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SEABIRDS AS POSSIBLE RESERVOIRS OF *ERYSIPELOTHRIX RHUSIOPATHIAE* ON ISLANDS USED FOR CONSERVATION TRANSLOCATIONS IN NEW ZEALAND

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ABSTRACT: *Erysipelothrix rhusiopathiae*, the causative agent of the disease erysipelas, is a gram-positive bacillus, and an opportunistic pathogen in diverse species of animals. In New Zealand, *E. rhusiopathiae* has killed endangered birds on offshore islands, including Kākāpō (*Strigops habroptilus*), Takahē (*Porphyrio hochstetteri*), and Kiwi (*Apteryx* spp.). The source of infection is uncertain, and the prevalence of *E. rhusiopathiae* among wild birds is currently unknown. During October 2018 to December 2018, we surveyed dead and live seabirds that visit two of New Zealand's offshore islands used for Kākāpō conservation with the goal of determining the prevalence of *E. rhusiopathiae*. Bone marrow from dead birds was cultivated on selective agar, and organisms cultured were identified using matrix-assisted laser desorption/ionization with time-of-flight mass spectrometry. The prevalence of *E. rhusiopathiae* was calculated in different species for each island. We surveyed live birds using PCR with *Erysipelothrix* spp.–specific primers on blood samples. The prevalence of *E. rhusiopathiae* in dead seabirds on Whenua Hou and Te Hauturu-o-Toi was 3.4% (3/86) and 11.4% (5/44), respectively. On Whenua Hou, *E. rhusiopathiae* was detected in Sooty Shearwaters (*Puffinus griseus*; 5.9%, 2/34) and Mottled Petrels (*Pterodroma inexpectata*; 2.7%, 1/36) while it was detected only in Cook's Petrels (*Pterodroma cookie*; 13.5%, 5/37) on Te Hauturu-o-Toi. Blood samples were collected from two seabird species; only one of 50 Mottled Petrels (2.0%) was positive for the presence of *Erysipelothrix* spp. Our findings confirm that burrowing seabirds are possible reservoirs of *E. rhusiopathiae* on both islands studied and may be the source of spillover to other species on the island. The differences in observed prevalence suggest the species composition of the reservoir of *E. rhusiopathiae* may vary geographically.

Key words: Codfish Island, Cook's Petrel, erysipelas, *Erysipelothrix rhusiopathiae*, Little Barrier Island, Mottled Petrel, seabirds, Sooty Shearwater.

INTRODUCTION

Erysipelas is the infectious disease caused by the bacterium *Erysipelothrix rhusiopathiae* in a wide range of mammals, birds, some reptiles, and fish (Bricker and Saif 2013; Chong et al. 2015). This gram-positive bacillus is ubiquitous in nature and considered to be an opportunistic pathogen (Wood 1992). The organism can be found in the tonsils or the intestinal tract of healthy carrier animals (Brooke and Riley 1999). It has been suggested that *E. rhusiopathiae* can multiply in alkaline soils during warm weather, and it can survive up to 35 days in soil under various conditions (Wood 1973). Transmission of *E. rhusiopathiae* is thought to be through the gastrointestinal tract after ingestion of contaminated food or water, or through breaks in the mucous membranes or skin (Bricker and Saif 2013). Asymptomatic carriage by animals, with subsequent dissemination into the environment and other animals, has been suggested as an important mechanism for the maintenance of *E. rhusiopathiae* (Brooke and Riley 1999). Although the main route of shedding has not been established, *E. rhusiopathiae* might enter the environment through infected carcasses and through saliva and droppings of live birds. *Erysipelothrix rhusiopathiae* has been isolated most frequently from the cecal tonsils, liver, large intestine, heart, and blood of carrier birds (Bricker and Saif 2013). Erysipelas in birds is characterized by either acute, fulminating infections or, more rarely, chronic infections causing infertility in male birds and reduced egg production in female birds (Bricker and Saif 2013).

