

First record of white stork in a birdstrike in South Africa above 3,300 m AGL

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Abstract: Birdstrikes to aircrafts are increasing on an annual basis and pose significant aviation safety risks. Identification of the birds involved is key to developing mitigation strategies. Often the only information available to make identifications are feather and/or tissue samples. Relying on feathers alone to identify the bird species requires special expertise and access to museum collections for specimen comparisons. In 2017, feathers and tissue samples were recovered from the engine cowling of an airplane that had just landed at the Oliver Reginald (O. R.) Tambo international airport in South Africa after striking a bird at 3,353 m. To confirm the bird species, we sequenced a region of cytochrome c oxidase I (COI) for the unknown sample and compared the results to the Barcode of Life Database (BOLD) and National Centre for Biotechnology Information (NCBI) GenBank databases. Comparisons to these databases indicated that the species involved in the birdstrike incident was a white stork (*Ciconia ciconia*; 99.6–100% similarity). This was the first known record of a white stork involved in a birdstrike in South Africa and is important because it provided evidence of a high-altitude birdstrike. Availability of publicly accessible DNA barcoding databases that include all potential bird species from various geographic regions is a valuable tool in species identification and can aid wildlife management strategies at airports to reduce the risks associated with birdstrikes.

Key words: birdstrikes, *Ciconia ciconia*, mitochondrial DNA, South Africa, white stork

BIRD–AIRCRAFT COLLISIONS (birdstrikes) cause economic losses and damage to the aircraft (Robinson 2000, Allan 2006, DeVault et al. 2018) and can lead to human fatalities (Neubauer 1990). It has been reported that several bird species use aerodrome habitats for foraging, perching, roosting, and as nesting sites despite potential lethal threats from aircrafts due to very low levels of human presence and incidental disturbance (Kershner and Bollinger 1998, Sodhi 2002, Soldatini et al. 2010, DeVault et al. 2017).

Areas surrounding most airports in developing countries are densely populated and industrialized and often, despite International Civil Aviation Authority best practice guidelines, contain refuse dumps and landfills that may attract several bird species (Froneman 2000, Baxter 2001). In addition, birdstrikes likely occur due to the increased speed and reduction of engine noise levels of modern aircrafts (Thorpe 1997). To alleviate birdstrike risks, accurate species identification and determination of why these species may be attracted to these areas is



Figure 1. White stork (*Ciconia ciconia*; photo courtesy of Albert Froneman).

of importance in order to employ a variety of control and mitigation measures. For example, it has been reported that manipulation of a single food source within airport property reduced use by tree-swallows (*Tachycineta bicolor*; Bernhardt et al. 2009).

In South Africa, Airports Company of South Africa (ACSA) has implemented an integrated wildlife hazard management program, which includes both reactive and proactive bird hazard management techniques (Froneman 2000). Analysis of bird presence is based on an electronic monitoring program (Oliver Reginald [O. R.] Tambo International Airport and Durban International Airport), which includes data on bird species, number present, grid reference, and behavior collected by bird patrols (3 or 4 times/day). Analysis of this data has identified potentially hazardous species on South African airports, which include the Blacksmith lapwing (*Vanellus armatus*), crowned lapwing (*V. coronatus*), black-headed heron (*Ardea melanocephala*), Hadeda ibis (*Bostrychia hagedash*), African sacred ibis (*Threskiornis aethiopicus*), helmeted guineafowl (*Numida meleagris*), and yellow-billed kite (*Milvus aegyptius*; Froneman 2000).

Currently, ACSA—in collaboration with the South African National Biodiversity Institute, National Zoological Garden (SANBI NZG) and BirdLife South Africa—are in the process of expanding current DNA barcoding databases such as the Barcode of Life Database (BOLD) and National Centre for Biotechnology

Information (NCBI) GenBank by adding sequence data (using protocols similar to those developed in Australia; Christidis et al. 2006) from voucher specimens obtained from various geographic areas in South Africa that are currently not represented. These databases can be used for the identification of bird remains involved in birdstrikes at South African airports.

