



Feeding Habits and Diet Composition of the African Sacred Ibis in Selected Water-Logged Sites in Dar es Salaam, Tanzania

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Received 26 Oct 2022, Revised 24 May 2023, Accepted 27 May 2023 Published June 2023

DOI: <https://dx.doi.org/10.4314/tjs.v49i2.9>

Abstract

The African Sacred Ibises *Threskiornis aethiopicus* are generalist birds that feed on many invertebrates and some vertebrates. However, the diet composition of local *T. aethiopicus* populations in wetland areas in Dar es Salaam Tanzania was unknown. Sacred Ibises from eight water-logged areas in Dar es Salaam City, Tanzania were studied to determine their feeding habits and diet composition using direct observations and molecular analysis of faecal samples. Snails species identified from sites frequented by *T. aethiopicus* included *Lymnaea natalensis* (61.62%), *Melanoides tuberculatus* (20.64%), *Bulinus* sp. (17.66%) and *Anisus* sp. (0.08%), while molecular analysis of the birds' diet revealed presence of mostly non-native invasive molluscs (*Physa acuta*, *Planorbella trivolvis* and *Radix natalensis*), fungus (*Candida parapsilosis*) and protozoan ciliate (*Vorticella striata*). There was no significant association between bird and snail abundance ($r = 0.2141$, $p > 0.05$, ns) although snail diversity showed strong negative relationship ($r = -0.7904$, $p < 0.05$). Our findings indicate that the African Sacred Ibises do not preferentially feed on snails, but do so as part of their broader diet. The results confirm that the African Sacred Ibis is a generalist feeder and therefore, ecological impacts of its feeding strategies particularly on invasive snails should be further investigated.

Keywords: African Sacred Ibis, feeding, diet, molluscs, generalist.

Introduction

The African Sacred Ibis (*Threskiornis aethiopicus*) is a wading bird of the order Ciconiiformes and family Threskiornithidae (Smits et al. 2010). It is naturally found in marshy or swampy areas of Africa including Tanzania (Kopij 1999, Tanzania Bird Atlas 2013). The population of African Sacred Ibis in the country has increased in the past three decades partly due to availability of food in garbage dumps (Tanzania Bird Atlas 2013). As detailed elsewhere (Birdlife International 2012, Ndinadyo 2019), the African Sacred Ibises are known to have a broad diet composition. They feed mostly on invertebrates and some few vertebrates

(Williams and Ward 2006). Their diet includes insects, fresh-water beetles, crustaceans, fish, frogs, eggs and nestlings of other bird species (Birdlife International 2012). The birds also forage and probe for different types of aquatic invertebrates including some molluscs (Marion 2013). According to Clergeau and Yésou (2006), the African Sacred Ibises have a wide diet spectrum and are able to use various habitats such as meadows, rubbish dumps, marshes, seashore and ploughed fields. A research survey conducted in South Africa found large numbers of the African Sacred Ibis at sewage farms, piggeries, garbage dumps as well as in the marshes and dams albeit in few numbers

(Clarke and Clarke 1979). The birds can feed in a solitary manner although at other times they can congregate in small groups as reported elsewhere (Williams and Ward 2006). When in groups, the African Sacred Ibises are known to scare mother cormorants off their nest and then eat their eggs or chicks (Williams and Ward 2006).

In general, most bird species including possibly *T. aethiopicus*, feed on various types of snails and other molluscan species (Clergeau and Yésou 2006, Smits et al. 2010). Snails are particularly known to form a major and an essential part of the diet of many avian species (Barker 2004). Some studies have reported on the importance of snails as the principle source of calcium for avian species (González-Solis et al. 1996). Calcium from snail shell is necessary for reproduction in birds. It is the major determinant of reproductive success in birds such that if the amount of calcium becomes low, the birds may delay breeding (Pelicia et al. 2009). At one time, the absence of bird species such as oystercatchers, curlews and plovers that fed on salt marsh snails led to the latter overgrazing the grasses turning productive marshes into mudflats (Silliman and Bertness 2002). This was an evidence that, predation of snails by birds controls their numbers thereby regulating salt marshes productivity and ecosystem health. It has been concluded therefore that, the presence of predatory birds in some habitats guaranteed the ecosystem functioning and their services (Silliman and Bertness 2002). Despite this level of understanding, there are various aspects related to avian feeding ecology that are still unknown including local diet composition and feeding strategies for some birds such as the African Sacred Ibis.

