Japanese encephalitis virus infection of boars

2024 Australian Pig Vets Conference

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Some history…

- Japanese encephalitis first emerged in 1870s
- The earliest association with disease in humans, horses and stillbirths in pigs dates back to 1946-47. A similar disease among swine was also noticed during the great JE epidemic in Japan in 1935.
- Initially, the seasonal occurrence of reproductive failure in boars, particularly in animals imported from England, were confused with hyperthermia and heat stress as it coincided with the summer months.
- Experimental studies in the 1970s confirmed JEV as the causative agent of the seasonal reproductive failure in boars
- Reports of disease in boars ceased following the introduction of vaccination strategies

Published findings…

- Limited studies that specifically focus on the boar (3 natural vs 3 experimental infection)
- Most reports <5 animals; largest n=40 (natural infection)
- Impact on sperm quality (count, motility, malformation rate)
- Limited experimental studies that examine histopathology changes and IHC

Slight pyrexia and reduced appetite (60-70%); no change in collection volume; reduced libido occasionally reported

Decreased sperm count to azoospermia (45%); malformations (10%); reduced motility (20%)

Swelling of head of epididymis most commonly described; scrotal hyperaemia, scrotal swelling, testis atrophy also observed

Viable virus recovered from 3 animals (n=7) btw days 3 and 17 (VI not PCR)

Time for semen to return to normal ranged from ~90-150 days (n=4)

Impact on semen…

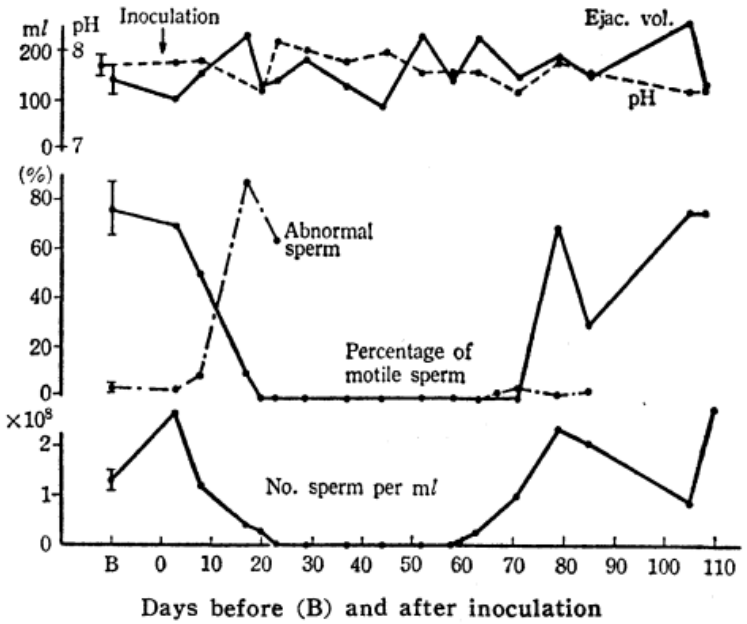


Fig. 2. Semen characteristics in boar No. 4 infected with JEV.

