LOCAL MOVEMENTS, FORAGING PATTERNS, AND HEAVY METALS EXPOSURE IN CASPIAN TERNS *HYDROPROGNE CASPIA* BREEDING ON PENGUIN ISLAND, WESTERN AUSTRALIA

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SUMMARY

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Caspian Terns from a breeding colony on Penguin Island in the southern metropolitan coastal waters of Perth, Western Australian, dispersed primarily onto the nearby Peel-Harvey Estuary. The terns preyed on benthic fishes associated with marine seagrass meadows and estuarine shallows. The diet was dominated by detritivores, in particular sea mullet *Mugil cephalus* and Perth herring *Nematalosa vlaminghi*. Stable isotope values from adult feathers also indicated the importance of perennial or annual seagrass habitats for foraging. Cadmium and lead levels in feathers were low. However, most breeding adults had elevated levels of mercury in their tail feathers. Mercury concentrations were strongly correlated with δ^{15} N, indicating a link between exposure and the mineralised nitrogen associated with the relative eutrophication of the foraging habitats. Mercury concentrations were strongly correlated with selenium in the adult feathers, suggesting that selenium was being regulated in response to mercury exposure.

Key words: Caspian Tern, movements, foraging, diet, metals, mercury, selenium, monitoring

INTRODUCTION

Caspian Terns *Hydroprogne caspia* inhabit coastal and inland waters in subtropical and temperate regions around the world. These terns are piscivorous mesopredators that forage by plunge-diving into shallow water (Cramp 1985).

In the southern portion of Western Australia (WA), all Caspian Tern nesting sites are on offshore islands, and most colonies are limited to a few pairs (Burbidge *et al.* 1996), probably reflecting the low productivity of the oligotrophic coastal waters in the region (Pearce *et al.* 2000). One exception is the breeding colony in Shoalwater Bay, most recently (since around 2005) located on Penguin Island (32°18′19.99″S, 115°41′34.92″E; Fig. 1), which includes up to 60 breeding pairs nesting between late July and early November. The higher Caspian Tern density in this area probably results from the proximity of two relatively productive (Pearce *et al.* 2000) estuaries, and especially the Peel-Harvey Estuary 30 km south of Penguin Island (Fig. 1).

During much of the year, Caspian Terns are conspicuous residents of the Swan-Canning and Peel-Harvey estuaries and penetrate further upstream than the other neritic terns, sometimes moving beyond the salt wedge into brackish waters (Serventy *et al.* 1971, Johnstone & Storr 1998, Hale & Butcher 2007, J.N. Dunlop pers. obs.). The estuaries and some marine embayments in the region (particularly Cockburn Sound) are highly eutrophic as the result of nitrogen and/or phosphorus inputs from industry, urban development, and agriculture (Department of Environmental Protection 1996, Hale & Butcher 2007). Despite some major management interventions, including the engineering of a second inlet in the 1990s (the Dawesville Cut), the water quality of the Peel-Harvey Estuary remains compromised by high phosphorus inputs. As a consequence, algal blooms, high organic carbon, anoxic and reducing surface sediments, toxic monosulphidic black ooze, and fish kills are evident (Hale & Butcher 2007). These conditions are particularly apparent near the river deltas in the upper reaches of the Peel Inlet. Monitoring data on the ecological consequences of these environmental impacts in the southern metropolitan coastal waters and the Peel-Harvey Estuary (Hale 2008) are unavailable, particularly with respect to aquatic wildlife.

Monitoring predators such as Caspian Terns has the capacity to integrate the effects of a wide range of changes in coastal aquatic ecosystems. These changes include climate shifts, habitat change, altered trophic pathways and prey abundance, fishing pressures (Boyd *et al.* 2006), and contamination by pollutants (Nisbet *et al.* 2002, Ohlendorf *et al.* 1988, Burger & Gochfeld 2004). This investigation was conducted to evaluate the suitability and effectiveness of using the Caspian Tern population breeding on islands in Shoalwater Bay (Fig. 1) as ecological indicators to monitor impacts to coastal and estuarine waters south of the Perth metropolitan area.

METHODS

Caspian Tern banding

Breeding Caspian Terns were captured in walk-in nest traps and banded in the colony on Penguin Island (Fig. 1) between August and October in the years 2012, 2013, and 2016. Chicks were handcaptured at night using night-lights to prevent them from running outside the colony area. Trapping was suspended in 2014 and 2015 to avoid further disturbance as the conservation managers dealt with an invasion of black rats *Rattus rattus*. In 2012, both adults and running chicks were given individually numbered stainless steel bands supplied by the Australian Bird and Bat Banding Scheme (ABBBS). Colour-banding of adults commenced in 2013 using a three-colour schema registered with ABBBS.

