The effect of conservation management on Little Blue Penguins (*Eudyptula minor*) on North Island, New Zealand

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TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

ABSTRACT

This study aimed to fill a gap in research, particularly on the size of breeding populations of Little Blue Penguins in the North Auckland area, while also providing recommendations for the conservation management of Little Blue Penguins. Three sites North of Auckland were used for the majority of this study; Tawharanui North, Tawharanui South and Goat Island

The abundance and nest density of Little Blue Penguins was estimated for the three sites north of Auckland. All three sites had a very low nest density: 0.0096 nests/100m^2 at Goat Island, 0.0064 nests/100m^2 at Tawharanui North and 0.0048 nests/100m^2 at Tawharanui South. Abundance showed a similar trend, with Goat Island having the highest average number of birds coming ashore over the year (2.25 birds per night), followed by Tawharanui North (1.81 birds per night) and finally Tawharanui South (1.19 birds per night).

Habitat surveys were conducted at the three sites and differences in habitat structure were found between all three. Tawharanui North was predominantly sand and flax and Tawharanui South was predominantly pebbles and scrub, while Goat Island displayed mostly sand with no vegetation. The results suggested that grass and boulders make up the Little Blue Penguins' preferred nesting habitat and it was predicted that sites containing more of this habitat type would have higher nest densities and abundance, although small sample sizes prevented this from being validated statistically.

The nest success was calculated, again for the three sites, using the Mayfield method. The key results showed that the incubation period was associated with the lowest success rate of any of the nesting stages observed. Success rates for the incubation stage at the three sites fell

between 0.11 and 0.66, success rates for the guard stage fell between 0.74 and 1.00, and success rates for the post-guard stage fell between 0.62 and 1.00. Findings also revealed that nest success was greater at the two Tawharanui sites (where similar rates of nest success were observed) than at the Goat Island site

The number of carcasses recovered was recorded as follows: Goat Island, 0.67 birds/km; Tawharanui North, 0.63 birds/km; and Tawharanui South, 0.43 birds/km. There was no statistically significant difference between the rates of carcass recovery and the study site, nor was there a statistically significant difference between the rates of carcass recovery and the season. The rates of carcass recovery were also determined to be lower than historically found for these sites.

As an aside to the rest of this study the acute stress response of Little Blue Penguins to pit tag and metal flipper band application was assessed through measuring corticosterone levels in the blood of the birds. The results showed that implantation of a pit tag produced a significantly greater acute stress response than banding with the traditional metal flipper bands.

This study makes recommendations regarding the study and conservation of Little Blue Penguins. These recommendations are found throughout this thesis; however, key recommendations are repeated here for convenience.

- Re-assess density and abundance measures for the Tawharanui and Goat Island sites in 5 to 10 years time

- Research what makes a particular habitat type more preferable than another for Little Blue Penguins
- Place nesting boxes around the Tawharanui coast to increase suitable nesting habitat
- Carry out ongoing carcass recovery observations and necropsies on penguin populations in Auckland to better understand the causes of mortality
- Engage in further study regarding both the acute and chronic stress levels induced by different identification techniques
- Engage in further study into possible alternatives to both pit tags and metal bands for penguin identification
- Protect Little Blue Penguins from predators on the mainland where possible

CHAPTER 1 - INTRODUCTION

1 General Overview

Conservation Biology is a relatively new science and has been a recognised discipline only since the mid 1980s (Soule, 1985). It is a multidisciplinary approach that draws on many facets of science, with a goal of providing principles and tools for preserving biological diversity (Soule). Conservation biology is also described as a "crisis discipline", suggesting that within the realm of the study of biology, it is often necessary to act before all knowledge is realised (Soule). In the relatively short history of the study and practice of conservation, the world has seen many success stories come out of conservation biology, and New Zealand is no exception. Tiritiri Matangi (a small island in Auckland Hauraki Gulf) is now a world renowned, open, offshore island sanctuary with many successful translocations; Tawharanui (a Peninsular to the North of Auckland) has become New Zealand's first mainland island sanctuary to integrate conservation, public recreation and farming; and Karori Wildlife sanctuary has succeeded in becoming an open mainland island sanctuary in the heart of Wellington.

Offshore island sanctuaries have been, and still are, a valuable tool in the conservation of much of New Zealand's biodiversity. Some species owe their current survival to translocations to such islands (for example, the Kakapo (*Strigops habroptila*)) (Powlesland, Merton, & Cockrem, 2005). If a fully representative collection of New Zealand's biodiversity is to be protected, the successes of these offshore island sanctuaries needs to be replicated on the mainland (Saunders & Norton, 2001). The recent use of mainland islands is allowing this objective to come within our reach. Saunders and Norton suggest that mainland island sanctuaries have, so far, succeeded in slowing the decline of monitored species, therefore revitalising the ecological process.

2 Rationale and Significance of Study

There have been many studies on the fundamental aspects of Little Blue Penguin biology and behaviour. However, the majority of these studies have been conducted in the South Island of New Zealand or in southern Australia and little is known about northern New Zealand populations. According to Taylor (2000), the size of all northern Blue Penguin populations is unknown and accurate estimates are needed from as many breeding localities as possible.

Predator control and nest boxes have been used in some habitat restoration efforts (Jessop & Dann, 2001) and some studies have looked at how Little Blue Penguins respond to translocation (Hull et al., 1997). However, very little research has examined how Little Blue Penguin populations respond to different levels of conservation management and predator control, or investigate the benefit these actions have on penguin populations (Williamson & Wilson, 2001), especially on the northern Little Blue Penguin populations. Powlesland (1984) commented that it would be interesting to compare the breeding success of a predator-free population with that of a nearby population to which predators have access. This study aimed to fill this gap in research and provide recommendations for the conservation management of Little Blue Penguins. The Department of Conservation (DOC) and the International Union for the Conservation of Nature (IUCN) have ranked the Little Blue Penguin as “Threatened – Gradual Decline”. The subspecies *Eudyptula minor albosignatais* is listed as endangered (Hitchmough, 2002).

Comparisons of abundance, nest density, habitat and breeding success were made between sites under different levels of terrestrial conservation management. In addition, comparisons were made between an open mainland island sanctuary and a non-protected regional park. Mortality was also investigated to assess threats faced by the penguins and discover whether these differed between sites.

It is important to acknowledge that there have been many studies into Little Blue Penguin abundance, breeding success, habitat, and mortality which have set the platform for this study, including Chiaradia and Kerry (1999), Dann (1994), Fraser and Lalas (2004), Harrigan (1992), Kinsky (1960), Richdale (1940), and Weavers (1992). Although much of the current study is not entirely new, it does fill a gap in the literature on the northern Little Blue Penguin and, in particular, populations around the Auckland area. The study also takes a fresh approach, and considers populations under different conservation management.

Conservation management and research often require manipulation of animals, such as capturing, marking, taking measurements and relocating. Having a better understanding of the degree of stress that these manipulations inflict on target study species would help improve research and management methodology. Any animal- marking technique has costs, whether they be financial or ethical (Peterson et al., 2005).

There are currently six different categories of penguin marking techniques that are used across different species. Peterson et al. (2005) identifies these as:

- Remote tracking
- Flipper bands
- Implanted transponders
- Dyes

- Foot webbing tags
- Photo identification

The final part of this study investigated and compared the acute stress associated with banding and applying micro-chips (pit tags) in an attempt to ascertain which is perceived to be more stressful by the bird. Long term chronic stress and damage in Little Blue Penguins as a result of banding or micro-chipping has been documented in some studies (Clarke & Kerry, 1998; Stonehouse, 1999). However, to date there has been no study comparing the differences in acute stress caused by the use of these two different methods in the field.

2.1 Benefits of the Study

This study offers many practical benefits, a few of which are outlined below.

- Abundance estimates from each of the study sites help address Taylor's recommendation of assessing the abundance of the northern Little Blue Penguin populations (Taylor, 2000), (see Chapter 2).
- By drawing comparisons between populations of Little Blue Penguins under different levels of terrestrial protection, this study gives insight into the benefits of predator control and conservation management and how these factors impact breeding success and population. This allows for recommendations to be made about appropriate conservation management (see Chapters 2 and 4).
- This study provides baseline information for the Little Blue Penguin populations located within the mainland island at Tawharanui, allowing for future studies into how the Little Blue Penguin population proceeds with the predator-exclusion fence and ongoing

management regimes. This information is also useful for making future research and management decisions (see Chapters 2 and 4). Field work for this study commenced shortly after the predator exclusion fence was erected.

- Study into Little Blue Penguin habitat provides information to allow recommendations on future sites suitable for protection and release of Little Blue Penguins (see Chapter 3).
- Large population die-offs are common for Little Blue Penguins; therefore, data collected from this study is useful for understanding such mortality events (see Chapter 5).
- The investigation into the acute stress of banding and micro-chipping birds for identification will add fresh material to the literature on the advantages and disadvantages of various identification techniques. Ultimately this will benefit the Little Blue Penguin by allowing researchers to make more informed decisions about the ethical treatment of the species (see Chapter 6).

3 Study species

3.1 General

Eudyptula minor, also known as the Little Penguin, Blue Penguin, Little Blue Penguin, Fairy Penguin and Korora, is the world's smallest penguin species. It is native to New Zealand and southern parts of Australia, and can be found on the shorelines and offshore islands of New Zealand, including the Chatham Islands; southern parts of Australia; and Tasmania and its associated offshore islands. A healthy adult Little Blue Penguin usually weighs approximately 1100g and stands approximately 40cm tall (Barrie & Robertson, 2000).

Sexing has been distinguished through bill size (Kinsky, 1960). However, this can be difficult, as the level of dimorphism is only small, with the male being only slightly larger and having a deeper bill. Fledglings are similar to adults with the exception of a shorter bill. Their dorsal feathers are also slightly lighter with a more blue appearance that fades with wear (Marchant & Higgins, 1990). Much of the knowledge of the general biology and behaviour of the Little Blue Penguin can be credited to the early work of biologists such as Kinsky, and Richdale (1940).

3.2 Breeding

Nests are usually made up of a natural cavity filled with a collection of nesting materials including dried grass, seaweed and small sticks (Kinsky, 1960). Little Blue Penguins usually breed on offshore islands and along parts of the mainland that are inaccessible to predators (although this is less common). In the Otago region, Little Blue Penguins no longer breed at seven previously occupied mainland breeding sites (Dann, 1994). The male often courts the female through a display of vocal and visual signals. Once mating has occurred, a long-term monogamous bond is formed between the pair which can last for life (Marchant & Higgins, 1990).

Laying generally takes place between July and December (Barrie & Robertson, 2000), but this does differ with locality (Davis & Renner, 2003). Factors such as weather patterns, temperature, food abundance and body condition can all have an effect on the timing, length and success of breeding (Perriman, Houston, Steen, & Johannesen, 2000; Robinson, Chiaradia, & Hindell, 2005).

Little Blue Penguins first breed at two to three years of age. One or two eggs are laid three to five days apart. The chicks hatch approximately 36 days later and are brooded for the first ten days, then guarded continuously for one to two weeks (guard stage), followed by a period of nocturnal guarding only (post-guard stage) (Barrie & Robertson, 2000).

The chicks fledge at approximately 54 days old. It is common for only one chick from each nest to survive to fledging, as one chick of laying clutch of two may receive four to eight times the quantity of food that was given to the other (Muller-Schwarze, 1984). Usually two clutches are laid in each breeding season, with the second functioning as either a replacement clutch or a double brood (Davis & Renner, 2003). Dann, Collins, and Cullen (2001) have shown that only 18 – 20% of Little Blue Penguin fledglings contribute to the next generation and suggest that survival is lowest soon after fledging, with the mass at fledging and the time of fledgling influencing survival probability.

3.3 Behaviour

Little Blue Penguins forage during the day and take refuge in nest or roost sites during the night. They usually forage within 15km of their burrow, but have been known to travel much larger distances (Weavers, 1992). Foraging trips generally last one to two days during the breeding season (Weavers), but can be much longer at other times of the year, especially post-moult. It is common for Little Blue Penguins to go without foraging during a period of bad weather, during which they take shelter on land (Reilly, 1983).

The diet of the Little Blue Penguin differs with geographical location; however, it usually constitutes small fish, squid, and, less commonly, crustaceans (Davis & Renner, 2003). Seasonal and yearly variation in food availability may account for the relatively low metabolic rate of Little Blue Penguins (Marchant & Higgins, 1990). The Little Blue Penguin can be seen as an opportunist of small, near-shore pelagic and schooling fish, targeting those species that are abundant for the area, season and year.

Nests are often found close to the shoreline above the high tide mark; however, anecdotal evidence suggests that Little Blue Penguins can nest up to 1.5 km inland and 550m above sea level (T. Lovegrove, personal communication, October 7, 2005). In Otago and southern parts of Australia, they live in colonies and come ashore at night in large numbers. The northern New Zealand populations appear to nest more sparsely, with nests separated by hundreds of meters. The birds are known to make audible calls while coming ashore, although this is more apparent in heavily populated colonies. Both the male and the female share in the incubation and guarding of chicks, with the frequency of change-over becoming greater as incubation approaches (Chiaradia & Kerry, 1999).

Moulting usually takes place between December and March and lasts approximately 15 days. During this time, the bird is confined to land and unable to forage; therefore, it must store enough energy reserves leading up to the moult to make sure it endures the moult with adequate energy to forage effectively upon completion. Little Blue Penguins can be found starving and underweight following the moult (S. Durrant, personal communication, July 26, 2006). Richdale (1940) proposed that eggs may be readily deserted but chicks will not be abandoned. However, observations during this Masters study have suggested that the early onset of moult can cause the abandonment of chicks.

3.4 Taxonomy

Much debate has been made over the taxonomy of Little Blue Penguins, most of which has occurred on the sub-species level, although there are still differing views on the classification at the species level. Up to six sub-species have been described by Kinsky and Falla (1976):

- *Albosignata* – The white-flippered penguin found in Banks peninsula
- Novaehollandiea - Australia
- *Minor* – Southern and Western South Island
- *Chathamensis* – Chatham Islands
- Variabilis – Southern North Island
- *Iredalei* – Northern North Island

These sub-species are defined by back colour, flipper pattern and size measurements (Barrie & Robertson, 2000). The most noticeable distinction can be seen between the white-flippered penguin and the other sub-species. Genetic evidence for this distinction of six sub-species is limited, and even though the six sub-species classification has been deleted, it still remains a common classification in the current literature. Some authors suggest there are only two sub-species. Banks, Mitchell, Waas, and Paterson (2001) found evidence for two distinct clades, one including the subspecies *albosignata* and *novaehollandiae*; and a second including the other four sub-species mentioned above.

Some authors consider the genus *Eudyptula minor* a monotypic genus with a single species, *Eudyptula minor* (Davis & Renner, 2003), while other authors make a species distinction between *Eudyptula albosignata* (the white-flippered penguin) and *Eudyptula minor* (all other

Little Blue Penguins) (Muller-Schwarze, 1984). Still other authors do not make any distinction based on molecular analysis (Jacob & Hoerschelmann, 1981). The remainder of this thesis will consider the northern Little Blue Penguin to a species level of *Eudyptula minor*.

3.5 Threats to Little Blue Penguin populations

Like many other flightless birds, Little Blue Penguins are very susceptible to predation by introduced mammals such as mustelids and dogs (Harrigan, 1992; Perriman & Steen, 2000). It is therefore intuitive to conclude that Little Penguins will obtain some form of benefit from predator control. Interestingly, there has been no published evidence to suggest that rats predate adult or juvenile Little Blue Penguins or even lower hatching success (D. Houston, personal communication, August 4, 2006). Most larger colonies are now found only in areas that are not accessible by predators or in areas with some form of predator management (Dann, 1994). While it remains important to manage introduced predators on the mainland, additional efforts must focus on terrestrial conservation management and the effects of tourism, coastal sub-division, climate change, disease, fisheries' by-catch and food availability (D. Houston, personal communication).

Furness (1978) suggests that, due to the limited foraging ranges of Little Blue Penguins, they are more susceptible to suffering consequences from food shortages. Thus, it is likely that Little Blue Penguins will benefit from nesting near a reliable food source. Some authors suggest that food shortages have led to large die-offs (Dann, Cullen, Thoday, & Jessop, 1992; Dann, Norman, Cullen, Neira, & Chiaradia, 2000; Mickelson, Dann, & Cullen, 1992). However, Norman, Cullen, and Dann (1992) suggest that the reasons for such large mortality events may vary between adults and juveniles, and may even be related to high levels of parasitism.

4 Study sites

In the current study, comparisons of the abundance, nest density and habitat of Little Blue Penguin populations were made between three main sites: Cape Rodney-Okakari Point Marine Reserve and two sites at Tawharanui Regional Park - Mainland Island. The Cape Rodney-Okakari Point Marine Reserve site spans the coastline from the public access point to the south for a distance of 2.5km. For the remainder of the document, this site will be referred to as Goat Island. Tawharanui Regional Park - Mainland Island consists of two sub-sites. The first sub-site is on the northern side of the peninsula from Anchor Bay for 2.5km to the east. The second sub-site is on the southern side of the peninsula and ranges from Jones Bay for 2.5km to the east. For the remainder of the document, these two sites will be referred to as Tawharanui North and Tawharanui South, respectively. As can be seen above, each of the three sites covers a total coastline distance of 2.5km.

A fourth and separate study site was used in the acute stress hormone component of this research (Chapter 6). A separate site was chosen once it became clear after initiating the research that the Tawharanui and Goat Island sites did not have population densities high enough to make stress analysis feasible. Thus, Tiritiri Matangi Island was chosen for the stress analysis research component of the study (Chapter 6).

4.1 Goat Island

Rodney-Okakari Point Marine Reserve is also known as Goat Island Marine Reserve and Leigh Marine Reserve, and is located approximately 100km north of Auckland’s central business district (CBD).

The mainland shore was originally coastal farmland; however, since the 1980s it has gradually been divided up into small titles to meet the demand for private property ownership in this desirable area. The island itself has a land area of 9.3ha and in the past has been stripped of native vegetation and grazed. The dominant rock types in the area are both greywacke and Waitemata conglomerates. The crown acquired the island in 1971 and now the island and the associated recreational reserve are administered by the Department of Conservation (DOC). Goat Island was New Zealand’s first marine reserve and was gazetted in 1975 (Anthoni, 1997).

The marine reserve itself covers 5km of coastline and an area of approximately 500ha. This area appears to include many diverse marine habitats such as deep reefs and sand-flats. Anecdotal evidence suggests that during the 1950s, the area was heavily over-fished, resulting in the loss of much of the marine life that populated the area prior to that time (Anthoni, 1997).

4.2 Tawharanui Regional Park – Mainland Island

Tawharanui Regional Park – Mainland Island is also known as Tawharanui Open Sanctuary, and is located 90km (driving distance) from Auckland’s CBD.

After 1870 the peninsula was developed as a farm, Kauri trees were milled and shingle was extracted. The Auckland Regional Council (ARC) purchased the land in 1973 and the resultant 588ha park has become New Zealand’s first integrated mainland island sanctuary where conservation, farming and recreation co-exist in the same space. A new predator exclusion fence was completed in August 2004 and an extensive pest eradication program was then launched, although mice and rabbits still persist in fairly large numbers (Auckland Regional Council 2006). Significant restoration and planting has been done and some translocations have already been made, with more planned for the future. Some species, such as Bellbirds (*Anthornis melanura*), have already returned to the park of their own accord. On the northern shores of the park is a marine reserve which was gazetted in 1981 and covers approximately 350ha. The southern shores of the park are not under marine reserve protection and fishing is permitted (Auckland Regional Council 2006).

Tawharanui is administered by the ARC, with a lot of support from a volunteer program organized by a non-profit organisation called The Tawharanui Open Sanctuary Society Incorporated (TOSSI) (Auckland Regional Council 2006).

4.3 Tiritiri Matangi Island

Tiritiri Matangi Island is an island sanctuary within Auckland's Hauraki Gulf and is located approximately 4km off the end of the Whangaparaoa Peninsula and 30km north-east of Auckland's CBD. The island has a land area of approximately 220ha and was farmed continuously from the mid 1850s through until the 1970s.

Tiritiri Matangi was gazetted as an open scientific reserve in 1980 and the removal of farm animals and extensive planting between 1984 and 1994 saw the native vegetation cover increase from just 6% to 60%. The predator control program has now eradicated all introduced predators from the island, and surprisingly, no evidence of mice, European rats or possums has been found on the island since (Armstrong, 1999).

Tiritiri Matangi is one of only two open scientific reserves in New Zealand accessible to the public. Tiritiri Matangi is now home to a large number of native species, including Takahe (*Porphyrio mantelli*), North Island Robin (*Petroica australis longipes*), Little Spotted Kiwi (*Apteryx owenii*) and Tuatara (*Sphenodon punctatus*). Some of the species translocated to the island were not found there historically. The restoration of the island is considered by some as one of the most successful conservation stories in the world to date. Tiritiri Matangi is part of the DOC estate; however, a volunteer organisation called Supporters of Tiritiri Matangi participate to a great extent in actions and decisions concerning the island (Armstrong, 1999).

5 Scope of Study

The structure of this thesis takes on the form of a series of short papers, each covering a concise topic in its own separate chapter. Each of the chapters contains their own abstract and the hypotheses for each section are given within the appropriate chapters.

Chapter Two investigates abundance and nest density across three North Auckland populations of Little Blue Penguins, and Chapter Three looks at the differences and similarities in habitat type for the same three sites in an attempt to explain any variation in abundance and nest density. Chapter Four investigates the nest success across the three North Auckland sites, while Chapter Five discusses the issues impacting mortality in Little Blue Penguin populations and reports on some of the necropsy results performed during this study. Finally, Chapter Six looks at the issues surrounding current methods of identifying Little Blue Penguins and investigates levels of acute stress associated with two of these methods.

Little Blue Penguins nest on land, yet spend much of their time foraging at sea. Therefore, a land-based study such as this may miss some of the information that can help explain the bigger picture. For example, there may also be factors at sea such as food availability affecting the number of nests present at a site. Therefore, the conclusions and relationships outlined in this study may be only a small part of the bigger picture of Little Blue Penguin ecology at the sites studied.

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CHAPTER 2 - COMPARISON OF LITTLE BLUE PENGUIN (*EUDYPTULA MINOR*) ABUNDANCE & NEST DENSITY AT THREE NORTHERN AUCKLAND SITES

1 Abstract

The abundance and nest density of Little Blue Penguins (*Eudyptula minor*) was estimated for three sites north of Auckland: Tawharanui North, Tawharanui South and Goat Island. Little Blue Penguins are known to inhabit these three sites. Abundance was calculated through a series of transect counts. The transect counts were conducted at night, each season, over the course of a year. The nest density was calculated as the number of active nests per kilometer of coast. All three sites had a very low nest density: 0.0096 nests/100m^2 at Goat Island, 0.0064 nests/100m^2 at Tawharanui North and 0.0048 nests/100m^2 at Tawharanui South. Abundance showed a similar trend, with Goat Island having the highest average number of birds coming ashore over the year (2.25 birds per night), followed by Tawharanui North (1.81 birds per night) and finally Tawharanui South (1.19 birds per night). This result is in stark contrast to the average number of birds coming ashore over the last 15 years at the Oamaru Little Blue Penguin Colony (51.5 birds per night). There was a statistically significant difference between abundance and season; however, no significant difference between abundance and site. More information is needed to assess the reasons why abundance and nest densities are extremely low at all the sites investigated.