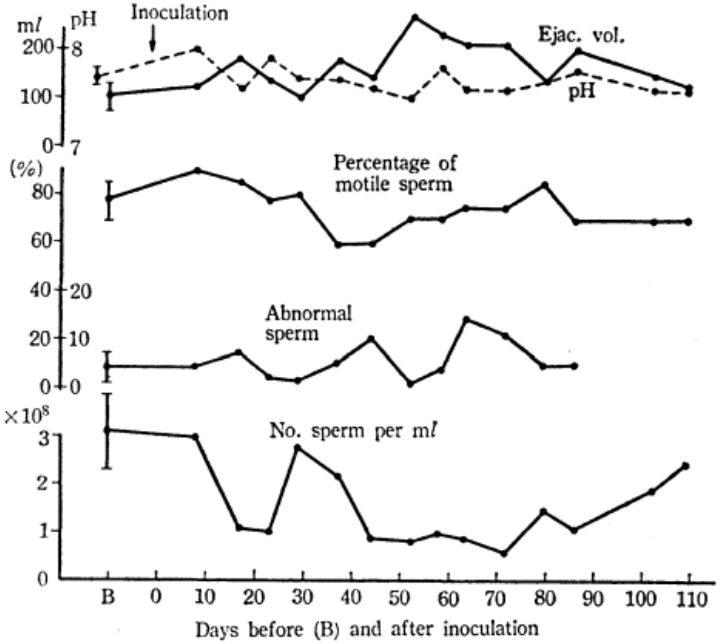


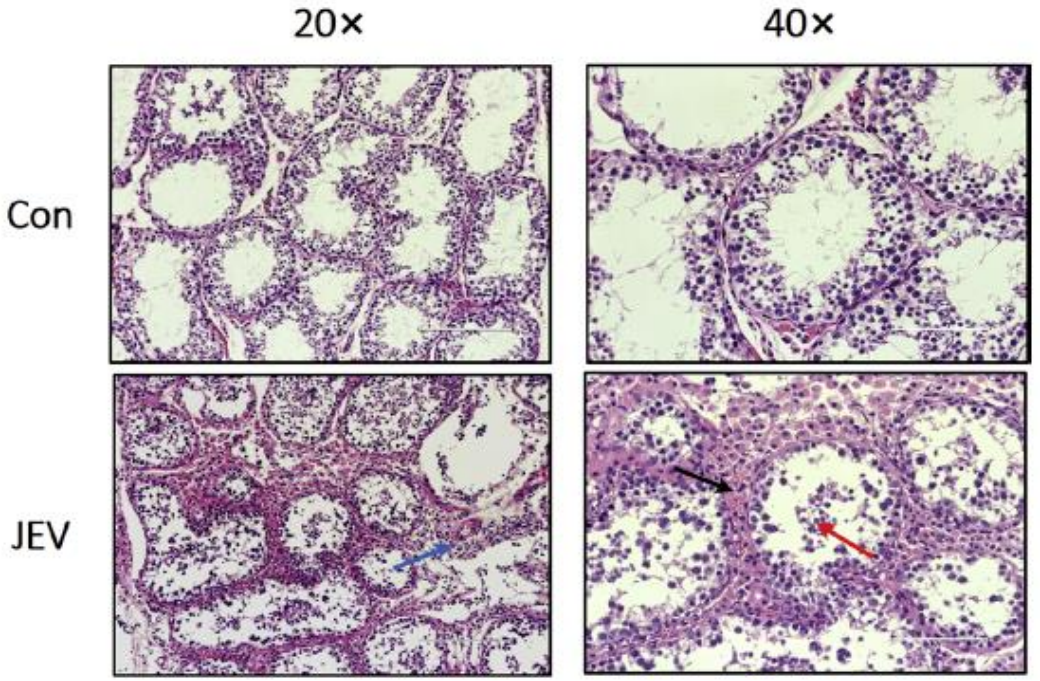
Fig. 3. Semen characteristics in boar No. 5 infected with JEV.

Ogasa, A., Yokoki, Y., Fujisaki, Y., Habu, A., J.J.J.o.A.R, 1977. Reproductive Disorders in Boars Infected Experimentally With Japanese Encephalitis Virus Vol.23. pp. 171–175.