Beginning in 2013, members of BirdLife WA, including the Peel and Bunbury subgroups, were invited to report (and, if possible, photograph) banded Caspian Terns. No coordinated searches were conducted. One author also intermittently searched the Canning River (a branch of the Swan-Canning Estuary) and Rottnest Island during the summer months for the Penguin Island terns.

Diet investigations

Three approaches were used to investigate the prey taken by Caspian Terns while nesting on Penguin Island: fresh regurgitations, regurgitated pellets, and records of bill-carried fishes.

Adults frequently regurgitated entire or partial prey items while being captured or processed in banding operations. These fish were invariably common species that were readily identified and recorded (by frequency of occurrence). Total lengths were measured on entire specimens or estimated from relative body proportions from partial ones.

In the 2016 season, intact regurgitated pellets were collected from the nest sites of incubating or brooding pairs in the colony. These consisted of scales, skin, bones, and otoliths. The pellet material was examined using a dissecting microscope, and the fish species present in the pellets were identified using scales (compared with a reference collection from intact specimens), fin rays, or otoliths (Furlani *et al.* 2007). No attempt was made to quantify the number of individuals in each pellet or to estimate the size of the fishes taken.

Fish carried by Caspian Terns for courtship or chick feeding were photographed in the bill using a digiscope (a digital camera mounted on a spotting scope; Larson & Craig 2006). Watches to visually identify prey species were undertaken in 2013 and 2016, and generally during mid-morning or late afternoon. Bill-loaded fish were included in the data only when the images provided a reliable "digi-voucher" to confirm the identification. Fish lengths were estimated by comparison with bill length (mean culmen length = 77.8 mm measured on banded birds using vernier calipers).

Stable isotope analysis

The carbon (δ^{13} C) and nitrogen (δ^{15} N) stable isotope ratios of feather keratin were used to infer the characteristics (e.g., producers and nitrogen sources) of the foraging habitats used by Caspian Terns, as well as their mean trophic level (for a review of this methodology see Bond & Jones 2009).

Both δ^{13} C and δ^{15} N values in consumer tissues can be used to infer the sources of carbon (energy) in food chains if the producer signatures (the isotopic baselines) are known (Hobson *et al.* 1994). The δ^{15} N value is a combined indicator of nitrate source, availability, and uptake that also undergoes stepwise increase with trophic level (Graham *et al.* 2010).

The fifth tail feathers (R5) were drawn from adult Caspian Terns during the banding operations in 2012 and 2016. This feather in

adults is replaced outside the breeding season, when the adults are undergoing their basic moult (December to February), and provided enough tissue for both stable isotope and metals analysis. In 2012, down and/or breast feathers were also collected from some Caspian Tern chicks that were 1–3 weeks old.

Before processing, the feathers were inspected, physically cleaned of any extraneous matter and, if there was any surface staining, washed in de-ionized water and air-dried. The stable isotope sample from the R5 feathers was trimmed from the outer vane close to its base. The remainder of the feather was used for heavy metals analysis (see below). Chick feathers were used in their entirety.

The analysis of Caspian Tern feathers for their δ^{13} C and δ^{15} N ratios was conducted at the Natural Isotopes Laboratory at Edith Cowan University in Western Australia in 2012 and at the Water Studies Centre Laboratory at Monash University in Victoria in 2016. The two laboratories employed the same methodology. Samples were weighed, placed in tin capsules, and combusted by an elemental analyser to N₂ and CO₂. The N₂ and CO₂ were then purified by gas chromatography, and the nitrogen and carbon elemental composition and isotope ratios were determined by continuous-flow isotope ratio mass spectrometry. International standard reference materials of known elemental composition were used for stable isotope analysis. The determination of the relative δ^{13} C and δ^{15} N ratios in the study tissues was interspaced with the reference samples for continuous calibration (Boutton 1991, Brand 1996).

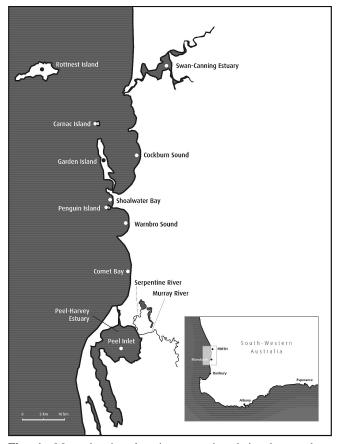


Fig. 1. Map showing locations mentioned in the southern metropolitan coastal waters of Perth, Western Australia.