2 Introduction

The number of individuals in a population (abundance) is one of the most important aspects in population ecology and is an important parameter for assessing the viability of a population. Many previous studies of Little Blue Penguins have included estimates of abundance and nest density (Challies & Burleigh, 2004; Dann, 1994a; Winter, 2000) . A common method of recording a relative measure of abundance over seasons or months is to count the number of birds arriving on shore each night; this method has been used by the team at the Oamaru Blue Penguin breeding colony (D. Houston, personal communication, July 14, 2006). Nest density can be calculated as the number of nests located per area (Winter). Using line transects to calculate the nest density of ground nesting birds is also common (Bibby, Burgess, Hill, & Mustoe, 2000; Harris & Bode, 1981).

According to Taylor (2000), the size of all northern Little Blue Penguin populations is unknown and accurate estimates are needed from as many breeding localities as possible to understand the status of this species in New Zealand. The current study contributes to this goal by providing abundance and nest density estimates of Little Blue Penguins at three new sites in the northern Auckland region. The three sites used and surveyed in this study were: Goat Island (marine reserve, no terrestrial predator control); Tawharanui North (marine reserve with terrestrial predator control); and Tawharanui South (with terrestrial predator control). These three sites are described in detail in the introduction chapter (Chapter 1).

This study aimed to test the following hypotheses:

1. Tawharanui coasts (Northern and Southern) will have a greater abundance and nest density of Little Blue Penguins as compared to Goat Island.

This hypothesis was based on the fact that predators have access to Goat Island while predator access is heavily restricted at Tawharanui. Dann (1994a) found that density was higher in areas where predator access was reduced or nonexistent. It is, however, unlikely that predator access is the only contributing factor to parameters such as abundance and nest density, especially for marine species such as Little Blue Penguins.

2. Both Tawharanui sites would have fairly equal abundance and nest density.

Although it is a marine reserve off the southern coast, the distance between the northern and southern coasts at Tawharanui is very small. Due to this fact, the movement of individuals between the two coasts is possible.

3. The summer season would see the greatest number of birds coming ashore while the winter season would prove to have the lowest numbers of birds coming ashore.

This hypothesis was made based on previous studies on the yearly life cycles and behavioural biology of Little Blue Penguins (Kinsky, 1960; Richdale, 1940), as well as long term data previously obtained from the Oamaru Little Blue Penguin colony

3 Methods

Although it is not feasible to census a population over a large area and therefore the actual numbers of birds may never be known, a relative measure of abundance or density allows for comparisons between sites. However, the same measurement methodology must be used for each site. The following methods were chosen from many other possibilities for calculating abundance and density, because they appear to be the most commonly used in current and previous literature on Little Blue Penguins. This allows comparisons to be made more readily between this thesis and other studies.

3.1 Data collection

The collection of abundance and nest density data involved two parts. Firstly, nest density was calculated as the number of nests located during the 2006/2007 breeding season for each 2.5km study area. Care was taken to ensure equal time and effort was spent at each site on each of the weekly to fortnightly field trips in an attempt to reduce bias in locating nests. Suitable nesting habitat such as boulders, crevasses and vegetation were thoroughly searched along the coast line of the study sites and an area of 50m inland was also searched (sometimes further if a track or guano trail lead one to believe there could be a nest further inland). Little Blue Penguins have been known to commonly nest at much greater distances from the shore (personal observation; T. Lovegrove, personal communication, November 22, 2005). However, due to time constraints, any further increase to the search area would have been impractical. The method of nest density data collection described above does, however, allow for a relative comparison to be made between the study sites. Previous studies have used similar methods for calculating the numbers and distribution of Little Blue Penguin nests. Winter (2000) used a systematic and detailed search of 40 meters from the shoreline, mainly locating burrows from worn paths and guano stains.

Data collection for measuring abundance consisted of walking ‘night transects’. Such night transects included walking the 2.5km of shore line with spotlights and recording the number of birds seen coming ashore. Measures of effort were taken for each transect; both the number of people present and the number of hours spent searching were noted. Transects were run four times for each season (from March 2006 till February 2007) at each of the three study sites, yielding a total of 48 transect runs. Transects for the three study sites were conducted over three consecutive nights, with one night spent at each study site; this was done to reduce temporal variation and bias between samples.

3.2 Data analysis

Manipulation and graphical analysis of the data was conducted using Microsoft Excel 2000, while statistical analysis was done using Minitab 13. A Chi-Square goodness of fit test was performed on the three nest density values. This test determines whether a statistical model fits the data set by analyzing the difference between your observed values and their expected values in the model. The test showed that the three values are not equal X^2(df=2) = 0.86 $P>0.05$. An Anderson-darling test for normality was performed on the abundance data set. This was found not to be normally distributed ($P<0.005$). The data was transformed by a common factor of +1 as it contained zero values and then further log transformed. Again this transformed data was found not to be normally distributed ($P<0.005$). A Kruskall-Wallis test was then used as a nonparametric alternative to a one-way ANOVA.

4 Results

4.1 Nest Density

The density of nests per kilometer of shoreline searched was low at all three study sites. Goat Island had the highest density at 4.8 nests per kilometer (n=12), followed by the northern coast of Tawharanui with 3.2 nests per kilometer (n=8). Finally, the southern coast of Tawharanui had the lowest nest density at 2.4 nests per kilometer (n=6tal). These differences were shown to significantly differ from the expected equal values ($X^2(df=2) = 0.86$, $P>0.05$). These densities were much lower than some of the other populations around NZ, even the Auckland population at Tiritiri Matangi. It is also interesting to note that no nests were located within the first 500m from the point of access at any of the study sites. All points of access were public access points.

Although the results of this study for nest density are reported in nests per kilometer of survey, they can be converted into nests per square 100m for comparison with other studies. To calculate the area, the length of the sites (2.5km) was multiplied by the average depth surveyed (50m). This conversion is as follows: Goat Island, 0.0096 nests/100m^2; northern coast of Tawharanui, 0.0064 nests/100m^2; and southern coast of Tawharanui, 0.0048 nests/100m^2.

4.2 Abundance

Again, the number of birds coming ashore at night during each season was very low (our measure of abundance). The mean number of birds sighted on each transect for any season never exceeded four birds a night. The average number of birds coming ashore for the entire year was 2.25 birds per night for Goat Island, followed by 1.81 birds per night for Tawharanui North and finally 1.19 birds per night for Tawharanui South.

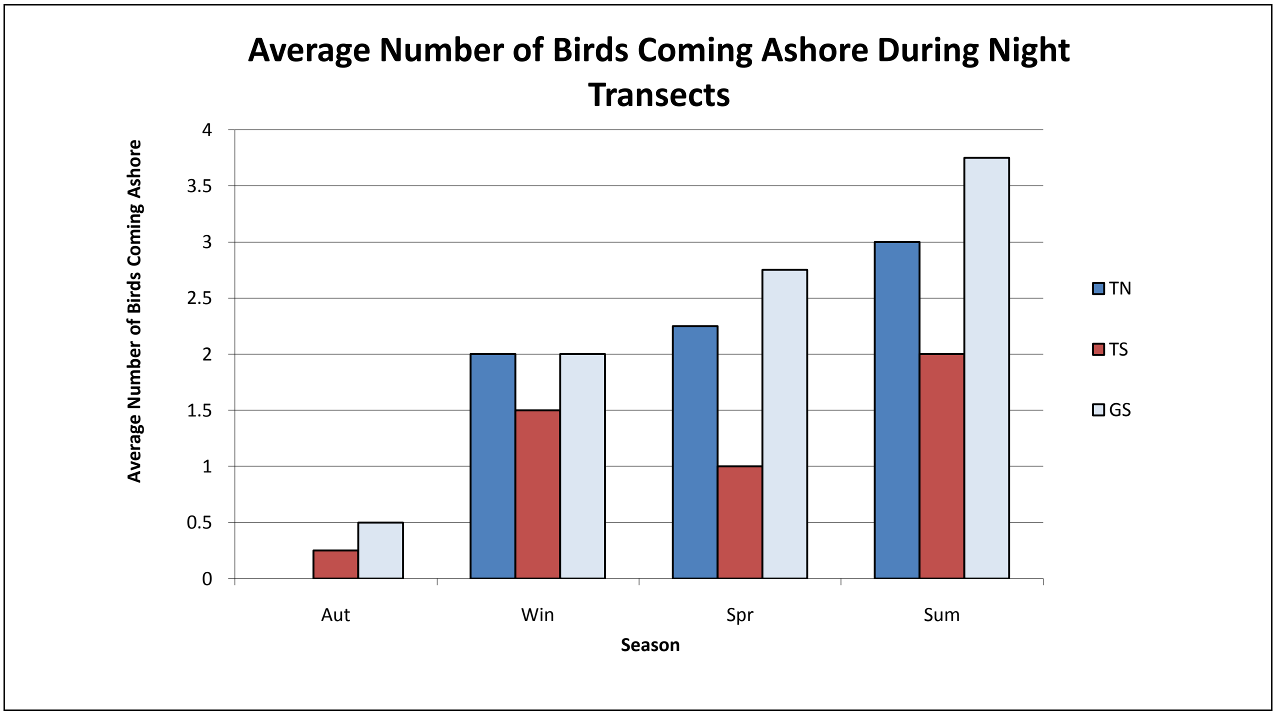

Figure 2.1: Mean number of Little Blue Penguins coming ashore for each season (autumn, winter, spring and summer) for each of the three study sites (Tawharanui North, Tawharanui South, and Goat Island mainland) from March 06 – Feb 07. TN = Tawharanui North, TS = Tawharanui South, GS = Goat Island.

A general graphical trend was identified (Fig 2.1), where for most seasons Goat Island had the highest number of birds arriving ashore, followed by the northern coast of Tawharanui and finally the southern coast of Tawharanui. This follows nest density trends reported above and again was an unexpected divergence from the hypothesis that both coasts of Tawharanui would have a similar abundance and that this abundance would be higher than the abundance at the Goat Island site. However a Kruskal-Wallis test showed that there was no significant difference in the number of birds coming ashore was found between the three sites, df=*2*, P>0.05

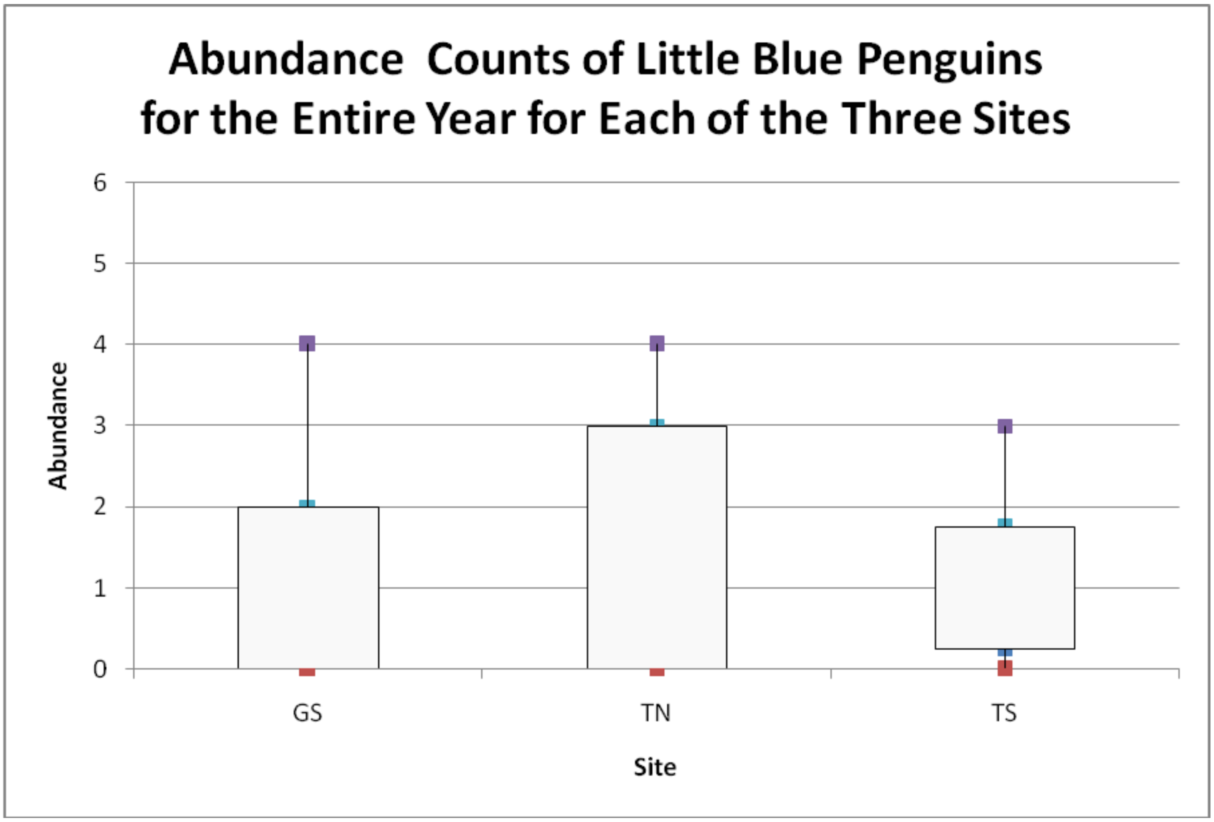

Figure 2.2: Distribution of abundance counts for each of the three sites (Tawharanui North, Tawharanui South, and Goat Island mainland) over the entire year (autumn, winter, spring and summer data combined) from March 06 – Feb 07. TN = Tawharanui North, TS = Tawharanui South, GS = Goat Island. Minimum, 1st Quartile, Medium, 3rd Quartile, and Maximum values are all displayed.

Graphically (Fig 2.3), abundance over seasons was lowest in autumn and increased over time throughout the seasons right through until summer. The exception to this was the southern coast of Tawharanui where very surprisingly, winter was the season with the second highest abundance recording. These results differed from the hypothesis that winter would correlate with the fewest number of birds coming ashore. However, data revealed that indeed, the highest numbers of birds came ashore in summer, as expected. A Kruskal-Wallis test showed there was a significant difference in the number of birds coming ashore (our measure of abundance) during the four different seasons, df= *2*, P<0.05

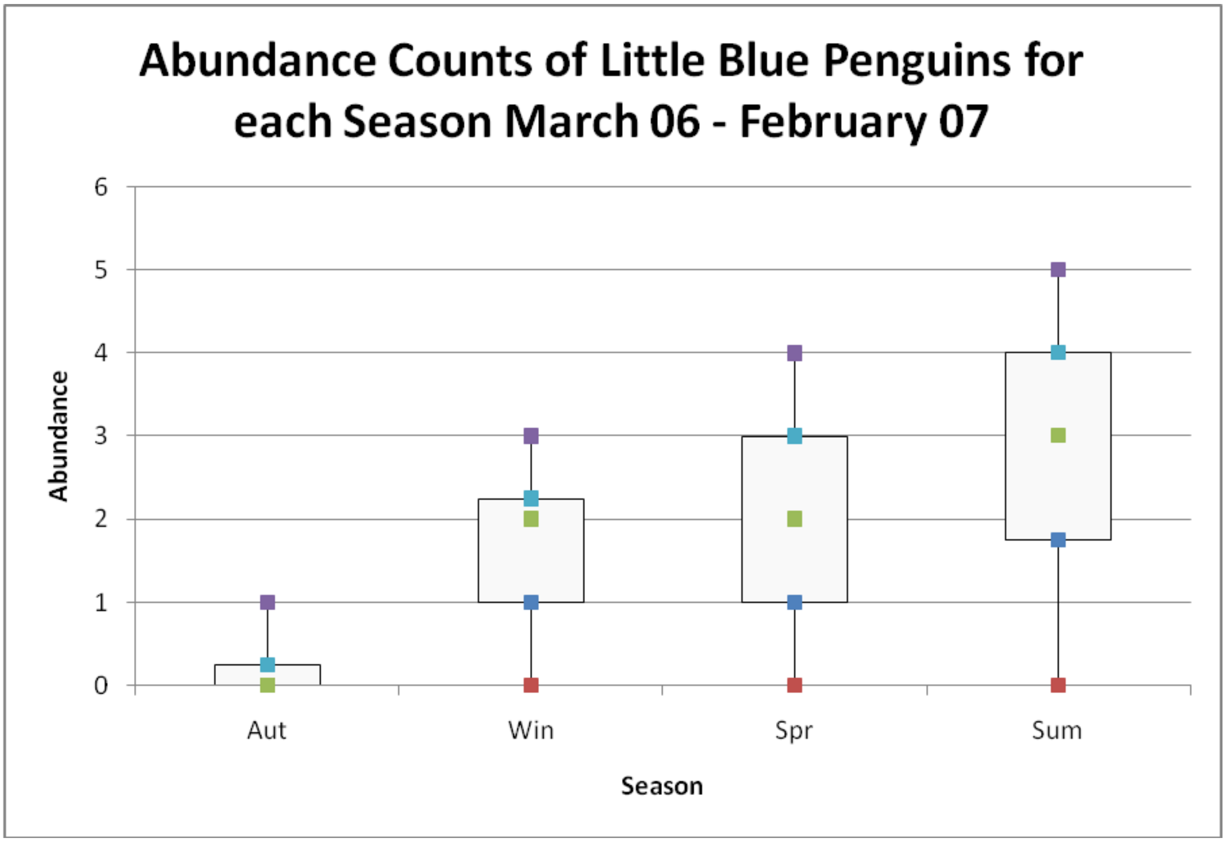

Figure 2.3: Distribution of abundance counts for each of the four seasons (autumn, winter, spring and summer) across all three sites (Tawharanui North, Tawharanui South, and Goat Island mainland combined from March 06 – February 07. Minimum, 1st Quartile, Medium, 3rd Quartile, and Maximum values are all displayed).

Measures of effort were recorded for each of the night transects (time spent searching and the number of people present). Calculating the number of birds coming ashore per unit of effort did not aid the interpretation of the results. This may have been confounded by the fact that the sample sizes are small.

5 Discussion

Comparing the results of the current abundance survey with data on the average abundance of Blue Penguins in Oamaru over the last 14 years (Fig 2.4), yielded both commonalities and differences. In both cases, the most common season to have the highest abundance figures was summer; however, the season with the lowest abundance differed by site. Autumn had the lowest abundance for the three northern study sites, while winter had the lowest abundance for the Oamaru population. The abundance of birds coming ashore was much greater at the Oamaru colony. The average number of birds coming ashore per night over an entire year was 51.5 birds for Oamaru (over the period 1993 - 2006), while in this shorter term study an average of only 2.25 birds came ashore per night for Goat Island, 1.81 birds per night for Tawharanui North, and 1.19 birds per night for Tawharanui South.

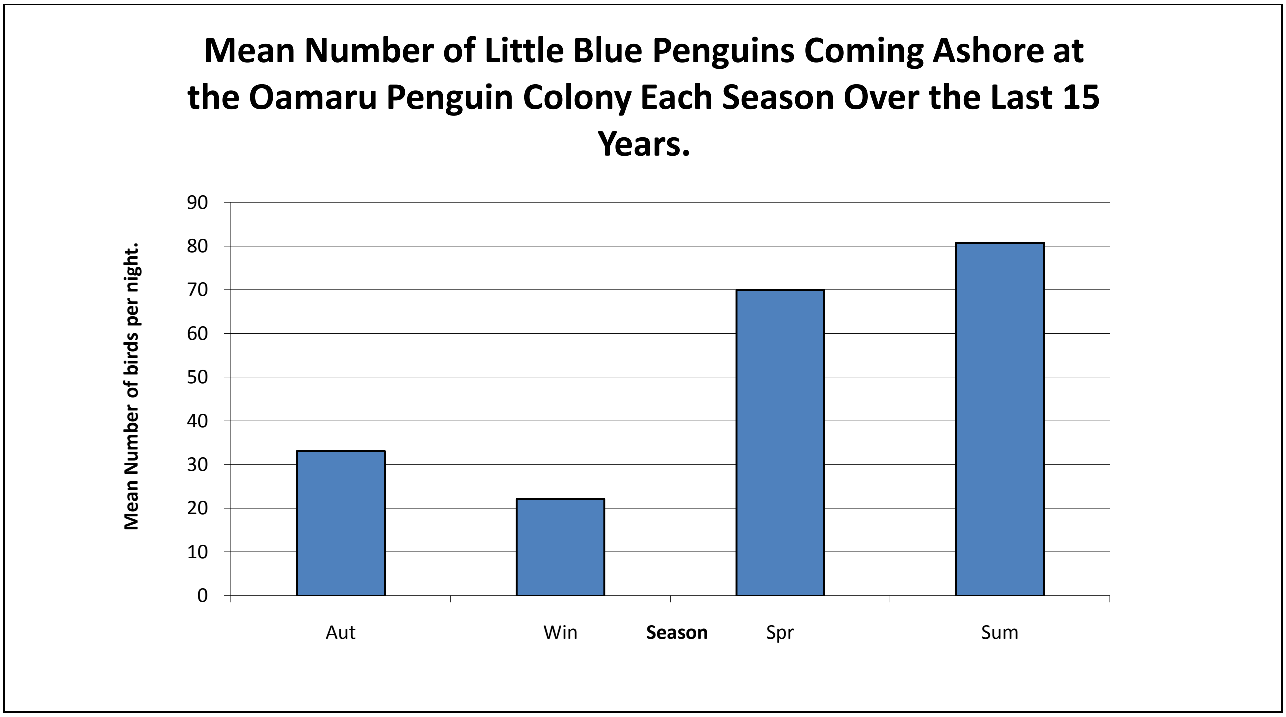

Figure 2.4: Mean number of Little Blue Penguins coming ashore for each season (autumn, winter, spring and summer) for the Oamaru Little Blue Penguin colony. The figures have been averaged from data collected from 1993 to 2006.

The nest densities discovered at each of the three sites were considerably low, all three being less than 0.01 nests/100m^2. This was much lower than densities reported in other studies in different locations. For example, nest density on Taieri Island (Australia) was reported as 2.17 nests/100 m^2 (Dann, 1994b), and Philip Island (Australia) has been found to have a density of 4.1 nests/100 m^2 (Marchant & Higgins, 1990), while the density on Bowen Island (Australia) has been recorded even higher again, at 9-12 nests/100 m^2 (Fortescue, 1999).

The results of abundance and nest density measurements in this present study were unexpected and did not fit with the hypothesis that levels of predator access would determine such parameters. It is most likely that a single variable, such as predation by terrestrial predators, is not the sole factor affecting the abundance and nest density of Little Blue Penguins. What is clear from these results is that, in the case of comparison between the three study sites at Tawharanui and Goat Island, predator access is not the variable most strongly affecting population abundance and nest density.

Examining the variables that have a greater effect on abundance and nest density of Little Blue Penguins in the northern region would make an interesting study. Such a study would have many factors to consider; for example, Dann, Norman, Cullen, Neira, and Chiaradia (2000) found that shortage in a food source caused mortality and breeding failure in Little Blue Penguins. Given that Little Blue Penguins tend to spend a large amount of time foraging for food within 15km of their burrows (Weavers, 1992), it is likely that local food supply plays a significant part in the abundance and nest density of a site. Although this study found that terrestrial predators were not the sole factor affecting abundance and nest density of Little Blue Penguins, marine predation was not taken into account and may also play a part. Additionally, it may be important to consider the dispersal of chicks. Reilly and Cullen (1982) found that after dispersal, some birds return to their natal colony to breed, while others breed elsewhere. The percentage of chicks returning to breed at their respective natal colonies for Tawharanui and Goat Island is not known.