Published findings - Histopathology

- 4-month-old boars
- 7 days post-infection

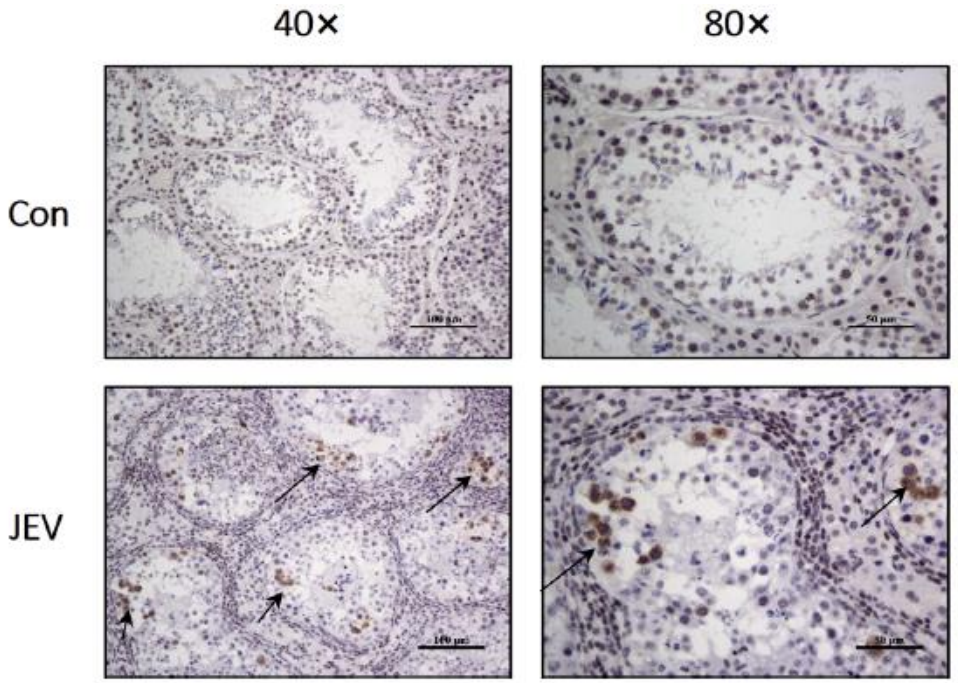
- Large number of spermatogonium are shed in centre of seminiferous tubules (red arrow)
- A lot of RBCs in the testicular stroma (black arrow)
- Inflammatory cell infiltration around the blood vessels (blue arrow)



https://doi.org/10.1016/j.vetmic.2019.108430

Published findings - Immunohistochemistry

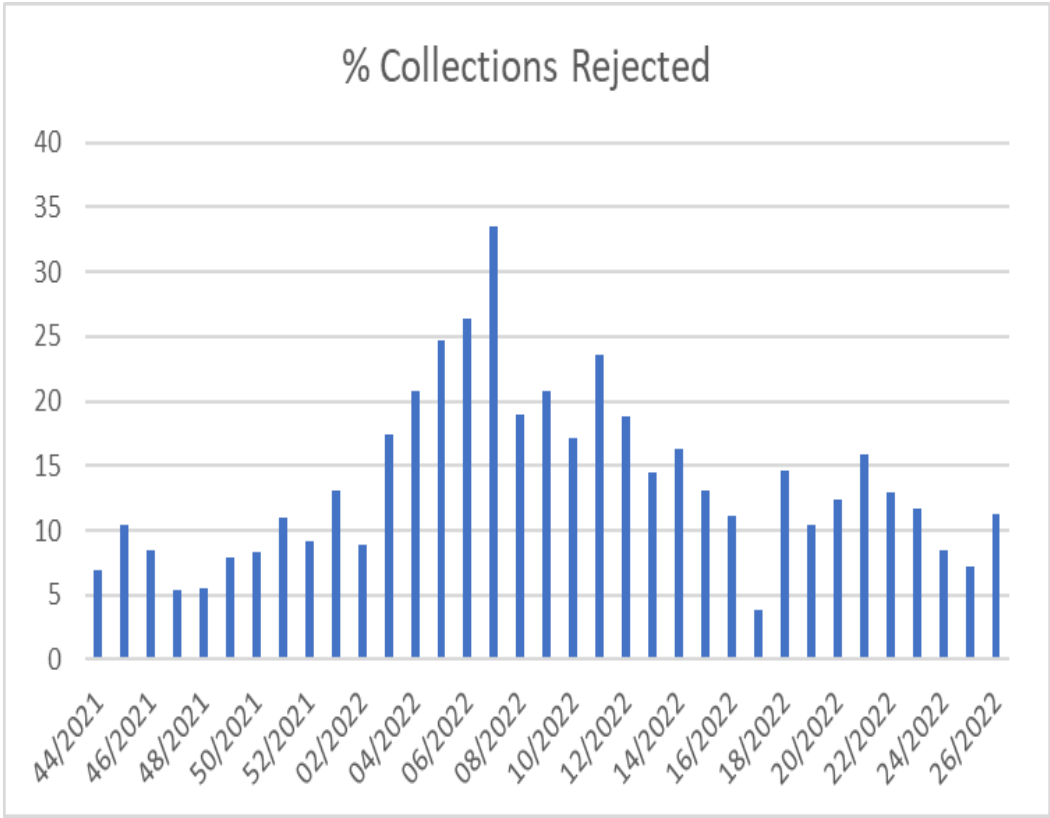
- JEV antigen mainly detected in spermatogenic cells and Sertoli cells following infection
- This explains the impact on spermatogenesis that in some cases leads to azoospermia