Studies on the feeding behaviour of most avian species including the African Sacred Ibis have largely been too general looking mostly at the birds' overall foraging habits. Thus, the diet composition of the African Sacred Ibis in Dar es Salaam City, Tanzania has not been studied. Furthermore, the impact of the African Sacred Ibis' feeding strategy on the abundance of its local prey was also not known. The present study was thus

initiated to establish the feeding strategies and diet composition of the African Sacred Ibis in selected water-logged areas of Dar es Salaam City. Behavioural observations and DNA-based molecular procedures were used to study the birds' feeding habits and diet composition, respectively. The data obtained were also used to draw conclusions on the ecological impacts of the bird's predatory habits on abundance and diversity of invertebrates such snails in the study area. The study's findings provide baseline information on the feeding habits of the local population of the African Sacred Ibis in Dar es Salaam Tanzania. The information broadens our understanding of the diet and feeding ecology of the African Sacred Ibis in its natural habitats in an urban setting.

Materials and Methods

Study area

The study was conducted at eight sampling sites in Dar es Salaam City, Tanzania (Figure 1). The sites were chosen based on the availability of suitable conditions and microhabitats for some invertebrates and the confirmed presence of the African Sacred Ibis in the area (Tanzania Bird Atlas 2013). The range of African Sacred Ibis and the map of certain molluscan species naturally overlap since they both share a preference for wetland habitats (Tanzania Bird Atlas 2013), which suggests that the birds might influence the distribution and abundance of the invertebrates in the study area. The studied sites included Kigogo (along Msimbazi River), Makurumla, Jangwani, Kinondoni, Vingunguti, Sipenko, Msewe and Bwawa la Boko (Figure 1). Dar es Salaam City, which is also the commercial capital of Tanzania, experiences two rainy seasons; the short rainy season (October–December) and main rainy season (March–May) with an annual average rainfall of 1000 mm (Hamisi 2013). The lowland areas of Msimbazi River and Jangwani wetland are some of the ecological zones characterizing the city and have favourable habitats for occurrence and survival of freshwater invertebrates such as snails.

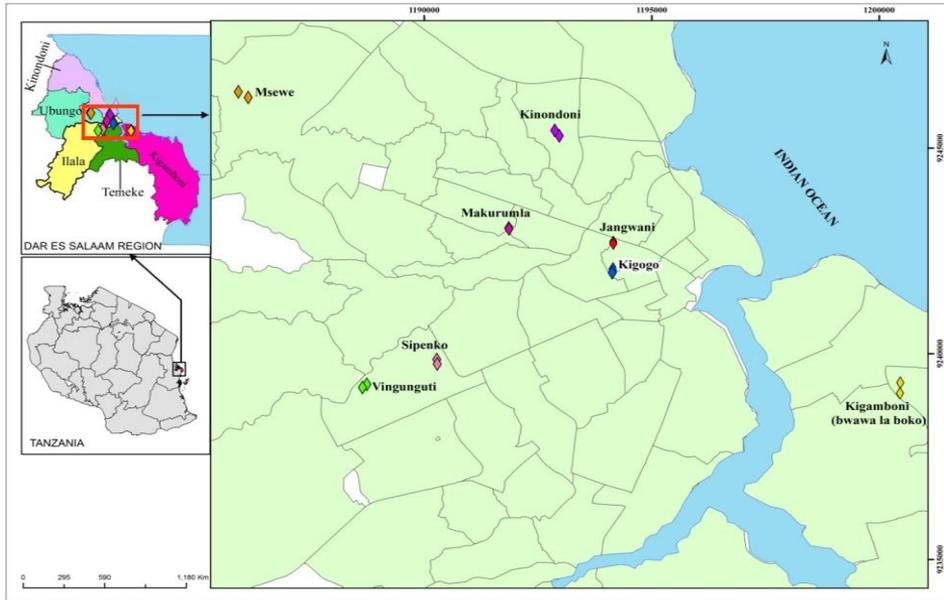


Figure 1: Location of Dar es Salaam Region in Tanzania (bottom left inset) and study area demarcated by a square (top left inset) and the sampling sites indicated by diamond shapes (right).