In July 2004, an outbreak of erysipelas affected the success of a translocation of juvenile Kākāpō (*Strigops habroptilus*) from Whenua Hou (Codfish Island) to Chalky Island, killing 3/19 birds translocated (Gartrell et al. 2005). Translocation of juvenile birds to islands where breeding success has been lower is one of the management practices used by the Kākāpō recovery team to enhance the chances of successful reproduction in adult birds (Cresswell 1996; Clout and Merton 1998). This was the first time that an infectious disease had affected a translocation event in Kākāpō and the first time that erysipelas was reported in this critically endangered New Zealand parrot (Gartrell at el. 2005). Subsequently, sporadic deaths from erysipelas have been seen in Takahē (*Porphyrio hochstetteri*; McLelland et al. 2011), Kākāpō from two islands studied, and Kiwi (*Apteryx* spp.) from another location, indicating the persistence of this bacterium in New Zealand ecosystems.

Many species of wild birds and mammals in various geographic regions have been reported to carry *E. rhusiopathiae* and act as reservoirs in natural ecosystems (Wolcott 2007), but the prevalence in wild bird populations is unknown. Surveillance of local fauna and ectoparasites (Chirico et al. 2003; Eriksson et al. 2009) has been recommended to learn more about the epidemiology of the disease (Gartrell et al. 2005). During the erysipelas outbreak in 2004, seabirds were considered the most likely source of infection for the Kākāpō, as both the source and the destination islands have a diverse fauna of marine and terrestrial birds, and 10 of 15 samples of ulnar bone marrow from seabird carcasses recovered from Whenua Hou at the time of the Kākāpō mortalities were culture-positive for *E. rhusiopathiae* (Gartrell et al. 2005).

We aimed to survey pelagic and coastal seabirds in two offshore islands used for conservation of native New Zealand fauna: Whenua Hou and Te Hauturu-o-Toi (Little Barrier Island) to estimate the prevalence of *E. rhusiopathiae* in these species. These two islands are the home for several translocated native bird species threatened with extinction, including Kākāpō, and both are home to large populations of nesting seabirds (Rayner et al. 2007b, 2008; Scott et al. 2009; Sagar et al. 2015).

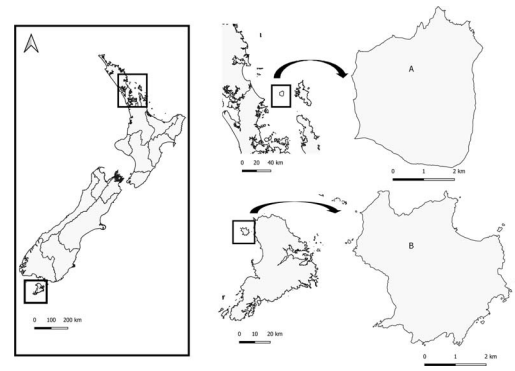

FIGURE 1. Map of New Zealand showing the locations of the two offshore islands on which seabirds were sampled in October 2018 to December 2018 to determine the prevalence of infection with *Erysipelothrix rhuseopathieae*. (A) Te Hauturu-o-Toi (Little Barrier Island). (B) Whenua Hou (Codfish Island).

MATERIALS AND METHODS

Study locations

Study samples were collected from two predator-free offshore islands (Fig. 1), Te Hauturu-o-Toi (36.1991°S, 175.0814°E) and Whenua Hou (46.7833°S, 167.6333°E). These two islands, both nature reserves, are located approximately 1,000 km apart (Taylor 2000). Samples were collected under a Massey University animal ethics permit (MUAEC protocol no. 17/51) and Department of Conservation, New Zealand (permit nos. 65400-DOA and 65402-FAU for dead and live seabird sampling) from the two nature reserves.

Te Hauturu-o-Toi (Little Barrier Island)

Te Hauturu-o-Toi is a nature reserve with an area of 28 km^2 located off the northeastern coast of New Zealand, 80 km to the north of Auckland. It is a highly eroded, extinct volcanic island, and a series of steep ridges radiating from a central range toward the coast have deeply dissected the land. An altitudinal gradient can be seen in the forest, and the forest cover in higher altitude areas is considered intact (Moorhouse and Powlesland 1991). It is a refuge for more than 350 species of plants and several species of threatened New Zealand birds. Saddleback (*Philesturnus rufusater*; Lovegrove 1996; Hoosen et al. 2003) and