Heavy metals analysis

The R5 feathers collected from adult Caspian Terns in 2012 (n = 24) and 2016 (n = 15) were analysed for concentrations (milligrams per kilogram dry weight) of the heavy metals cadmium, lead, and mercury, as well as selenium. Selenium was included because organic forms of this element can replace mercury in vertebrate animals, reducing the rate of bio-transfer or assimilation. There is evidence that the uptake of selenium is regulated in seabirds to reduce exposure to methylmercury toxicity (Cuvin-Aralar & Furness 1991).

The heavy metals analysis of feathers was conducted by the Chemistry Centre of Western Australia. The Caspian Tern feathers were analysed by mixed-acid microwave digestion, followed by inductively coupled plasma mass spectrometry (ICPMS). Results were reported as milligrams per kilogram dry weight.

RESULTS

Banding-recaptures

Between August 2012 and October 2016, there were 74 sight/ digital photograph recaptures of at least 16 adults (28.5% of the 56 banded) reported to the authors. Seventy-three (98.6%) of these were from Peel Inlet, and all but one were from the deltaic fluvial shelf (Coodanup) where the Serpentine and Murray rivers meet the estuary (Fig. 1). There was only one record from elsewhere, the upper Canning River on the Swan-Canning Estuary, north of Penguin Island (Fig. 1). All but six records were from the spring-summer period, a pattern that probably reflects the activity period of bird-watchers targeting shorebirds rather than the seasonal distribution of the terns. Adults were recorded in the Peel Inlet within a few days of banding at the breeding colony, and banded young accompanied by adults were observed to move quickly onto the Peel-Harvey Estuary after fledging. There were no records of banded Caspian Terns at islands or colonies other than Penguin Island during the observation period.

Diet investigations

Thirteen fish species were identified in the diet of the nesting Caspian Terns (Table 1). Prey sizes were 60–260 mm (Table 1). All fish taken were benthic species associated with the nearshore seagrass (*Posidonia* and *Amphibolus*) meadows, sea wrack and sandy patches, and banks in the Shoalwater Bay area (Hyndes & Lavery 2005). The most frequently recorded prey taken were sea mullet *Mugil cephalus* (49.6%) and Perth herring *Nematalosa vlaminghi* (12.6%). Both species are detritivores that ingest fine organic matter by sieving surface sediments (Waltham *et al.* 2013, Department of Water WA 2009).

Stable isotope analysis

The R5 feathers of the adult Caspian Terns had a mean δ^{13} C value of -12.15 (standard deviation [SD] 1.47), while in the runner feathers the mean δ^{13} C was -13.52 (SD 0.78). The difference was not significant, primarily because of the high variability in adult δ^{13} C values.

The R5 feathers of the adult Caspian Terns had a mean δ^{15} N value of 11.75 (SD 1.17), while in the chick feathers the mean δ^{15} N was higher, at 12.98 (SD 0.98). Again, the differences were not statistically significant because of high variability in the adults.

Common name	Scientific name	Regurgitates		Bill-loaded		Pellets
		n	Size range (mm)	n	Size range (mm) ^a	n
Perth Herring	Nematalosa vlaminghi	4	100–190	1	220	7
Ogilby's Hardyhead	Pranesus ogilbyi	1	160	6	60–120	0
Flathead	Platycephalus sp.	0		0		1
Striped Trumpeter	Pelatus sexlineatus	2	70-80	2	90-100	0
Sea Trumpeter	Pelsartia humeralis	0		2	90-140	0
Trumpeter Whiting	Sillago maculata	0		4	170-180	0
Yellow-finned Whiting	Sillago schomburgkii	5	200-205	0		0
King George Whiting	Sillaginodes punctata	5	150-220	2	180-200	0
Sand Trevally	Pseudocaranx wrighti	0		1	210	0
Roach	Gerres subfasciatus	2	140	0		0
Yellow-eyed Mullet	Aldrichetta forsteri	0		1	110	0
Sea Mullet	Mugil cephalus	17	200-260	6	180-220	24
Goby	Gobidae	0		1	90	0
Unidentified crustacean				1	100	0

TABLE 1 Fish species recorded from specimens regurgitated during handling, photographed while being bill-carried, or identified from hard parts in regurgitated pellets

^a The ranges of total lengths are reported for regurgitated and "digi-vouchered" specimens.

