Infectivity of SARS-CoV-2 in pigs (?)

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Do pigs get infected with SARS-CoV-2?

- Pigs must have been exposed to SARS-CoV-2 from infected people (as with mink) but no evidence for SARS-CoV-2 infection in the "field" from around the world - maybe lack of extensive investigation but no signs of disease at least

- Other coronaviruses can infect pigs (e.g. porcine epidemic diarrhoea virus (PEDV), transmissible gastroenteritis virus (TGEV), porcine respiratory coronavirus, porcine deltacoronavirus)- only PRCV detected in DK

- Several experimental studies have described inoculation of pigs with SARS-CoV-2 and examined the outcome

Experimental studies to determine susceptibility of pigs to SARS-CoV-2

At least 5 separate experimental studies have been reported:

1) Shi et al.,(2020) Susceptibility of ferrets, cats, dogs, and other domesticated animals to SARS-coronavirus 2. Science. 368(6494):1016-1020

2) Schlottau K et al. (2020) SARS-CoV-2 in fruit bats, ferrets, pigs, and chickens: an experimental transmission study. Lancet Microbe. 2020 1(5):e218-e225.

3) Meekins DA et al (2020) Susceptibility of swine cells and domestic pigs to SARS-CoV-2, Emerging Microbes & Infections, 9:1, 2278-2288

4) Vergara-Alert J et al. (2020) Pigs are not susceptible to SARS-CoV-2 infection but are a model for viral immunogenicity studies. TBED (epub)

5) Pickering BS et al. (2021) Susceptibility of Domestic Swine to Experimental Infection with Severe Acute Respiratory Syndrome Coronavirus 2. Emerg Inf Dis 27 (1) 104-111.

Experimental design

- 1) Inoculation of pigs with known dose of virus

- 2) Assessment of:

a) infection/replication

b) virus shedding

c) virus transmission

d) clinical signs/pathology

e) seroconversion

- 3) Draw conclusions
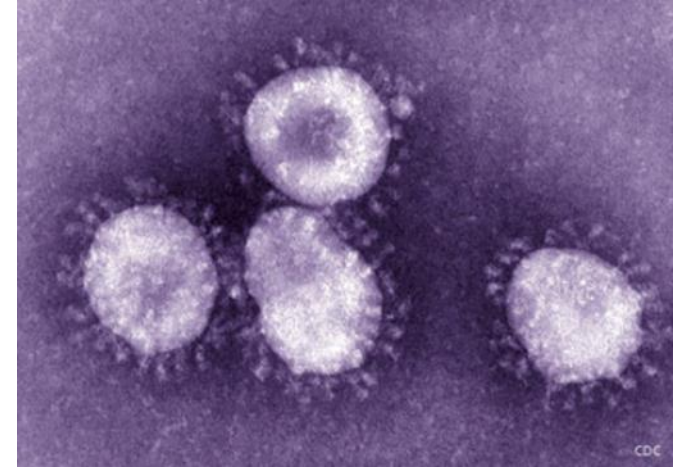
Shi et al.,(2020)

- 5 pigs inoculated intranasally with 10^5 pfu virus

- No viral RNA detectable in oropharyngeal swabs >3 dpi

- No viral RNA in rectal swabs (2-14 dpi)

- No transmission of virus to 3 "in-contact" pigs

- No evidence for seroconversion (14 dpi)

- The same virus was used to inoculate ferrets and cats and evidence of infection and seroconversion was obtained

- Conclusion: No evidence for infection of pigs by SARS-CoV-2

Schlottau et al (2020)

- 9 pigs inoculated intranasally with 10^5 pfu virus

- No viral RNA detected in oropharyngeal swabs 2-21 dpi

- No viral RNA detected in rectal swabs (2-21 dpi)

- No transmission of virus to 3 "in-contact" pigs

- No evidence for seroconversion

- No clinical signs of disease

- Fruit bats and ferrets were shown to be infected in parallel studies

- Conclusion: Pigs were declared not susceptible to infection by SARS-CoV-2

Meekins DA et al., (2020)