Winter (2000) found that areas more densely populated with Little Blue Penguins were also more heavily utilized by humans. This study found a similar pattern, with the less frequented southern coast of Tawharanui having the lowest abundance and density, while the more popular northern coast had a higher abundance and nest density. This is in contrast to other studies which conclude that human disturbance can reduce the number of Little Blue Penguins present at colonies (Boersma, 1991; Williams, 1995). This conclusion may, however, be premature as each of the three study areas began at a point of public access and an interesting observation (noted above) was that no nests were found to be located within the first 500m of the point of access at any of the study sites. This could be a result of avoidance of human disturbance.

It was hypothesised that the northern and southern coasts of Tawharanui would have similar abundance and nest density of Little Blue Penguins, as the sites are in such close proximity to each other. This hypothesis did not hold true, and explanations as to why may include any or all of the differences between the two coasts. A “no take” marine reserve is present on the northern coast, while no such reserve exists on the southern coast. Also, it has been observed that the southern coast is much more exposed; with stronger onshore winds often present (personal observation). Challies and Burleigh (2004) found that colonies located at more sheltered sites tended to be larger than those at more exposed sites. Lastly, although the sites are in close proximity to each other, there may be substantial variations in habitat or nest availability that could lead to the noted differences in abundance and nest density.

The seasonal changes in abundance recorded in the current study differed from the long term data collected in Oamaru. Most noticeably, the winter season was not associated with the lowest records of abundance in the present study. The abundance for Oamaru was averaged over a long term study, while the data used in this study was collected over only one year; so it is possible that the data collected from the single year does not accurately represent the normal pattern of the populations.

As more night transects were conducted, it became obvious the birds that were sighted coming ashore were known individuals. These individuals were often seen coming ashore near their nests or regular roost sites (personal observation). This observation also suggested that the density and abundance of Little Blue Penguins at all three of the study sites was low. Furthermore, it may provide a certain confidence that the abundance and densities reported here are relatively accurate, as unknown individuals with un-located nests were seldom seen coming ashore during the night transects.

In larger colonies of Little Blue Penguins, synchronized arrivals and departures are common. The behaviour of "raft" formation and synchronized arrival ashore may act as a predator-distraction (Klomp & Wooller, 1991). This behaviour occurs occasionally on Tiritiri Matangi Island in Auckland's Gulf; however, it has not once been observed at either Tawharanui or Goat Island (personal observation). This could possibly be another confirmation that abundance and density are low at both of the Tawharanui and the Goat Island locations, assuming that the lack of synchronization is a result of low population densities and greater space between nests.

5.1 Issues

Weerheim, Klomp, Brunsting, and Komdeur (2003) cite that fewer nests were found in each month of searching when compared to the cumulative total, indicating that a single population count will underestimate a population's size. Due to the fact that the number of nest sites varies over the duration of the breeding season, the timing and length of a population count will certainly affect the outcome. Judging the number of penguins in an area can be a very difficult task to undertake successfully. Although the estimates obtained in this study may not be 100% accurate and are not based on large sample sizes, they still function well as relative measures. Additionally, as mentioned above, the fact the same individuals were sighted regularly during night transects gives confidence that the estimates are accurate in depicting low levels of abundance and nest density.

Even though due care was taken to spend equal time and effort searching for nests at each study site, it is possible that this could be one of a number of sources of bias in locating of nests. Another such bias could occur because tracks of flattened grass and guano trails make it easier to find nests that are frequented more often. Guano stains are the most easily recognisable sign of penguin activity along a shore line. They are also more noticeable in areas of large boulders or flat rock, as opposed to fine sand or grass. It is uncertain if more nests were found to be located amongst boulders and rock crevasses as a result of the fact that they were easier to identify in these locations, or because more nests were, in fact, built at these sites.

Nest density counts only measure those nests that were being used by active breeders, therefore underestimating the true density of the population. Dann (1994b), along with Johannesen, Steen, and Perriman (2002), found that population estimates were most precise (with the narrowest confidence intervals) when taken at the time when most adults had finished guarding chicks and were making daily foraging trips to feed the chicks each night. This indicated that the summer season may give the most precise estimate of abundance.

5.2 Implications and Recommendations

The main implication of this data is that, although terrestrial predator control may improve breeding success, it cannot be assumed that reduced terrestrial predation will in itself be enough to confer a greater nest density and abundance in Little Blue Penguins. More research is needed into what makes one habitat more preferable than another for Little Blue Penguins. As the erection of the predator exclusion fence and the establishment of a pest management program at Tawharanui only occurred in late 2004, it is possible the effect of the predator management program there is yet to be seen and nest density and abundance has remained low secondary to predation. Another study comparing Goat Island and Tawharanui should be conducted in five to ten years to determine if this is the case.

6 Summary

Goat Island had the highest nest density and abundance of the three study sites, followed by the northern coast of Tawharanui, and finally the southern coast, although none of these graphical trends were proven to be statistically significant. The differences in season, however, were shown to be statistically significant. The months of summer saw the greatest number of birds coming ashore across all three study sites, while the months of autumn recorded the lowest number of birds coming ashore.

Although there was no significant difference between sites in terms of abundance or nest density, it is clear that all three sites investigated were much lower in abundance that the Oamaru Penguin Colony. It is unlikely that one factor alone could be responsible for any such difference or reduction in abundance and nest density. The scope of relevant factors includes shelter, habitat type, resource availability, dispersal of chicks and predator (marine and terrestrial) access, among others. The next chapter will investigate one of these variables—habitat differences between the sites.

The current study of abundance and nest density at Tawharanui and Goat Island highlights that developing hypotheses about even basic biological measurements is not always as simple and clean cut as one may initially presume. Thus, the truth that biology often involves confounding, and sometimes unseen, variables that impact results is manifest in these findings.

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CHAPTER 3 - AN INVESTIGATION INTO GENERAL HABITAT & LITTLE BLUE PENGUIN (*EUDYPTULA MINOR)* NEST HABITAT COMPOSITION AT THREE NORTH AUCKLAND SITES

1 Abstract

Habitat surveys were conducted at three sites: Tawharanui North, Tawharanui South and Goat Island. These locations north of Auckland were known to be inhabited by Little Blue Penguins. The investigation included exploration of the substrate and vegetation structure of the three sites as well as the substrate and vegetation structure at known active nests within the three sites. Differences in habitat structure were found at all three sites. Tawharanui North was predominantly sand and flax, and Tawharanui South was predominantly pebbles and scrub, while Goat Island displayed mostly sand with no vegetation. The results suggested that grass and boulders make up the Little Blue Penguins' preferred nesting habitat and it was predicted that sites containing more of this habitat type would have higher nest densities and abundance, although small sample sizes prevented this from being validated statistically. Parameters such as nest density and abundance are likely to be affected by many factors; therefore, correlation with increases in these levels could be only partially explained by differences in nesting habitat.

2 Introduction

As previously mentioned in Chapter 2, predator access did not provide a good basis for predicting which of the three sites (Goat Island, Tawharanui North and Tawharanui South) would have the greater nest density and abundance of Little Blue Penguins. This chapter investigates the habitat structure (substrate and vegetation) present at each of the study sites and the micro-habitat structure located at nest sites. The rationale for this is firstly, to determine any similarities or differences between the sites in terms of habitat structure, and secondly, to determine if there is a preferred habitat type for nesting.

Habitat type and quality have been shown to play a part in determining parameters such as nest abundance and breeding success in previous studies (Bull, 2000; Knight & Rogers, 2004; Trezise, 1999). Trezise found that the number of Little Blue Penguin nests was inversely related to the height of the kikuyu grass growing in the area. His study also found entanglement in kikuyu to be a cause of death for Little Blue Penguins. Little Blue Penguins have been found nesting in kikuyu at Tawharanui and Goat Island (personal observation) and on Montague Island (Weerheim, Klomp, Brunsting, & Komdeur, 2003). However, current literature also suggested that as kikuyu matures and becomes dense, the suitability of such habitat decreases (Weerheim et al., 2003).

This study aimed to test the following hypotheses:

1. The habitat structure of Goat Island will differ significantly from that of Tawharanui,
2. The habitat structure of the two coasts of Tawharanui will be fairly similar.

These predictions were based on the relative geographical location of the sites.

3. There will be a nesting habitat preferred by Little Blue Penguins and such habitat types will be more abundant at Goat Island.

This second hypothesis was based on the higher density of nests at Goat Island (see Chapter 2), and previous research that showed habitat type could have an effect on nest density (Trezise, 1999) and breeding success (Bull, 2000; Knight & Rogers, 2004).

3 Methods

3.1 Data collection

Data collection for the current study involved two parts, the first of which was collection of data on the general habitats at each of the three study sites, and the second of which was collection of data on the micro-habitats surrounding each nest that was located during the 2006/2007 breeding season. Weerheim et. al (2003) noted the value of using a similar method when he commented on using line transects to investigate the relationship between Little Blue Penguins and their breeding habitat.

3.1.1 General habitat

Surveying general habitat at the study sites involved the set up of a transect that measured 2.5 km along the coast at each study site. Habitat type was surveyed and recorded every 50 m from the beginning of the transect, resulting in 51 survey points at each site. The substrate type was recorded for an area that ranged from the mean high tide mark to the beginning of vegetation. The vegetation type was recorded for the area from the beginning of the vegetation to approximately 30 m behind the vegetation.

Substrate types were classified into the following categories: sand, rock (continuous rock), pebbles (an average diameter between 2 cm and 50 cm) and boulders (a diameter greater than 50 cm). These classifications were chosen as they were present in at least one of the sites. Often there would be more than one classification present and in these cases, only the dominant type was recorded.

The vegetation types were classified into categories of dominant plant types: flax (dominated by *Phormium tenax*), grass (dominated by pasture grasses, mostly Kikuya [*Pennisetum clandestinum*]), scrub (dominating species included Karamu [*Coprosma robust*], Manuka *[Leptospermum scoparium]*, Bracken [*Pteridium esculentum*]) and lack of vegetation. These classifications were chosen because they were present in at least one of the sites.

3.1.2 Micro-habitat at nest sites

The dominant vegetation and the dominant substrate types were both recorded for each nest site. The same categories were used in recording nest habitat type and general habitat survey.

3.2 Data analysis

Manipulation and graphical analysis of the data was conducted using Microsoft excel 2000, while statistical analysis was done using Minitab 13. An Anderson-darling test for normality showed that the occurrence of substrate and vegetation types for the general habitat at each of the three sites is normally distributed ($P>0.05$ for both) while the distribution of substrate and vegetation types at active nest sites is not normally distributed ($P<0.05$ for both). Chi-Squared tests were used to test the independence between the tabulated categorical variables of the general habitat data. The assumptions that the sample is randomly selected from the population and that each cell has a value larger than 5 are met. The sample sizes for the substrate and vegetation types at active nest sites were too small to allow for statistical analysis.

4 Results

4.1 General Habitat

4.1.1 Substrate

Sand was the most frequently observed substrate type along the 2.5 km transect at the Goat Island (45%) and Tawharanui North (33%) study sites. However, pebbles (69%) were the most common substrate type on the southern coast of Tawharanui. Goat Island had the highest percentage of boulders (37%), while the southern coast of Tawharanui had the highest percentage of pebbles. Pebbles and boulders were the substrate types most often found at active nest sites. Statistically significant differences occurred when the substrate composition of the three sites was compared: $X^2(\mathrm{df}=6, N = 153) = 57.534$, $P<0.05$. A chi squared test between Tawharanui North and Tawharanui South showed there was also a statistically significant difference between the substrate compositions at each of these two sites: $X^2(\mathrm{df}=3, N = 102) = 1.805$, $P<0.05$.

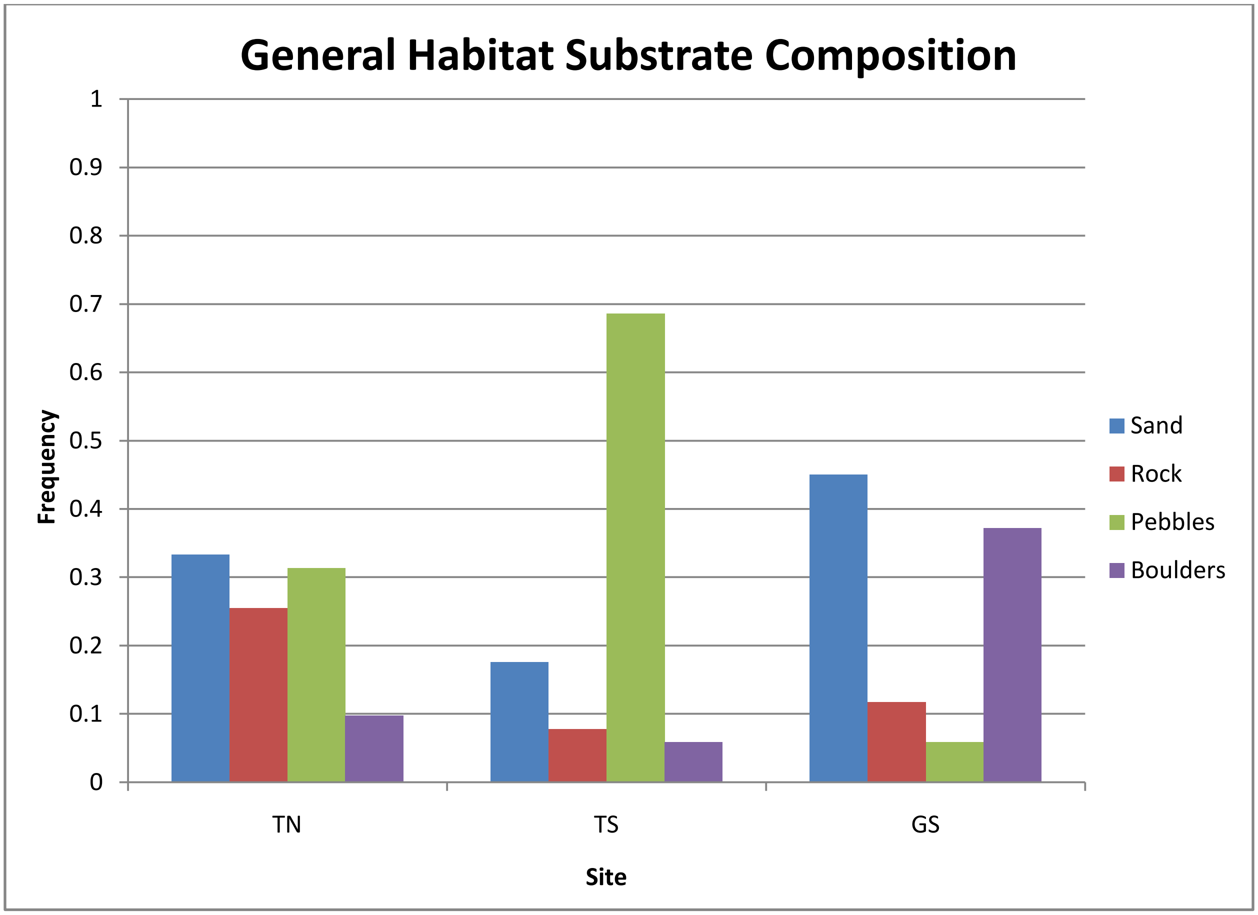

Figure 3.1: Frequency of dominant substrate types present at each of the three study sites. The graph shows the point data that was collected every 50m along the 2.5km study sites. TN = Tawharanui North, TS = Tawharanui South, GS = Goat Island.

4.1.2 Vegetation

Flax was the most common vegetation type along the 2.5km coastline of the study site on the northern coast of Tawharanui, while scrub was the most common along the southern coast of Tawharanui. Vegetation was sparse at the Goat Island site and 'no vegetation' was the most common category recorded there, followed closely by the 'grass' category. The difference in vegetation composition at the three sites was statistically significant: X^2(df=6, $N = 153) = 35.518$, P<0.05. A chi squared test between Tawharanui North and Tawharanui South showed there was also a statistically significant difference between the vegetation composition at each of these two sites X^2(df=3, N = 102) = 17.030, P<0.05

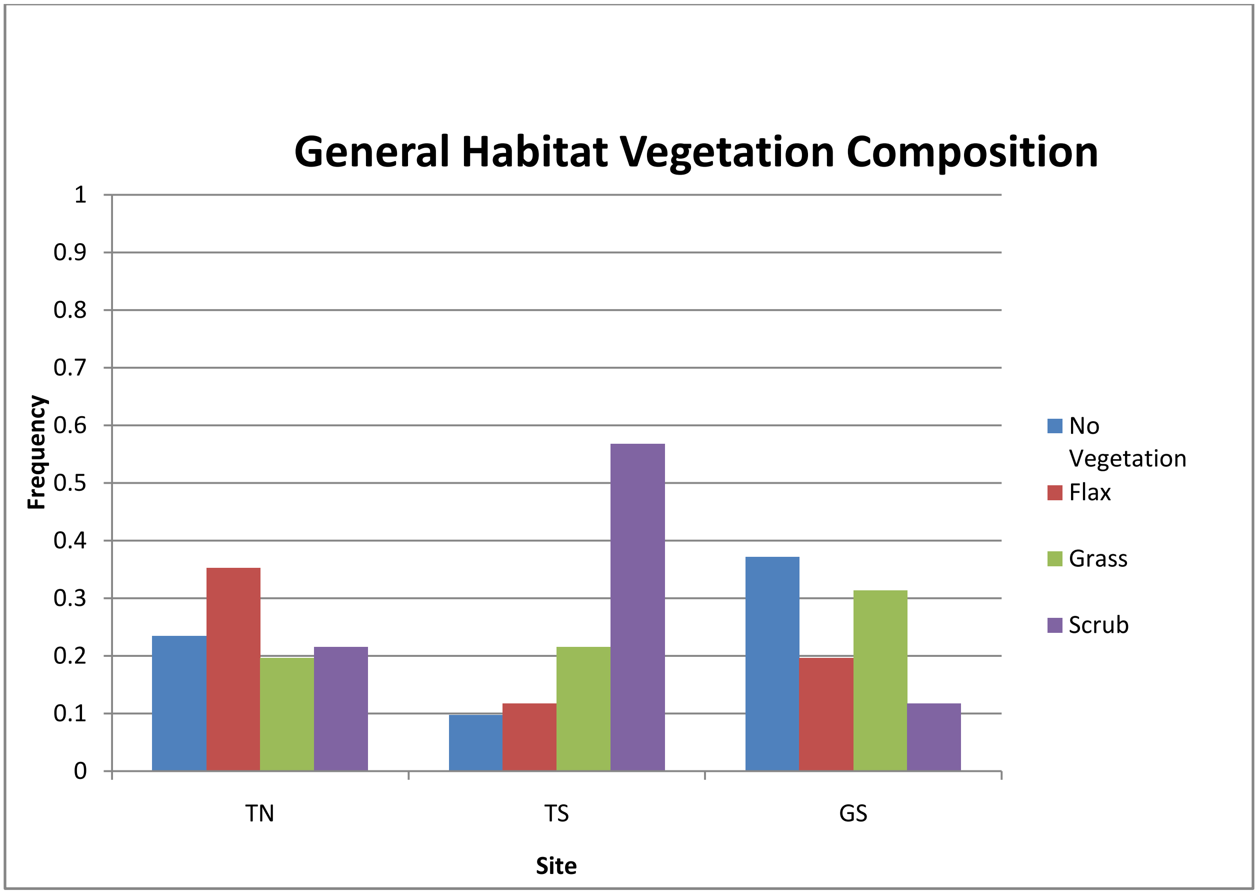

Figure 3.2: Frequency of dominant vegetation types present at each of the three study sites. The graph shows the point data that was collected every 50m along the 2.5km study sites. TN = Tawharanui North, TS = Tawharanui South, GS = Goat Island.

4.1 Nest Micro-Habitat

4.2.1 Substrate

Over the 2006/2007 breeding season, habitat surveys were carried out on eight active nests at Tawharanui North, six at Tawharanui South and 12 at the Goat Island site.

The two most frequent substrate types found at active nest sites were pebbles and boulders (fig3.3). Pebbles were the dominant substrate present at active nests at Tawharanui North and Tawharanui South, while the dominant substrate at active nests at Goat Island was boulders. Tawharanui North and South appeared to be similar in substrate structure surrounding nest sites, while Goat Island appeared to differ from Tawharanui North and South in this variable. However, sample sizes were too small for statistical analysis.

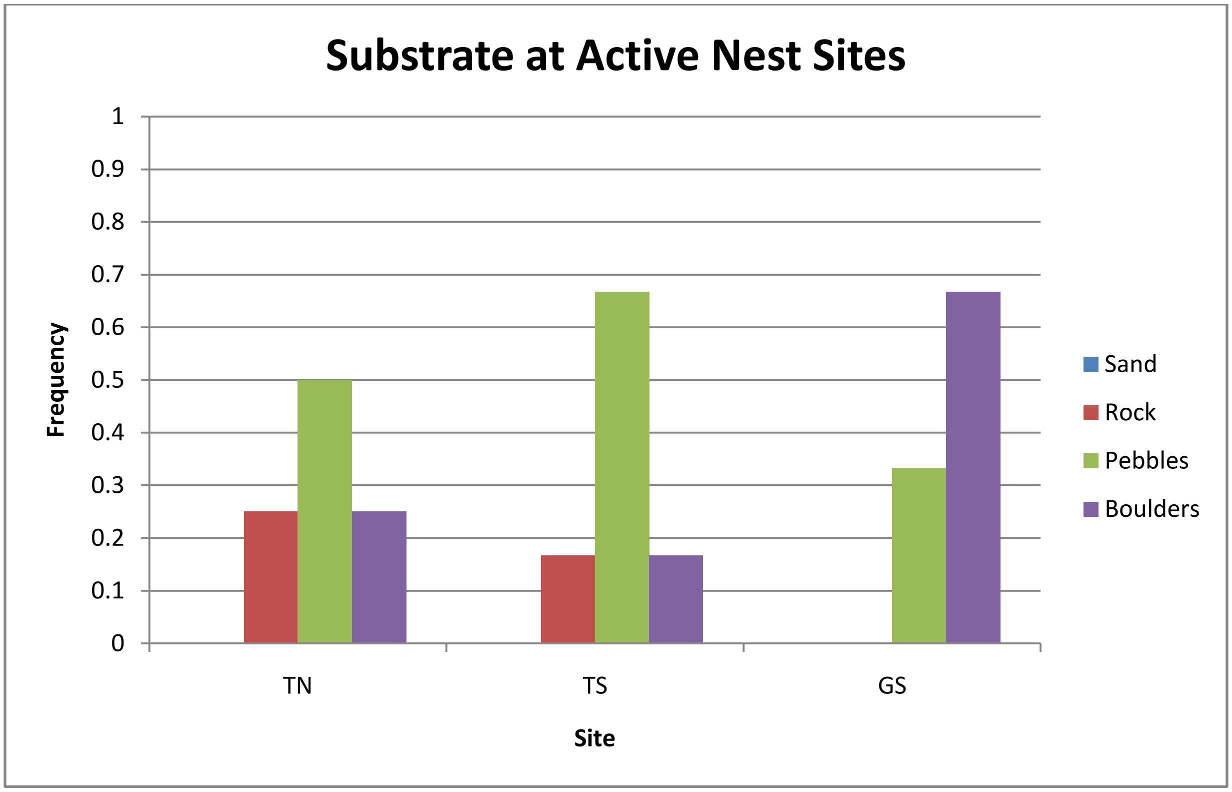

Figure 3.3: Dominant substrate types found at and around (5m radius) active nest sites from all three study sites. Definitions of the substrate types are made above in the methods section. The ‘y’ axis gives the frequency of nests with that substrate present. TN = Tawharanui North, TS = Tawharanui South, GS = Goat Island.