Heavy metals analysis

Cadmium levels in the feathers of the adult Caspian Terns were all below the detection limit of 0.001 mg/kg. Mean lead concentration was low, at 0.074 mg/kg (SD 0.13), and mean selenium concentration was 0.82 mg/kg (SD 0.24).

Mean mercury concentration was 2.27 mg/kg (SD 1.44) in feathers (Fig. 2). Neither the mercury nor selenium values were normally distributed, indicating that there may have been more than one sample population in the data.

There was a highly significant positive correlation in $\delta^{15}N$ values against mercury concentrations for the Caspian Tern R5 feathers ($R^2 = 0.51$, P < 0.005, n = 37; Fig. 3); i.e., mercury sequestration increased with increasing $\delta^{15}N$.

Mercury and selenium concentrations in the R5 Caspian Tern feathers were also strongly correlated ($R^2 = 0.78$, P < 0.005, n = 39; Fig. 4), with selenium concentrations increasing with increased mercury.

DISCUSSION

Banded adult and juvenile Caspian Terns from the colony on Penguin Island were frequently resighted on the Peel Inlet (30 km to the south), with only one record from the Swan-Canning Estuary (38 km to the north). Individual terns (and their fledglings) were observed to move onto the Peel-Harvey Estuary soon after completion of a nesting attempt. Andersen *et al.* (2007) reported mean trip distances in breeding Caspian Terns of 16–28 km, with the shortest distances during chick rearing. While the waters of the Peel-Harvey Estuary are within the extreme trip distances reported, foraging was more likely to have occurred closer to the colony (and therefore in the marine environment) when the birds were provisioning their young (Anderson *et al.* 2007). Estuarine prey would have been within range for most of the year but probably not during the chick-rearing period.

Caspian Terns foraging from Penguin Island captured an array of benthic species associated with the nearby, nearshore, perennial seagrass meadows and sand patches. However, the diet was dominated by sea mullet and Perth herring. Both of these fishes

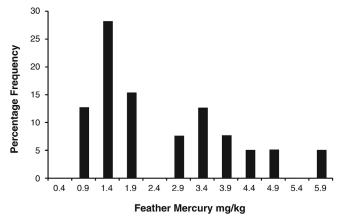


Fig. 2. Distribution of mercury values in adult Caspian Tern R5 feathers (n = 39).

are detritivores that ingest organic matter from surface sediments. The authors have no specific data on the prey taken by the Caspian Terns outside the breeding season, although these fishes are among the most abundant fishes in the local estuaries, and sea mullet is a target species for a commercial fishery operating in the Peel-Harvey system (Chubb & Potter 1984, SCS Global Services 2016). The producer-based diet of the main prey species probably explains the relatively low trophic level observed in the adult Caspian Terns while foraging on the estuary.

The δ^{13} C values in the Caspian Tern tail feathers were within the C4 (photosynthetic pathway) range for producers in the local estuaries (Brearly 2005), indicating that seagrass biomass is the dominant contributor to the food web for adult Caspian Terns, while the more depleted δ^{13} C value in the chicks probably indicates an increased contribution from algae in the marine environment around Penguin Island relative to that in the estuaries.

In the estuarine context, the seagrasses are mainly annual species such as *Halophila* and *Ruppia* (Brearly 2005). Perennial seagrasses, such as *Posidonia* and *Amphibolus*, (with a contribution from epiphytic algae), characterize the producer base in the marine environment around Penguin Island (Hyndes & Lavery 2005).

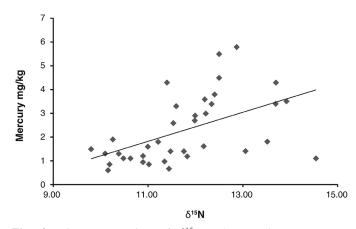


Fig. 3. Linear regression of δ^{15} N values against mercury concentrations for Caspian Tern R5 features ($R^2 = 0.51$, P < 0.005, n = 37).

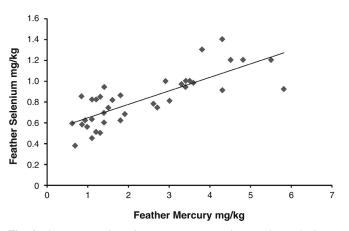


Fig. 4. Linear regression of mercury concentrations against selenium concentrations in Caspian Tern R5 feathers ($R^2 = 0.78$, P < 0.005, n = 39).

