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Prevalence of Pathogens in Great White Pelicans *(Pelecanus onocrotalus)* from the Western Cape, South Africa

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Abstract

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Great White pelicans ***(Pelecanus onocrotalus)*** *breed in Africa, Europe and Asia from Greece to Vietnam. In Africa, the Western Cape is probably the only place in the world where pelican numbers have shown a sustained increase over the past few decades. Nothing is known regarding the prevalence of pathogens present in these populations. Therefore, 50 Great White Pelicans from Western Cape were tested for the presence of various bacteria and viruses by polymerase chain reaction (PCR). It was observed that 49 pelicans were positive for* ***Mycoplasma*** *spp., 22 for* ***Salmonella*** *spp. and 3 for NDV, making these a potential risk to domesticated avian species, as well as human beings. This information is to be considered while planning any expansion/control programme for this species.*

Key words: Great white pelicans, pathogens, *Salmonella*, *Mycoplasma*, NDV.

Introduction

Great White pelicans (*Pelecanus onocrotalus*) breed in Africa, Europe and Asia from Greece to Vietman. The world's population is believed to be about 85 000 pairs, of which about 80% are in Africa (del Hoyo *et al.*, 1992).

Although the population of Great White pelican is on decline worldwide, their numbers have increased substantially in the Western Cape in the last decades.

Traditionally, they feed at freshwater wetlands and at estuaries (Crawford *et al.*, 1995). The construction of a plethora of farm dams and the practice of stocking them with freshwater Carp *Cyprinus carpio* has increased food availability for the pelicans. Also, in recent years, they have started feeding on offals at pig and chicken farms in the Greater Cape Town area. This behaviour

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places them at risk to mass poisoning incidents. On the other hand, wild birds are a serious threat to public health because they carry emerging zoonotic pathogens, either as a reservoir host or by dispersing infected arthropod vectors (Reed *et al.*, 2003).

In order to better understand and conserve the population of Great White Pelican in the Western Cape and for ecological reasons, it is essential to study, not only the impact that dumped offal is having on breeding productivity, but also the prevalence of pathogens present in these animals.

Materials and Methods

In the present study we investigated the prevalence of *Mycoplasma spp.*, *Salmonella spp.*, *Chlamydia psittacci*, avian infectious laryngotracheitis virus (ILTV, *Herpesvirus*), Newcastle disease virus (NDV, *Paramixovirus type I*), infectious bronchitis virus (IBV, *Coronavirus*), and avian influenza virus (AIV, *Influenza*, A type) by polymerase chain reaction (PCR) in a community of Great White pelicans in the Western Cape, South Africa, capturing 50 birds using modified leg traps (King *et al.*, 1998) on a pig farm.

A total of 50 tracheal and 50 cloacal samples were taken from apparently healthy pelicans using sterile swabs. The samples were preserved in a cell growth medium RPMI-1640 (Roswell Park Memorial Institute, Sigma, St. Louis, MO, USA) with 10% horse serum.

DNA extractions were performed following the protocol developed by Tola *et al.* (1997), RNA extractions were performed with Tripure isolation reagent (Roche, IN, USA) and the RNA was transcribed to cDNA with the iScript cDNA synthesis kit (Biorad, CA, USA) according to the instructions of the manufacturers. A 5 µl of DNA sample was amplified in a 25 µl reaction mixture containing 10 pM of each primer and 12.5 µl of iQ SYBR Green supermix (Biorad, CA, USA). The primers and conditions for the PCR were followed according to the protocols described previously (Table 1). PCR was performed in a MyiQ Single-Color Real-Time detection system (Biorad, CA, USA).

Results and Discussion

The PCR results showed that 49 (98%) pelicans were positive to *Mycoplasmas spp.*, 22 (44%) to *Salmonella spp.* and 3 (6%) to NDV. All birds were negative for *Chlamydia psittacci*, ILTV, IBV and AIV.

Regarding avian mycoplasma, only four species (*M. gallisepticum*, *M. synoviae*, *M. meleagridis* and *M. iowae*) are known to be pathogenic (Bradbury, 2005). Although we have not determined yet which Mycoplasma species were present in the studied population, it is known that the adaptation of, for example, *M. gallisepticum* to a free-flying avian host presents potential problems for the control of mycoplasmosis in commercial poultry (Luttrell *et al.*, 2001).

NDV, a pathogen of social and economical importance, is known to be disseminated throughout the world by migratory and wild birds (Soares *et al.*, 2005). There is also a general concern that NDV wild strains, circulating among wild birds, can become highly pathogenic in poultry (Shengqing *et al.*, 2002). The main threat comes from psittacine species and racing pigeons (Cross, 1991), but in some regions, other wild birds may represent an important role in spreading the disease. Pfitzer *et al.* (2000) reported the presence of NDV antibodies in wild aquatic birds in an intensive ostrich farming area in South Africa and it was suggested that these birds could represent an important reservoir of NDV in the area.