4.2.2 Vegetation

Vegetation at Goat Island was characterized by relatively equal amounts of grass and flax; Tawharanui South had more grass and scrub, while ‘no vegetation’ was the leading category at Tawharanui North (Fig 3.4). Again, sample sizes were too small for statistical analysis.

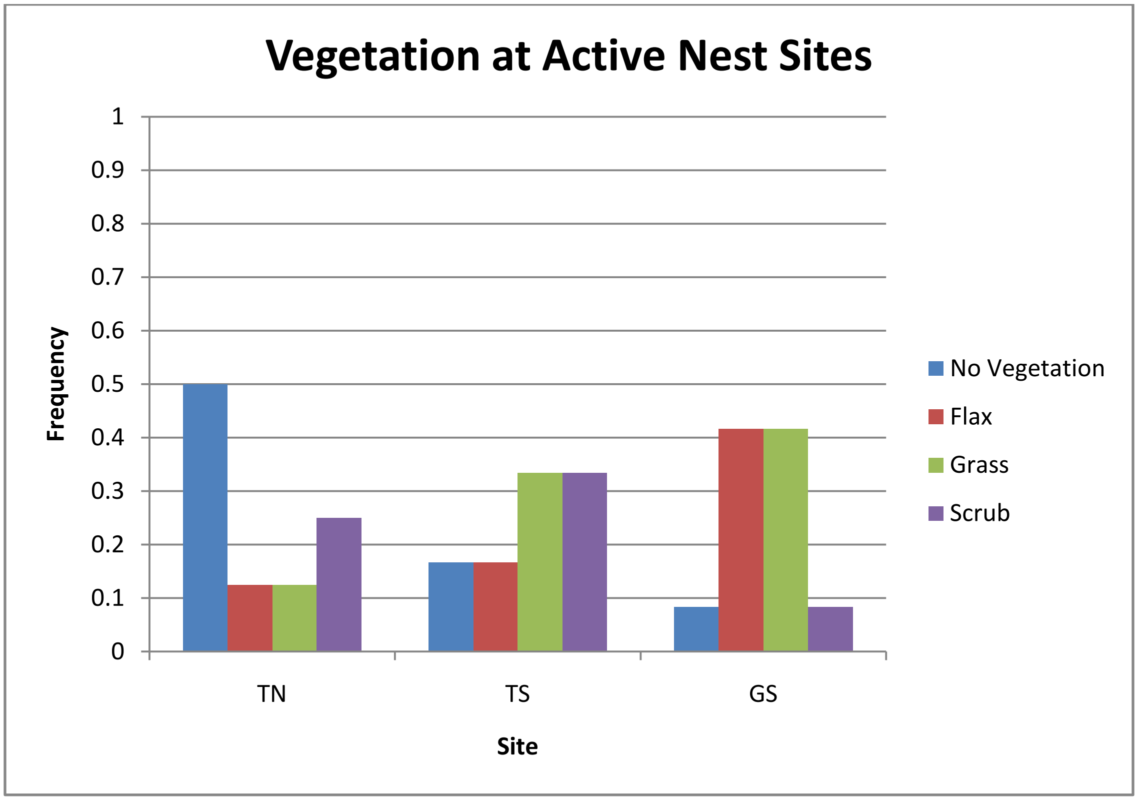

Figure 3.4: Dominant vegetation types found at active nest sites from all three study sites. The ‘y’ axis gives the frequency of nests with that vegetation type present. TN = Tawharanui North, TS = Tawharanui South, GS = Goat Island.

4.3 Summary of results

Table 3.1
Summary of the Dominant Substrates and Vegetation Types

	Dominant substrate type along coast	**Dominant substrate type at nest sites**	**Dominant vegetation type along coast**	**Dominant vegetation type at nest sites**
TN	Sand	Pebbles	Flax	No Vegetation
TS	Pebbles	Pebbles	Scrub	Grass/Scrub
GS	Sand	Boulders	No Vegetation	Flax/Grass

Table 3.1 Summary of the dominant substrates and vegetation types for both the general coast surveys and the micro-habitat nest surveys. Sample sizes (for micro-habitat composition at active nests) were too small to allow for statical analysis between the general habitat composition and the composition of habitat found at active nests. TN = Tawharanui North, TS = Tawharanui South, GS = Goat Island.

A statistically significant difference in the proportions of substrate and vegetation present at each of the three sites was demonstrated. Grass was common at all active nests but was represented in the greatest quantities at the Goat Island site. Boulders were also regularly found at active nests, once again appearing more often at the Goat Island site than at the other two sites. Sample sizes for the recording of substrate and vegetation at the actual nest were not large enough to allow for any statistical evaluation. However, graphically and descriptively it appeared that pebbles and boulders were the most frequently observed substrate present at active nests, while grass and flax were the most frequently observed vegetation types. Again, it appeared graphically that active nests at Tawharanui North and Tawharanui South were similar in their substrate and vegetation types, while Goat Island differed from these two sites (although this cannot be statistically verified due to the small sample sizes). Statistical tests on the make up of substrate and vegetation compositions present at Tawharanui North, and Tawharanui South showed there was a significant difference between these two sites in the frequency of various composition types.

5 Discussion

This study investigated whether or not any habitat differences between Tawharanui and Goat Island could explain why a higher nest density and abundance of Little Blue Penguins was found at the Goat Island site (where predator access is greater than at Tawharanui). Habitat type and quality have been shown to play a part in determining parameters such as nest abundance and breeding success in previous studies (Bull, 2000; Knight & Rogers, 2004; Trezise, 1999).

Many penguin colonies are being threatened by loss of habitat. Harris and Bode (1979) attributed such a loss on Phillip Island (Australia) to real estate development and tourism. Differences in nest density and abundance are likely to be caused by many things, including food supply, predation, parasite load, habitat loss and habitat type, so a single factor is unlikely to be the sole determinant. However, habitat type has been shown to have an impact on such parameters (Bull, 2000; Knight & Rogers, 2004; Trezise, 1999).

5.1 Substrate

Boulders were found to be one of the most common substrate types observed at active nest sites. Boulders were more prevalent at the Goat Island site than at the Northern and Southern coasts of Tawharanui. A comparison of boulder frequency shows that Goat Island has the highest percentage of boulders present within its 2.5km of study site, followed by the northern coast of Tawharanui, and finally the southern coast of Tawharanui. This pattern is in direct correlation with the patterns of nest density and abundance rates at the three study sites. Thus, the relatively higher number of boulders at the Goat Island site could account in some part for the fact that nest abundance is also higher at the Goat Island site.

However, pebbles were also frequently associated with active nest sites; this substrate type was the most pervasive type observed on the southern coast of Tawharanui where nest density and abundance were lowest. It is possible that active nests at Tawharanui (North and South) showed a high use of pebble substrate because boulders were low in number and therefore, not as available.

It was expected that the general substrate types used for nest building would be similar at the two Tawharanui sites. However, a chi squared test performed on the proportions of substrate classifications at the two Tawharanui sites showed that this was not the case.

Bull (2000) made personal observations that penguins who nested in soil burrows that collapsed or in rock nests that shifted, deserted the nest and did not return. During data collection for this study it was observed that some types of nests appeared more resistant to environmental disturbances than others and that boulders provided a safer and more stable nesting habitat. Nests deserted due to trampling by sheep, and adult penguins killed in their nests due to slips of rock and loose material were both witnessed during the course of this research.

5.2 Vegetation

One of the most abundant vegetation types found around active nests was grass and as reported above, grass was present more often at the Goat Island site than at either of the other sites. Again, the presence of more grass at Goat Island could account in part for the greater abundance that was recorded at that site. The two coasts of Tawharanui were fairly similar in the percentages of vegetation that were recorded as grass; however, the southern coast of Tawharanui had a slightly higher percentage of it. This caused calculations to reveal a non-direct relationship between the percentage of grass present at a study site and the levels of nest density and abundance. Flax was also often found at active nest sites; yet, it was more abundant on the northern coast of Tawharanui than it is at the Goat Island site.

Similar to the findings concerning substrate types described above, it was expected that the levels and combinations of general vegetation types would be similar at the two Tawharanui sites. However, once again a chi squared test performed on the proportions of vegetation classifications observed at the two Tawharanui sites showed this was not the case.

5.3 Issues

Similar to the discussion in the previous chapter on nest density and abundance, it must be acknowledged here that nests are more easily located in surroundings that feature pebbles, rock and boulders, due to their visual contrast with the guano stains that accompany active nesting.

Therefore, the finding that boulders and large pebbles were the most common substrate located around active nest sites may reflect such an artifact and bias in locating nests.

Additionally, the sample size of active nests was relatively small (26 across all three sites). A sample size this small leads to difficulties in drawing conclusions and statistically validating them. Yet when investigating a population in an effort to find out why it is so small, sample size, by implication, will necessarily be low—an interesting paradox!

5.4 Implications & Recommendations

As discussed in Chapter 2, many factors may influence the abundance and nest density of any particular site. If habitat structure or nest availability has proved to be a limiting factor at Tawharanui, then these issues will affect population increase at the reserve in the future. As data from the current study has suggested that boulders are a preferred nesting habitat, it is recommended that nesting boxes resembling boulders be placed around the Tawharanui coast to increase suitable nesting habitat, potentially contributing to increased abundance of Little Blue Penguins in the area. It would be interesting to see if such nesting boxes would in fact be used along the Tawharanui coast and whether or not they help produce an increase in abundance levels. If such nesting boxes are used, it may also make studying Little Blue Penguins at Tawharanui easier, as many of the nests located during this study were either difficult to access, or actually totally inaccessible.

6 Summary

It was expected that the habitat structure of Goat Island would differ significantly from that at Tawharanui, while the habitat structure of the two coasts of Tawharanui would be fairly similar. In fact, all three sites differed significantly from each other regarding the proportions of habitat substrates and vegetation categories found in the area and at active nest sites. It was also predicted that a preferred nesting habitat would be identified, and that this habitat type would be observed more frequently at Goat Island, where abundance and nest density levels were highest. Unfortunately, small sample sizes prevented the identification of a statistically validated preference in habitat substrate, and therefore in nesting habitat.

A general trend was identified, however, revealing that a higher percentage of boulders and grass correlated with a higher level of nest density and abundance. This may indicate that these categories of substrate and vegetation, regularly found at active nest sites, are important factors in achieving optimal nest density and abundance. However, such a conclusion should be considered with caution, because other nest habitat categories frequently found at active nest sites did not follow the same trend, indicating that this relationship may not be reliable or generalisable. For example, pebbles and flax were also commonly found at active nests; however, they were not the most frequent characteristics of active nest sites at Goat Island, where nest density and abundance levels were highest. Nonetheless, it appears that many of the active nests at both Tawharanui and Goat Island—especially the more successful ones—were located amongst large boulders, an observation that adds weight to the conclusion that the amount of boulders present at a site will have a direct affect on the nest density and abundance levels of Little Blue Penguins in the area.

To reiterate, active nests were most often found on and around substrate types classified as boulders and large pebbles, and vegetation types categorized as grass, flax, and rock faces. A statistically significant direct relationship was identified, such that a higher percentage of boulders correlated with higher nest density and abundance levels at all three sites. The same relationship did not hold true for pebbles, flax or grass.

The issues of possible bias and low sample size have been discussed above. Had the sample sizes been larger, it may have been possible to compare nesting habitat with nesting success. The next chapter will take a closer look at rates of nesting success at each of the three study sites.

7 References

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CHAPTER 4 - AN INVESTIGATION OF LITTLE BLUE PENGUIN (*EUDYPTULA MINOR*) NEST SUCCESS AT THREE NORTH AUCKLAND SITES

1 Abstract

The nest success of Little Blue Penguins (*Eudyptula minor*) was calculated for three sites, Tawharanui North, Tawharanui South and Goat Island. These three sites located north of Auckland were known to be inhabited by Little Blue Penguins. Nest success was evaluated using the Mayfield method.

This study was conducted to expand current knowledge of northern Little Blue Penguins, specifically at the three sites investigated in this study. Data was collected during regular visits and nest searches at the three sites. Key results showed that the incubation period was associated with the lowest success rate of any of the nesting stages observed. Success rates for the incubation stage at the three sites fell between 0.11 and 0.66, success rates for the guard stage fell between 0.74 and 1.00, and success rates for the post-guard stage fell between 0.62 and 1.00.

Findings also revealed that nest success was greater at the two Tawharanui sites (where similar rates of nest success were observed) than at the Goat Island site. The actual nest success rates (calculated through the Mayfield method) were 0.58 for Tawharanui North, 0.51 for Tawharanui South and 0.05 for Goat Island. Therefore, confidence in the results is uncertain. Factors most strongly influencing nest success at these sites were thought to be habitat and predation, although many other factors were likely to have contributed.

2 Introduction

Little Blue Penguin nests are usually made up of a natural cavity filled with a collection of nesting material including dry grass, seaweed and small sticks (Kinsky, 1960). Little Blue Penguins usually breed on offshore islands and, less commonly, along parts of the mainland that are inaccessible to predators. For example, in the Otago region Little Blue Penguins no longer breed at seven previously occupied mainland breeding sites (Dann, 1994). The male often courts the female through a display of vocal and visual signals. Once mating has occurred, a long-term monogamous bond is formed between the pair which can last for life (Marchant & Higgins, 1990).

Laying generally takes place between July and December (Barrie & Robertson, 2000) but this does differ with locality (Davis & Renner, 2003). Factors such as weather patterns, temperature, food abundance and body condition can all have an effect on the timing, and the length and success of breeding. Little Blue Penguins begin breeding between the ages of two and three. One to two eggs are laid three to five days apart and the chicks hatch approximately 36 days later. The chicks are brooded for the first 10 days, then guarded continuously for one to two weeks (guard stage), followed by a period of being guarded only at night (post-guard stage) (Barrie & Robertson).

The chicks fledge at approximately 54 days. It is common that only one chick survives to fledging, as one chick may receive four to eight times the quantity of food received by the other (Muller-Schwarze, 1984). Usually two clutches are laid each breeding season, with the second serving either as a replacement clutch or a double brood (Davis & Renner, 2003).

Dann, Collins and Cullen in *The Royal Society of New Zealand* (2001), has shown that only 18–20% of Little Blue Penguin fledglings contribute to the next generation, and suggested that survival is lowest soon after fledging. The mass at fledging and the time of fledging influences survival probability.

Nests are often built close to the shoreline above the high tide mark; however, anecdotal evidence suggests that they can be found up to 1.5 km inland and 550 m above sea level (T. Lovegrove, personal communication, November 11, 2005). In Otago and southern parts of Australia, Little Blue Penguins live in colonies and come ashore at night in large numbers. However, the northern New Zealand populations appear to nest more sparsely, with nests separated by hundreds of meters. The birds are known to make audible calls while coming ashore, and this behaviour is more apparent in more heavily populated colonies.

A rich body of literature exists regarding Little Blue Penguin populations in other regions. However, a deficit in breeding information on northern penguin populations remains. This deficit was noted by Taylor (2000), who stated that little is known of the size of northern Blue Penguin populations and recommended that accurate estimates be obtained from as many breeding localities as possible in order to fill this knowledge gap.

It is within this context that this chapter investigates nest success at three different sites, Tawharanui North, Tawharanui South and Goat Island. These three sites are located north of Auckland and are known to be inhabited by Little Blue Penguins (see the Introduction chapter for a full description of these sites).

This study aims to test the following hypothesis:

> Nest success rates will be greater at the two Tawharanui sites (Tawharanui North and Tawharanui South) than at the Goat Island site. Furthermore, nest success rates at the two Tawharanui sites will be similar.

This hypothesis is based on the fact that the Tawharanui sites are heavily protected and monitored for introduced predators, while Goat Island is not.

3 Methods

3.1 Background to method

Reliable estimates of nesting success are an important part of assessment strategies that contribute to successful conservation efforts (Jehle, Adams, Savidge, & Skagen, 2004). In the early 1960s, Harold Mayfield developed a method that was to herald a significant advance in nesting success calculation methodology (Mayfield, 1960; Mayfield, 1961). Later, when the use of his method became more popular, Mayfield felt it necessary to revise and provide further guidance on the application of his method in the field (Mayfield, 1975). Before the introduction of the Mayfield method, the most common means of calculating nest success was “apparent nest success”. This counting method simply takes the number of successful nests (defined as nests in which at least one young survives to fledging) and describes this as a percentage of the number of nests observed.

The apparent nest success method tends to overestimate true nest success, as many nests fail before they are detected. This causes the calculated percentage of success to be higher than it actually is in reality. This bias can be between 9% and 27% and is exaggerated by low daily survival rates (Nichols, Percival, Coon, Conroy, Hensler, & Hines, 1984). The inaccuracy in estimation was identified by Coulson in (1956) and Mayfield later developed his method to address and avoid this positive bias. He overcame the bias by taking into account the length of exposure as “nest days” or “egg days”. The method is explained in more detail below and has been widely accepted for comparing the nest success rates of different areas (Hoover, Brittinghan, & Goodrich, 1995; Robinson, Thompson, Donovan, Whitehead, & Faaborg, 1995). Thus, it was considered appropriate for use in this study.

3.2 Assumptions of method

The main assumption of the Mayfield method is that every “nest day” or “egg day” is independent, and the survival probability for each day is constant throughout both the breeding season and the nests observed. This assumption would be violated if, for example, younger breeders were more likely to fail than older breeders (Farnsworth, Weeks, & Simons, 2000). Having constant survival probabilities means that the survival for any one day cannot be dependent on the survival of the previous days, but this may not be realistic. The Mayfield method does not lend itself well to colony nesters, where catastrophe may violate this central assumption (Johnson & Shaffer, 1990).

The Mayfield method also assumes that all fledging or failure dates are known. Daily observations may not always be possible within a field regime, and daily checks may influence survival, thus violating another assumption. It is assumed that the nests found and observed are a random selection representative of all possible nests. However, observed nests are likely to be more easily found than others, and thus could be more prone to failure than nests that go undetected. In addition, the Mayfield method also assumes that finding and monitoring a nest does not have any effect on its outcome; this is an assumption that has been challenged in the past (Nichols et al., 1984). Finally, if a nest fails between observations, an assumption is made that the failure date falls midway between the two observation dates. However, Miller & Johnson (1978) proposed and demonstrated that when longer periods of time pass between observations (> 15 days), a failure is more accurately estimated to occur 40% of the way through the exposure period. Some authors have commented that the assumptions of the Mayfield method are restrictive (Jehle et al., 2004). As a result, many new additions and modifications have been developed to relax the assumptions, and a few of these are discussed below.

3.3 Alternative Methods

The Kaplan-Meier method (Kaplan & Meier, 1958) with a staggered entry (Pollock, Winterstein, Bunck, & Paul, 1989) is often used for calculating the survival of radio-tagged animals (Pollock, Bunck, Winterstein, & Chen, 1995). Cornelius (1993) adapted this method to estimate nest success. Its advantage is that it does not require the assumption that nest survival is constant throughout the breeding season.

Klett and Johnson (1982) suggested a piecewise application of the Mayfield method when an identifiable factor causes differences in survival during different breeding stages, and the survival probability remains constant within the different stages. The piecewise application involves separately considering each stage, such as incubation, guard and post-guard.

Another alternative is MARK, a very powerful program that can be used to calculate nest success (Dinsmore, White, & Knopf, 2002). Using MARK, survival can be modelled as a function of biological variables. A generous amount of data is needed for this method, and nests with uncertain fates must be discarded (Jehle et al., 2004).

Hazler (2004) presented a method that combines the strengths of both the Mayfield method and logistic regression. This method lends itself well to situations in which nests are found at various times (fragmented data), and the data is used for exploring multiple variables on nesting success (age, season, habitat, etc.).

The above methods were analysed and considered for use within this study; however, the restricted size of the data set had to be considered. Thus, many of these methods were rendered unsuitable. The Mayfield method is able to cope with relatively smaller data sets (as compared to the method presented by Hazler (2004) and the MARK program). It was decided that, given the small sample size, a piecewise application of the Mayfield method using the 40% exposure calculation for failed nests would provide the best indication of nest success.

3.4 Data collection

Regular visits were conducted at each of the three study sites leading up to and during the 2006/2007 breeding season (from August 2006 until March 2007). Each of the three sites was visited on consecutive days and the 2.5 km length of each study area was searched for new nest locations. If a new nest was located, its position was recorded, along with a description of the nest and its state (number of eggs or chicks, and whether or not the young were guarded). A site was only considered to qualify as a nest if eggs or chicks were present, thus avoiding the mistake of including rest or roost sites. Each located nest was observed on all subsequent visits and a description of their current state recorded. Many of the nests were difficult to access visually, and a fiberoptic camera was used to observe the nest status.

This process continued until the nest had either failed, or produced fledglings. Failed nests continued to be checked on the chance that new individuals might use the burrow for nesting, or that the original individuals might lay a second or replacement clutch. The length of time between site visits was between 10 and 15 days. Only one live fledgling was required from each nest for it to be regarded it as a success.

3.5 Data analysis

Manipulation of the data, including performing the Mayfield method of calculating nest success, was performed using Microsoft Excel 2007. Graphical analysis was also performed using Microsoft Excel 2007

The Mayfield 40% method (Johnson, 1979; Miller & Johnson, 1978), was used during the analysis of nest success (although there were not many occasions where length between observations reached 15 days or greater). Nest success was calculated separately for the three stages of nesting observed (incubation, guard, and post guard). The results from each of these stages were multiplied together to compute the total nest success score.

4 Results

Over the 2006/2007 breeding season, eight nest locations were found at Tawharanui North, six at Tawharanui South and 12 at Goat Island. Of the eight nests found at Tawharanui North, two were used twice. Whether or not this was by new individuals or the same individuals laying a second clutch was unclear, as physical access to band the birds was made impossible by their location and identification was therefore not viable. This yielded a total of ten nesting attempts for Tawharanui North. Of the ten nesting attempts, three attempts failed and the remaining 12 chicks fledged successfully. The Tawharanui South site had six nesting sites; one of these nest locations was used in a second nesting attempt. Of the seven nesting attempts for Tawharanui South, three attempts failed and seven chicks fledged. The Goat Island site had the greatest occurrence of second nesting attempts in the same nest location. Of the 12 nesting sites located, six were used in a second nesting attempt. Of the 18 nesting attempts at Goat Island, 13 attempts failed and 11 chicks fledged successfully. This data is summarised in the table below.

Table 4.1
Nesting attempts and successes at the three study sites

	Physical Nests	**Nesting Attempts**	**Failed Nesting Attempts**	**Successful fledglings**	**Fledglings per Nesting Attempt**
TN	8	10	3	12	1.2
TS	6	7	3	7	1.0
GS	12	18	13	11	0.6

Table (4.1). Nesting attempts and successes at the three study sites, Tawharanui North, Tawharanui South and Goat Island. Physical nests are the total number of actual nesting locations found, while nesting attempts include second clutch by the same pair and/or reusing of the nesting sites by a different pair. Successful fledglings describes the total number of fledglings produced (some nests produced two fledglings). TN = Tawharanui North, TS = Tawharanui South, GS = Goat Island.