Techniques used to identify species involved in birdstrikes traditionally included microscopic identification of feathers and feather fragments collected after collisions (Manville 1963, Laybourne 1974, Brom 1986), keratin electrophoresis (Ouellet 1994), and morphological analysis where whole carcasses, feathers, or feather fragments are obtained. More recently, application of DNA barcoding for use in birdstrike identification have been described in Dove et al. (2008a, b). The DNA barcoding provided a reliable method to identify species using material such as blood, feathers, and tissue (Hebert et al. 2004). Amplification and sequencing using universal primers cytochrome c oxidase I (COI) provide a reliable test for the identification of species in wildlife forensic cases (Branicki et al. 2003, Verma and Singh 2003, Wu et al. 2005). In addition, the COI gene can also be used to barcode most animal life (Hebert et al. 2003a, b) and has been used to identify bird species involved in airstrikes (Dove et al. 2008a, b).

In this study, the pilot reported that the plane struck a flock of 2–10 birds at approximately

3,353 m above ground level (AGL) following departure from O. R. Tambo International Airport. Immediately following the incident, engine indicators were inspected and were observed to be normal; however, damage to the number 1 engine cowling was subsequently reported to the pilot by a crew member. Due to the unknown damage to the engine, the plane returned to O. R. Tambo International Airport and an uneventful landing was made.

Most birdstrikes (74%) occur within 153 m AGL and only 7% above 1,067 m AGL (Dolbeer 2006). Birdstrikes with vultures have been reported at 11,280 m AGL in Africa with a Rüppell's griffon vulture (*Gyps rueppelli*; Laybourne 1974). Dolbeer (2006) reported only 2 Ciconiiformes (Hérons/egrets/bitterns/storks), which were struck in the United States between 1990 and 2004 above 1,067 m AGL. The International Civil Aviation Organization (ICAO) Ibis database reported 4 white stork (*Ciconia ciconia*; Figure 1) strike incidents between 2008 and 2015 (ICAO 2017) thus indicating that white storks were not regarded as a high-risk birdstrike species, especially in situations where birds are flying at a high altitude over an aerodrome, as was the case with the incident discussed in this study. In this study, we report, to our knowledge, the first known record of a white stork birdstrike in South Africa at such high altitude and the current DNA methods used to identify species in the bird collision with an aircraft in South Africa.

Materials and methods

The airline operator (Mango Airlines) collected tissue and black and white feathers following a birdstrike incident and submitted the samples to the bird and wildlife control staff employed by ACSA on November 19, 2017. Some local airlines carry DNA sampling kits onboard and have assisted in collecting valuable DNA samples for analysis. The unknown sample consisted of tissue and feathers and was given accession number EXT02476 upon receipt by the SANBI NZG laboratory and stored at -20°C at the SANBI NZG biobank. Genomic DNA was isolated from the samples using the Applied Biosystems™ PrepFiler™ Forensic DNA extraction kit (Thermo Fisher Scientific, Waltham, Massachusetts, USA) following the

manufacturer's protocol.

In the present study, a region of the mitochondrial gene COI was amplified and sequenced using primer sets provided in Hebert et al. (2004) and Bitanyi et al. (2011) using the Applied Biosystems 2720 Thermal Cycler. Polymerase chain reaction (PCR) amplification was carried out in a total volume of 25 micro litres (µL). The PCR was conducted with Thermo Scientific DreamTaq Green PCR Master Mix (2X), which contains DreamTaq DNA polymerase, DreamTaq Green buffer, 4 mM MgCl₂ and dNTPs. The final reaction conditions were as follows: 1 X PCR Mastermix, 10 pico mol (pmol) of each of the forward and reverse primer, and 50–100 nano gram (ng) genomic DNA template. The conditions for PCR amplification were as follows; 5 minutes at 95°C denaturation, 35 cycles for 30 seconds at 95°C, 30 seconds at 50°C, and 1 minute at 72°C. The PCR products were separated by electrophoresis in a 2% agarose gel for 30 minutes at 100 volts in 1 x Tris-Borate-EDTA Buffer (TBE) followed by clean-up of PCR products using the ExoSAP protocol. The purified PCR products were cycle sequenced using Applied Biosystems™ BigDye™ Terminator v3.1. Cycle Sequencing Kit (Thermo Fisher Scientific, Waltham, Massachusetts, USA). The cycle sequencing products were purified with the BigDye XTerminator Purification Kit (Life Technologies) before sequencing on an ABI 3500 genetic analyzer (Life Technologies) in forward and reverse. The raw sequence data for each mtDNA gene were edited and aligned in GENEIOUS v10.2.3. The sequences obtained for the unknown sample were blasted against the mitochondrial DNA sequence database of NCBI using the Basic Local Alignment Search Tool (BLAST) program (Altschul et al. 1990) and the BOLD database (Ratnasingham and Hebert 2007).

