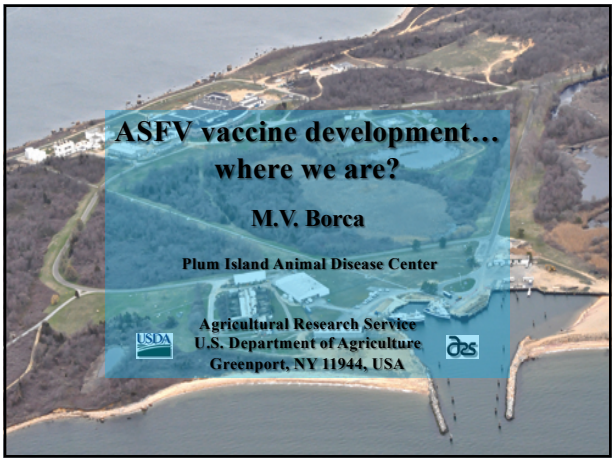
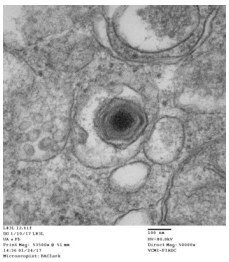


1

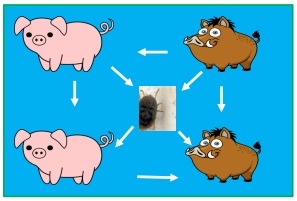
African Swine Fever Virus

- African swine fever virus: Asfarviridae family
  - dsDNA genome170-190kb
  - 150+ genes
  - Very few have been studied in detail ~20
- Highly lethal (100%) to subclinical infection
- Hemorrhage, edema, ascites and shock
- Wild Boar, Warthogs, domestic pigs
- Virus/Host factors responsible for different outcomes
  - Remain poorly understood
  - Different strains have different virulence
  - Varying host species symptoms/lethality
- Long-term persistent/latent infection
- Rapid and efficient transmission among pigs



2

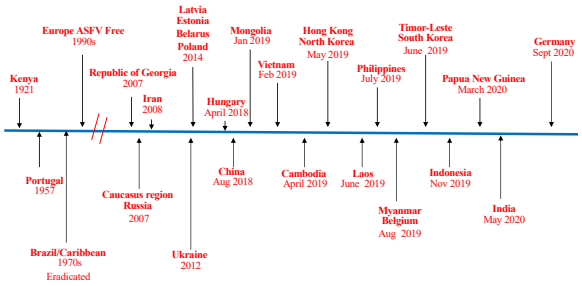
African swine fever virus: transmission



- Several days in feces
- Months in contaminated pens
- Years in frozen carcasses
- Pork products: Found over 140 days in salted/dried hams
- Feeding swill, garbage, waste

3

ASFV outbreaks



4

2019: African swine fever in China

Map courtesy of Pig Progress

- 104 outbreaks
- 360,000 culled on infected farms
- 1 Million culled to prevent spread
- 26 Provinces of China
- Backyard to ~70,000 animals/farm
- ASFV found in pig feed
- 50% reduction of China's swine heard
  - 1yr after initial outbreak

- China produced 639.82 million pigs/ year prior to ASFV

5

Live attenuated vaccines (LAV) are effective preventing homologous challenge

Attenuated isolates were produced by:

- Low virulence field isolates
- Viruses attenuated by tissue culture passages
- Viruses with genetically engineered deletions

6

Live attenuated vaccines (LAV) are effective preventing homologous challenge

Attenuated isolates were produced by:

- Lo

In this study we investigated whether deletion of genes from the ASFV low virulence isolate OUR T88/3 would reduce adverse clinical reactions post-immunisation but maintain induction of high levels of protection post-challenge with virulent isolate OUR T88/1. We took advantage of the adjacent genomic location of

Immunisation with OUR T88/3ΔDP2 did not appear to significantly reduce the adverse clinical reactions observed in pigs compared to parental OUR T88/3 since two pigs in each of groups 1 and 2 had transient joint swelling. Although it is possible that

fever virus isolate OUR T88/3 decreases its ability to protect against

deletion virus OUR T88/3ΔDP2 compared to 100% protection with the parental virus OUR T88/3. Thus the deletion of the two genes DP71L and DP96R from OUR T88/3 strain reduced its ability to protect pigs against challenge with virulent virus.