Study permits and approval

The approval to conduct the study was granted by the University of Dar es Salaam (Reg. No. DUCE-18087). The permission to conduct fieldwork surveys in swamps and ponds in the study sites was obtained from the Directors of respective Municipal Councils in Dar es Salaam City. Samples were obtained from snails and birds in accordance with accepted animal welfare and guidelines for working with wildlife subjects for research purposes (TAWIRI 2012). Collected snails were immediately returned to their original sites after identification and counting, while only a few representative snails were taken to the laboratory for morphological and molecular identification processes.

Study design and sampling strategy

A pilot study was conducted in the study area from February to March 2017 to identify suitable sampling sites for invertebrates (freshwater snails) and the presence or absence of the African Sacred Ibises. The convenience and purposive sampling strategies were used by choosing study sites based on informants' reports on the presence

of the African Sacred Ibis in the area and our own observations on the availability of appropriate snail micro-habitats such as grass, litter, aquatic vegetation and sediments following guidelines in Oloyede et al. (2017). Snail sampling was conducted four times in each site with an interval of two weeks to coincide with snail oviposition and hatching of juveniles, which usually takes 6–14 days (Brown 1994). As described in Chane and Balakrishan (2016), each site was visited three times per day especially in the morning and evening when most of the birds are active. Visitations were made, first in the morning (06:30–10:30 hours), then afternoon (12:30–14:30 hours) and evening time (16:30–18:30 hours), resulting in a total of twelve visits per site. If the birds were sighted in a site, three hours were spent while watching them as they fed into water until they moved out onto land for defaecation (Ndinadyo 2019). The number of the African Sacred Ibises present in the flock at each site during each visit was counted, added and averaged daily to determine the birds' daily and overall abundance as described in Bibby et al. (2000) and Bart et al. (2004).

Snail sampling

In sites harbouring the African Sacred Ibis and those where the birds were not observed, snails were sampled using the quadrat method as described in literature (Dalila 2009, Clergeau et al. 2011). The quadrats, which were 1 m x 1 m in sizes, were placed at fixed intervals of 5 m and snails searched for within it for 15 minutes and counted, as suggested in Ndalila (2009) and Bakuza et al. (2017). A total of 10 quadrats were sampled per site. Snails were picked-up using a scoop measuring 24.5 cm long and 23 cm wide covered with a mesh wire of 0.2 cm x 0.2 cm pore size as recommended by Madsen et al. (2001). In places where scooping was not possible such as muddy sites, snails were picked-up by hand using long forceps as explained in Oloyede et al. (2017). Collected snails were kept into plastic containers that had perforated lids for fresh air entry while separating live snails from dead ones (empty shells). Snails from the same site were placed together in the same plastic containers (250 ml) containing mud and water from the area (Ngele et al. 2012). Representative snails were then taken to the laboratory for further identification. Snails were identified based on the shape of the shell, number of whorls and the position of the aperture by noting whether it was dextrally or sinistrially coiled as recommended (Mandahl-Barth 1962, Brown 1994).

Observations of African Sacred Ibis feeding strategies

As the Sacred Ibises were feeding mostly in swamps, a high-powered binocular was used to observe their diet from a distance of about 50 m away. Focal sampling was used by watching an individual bird feeding for up to 10 minutes to observe and record the type of food items (Altmann 1974, Chane and Balakrishan 2016). However, most birds remained in the flock, so focal sampling was not effectively carried out. Although longer distances of 100–200 m have been recommended during observations and counting of water birds in inland sites (Bart et al. 2004) and shorter distances (20–30 m) have also been used (Chane and Balakrishan

2016), the 50 m distance was used in the present study as the birds are less habituated to the presence of humans since they live or forage in or near urban densely populated areas (Clarke and Clarke 1979, Calle and Gawlik 2011, Figure 2). It was therefore possible to approach the flock up to approximately 50 m (Figure 3). However, the quick speed at which the birds captured and swallowed food, made it difficult to identify the type of food being consumed as also reported elsewhere (Murray and Shaw 2009).

