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**The Feeding and Breeding Ecology of Little Blue
Penguins (*Eudyptula minor*) from Tiritiri Matangi
Island, New Zealand**

A thesis submitted in partial fulfilment of the requirements for the degree of

Master of Science in Conservation Ecology.

Massey University, Auckland.

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Arara te manu whakarongo tipuna, te manu nei a te kororā

Kororā-ā-uta, kororā-ā-tai

Kowhetewhete māi o ngutu

Nau na Tane, naku na Tane

Hui e taiki e

Listen attentively for it is the bird that converses with our ancestors

This bird the blue penguin

The *land* blue penguin, the *sea* blue penguin

Your lips are murmuring to us

You are of Tane and we are of Tane

Bind us together

Let it be so!

(By Laurie Porima and Jacqueline Courts)



Finding the connection between the land and the sea

Abstract

At present the New Zealand populations of Little Blue Penguins (LBP: *Eudyptula minor*) are classified as 'Threatened' and in 'Gradual Decline' by the Department of Conservation. Effective conservation management of the North Island sub-species requires an understanding of the factors affecting their survival and breeding success. There is little information on the breeding ecology of the *E. minor*, especially in the North Island of New Zealand. The overall goal of this study was to establish baseline data on a North Island population of LBP in New Zealand. The aims of this study were to 1) identify population demographics, 2) quantify breeding success and identify abiotic and biotic parameters influencing nesting success, 3) identify feeding ecology based on diet and trophic level assessment, and 4) identify cause of death and underlying patterns associated with mass mortalities of the LBP species. Breeding success was quantified by monitoring the nesting activity of 87 nesting attempts during the 2005/06 breeding season. Nest monitoring also involved identifying risks associated with both the egg and chick stage. Diet analysis involved comparing stomach regurgitation samples and isotope samples of feathers spanning a 120 year period. The cause of death for the mass occurrence of beach wrecked birds found during 2005/06 was established through necropsies and histological tests. The major cause of death was compared to patterns of past beach wreck events that has occurred in New Zealand over a 33 year period, obtained through the Ornithological Society of New Zealand. Where possible, both short- and long-term comparisons were made to establish a sound understanding of the key factors that are influencing breeding success, foraging, and survival.

Results showed that 2005/06 was a poor breeding year which was the result of a large number of nest desertions. Furthermore, analysis of stable isotopes shows that the LBP

have been feeding at low trophic levels over the past 120 years and that 2005 was significantly lower in carbon levels suggesting a low year of marine productivity. The largest cause of death associated with mass beach wrecks was starvation. Analysis of past beach wrecks suggest that during the year LBP are at a greater risk of death after the breeding season, after moult, and during winter which are energetically expensive periods. A more long-term study is required to identify the trends in LBP breeding success and to ascertain the primary reason as to why they are unable to obtain enough food. Seabirds are increasingly being used as biological indicators since they are largely influenced by changes associated with the marine environment. The use of LBP as biological indicators may have limitations depending on the parameters being used. However stable isotope measures may be one of the easiest methods to achieve this and allows for reconstruction of past ecological histories through analysis of historical tissues.

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CHAPTER 1 General overview



Plate 1.1. Adult Little Blue Penguins. Photo by J.Geurts 2005.

1.1 ABSTRACT

At present the New Zealand population of Little Blue Penguins (LBP) (*Eudyptula minor*) is classified as “threatened” and in gradual decline by the Department of Conservation (DoC), New Zealand. Populations are declining on the mainland with subspecies ranging from nationally vulnerable in Australia to gradual decline in New Zealand (Hitchmough in press). Effective conservation management of these subspecies requires an understanding of the factors affecting their survival and breeding success throughout their geographical range. There is little information on the feeding and breeding ecology of the LBP, especially in the northern region of New Zealand. The aims of this study were to: 1) quantify LBP breeding success on a protected northern New Zealand island, 2) identify the most influential parameters of breeding success and adult survival, 3) determine prey types of LBP, and 4) investigate the short-term and long-term patterns of breeding, feeding ecology and survival. This study targeted the LBP population on Tiritiri Matangi Island in New Zealand’s Hauraki Gulf. Breeding parameters were measured for a large number of breeding attempts ($n = 87$). Feeding ecology was based on stomach content samples taken after foraging and isotope analysis from feathers. Findings will provide valuable baseline information for future studies and a better understanding of the low success rate associated with the population studied.

1.2 INTRODUCTION

1.2.1 General Overview

Penguins are classified in the Order Sphenisciformes which evolved from flying birds (Order Procellariiformes) (Marchant and Higgins 1990). There are 17 living species of penguins distributed throughout the Southern Hemisphere, ranging from Antarctica to the Galapagos Islands (Marchant and Higgins 1990). Penguins have adapted to one of the most aquatic lifestyles of all birds while still relying on the land to carry out important behaviours associated with their life-cycle.