7

Live attenuated vaccines (LAV) are effective preventing homologous challenge

Journal of General Virology (2001), **82**, 513–523. Printed in Great Britain

In the current study, we investigated the effect of deletion of genes (*A238L* [28–30]; *A224L* [31,32]; *EP153R* [33,34] and *A276R* [35]), involved in virus-host interaction and immune system control on protection and virulence of the naturally-attenuated ASFV strain NH/P68.

•

Vaccine

journal homepage: www.elsevier.com/locate/vaccine

challenge. Pigs immunized with 10^6 $TCID_{50}$ of either vaccine candidates produced in COS7 cells or parental NH/P68 (produced in PAM), developed mild clinical signs (slightly raised body temperatures, necrotic skin areas and joint swelling, associated with chronic ASFV infection [25,27,45,46]) or remained asymptomatic.

8

Live attenuated vaccines (LAV) are effective preventing homologous challenge

Atten uated isolates were produced by:

- L

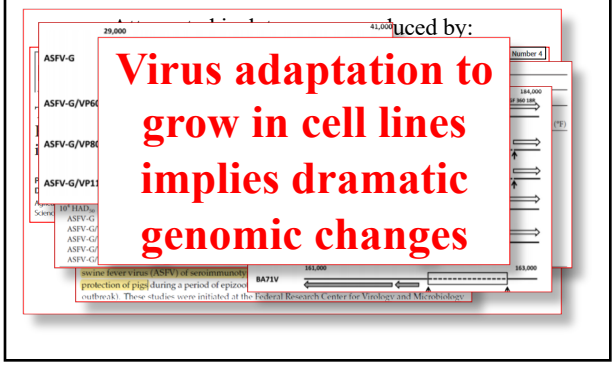
humoral immune response are provided in Figure 1. Specifically, one inoculated pig (PW17) showed weak peaks of fever (40.3–40.7°C) from 8 to 12 dpi accompanied by the appearance of cyanosis in

Low virulent field isolates retain residual virulence

fever at 10 dpe (PW16). Despite their apparent health status, intermittent weak viremia (Ct > 35) was detected in PW15 from 22 to 94 dpe, while PW18 remained aviraemic on all analysis days (up to 101 dpe). The remaining in-contact animals (PW14 and PW16) presented mild transitory clinical signs, specifically joint swelling, from 7 to 16 dpe, particularly in PW14. In PW14, the viremia lasted for