https://doi.org/10.1016/j.vetmic.2019.108430

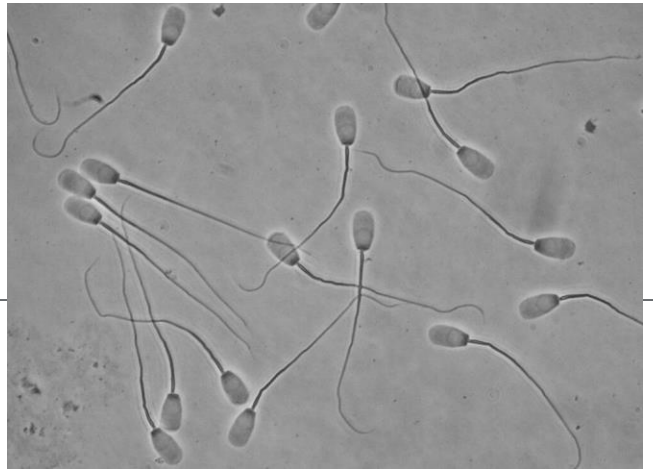
Was there an impact on boars in Australia in 2022?

- 24 February 2022 – large boar stud (n=~200) in Corowa NSW (phone call)
- “A few weeks ago a few boars (n=5-6) were lethargic, no fever, now back on food. Concerned about heat stress as it’s the biggest, heaviest boars that were affected”
- “2 weeks ago 17%, 1 week ago 23% and this week 50% semen rejected”
- “Abnormal bent tails, no sperm rich fraction at all…”

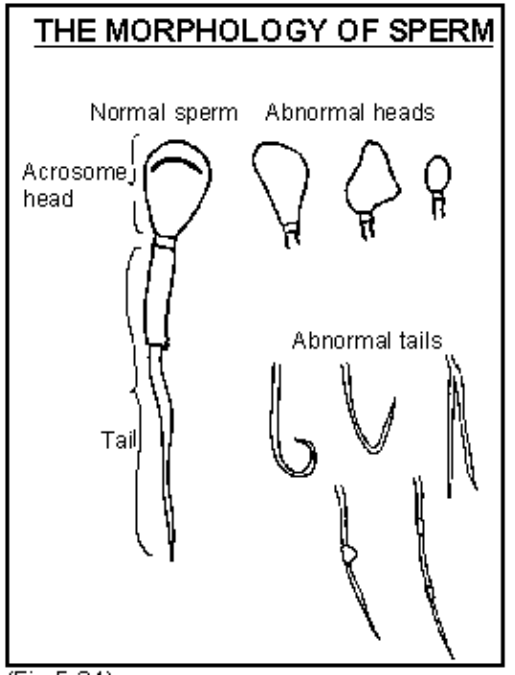


Data from boar stud (Site 1)

- All sampled animals (n=112) at boar stud were seropositive on 28/2/2022 (N=~199)
- A related quarantine station had first seropositive animals detected 27/12/2021 (2/24)
- Recovery period:
  - This varied and was boar specific:
  - ~50% of affected boars recovered in 4 weeks
  - ~50% never recovered after giving them 2-3 months before culling
- Impact – when semen rejection was at its highest there was not sufficient semen to maintain normal breeding



https://aces.illinois.edu/news/sugar-promotes-sperm-longevity-pig-reproductive-tract



