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Molecular and Immunodiagnostic Methods for Scrapie in Sheep: Applications of PCR and ELISA

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Abstract: *Scrapie is a fatal neurodegenerative disease of sheep associated with the accumulation of an abnormal form of the prion protein. Laboratory testing is essential for disease surveillance, while molecular analysis provides valuable information on genetic susceptibility. This review examines the principles, applications, and limitations of PCR-based methods and enzyme-linked immunosorbent assays (ELISA) for assessing scrapie in sheep.*

The molecular component focuses on analysing the prion protein gene (PRNP), particularly polymorphisms at codons 136, 154, and 171 associated with susceptibility to classical scrapie. The review discusses conventional PCR followed by Sanger sequencing and real-time PCR with allele-specific fluorescent probes as approaches for identifying PRNP variants. The review considers their applications in genetic risk assessment and breeding programmes, along with factors that affect analytical reliability and genotype interpretation.

The immunodiagnostic component addresses ELISA-based detection of disease-associated prion protein in tissue samples, emphasising its role in rapid screening and surveillance. The paper discusses sample selection, assay performance, and the need for confirmatory testing, as well as differences between classical and atypical scrapie that may affect detection and interpretation.

The review focuses on the distinction between identifying genetic susceptibility and detecting disease-associated prion protein. PRNP genotyping does not establish infection status, whereas ELISA screening requires interpretation within an appropriate diagnostic framework. By examining these complementary approaches, the review provides a foundation for understanding how molecular genetics and immunodiagnostics contribute to scrapie surveillance, prevention and control.

• Introduction

Scrapie is a naturally occurring prion disease of sheep and goats that poses an important challenge to animal health and livestock management. Its prolonged incubation period and the limited specificity of clinical signs make early recognition difficult, highlighting the importance of reliable laboratory testing and effective surveillance programmes. [Benestad et al., 2008; EFSA Panel on Biological Hazards, 2014; WOAH, 2022].

• Results and discussions

PRNP genotyping: Polymorphisms at codons 136, 154, and 171 are important genetic markers associated with susceptibility to classical scrapie. PCR followed by DNA sequencing allows the identification of these variants, while allele-specific real-time PCR offers a faster approach for targeted genotyping.

These methods support genetic risk assessment and selective breeding programmes but do not determine infection status [Dawson et al., 1998].

ELISA-based diagnosis: ELISA rapid tests support scrapie surveillance by detecting disease-associated prion protein in tissue samples. Their reliability depends on appropriate sample selection, assay performance, and the disease form. Reactive screening results require interpretation within the established diagnostic framework and, where indicated, confirmatory testing [WOAH, 2022].

Comparative assessment: PCR-based genotyping and ELISA serve complementary purposes. Genotyping estimates genetic susceptibility, whereas ELISA provides evidence of disease-associated prion protein. Differences between classical and atypical scrapie may affect tissue distribution and detection, emphasizing the importance of appropriate sampling and diagnostic interpretation [Benestad et al., 2008; EFSA Panel on Biological Hazards, 2014].

• Conclusions

PCR genotyping assesses genetic susceptibility, while ELISA supports scrapie detection. Both contribute to effective disease control.