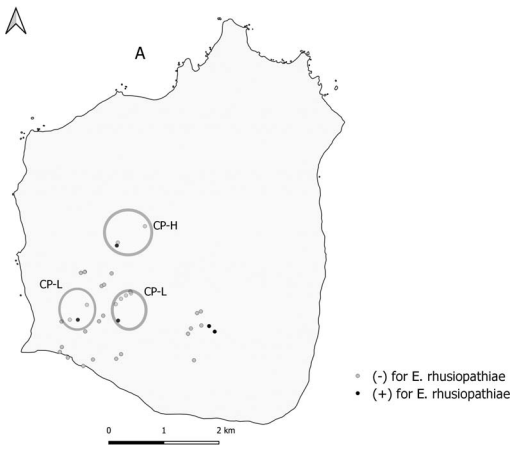

FIGURE 2. The areas of dead seabird collection on Te Hauturu-o-Toi. Dots correspond to the collection location of *Erysipelothrix rhuseopathieae*–positive (dark) and –negative (pale) samples. Circled areas indicate low altitude (CP-L) and high altitude (CP-H) breeding areas of Cook's Petrels (*Pterodroma cookie*) sampled for the study. Boundaries of the colonies are not exact.

Kākāpō (Moorhouse and Powlesland 1991) are two of the endemic bird species that have been translocated to this island.

Te Hauturu-o-Toi is the most important breeding ground for the endangered Cook's Petrel (*Pterodroma cookii*), with more than 50,000 breeding pairs in higher-altitude areas (Rayner et al. 2007b). Other burrowing seabird species found on the island include the Black Petrel (*Procellaria parkinsoni*; Rayner et al. 2007b) and the Grey Faced Petrel (*Pterodroma macroptera*; Rayner et al. 2008). A population of critically endangered New Zealand Storm Petrels (*Fregetta maoriana*) was identified breeding on the island in 2003 (Flood 2003). Additionally, Little Blue Penguin (*Eudyptula minor*) breed in the forest areas.

Whenua Hou (Codfish Island)

Whenua Hou has a land area of 14 km^2 and is situated in southern New Zealand, 3 km west from the northwest coast of Rakiura (Stewart Island). This nature reserve was identified as a potential location for native species in 1960, and measures were taken for island restoration, including a restriction on unauthorized access, in 1968. The summit of the island is close to the south coastal area and reaches a height of 250 m. A large valley in the northeast side and steep coastal cliffs on the southwest are the predominant features (Sedgeley et al. 2006; Scott et al. 2009). It is home to Southern Short-tailed Bats (*Mystacina tuberculata tuberculata*), Kākā (*Nestor meridionalis*), Fernbirds (*Bowdleria punctata*), Red-crowned Parakeet, Yellow-crowned Parakeet (*Cyanoramphus auriceps*), Pacific Black Ducks (*Anas superciliosa*), and a breeding population of Kākāpō.

It also provides a home for translocated populations of endangered Mohua (*Mohoua ochrocephala*) and Campbell Island Teal (*Anas nesiotis*). It is important as a breeding ground for several species of seabirds, including endemic species. Common Diving Petrels (*Pelecanoides urinatrix*), South Georgian Diving Petrels (*Pelecanoides georgicus*), Cook's Petrels, Mottled Petrels (*Pterodroma inexpectata*), and Sooty Shearwaters (*Puffinus griseus*) are the burrowing seabirds found on this island. Diving petrel and prions are limited to areas near the coast, whereas burrows of forest-nesting species, such as Cook's Petrel and Mottled Petrel, are distributed across large areas of slope and ridge-top habitats, mostly at higher altitudes (Rayner et al. 2008; Scott et al. 2009).

Time of sampling

We sampled dead and live seabirds on Whenua Hou and Te Hauturu-o-Toi during the Austral spring and early summer of 2018, from 15 October 2018 to 1 November 2018 and from 21 November 2018 to 6 December 2018, respectively.

Dead seabird sampling

On both islands, dead sea birds were sampled from seabird colonies and along the tracks outside the colonies. On Whenua Hou, large Mottled Petrel colonies sampled were distributed near the summit area (250 m). Cook's Petrel colonies on this island have a patchy distribution within the forest area. Sooty Shearwater colonies were found near the coastal side on the southwest and eastern sides of the island. Some of the Sooty Shearwater colonies were observed closer to the Cook's Petrel colonies. On Te Hauturu-o-Toi, dead birds were sampled from two low-altitude colonies, two high-altitude colonies, and along the tracks outside the colony areas (Figs. 2, 3). Black Petrel colonies were observed a few meters above the higher-altitude Cook's Petrel colonies.