After moving out of the stream or sewage ponds (Figure 3), the birds defecated and a single stool (approximately 100 g) was picked-up using a spatula and immediately preserved in 250 mL plastic vials containing 150 mL of absolute alcohol as recommended in King et al. (2015). To ensure that each sample was collected from an independent bird, stool was picked once, immediately after defaecation.

Molecular analysis of the African Sacred Ibis stool

A total of 96 faecal samples from 96 African Sacred Ibis at Vingunguti, Jangwani, Sipenko and Bwawa la Boko sites were processed, and DNA extracted from them using the Zymo kit guidelines and protocol (Jedlicka et al. 2013). Recovered DNA was quantified and checked for purity using NANODROP 2000 spectrophotometer. The conserved region of mitochondrial cytochrome oxidase 1 (MCO1) was amplified using the general invertebrate primers LCO1490–GTCAACAAATCATAAAGATATTGG and HCO2198–TAAACTTCAGGGTGACCAAAAAATCA and the specific primer for snails ETTS1 TGCTTAAGTTCAGCGGGT and ETTS2 TAACAAGGTTTCCGTAGGTGAA as described in Velásquez et al. (2002). The primers were used to amplify a 740 bp region of the COI gene from invertebrate remains in the birds' faecal DNA as described in Deagle et al. (2007). Details on PCR reactions and other analysis details can be found in Ndinadyo (2019). The samples were

sequenced using sequencer instrumental Model/ name: 3730X1/ Bioneer.

Data analysis

The significance of the difference in the mean of snail abundance between study sites with and without the African Sacred Ibis and the importance of variations in snail abundance among sites with the birds were analysed using one-way ANOVA test assuming equal means between the two snail populations. Moreover, Spearman rank

correlation test was applied to assess for the relationships between the abundance of the African Sacred Ibis and snail abundance and diversity. For molecular analysis data, sequences were aligned and trimmed on equal length of each nucleotide by using W function version 5.09. Edited sequences were further analysed using Basic Local Alignment Search Tool (BLAST) program optimized for highly similar sequences (BLASTN) as recommended by Jedlicka et al. (2013) and King et al. (2015).



Figure 2: The African Sacred Ibis flock feeding in their natural habitat at Sipenko site close to human settlement.



Figure 3: The African Sacred Ibis flock outside of water during which time, faecal samples were collected from them at Sipenko sewage pond, Dar es Salaam.

Results

Diet composition of the African Sacred Ibis

No food items consumed by the African Sacred Ibis were identified based on direct observations. On the other hand, PCR products amplified using general invertebrate primers displayed unclear bands (Figure 4). However, the primers ETTS1 and ETTS2 showed visible bands for 12 out of 16 samples of amplified PCR products (Figure 5). As shown in Table 1, matching of the

nucleotide sequences in GenBank for the African Sacred Ibis diet showed the presence of some molluscan species, namely; *Physa acuta* (Gastropoda: Physidae), *Radix natalensis* (Gastropoda: Lymnaeidae) and *Planorbella trivolvis* (Gastropoda: Planorbidae). Other organisms identified molecularly from the African Sacred Ibis diet included a ciliate *Vorticella striata* (Oligohymenophorea: Vorticellidae) and fungus *Candida parapsilosis* (Saccharomycetes: Saccharomycetaceae).

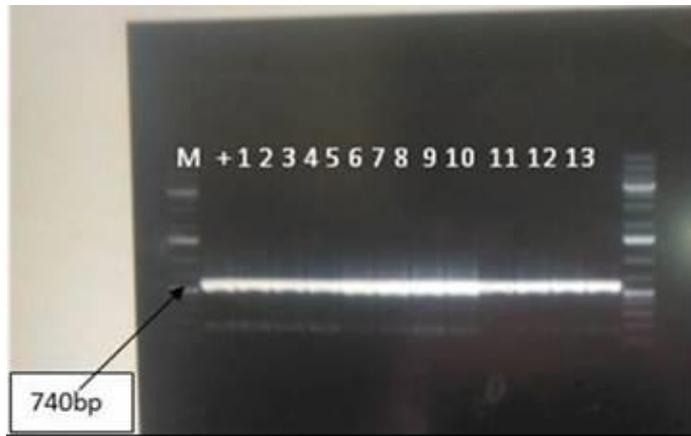


Figure 4: PCR products of the African Sacred Ibis stool DNA amplified by LCO 1490 and HCO 2198 primers.