Adaptations to both terrestrial and marine environments allow them to utilise the land for breeding, moulting, and roosting, while feeding primarily at sea. However, the alternation between terrestrial and aquatic lifestyles poses many problems. The energetic expenditure and physiological changes that occur when moving between these environments, largely influences penguin behavioural and ecological lifestyles (Jones 1978). For example, energy requirements change depending on the type of locomotion required. Within water the energy expenditure is less for penguins compared to walking on land (Baudinette *et al.* 1986). Physiological processes such as thermoregulation are also affected by the change in medium since water requires thermal insulation to keep warm, while on land this is decreased (Frost *et al.* 1975; Jones unpubl. data 1978). Without effective thermoregulation between the two different medium, an individual can suffer hypothermia or hyperthermia, respectively.

The change between land and sea over the course of the year for penguins, means that behaviours associated with each environment will be traded off and influence each other. The ability of a penguin to forage at sea will impact on behaviours

such as uniting of pair bonds, breeding (courtship, egg laying, incubation, chick rearing), moulting, and individual survival. These land based activities will in turn influence the foraging requirements. Studies of penguins are therefore both interesting and difficult to conduct given the interactions resulting from varying needs and habitat use.

1.2.2 Status of Penguin Species

Declines in many populations of penguin species have been identified and investigated to elucidate causes. Several themes are re-occurring and include habitat loss (e.g. Little Blue Penguin *Eudyptula minor*) (Dann 1992), predation by introduced predators (e.g. Magellanic Penguin *Spheniscus magellanicus*) (Boersma *et al.* 1990; Dann 1992), and prey availability (e.g. Jackass Penguin *Spheniscus demersus* (Crawford and Shelton 1978, 1987) and Rockhopper Penguin *Eudyptes chrysocome* (Cunningham and Moors 1994, Dann 1992). Of the 25 globally threatened species (Table 1.1. IUCN 2006) six belong to the penguin Genus *Eudyptes* (Hilton *et al.* 2006).

The Little Blue Penguin is the smallest of the penguin species and is found along the Southern Australian Coast and around New Zealand (Marchant and Higgins 1990). Classification of this species has been based on morphological analysis and distribution (Kinsky and Falla 1976). Until a genetic based classification is accepted this classification with six sub-species will be used. These sub-species include; *Eudyptula minor novaehollonsiae* (South Australia), *E. m. iredalei* (North Island New Zealand), *E. m. minor* (South Island NZ), *E. m. albosignata* (Banks Peninsula), *E. m. chathamensis* (Chatham Islands). Recent molecular analysis by Banks *et al.* (2002) has suggested that there are two clades of LBP. The clades group the Australian sub-species and the Otago sub-species, with the second clade grouping the North Island, Cook Strait, Chatham

Island, and Banks Peninsula sub-species together. This divergence has been suggested to have occurred 2.38 million years ago and is substantiated by vocalisation and morphological analysis (Banks *et al.* 2002).

Although sub-species are reported for within *Eudyptula* no research has suggested that they are reproductively isolated from each other. This is especially true for the New Zealand coast where potentially there is mixing of populations (Jones 1978). Many common names are used for *E. minor*. In Australia it has been commonly known as ‘Fairy penguin’, and ‘Blue Penguin’. In New Zealand the Maori name is ‘Korora’ but it is also referred to as ‘Little Penguin’ and ‘Little Blue Penguin’. In this thesis for consistency reasons with other North Island research *E. minor* will be known as Little Blue Penguin (LBP). Any reference to the general terms ‘penguin’ or ‘bird’ will mean LBP, unless stated otherwise.

The North Island sub-species of LBP *E. m. iredalei* is currently considered as ‘Threatened’ and in ‘Gradual Decline’ under by the Department of Conservation (Hitchmough 2002) (Table 1.1). However under the international IUCN Red List classification LBP are considered of ‘Low Concern’ (IUCN 2006).

Table 1.1. IUCN Red List (IUCN 2006 <http://www.IUCNredlist.org>) classification of all penguin species and the Department of Conservation classification, note: applicable to New Zealand species. Abbreviations: EX = extinct, EW = extinct in wild, CE = critically endangered, EN = endangered, VU = vulnerable, NT = near threatened, LC = least concern. Threat classification; 1 = Nationally Critical, 2 = Nationally Endangered, 3 = Nationally Vulnerable, 4 = Serious Decline, 5 = Gradual Decline, 6 = Sparse, 7 = Range Restricted.