We used opportunistic sampling because there was no prior data on this disease in particular species or spillover events. On both islands, seabirds found dead were sampled; one long bone, preferably an ulna, was placed in thick zip-lock plastic bags for transport to the laboratory. Species were identified by the color of the plumage and by using skeletal and bill morphometrics. Age or age group of the sampled dead

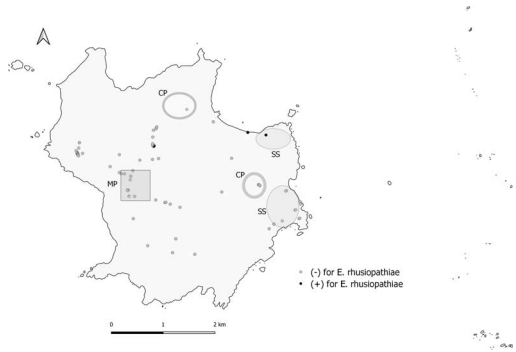

FIGURE 3. A map showing the areas of dead seabird collection on Whenua Hou with dots corresponding to the collection location of positive and negative samples. Circled and squared areas indicate breeding areas of Sooty Shearwaters (SS; *Puffinus griseus*), Mottled Petrel (MP; *Pterodroma inexpectata*), and Cook's Petrel (CP; *Pterodroma cookie*) sampled for the study. Boundaries of the colonies are not exact.

birds could not be determined in many cases, although most could be categorized as adults.

Live seabird sampling

On Whenua Hou, live-bird sampling was performed in Mottled Petrel colonies located near the summit and the lower and upper loop tracks of the island. On Te Hauturu-o-Toi, Cook's Petrel colonies of the lower-valley track (80–100 m above sea level) and higher-altitude (more than 500 m above sea level) areas in the Thumb track (Rayner et al. 2007a) were used for live bird sampling. Adult birds were caught inside their burrows and restrained by controlling their head, wings, and feet; thick gloves and towels were used for handling. Birds were checked for identification bands and 0.2–0.3 mL of blood was collected from the cutaneous ulnar vein of each bird. If the birds were not banded, they were microchipped using a Minichip (Allflex, Somerset West, South Africa). Collected samples were stored in calcium ethylenediaminetetraacetic acid anticoagulant tubes (1.3 mL SC Micro tube K3, Sarstedt Inc., Newton, North Carolina, USA) at 4 C until they were transported to the laboratory. Storage time for collected blood samples ranged from 3 to 6 days.

Isolation and identification of *E. rhusiopathiae* in bone-marrow samples

Laboratory isolation and identification of *E. rhusiopathiae* was conducted in the ^{m}EpiLab (Hopkirk Institute, Massey University, Palmerston North, New Zealand). Bone marrow from the collected bones was cultured in brain-heart infusion broth (Fort Richard Laboratories, Auckland, New Zealand) and incubated at 37 C for 48 h. The broth was subcultured on brain-heart infusion agar with kanamycin and vancomycin (Fort Richard Laboratories) and on Columbia horse-blood agar (Fort Richard Laboratories) and incubated at 37 C with 5% CO2. Cultures were checked for colonies showing morphology typical of *E. rhusiopathiae* (tiny or small, colorless or grey, circular colonies) at 24 and 48 h intervals. All cultures similar to *E. rhusiopathiae* on brain-heart infusion or blood agar were subjected to a catalase test. Because *E. rhusiopathiae* is catalase negative, all the catalase-negative isolates were sent for confirmation by matrix-assisted laser desorption/ionization with time-of-flight mass spectrometry (MALDI-TOF MS).

Matrix-assisted laser desorption/ionization with time-of-flight mass spectrometry

A protein profile of bacteria can be obtained directly from bacterial colonies using MALDI-TOF MS. This allows rapid and accurate identification of bacteria within a few hours, based on their molecular size and electrical charge (Holland et al. 1996; Bizzini et al. 2010). A bacterial colony from a pure culture was homogenized with 300 μL of sterile water and, then, mixed with 900 μL of 100% ethanol and stored at –20 C, before being further processed and identified by MALDI–TOF MS in a MALDI Biotyper 3.0 (Bruker Daltonics, Billerica, Massachusetts, USA) as described by the manufacturer (Stępień-Pyśniak et al. 2017), using the laboratories of Food Assurance at the Fonterra Research and Development Centre (Fitzherbert, Palmerston North, New Zealand). In the MALDI Biotyper system, the reliability of identification of the bacterial isolates is expressed in points: an isolate is considered identified correctly to the species level if the score (log) was between 2.0 and 3.00. A score (log) between 1.70 and 1.99 indicates identification to the genus level, whereas a score (log) <1.70 indicates that no organism identification was possible.

