......................  251

Query  252  YPSYISPIGCLPAHLLGDMWGRFWTNLYSLTVPFGQKPNIDVTDAMVDQAWDAQRIFKEA  311
Sbjct  252  .......T.......................................N...N........  311

Query  312  EKFFVSVGLPNMTQGFWENSMLTDPGNVQKAVCHPTAWDLGKGDFRILMCTKVTMDDFLT  371
Sbjct  312  ..............................V................I............  371

Query  372  AHHEMGHIQYDMAYAAQPFLLRNGANEGFHEAVGEIMSLSAATPKHLKSIGLLSPDFQED  431
Sbjct  372  ............................................................  431

Query  432  NETEINFLLKQALTIVGTLPFTYMLEKWRWMVFKGEIPKDQWMKKWWEMKREIVGVVEPV  491
Sbjct  432  ............................................................  491

Query  492  PHDETYCDPASLFHVSNDYSFIRYYTRTLYQFQFQEALCQAAKHEGPLHKCDISNSTEAG  551
Sbjct  492  ............................................................  551

Query  552  QKLFNMLRLGKSEPWTLALENVVGAKNMNVRPLLNYFEPLFTWLKDQNKNSFVGWSTDWS  611
Sbjct  552  ...L...K....................................................  611

Query  612  PYADQSIKVRISLKSALGDKAYEWNDNEMYLFRSSVAYAMRQYFLKVKNQMILFGEEDVR  671
Sbjct  612  .........................................T...EI.H.T.........  671

Query  672  VANLKPRISFNFFVTAPKNVSDIIPRTEVEKAIRMSRSRINDAFRLNDNSLEFLGIQPTL  731
Sbjct  672  ..D.........Y.................E...I......................T..  731

Query  732  GPPNQPPVSIWLIVFGVVMGVIVVGIVILIFTGIRDRKKKNKARSGENPYASIDISKGEN  791
Sbjct  732  A..Y.S..TT.............A...V.............Q...E.........N....  791

Query  792  NPGFQNTDDVQTSF  805
Sbjct  792  ..............  805
```

angiotensin I converting enzyme 2 [Oryctolagus cuniculus]

Sequence ID: QHX39726.1 Length: 805 Number of Matches: 1

Range 1: 12 to 805 GenPept Graphics ▼ Next Match ▲ Previous Match

Score	Expect	Method	Identities	Positives	Gaps
1440 bits(3727)	0.0	Compositional matrix adjust.	673/794(85%)	733/794(92%)	0/794(0%)

```
Query  12   VAVTAAQSTIEEQAKTFLDKFNHEAEDLFYQSSLASWNYNTNITEENVQNMNNAGDKWSA  71
Sbjct  12   ............L.....E...Q.....S...A....D...........K..D.EA....  71

Query  72   FLKEQSTLAQMYPLQEIQNLTVKLQLQALQQNGSSVLSEDKSKRLNTILNTMSTIYSTGK  131
Sbjct  72   .YE...K..KT..S..V......R.......S...A..A....Q.....S..........  131

Query  132  VCNPDNPQECLLLEPGLNEIMANSLDYNERLWAWESWRSEVGKQLRPLYEEYVVLKNEMA  191
Sbjct  132  ...QS.....F......D....K.T..........G...V....................  191

Query  192  RANHYEDYGDYWRGDYEVNGVDGYDYSRGQLIEDVEHTFEEIKPLYEHLHAYVRAKLMNA  251
Sbjct  192  ...N.........A...AE.A.......S...D...R..S.......Q......T...D.  251

Query  252  YPSYISPIGCLPAHLLGDMWGRFWTNLYSLTVPFGQKPNIDVTDAMVDQAWDAQRIFKEA  311
Sbjct  252  ...R...T....................................T..N.G...E......  311

Query  312  EKFFVSVGLPNMTQGFWENSMLTDPGNVQKAVCHPTAWDLGKGDFRILMCTKVTMDDFLT  371
Sbjct  312  ..........S..H........PES.DGR.V...........R....K........N...  371

Query  372  AHHEMGHIQYDMAYAAQPFLLRNGANEGFHEAVGEIMSLSAATPKHLKSIGLLSPDFQED  431
Sbjct  372  ...............T............................E........PY..H..  431

Query  432  NETEINFLLKQALTIVGTLPFTYMLEKWRWMVFKGEIPKDQWMKKWWEMKREIVGVVEPV  491
Sbjct  432  .......................................E...Q...............M  491