| Species | | IUCN | DoC Threat |
|---------------------------|-----------------------------|------|------------|
| Emperor Penguin | <i>Aptenodytes forsteri</i> | LC | N/A |
| King Penguin | <i>A. patagonicus</i> | LC | N/A |
| Rockhopper Penguin | <i>Eudyptes chrysocome</i> | VU | 4 |
| Macaroni Penguin | <i>E. chrysolophus</i> | VU | N/A |
| Fiordland crested Penguin | <i>E. pachyryhynchus</i> | VU | 5 |
| Snares Crested Penguin | <i>E. robustus</i> | VU | 7 |
| Royal Penguin | <i>E. schlegeli</i> | VU | N/A |
| Erect-crested penguin | <i>E. sclateri</i> | EN | 2 |
| Little Penguin | <i>E. minor</i> | LC | 5 |
| Yellow-eyed Penguin | <i>Megadyptes antipodes</i> | EN | 1 |
| Adélie Penguin | <i>Pygoscelis adeliae</i> | LC | N/A |
| Chinstrap Penguin | <i>P. antarcticus</i> | LC | N/A |
| Gentoo Penguin | <i>P. papua</i> | NT | N/A |
| African Penguin | <i>Spheniscus demersus</i> | VU | N/A |
| Humbolt Penguin | <i>S. humboldti</i> | VU | N/A |
| Magellanic Penguin | <i>S. magellanicus</i> | NT | N/A |
| Galapagos Penguin | <i>S. mendiculus</i> | EN | N/A |

1.2.3 Life Chronology

The LBP life-cycle chronology includes; foraging time at sea, pair bond formation, courtship, breeding, and moulting. Circadian activity involves feeding at sea during the day and roosting ashore at night. The number of penguins coming ashore at night has been found to vary with the time of year and hence breeding activity. Jones (1978) found that the number of LBP coming ashore during 1974 - 1976 is lowest during March and April with the birds primarily feeding at sea. Numbers would then peak around May, before breeding, and level off around June and July when courtship and burrow occupation began. Low numbers of penguins come ashore during November when birds forage prior to moult to build fat stores for survival during the fast required for the moult. Annual moult occurs from late December until early March and takes on average 15 days ashore over which time LBP can not return to the sea to breed. Peak abundance is around January which coincides with the start of moult. Body weight also varies with the time of year and a pre-breeding peak in body weight occurs in June, dropping throughout the breeding season as a result of egg laying, incubation, and chick rearing (Jones 1978).

1.2.4 Biology

Little Blue Penguins are on average around 40cm high and weigh around 1000g. LBP do not show high levels of sexual dimorphism and have monomorphic plumage, however, males are generally larger than the females at maturity (Agnew and Kerny, 1995; Miyazaki and Waas 2003b). LBP have also been found to show sexual dimorphism based on bill size (Kinsky and Falla 1976; Gales 1988; Renner and Davis 1999; Hocken and Russel 2002) but can be difficult to judge as it is not obvious. This varies geographically (Renner 1998) and requires a separate discriminant functional

analysis for each sub-species (Kinsky and Falla 1976; Meredith and Sin 1998; Renner 1998).

1.2.5 Breeding Ecology

Breeding in *E. minor* starts after 2 years of age and occurs generally from July through to January (Klomp and Wooller 1991; Miyazaki and Waas 2003a; Chen 2004). The annual life cycle includes a breeding season followed by moult and then a non breeding period spent mostly foraging at sea. The breeding season begins with a courtship period where old pair bonds will be re-established or new pair bonds formed. Established pairs reunite at the old nest site while males forming a new relationship will find a nest site prior to attracting a new mate (Marchant and Higgins 1990).

LBP breed in various surroundings (Kinsky 1960; Marchant and Higgins 1990; Miyazaki and Waas 2003) such as deep burrows or sand banks, human made nests boxes, dense vegetation, rock caves or under boulders (Marchant and Higgins 1990). Because LBP can live in burrows that can be several metres long, direct observations are difficult to obtain (Dann 1994; Perriman and Steen 2000). Although in some populations, including Tiri, research has been facilitated by the use of nesting boxes (Jones 1978).

LBP in Australia and New Zealand are highly philopatric (Dann 1992), returning from sea to breed at their natal site (Pledger and Bullen 1998). Penguins may occupy several sites before they settle on one for breeding (Dann 1994; Perriman and Steen 2000). LBP can lay one to three eggs but will lay on average two eggs. LBP are a monogamous species and parental care duties are shared between the sexes, which include incubation, chick brooding, and feeding (Miyazaki and Waas. 2003). Adults change duties every one to two days during the early evening when they return from sea

(Miyazaki and Waas 2003), then before dawn the other partner will head out to sea to forage and replenish food stores. Food demand of the chicks is constantly growing with age (Culik 1994; Mattern 1991), therefore the foraging efforts of both adults increase with chick age and clutch size.