Furthermore, it can be noted from the Table 4.1, that Tawharanui North had the greatest number of successfully fledged chicks per nesting attempt, closely followed by Tawharanui

South, while Goat Island yielded almost half the number of successful fledglings per nesting attempt.

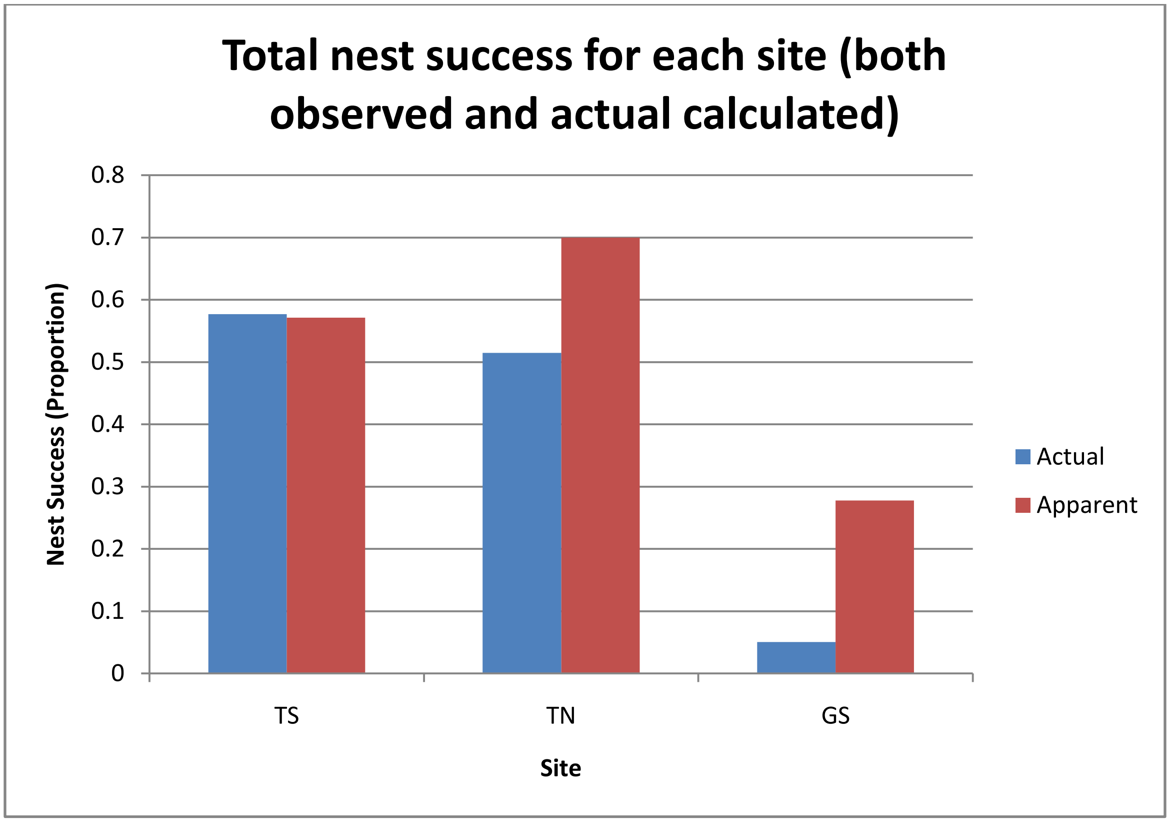

Figure 4.1 Actual and apparent total nest success for the three study sites (Tawharanui North, Tawharanui South, and Goat Island) The measure of success given is the chance that a nest will survive to produce a successful fledgling. For example (using the actual data) a nest at Tawharanui South has a 58% (0.58) chance of producing a fledgling. The actual nest success is the total nest success calculated via the Mayfield method, while the observed nest success is simply the inverse ratio of the number of failed attempts per nesting attempts. TN = Tawharanui North, TS = Tawharanui South, GS = Goat Island.

The actual nest success is the total nest success calculated via the Mayfield method, while the observed nest success is simply the inverse ratio of the number of failed attempts per nesting attempts. As mentioned earlier, the apparent nest success method (observed nest success) tends to overestimate nest success, as many nests may fail before detection. The figure above illustrates this (with the exception of the Tawharanui North site), and apparent nest success data was included on the graph for this purpose. However, when comparing nest success between sites, the actual calculated nest success should always be used, as it is known to be more accurate. Tawharanui South had the greatest actual nest success at 0.58, and this was closely followed by Tawharanui North with a total actual nest success of 0.51. Goat Island showed a much lower nest success of only 0.05.

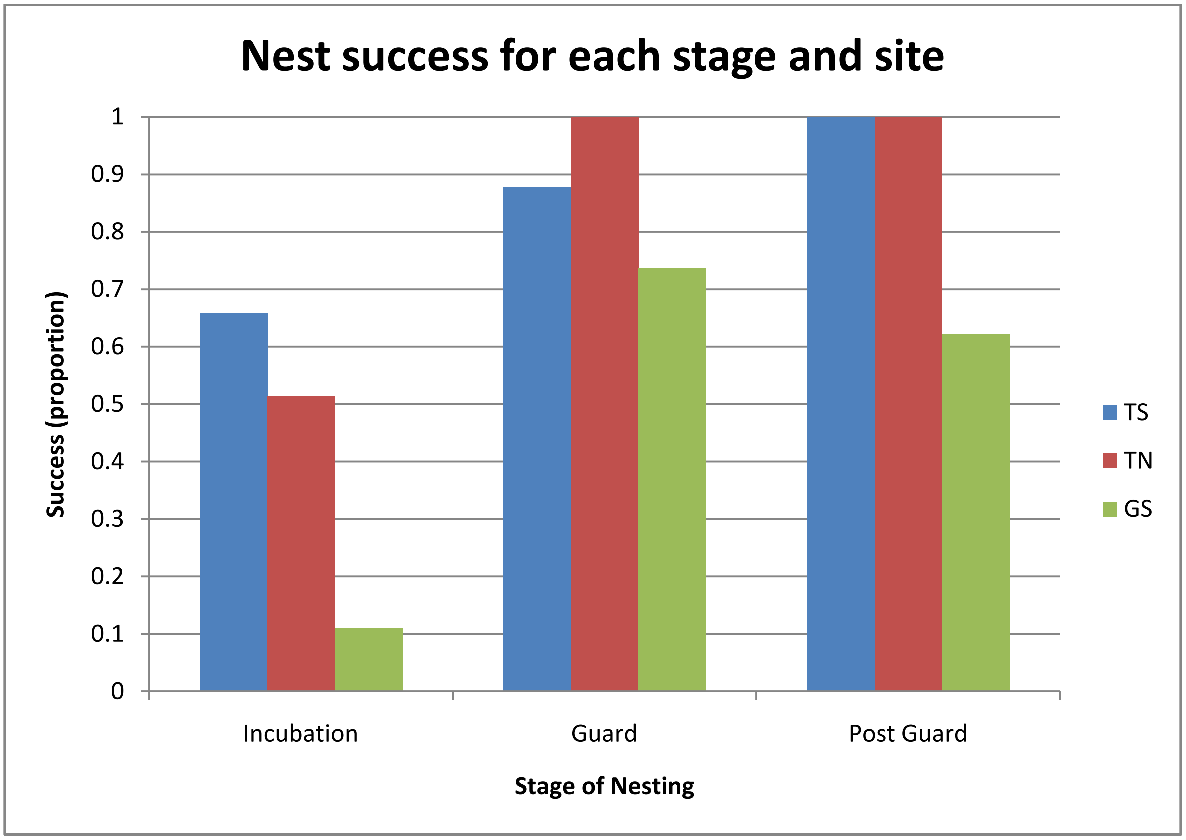

Figure 4.2 Nest success for each of the three stages of nesting observed (incubation, guard and post guard) across the three study sites (Tawharanui North, Tawharanui South, and Goat Island). The measure of success given is the chance that a nest will survive that period of nesting. For example a nest at Tawharanui South has a 66% (0.66) chance of surviving the incubation period.

The nesting success rates for each of the three observed stages of nesting are displayed in Figure 4.2. Trends in total nest success were apparent and these appeared similar to those detailed in Table 4.1. The two Tawharanui sites (North and South) were comparable in rates of nest success for each of the three stages observed, while again, Goat Island nesting success seemed to be considerably lower. For all three sites, the lowest levels of nest success were recorded at the incubation stages.

Due to the small sample sizes, no statistical test could be performed. However, apparent trends in the data are consistent between the figure and table above.

5 Discussion

It was expected that nest success would be greater at the two Tawharanui sites (North and South) than at the Goat Island site, and the results presented showed this to be the case. This hypothesis was based on the fact that the Tawharanui site is heavily protected and monitored for introduced predators, while Goat Island is not. It is likely that this difference in predator access plays a significant part in the differences in nest success between the Tawharanui sites and Goat Island. During this study, Goat Island had greater nest density and abundance than the two Tawharanui sites (see Chapter 2), suggesting that if a predator is responsible for the lower nest success at Goat Island it is one which preys on young and/or takes eggs, but not adult birds.

It should be noted that many factors other than predator access may have a considerable impact on nest success, such as food availability, habitat type and weather conditions. Due to the similarities between Tawharanui North and Tawharanui South and the close proximity between the sites, it was expected that nest success would be similar between the two. The two sites are on opposite coasts of the Tawharanui peninsula and are only separated by a small distance. Although differing in wind exposure and marine reserve status, the habitat—vegetation and substrate—is fairly similar.

From the results presented it can be seen that the two Tawharanui sites were similar in actual nest success, with Tawharanui North having a slightly higher rate of nest success than Tawharanui South (0.58 and 0.51, respectively). Both the Tawharanui North and Goat Island sites are located at marine reserves. Marine protection could have an effect on nest success for two reasons. Firstly, marine protection could result in a greater abundance of food species, therefore allowing young to be adequately fed. Secondly, marine protection could result in higher predator abundance, producing higher adult mortality which leaves more nests unattended. Little Blue Penguins on the East Coast of Auckland have one less predator—the fur seal (*Arctocephalus forster*). Due to the small sample sizes of this study and the lack of abundance measures for food and predator species, a definite conclusion cannot be drawn on how marine protection affects nest success.

The incubation stage had the highest probability of failure when compared to other nesting stages. This trend occurred across all three study sites. As well as predation, disturbance may cause nest failure. There is evidence that some birds will interrupt nesting when disturbance occurs, be it predatory, environmental, or food shortage in nature. During these events, the birds in nests that fail have made the decision to abandon the chance of contributing offspring to the next generation in exchange for protecting themselves so they can breed again in the future (Groscolas, Lacroix, & Robin, 2008). A higher rate of failure at the incubation stage may be due the stronger stimulus to remain on the nest that is presented by a chick as opposed to an egg; thus, a bird would be less likely give up a nest in the guard or post-guard stage (Groscolas et al.).

Chiaradia & Kerry (1999) showed that Little Blue Penguins were more likely to be successful breeders when they started breeding earlier, and took more foraging trips and swapped over parental care more often. This may indicate that the need to forage for food is an important factor in the abandonment of nests. Bull (2000) investigated factors that influence egg success (identical to the incubation stage in this study) and found that of the factors measured (nest type, clutch size, date and year of laying), only nest type showed a relationship with the fate of an egg. Bull did not, however, test predator abundance or predator access across several sites. Knight & Rogers (2004) conducted a similar study in which they investigated factors influencing fledgling production rather than egg success. Of the factors examined (year, date of laying, years since banding and habitat), it was found that the date of laying provided the strongest indicator of fledgling numbers, while habitat and year also appeared related. However, they did not investigate the relationship between fledgling numbers and predator abundance or access, either.

Perriman, Houston, Steen, & Johannesen, (2000), looked at the relationship between climatic fluctuations and nest success, while Robinson, Chiaradia, & Hindell, (2005) investigated the relationship between body condition and nest success. The length of foraging trips appears to also influence egg success, as longer foraging trips (usually brought on by poor body condition), were found to result in the fasting bird who was in charge of incubation needing to leave the nest before its partner returned (Numata, Davis, & Renner, 2000). Although studies have tried to identify a single factor or combination of factors influencing nesting success, it is difficult to establish finite confidence in them, as a large number of variables and interconnected relationships comes in to play.

During the current study, penguins would occasionally make a second attempt to nest in a location that had been previously categorized as failed. Typically, when this occurred, it could not be determined whether the same individuals were laying a second clutch or new individuals were laying their first clutch at that nest, because the location and situation of many of the nests did not allow for the removal of the birds for banding. It can be noted from Table (4.1) that second nesting attempts in the same nest site were more common at Goat Island. This may have been because the higher predation pressure caused a higher rate of nesting failure, leading to a greater number of unused ready-made nests available for re-nesting attempts. It is possible that this increased number of re-nesting attempts at Goat Island was a compensation measure, with the birds at that location encountering higher costs for maintaining a greater reproductive output than the birds nesting at the two Tawharanui sites. Johannesen, Houston and Russell (2003) pointed out that Little Blue Penguins are unique among penguins in being able to produce fledglings from two separate clutches in the same breeding season. (Johannesen et al.) also showed that there was an increase in survival and breeding performance if a pair had laid a double clutch the previous year.

5.1 Issues

Knadle, Brubaker, & Brubaker (2001) suggested that nest success is a poor measure for comparing productivity between areas or populations. They postulated that nest success is poorly distinguished from annual reproductive success and that authors frequently do not address the influences of re-nesting or multiple brooders. Murray (2000) showed that differences in egg or nest success do not necessarily result in a difference in annual reproductive success, as these are only two of many relevant factors. Murray further commented that determining reproductive success on a measure of egg or nest success could be misleading. If nest success differed between two populations whose annual reproductive success was the same, the researcher should endeavor to find the balancing factor. Even Mayfield himself (Mayfield,1975) wrote that, ideally, every nest should be found and observed and the fate of each fledgling determined, in order to make research complete.

The Mayfield method was chosen to analyse data on nest success due to its ease of use and relative accuracy. The 40% method as described by Miller & Johnson, (1978) was used where appropriate, and the piecewise application as described by Klett & Johnson (1982) was used to assess the differing success of each of the three nesting stages that were under observation in this study. It should also be noted that, as Stanley (2000) pointed out, the use of a piecewise application of the Mayfield method is inappropriate when the transition between stages occurs between visits. Therefore, the stage-specific data in this study should only be used as a relative comparison between sites, and confidence should not be given to the actual values of success for each of the individual stages.

Hensler & Nichols (1981) recommended that sample sizes of at least 20 nests be used for calculating nest success using the Mayfield method. None of the three sites included in this study had a sample size greater than 20; therefore, it is possible that the nest success values given are not 100% accurate. However, they do provide a relative measure for comparing the differences between the three sites.

Finally, the length of time between observational visits could have affected the accuracy of the data collected (citation here). Maintaining a shorter time period between visits might have resulted in an increased ability to achieve truly representative data.

5.2 Implications and Recommendations

Previous studies have shown that habitat or nest type play a part in parameters of nest success (Bull, 2000; Knight & Rogers, 2004). It is therefore recommended that suitable habitat be made more readily available at the three sites, given that nest success rates were unexpectedly low throughout this research timeline. Bull suggested that creeping, low-lying scrub such as *Muehlenbeckia complexa* provides an ideal site for nesting and it would thus make sense to replant such vegetation at the penguin habitat sites. As described in Chapter 3, low scrub did not appear to be the preferred habitat for nesting, judging by the data that was collected (although very little *Muehlenbeckia* was present). Therefore, it is also recommended that secure and suitable nesting boxes, resembling boulders, be placed throughout the three sites.

It is likely (although not fully validated by this study) that predation is also affecting nest success at Goat Island. It is therefore recommended that some form of ongoing predator control take place at Goat Island.

6 Summary

Nest success was expected to be greater at the two Tawharanui sites (North and South) than at the Goat Island site. It was also expected that nest success at the two Tawharanui sites would be relatively similar. Both of these hypotheses held true. These hypotheses were based on levels of predator access to the sites; however, previous studies have shown that habitat and nest type influence nest success. This study has not definitively shown that predator access influences nest success in terms of the number of chicks produced. However, the population at the Goat Island site appeared to be compensating for a higher nest failure rate that was postulated to be related to predation, with more re-nesting attempts. Therefore, birds at the Goat Island site incurred higher costs to reach a similar, if not somewhat reduced, reproductive output when compared to the two Tawharanui sites. Given the findings reported in the previous chapter which revealed Goat Island's higher incidence of preferred nesting habitat, it makes more intuitive sense that predator access was the key factor, affecting nesting success to a greater degree than habitat or nest type. Overall, however, there were many factors affecting nest success, and it is likely a combination of these produced the observed nest success rates.

There are many issues and debates concerning the best and most accurate ways of measuring nest success, including whether or not nest success should be measured at all (the alternative being survival rates of the young). Although the use of a piecewise application in this study could be criticised (as the length of time between visits does not allow for the actual date of transition between stages to be known), it has been stated that these results should not be taken as absolute values, but rather be used as relative values to make comparisons between the three sites.

This chapter has offered insight into the differences and similarities between nesting success (or failure) rates at three North Auckland Little Blue Penguin nesting sites. The following chapter will investigate the causes and rates of mortality at these same sites.

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CHAPTER 5 - CAUSES OF MORTALITY AND RATES OF CARCASS RECOVERY IN LITTLE BLUE PENGUINS (*EUDYPTULA MINOR*) FOR THREE NORTH AUCKLAND SITES

1 Abstract

This study investigated the mortality of Little Blue Penguins (*Eudyptula minor*) by counting the number of carcasses found on the coast at three Auckland sites and establishing various causes of mortality through necropsies of these carcasses. The investigation was carried out at Tawharanui North, Tawharanui South and Goat Island over the period of one year. These three sites located north of Auckland were known to be inhabited by Little Blue Penguins. Once a month for the year of 2006, a 2.5km section of each site was searched in an attempt to recover carcasses of Little Blue Penguins. The number of carcasses recovered was recorded as follows: Goat Island, 0.67 birds/km; Tawharanui North, 0.63 birds/km; and Tawharanui South, 0.43 birds/km. These rates are lower than historic rates for this area. There were two peaks in the number of carcasses found during the year, one in June and another in September

2 Introduction

The viability of a population is affected by the birth and the death rate for that location (demographic factors), as well as other issues. Further, in small populations, the survival and reproductive capacities of each individual have a large impact on the population as a whole. The population dynamics of a small population are more uncertain than those of a larger one; this is simply a result of the law of averages (Caughley, 1994). As well as demographic factors, population dynamics are also affected by environmental and spatial factors. Because of the small size of the population involved in this study, its viability is uncertain and factors such as mortality have had a large impact upon the reported results.

Harrigan (1992) found that the most common causes of death in Little Blue Penguins were predation and road trauma. In addition, juveniles were often found with heavy parasite loads which could have either caused the death or contributed to it as a compounding factor. Starvation was found to be a common cause of death in adult penguins. Obendorf and McColl (1980a) suggested that increased mortality could be attributed to the compounding of starvation with high parasite loads. Dann, Norman, Cullen, Neira, and Chiaradia (2000) found after performing 29 necropsies that 89% of deaths were caused by starvation associated with the presence of gastro-intestinal parasites. Dann et al. investigated the raised death rate among Little Blue Penguins during a high mortality of pilchard which may have been caused by a virus (Whittington, Jones, Hine, & Hyatt, 1997). Most interestingly, only the larger pilchards were affected by the virus (Fletcher, Jones, Pearce, & Hosja, 1997). These larger pilchards were not thought to be heavily predated upon by Little Blue penguins (Cullen, Montague, & Hull, 1992), and Dann et al. suggested that penguin mortality at that time rose out of increased inter-specific competition for the smaller pilchards—those that did not have the potential to be affected by the virus. What remains unclear is why Little Penguins were more heavily affected during that period of time than other birds and fish that also fed upon pilchard. Identifying a negative consequence that is the direct result of competition between penguins and human fisheries is difficult, but there is clear evidence that fisheries have an impact on penguin mortality through by-catch (D. Houston, personal communication, July 14, 2006).

On the mainland and some offshore islands, predation is a very real factor contributing to penguin mortality. Mustilids and dogs are known to prey upon penguins, and in Australia, foxes are also a problem. Phillip Island currently has a fox control program in place (A. Chiardia, personal communication, September 18, 2006). To date, the literature has not reported any evidence of rat predation on either eggs or chicks of Little Blue Penguins.

In addition to predators, disease has contributed to mortality rates. For example, the respiratory disease aspergillosis has been known to cause death in penguins (Hocken, 2000). Other documented causes of mortality include the effects of environmental pollution, such as oil spills, and discarded plastic and nylon waste. Lead poisoning was found to cause the death of at least one penguin which had ingested a piece of fishing sinker made out of lead (Harrigan, 1992). Also, pulmonary oedema caused by asphyxiation from choking on a fish was the documented cause of death of another bird (Dann et al., 2000).

Little Blue Penguins are known for large scale “die-offs”. Such die-offs occur every three to four years, with larger peaks also occurring cyclically every 10 to 11 years (J. Geurts, personal communication, January 2009). However, the die-offs that occur periodically in the Northern and Auckland areas do not seem to occur in the Otago region (D. Houston, personal communication, July 14, 2006). There are many possible causes for these die-offs, including storm events and a lack of food species, and more research is needed in this area.

Assessing which factors are most significant in supporting health and growth in a population is important when selecting appropriate conservation regimes. These factors differ spatially and temporally and it is therefore intuitive to postulate that death rates also differ spatially and temporally. Thus, investigating the differences in birth and death rates between sites under different types of conservation management could make an interesting study. Dann and Cullen (1990) estimated first year mortality in Little Blue Penguins at approximately 50% and adult mortality at approximately 25%. Previous studies have investigated the causes of mortality in little penguins in Otago (Hocken, 2000) and Australia (Harrigan, 1992; Obendorf & McColl, 1980a); however, no such literature exists on the northern Little Blue Penguin.

This study investigated the causes of mortality for the northern Little Blue Penguins and also explored how these causes differ between sites, including comparisons of mortality rates and the number of carcasses recorded at each of the three sites.

This study aims to test the following hypotheses:

1. There will be a trend demonstrating the affect of starvation compounded by heavy parasite load as the leading causes of death.

This hypothesis was based on the findings of Obendorf and McColl (1980b) which showed that 77% of all Little Blue Penguins necropsied were in poor body condition with moderate to heavy parasite burdens.

2. The level of mortality caused by predation will be higher at the Goat Island site than it is at either of the Tawharanui sites.

This hypothesis was based on the fact that there is no predator control at Goat Island, meaning predators have free access to penguins and other wildlife there. The Tawharanui sites have extensive predator control measures in place, including a predator exclusion fence and trapping regimes.

3. There will be an increased rate of mortality (shown through an increased rate of carcasses found) from January through to March.

Kinsky (1960) and Jones (1978) both reported that the highest rates of penguin carcasses found on the beach occurred from January to March, a time when both adults and fledglings may be struggling to obtain food resources.

In testing these hypotheses it is hoped that a better understanding of the causes of mortality in Little Blue Penguins on the east coast of Auckland will be achieved. Perhaps the current findings will contribute to furthering knowledge about the ways in which different conservation management regimes affect mortality rates either positively or negatively and thus help identify which management regimes are more effective in reducing mortality.