Results

Amplification was successful for the unknown sample. Sequence analysis of the unknown sample using BLAST of the NCBI nucleotide database (GenBank) identified the closest species match was a white stork (99.6% match). Analysis in BOLD confirmed the match (100% match; Figure 2). We concluded that a white stork was involved in the birdstrike,

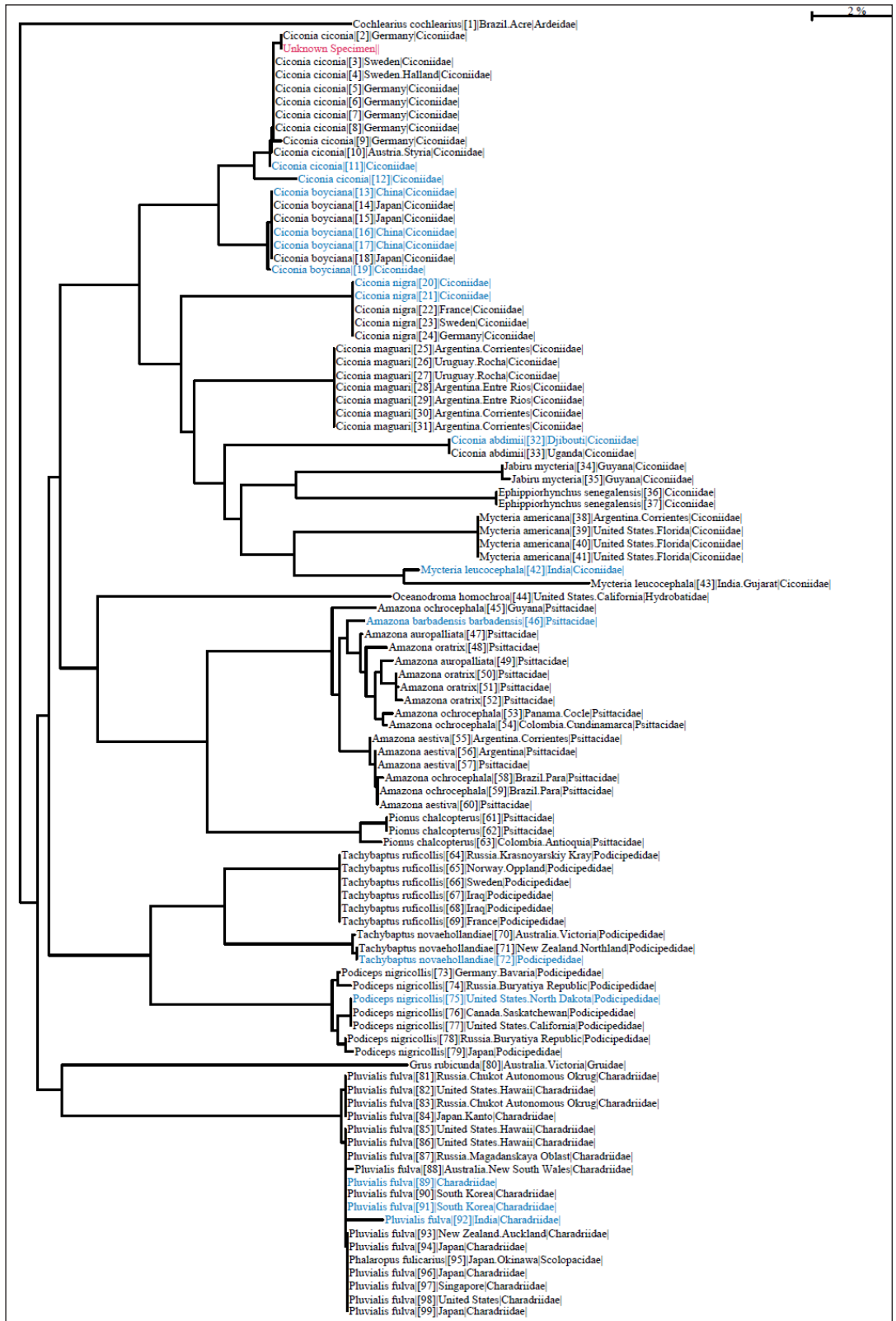


Figure 2. Taxon ID tree of barcode records generated from the Barcode of Life Database (BOLD), indicating the assignment of the unknown sample to the white stork (*Ciconia ciconia*) clade.

which, in addition, was consistent with the coloration of the feather with this species.

Discussion

This is, to our knowledge, the first record of a birdstrike incident involving white stork in South Africa and documents an altitude record for this species. It has been reported that on average, 1 strike occurs every 18 days in South Africa (Byron and Downs 2002). However, the number of birdstrikes is continually increasing at a global level (ICAO 2017). Bird collisions are a threat to commercial aircrafts at several South African airports with various techniques being employed to control bird numbers on airfields to reduce the number of birdstrikes (Byron and Downs 2002).

Airports Company South Africa has employed full-time bird and wildlife officers at ACSA airports to manage the associated risk with bird and wildlife strikes. Certain airports both in South Africa and internationally deploy potential predators such as trained domestic dogs (*Canis lupus familiaris*) and many other methods such as fake birds, remote controlled aircrafts etc. to reduce the risk of birds feeding and breeding on airside (Froneman and van Rooyen 2003). This, in turn, assists in reducing the number of birds that may be present on the aerodrome. Populations of birds are closely monitored, and trends in bird species and numbers present are utilized to analyze predictive trends.

