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DEVELOPMENT OF A REAL-TIME RT-PCR FOR DIFFERENTIAL DETECTION OF REASSORTANT H1N2 (H1N2R) SWINE INFLUENZA VIRUS

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Introduction

Endemic swine influenza virus (swIAV) strains including $H1_{av}N1$, $H1_{hu}N2$ have been circulating in Great Britain (GB) for decades. In 2009, H1N1pdm09 became the dominant strain detected followed by H1N2 and the rapid decline of $H1_{av}N1$ detections. An H1N2-H1N1pdm09 virus reassortant was detected in 2010; comprising H1 haemagglutinin (HA) and N2 neuraminidase (NA) surface glycoproteins with the H1N1pdm09 internal cassette. Identification of the swIAV sub-type is important for surveillance, epidemiological investigations and decisions regarding vaccination, animal welfare and public health implications. While real-time reverse transcription polymerase chain reaction (RRT-PCR) assays have recently improved the sensitivity and speed of swIAV sub-typing, these protocols cannot specifically identify reassortant H1N2 (H1N2r) swIAVs. Both the conventional $H1_{hu}N2$ and H1N2r now co-circulate in GB. A RRT-PCR for differential detection of H1N2r in the GB pig population was therefore developed for use in conjunction with H1N2 sub-typing RRT-PCR assays.

Material & Methods

An RRT-PCR to specifically detect the nucleoprotein (NP) internal gene of H1N1pdm09 was developed to distinguish between conventional H1N2 and H1N2r swIAVs using a modification of previously-published primers incorporating a re-designed locked nucleic acid probe to impart maximal discriminatory power.

Results

The H1N1pdm09-NP RRT-PCR assay correctly identified the H1N1pdm09-NP gene segment in H1N2r viruses from a panel of four H1N1pdm09 control viruses and a further panel of 12 conventionally-typed H1N2 viruses. This will be expanded to cover all H1N2 detections since 2012 (~60 viruses).

Discussion & Conclusion The RRT-PCR will provide added value for influenza A surveillance in GB by building on the sub-typing RRT-PCR protocols to differentiate between the conventional H1N2 swIAVs and H1N2r viruses which will be of relevance to animal welfare and zoonotic risk potential.