Query  492  PHDETYCDPASLFHVSNDYSFIRYYTRTLYQFQFQEALCQAAKHEGPLHKCDISNSTEAG  551
Sbjct  492  ..........A....A............I.............Q.................  551

Query  552  QKLFNMLRLGKSEPWTLALENVVGAKNMNVRPLLNYFEPLFTWLKDQNKNSFVGWSTDWS  611
Sbjct  552  ...L......R.................D................E..R........E.T  611

Query  612  PYADQSIKVRISLKSALGDKAYEWNDNEMYLFRSSVAYAMRQYFLKVKNQMILFGEEDVR  671
Sbjct  612  ..............T....Q......S..............K..SE....T.........  671

Query  672  VANLKPRISFNFFVTAPKNVSDIIPRTEVEKAIRMSRSRINDAFRLNDNSLEFLGIQPTL  731
Sbjct  672  .SD..............N..N.....N...E..S........I...D......V......  731

Query  732  GPPNQPPVSIWLIVFGVVMGVIVVGIVILIFTGIRDRKKKNKARSGENPYASIDISKGEN  791
Sbjct  732  E..YES..P...V.......M..I...V......K..R.QKQ.KRE....GFV.M.....  791

Query  792  NPGFQNTDDVQTSF  805
Sbjct  792  .S....S..I....  805
```

angiotensin-converting enzyme 2 [Mesocricetus auratus]

Sequence ID: XP_005074266.1 Length: 805 Number of Matches: 1

Range 1: 12 to 805 GenPept Graphics ▼ Next Match ▲ Previous Match

Score	Expect	Method	Identities	Positives	Gaps
1398 bits(3619)	0.0	Compositional matrix adjust.	669/794(84%)	727/794(91%)	0/794(0%)

```
Query  12   VAVTAAQSTIEEQAKTFLDKFNHEAEDLFYQSSLASWNYNTNITEENVQNMNNAGDKWSA  71
Sbjct  12   ....T...I.............Q.....S...A..............A.K..E.AA....  71

Query  72   FLKEQSTLAQMYPLQEIQNLTVKLQLQALQQNGSSVLSEDKSKRLNTILNTMSTIYSTGK  131
Sbjct  72   .YE...K..KN.S...V....I.R.......S...A..A..N.Q................  131

Query  132  VCNPDNPQECLLLEPGLNEIMANSLDYNERLWAWESWRSEVGKQLRPLYEEYVVLKNEMA  191
Sbjct  132  ....K............DD...T.T..........G..A.....................  191

Query  192  RANHYEDYGDYWRGDYEVNGVDGYDYSRGQLIEDVEHTFEEIKPLYEHLHAYVRAKLMNA  251
Sbjct  192  ...N.............AE.A...N.NGN.......R..K.......Q......T....T  251

Query  252  YPSYISPIGCLPAHLLGDMWGRFWTNLYSLTVPFGQKPNIDVTDAMVDQAWDAQRIFKEA  311
Sbjct  252  .......T....................P..................N.G.N.E......  311

Query  312  EKFFVSVGLPNMTQGFWENSMLTDPGNVQKAVCHPTAWDLGKGDFRILMCTKVTMDDFLT  371
Sbjct  312  ..........Y...............DDR.V................K........N...  371

Query  372  AHHEMGHIQYDMAYAAQPFLLRNGANEGFHEAVGEIMSLSAATPKHLKSIGLLSPDFQED  431
Sbjct  372  ...............T............................E........PS.....  431

Query  432  NETEINFLLKQALTIVGTLPFTYMLEKWRWMVFKGEIPKDQWMKKWWEMKREIVGVVEPV  491
Sbjct  432  ...................................D...E...E...............L  491

Query  492  PHDETYCDPASLFHVSNDYSFIRYYTRTLYQFQFQEALCQAAKHEGPLHKCDISNSTEAG  551
Sbjct  492  ..........A.................I...............D...............  551

Query  552  QKLFNMLRLGKSEPWTLALENVVGAKNMNVRPLLNYFEPLFTWLKDQNKNSFVGWSTDWS  611
Sbjct  552  ...L.....................R..D...........SV...E.........N....  611

Query  612  PYADQSIKVRISLKSALGDKAYEWNDNEMYLFRSSVAYAMRQYFLKVKNQMILFGEEDVR  671
Sbjct  612  ..................EN....D........A.......V..A.N.T.TVP..V..I.  671