Figure 5: PCR products of African Sacred Ibis stool amplified by ETTS1 and ETTS2 primers.

Table 1: Summarized results of blasted DNA sequences in GenBank with the marker O' Gene Ruler 1Kb ladder

Sample number	Species	Query cover	Expect (E)- value	Similarity (%)	Accession number
1	<i>Physa acuta</i> (Mollusc)	57	0.0	97	HQ 283259.1
2	<i>Candida parapsilosis</i> (Fungus)	98	0.0	99	KP 131777.1
3	<i>Vorticella striata</i> (Ciliate)	95	5e-90	81	KF 524427.1
4	<i>Radix natalensis</i> (Mollusc)	49	0.0	97	HQ 283257.1
5	<i>Planorbella trivolvis</i> (Mollusc)	83	0.0	90	AY 030403.1

Occurrence of African Sacred Ibis and snail abundance

A total of 115 African Sacred Ibises were observed at Jangwani, Vingunguti, Sipenko and Bwawa la Boko sites. Snails collected from the sites (n = 7191) were identified as *Lymnaea natalensis*: Gastropoda: Lymnaeidae (61.62%), *Melanooides tuberculatus*: Gastropoda: Thiariidae (20.64%), *Bulinus* sp.: Gastropoda: Planorbidae (17.66%) and *Anisus* sp.: Gastropoda: Planorbidae (0.08%). The sites with African Sacred Ibis populations had lower snail abundance compared to those lacking the birds (Figure 6) although no

association was observed between the abundance of snails and that of the Sacred Ibis (Spearman $r = -0.2141$, $p > 0.05$, ns; Figure 6). However, there was a statistically strong negative relationship between the African Sacred Ibis abundance and snail diversity ($r = -0.7904$, $p < 0.05$). Furthermore, the difference in the mean of snail abundance between the two categories of sites was negligible (One factor ANOVA test, $p > 0.05$, ns). One-way ANOVA indicated significant variations in snail abundance among sites harbouring the African Sacred Ibis (Tukey-Kramer test, $p < 0.05$).

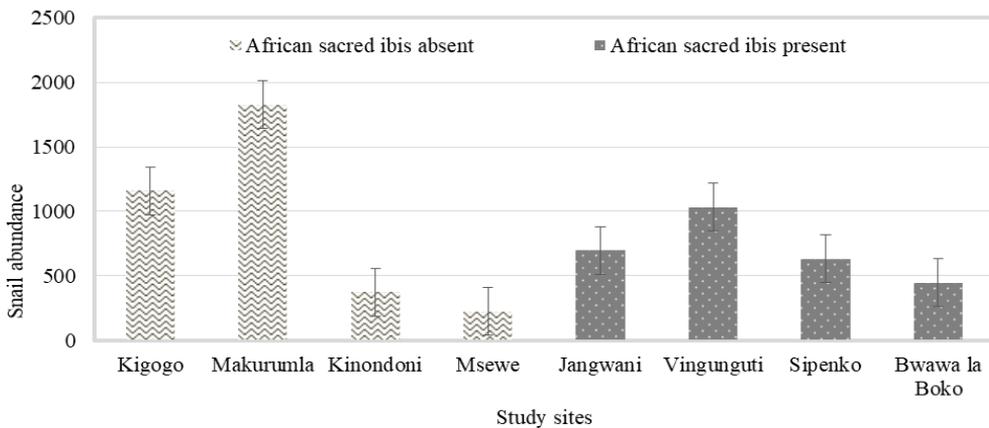


Figure 6: Variations of number of snails (vertical axis) in sites with and without African Sacred Ibis.

