



Transmissible Spongiform Encephalopathies rapid post mortem tests in goats: Strategies to improve the efficiency of the active surveillance statutory programme

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ancillary target for scrapie selection. Moreover, our findings support the idea that AGER is involved in the mechanism of neuronal dysfunction associated with prion diseases.

P.105: Transmissible Spongiform Encephalopathies rapid post mortem tests in goats: Strategies to improve the efficiency of the active surveillance statutory programme

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Annex X to Regulation (EC) No 999/2001 lists the approved rapid post mortem tests which may be used within the framework of the EU monitoring programmes. Little is known regarding the efficiency of different rapid methods on goats. The authors compared the performance of the *IDEXX HerdChek*[®] BSE-scrapie, the *Bio-Rad*[®] *TeSeE*TM *SAP* and the *Bio-Rad*[®] *TeSeE*TM *Sheep/Goat* tests over 96 central nervous system goat tissues including 20 experimentally obtained BSE positives, 41 scrapie positives and 35 negative samples prepared following the European reference laboratory standard method of homogenate preparation (50% w/v protocol). The relation between age, prion type, PNRP genotype and test performance were further investigated. Clear differences in relative diagnostic sensitivity were shown, being the *IDEXX HerdChek*[®] BSE-scrapie the best performing system setting its 95% IC sensitivity at 100%. Interestingly, *BioRad*[®] *TeSeE*TM *Sheep/Goat* test showed a 95% IC sensitivity of 85% on BSE positive samples and an optimistic 82.9% on scrapie ones (considering the only "suspect" result as positive), while *BioRad*[®] *TeSeE*TM *SAP* reached only a 75% sensitivity on BSE samples but a 90.2% on scrapie positives. The reproducibility was not absolute as some discrepant results in any combination of tests was shown, nevertheless, the lower limit of the 95% CI was always above 0.78 considering the samples as a whole, and always above 0.74 when the stratification by PrP^{Sc} profile was applied, values consistent with a substantial-good-fair to good agreement. All the tests correctly identified the 35 negative samples, showing a 100% specificity. Our findings on BSE samples do not suggest any contribution of the original inocula (bovine or goat BSE) on the test results whereas the mean age of the natural scrapie positive animals set at 70 months, confirming the hypothesis that the

sensitivity of detection using brainstem appeared to be dependent on the age of tested individuals. The studies on the correlation between rapid test performance and genotype is ongoing. Considering the current lack of genetic selection for eradicating/controlling classical goat scrapie at population level, the efficiency of the surveillance in place would be crucially dependent on the system used to identify infected flocks.

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P.106: In vitro generated mouse prion protein causes brain neurodegeneration in FVB/N female mice

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Recently we reported that lipopolysaccharide (LPS) from *Escherichia coli* 0111:B4 was able to instantly convert mouse prion protein into a beta-rich isoform (moPrP^B) resistant to proteinase K digestion under normal physiological conditions. In this study we tested whether subcutaneously (sc) administered moPrP^B (29-232) is able to cause prion-like pathology in vivo. Six groups of 15 FVB/N female mice each were treated sc with: (1) Saline, (2) LPS (0.1 µg/g of body weight), (3) moPrP^B (45 µg/mice), (4) LPS+moPrP^B, (5) RML (Rocky Mountain Laboratory scrapie prions at 10⁷ ID units), and (6) LPS+RML. Saline and LPS were administered over a period of 6wk using ALZET[®] osmotic minipumps (ALZET, Cupertino, CA) implanted sc, whereas moPrP^B and RML were administered as one time sc injection at the beginning of LPS infusion. Five animals from each treatment group were euthanized at 11wk post-inoculation (pi), with no clinical signs of prion disease. The rest of the mice were left to develop clinical signs associated with prion disease until terminal sickness. Hematoxylin & eosin (H&E) and PrP^{Sc}-stainings were conducted from brain tissues to determine presence of vacuolation and PrP^{Sc} accumulation, respectively. Western blot (WB) analysis and scrapie cell assay (SCA) using L929 cells were also conducted to evaluate presence of resistant PrP (PrP-res). All treatment groups, except for saline, showed mild brain vacuolation at 11wk pi in the cerebral cortex (Cc), thalamus (Th), midbrain (Mb), and cerebellum (Cr) and mild PrP^{Sc} accumulation only in the LPS+RML treatment. Terminally sick mice exhibiting clinical signs of prion disease from the LPS, moPrP^B, and LPS+moPrP^B treated groups showed widespread brain neurodegeneration in the Cc, Th, Mb, and Cr comparable to positive controls. Computer evaluated vacuolation of terminally sick mice showed numerically larger size vacuoles in the Cr and Mb brain regions of LPS, moPrP^B, LPS+PrP^B, and LPS+RML treated mice