Query  672  VANLKPRISFNFFVTAPKNVSDIIPRTEVEKAIRMSRSRINDAFRLNDNSLEFLGIQPTL  731
Sbjct  672  .SD....V.......S.Q........N...E.V.L..G....V.G.D.........N...  731

Query  732  GPPNQPPVSIWLIVFGVVMGVIVVGIVILIFTGIRDRKKKNKARSGENPYASIDISKGEN  791
Sbjct  732  S..Y....T....I......IV....I.......KG.....ETKRE....D.V..G...S  791

Query  792  NPGFQNTDDVQTSF  805
Sbjct  792  .A..LSN..A....  805
```

Comparison of ACE2 sequences in humans vs. small animal models

angiotensin-converting enzyme 2 [Cricetulus griseus]

Sequence ID: XP_027288607.1 Length: 805 Number of Matches: 1

See 1 more title(s) See all Identical Proteins(IPG)

Range 1: 12 to 805 GenPept Graphics ▼ Next Match ▲ Previous Match

Score	Expect	Method	Identities	Positives	Gaps
1394 bits(3609)	0.0	Compositional matrix adjust.	667/794(84%)	726/794(91%)	0/794(0%)

```
Query  12   VAVTAAQSTIEEQAKTFLDKFNHEAEDLFYQSSLASWNYNTNITEENVQNMNNAGDKWSA  71
Sbjct  12   ....T...I.............Q.....S...A..............A.K..E.AA....  71

Query  72   FLKEQSTLAQMYPLQEIQNLTVKLQLQALQQNGSSVLSEDKSKRLNTILNTMSTIYSTGK  131
Sbjct  72   .YE...K..KN.S...V...II.R.......S...A..A..N.Q................  131

Query  132  VCNPDNPQECLLLEPGLNEIMANSLDYNERLWAWESWRSEVGKQLRPLYEEYVVLKNEMA  191
Sbjct  132  ....K............DD...T.T..........G..A.....................  191

Query  192  RANHYEDYGDYWRGDYEVNGVDGYDYSRGQLIEDVEHTFEEIKPLYEHLHAYVRAKLMNA  251
Sbjct  192  ...N.K...........AE.A...N.NGN.......R..K.......Q......T...DT  251

Query  252  YPSYISPIGCLPAHLLGDMWGRFWTNLYSLTVPFGQKPNIDVTDAMVDQAWDAQRIFKEA  311
Sbjct  252  ...F...T....................P..................N.G...E......  311

Query  312  EKFFVSVGLPNMTQGFWENSMLTDPGNVQKAVCHPTAWDLGKGDFRILMCTKVTMDDFLT  371
Sbjct  312  ..........H......G........DDR.V................K........N...  371

Query  372  AHHEMGHIQYDMAYAAQPFLLRNGANEGFHEAVGEIMSLSAATPKHLKSIGLLSPDFQED  431
Sbjct  372  ...............T.....................................PSN.H..  431

Query  432  NETEINFLLKQALTIVGTLPFTYMLEKWRWMVFKGEIPKDQWMKKWWEMKREIVGVVEPV  491
Sbjct  432  ...................................D...EK..E...............L  491

Query  492  PHDETYCDPASLFHVSNDYSFIRYYTRTLYQFQFQEALCQAAKHEGPLHKCDISNSTEAG  551
Sbjct  492  ..........A.................I...............D...............  551

Query  552  QKLFNMLRLGKSEPWTLALENVVGAKNMNVRPLLNYFEPLFTWLKDQNKNSFVGWSTDWS  611
Sbjct  552  ...L.....................R..D...........SV...E.........N....  611

Query  612  PYADQSIKVRISLKSALGDKAYEWNDNEMYLFRSSVAYAMRQYFLKVKNQMILFGEEDVR  671
Sbjct  612  ..................EN.............AT......V..A.N.T.TV...V..I.  671

Query  672  VANLKPRISFNFFVTAPKNVSDIIPRTEVEKAIRMSRSRINDAFRLNDNSLEFLGIQPTL  731
Sbjct  672  .SD....V.......S.Q........N...E.V.F..G....V.G.D.........N...  731

Query  732  GPPNQPPVSIWLIVFGVVMGVIVVGIVILIFTGIRDRKKKNKARSGENPYASIDISKGEN  791
Sbjct  732  A..Y....T....I......IV........V....A...N.E.KRE....D.V..G...S  791