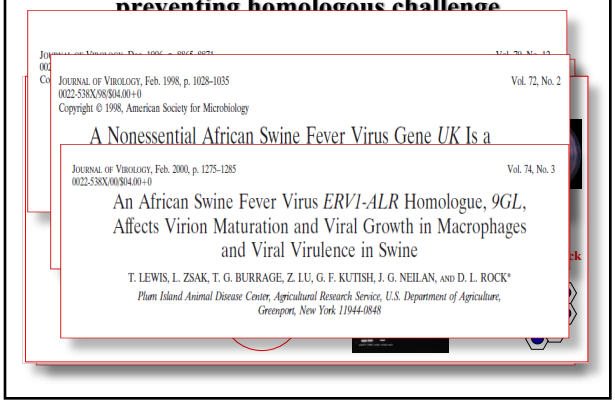
9

Live attenuated vaccines (LAV) are effective preventing homologous challenge

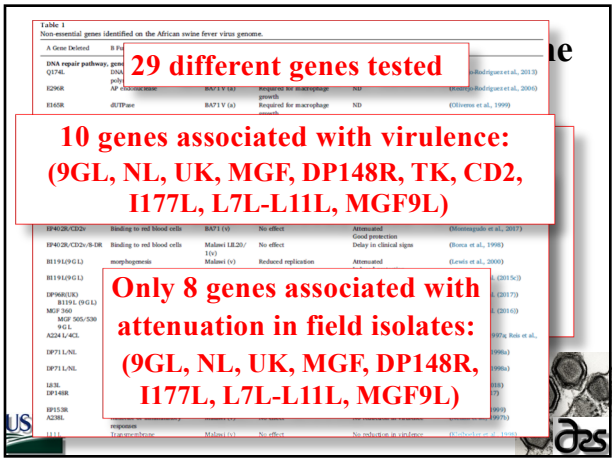


10

Live attenuated vaccines (LAV) are effective preventing homologous challenge

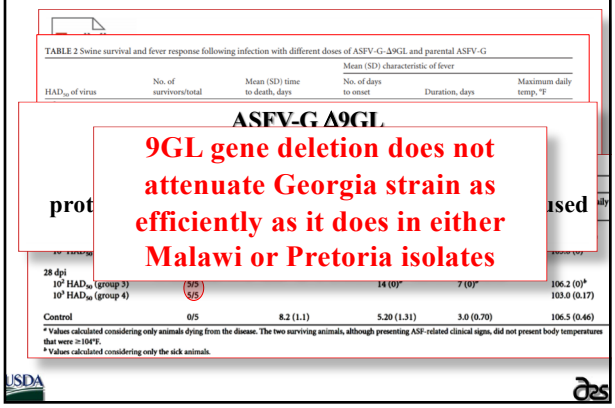


11



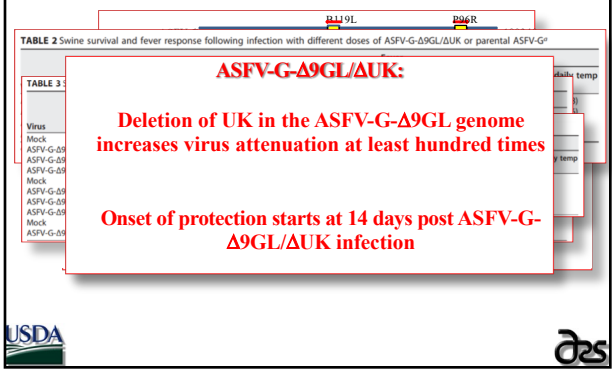
12

ASFV-G-Δ9GL



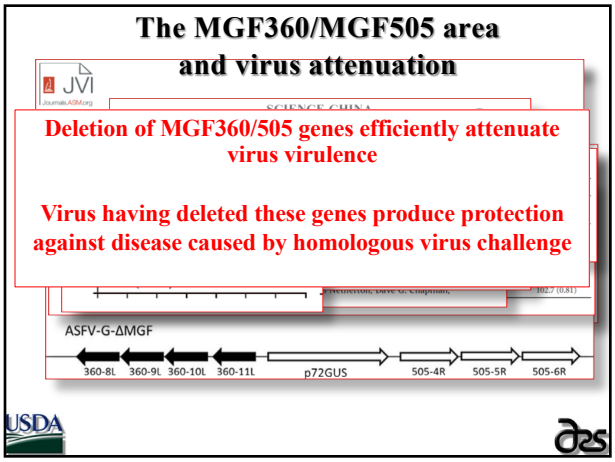
13

ASFV-G Δ9GL/ΔUK



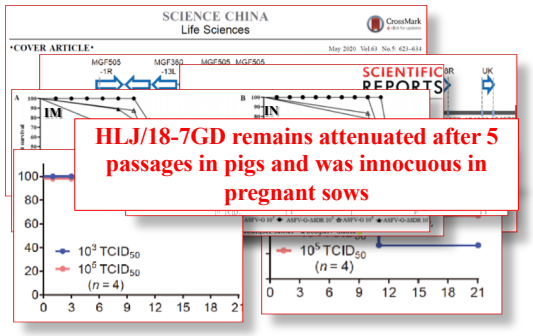
14

The MGF360/MGF505 area and virus attenuation



15

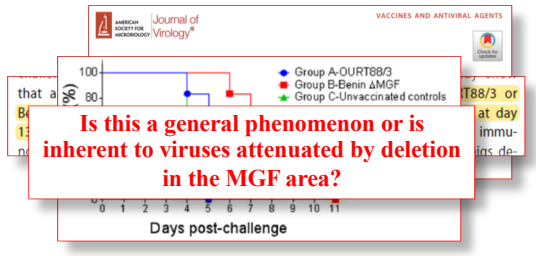
HLJ/18-7GD



HLJ/18-7GD remains attenuated after 5 passages in pigs and was innocuous in pregnant sows