Discussion

Composition of the African Sacred Ibis diet

It was not possible to deduce the type of prey consumed by the Sacred Ibis based on

direct observations. While direct observations of bird's feeding may provide valuable information on the food-stuff they consume (Freitag et al. 2001), the method can have some shortcomings as well, rendering it

ineffective. Rosenberg and Cooper (1990) and Murray and Shaw (2009) have argued that, direct observation of avian diet is often restricted to predator-prey relationships as large or common prey may be easy to distinguish. Furthermore, while direct observations are effective in studying and identifying the diet of frugivorous and nectarivorous birds, the methods may not be suitable for elucidating the proportions of animal foods in the diets in birds (Rosenberg and Cooper 1990). This is because of the quick speed at which the birds capture food and swallow it, which makes it difficult for an observer to identify the type of food being ingested, particularly for small preys (Murray and Shaw 2009, Ndinadyo 2019). It has therefore been concluded that, while it is more advantageous to study the diet of wading birds (including the African Sacred Ibis) in the field by direct observations, the method should be used with caution. Researchers using the method should be aware of these limitations and state clearly the influence this could have on the conclusions made from their studies (Cezilly and Wallace 1988).

In contrast, PCR procedures in the present study identified several organisms as part of the Sacred Ibis diet (Figure 4). Sequences of DNA from the African Sacred Ibis faecal matter indicated that the birds included some molluscan species in their diet (Table 1). These were identified as *Physa acuta*, *Radix natalensis* and *Planorbella trivolvis* (Table 1). These findings are similar to those of Pungetti and MacIvor (2007) who reported that most bird species, feed on some snails. Snails are particularly known to form a major and an essential part of the diet of many species of birds (Barker 2004), and thus avian predation on snail is a widespread ecological phenomenon (Rosin et al. 2017). Literature has shown that, the inclusion of gastropods (snails) in the diet of birds has been known for many years (Wasef et al. 2015).

Studies have also shown that, different species of birds feed on various types of molluscan species (Rosin et al. 2011, Wada et al. 2012). Some birds, e.g. the blackbirds which belong to the genus *Turdus* are

believed to opportunistically feed on snails as also reported elsewhere (Cameron 1969). This could suggest that, the availability of food sources in the birds' habitats determines their preference for molluscs. For instance, Rosin et al. (2017) reported that, birds were found to occur in habitats where snails are abundant. Esayas (2017) further suggested that, the presence and preference of certain food types by individual birds were largely influenced by seasons. For instance, during the rainy season when stream water increase, worms and molluscs become available; hence, they serve as the major diet for birds. Some birds however, consume molluscs when other food sources become scarce (Bergmann et al. 2013), while to others, snails are their main source of energy. For instance, when water volume decreases during the dry season, insects and fruits rather than molluscs serve as the main diet for most birds. The African Sacred Ibis has been described as an opportunistic feeder that is able to switch to non-natural food items when the abundance of the prey is depleted (Chane and Balakrishan 2016). Thus, such variations of the avian diet with seasons or space confirm some claims that most birds' species are generalist feeders suggesting that what comes across them in a particular time becomes their staple food.

Likewise, Barker (2004) indicated that, bird predation on gastropods, especially terrestrial snails, may serve as the means of obtaining water. Terrestrial gastropods have developed a physiological mechanism for water retention into their body tissues, especially in hot and arid environments, which is why water content in the snails is higher compared to other food sources. Some species of birds such as *Pycnonotus* sp. and Song thrush (*Turdus philomelos*) prefer terrestrial gastropods over other food stuff. Thus, molluscs, and snails in particular, are ecologically important in maintaining food web and food chain intricacies partly through their capability of transferring calcium (and potentially other minerals) to higher trophic levels such as predatory birds (Adams and Wall 2000, Bureš and Weidinger 2003).

Other food types consumed by the African Sacred Ibis

DNA sequencing in the present study has revealed that, food items other than molluscs form key components of the African Sacred Ibis diet. The PCR analysis indicated that, in addition to molluscs, some ciliates and fungi are possibly accidentally swallowed in equal volumes by the African Sacred Ibis. The presence of the range of these molluscs, ciliates and fungus in the African Sacred Ibises reflect the diversity of the birds' diet. The results corroborate conclusions from other studies on the diet of the African Sacred Ibises and other birds, that, in general, birds have a broad diet spectrum, as they are adaptive and flexible in their feeding habits (Smits et al. 2010). Most of them are also known to feed on various vertebrates such as reptiles, lizards and frogs. They also consume several invertebrates such as grasshoppers, worms, crustaceans and snails (Clergeau and Yésou 2006, Smits et al. 2010).