Query  792  NPGFQNTDDVQTSF  805
Sbjct  792  .A...SN.......  805
```

angiotensin-converting enzyme 2 [Onychomys torridus]

Sequence ID: XP_036030683.1 Length: 805 Number of Matches: 1

Range 1: 12 to 805 GenPept Graphics ▼ Next Match ▲ Previous Match

Score	Expect	Method	Identities	Positives	Gaps
1388 bits(3593)	0.0	Compositional matrix adjust.	665/794(84%)	719/794(90%)	0/794(0%)

```
Query  12   VAVTAAQSTIEEQAKTFLDKFNHEAEDLFYQSSLASWNYNTNITEENVQNMNNAGDKWSA  71
Sbjct  12   ....T...I.............Q.....S...A..............A.K..E.AA....  71

Query  72   FLKEQSTLAQMYPLQEIQNLTVKLQLQALQQNGSSVLSEDKSKRLNTILNTMSTIYSTGK  131
Sbjct  72   .YE...K..KN.S.......II.R.......S...A..A..N.Q......A.........  131

Query  132  VCNPDNPQECLLLEPGLNEIMANSLDYNERLWAWESWRSEVGKQLRPLYEEYVVLKNEMA  191
Sbjct  132  ..K.K............DV...T.T..........G..A.....................  191

Query  192  RANHYEDYGDYWRGDYEVNGVDGYDYSRGQLIEDVEHTFEEIKPLYEHLHAYVRAKLMNA  251
Sbjct  192  ...N.K........G..AE.AE..N.N.N.......R..Q.......Q......T...DT  251

Query  252  YPSYISPIGCLPAHLLGDMWGRFWTNLYSLTVPFGQKPNIDVTDAMVDQAWDAQRIFKEA  311
Sbjct  252  .....N.T......................................MK.G...E......  311

Query  312  EKFFVSVGLPNMTQGFWENSMLTDPGNVQKAVCHPTAWDLGKGDFRILMCTKVTMDDFLT  371
Sbjct  312  ..........H...........V...DDR.V................K........N...  371

Query  372  AHHEMGHIQYDMAYAAQPFLLRNGANEGFHEAVGEIMSLSAATPKHLKSIGLLSPDFQED  431
Sbjct  372  ...............T............................E........PS..H..  431

Query  432  NETEINFLLKQALTIVGTLPFTYMLEKWRWMVFKGEIPKDQWMKKWWEMKREIVGVVEPV  491
Sbjct  432  S................................T.....E...Q...............L  491

Query  492  PHDETYCDPASLFHVSNDYSFIRYYTRTLYQFQFQEALCQAAKHEGPLHKCDISNSTEAG  551
Sbjct  492  ..........A.................I..............YD...............  551

Query  552  QKLFNMLRLGKSEPWTLALENVVGAKNMNVRPLLNYFEPLFTWLKDQNKNSFVGWSTDWS  611
Sbjct  552  ...L......N..............R..D............A...E.....L...N....  611

Query  612  PYADQSIKVRISLKSALGDKAYEWNDNEMYLFRSSVAYAMRQYFLKVKNQMILFGEEDVR  671
Sbjct  612  ..................KN.....................V..A.N.T.IVP..V..IH  671

Query  672  VANLKPRISFNFFVTAPKNVSDIIPRTEVEKAIRMSRSRINDAFRLNDNSLEFLGIQPTL  731
Sbjct  672  .SD....V.......S.Q........K...D...F..G....V.G.D.........Y...  731

Query  732  GPPNQPPVSIWLIVFGVVMGVIVVGIVILIFTGIRDRKKKNKARSGENPYASIDISKGEN  791
Sbjct  732  A..Y....T....I......IV............KG.....ETKRE....D.M..G...S  791

Query  792  NPGFQNTDDVQTSF  805
Sbjct  792  .A....N.......  805
```

angiotensin-converting enzyme 2 [Peromyscus leucopus]

Sequence ID: XP_028743609.1 Length: 805 Number of Matches: 1

Range 1: 12 to 805 GenPept Graphics ▼ Next Match ▲ Previous Match

Score	Expect	Method	Identities	Positives	Gaps
1373 bits(3554)	0.0	Compositional matrix adjust.	658/794(83%)	712/794(89%)	0/794(0%)

```
Query  12   VAVTAAQSTIEEQAKTFLDKFNHEAEDLFYQSSLASWNYNTNITEENVQNMNNAGDKWSA  71
Sbjct  12   ....T...I......I......Q.....S...A..............A.K..E.SA....  71