16

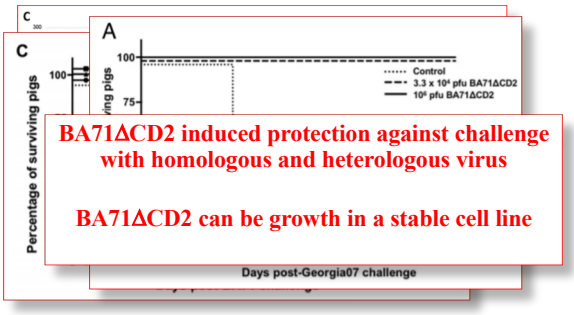
Length of protection induced by vaccine candidates harboring deletion of the MGF area



Is this a general phenomenon or is inherent to viruses attenuated by deletion in the MGF area?

17

BA71ΔCD2

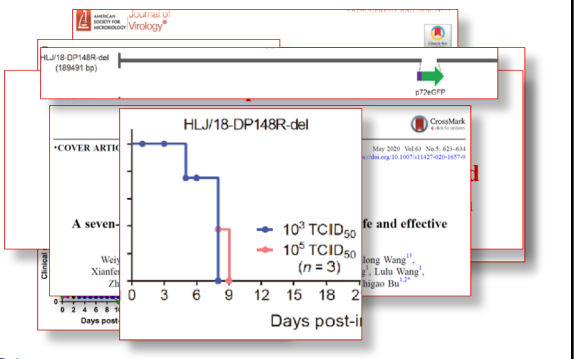


BA71ΔCD2 induced protection against challenge with homologous and heterologous virus

BA71ΔCD2 can be growth in a stable cell line

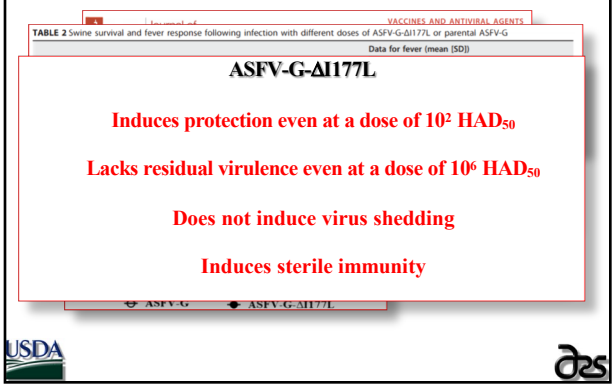
18

BeninΔDP148R



19

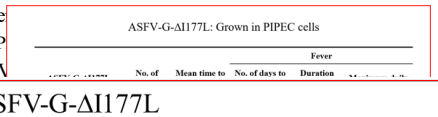
ASFV-G-ΔI177L



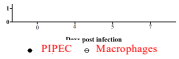
20

Production of ASFV-G-ΔI177L in cell culture

- Scree
  - PIP
- ASFV
-



ASFV-G-ΔI177L
- Vaccine produced in Macrophages or PIPEC
- 100% protection at 10^2
- PIPEC suitable cell line for Vaccine production



- PIPEC are a suitable cell line for vaccine production

21

Gaps

Study critical aspects of vaccine candidates (onset and duration of immunity, genetic stability, attenuation stability, minimal protection doses, possible routes of inoculation)

Standardization of methodologies to test efficacy of vaccine candidates

Develop vaccine candidates with DIVA capability

Investigate the bases of the differential effect in virus virulence of deleting the same genes among different virus isolates (9GL, CD2, NL, UK, TK, DP148R)

Continue the discovery and characterization of virulence associate genes

22

ARS vaccine candidates

ASFV-G-Δ9GL
ASFV-G-ASFV-G-Δ9GL/ΔUK
ASFV-G-ΔMGF
ASFV-G-ΔI177L
ASFV-G-ΔI177L/ΔLVR

All these candidates have been patented and are being in the process of being licensed by veterinary pharmaceutical companies

Thank you

Questions?

23