3 Methodology

3.1 Data collection

3.1.1 Field collection of data

Once a month for the year of 2006, each site was explored in an attempt to recover dead Little Blue Penguins. At each site, a 2.5km section of coast was carefully searched. When a Little Blue Penguin carcass was found within the study site area, it was assigned a unique identification number, and the date and time were recorded. A GPS reading was taken of the location to allow for mapping and spatial analysis at a later time. The state of the body was noted, including the condition of the feathers (oily, damaged, weathered, etc.), the condition of the skeleton (whole, weathered, bones missing or broken, etc.) and the presence or absence of maggots. Also, a description of the physical location from which the carcass was recovered was recorded as this can prove useful in determining a cause of death.

If the bird was "fresh" with no visible maggots in the mouth, an initial examination was conducted at the site of recovery, including an assessment of the condition of the eyes as a means of estimating the length of time between death and carcass recovery. The following categories of eye condition were recorded: fresh and bright, dull, slightly sunken, very sunken, or absent. The ears were also examined for the presence or absence of ecto-parasites, and the condition of the plumage was noted as to whether it was wet, dirty, sandy, dull, bright, pre-moult, post-moult or balding. The mouth was opened and the colour of the gape ranked between 1 and 5, with white (anemic) ranked as 1 and red (healthy) ranked as 5. Weight and standard external morphometric measurements were taken of the tarsus, wing, head width, head length, beak depth, beak length and nose-to-tip (Jones, 1978). Finally, any external wound or bleeding was noted and recorded.

After completion of the initial examination, the carcass was placed in an airtight bag to prevent it from drying out and minimize any distortion of tissue (Hocken, 2002). It was then frozen at the first opportunity, pending necropsy.

3.1.2 Necropsy

The method of necropsy examination utilised in this research was adapted from post-mortem examination of penguins (Hocken, 2002, Geurts, 2006).

To begin the necropsy, the bird was placed ventral side up and the sagittal ventral feathers soaked with water and soap to help hold them down so as not to obstruct vision. A shallow incision was made through the skin from the vent to chin. The skin was then pulled away from the body, exposing the pectoralis major muscle and the abdominal viscera. At this stage, fat stores were ranked from 1 to 5, with 1 indicating no fat stores were present and 5 indicating the animal was well covered with fat. Fat stores can best be located subcutaneously or in the abdomen. The pectoral muscles (pectoralis major, pectoralis minor and pectoralis brachialis) were also scored from 1 to 5, with 1 signifying severe wasting of the pectoral muscles resulting in a concave shape, and 5 indicating the bird was well built, with the pectoral muscle rising from the keel (ventral process of the sternum) in a convex manner.

The ribcage and pectoral muscles were then removed by cutting through the ribs with scissors and the furcula (fused clavicals) with a pair of bone cutters. Removing the ribcage exposed most of the internal organs.

At this point the absence or presence and location of maggots and fly eggs was noted and the amount of autolytic change ranked from 1 to 5, with a score of 1 indicating no change, and a score of 5 signifying the most autolytic distortion possible in the context of necropsy. The liver was then removed and examined inside and out for any abnormalities (see below), and the same was carried out for the lungs and heart.

The trachea was then opened to look for any abnormalities or blockages, and the digestive tract was removed from the top of the oesophagus to the anus. It was noted that removal of the stomach required that it first be lifted and the mesentery beneath it cut. Following this, the oesophagus was opened and examined, followed by the stomach. The inside of the stomach was rinsed and further examined for lesions (possibly caused by intestinal parasites) and abnormalities. The upper (cranial) portion of the stomach was expected to be glandular, with the lower portion being smooth (Hocken, 2002). If intestinal parasites were present, samples were stored in 10% formalin saline solution for identification at a later stage.

With removal of the digestive tract, the kidneys (located on either side of the spine) could be examined and the sex determined from the genitals (a feather sample was taken and labeled if sex could not be determined visually). Females possessed a single ovary on the left side. A thin-walled and relatively straight oviduct (not convoluted and without thickening of the wall) suggested the bird had never bred before and therefore was likely to be immature (Hocken, 2002).

The ovary of an immature bird is characterised by a fine, uniform texture and a yellow colour, while the ovaries of mature females differ with age and season (Hocken, 2002). Males posses two testes, the left of which is always larger than the right. In mature males they are grey and easily located but the testes of immature males are black in colour and are less obvious; they may be nothing more than flat structures on the posterior abdominal wall (Hocken, 2002).

Finally, the back of the skull was removed using a pair of scissors and the brain examined for any signs of trauma.

3.1.3 Causes of mortality and interpreting findings

A penguin that is in a healthy nutritional state should have subcutaneous and abdominal fat stores and a well-defined, convex set of pectoral muscles (Hocken, 2002). Wasting muscle mass indicates a degree of protein malnutrition, and if found in conjunction with an absence of fat stores, can suggest a long period of starvation. However, muscle wasting combined with the presence of fat stores may indicate a short-term high demand for protein, possibly in response to severe infection. Poor nutrition will weaken a bird and reduce its ability to effectively cope with risk factors, ultimately increasing its vulnerability and the likelihood of death. For starvation to be considered as the proximate cause of death there must be signs of bleeding into the stomach—specifically, the presence of dark red or black material. Lesions within the stomach wall can indicate a high intestinal parasite load and although endoparisitism may contribute to the mortality of a penguin, it is unlikely to be the proximate cause.

The lungs of a healthy penguin should be light red in colour and spongy in texture. Upon opening the lungs, the observation of fluid in them indicates that drowning may have been the cause of death. The lungs can also hold evidence of aspergillosis, a disease that is sometimes stress-induced with indicators that are easily recognisable in wild penguins on postmortem. There are three common signs of this respiratory disease, the first being the presence of granulomatous tumours (Hocken, 2002). These are observed as white masses arising from the lungs or air sacs. On excision and dissection of the tumour, its cross section shows concentric lines. Such tumours result from a chronic form of aspergillosis and their appearance usually coincides with evidence of malnutrition (Hocken). There are two other signs of the disease: the first is white plaque appearing as a mould on the lining of the air sacs in the lungs, and the second is seed-sized nodules scattered across the surface of the air sacs and lung tissue, indicating a rapidly acting form of the disease.

The heart of a fit and well penguin is dark red and has a smooth and shiny appearance. Trauma to the heart or its associated vessels, or the presence of an internal clot, indicates the cause of death. The liver in a healthy bird is also dark red, while brain tissue should be off-white in colour.

The removal of the neck muscle and internal muscles of the trunk, with the skin remaining relatively intact, is consistent with signs of mustelid predation. Ferrets, stoats and weasels attack with a bite to the back of the neck that penetrates the spine below the skull. Weasels are small, however, and unlikely to be able to predate an adult Little Blue Penguin. Identifying a mustilid predator to species is a challenge, with canine spacing providing the best clues. Canine spacing for ferrets, stoats and weasels are 9-15 mm, 5-10 mm and 3-7 mm respectively. Cats kill by cervical dislocation, or by thoracic trauma resulting in bruised and/or chipped vertebrae and ultimately, asphyxia. Cats usually consume large areas of flesh (mainly the large muscles) and bone (M. Alley, personal communication, September 22, 2007). The canine spacing of a cat is usually between 13 and 20 mm. Predation by dog will usually leave little or no external injury; however, it is likely that the ribs will be crushed and/or internal muscles will appear torn and no longer smooth in appearance (especially the pectoralis major). Bleeding from the mouth is also common as a result of the lungs being crushed. Predation by large fish will usually leave linear knife-like cuts in the skin and feathers. There is no published evidence that suggests rats predate adult or juvenile Little Penguins, or that rats reduce hatching success (D. Houston, personal communication, July 14, 2006).

3.2 Methods of data analysis

Manipulation and graphical analysis of the data was conducted using Microsoft Excel 2000; while statistical analysis was accomplished using Minitab 13. An Anderson-Darling test for normality was performed on the data set of carcass recovery numbers. The test showed that the data was not normally distributed $P<0.005$). The data set was then transformed by a common factor of +1 as it included zero values and then further log transformed. This transformed data set also did not follow a normal distribution ($P<0.005$). A Friedman test was them used as a nonparametric alternative to a two-way ANOVA to test.

4 Results

4.1 Recoveries of dead birds

In all, 52 dead penguins were recovered from the three sites included in this study over the year 2006. There were 20 individuals from Goat Island, 19 individuals from Tawharanui North and 13 individuals from Tawharanui South. The 2.5km section designated at each site was searched once a month, yielding a total of 12 searches per site during the year. From this it can be calculated that the rates of recovery for the year were as follows: Goat Island, 0.67 birds/km; Tawharanui North, 0.63 birds/km; and Tawharanui South, 0.43 birds/km.

There are a number of interesting trends that can be noted from the data displayed in the graph of Figure (5.1). Firstly, there are several large peaks in the graph (June and September for Goat Island, and September for Tawharanui North). Secondly, the number of recoveries was much lower in the months between March and May; and finally, no recoveries were made at Goat Island for the first five months of the year. A Friedman test showed that there was no significant difference in the number of recoveries with respect to site: *S*(df=*2*) = 0.54, P>0.05 (adjusted for ties) or, significant difference in the number of recoveries with respect to season: *S*(df=*11*) = 18.76, P>0.05 (adjusted for ties).

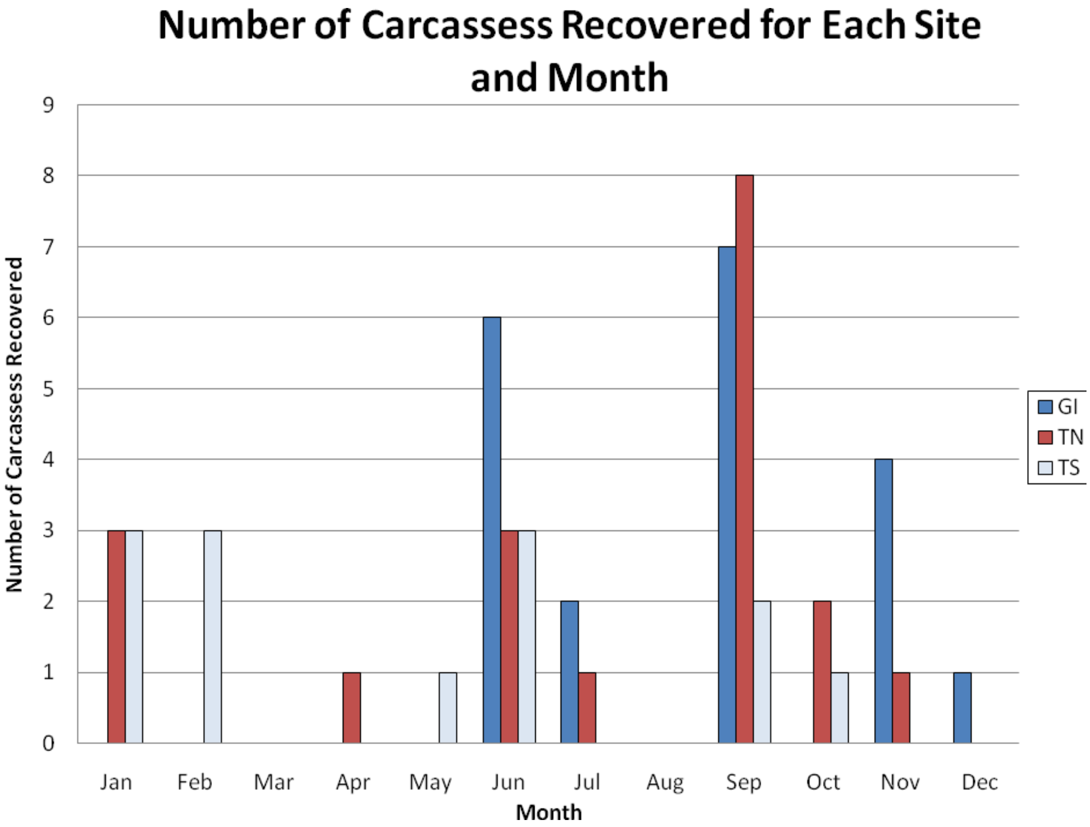

Figure 5.1: Number of Little Blue Penguins found dead for each month at each of the three study sites (Tawharanui North, Tawharanui South and Goat Island mainland) from Jan 06 – Dec 06. TN = Tawharanui North, TS = Tawharanui South, GS = Goat Island.

4.2 Necropsies

Of the 52 birds recovered over the 2006 period, only 5 were in suitable condition for necropsy. The causes of death are outlined in table 5.1. Many of the birds that underwent necropsy were in poor body condition. No internal parasites were found within any of the 5 birds that underwent necropsy.

Table 5.1
Causes of mortality of Little Blue Penguins examined post-mortem

Date Recovered	**Site**	**Cause of Death**	**Fat Score**	**Pectoral Score**
21.2.06	TN	Drowned (compounded through starvation) (inferred)	1	2
24.2.06	TS	Starvation (inferred)	1	2
27.3.06	TN	Drowned (from trauma at sea) (inferred)	3.5	3
14.6.06	TN	Trauma (through being caught in a stoat trap)	1	2
14.6.06	GI	Predated (by mustelid spp)	3	Absent

Table 5.1: Cause of mortality, fat score, pectoral score, site found and date found for each of the five necropsies carried out during 2006. TN = Tawharanui North, TS = Tawharanui South, GS = Goat Island.

4.3 Summary of results

Surprisingly, there were no bird carcasses recovered from Goat Island from January to May, although, generally, more birds were recovered from Goat Island than the other two sites over the months from June to December. Therefore, it would be difficult to say that the rate of recovery was greater at Goat Island, even though Goat Island did have the highest total number of recoveries (20 individuals). Another unexpected result was the finding that the months from January to March did not show the highest rate of carcass recovery. Of those found, only a few individuals were in suitable condition to undergo necropsy; however, these necropsies will provide interesting points of discussion below.

5 Discussion

Simply dissecting a penguin does not provide enough evidence to conclusively identify the cause of death. One must first recognise the anatomical characteristics of a normal, healthy penguin before one can identify a penguin that is not so. Subsequently, one must be able to interpret these differences accurately to identify a probable cause of death. It is important to realize that not every necropsy will yield conclusive results. For example, in one study approximately 16% of necropsies on the Little Blue Penguins from Otago resulted in failure to identify the cause of death. Additionally, many of the necropsies that identified a cause of death cited support for the final decision that was based largely on the situation in which the bird was found (road kill or by-catch, for example) (Hocken, 2000).

Identifying a predator as a cause of death typically involves utilising circumstantial evidence, and in these situations, one should be conservative in applying predation as the proximal cause of mortality. Evidence that appears to point to predation could also have been caused by secondary scavenging after the penguin had already died. Predation is defined in the context of this study to include animal attacks that may not necessarily have been preformed for food (dog attacks, for example). Many of the considerations that must be taken into account when assessing the possibility of predation are outlined below.

Ratz, Moller, and Fletcher (1999) state that there are only two methods for unequivocally identifying a predator: firstly, direct observation (live in field or recorded), and secondly, recovery of the predator’s gut contents. The second of these in and of itself cannot distinguish between predator and scavenger, and therefore, the only completely unequivocal method of identifying a predator is through direct observation.

Another possible method for identifying a predator is the use of canine spacing. It provides the potential for ascertaining predator type, but the method has its limitations. Firstly, in canine measurements there is considerable overlap between species; and secondly, the carcass skin must have been left intact if there is to be any chance of identifying a predator through this method (Ratz et al., 1999). Another assumption made when using the canine spacing method is that only one species has participated in the predation. Secondary scavenging and maggots can make identification of a predator difficult or even impossible (Brown, Moller, & Innes, 1996; Major, 1991; Ratz et al.).

Understanding the unique behavioural characteristics of specific predators can help in species identification. Unlike cats, mustilids are likely to kill animals in very wet sites. Stoats are more abundant in forests, whereas ferrets are largely restricted pastoral habitat and forest margins (King, 1990; Lavers & Clapperton, 1990). North Island brown kiwi (*Apteryx manteli*) over the size of 800 g are reported to be able to defend against stoat attacks (McLennan, Dew, Miles, Gillingham, & Waiwai, 2004); therefore, it can be postulated that adult Little Blue Penguins of a similar size are capable of defending against stoat attacks as well.

5.1 Recovery of Carcasses

The rates of carcass recovery identified in this study were 0.67 birds/km at the Goat Island site, 0.63 birds/km at the Tawharanui North site and 0.43 birds/km at the Tawharanui South site. In comparison, the rate of penguin carcass recovery by the ornithological society on the east coast of Auckland in 1982 was 2.03 birds/km of coast covered, while that for Otago was 0.43 birds/km. Even when a large scale die-off is excluded from the east coast of Auckland statistics, the rate is still the highest recorded for any location during 1982 at 1.97/km. Powlesland (1984) suggests that such a high rate of carcass recovery could reflect a greater density in Little Blue Penguin population numbers. This explanation is possible given that Goat Island had the greatest number of carcass recoveries and also the greatest population density (Chapter 2). In addition, it must be noted that differences in the rates of carcass recovery could be due to a difference in the amount of search effort.

Comparing the range of carcass recovery rates recorded in this study (0.43 – 0.67 birds/km) with those reported by Powlesland (1984), it appears the rates of the current study fit more closely with those reported from Otago in 1982 than with those from the east coast of Auckland.

There were two peaks in the number of carcasses found during the year. The first peak occurred in June at Goat Island, and necropsy of one carcass indicated that the cause of death was predation by mustilid species. It is therefore possible that the peak in the number of carcasses recorded at this site was caused by an increased occurrence of predation. The second peak took place in September at both Goat Island and Tawharanui North. This peak is most likely related to storm conditions observed along the coastline during this period. The southern side of Tawharanui is more sheltered, which may explain the lack of a September peak at this site.

An increased rate of carcass recoveries in September was also reported by Powlesland (1984), however, Kinsky (1960) and Jones (1978) both recorded their highest rates of dead penguin recovery from January to March, a time when both adults and fledglings may be struggling to obtain food resources. It is possible that mortality typically peaks during both these time periods, depending on the severity of the weather and food shortages for that year.

5.2 Necropsies

An early study by Crockett and Kearns (1975) linked the high mortality of northern Little Blue Penguins in 1973 and 1974 to starvation, heavy parasite loads and rough weather. Obendorf and McColl (1980a) also suggested that increased mortality may be linked to starvation and increased parasite loads. Although this pattern may occur in many places, it is expected that types of mortality will differ with geographical location. For example, the presence, absence and abundance of parasites and predators changes widely across different habitats, which in turn vary according to location.

Hocken (2000) suggests there is spatial variation of parasites. From the few necropsies that were conducted during this study, it would appear that, similar to the findings of Crockett and Kearns (1975), starvation and rough weather played a large part in the mortality of Little Blue Penguins during the time period under investigation; however, no internal parasite loads were discovered in any of the birds that underwent necropsy. These findings also suggest that predation plays a significant part in the mortality of Little Blue Penguins on the mainland.

Finally, it should be noted that many of the birds that underwent necropsy in this study had low fat and pectoral scores, which may be indicative that starvation was a compounding factor in mortality.

5.3 Issues

There were simply not enough suitable Little Blue Penguin carcasses available for necropsy to allow for any comparisons between the three sites. Therefore, the original aim—to identify causes of mortality for Little Blue Penguins at both Tawharanui Regional Park and Goat Island, and to investigate any differences in mortality—could not be answered.

No measure of effort (other than kilometers searched) was recorded during this part of the study, so it is impossible to tell if differences in the amount of effort expended during searches at the three sites during different months affected carcass recovery rates. In hindsight, it would have been useful to record the number of hours spent searching and the number of people present to allow this possibility to be tested.

5.4 Implications and Recommendations

Introduced predators pose a serious threat to many of New Zealand's native fauna, and Little Blue Penguins are no exception. They are very susceptible to predation by introduced mammals such as mustelids and dogs (Harrigan, 1992; Perriman & Steen, 2000). Despite this, Little Blue Penguins still persist on the mainland and identification of precisely which introduced species are predating Little Blue Penguins is essential for making effective conservation management decisions (Ratz et al., 1999). It is recommended that Little Blue Penguins be protected from predators on the mainland where possible, and that ongoing carcass recovery observations and necropsies be carried out on penguin populations in Auckland to better understand the causes of mortality.

6 Summary

This study aimed to identify the causes of mortality in Little Blue Penguins at both Tawharanui Regional Park and Goat Island and to investigate differences in mortality and rates of carcass recovery. The study was unable to ascertain any statistically significant differences in the types of mortality between sites due to the small sample sizes. There was no significant difference in the number of carcass recoveries in respect to season. There was also no significant difference in the number of recoveries with respect to the site.

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CHAPTER 6 - QUANTIFYING STRESS – CORTICOSTERONE LEVELS IN LITTLE BLUE PENGUINS (*EUDYPTULA MINOR*) SUBJECT TO IDENTIFICATION MANIPULATIONS

1 Abstract

Metal flipper bands and micro chip (pit tag) implantation are two of the methods commonly used for identification of penguins. This study assessed the acute stress response of Little Blue Penguins (*Eudyptula minor*) to these two methods through measuring corticosterone levels in the blood of the birds. The results showed that implantation of a pit tag produced a significantly greater acute stress response than banding with the traditional metal flipper bands. Interestingly, the control using handling showed a greater stress response than the banding treatment. Although banding appeared to cause less acute stress than pit-tagging, the chronic stress and the long term effect of these two methods need to be investigated before a recommendation on which is a better can be made.

2 Introduction

There have been a few studies into the medium to long term costs incurred by the different types of bands used on penguins (including the trial of a new rubber flipper band) (Hindel, Lea, & Hull, 1996; Jackson & Wilson, 2002; Peterson, Branch, Ainley, Dee Boersma, & Woehler, 2005; Stonehouse, 1999); and a few studies have investigated the stress levels of penguins in relation to factors such as tourism (Cockrem, Potter, & Candy, 2006; Nimon, Schroter, & Stonehouse, 1995). However, no study has looked at the differences in acute stress caused by identification techniques. I feel this study is much needed and could lead to more informed choices about penguin identification techniques.

It is a requirement to obtain the appropriate permits and permission from an ethics committee before conducting research on any vertebrate. Langkilde and Shine (2006), urge all researchers to obtain objective information about the stress they are thinking of inflicting on their study species.

Not only do raised stress levels in study species cause a genuine ethical concern, they can also cause confounding factors that might interfere with answering research questions and add bias to data (Guillette, Cree, & Rooney, 1995; Hindel et al., 1996). According to Hindel et al., a commonly made assumption in ornithological research is that the methods used to mark birds do not interfere with the normal behaviour of the study species. However, this is often unlikely to be the case.

In this study, the corticosterone levels following either the application of a traditional metal flipper band or a microchip (pit tag) were analysed and compared between individual penguins. The study was conducted on Tiritiri Matangi Island (Auckland Haraki Gulf) to allow a large enough sample size to be collected, as the other three sites used throughout this study did not have population sizes adequate for this part of the study.

This study aims to test the following hypotheses:

1. The application of metal flipper bands or microchips will cause a rise in the acute stress levels of Little Blue Penguins, shown by a rise in blood corticosterone levels.

This hypothesis is based on the fact that these identification manipulations are unnatural for the birds and therefore should be perceived as stressful.

2. It is expected that the birds that undergo the application of pit tags will show a higher level of blood corticosterone post-application than that of birds that are banded with traditional metal flipper bands.