The δ^{15} N value indicates a mean trophic level for Caspian Terns that is quite low for a large piscivorous bird, reflecting the producer detritus in the diet of its main prey species. By comparison, the mean δ^{15} N values in adult Crested Tern *Sterna bergii* feathers from Penguin Island ranged from 12.97 to 14.77, while the down of Little Penguins *Eudyptula minor* in the southern metropolitan waters ranged from 13.97 to 14.96 (J.N. Dunlop, unpubl. data). A higher proportion of predatory fishes (e.g., whiting; Table 1) were likely taken from the marine environment around the island during breeding, increasing the trophic level in the chicks.

The mean feather mercury concentration in adult Caspian Terns breeding on Penguin Island was 2.27 mg/kg. Bridled Terns Onychoprion anaethetus feeding over shelf waters off the Perth metropolitan region had a mean P6 feather mercury concentration of 0.71 mg/kg (SD 0.53, n = 10; J.N. Dunlop, unpubl. data). All but two adult Caspian Terns sampled on Penguin Island had feather mercury levels above this indicator of uncontaminated conditions. Two of the sampled Caspian Terns had feather mercury levels above the threshold considered capable of producing deleterious effects (5 mg/kg, Burger & Gochfeld 2004); four individuals registered above 4 mg/kg. One bird with a feather mercury of 5.8 mg/kg was observed dying on the Peel Inlet about 3 months after sampling, with symptoms consistent with nervous disintegration caused by mercury intoxication (Burger & Gochfeld 2004). These observations indicate that the Caspian Terns foraging in the southern metropolitan coastal waters of Perth may well be subject to an elevated level of mercury exposure.

The mercury concentrations in the adult Caspian Tern feathers were not normally distributed, indicating that there may have been more than one sample population in the data. These differences in exposure patterns are probably the result of differences in foraging behaviour and locations between adult individuals. Indeed, Caspian Terns have been reported to occupy temporary foraging territories, and this might underpin individual variations in mercury contamination (McNicholl 1990).

The mercury concentrations observed were strongly correlated with the $\delta^{15}N$ values in the same feathers. This suggests that variations in the relative abundance of mineralised nitrogen in the food chain at different locations may be more important than individual differences in trophic level in determining mercury concentrations in adult Caspian Terns. The $\delta^{15}N$ values in the Caspian Tern feathers were probably indicative of the level of nitrate contamination in the foraging habitat. High mercury exposure would appear to be related to the increased eutrophication in these aquatic ecosystems.

The biotransference and amplification of mercury into aquatic food chains is facilitated by microbial conversion of elemental forms into organic forms, principally methylmercury (Costa *et al.* 2012). Almost all of the mercury in birds and bird feathers is in organo-mercury compounds (Burger & Gochfeld 2004). Microbial methylation of mercury tends to occur in high-carbon, anoxic environments in surface sediments or biofilms (Costa *et al.* 2012). Such conditions frequently develop in the upper reaches of the Peel-Harvey Estuary as a result of the de-oxygenation of the surface sediments from the decomposition of excessive algal biomass and the oxidation of monosulphidic black ooze (Hale & Butcher 2007). The detritivorous fishes consumed by the Caspian Terns are likely to ingest contaminated surface sediment and assimilate methylmercury from affected areas into their tissues. The principal source of mercury contamination in Caspian Terns in the southern metropolitan coastal waters is therefore probably the

Peel-Harvey Estuary. The variations in exposure probably relate to the time different individuals spend foraging in the estuary.

Selenium is a strong anti-oxidant and an important animal trace element. This element is relatively abundant in seawater and in marine biota but less abundant, and potentially toxic, in lowsalinity environments. Selenium can replace mercury in vertebrate animals, reducing the rate of biotransfer or assimilation of the latter. There is evidence that the uptake of selenium is regulated in seabirds to reduce exposure to methylmercury toxicity (Cuvin-Aralar & Furness 1991). This was evidently the case with the adult Caspian Terns sampled in this study, as there was a strong positive correlation between feather mercury and selenium concentrations. The high selenium availability from marine prey likely provides some protection from mercury toxicity for Caspian Terns while they are in the estuaries and may also mask some of the environmental exposure. Individual terns with a high mercury burden may be more at risk when they shift from mostly marine foraging when breeding to estuarine environments with lower availability of selenium.

CONCLUSION

Caspian Terns breeding on Penguin Island in Shoalwater Bay and foraging for much of the non-breeding period in the Peel-Harvey Estuary showed significant potential as indicators of the ecological condition of the coastal waters south of the Perth metropolitan region. Combining banding and dietary analysis with stable isotope approaches to identify foraging habitats was particularly useful in interpreting the spatial and ecological factors underpinning mercury exposure. Repeated observations could readily be incorporated into a monitoring framework, particularly for the Peel-Harvey Estuary.

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