Query  72   FLKEQSTLAQMYPLQEIQNLTVKLQLQALQQNGSSVLSEDKSKRLNTILNTMSTIYSTGK  131
Sbjct  72   .YE...K..KN.S....P..II.R.......S...A..A..T.Q......A.........  131

Query  132  VCNPDNPQECLLLEPGLNEIMANSLDYNERLWAWESWRSEVGKQLRPLYEEYVVLKNEMA  191
Sbjct  132  ..K.K............DV...T.T..........G..A.....................  191

Query  192  RANHYEDYGDYWRGDYEVNGVDGYDYSRGQLIEDVEHTFEEIKPLYEHLHAYVRAKLMNA  251
Sbjct  192  ...N.R...........AE.AE..N.N.N.......RI.Q..............T...DT  251

Query  252  YPSYISPIGCLPAHLLGDMWGRFWTNLYSLTVPFGQKPNIDVTDAMVDQAWDAQRIFKEA  311
Sbjct  252  .....N.T....................P.................LK.G...E......  311

Query  312  EKFFVSVGLPNMTQGFWENSMLTDPGNVQKAVCHPTAWDLGKGDFRILMCTKVTMDDFLT  371
Sbjct  312  ......I...P...........V...DDR.V................K...T....N...  371

Query  372  AHHEMGHIQYDMAYAAQPFLLRNGANEGFHEAVGEIMSLSAATPKHLKSIGLLSPDFQED  431
Sbjct  372  ...............T............................E........PS..R..  431

Query  432  NETEINFLLKQALTIVGTLPFTYMLEKWRWMVFKGEIPKDQWMKKWWEMKREIVGVVEPV  491
Sbjct  432  S................................T.....E...Q...............L  491

Query  492  PHDETYCDPASLFHVSNDYSFIRYYTRTLYQFQFQEALCQAAKHEGPLHKCDISNSTEAG  551
Sbjct  492  ..........A.................I...............D...............  551

Query  552  QKLFNMLRLGKSEPWTLALENVVGAKNMNVRPLLNYFEPLFTWLKDQNKNSFVGWSTDWS  611
Sbjct  552  ...L......N..............R..D............A...E.....L...N....  611

Query  612  PYADQSIKVRISLKSALGDKAYEWNDNEMYLFRSSVAYAMRQYFLKVKNQMILFGEEDVR  671
Sbjct  612  ..................KN.....................V..AMN.T.NVP..V..IH  671

Query  672  VANLKPRISFNFFVTAPKNVSDIIPRTEVEKAIRMSRSRINDAFRLNDNSLEFLGIQPTL  731
Sbjct  672  .SD....V.......S.Q........KD..D...F..G....V.G.D.........Y...  731

Query  732  GPPNQPPVSIWLIVFGVVMGVIVVGIVILIFTGIRDRKKKNKARSGENPYASIDISKGEN  791
Sbjct  732  A..Y....T....I..I...IV............KG.....ETKRE....D.M..G...S  791

Query  792  NPGFQNTDDVQTSF  805
Sbjct  792  .A....N..A....  805
```

Comparison of ACE2 sequences in humans vs. small animal models

angiotensin-converting enzyme 2 precursor [Mustela putorius furo]

Sequence ID: NP_001297119.1 Length: 805 Number of Matches: 1

See 3 more title(s) See all Identical Proteins(IPG)

Range 1: 18 to 805 GenPept Graphics ▼ Next Match ▲ Previous

Score	Expect	Method	Identities	Positives	Gaps
1369 bits(3544)	0.0	Compositional matrix adjust.	652/788(83%)	723/788(91%)	0/788(0%)

```
Query  18   QSTIEEQAKTFLDKFNHEAEDLFYQSSLASWNYNTNITEENVQNMNNAGDKWSAFLKEQS  77
Sbjct  18   ...T.DL.....E...Y...E.S..N...........D..I.K..I..A.....YE.E.  77

Query  78   TLAQMYPLQEIQNLTVKLQLQALQQNGSSVLSEDKSKRLNTILNTMSTIYSTGKVCNPDN  137
Sbjct  78   QH.KT...E...DPII.R..R....S......A..RE.......A.........A...N.  137

Query  138  PQECLLLEPGLNEIMANSLDYNERLWAWESWRSEVGKQLRPLYEEYVVLKNEMARANHYE  197
Sbjct  138  ...........DD..E..K.........G.................A.........N..  197

