

## **6b Summary**

### Equine Herpes virus Type 2 (EHV-2): Studies of the prevalence in eye swabs and the tissue- and cells tropism, particularly in cases of equine Keratokonjunktivitis

The aim of this project was to further investigate the etiopathogenetic role of EHV-2. The main goal was to exam the clinical relevance of EHV-2 in the genesis of conjunctivitis and keratitis, tissue tropism and possible sites of latency in horses. Blood samples and swabs from 28 eye diseased test group animals and 28 healthy control group horses were taken twice within 4-5 weeks in the horse clinic of the Tierärztliche Hochschule Hanover. These samples were virologically investigated in the Institute of Virology, Faculty of Veterinary Medicine, Freie Universität Berlin. Furthermore, *post-mortem* tissues from 20 naturally infected horses were analysed for EHV-2.

EHV-2 was detected by various serological methods, nested PCR (nPCR) and co-cultivation in both the test and control group. A statistically significant correlation between infection and tested groups was not observed. Interestingly, EHV-2 was detected in mules using the aforementioned methods, implicating that mules appear to be permissive for EHV-2.

The sampling of the eye was carried out in parallel with cotton wool swabs and Cytobrushes from the conjunctival fossa. In both groups EHV-2 was detected significantly more often in the samples obtained by the Cytobrush. As a Cytobrush sample contains much more cell material than a cotton wool swab sample, it is likely that the higher frequency of EHV-2 detection in Cytobrush samples indicates an intracellular virus load of the deeper stratum of the conjunctiva. Therefore, the conjunctiva might be a site of latency of EHV-2.

The fact that EHV-2 was detected more frequently in Cytobrush samples from the conjunctival fossa than in PBMC, an already known site of latency, provides further evidence that the conjunctiva may be a site of latency.

In *post-mortem* tissues, EHV-2 was found more often in mucous membrane (56% of the conjunctiva, 2/2 noses) than in the lachrymal glands (31%), the lung (24%), nerve tissue (20% of the trigeminal ganglia, 10% of the optic nerves) and the aqueous fluid (0%) via nPCR. These results demonstrate that EHV-2 was detected most frequently in the conjunctiva of *post-mortem* tissues.

In tissues analysed from the lachrymal gland, nose and optic nerve, EHV-2 was present only in conjunction with areas which were observed to have an EHV-2-positive conjunctiva. This suggests that the conjunctiva might act as a portal for EHV-2, and that the virus spreads from the conjunctiva to other tissues. Another indicator for virus spread via the lacrimal-nose-channel was indicated by the fact that 75% of horses with EHV-2-positive nose swabs were also positive for EHV-2 in the eye swab sample.

EHV-2 was detected in a naturally-intrauterine-infected foetus, which was aborted during the eighth month of pregnancy. EHV-2 was detected in the trigeminal ganglia of this foetus.

Genetically differing EHV-2 serovars were present in both swabs and in blood samples taken from the same horse simultaneously as well as in eye- and nose- and blood-samples taken from one animal at various time points. No genetic similarities were observed by using the described primers and restriction enzymes. Thus no conclusions concerning the virulence and tissue tropism of different EHV-2 serovars were revealed.