This hypothesis is based on the fact that application of a microchip is physically more invasive than the application of a metal flipper band.

2.1 General Endocrinology

Behavioural neuroendocrinology is a field that considers interactions between behaviour and neuroendocrine function in animals (Cockrem, 2005). Such studies answer questions of how behaviours interact with hormones and the reproductive status of an individual, and how external and internal factors affect hormones.

Cockrem and Ishii (1999) describe a new field of endocrinology that has recently emerged and taken on a multidisciplinary approach. They define conservation endocrinology as “endocrine studies that can contribute to conservation”. This new field draws on areas of endocrinology, neuroendocrinology and conservation biology. The most direct contributions from endocrinology to conservation biology occur when research outcomes directly affect management decisions.

With the use of endocrinology, it is possible to identify stressors that may not be detected by behaviour alone (Walker, Boersma, & Wingfield, 2005).

The adrenal gland is comprised of two distinctive regions, the cortex and the medulla. The cortex is responsible for producing steroid hormones, while the medulla produces the sympathetic hormones epinephrine and norepinephrine (Hickman, Roberts, & Larson, 2001). Although penguins are not mammals, they do have the same components of the adrenal gland as mammals have, they are just not arranged in the distinct cortex and mudella regions (Hickman et al.). Corticosterone is a steroid hormone of the glucocorticoid family. Glucocorticoids promote gluconeogenesis, a process which increases the level of glucose in the bloodstream, providing energy for muscles and nervous tissue. The secretions of glucocorticoids are controlled through the hypothalamo-adrenal axis. Adrenocorticotropic hormone (ATCH) of the anterior pituitary gland controls the synthesis and secretion of glucocorticoids, and ATCH is itself controlled by corticotrophin releasing hormone (CRH) of the hypothalamus. A negative feedback loop exists between glucocorticoids and both ATCH and CRH (Hickman et al.).

Glucocorticosteriods, along with other hormones of the hypothalamic-pituitary-adrenal axis, cause behavioural and physiological changes that allow an organism to deal with a disturbance (Sapolsky, Romero, & Munck, 2000; Wingfield & Romero, 2000). Baseline levels of glucocorticosteriods are necessary in animals for them to maintain homeostasis (Dallman et al., 1993). However, chronically elevated levels can induce muscle wasting, depressed immune systems and reduced growth, although chronic stressors are unlikely to occur in nature (Sapolsky et al.; Wingfield & Romero).

Carsia & Harvey (2000) state that corticorsterone is the major adrenocorticoid hormone among birds and the typical corticosterone response to a stressful stimulus is a rise in its levels, typically a few minutes after the stimulus with a peak in approximately 15 – 20 minutes. In free-living wild animals it is common for levels to still be elevated an hour after stress is experienced (Cockrem & Silverin, 2002b). It is well known that glucocorticoids are good indicators of physiological stress (Belliure, Meylan, & Clobert, 2004).

2.2 Identification Techniques

Any animal marking technique has costs, whether these be financial or ethical (Peterson et al., 2005). Part of the job of a researcher is to decide if the benefits adequately outweigh the costs. There are currently six different categories of penguin marking techniques that are used across different species. Peterson et al. identifies these as remote tracking, flipper bands, implanted transponders, dyes, foot webbing tags and photo identification.

Penguins were first banded in 1908 by L.Gain who used tarsus bands. The first metal flipper bands were used nearly 60 years ago by W. Sladen (Stonehouse, 1999). The move from tarsus bands to flipper bands came about because of the difficulty of viewing the tarsus bands in the field (Peterson et al., 2005). As well as bands, both paint and holes punched into the foot webbing have been used as methods to mark penguins (Stonehouse). Some improvements have been made upon the earlier flipper band designs; early flipper bands fitted closely to the flipper and did not allow room for swelling during molting (Stonehouse). Current flipper bands are molded from metal and sit loosely on the auxiliary portion of the flipper (Jackson & Wilson, 2002). Culik, Wilson, and Bannasch, (1993) showed that flipper bands on Adelie Penguins caused a significant increase in power output during swimming. It is likely that the mass of the flipper bands is less important than the streamlining, as loads carried for chicks can reach as high as 30% body mass (Jackson & Wilson). During the present study damage to both the feathers and the bodies of Little Blue Penguins as a clear result of interference from flipper bands has been observed (M. Lowe, Pers. Ob.). Other authors have also noted similar observations in other species of penguins (see also Sallaberry & Valencia, 1985).

Wolfaardt and Nel (2003) and Gauthier-Clerc et al. (2004) reported that the negative effects of flipper banding only occurred in years of food shortage and poor conditions, contrary to the findings of (Peterson et al., 2005). Dugger, Ballard, Ainley and Barton, (2006) suggested that the negative effects of flipper bands were only a factor in the first year or so of application, as after this time the birds learned and adapted to life with a flipper band. Other research has concluded the effects of flipper bands may be cumulative and not simply limited to the first year of application; alternatively, they may only become apparent after several years (Hindel et al., 1996; see also Dugger et al.). It is obvious from these contradictory findings that there is still a large amount of debate surrounding this issue.

Research into the effects of flipper bands on penguins has been carried out upon six different species. These studies have found negative effects on survival (Ainley, 2002; Ainley, Lereche, & Sladen, 1983; Clarke & Kerry, 1998; Dugger et al., 2006), energetic costs (Bannasch, 1995; Culik et al., 1993), and breeding (Froget, Gauthier-Clerc, Lehaho, & Handrich, 1998; Gauthier-Clerc, Gender, Gilly, Le Bohec, & Le Maho, 2001; Gauthier-Clerc et al., 2004). There are also studies which reported no significant negative effects (Boersma & Van Buren, 2004; Hindel et al., 1996; Wolfaardt & Nel, 2003).

These studies illustrate there is no consistent rule concerning the negative effects of bands. The issue seems to be more complex than one might initially think; with no consistancy across species or experimental methods. Confounding factors such as age, sex and body mass make this issue even more difficult to unravel. Additionally, there are different band designs, the effects of which have been investigated using different experimental and analytical methods (Dugger et al., 2006). Perhaps as Dugger et al., suggest the true issue of concern is not whether flipper bands negatively affect penguins, but instead, the identification of those mechanisms by which flipper bands cause negative effects. This path of investigation will help aid the development of new improved methods of penguin marking.

Microchips (pit tags, transponders) can be inserted subcutaneously and therefore do not add extra hydrodynamic drag in the water (Jackson & Wilson, 2002). However, transponders create a dilemma in that tagged and un-tagged birds are indistinguishable from a distance (Jackson & Wilson). This point is often used as an argument in favor of flipper bands over transponders, since they provide instant visual identification from a distance. From my own experience, I find the flipper bands used for Little Blue Penguins difficult to read, often making it necessary for me to remove a bird from its nest to read the tag. Transponders offer the advantage of being scanned without disturbing the bird by removing it from its nest.

Clarke and Kerry (1998) found that over five seasons, the survival rate of Adelie Penguins carrying transponders was equal to or better than that of birds with flipper bands. However, they suggested that the use of transponders may not be suitable in species with long life expectancies. In fact, they reported that a removed transponder had developed a bio-film containing potentially pathogenic bacteria. This finding points to the fact that if transponders are not inserted correctly they can have negative consequences on the study animal.

3 Methods

The two most common methods for assessing hormone levels are the analysis of faecal matter and the more invasive analysis of blood plasma, both of which have advantages and disadvantages. Hormone levels in both sources are affected by a wide range of factors (Khan, Altmann, Ishii, & Yu, 2002; Langkilde & Shine, 2006; Millspaugh & Washburn, 2004; Tempel & Gutierrez, 2004; Woodley & Moore, 2002) and may naturally fluctuate with season (Walker et al., 2005). However, the resolution of plasma steroid measurements is much greater than that of fecal steroid measurements (Cockrem, 2005). Therefore this study will use blood plasma to analyse the steroid level of Little Blue Penguins under various manipulations.

3.1 Data collection

Blood sampling was carried out during the first two weeks of November 2007 on Tiritiri Matangi Island. Care was taken to ensure birds were not molting, as that is a period of time when the birds are too sensitive to be handled.

For consistency and ease, time zero was set as the moment a bird was first captured. This was important in determining when samples were to be taken. However, a bird may begin to experience stress from the moment it senses danger, and therefore effort was made to capture birds as efficiently and quickly as possible. Also, for the sake of consistence, the same person was used throughout the study to time each of the steps. The following times were recorded using a stopwatch:

Capture time: The time from when the bird starts to move away from the person attempting to capture it to having the bird at the sampling station.

Sample time: The time between when the bird was picked up for sampling and when it was placed back in the holding bag.

Band/Handling time: The time required to insert the microchip or apply the metal flipper band. This was also the length of handling time for those birds in the 'handling only' control group (see below).

Birds coming ashore were captured as soon as possible using two hands around the wings and approaching from behind. The bird was then immediately placed into a cloth bag (pillow slip) allowing for easier handling of the bird. Only birds that had not previously been tagged were sampled. There was no way to tell the age or sex of bird was it was coming ashore or being sampled.

The first blood sample was taken immediately from a vein on the dorsal side of the foot. A 27.5 gauge needle (0.4x13mm) was used to pierce the vein and blood was collected using 75ul heparin-coated capillary tubes. To get an accurate baseline measurement of corticosterone levels, the initial blood sample was taken within 2 minutes of the bird being captured. A minimum of two 75ul capillary tubes were needed per sample; however, whenever possible the ideal total of three or four tubes was collected.

The bird was kept secure within the cloth bag during sampling and care was taken not to apply pressure to the ventral chest area. The foot to be sampled was held secure around the ankle while the blood sample was taken. The site on the foot to be sampled was wiped with ethanol using a cotton pad prior to and following the taking of blood. A sterile cotton pad was held on the site until the bleeding stopped following the sampling.

The blood collected within the capillary tubes was blown into 1.5ml heparin coated eppendorf tubes and immediately stored on ice until they could be centrifuged (maximum of 3-4 hours later). The heparin tubes were prepared a few days before field work commenced using a drop of 1,000 IU/ml heparin solution.

Each bird was randomly assigned to one of four treatments: pit tagging (microchip), banding with traditional metal flipper band, control for capture, or control for handling.

1. Pit Tagging Treatment: Following the initial sample, a pit tag (microchip) was applied subcutaneously in the area over the pectoral muscle. The birds were then kept secure in the cloth bag until 15 minutes had elapsed, when a second sample was taken. A further two samples were taken 30 and 60 minutes from when the bird was captured. Two samples were taken from each foot.

2. Traditional Metal Flipper Banding Treatment: The same procedure was followed as for the pit tag treatment; however, instead of inserting a microchip, a traditional metal flipper band was applied using banding pliers and two sets of standard pliers in a method commonly used and affectionately known as the “sushi method”.

3. Control for Capturing: The birds were captured and bled at time zero, then placed into a cloth bag. The birds were then bled again at times 15, 30 and 60 minutes post-capture without additional manipulations, while being returned to the cloth bag between sampling.

4. Control for Handling: The birds were captured and bled at time zero, and then handled in the same manner as the birds in the banding and tagging treatments for a similar length of time. The birds were then placed into a cloth bag to be sampled again at times 15, 30 and 60 minutes post-capture.

A minimum of ten birds for each treatment group were sampled.

If a pit tag was inserted into the bird, then following the final blood sample, a traditional metal band was also applied to allow for easy and quick identification and to make sure the bird was not sampled again. Before releasing any bird from either of the two control groups, a traditional metal flipper band was also applied for the same reasons.

Other data recorded for each bird included weather conditions, tide, time, handlers, body condition (1-5 based on pectoral muscle mass), weight and basic body size measurements (beak length and depth, tarsus length etc). The measurements of the bird were conducted after the final sampling.

The blood samples were centrifuged within 3-4 hours of being taken to separate the plasma, which was then transferred to a fresh eppendorf tube (0.7ml) using a 20ul micro pipette. To make sure each bird was handled in the same manner, the same person preformed the same job for each sampling event whenever possible.

This protocol is a common method for analysing stress responses. The method assumes that a greater expression of corticosterone is relative to a greater perception of stress in the animal (Walker et al., 2005). The methodology has been partly developed and critiqued by John Cockrem, an expert in bird stress hormone levels at Massey University, NZ.

3.3 Data analysis

Manipulation and graphical analysis of the data was conducted using Microsoft excel 2000, while statistical analysis was done using Minitab 13 and SPSS.

Many biological variables do not meet the assumptions of parametric statistical tests. The data sets may not be normally distributed or the variances may not be homogeneous. Using a parametric statistical test (such as an ANOVA) on such data may give a misleading result. In some cases, transforming the data will make it fit the assumptions better. The raw data in this study was not normally distributed, and for this reason a log transformation (Log_{10}) was used in the corticosterone response data; this log data was then used throughout the analysis.

A Levene's test of equality of error variances was used to test for homogeneity of variances within time (Log_{10}). Of the four time groups, none were significant for this test, meaning that the null hypothesis must be accepted. Therefore, the variance within time is homogenous. Similarly, the test for homogeneity within treatment groups showed no significant differences in variances—a Hochberg homogeneous subsets test was used to test for homogeneity of variance within the treatment groups.

Table 6.1

Homogeneity of Variances Within Time (Log_{10})

Time Group (Log_{10})	F	Sig
0	2.78	0.057
15	0.67	0.575
30	0.94	0.432
60	0.46	0.714

Table 6.1: Results the Levene's test of equality of error variances. This test was used to test for homogeneity of variances within time (Log_{10}).

Table 6.2

Homogeneity of Variance Within Treatment Groups

Treatment Group	Sig
Capture	0.48
Handle	0.61
Band	0.30
Tag	0.61

Table 6.2: Results from the Hochberg homogeneous subsets test which was used to test for homogeneity of variance within the treatment groups

An Anderson-Darling test for normality was performed on each of the time and treatment groups. Of the eight groups tested, only three groups significantly deviated from a normal distribution: the zero time group, the band treatment and the tag treatment. Given that the majority of groups could be considered normally distributed, it was accepted that the log-transferred data set as a whole was normally distributed.

Table 6.3
Normal Distribution in Groups by Time and Treatment

Time Group	Sig	Treatment Group	Sig
0	0.000*	Capture	0.134
15	0.077	Handle	0.073
30	0.588	Band	0.005*
60	0.127	Tag	0.000*

Table 6.3: The table above shows the results from the Anderson-Darling test for normality. (*) denotes a significant difference.

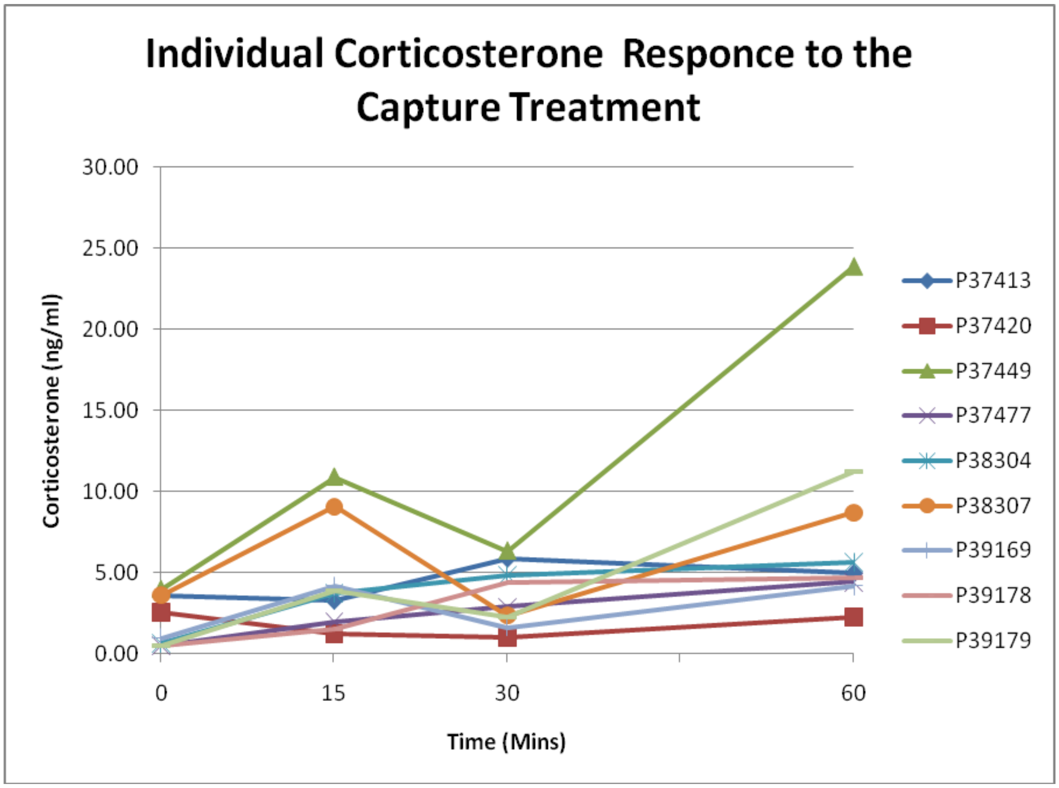
As both the tests for normality and homogeneity were met, it was possible to use ANOVA to calculate any significance between differences in the data set. However, to increase confidence in the results, Mauchly's test of sphericity was performed. This test is similar to a normality test, but considers both the factor (treatment) and repeated measure (time). The test returned an epsilon value greater than 0.75, indicating that the Huynh-Feldt correction must be used on the ANOVA.

4 Results

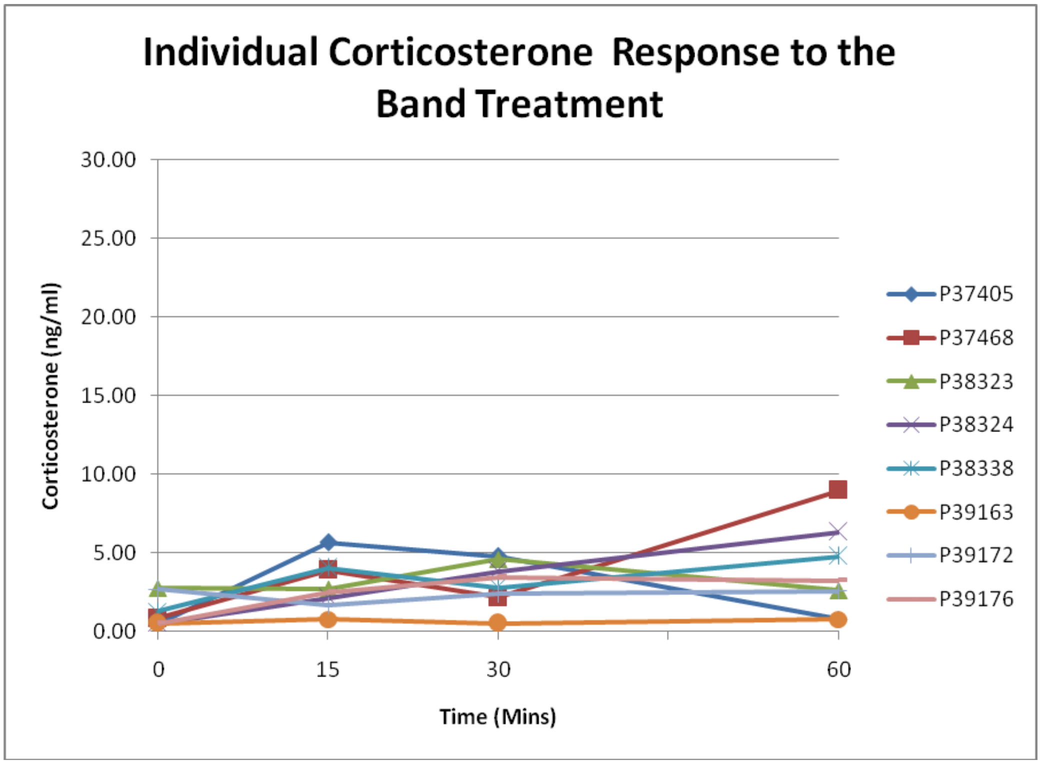
Forty birds were sampled over the two weeks of fieldwork, giving a total of 160 samples.

Figure 6.1 shows the individual corticosterone responses for each of the subjects sampled. Subjects with one or more samples missing have been excluded from this analysis.

A

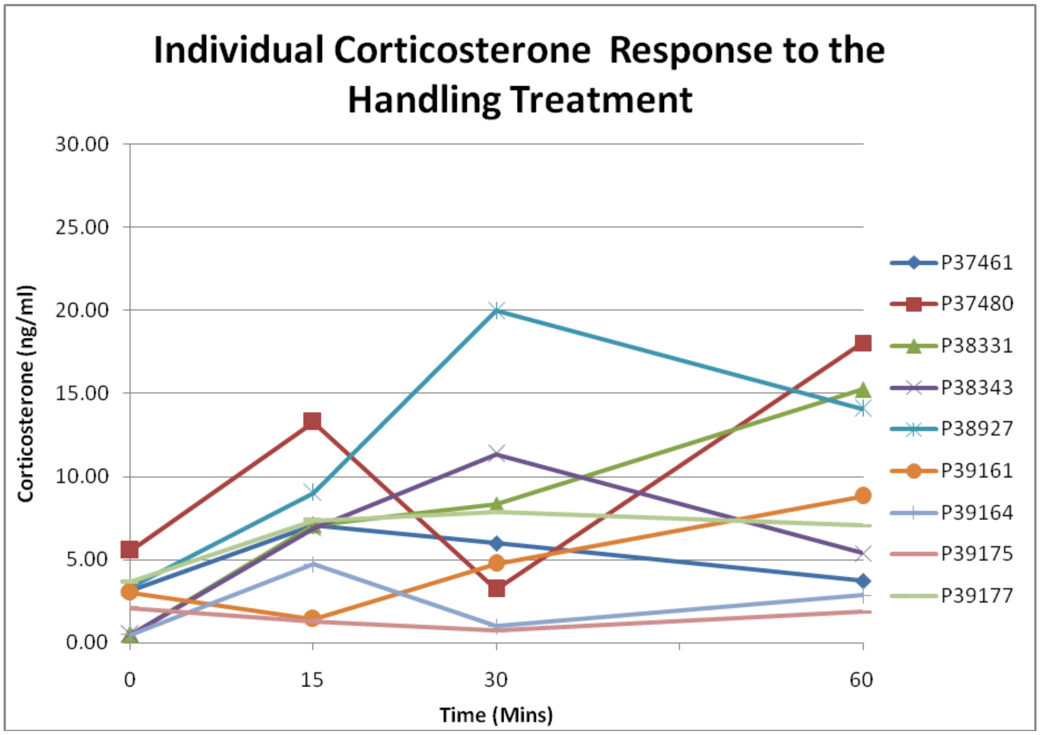


B



Individual Corticosterone Response to the Band Treatment
Corticosterone (ng/ml)
30.00
25.00
20.00
15.00
10.00
5.00
0.00
0
15
30
60
Time (Mins)
P37405
P37468
P38323
P38324
P38338
P39163
P39172
P39176

C



Individual Corticosterone Response to the Handling Treatment
Corticosterone (ng/ml)
30.00
25.00
20.00
15.00
10.00
5.00
0.00
0
15
30
60
Time (Mins)
P37461
P37480
P38331
P38343
P38927
P39161
P39164
P39175
P39177

D

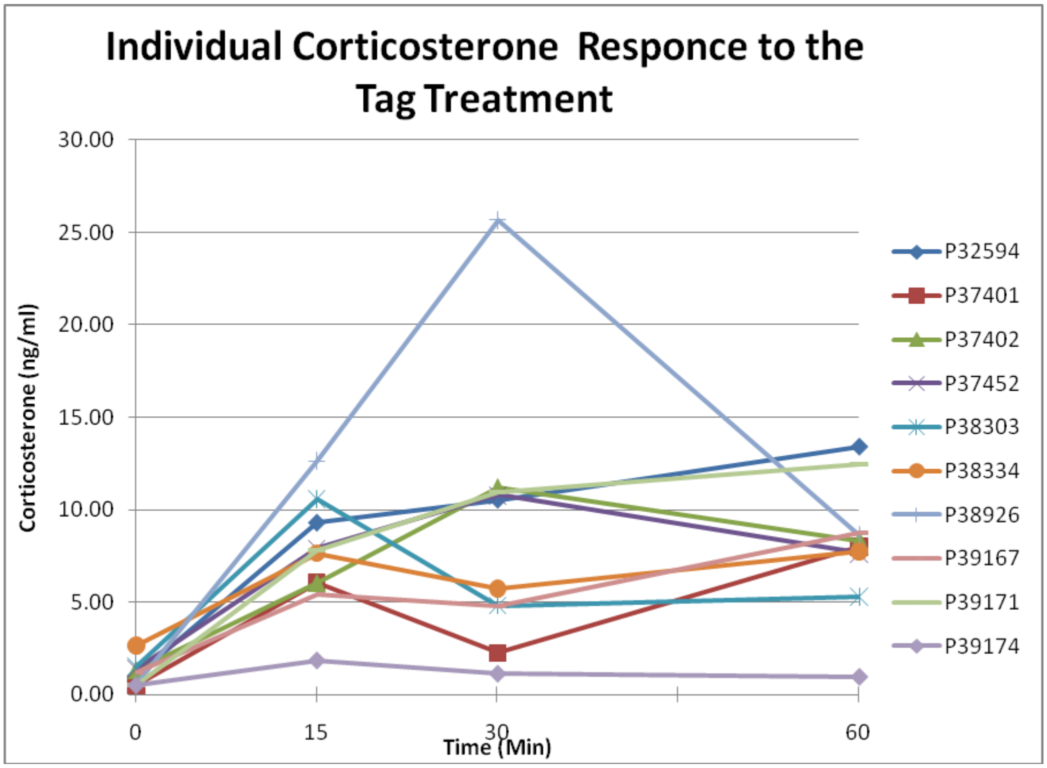


Figure 6.1: Individual corticosterone responses for each of the subjects sampled for each of the four treatments (a) Capture, (b) Banding, (c) Handling, (d) Tagging. Individuals which had one or more samples missing have been excluded from the data set and analysis. P37402 (Tag), P39178 (Capture), P388331 (Handle) were all noted as being extremely calm during the treatment. P38338 (Band) was noted as having diarrhoea throughout the treatment. P38324 (Band) was noted as making vocal calls throughout the treatment. P39161 (Handle) was noted as an extremely aggressive and combative individual for the duration of the treatment.