Query  198  DYGDYWRGDYEVNGVDGYDYSRGQLIEDVEHTFEEIKPLYEHLHAYVRAKLMNAYPSYIS  257
Sbjct  198  ...........EEWA...S...N..........TQ.................D....R..  257

Query  258  PIGCLPAHLLGDMWGRFWTNLYSLTVPFGQKPNIDVTDAMVDQAWDAQRIFKEAEKFFVS  317
Sbjct  258  .T...................P.M...R............N.S...R...E...T....  317

Query  318  VGLPNMTQGFWENSMLTDPGNVQKAVCHPTAWDLGKGDFRILMCTKVTMDDFLTAHHEMG  377
Sbjct  318  .......E...Q.....E..DNR.V...........R....K.................  377

Query  378  HIQYDMAYAAQPFLLRNGANEGFHEAVGEIMSLSAATPKHLKSIGLLSPDFQEDNETEIN  437
Sbjct  378  .........E...........................N...N....P...S..S..D..  437

Query  438  FLLKQALTIVGTLPFTYMLEKWRWMVFKGEIPKDQWMKKWWEMKREIVGVVEPVPHDETY  497
Sbjct  438  ..............................E...Q.......D.......L......  497

Query  498  CDPASLFHVSNDYSFIRYYTRTLYQFQFQEALCQAAKHEGPLHKCDISNSTEAGQKLFNM  557
Sbjct  498  ....A....A............I..........I.......Y.......S......HE.  557

Query  558  LRLGKSEPWTLALENVVGAKNMNVRPLLNYFEPLFTWLKDQNKNSFVGWSTDWSPYADQS  617
Sbjct  558  .S..R.K...F...R.....T.D...............E..R......N.........  617

Query  618  IKVRISLKSALGDKAYEWNDNEMYLFRSSVAYAMRQYFLKVKNQMILFGEEDVRVANLKP  677
Sbjct  618  ...........E..........F.Q..I.....E..S.....T.P.VGK....SD...  677

Query  678  RISFNFFVTAPKNVSDIIPRTEVEKAIRMSRSRINDAFRLNDNSLEFLGIQPTLGPPNQP  737
Sbjct  678  ......I..S.E.M......AD..E...K..G........D.............E..Y..  737

Query  738  PVSIWLIVFGVVMGVIVVGIVILIFTGIRDRKKKNKARSGENPYASIDISKGENNPGFQN  797
Sbjct  738  ..T............V....FL...S...N.R.N.Q...E......V.L..........  797

Query  798  TDDVQTSF  805
Sbjct  798  V.......  805
```

angiotensin-converting enzyme 2 [Rattus rattus]

Sequence ID: XP_032746145.1 Length: 793 Number of Matches: 1

Range 1: 1 to 793 GenPept Graphics ▼ Next Match ▲ Previous M

Score	Expect	Method	Identities	Positives	Gaps
1320 bits(3416)	0.0	Compositional matrix adjust.	640/807(79%)	709/807(87%)	16/807(1%)

```
Query  1    MSSSSWLLLSLVAVTAAQSTIEEQAKTFLDKFNHEAEDLFYQSSLASWNYNTNITEENVQ  60
Sbjct  1    ..R.P.........AT...L...K.ES..N...Q.....S..................A.  60

Query  61   NMNNAGDKWSAFLKEQSTLAQMYPLQEIQNLTVKLQLQALQQNGSSVLSEDKSKRLNTIL  120
Sbjct  61   K..E.AA.....YE...KI..NFS.....DA.I.R..K....S...A..P..N.Q.....  120

Query  121  NTMSTIYSTGKVCNPDNPQECLLLEPGLNEIMANSLDYNERLWAWESWRSEVGKQLRPLY  180
Sbjct  121  ..............SM.....FV.....D....T.T...R......G..A..........  180

Query  181  EEYVVLKNEMARANHYEDYGDYWRGDYEVNGVDGYDYSRGQLIEDVEHTFEEIKPLYEHL  240
Sbjct  181  ..............N.............AE..E..N.N.N.......N..K.......Q.  240

Query  241  HAYVRAKLMNAYPSYISPIGCLPAHLLGDMWGRFWTNLYSLTVPFGQKPNIDVTDAMVDQ  300
Sbjct  241  .....T...DV.......T....................P..T..L............N.  300

Query  301  AWDAQRIFKEAEKFFVSVGLPNMTQGFWENSMLTDPGNVQKAVCHPTAWDLGKGDFRILM  360
Sbjct  301  S...E................Q..P...T.....E..DDR.V..........H.....K.  360