Extraction of DNA from blood samples and identification using PCR

We extracted DNA from the seabird blood samples using the QIAamp® DNA Blood Mini Kit (Qiagen, Valencia, California, USA), following the manufacturer's recommendations. Extracted DNA was subjected to PCR using *Erysipelothrix* genus-specific primers MO101 (5′-AGATGCCATAGAAACTGGTA-3′) and M0102 (5′-CTGTATCCGCCATAACTA-3′) as previously de-

scribed (Makino et al. 1994). The primers are based on the DNA sequence coding for 16S rRNA gene *Erysipelothrix* spp.

The PCR method described by Makino et al. (1994) was used with some modifications to the PCR reaction mixture. A Sensoquest Labcycler Thermocycler (Sensoquest, Gottingen, Germany) was used. For each sample, the PCR process used 20 μL of reaction mixture containing 4 μL HOT FIREPol® Blend Master Mix (Solis BioDyne, Tartu, Estonia), 50 pmol of oligonucleotide primers in a volume of 1 μL, 13 μL of water, and 2 μL of sample DNA. After initial heating at 94 C for 2 min, the process followed was denaturation at 94 C for 1.0 min, annealing at 54 C for 2 min, and extension at 72 C for 2 min; amplification was repeated for 30 cycles. Finally, an additional extension step was performed at 72 C for 7 min. The amplified product was electrophoresed at 110 v on 1% (w/v) agarose gels in Tris borate buffer, and bands were photographed under ultraviolet light.

Statistical analysis

Apparent prevalence estimates and the 95% confidence intervals (Wilson method) for *E. rhusiopathiae* on the two islands and in different species of seabirds were calculated in R version 3.4.1 (RStudio Team 2016). We used the package 'prevalence' version 0.4.0 (Devleesschauwer et al. 2014).

RESULTS

Dead seabird sampling

In total, 86 dead seabirds were collected on Whenua Hou over 3 wk. Most of the birds sampled were Mottled Petrels (36/86), followed by Sooty Shearwaters (34/86) and Cook's Petrels (4/86). Twelve of the 86 carcasses found were extremely decomposed and could not be identified. On Te Hauturu-o-Toi, 44 dead seabirds were collected during the study period, mainly Cook's Petrels (37/44), with single samples each of a Black Petrel, a Little Blue Penguin, and an Australasian Gannet. Four carcasses from Te Hauturu-o-Toi could not be identified.

Live bird sampling

A total of 50 blood samples were collected from Mottled Petrels on Whenua Hou. On Te Hauturu-o-Toi, 18 blood samples were collected from Cook's Petrels.

Prevalence of *E. rhusiopathiae* on Whenua Hou and Te Hauturu-o-Toi

Using MALDI-TOF MS, *E. rhusiopathiae* was identified from 3/86 dead seabirds on Whenua Hou (3.5%, 95% confidence interval [CI] 1.1–9.7) and 5/44 on Te Hauturu-o-Toi (11.4%, 95% CI 4.9–23.9). On Whenua Hou, positive samples were found in two Sooty Shearwaters (2/34) and a Mottled Petrel (1/36). On Te Hauturu-o-Toi, all the birds found to be positive for *E. rhusiopathiae* were Cook's Petrels (5/37). In total, eight seabirds belonging to three species were infected with this bacterium in both study locations (Table 1).

Of 68 blood samples, one Mottled Petrel (1/50) was positive with *Erysipelothrix spp*. specific primers in PCR (2.0%, 95% CI 0.36–10.5).