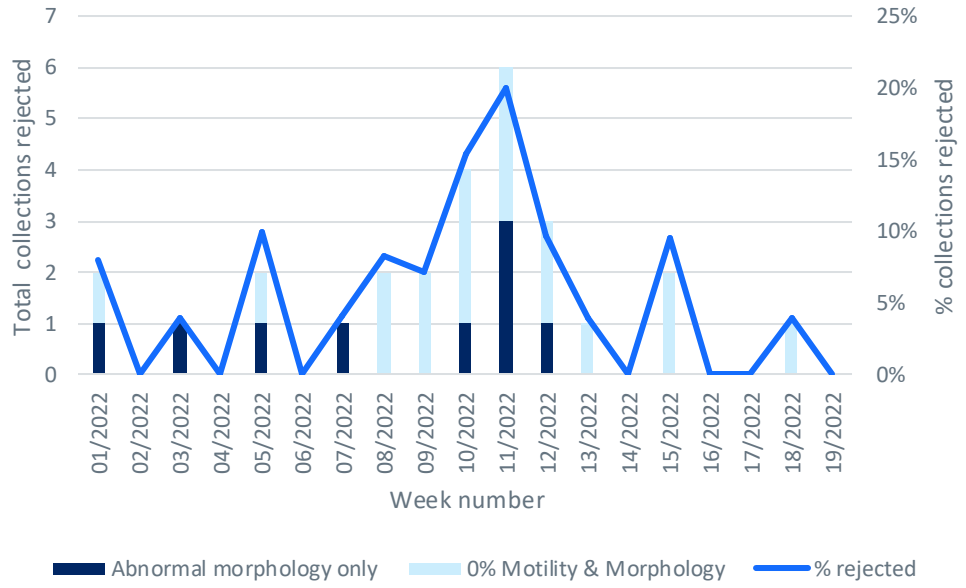
(Fig.5-34)

From: https://www.thepigsite.com/genetics-and-reproduction/insemination/facts-about-semen

Data from boar studs

- JEV PCR (Site 1) – 68 semen samples collected between 23/2-14/3/2022
  - 4 +ve (Ct values - 25.2, 32.0, 32.6, 36.7)
- JEV PCR (Site 2) – poor quality semen samples collected 2nd week of March 2022
  - 2/5 +ve (Ct values - 36.2, 36.5)

Collection Date	Boar Tag	week	BREED5	Ejaculate Volume	Density	Motility Day 1		Comments	Doses Produced	Normal	Distal Proximal	Distal Bent	Ab Head	Coiled	Other Damage Ac	Tailless
23/03/2022			5D4105				PEN	296								
23-Mar-22	2117	5D4105		206	0.061	85	0	reject Thin	Rejected	34	36	18	12	0	0	0	0	0
23-Feb-22	2113	5D4105		219	0.127	5	0	reject SQ Bact +++	Rejected	10	4	2	6	0	0	0	78	0
10-Feb-22	2111	5D4105		114	0.288	85	0	reject - SQ	Rejected	40	0	60	0	0	0	0	0	0
31-Jan-22	2110	5D4105		115	0.233	80	0	Reject - SQ	Rejected	58	4	32	6	0	0	0	0	0
25-Jan-22	2109	5D4105		173	0.408	85	0		30	80	0	18	2	0	0	0	0	0
20-Jan-22	2108	5D4105		110	0.439	85	0		20	86	0	12	2	0	0	0	0	0
7-Jan-22	2106	5D4105		153	0.448	85	0		29	90	4	2	4	0	0	0	0	0
31-Dec-21	2105	5D4105		153	0.333	85	0		22	90	0	4	4	0	0	0	2	0
24-Dec-21	2104	5D4105		191	0.457	85	0		37	92	0	2	6	0	0	0	0	0
15-Dec-21	2103	5D4105		155	0.441	85	0		29	96	0	0	4	0	0	0	0	0



Histopathology

- Submission 5/1/2022
- Gilgandra NSW
- The boar was mildly unwell with swollen testicles and recent history of mingling with a feral boar
  - Tissues were submitted for a Brucella exclusion
  - Brucella suis PCR and culture negative
  - Histo was subsequently requested, which showed intratubular orchitis
  - Then in light of JE outbreak, JEV PCR was done, which came back positive
- Histopathology - severe inflammation, with necrotic debris filling seminiferous tubules