Habitat management is an indirect method for managing the presence of birds and wildlife on airside, but first the species involved must be identified. Environmental factors such as standing water, vegetation management, which includes grass height management, refuse site management, and areas that may be attractive for breeding and perching are managed to prevent the attractiveness of these areas (Brown et al. 2001). Each airport site may require different management strategies in birdstrike prevention based on species involved in order to minimize animal impact on the aviation industry. However, the ability to identify bird remains on aircrafts to species level is fundamental for risk identification and risk mitigation, as the species involved could potentially assist with identifying off-airport feeding and breeding sites and possibly flight paths of bird species involved (Byron and

Downs 2002, Dove et al. 2008a, b).

The use of molecular analysis to identify species involved in birdstrikes and the continued expansion of comprehensive birdstrike reference barcode databases will greatly assist in identifying species most frequently involved in birdstrikes at the respective ACSA airports across South Africa. This case study also underscores the importance of documenting information on altitudes of birdstrikes to enhance our ornithological knowledge of bird flight altitudes and behavior. Such information will form the baseline for the development of site and/or species-specific management guidelines, which will lead to the implementation of the correct site-specific strategy and will assist with future aircraft collision investigations in South Africa as it does in other parts of the world.

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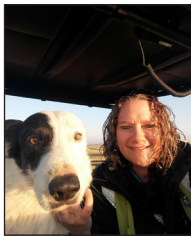


and phylogenetic relationships among species or populations. She is also using genetic technologies in conservation management and for forensic science applications to support enforcement for illegal wildlife activities. She is also a member of the Scientific Authority of South Africa and the IUCN African Conservation Genetics Specialist Group.

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