Immunisation of African indigenous pigs with attenuated genotype I African swine fever virus OURT88/3 induces protection against challenge with virulent strains of genotype I

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African swine fever virus (ASFV)

- member of Asfivirus genus, *Asfarviridae family*
- **DNA** virus (170 – 193 Kbp), with highly conserved central region (125 Kbp) with two terminal variable regions (∼ 30 Kbp) → currently they are **22 different genotypes**
- causes an **acute haemorrhagic fever in pigs** and can induce high mortality
- with **severe socio-economic impacts**
- **endemic** in many sub-Saharan African countries, Sardinia and parts of the Trans Caucasus region and Russian Federation

- Currently **no vaccine is available** to protect pigs against ASF and this limits options for disease control
Background and aim

• **Background:** The attenuated African swine fever virus genotype I strain OURT88/3 has previously been shown to induce 66 to 100% of protection of European breeds of domestic pigs depending of the virus isolates as challenge (Boinas et al., 2004; King et al., 2011).

   ⇢ This strain has a **large deletion** near the left genome end compared to virulent isolates and interruptions in three other **genes involved in immune evasion** (Chapman et al., 2008).

• **Research question:** What is the **efficacy** of the same attenuated vaccine strain in indigenous breeds of pigs from the DRC after challenge with autologous genotype 1 (2 different virulent isolates)?
Design and protocol of the study

• Appropriate BSL3 facilities (equivalent)
• Experiment approved by the ad hoc Authority
• Virus isolates: **OURT88/3** (attenuated strain) and two virulent strains from genotype I: **OURT88/1** previously described (Chapman et al., 2011) and a DRC field strain **085/10**.
• Maximum five times in primary pig macrophage cultures
• Indigenous pigs (at least 3 months old) included in the experiment coming from farms near Kinshasa and tested negative for ASFV by antibody detection ELISA and PCR (2 times at 30 days interval).
Groups of animals

21 pigs

8 vaccinated (Group A)
Vaccinated and 2 subsequent challenges

6 control challenge (Gr. B)
Control with one challenge (first virulent strain)

7 control challenge (Gr. C)
Control with one challenge (second virulent strain)
8 vaccinated (V 1)

**Vaccinated group**

- **IM** with $10^4$ TCID$_{50}$ **attenuated OURT88/3 strain**
- After **21 days** challenged **IM** with $10^4$ HAD$_{50}$ **OURT88/1 virulent strain**
- After a further **21 days**, remaining pigs were challenged **IM** with $10^4$ HAD$_{50}$ DRC strain **085/10 virulent strain**
In **group B**, six pigs were challenged IM with $10^4$ HAD$_{50}$ **virulent strain OURT88/1** 21 days after the start of the experiment, at the same time as those in group A

In **group C**, seven pigs were challenged IM with $10^4$ HAD$_{50}$ **virulent DRC strain 085/10** at 42 days at the same time that group A pigs were challenged with this strain
Follow up

• Pigs were monitored daily for clinical signs, and these were scored using a clinical scoring system previously reported (King et al., 2011).

• Blood samples were collected at different days post-immunization and challenge (ELISA). At termination, tissue samples were collected from spleen, lymph nodes and kidney (PCR).

• The ASFV genome copy numbers in blood and tissue samples were estimated by quantitative PCR (King et al., 2003).
Main results

• **21 days after the second challenge** (63 days after vaccine):
  - 4 pigs out 8 were immunized,
  - 4 pigs out 8 died (1 from an unknown cause)

• **Regarding the 4 immunized pigs** with zero antibodies (Abs) at the beginning of the trial, showed high titers 21 days after the second challenge, zero clinical signs, few DNA copies for some and zero for others.

• **Regarding the 4 died pigs**:
  - 1 of the 4 dead pigs was apparently killed by the sampling manipulation;
  - 3 killed by the challenges’ viruses (acute disease + severe signs + PCR positive).
Conclusions, perspectives and limitations

• Despite encouraging results in terms of “efficacy”
• The “safety” was not investigated
• Interference between vaccine and first challenge on the 2\textsuperscript{nd} challenge outcome was not investigated
• They are 22 genotypes of AFSV, with 3 genotypes in DRC (genotypes I = main, IX and XIV) \rightarrow more experiments should be devoted to the other main genotypes
• The genome of AFSV is the subject of variation

\rightarrow vaccination still a challenge
ORIGINAL ARTICLE

Immunization of African Indigenous Pigs with Attenuated Genotype I African Swine Fever Virus OURT88/3 Induces Protection Against Challenge with Virulent Strains of Genotype I

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Vaccinated and 2 challenge’s surviving pigs
Thank you for your attention