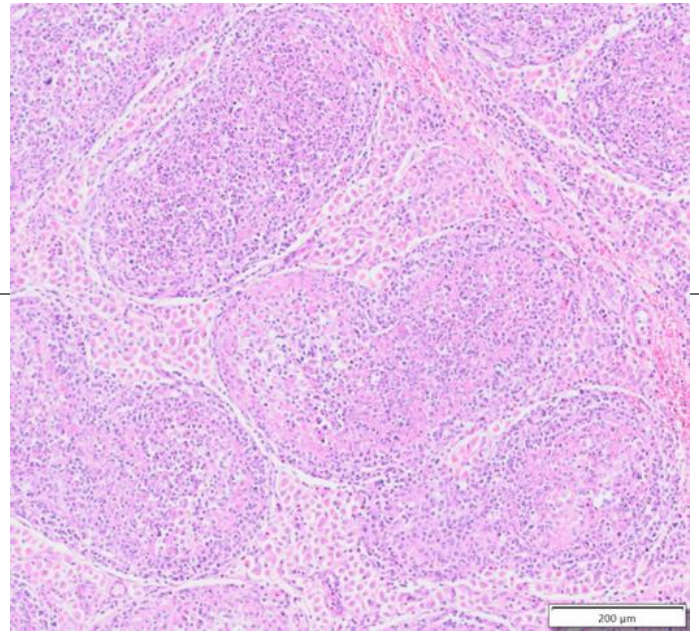
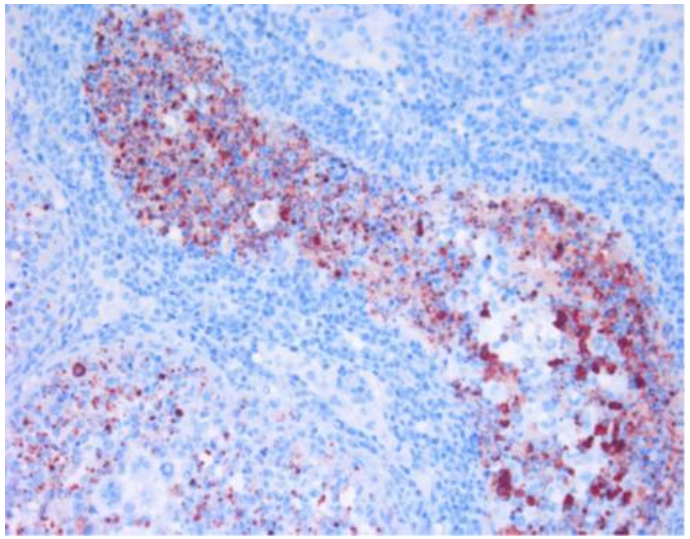


