Molecular epidemiology of Newcastle disease and avian influenza in South Africa

by

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Highly pathogenic avian influenza (HPAI) and velogenic Newcastle disease (ND) are devastating diseases of poultry that are notifiable to the World Animal Health Organization (Office International des Epizooties). RT-PCR, DNA sequencing, molecular characterisation and phylogenetic analyses were conducted on South African ND and AI virus strains isolated since the 1990s, to investigate the epidemiology of both diseases in the country. The first recorded outbreak of AI in chickens that started in 2002 was caused by two genetically distinct low pathogenicity avian influenza virus (LPAI) H6N2 genotypes that arose from a common ancestor. The ancestral virus appears to have been produced by reassortment between two ostrich viruses, A/Ostrich/South Africa/KK98/98 (H6N8), and A/Ostrich/South Africa/9508103/95 (H9N2). This highlighted the potential role that ostriches may play as mixing vessels for strains that may spill over into chickens when biosecurity breaks down. LPAI H3N8, H4N8 and H5N1 viruses isolated in 2004 from wild ducks in Gauteng were determined to be Eurasian in origin, but the LPAI H5N1 virus was not closely-related to the highly pathogenic avian influenza (HPAI) Asian (or Genotype Z) H5N1 strain. Pelagic shorebirds are implicated in the introduction of these viruses into South African wetlands that infect sympatric wild ducks and geese that in turn move extensively throughout the country. Interactions between wild ducks and ostriches are well-documented, and the transmission of AIV between these species was confirmed by the genesis of the HPAI

H5N2 ostrich outbreak strain of 2004 from the common ancestor of an LPAI H5N2 virus, isolated from an Egyptian goose (*Alopochen aegypticus*) in the same year.

Since the 1990s, outbreaks of velogenic Newcastle disease virus (NDV) in South Africa were caused by three distinct genotypes. The historic genotype VIII (lineage 3), enzootic since the 1970s, was replaced by genotype VIIb (lineage 5b) in the early 1990s during a panzootic, and VIIb was replaced by genotype VIId (lineage 5d) in 1999. Lineage 5d reemerged in 2003, causing an outbreak in 2004 in commercial and backyard flocks. Phylogenetic evidence indicated that pigeon paramyxmoviruses (ND), similar to the aforementioned genotypes, were introduced into South Africa from Eurasian sources on several occasions. The consecutive replacement of Eurasian NDV genotypes, the lack of a unique South African lineage and the lack of conclusive evidence of a true cyclic reservoir challenges the mindset that NDV is enzootic in South Africa. This is the first time that molecular techniques were used to elucidate the status of these two important diseases in South Africa. It is evident that the region is at risk to the introduction of HPAI strains carried by migratory birds, but that with improved control, the enzootic status of NDV could be reversed, with important economic implications for the poultry industry. Improved biosecurity is therefore key to the prevention of future outbreaks of AI and ND in South Africa.

PAPERS AND CONFERENCE PROCEEDINGS

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Posters

C Abolnik, E Cornelius SPR Bisshopp, M Romito, D Verwoerd. Phylogenetic analyses of genes from South African LPAI viruses isolated in 2004 from wild aquatic birds suggests introduction by Eurasian migrants. OIE/FAO International Scientific Conference on Avian Influenza, 7-8 April 2005, Paris.

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LIST OF ABBREVIATIONS

°C	:	degrees Celcius
%	:	percentage
A	:	adenine
AGID	:	agar gel immunodiffusion test
AI	:	avian influenza
AIV	:	avian influenza virus
APMV-1	:	avian paramyxovirus type 1
bp	:	base pair
C	:	cytosine
cRNA	:	complimentary RNA
DIVA	:	differentiating infected from vaccinated animals
e.g.	:	for example
ELISA	:	enzyme-linked immunosorbent assay
EM	:	electron microscope
ER	:	endoplasmic reticulum
EU	:	European Union
F_0	:	fusion protein precursor
G	:	guanidine
GIS	:	geographic Information System
GPMV	:	goose paramyxovirus
H_0	:	hemagglutinin protein precursor
HA	:	hemagglutinin protein
HA	:	hemagglutination
HAU	:	hemagglutiniating units
HI	:	hemagglutinin inhibition
HN	:	hemagglutinin-neuraminidase protein
HPAI	:	high pathogenic avian influenza
HPNAI	:	high pathogenic notifiable avian influenza
ICPI	:	intracerebral pathogenicity index
IVPI	:	intravenous pathogenicity index
kDA	:	kilodaltons
KZN	:	KwaZulu-Natal province
L	:	large polymerase protein
LPAI	:	low pathogenic avian influenza
LPNAI	:	low pathogenic notifiable avian influenza

M	:	matrix protein
M1	:	matrix protein-1
M2	:	matrix protein-2
MAb	:	monoclonal antibody
MDT	:	mean death time
min	:	minutes
M-MLV	:	mouse Moloney murine leukemia virus
mRNA	:	messenger RNA
NA	:	neuraminidase protein
NASBA	:	nucleic acid sequence-based assay
NDA	:	National Department of Agriculture
NeuAc	:	N-acetylneuraminic acid
NeuGc	:	N-glycolylneuraminic acid
NI	:	neuraminidase inhibition
ND	:	Newcastle disease
NDV	:	Newcastle disease virus
NP	:	nucleoprotein
NS	:	non-structural
nt	:	nucleotide
NWP	:	North-West province
OIE	:	Office International des Epizooties (World animal health organization)
ORF	:	open reading frame
OVI	:	Onderstepoort Veterinary Institute
P	:	phosphoprotein
PA	:	Polymerase A
PB1	:	Polymerase B1
PB2	:	Polymerase B2
PCR	:	polymerase chain reaction
PPMV-1	:	pigeon paramyxovirus type 1
RBS	:	receptor binding site
RNA	:	ribonucleic acid
RNP	:	ribonucleoprotein
RT-PCR	:	reverse transcription polymerase chain reaction
rRT-PCR	:	real-time reverse transcription polymerase chain reaction
SA	:	sialic acid
SAN	:	specific antibody negative

SAPA	:	South African Poultry Association
sec	:	seconds
SPF	:	specific pathogen free
T	:	thymidine
TNFα	:	tumor necrosis factor alpha
UAE	:	United Arab Emirates
UTR	:	un-translated region
VLA	:	Veterinary Laboratory Agency
vRNA	:	viral ribonucleic acid
RNAP	:	viral RNA-dependent RNA polymerase
vRNP	:	viral ribonucleoprotein
WHO	:	World Health Organization

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