Query  361  CTKVTMDDFLTAHHEMGHIQYDMAYAAQPFLLRNGANEGFHEAVGEIMSLSAATPKHLKS  420
Sbjct  361  .......N..................K.................................  420

Query  421  IGLLSPDFQEDNETEINFLLKQALTIVGTLPFTYMLEKWRWMVFKGEIPKDQWMKKWWEM  480
Sbjct  421  ....PSN....D............I...................QDK..RE..TQ.....  480

Query  481  KREIVGVVEPVPHDETYCDPASLFHVSNDYSFIRYYTRTLYQFQFQEALCQAAKHEGPLH  540
Sbjct  481  ..........L...........................I...............D....  540

Query  541  KCDISNSTEAGQKLFNMLRLGKSEPWTLALENVVGAKNMNVRPLLNYFEPLFTWLKDQNK  600
Sbjct  541  ..............L...S..N.G...........SR..D.K......Q...V...E..S  600

Query  601  NSFVGWSTDWSP--YADQSIKVRISLKSALGDKAYEWNDNEMYLFRSSVAYAMRQYFLKV  658
Sbjct  601  ..T...N.....CDFT.CT-------------H...T................E..SRE  646

Query  659  KNQMILFGEEDVRVANLKPRISFNFFVTAPKNVSDIIPRTEVEKAIRMSRSRINDAFRLN  718
Sbjct  647  ...TVP...A..W.SD....V.......S..........S...E......G....I.G..  706

Query  719  DNSLEFLGIQPTLGPPNQPPVSIWLIVFGVVMGVIVVGIVILIFTGIRDRKKKNKARSGE  778
Sbjct  707  .........Y...K..YE...T....I......MV........V...KG.....ETKRE.  766

Query  779  NPYASIDISKGENNPGFQNTDDVQTSF  805
Sbjct  767  ...D.V..G...S.A....S..A....  793
```

LOW QUALITY PROTEIN: angiotensin-converting enzyme 2 [Cavia porcellus]

Sequence ID: XP_023417808.1 Length: 813 Number of Matches: 1

Range 1: 9 to 813 GenPept Graphics ▼ Next Match ▲ Previous Match

Score	Expect	Method	Identities	Positives	Gaps
1285 bits(3326)	0.0	Compositional matrix adjust.	622/805(77%)	704/805(87%)	0/805(0%)

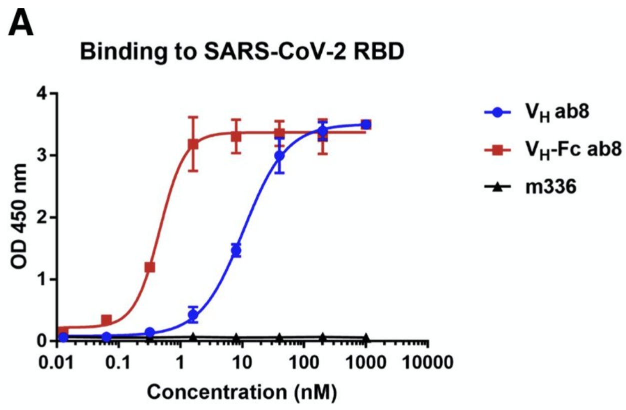
```
Query  1    MSSSSWLLLSLVAVTAAQSTIEEQAKTFLDKFNHEAEDLFYQSSLASWNYNTNITEENVQ  60
Sbjct  9    ..G.F.F..N.....T..FNL.........E..LK....Y...............D....  68

Query  61   NMNNAGDKWSAFLKEQSTLAQMYPLQEIQNLTVKLQLQALQQNGSSVLSEDKSKRLNTIL  120
Sbjct  69   K.SE..GIL...YE...N..KA....D.......R..RI...S...GF.A..N.Q.S...  128

Query  121  NTMSTIYSTGKVCNPDNPQECLLLEPGLNEIMANSLDYNERLWAWESWRSEVGKQLRPLY  180
Sbjct  129  .....L.......Y.SD...........AD..SK.T...L......G...K.........  188

Query  181  EEYVVLKNEMARANHYEDYGDYWRGDYEVNGVDGYDYSRGQLIEDVEHTFEEIKPLYEHL  240
Sbjct  189  ....A.........K.........R....EDM...N...N.......R..A.......Q.  248