Figurew 6.2 shows the mean corticosterone response for each of the four treatments. Graphically, it can be seen that the pit tag treatment resulted in the greatest corticosterone response, followed by the handling treatment, the capture treatment and finally, the band treatment. It also appears that the corticosterone response for the tag and handling treatments were similar, whereas the capture and band treatments showed differing corticosterone responses.

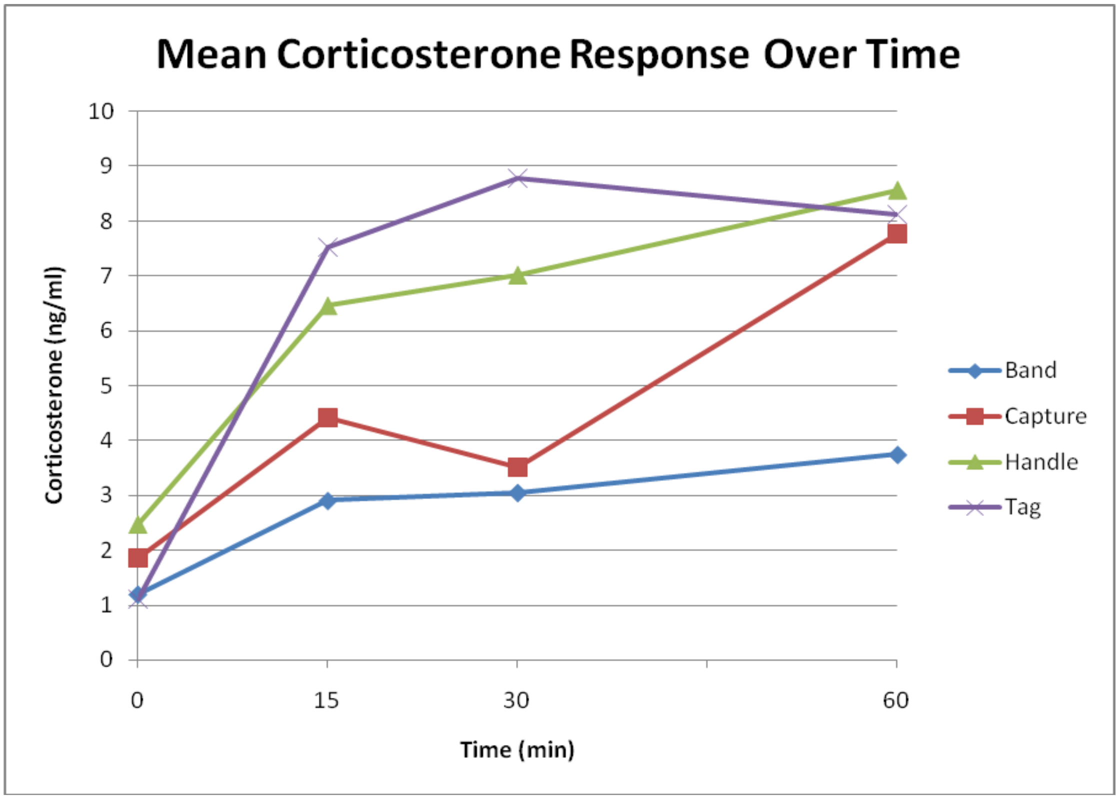


Figure 6.2: Mean corticosterone response over time for each of the four treatment groups. Again, individuals which had one or more samples missing have been excluded from the data set and analysis.

A one-way ANOVA with repeated measures was performed on the data set, with time being the repeated measure. Using the Huynh-Feldt correction, the ANOVA yielded a significant difference in corticosterone response with respect to time; $F(2.75)=41.291$, $P<0.05$ and also a significant difference in corticosterone response with respect to treatment $F(3.0)=2.99$ $P<0.05$. Furthermore there was no significant interaction between time and treatment $F(8.26)=1.59$, $P>0.05$.

Table 6.4

One-way ANOVA With Time as Repeated Measure

	df	F	Sig
Time	2.753	3.308	0.000*
Treatment	3.0	2.989	0.045*
Time x Treatment	8.259	0.123	0.154

Table 6.4: The results of the one-way ANOVA with repeated measures for time. (*) denotes a significant difference.

Pairwise comparisons were performed on each of the treatment comparisons (Table 6.5).

Table 6.5

Pairwise Comparisons Between Treatment Groups

	Capture	Handle	Band	Tag
Capture	/	0.264	0.154	0.247
Handle	/	/	0.015*	0.990
Band	/	/	/	0.013*
Tag	/	/	/	/

Table 6.5: The results of the pairwise comparisons between treatment groups. (*) denotes a significant difference.

From Table 6.5 it can be seen that the only significant differences between the total average corticosterone responses of the four treatment groups was (a) between the tag and the band treatments and (b) between the handle and the band treatments. There was no significant difference between the tag and handle treatments or between the capture treatment and any of the other treatments.

A test of correlation was performed in order to ascertain whether or not the amount of time taken for capture (time from bird sighting its capturer, to when the initial blood sample was taken) or treatment (time taken to perform the treatment) affected the corticosterone response for the subject. A correlation between capture time and log corticosterone levels at time zero was performed, as was a correlation between treatment time and log corticosterone levels at time 15. The correlation of capture time (M = 198.78, SD = 79.88, N = 36) and corticosterone response at time zero (M = 0.074, SD = 0.36, N = 36) was not significant ($P>0.05$). Furthermore, the correlation of treatment time (M = 148.97, SD = 92.79, N = 36) and corticosterone response at time 15 (M = 0.63, SD = 0.33, N = 36) was also not significant ($P>0.05$). This indicates that the differences in capture and treatment times did not affect the level of corticosterone response.

As there were a number of haemolysed samples and relatively few samples in total, it would have been unfeasible to exclude the haemolysed samples from the results. However, haemolysis can cause reduced amounts of corticosterone to be detected in a sample (J. Cockrem, personal communication, June 20, 2008). Therefore, it is possible that the distribution of the haemolysed samples could have affected the relationship between the corticosterone responses detected in the four treatment groups. The distribution of haemolysed samples amongst the other samples was graphed and as it appeared evenly spread across each of the four treatments, the possible effects of haemolysed samples can be ignored. This is a commonly adopted approach in corticosterone studies (J. Cockrem, personal communication, August 12, 2008).

5 Discussion

To reiterate, it was expected that the birds that underwent the application of pit tags would show a higher level of corticosterone post-application than the birds that were banded with traditional metal flipper bands. This hypothesis held true and was exactly what was discovered in this study. The control groups (for both capture and handling) have also showed a high level of corticosterone response. The group controlling for capture showed a similar corticosterone response to that for banding treatment, which indicates that banding was not significantly more stressful than capture itself. Surprisingly, the corticosterone response of the group controlling for handling was significantly higher than the group subjected to banding treatment. It is possible that the handling was more stressful than the banding treatment, due to the fact that the handling of the birds in the control group involved more handling movements than that applied to the individuals subjected to banding treatment. For example, the birds who received banding treatment were held relatively still while the band was fitted around their wings, while the birds for handling control were handled and moved for the whole time period (similar in length to the time taken to apply a metal flipper band or insert a pit tag).

Silverin (1998) suggested that birds may view capture by people as a predation event. This is understandable, and raises an ethical question about the level of stress researchers induce upon their study animals. A study by Cockrem and Silverin (2002a) on corticosterone response in the Great Tit showed that a response was dependent upon whether or not the bird perceived the stimulus to be a threat. The study also offered evidence to confirm the value of using corticosterone measurements in assessing whether or not birds perceive a situation or stimulus to be stressful.

The variation in mean corticosterone levels for each of the four treatments at time zero was very small, which indicates that the baseline samples (zero time blood samples) were accurate and thus increases confidence in the results. Cockrem and Silverin (2002b) found that the individual variation in corticosterone levels in their initial blood sampling was greater than that after 10 minutes. However, this was not the case in the current study. Results reported by Cockrem & Silverin also suggested there was possible habituation to the bleeding process, as the corticosterone response became weaker by the third time the birds were bled. This does not appear to be the case in the current study.

It has been noted that a typical corticosterone response to a stressful stimulus is a rise in levels a few minutes after stimulus with a peak in corticosterone levels after approximately 15 – 20 minutes. Additionally, it is common for levels to be still elevated after an hour in free-living wild animals (Cockrem & Silverin, 2002b). The results reported above demonstrate that corticosterone levels for this study did not peak after 15 – 20 minutes; in fact, the corticosterone levels appear to have still not peaked after 60 minutes. The reason for this disparity with previous research results is unknown; however, it is possible that very little or no habituation to the bleeding process took place and that each new bleeding event induced a new stress response that added to the previous effects.

Adams, Cockrem, Taylor, Candy, and Bridges (2005) revealed that hand-reared Grey-Faced Petrels experienced a lower corticosterone response to standard blood sampling than parent-reared birds. This was assumed to be a result of habitualisation to handling, but there may be other explanations. Kitaysky, Kitaiskaia, Wingfield, and Piatt (2001) suggested that such responses could be a result of dietary factors.

As mentioned above, time of capture, time of treatment and haemolysed samples did not have a significant effect on the results. However, it is possible that the males and females exhibit different corticosterone responses. There have been many previous studies where this has been the case (see Lance, Grumbles, & Rostal, 2001; Rivier, 2006). However, within the constraints of this study, testing for the sex of the birds was unfeasible and no analysis involving the sex of the birds could be conducted.

Another factor that may have affected the individual corticosterone responses to the stimulus was nest location. Individuals that nest near areas frequently visited by humans may be more habitulised to human presence than those who nest in areas that are less frequented by humans. Boersma (1991) and Williams (1995) both found that human disturbance could reduce the number of Little Blue Penguins present at colonies. This may indicate that Little Blue Penguins perceive humans as a threat. However, many Little Blue Penguins on Tiritiri Matangi do, in fact, nest near walking tracks and areas frequently visited by humans. The penguins tested in this study were unbanded birds taken as they returned from sea; therefore, no information on nest location was collected. Consequently, no analysis could be conducted to ascertain whether or not nest location might have affected individual corticosterone responses.

It should be noted that care must be taken when interpreting endocrine studies because there are so many factors that can influence the results. Even a description of a complete annual endocrine cycle may not provide the complete picture of the species under consideration (Cockrem, 2005). Furthermore, as pointed out by (Langkilde and Shine, 2006), it is difficult to measure stress subjectively or using researcher intuition. During the field work for this study, special notes were made on particularly calm, aggressive or visually stressed birds. It may have been expected that the extremely calm or aggressive birds would show extremes in corticosterone responses. Interestingly, these birds' blood tests did not yield extreme levels (high or low) in corticosterone response. This highlights that subjective and visual interpretation of stress is unreliable, which supports the research performed by (Langkilde and Shine). It was also interesting to see that some individuals showed very little or almost no elevation in corticosterone response to the stimulus. This is a normal and expected result amongst individual variations in corticosterone response, although the reasons for it are not known (J. Cockrem, personal communication, August 12, 2008).

It should also be noted that it is difficult to make generalisations about hormone response across species or investigations (Moore, Thompson, & Marler, 1991). Different methods of stress induction, the time of day, the sex of animals, domestication and housing are all factors that can make generalising across studies difficult.

5.1 Issues

Romero and Romero (2002) concluded that the time between capture and sampling is a critical variable in measuring the stress response of wild birds. The issue arises because plasma corticosterone levels quickly become elevated in response to handling (Wingfield & Romero, 2000). For many bird species it is assumed that sampling within the first 2-3 minutes will provide a reliable baseline (unstressed) corticosterone level (Schoech, Ketterson, & Nolan, 1999). However, this assumes the animal was not already stressed prior to sampling, and that all animals have a consistent level of stress (Romero & Romero). It was not always possible to collect the first blood sample within 2 minutes of the bird sighting the capturer. Firstly, Little Blue Penguins can be difficult to catch as they hop and run across the shore and secondly, some individuals struggle more than others making it hard to find a suitable vein and extract a blood sample. Finally, the birds' extremities were often cold after emerging from the sea, thus reducing blood perfusion and making venous access and blood extraction difficult.

It may be important to determine whether a population has already habituated—or will be able to habituate—to a stressor. Nisbet (2000) states tolerance is a separate concept to habituation, stating that tolerance is the intensity of a disturbance that an individual tolerates before responding, whereas habituation is learning to not respond. It is assumed that none of the birds sampled during data collection showed any habitualisation; in fact, this was the reason for choosing as subjects only those birds that were assumed to have not been handled previously.

5.2 Implications and Recommendations

This study has shown that applying microchips (pit tags) to Little Blue Penguins induces a significantly greater acute corticosterone response than applying traditional metal flipper bands. The implication of this result may be that traditional metal flipper bands should be used for identifying Little Blue Penguins rather than the newer microchip (pit tags) methodology.

However, Nisbet (2000) points out that cause and effect of disturbance on populations should not be assumed until sound control experiments have been carried out. Physiological and behavioural responses to a stressor may not be detrimental or adverse to the fitness of the individual if they do not cause a decrease in survival or reproductive success (Nisbet), such as abandonment of a nest or increased susceptibility to disease. Furthermore, Cockrem (2005) suggests that management decisions should not be made purely on mean characteristics of hormone studies, especially when sample sizes are small, because individual variation is important in the survival of wild populations. Therefore, although this study’s results yielded a statistically significant mean difference between the two methods, there is no reason, without further experiments, to recommend that the traditional metal flipper bands are used as an identification technique over pit tags.

In addition to the comments made by Nisbet (2000) and Cockrem (2005), there are several studies that highlight the long term detrimental effects of metal flipper bands on survival (Ainley, 2002; Ainley et al., 1983; Clarke & Kerry, 1998; Dugger et al., 2006); energy costs (Bannasch, 1995; Culik et al., 1993) and breeding (Froget et al., 1998; Gauthier-Clerc et al., 2001; Gauthier-Clerc et al., 2004). Therefore, although it is more stressful for birds to have pit tags implanted, once they are fitted they may be less irritating and cause fewer problems in the long run, as compared to flipper bands. Further study is necessary before a more robust conclusion can be made.

6 Summary

In this study, corticosterone levels following the application of either a traditional metal flipper band or a pit tag were analysed and compared. It was expected that those birds undergoing the application of pit tags would show a higher level of blood corticosterone post-application than birds banded with traditional metal flipper bands. This hypothesis was found to hold true. The results also indicated that capturing causes levels of stress similar to those of banding, and that continued handling causes higher levels of stress than a normal banding procedure. The length of capture time, treatment time and heamolised samples did not appear to have affected corticosterone levels. Differences in sex were not investigated in this study.

Although identification through the use of pit tags induces a significantly greater acute stress response, researchers should not be too hasty to abandon the use of them in favor of traditional metal flipper tags, as previous research has shown traditional metal flipper bands cause a number of chronic negative effects on areas such as survival, energy costs and breeding. The long term effect of pit tagging needs to be further investigated.

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CHAPTER 7 - SUMMARY

The structure of this thesis has taken the form of a series of short papers, each covering a concise topic in separate chapters. Although each of these chapters finishes with its own conclusion, I offer this chapter as a summary of the entire study, briefly summerising the rationale for the study, the main questions, the results and the recommendations.

This study began as a response to the lack of literature on how Little Blue Penguin populations respond to different levels of conservation management and predator control (Williamson & Wilson, 2001). It was also commented by Powlesland (1984) that it would be interesting to compare the breeding success of a predator-free population with that of a nearby population that predators have access to. This study set out to fill these gaps in the literature and provide recommendations for the conservation management of Little Blue Penguins.

Further into the study, I become interested in the amount of stress research causes study species. This concern arose from observing the stress birds are perceived to undergo during banding, measurements and manipulation, and was augmented by reading on the subject (Langkilde & Shine, 2006). Langkilde and Shine pointed out that it is difficult to measure stress subjectively or by using an intuitive analysis of field observations. Among other findings, Langkilde and Shine revealed that microchip implants in lizards were more stressful than toe clipping as forms of identification. By investigating the acute stress levels associated with the manipulations of different identification techniques, it was hoped the current study could offer more objective recommendations on identification practices.

The main research questions of this study were: (a) would locations that are protected by a greater level of conservation management show a greater density and abundance of Little Blue Penguins? (b) Does the habitat type of a location affect the density and abundance of Little Blue Penguins? (c) Would locations that are protected by a greater level of conservation management have a higher rate of nest success? (d) Are there any differences in the levels or causes of mortality between locations under different levels of conservation management? And finally, (e) What levels of acute stress are associated with different identification manipulations?

Results of the study indicated there was no statistically significant difference in nest density or abundance between the three sites studied. or in season. All three sites had a much lower abundance than the Oamaru Penguin Colony (South Island, New Zealand). Thus, it would appear in this instance that conservation management practices do not play a part in the nest density or abundance of Little Blue Penguins. Alternativly, it is possible that the management practices that have resently begun at Tawharanui Mainland Island have not been in place long enough for the resident population of Little Blue Penguins to show any effects. Why do Little Blue Penguin populations thrive on Tiritiri Matangi Island (Haraki Gulf, Auckland) and Oamaru (South Island, New Zealand) but not at Tawharanui or Goat Island?

Unfortunately, small sample sizes prevented the identification of a statistically validated preference in habitat substrate, and therefore also in nesting habitat. However, active nests were most often found on and around substrate types classified as boulders and large pebbles, and vegetation types categorised as grass, flax, and rock faces. Although sample sizes prevented validation of preferential habitat type, it was shown that the habitat structure at the three different study sites was significantly different.

Nest success was greater at the two Tawharanui sites (North and South) than at the Goat Island site, and these two Tawharanui sites were relatively similar in nesting density. This study has not definitively shown that predator access influences nest success in terms of the number of chicks produced; however, previous studies have shown that predation level influences nest success. It appeared that Little Blue Penguins at the Goat Island site were compensating for a higher nest failure rate, with more re-nesting attempts. Therefore, birds at the Goat Island site incurred higher costs to reach a similar, if not somewhat reduced, reproductive output as compared to the two Tawharanui sites. It is possible that as well as habitat and nest type, predator access was a factor in the nest success results obtained.

Unfortunately, small sample sizes again prevented the study from ascertaining if there was a statistically significant difference in the types of mortality between the sites. A Friedman test showed that there was no significant difference in the number of recoveries with respect to site or season.

Interestingly, nest success was greater at the two Tawharanui sites than the Goat Island site, yet there was no difference in Abundance, nest density or rates of carcass recovery. It is possible that the difference in habitat structure is responsible for the differences in nest success? This hypothesis could not be statistically proven as the numbers of active nests were too low at all three sites. Although the nest success is higher at Tawharanui than Goat Island, the reality is that all of the populations investigated in this study are very small. It is important to investigate why these populations are not fairing as well as other populations (Tiritiri Matangi Island and Oamaru for example).

This study showed that acute stress, identified and measured through blood corticosterone levels, was greater in birds that underwent the application of a pit tag than in birds fitted with a traditional metal flipper band. The study also showed that capturing causes similar levels of stress to banding, and that continued handling causes higher levels of stress than a normal banding procedure. Although identification through the use of pit tags induces a significantly greater acute stress response, researchers should not be too hasty to abandon the use of them in favour of traditional metal flipper tags, as previous research has shown traditional metal flipper bands cause a number of chronic negative effects to areas such as survival, energy costs and breeding.

The key recommendations offered as a result of this study are as follows:

- Re-assess density and abundance measures for the Tawharanui and Goat Island sites in 5 to 10 years time. This should give an indication as to whether or not the predator exclusion fence is having a positive impact on the population density of Little Blue Penguins.
- Research what makes a particular habitat type more preferable than another for Little Blue Penguins, this could provide useful information in nesting box design.
- Place nesting boxes around the Tawharanui coast to increase suitable nesting habitat. This should be included with an ongoing study to investigate if the placing of these nesting boxes increases the abundance or nesting success.
- Carry out ongoing carcass recovery observations and necropsies on penguin populations in Auckland to better understand the causes of mortality.
- Engage in further study regarding both the acute and chronic stress levels induced by different identification techniques.

- Engage in further study into possible alternatives to both pit tags and metal bands for penguin identification, ideally a design that provides quick and easy identification while only inducing very minor acute and no chronic stress.
- What is obvious from this study is that some mainland Little Blue Penguin populations are struggling with very low population densities and nest success; therefore it is important to protect Little Blue Penguins from predators on the mainland where possible. This could include trapping, providing more nesting habitat and in some cases predator exclusion fencing.

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