- 9 pigs inoculated with 10^6 TCID$_{50}$ virus (oral, intranasal and intratracheal)
- No viral RNA detected in nasal, throat or rectal swabs taken up to 14 dpi
- No transmission of virus to 6 "in-contact" pigs
- No evidence for seroconversion
- No pathological lesions and no detection of viral RNA or proteins in lungs

- Conclusion: Pigs did not become infected with SARS-CoV-2

Vergara-Alert J et al. (2020)

- 4 groups of 5 pigs, inoculated by different routes (intranasal, intratracheal, intramuscular, intravenous) with $10^{5.8}$ TCID$_{50}$

- None of the animals had nasal or rectal shedding of viral RNA beyond 1 dpi

- Low levels of antibodies could be detected in animals given virus IM or IV (like a vaccination) at 14 and 22 dpi but not in others

- Conclusion: SARS-CoV-2 unable to infect pigs by any of these routes

Pickering et al (2021)

- 16 pigs inoculated with 10^6 pfu intranasally

- 2 naïve animals introduced at 10 dpi

- Viral RNA assayed in nasal washings and in oral fluid collected on a "rope" (shared access)-

- At necropsy, various tissue samples collected – analysed by RT-qPCR

- Serology (VNT and surrogate VNT)

Pickering et al. (2021)

Detection of SARS-CoV-2 by RT-qPCR of samples from experimentally inoculated pigs

	Days postinoculation									
Samples	0	3	5	7	9	11	13	15	22	29
Swab samples	0/57	0/57	0/51	0/45	0/39	0/30	0/24	0/18	0/12	0/6
Nasal wash	0/16	**2/16**	0/14	0/12	0/12	0/10	0/8	0/6	0/4	0/2
Blood	0/16	0/16	0/14	0/12	0/12	0/10	0/8	0/6	0/4	0/2

N.B. pigs inoculated intranasally with virus
No infectious virus detected
Oral, nasal and rectal swabs sampled

Detection of SARS-CoV-2 by RT-qPCR in oral fluid samples from inoculated pigs

Pickering et al. EID (2021)

1 positive sample detected (at 3 dpi) from sampling over 29 days

dpi	No. samples tested	No. samples positive
0	2	0
1	2	0
2	2	0
3	2	1
4	2	0
5	2	0
6	2	0
7	2	0
8	2	0
9	2	0
10	2	0
11	2	0
12	2	0
13	2	0
14	2	0
15	2	0
16	2	0
17	2	0

Pickering et al., EID (2021)

- In serum samples from 19 pigs on dpi 7-29, one pig scored weakly +ve by VNT on days 13 and 15 (but then negative)

- In serum samples from 19 pigs on dpi 7-29, one pig scored weakly +ve by surrogate VNT on days 11 and 13 (but then negative) and one pig on day 15 (and then negative)

- Tissue samples (ca. 35 samples (from 2 pigs/day) tested from 3-29 dpi) by RT-qPCR, just one sample scored positive at 13 dpi (submandibular lymph node)- isolated SARS-CoV-2 (after 2 passages) in Vero cells.

- No shedding of infectious virus detected

- No clinical disease (neither fever nor respiratory symptoms)

- No transmission

- Conclusion: Domestic pigs are susceptible to SARS-CoV-2 infection !!

Susceptibility of porcine cells to SARS-CoV-2

- Various porcine cells (SK6 and swine testicular cells, maybe PK-15), in culture, can be infected with SARS-CoV-2

- HeLa cells (human) made to express the porcine ACE2 protein can be infected with SARS-CoV-2 (the human ACE2 protein is the cellular receptor for SARS-CoV-2)

- So what is the barrier to infection of pigs by SARS-CoV-2 (maybe receptor distribution? (not in respiratory system or digestive system?)

My conclusions

- Pigs have not been shown to be susceptible to infection by SARS-CoV-2

- However:

  Some pig cell lines can be infected in culture by the virus

  Porcine ACE2 can act as a receptor for the virus

Thanks for your attention

- Any questions?