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ANALYSIS OF VIREMIA AND TRANSPLACENTAL TRANSMISSION OF FIELD AND 
RESCUED STRAINS OF BTV-2 AND BTV-8 FOLLOWING INOCULATION OF 
PREGNANT SHEEP

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Objectives
Live bluetongue virus (BTV) vaccine-strains and also, surprisingly, the European strain of BTV- 
8 can cross the placental-barrier and thus pass from one generation of animals to another without 
involvement of the insect vector.
A better understanding of the genetic basis for the transmission characteristics of the virus would 
help to identify the risks posed by further BTV incursions and facilitate the design of better 
control strategies. The development of reverse genetics for BTV enables investigation of the 
genetic traits conferred by individual genome segments within rescued viruses by making 
defined reassortants. To date, only a few experiments have investigated whether field and 
rescued virus strains behave similarly in vivo.

Methods
Twenty-four sheep (in 4 groups of 6) were inoculated (s.c.) with 4 strains of BTV in late 
pregnancy (approx. 1 month before lambing). The viruses used were: BTV-2 wt (Italian field 
strain), BTV-2 (rescued), BTV-8 wt (field strain from the Netherlands) and BTV-8 (rescued). 
Four sheep were non-inoculated controls. Blood samples from the sheep were tested frequently 
for viremia and anti-BTV antibodies (by ELISA) in the period until lambing. Pre-colostral blood 
samples were collected from all newborn lambs, except for one born dead, to determine if 
transplacental transmission had occurred. Milk from ewes was collected daily for 7 days after 
lambing and blood samples from the lambs were collected on days 0, 3 and 7 after birth. All 
samples have being tested for the presence of anti-BTV antibodies and for virus (RT-qPCR).

Results
All inoculated animals developed viremia. The viremia was significantly higher at all sampling 
points following inoculation (p<0.01 or p< 0.05, Mann-Whitney's U Test) in animals inoculated 
with BTV-2 wt compared to animals inoculated with BTV-2 rescued, whereas no significant 
difference was detected between BTV-8 wt and BTV-8 rescued. Wild type virus infected animals 
had a longer lag phase before antibodies were detected but the response increased at a faster rate. 
Some of the animals displayed clinical signs of infection, e.g. fever and panting. All the ewes 
deivered one lamb each, a few lambs born early did not thrive and were euthanized but most 
appeared healthy. Seven of the 28 lambs had been infected transplacentally; 2 from ewes 
inoculated with BTV-2 wt, 3 from ewes inoculated with BTV-2 rescued and 1 from a ewe 
inoculated with BTV-8 wt. The last infected lamb was from a non-inoculated control sheep, in 
the same stable but physically separated from, the BTV-2 wt inoculated ewes and became 
viremic with BTV-2 10 days after the others were inoculated.

Conclusion
Both wild-type and rescued BTVs induced viremia. Surprisingly, transplacental transmission 
ocurred more frequently in ewes inoculated with BTV-2, both wt and rescued, than in ewes 
inoculated with BTV-8. The BTV-2 wt was passaged once in Kc and once in CPT-Tert cells. 
These very few passages may be enough to introduce changes enabling the virus to cross the 
placental barrier. This experiment indicates it will be difficult to identify a single BTV segment 
responsible for transplacental transmission in sheep using rescued BTV-2 and BTV-8 strains.