

Process from block collection to returning blocks to participating hospitals: following ethical approval and agreement of the participating hospitals to release paraffin blocks, they were collected by a team based at Public Health England. At Public Health England, blocks were allocated to bins of 50 blocks, and using database software, the PIN was allocated to a serial number for each block. Blocks were then sent to the participating laboratories, where they were sectioned and stained with haematoxylin and eosin, and immunolabelled for ICSM35 and KG9 (UCL Institute of Neurology) or for ICSM35 and KG9 only (Animal Health and Veterinary Laboratories Agency). One block underwent a quality control procedure, i.e. 8 unstained sections, and 2 x 10 µm paraffin rolls were prepared. Blocks of which preparations showed an immunoreactivity requiring further expert examination were also further sectioned (12 sections, 4 µm each) and 3 x 10µm paraffin rolls. At this stage, all blocks were returned to the Public Health England laboratory and were unlinked. Following the unlinking, the sections requiring further expert examination were presented to the consultant histopathologist (SB) or senior histologist (YS) who assessed the sections for specific PrP immunolabelling and confirmed a “negative, non-specific, suspect, or positive” sample, as defined in the Methods section. At this stage, additional sections were prepared with the antibodies 3F4 and 12F10. The result was communicated to Public Health England and suspect or positive as well as unstained slides were sent to the external experts (James Ironside, Edinburgh and then David Hilton, Plymouth). Additional immunohistochemical preparations were prepared in the laboratory in Edinburgh when necessary