In this context, the high percentage of *Salmonella* and *Mycoplasma spp.* and the presence of NDV shown by this Great White pelican population indicated that they were an important reservoir for these organisms.

Table 1
PCR conditions

Pathogen	Primers	Target	References	PCR conditions (40 cycles)*		
				Denaturation	Annealing	Extension
Mycoplasma spp.	5'-CCA GAT TCC TAC GGG CA-3' 5'-TGC GAG CAT ACT ACT CAG GC-3'	16S ribossome subunit	Spergser *et al.* (2002)	95C for 40 sec	64C for 40 sec	72C for 1 min
Salmonella spp.	5'-GCA AGA CAC TCC TCA AAG CC-3' 5'-CCT TCC CAC ATA GTG CCA TC-3'	InvA gene	Malorny *et al.* (2003)	95C for 40 sec	60C for 40 sec	72C for 40 sec
Chlamydia psittacci	5'-GTG AAA TTA TCG CCA CGT TCG GGC AA-3' 5'-TCA TCG CAC CGT CAA AGG AAC C-3'	OmpA gene	Hewinson *et al.* (1997)	95C for 40 sec	62C for 40 sec	72C for 40 sec
ILTV	5'-CGT GGC TTC ACC AGC AA-3' 5'-CGA GTA AGT AAT AGG CT-3'	Thymidine kinase gene	Scholz *et al.* (1994)	95C for 40 sec	57C for 40 sec	72C for 1 min
AIV (A Type)	5'-AGA TGA GTC TTC TAA CCG AGG TCG-3' 5'-TGC AAA AAC ATC TTC AAG TCT CTG-3'	Matrix gene	Spackman *et al.* (2003)	95C for 30 sec	60C for 30 sec	72C for 40 sec
NDV	5'- GGA GGA TGT TGG CAG CAT T-3' 5'- GTC AAC ATA TAC ACC TCA TC-3'	S1 glycoprotein gene	Pang *et al.* (2002)	95C for 40 sec	60C for 40 sec	72C for 40 sec
IBV	5'-TGG TTG GCA TTT ACA CG GGG-3' 5'-CAA TGG GTA ACA AAC AC-3'	F0 fusion protein coding sequence	Liu *et al.* (2003)	95C for 40 sec	60C for 40 sec	72C for 40 sec

*An initial cycle of 95C for 2 min was performed for all PCR.

Further sampling will reveal whether there are differences in the prevalence of pathogens between the three main southern African populations, as could be expected due to the different foraging behaviours observed. A comparison between the natural- and offal-feeding pelicans will help assess the risk of transmission of diseases from domestic animals to scavenging pelicans in the Western Cape.

It is concluded that though pelicans themselves are on decline worldwide, there are potential risk to domesticated avians and human beings for spread of the disease. These factors are to be borne in mind while planning their population expansion or control.

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पी. एस्सुनकावो, एम. मचादो, सी. दे ला फे, ए.एस. रामिरेज, आर.एस. रोसालेस, एन.टी. अंतुनेस, सी. पोवेदा, जे.बी. पोवेदा। पश्चिमी केप दक्षिणी अफ्रीका के श्वेत पेलिकन (पेलिकेनस ओनोक्रोटेलस) में रोगाणुओं की व्यापकता।

वृहत श्वेत पेलिकन (पेलिकेनस ओनोक्रोटेलस) में अफ्रीका, योरोप और एशिया में ग्रीस से लेकर वियतनाम तक संवर्धन होता है। विश्व में संभवत: अफ्रीका का पश्चिमी केप है जहाँ पर पिछले कई दशकों से पेलिकन की संख्या में प्रतिपालित वृद्धि हुई है। विभिन्न स्थानों पर पेलिकन में रोगाणुओं की व्यापकता की कोई भी जानकारी नहीं है। इसलिए पश्चिमी केप के 50 बृहत् श्वेत पेलिकन में पालीमरेज श्रृंखला प्रतिक्रिया से विभिन्न सूक्ष्माणुओं और विषाणुओं की उपस्थित के लिए जांच की गयी। उनमें 49 पेलिकनों में माइकोप्लाज्मा प्रजाति, 22 में साल्मोनेला प्रजाति और 3 में एनडीवी पाये जाने से यह दिखता है कि अन्य पक्षियों और मनुष्यों के लिए सशक्त खतरा हैं। इस प्रजाति के किसी प्रकार के विस्तार या नियंत्रण का कार्यक्रम बनाते समय इस सूचना को भी ध्यान में रखने की आवश्यकता है।