Image courtesy of Anne Jordan, EMAI

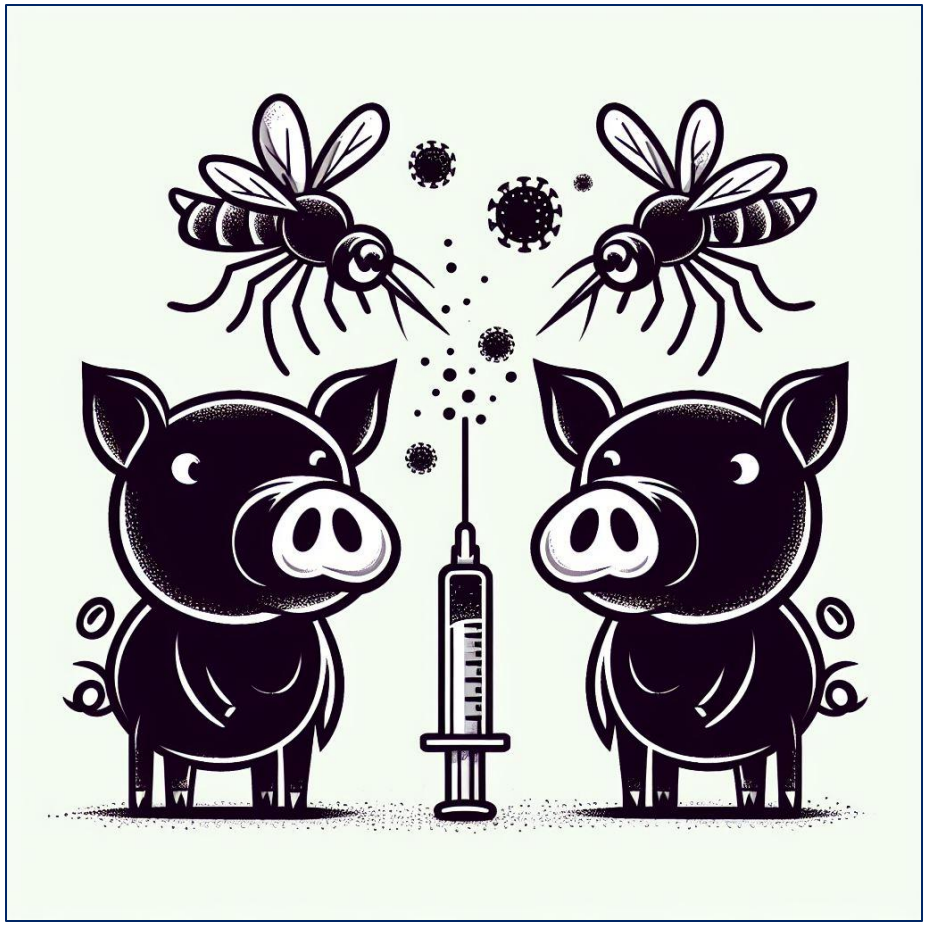


Flaviviral NS1 antigen in necrotic seminiferous tubules with peritubular inflammation.

22/03145

Summary

- Effect on boars in 2022 is similar to >50 years ago
- Limited host-pathogen interaction studies are available
- % of affected animals comparable (40%)
- Recovery period 30-150 days (sperm production takes up to 6 weeks)
- Decreased sperm count to azoospermia
- Tail abnormalities
- Frequency of pyrexia, lethargy and inappetence was uncommon in recent event



Acknowledgements

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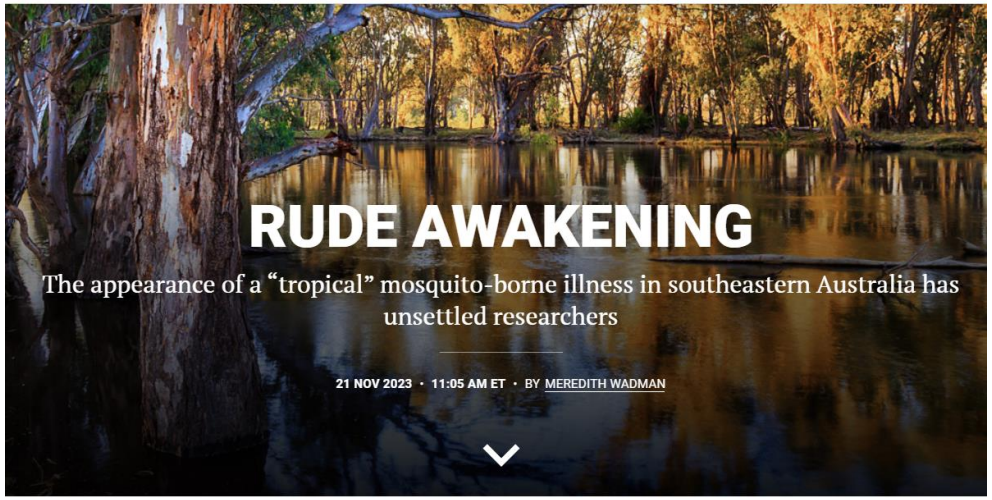
  - Greg Tuckett and Jade Marschall
  - Bernie Gleeson



https://www.science.org/content/article/how-rains-pigs-and-waterbirds-fueled-shocking-disease-outbreak-australia