DISCUSSION

Erysipelas is a sporadic cause of death in threatened species in New Zealand. Our study confirmed that burrowing seabirds inhabiting two of the offshore islands used for the conservation of Kākāpō and other species are a possible source of *E. rhusiopathiae* within these island environments. Although the prevalence of this bacterium in dead seabirds on Te Hauturu-o-Toi (11.4%) was comparatively higher than the overall prevalence on Whenua Hou (3.5%), *E. rhusiopathiae* was isolated from two seabird species on Whenua Hou, including the numerous Sooty Shearwaters. Sooty Shearwaters are one of the most widely distributed seabirds in the world, and they breed in large dense colonies on small islands in the south Pacific and south Atlantic Oceans, mainly around New Zealand, the Falkland Islands, Tierra del Fuego, and in the Auckland Islands and Phillip Island off Norfolk Island (Sagar 2013). They are used as a food source for humans in Australia and New Zealand, with the New Zealand harvest estimated at 250,000 birds/yr; in New Zealand, Sooty Shearwaters are important sea-

TABLE 1. The number of dead seabirds of different species sampled from Whenua Hou and Te Hauturu-o-Toi during October 2018 to December 2018 and the prevalence of *Erysipelothrix rhusiopathiae* in each species with 95% confidence intervals for Cook's Petrel (*Pterodroma cookie*), Black Petrel (*Procellaria parkinsoni*), Little Blue Penguin (*Eudyptula minor*), Australasian Gannet (*Morus serrator*), Sooty Shearwater (*Puffinus griseus*), Mottled Petrel (*Pterodroma inexpectata*).

Seabird species collected from two islands	No. dead seabirds sampled	No. dead seabirds positive for *E. rhusiopathiae*	Prevalence of *E. rhusiopathiae*, %	95% Confidence intervals
Te Hauturu-o-Toi				
Cook's Petrel	37	5	13.5	5.9–27.9
Black Petrel	1	0	0	0–7.3
Little Blue Penguin	1	0	0	0–7.3
Australasian Gannet	1	0	0	0–7.3
Other/unknown species	4	0	0	0–4.0
Whenua Hou				
Cook's Petrel	4	0	0	0–4.0
Sooty Shearwater	34	2	5.9	1.6–19.0
Mottled Petrel	36	1	2.8	0.4–14.1
Other/unknown species	12	0	0	0–1.8

birds both culturally and ecologically. Considering the public health significance, the implications of our study may extend beyond our two study sites.

We also detected *E. rhusiopathiae* in Mottled Petrel and Cook's Petrel. Mottled Petrels only breed on the Snares, Whenua Hou, and Big South Cape Island, with small colonies on other islands around Stewart Island and in Fiordland in southern New Zealand (Sagar 2013). Te Hauturu-o-Toi and Whenua Hou are the only breeding grounds for Cook's Petrels (Taylor 2000). We detected *E. rhusiopathiae* in Cook's Petrels from Te Hauturu-o-Toi, but not in those from Whenua Hou. However, fewer Cook's Petrels were sampled in Whenua Hou, and more samples would be needed to study the presence of this bacterium in that island.

The species-specific prevalence was higher in Cook's Petrels (13.5%) compared with that of the Sooty Shearwater (5.9%) and Mottled Petrels (2.8%). However, Cook's Petrels represent the bulk of the dead bird sample (84.0%) from Te Hauturu-o-Toi. Given this species bias in our opportunistic sampling, and that our study was limited to only two islands, we cannot make any accurate conclusions about the wider prevalence of *E. rhusiopathiae* in seabirds or the diversity of species affected. However, our results suggest wider studies are warranted, not only to understand the role of the seabirds for island conservation management but also to understand the effect of this infection on the seabird species.

In acute and lethal infections, *E. rhusiopathiae* bacteria will be present in high numbers in bone marrow (and in blood), but the birds only survive in this state for 2–3 days (Bricker and Saif 2013). To understand the role of seabirds as reservoirs, we need to be able to identify live birds that are carrying nonlethal infections. These birds are likely to have far fewer organisms in their blood; therefore, examination of blood samples from live birds was expected to produce lower prevalence estimates than that of the bone marrow samples from dead birds and, indeed, only 1/50 Mottled Petrels was positive. To understand the role and prevalence of live birds that are carrying this infection, there is a need to validate sensitive techniques that can detect and quantify low numbers of organisms in blood. Culturing the blood before PCR may improve the sensitivity of the assay used in this study. Alternatively, serologic investigations could be used as the next step to assess

the extent of current or previous infections of *E. rhusiopathiae* in these free-living seabird populations.