As observed by other authors, e.g. Smits et al. (2010), the African Sacred Ibis can be described as being rather generalist feeders, foraging on a wide range of food items including snails. However, the significant effects demonstrated in the present analysis on the diversity of snails could have been due to other factors such as environmental features (habitat characteristics) as explained in Ndinadyo (2019). According to Marion (2013), the African Sacred Ibises are opportunist feeders with some flexibility in their diet as they can feed on whichever food item is available. The present observations are consistent with reports by Murray and Shaw (2009) and Marion (2013) that, the African Sacred Ibises are not selective feeders, but instead, consume many different types of food items. Nonetheless, the DNA sequences amplified in the present study produced a lower matching that it was not possible to precisely specify the species of the identified organisms.

Ecological and medical implications of the African Sacred Ibis feeding strategies

The present analysis of the African Sacred Ibis feeding behaviour and its effects on snail

abundance has found no significant impact to that effect, although the correlation analysis indicated a significant negative impact by the birds' abundance on the diversity of snails. Although many bird species forage on snails largely to obtain calcium for various body needs, the ecological impacts of these biological interactions is worth being discussed here. Most studies on avian feeding on snails have highlighted the impacts their foraging strategies have in their habitats. For instance, Richards (1977) studied the predation of Migrant Song Thrushes and Redwings on the Holy Island snails, Northumberland UK and concluded that, high abundance of predatory birds exerted significant pressure on resident non-cryptic snail morphs on which they fed. On the other hand, Hamilton (2000) has found that, birds directly reduce abundance of mussels tilting the ecosystem balance. Other works (Anderson and Coppolino 2009, Wenny et al. 2011) have observed that, birds through their feeding strategies, are capable of regulating snail populations in their habitats. In a study by Nachuha et al. (2014) on the role of Ciconiiformes birds in the control of pests in rice plantations of Kibimba in Uganda, the low abundance of snails in the rice scheme was linked to feeding behaviour of water birds in the area. Given the abundance of birds and their position in ecosystem maintenance, particularly in wetland habitats (Anderson and Coppolino 2009), it is important to fully explore their feeding impacts on snails in tropical and sub-tropical habitats. This awareness not only broadens our knowledge of predator-prey interactions in birds but also offers the room for the possibility of using some of the predatory birds as keystone species in their ecosystems.

Furthermore, some birds, through their feeding can regulate the populations of snails including those which transmit parasites and diseases in humans. The high abundance of snails of medical and veterinary importance like *Bulinus* and *Lymnaea* observed in the present study implies potential threats to public and animal health. The snails of the genus *Bulinus* transmit various parasites such as schistosomes that cause urogenital

schistosomiasis in humans (Brown 1994, Madsen et al. 2001). On the other hand, many *Lymnaea* species are known to serve as intermediate hosts for trematodes of medical and veterinary importance including for instance *Lymnaea natalensis*, which is an intermediate host for *Fasciola gigantica*, the causative agent of fascioliasis in Africa (Ibrahim and Ahmed 2019). The present observation of the fungus *Candida parapsilosis* in particular, is of great ecological importance from the food diversity standpoint of the African Sacred Ibis. The observation also has some significance to public health as well since *C. parapsilosis* is a significant cause of sepsis and of wound and tissue infections in people with lower or compromised body immunity (Trofa et al. 2008).

Conclusion and recommendations

The present findings have confirmed previously held reports that the African Sacred Ibis include some molluscs, fungus and protozoan ciliates in their diet. There is evidence that avian feeding on various snail species can have significant impact including exerting pressure on the prey abundance and regulating snail populations in their habitats. The findings of this study provide information on the ecological role of the African Sacred Ibises in their ecosystems and also point towards their possible future use as biological control agents for snails of ecological and medical importance. As also recommended elsewhere (Wenny et al. 2011), future comprehensive studies to elucidate the feeding strategies of the birds and their impacts on the ecosystems where they live are necessary.

Acknowledgments

We are grateful to the Waterbird Society for funding the study, anonymous reviewers for critically reading the manuscript and all others for assistance during data collection and analysis.

Conflict of interest statement

There is no potential conflict of interests related to the publication of this article.

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