Query  241  HAYVRAKLMNAYPSYISPIGCLPAHLLGDMWGRFWTNLYSLTVPFGQKPNIDVTDAMVDQ  300
Sbjct  249  .....T...ET...R...V.................E........Q...........ES.  308

Query  301  AWDAQRIFKEAEKFFVSVGLPNMTQGFWENSMLTDPGNVQKAVCHPTAWDLGKGDFRILM  360
Sbjct  309  S...EK...............P......K.....E..DG..V........M..N....K.  368

Query  361  CTKVTMDDFLTAHHEMGHIQYDMAYAAQPFLLRNGANEGFHEAVGEIMSLSAATPKHLKS  420
Sbjct  369  .......H..................I......D.........I..........E....  428

Query  421  IGLLSPDFQEDNETEINFLLKQALTIVGTLPFTYMLEKWRWMVFKGEIPKDQWMKKWWEM  480
Sbjct  429  ....P...H...G.KX.SS......LL......F....GEG...T..N.QE..IE.X.Q.  488

Query  481  KREIVGVVEPVPHDETYCDPASLFHVSNDYSFIRYYTRTLYQFQFQEALCQAAKHEGPLH  540
Sbjct  489  ..........L...........................I..........K..N.V....  548

Query  541  KCDISNSTEAGQKLFNMLRLGKSEPWTLALENVVGAKNMNVRPLLNYFEPLFTWLKDQNK  600
Sbjct  549  ..............L...K...........SI..T...D.K......Q..S...Q...R  608

Query  601  NSFVGWSTDWSPYADQSIKVRISLKSALGDKAYEWNDNEMYLFRSSVAYAMRQYFLKVKN  660
Sbjct  609  ......N.E....SEE............ED..K.D...............K...D...  668

Query  661  QMILFGEEDVRVANLKPRISFNFFVTAPKNVSDIIPRTEVEKAIRMSRSRINDAFRLNDN  720
Sbjct  669  .TV..SW.....SDWTH.V..T....E.N...N...K....D...L.......V...GXX  728

Query  721  SLEFLGIQPTLGPPNQPPVSIWLIVFGVVMGVIVVGIVILIFTGIRDRKKKNKARSGENP  780
Sbjct  729  HSGVS..Y...S..YE...T...........LV.....V.VI......R..KQKQRE...  788

Query  781  YASIDISKGENNPGFQNTDDVQTSF  805
Sbjct  789  .S.V..G.....TA...SE.N....  813
```

Identifying key epitope regions within SARS-CoV-2

- SARS-CoV genome shares an 80% homology with SARS-CoV-2 (Grifoni et al., 2020)
    - Uses known antibody libraries and/or patient samples to select antibodies
- Most neutralizing antibodies target the RBD-A
    - competes with RBD of SARS-CoV-2
    - VHs may have an advantage for treatment of respiratory virus infections

SARS-CoV-2 Antibody Neutralizing Potencies

- Anti-RBD antibodies are mainly found in the V-gene family
  - Found enriched use of VH3-53 or VH3-66. VH3-53 and VH3-66 V-genes are closely related (Cao et al., 2020)
- Maintains similarities with human germline V-genes
  - VH-Fc ab8 shows complete neutralization at 36 mg/kg dosage in the lungs (Li et al., 2020)
  - STE90-C11 is tolerant to known RBD mutants



Antibody binding to SARS-CoV-2 RBD (Li et al. 2020)

Further Research Questions

- Can further clinical trials be held using small animal models transduced with human ACE2 receptors?
- Should we rely on the prophylactic efficacy of the V-gene antibodies to treat at risk populations?
- Are small animals good models for human immune responses to SARS-CoV-2?
    - dosage
    - Respiratory response

Future neutralizing antibody work using small animal models

- Identify different neutralizing antibody regions that obtained promising results in SARS-CoV-2 studies
    - Rogers et al indicates vaccine development should focus on the RBD
- Compare common antibody regions in small animal models to observe similarities
- Potential studies could identify which small animal models work most efficiently for vaccine development

Summary

- Efficacy of RBD nAb in vivo in Syrian hamsters shows promise for human studies
- Syrian hamsters as a model for studying respiratory diseases in humans
- Small animal model ACE2 sequences showed varied relationships
- Comparison of ACE2 sequences in humans vs. small animal models
- SARS-CoV-2 Antibody Neutralizing Potencies
- Future neutralizing antibody work using small animal models

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Acknowledgements

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BIOL 368 Class