The pathogen prevalence in the host species is one of the major determinants of spillover risk among species (Plowright et al. 2017). We found *E. rhusiopathiae* in burrowing seabird species in a New Zealand island ecosystem, prevalence appearing to vary among species. These birds share the same environment with endangered native birds, and there is evidence of direct contact between those species (Powlesland et al. 2006), providing possible routes for pathogen spillover. On Whenua Hou, close encounters between Cook's Petrels and Kākāpō have been recorded, with birds sharing the same nesting burrow on one occasion, whereas, in another record, a Cook's Petrel was killed by an aggressive female Kākāpō during the breeding season (Powlesland et al. 2006). Further studies are required to confirm whether seabirds act as reservoirs for *E. rhusiopathiae* and whether spillover is occurring among species and, if so, the exact mechanism of transmission.

The transmission pathways of *E. rhusiopathiae* under natural conditions are not well understood; widely accepted hypotheses include transmission through ingestion or via skin lesions or mucous membranes when bacteria are present in the environment. Our current hypothesis is that *E. rhusiopathiae* may be shed from decomposing infected seabirds' bodies, contaminating the soil, which may then be ingested by other species during preening or feeding. The organism can survive in damp soil for weeks to months (Reboli and Farrar 1989), and environmental studies of the persistence of the organism on the islands could provide evidence to support this hypothesis. Close proximity among different species in these island ecosystems may provide other opportunities for pathogen transmission between species.

We sampled dead and live seabirds during their courtship period. In practice, it is not possible to sample live birds in another season, because of prohibitions against disturbance during incubation and when birds are feeding nestlings and fledglings. Our study was timed during the birds' courtship period to avoid any significant disturbance to egg laying, incubation, or feeding of the juvenile birds. Dead bird sampling during the fledging period could possibly be affecting the number of birds sampled. However, sampling during such periods is problematic because access to Whenua Hou is only permitted when the Kākāpō recovery team is on the island.

Our finding of a potential reservoir of *E. rhusiopathiae* in these offshore islands should be considered when evaluating the need to vaccinate at-risk species (Livingston et al. 2013), such as Kākāpō and Takahē. This is particularly important for translocations of young birds, which are likely to be more susceptible (Benskin et al. 2009).

The prevalence of *E. rhusiopathiae* in wild populations of nesting seabirds in New Zealand has not, to our knowledge, been assessed previously. Previously, *E. rhusiopathiae* was found as a cause of extensive chick mortality in Indian Yellow Nosed Albatross (*Thalassarche carteri*) on Amsterdam Island in the southern Indian Ocean (Weimerskirch 2004), resulting in low breeding success and population decline (Jaeger et al. 2018). In addition, *E. rhusiopathiae* has been detected through PCR in oropharyngeal swabs (1/21) of Amsterdam Albatross (*Diomedea amsterdamensis*) chicks and adult Sooty Albatross (1/30) (*Phoebetria fusca*) and in cloacal swabs (3/30) of adult Northern Rockhopper Penguins (*Eudyptes moseleyi*) on Amsterdam Island (Jaeger et al. 2018).

Our study describes the presence of *E. rhusiopathiae* in dead Cook's Petrels from Te Hauturu-o-Toi, an important place for the conservation of threatened New Zealand birds. The Jaeger (2018) finding that erysipelas was a cause of mortality, low breeding success, and possible population decline in the Indian Yellow Nosed Albatross (*Thalassarche carteri*) may indicate the erysipelas is having a wider effect on seabird population dynamics than previously recognized. At present, the occurrence *E. rhusiopathiae* in seabirds in these island ecosystems is not monitored, and the effect of this bacterium on

the seabird population health is not known. Our results suggest that the effects of this bacterium on seabird breeding and survival are worthy of further study, particularly for threatened seabird species. There may be multiplication and increased transmission of the pathogen within seabirds during their breeding season because many gather in large colonies for several months. Longitudinal studies of the seabird populations are difficult because of their pelagic movements; however, a focused study during the breeding season could elucidate the dynamics of *E. rhusiopathiae* infection in the seabirds at that time.

Investigation of the genetic structure of *E. rhusiopathiae* on the two islands studied and in different seabird species could improve our understanding of the possible sources or origin of this pathogen in the island ecosystem, whether one or several introduction events has occurred, and might identify the epidemiologic networks of the bacteria. Such information is important for the assessment of transmission risk between local and introduced translocated populations, or between marine and terrestrial birds. Our results confirm that burrowing seabirds are possible reservoirs of *E. rhusiopathiae* on both islands studied and may be the source of spillover to other translocated species on the islands. This should be considered in the conservation management practice of translocation of endangered New Zealand birds to these islands and should inform vaccination and health-screening strategies.

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