Breeding success of this species varies between years. This may be influenced by colony specific factors such as the weather, burrow substrate type (Perriman and McKinlay 1995; Perriman and Steen 2000), and nest site selection (Miyazaki and Waas 2003), or non-colony specific factors such as food availability (Cullen *et al.* 1992; Perriman and Steen 2000). In addition the initial onset of breeding may relate to food availability, which according to Cullen *et al.* (1992) indicates that conditions are favourable (Cullen *et al.* 1992; Perriman and Steen. 2000). Studies in Motuora Island in the Marlborough Sounds and Oamaru, Otago have found that there is a difference in the length of time foraging and nest attendance between these two sites. Differences in the number of nest desertions between the two populations could be due to the differences in food abundance within surrounding waters (Numata *et al.* 2000). There is a greater chance that a nest will be deserted when foraging duration is increased. Therefore higher prey availability close to the breeding sites of the penguins is expected to play an important role in increasing breeding success (Weavers 1992; Mattern 2001).

1.2.6 Feeding Ecology

When considering the role of penguins within an ecosystem it is of particular importance to look at their diet (Gales 1985). This requires understanding the relationship of diet and the factors that may limit population sizes (Dann *et al.* 1992). Penguins often spend their days at sea feeding within relatively shallow waters. They

are considered top predators even though they themselves are prey for Orca (*Orcinus orca*), sharks and seals (Spellburg 1975; Jones 1978).

Their diet usually consists of small pelagic fishes (Perriman *et al.* 2000), krill, Anchovies (*Engraulis australis*), Pilchards (*Sardinops sagax*), plankton, small octopi, squid, and crustaceans (Montague and Cullen 1988). Studies looking at the diet of penguins have involved analysing stomach contents by stomach flushing, the use of emetics, and autopsies (Gales 1985; Cullen *et al.* 1992). Studies have shown that the penguins tend to be generalist predators (Cullen *et al.* 1992) and may change their diet seasonally since penguin foraging tactics may vary based on prey type and availability (Schreiber and Burger 2002).

LBP spend a significant amount of time at sea (Weavers 1992), but like all penguins they breed and moult ashore. Foraging trips can extend from days to weeks depending on the time of year. Although penguins are marine organisms little is known about their foraging ranges and general travel within the sea (Weavers 1992). LBP are capable of extending out past a 25-kilometer home range zone (Mattern 2001) but this may not be viable in terms of energetic requirements if they are foraging to feed chicks. With the onset of the breeding season, foraging trips will be typically shortened. Arrivals and departures are synchronised with the return to the nest and departure occurring just before sunrise and sunset, respectively (Klomp and Wooller 1991). They are only found to feed their chicks at night (Miyazaki and Waas 2003) after they have been out foraging.

1.2.7 Threats

Since LBP are highly philopatric, changes in their natal site through habitat modification, disturbance and predation have the potential to influence the breeding

success of local populations. Studies on the coastal populations of penguins near Wellington show that mortalities and disturbance led to a local decrease in numbers (Kinsky 1960). Kinsky's (1960) study on Mātū-Somes Island, Wellington, revealed that changes in the ecosystem may have influenced the breeding biology and status of the colonies in this area. Mammalian predators such as dogs, cats, stoats, foxes and ferrets are a factor in population declines (Dann 1992), e.g. Tasmania (Stahel and Gales 1987) and Victoria (Dann 1992).

At sea, penguins have been frequently caught in near-shore nets (Dann, 1990 1992; Taylor 1999), however they are unlikely to be caught in trawling or captured by line techniques. LBP are also highly vulnerable to oil spills (Dann 1990). At present there is a high shipping volume that occurs within Auckland's Hauraki Gulf, also Marsden Point contains an oil refinery (Taylor 2000). Hence penguins are not free from the potential risks of an oil spill within these waters.

The main concerns surrounding New Zealand LBP populations are the declining numbers on the mainland and the mass beach wrecks. Large numbers of dead birds are termed a 'beach wreck' and are thought to occur every 3 to 4 years (Taylor *pers. comm*). Several factors have been proposed for these die-offs in Australia (Dann *et al.* 2000; Norman *et al.* 2000) however little has been done in New Zealand. Potential causes include weather effects such as storms and El Niño events (Norman *et al.* 1992), die-off of prey species (Dann *et al.* 2000), starvation (Harrigan 1992), and disease or toxicology outbreaks (Norman *et al.* 1992; Hocken 2002). Some of these factors may be correlated, for example storm events can cause starvation, fatigue, and potential injury to birds, resulting in large numbers of deaths. Past studies in Australia has associated mass beach wrecks with a mass die-off of a major prey species, the Pilchard. A similar mortality event of Pilchards occurred in New Zealand in 1995 (Griffen *et al.* 1997;

Hyatt *et al.* 1997), however, the effects on LBP were not documented. A die-off in preferred prey items could decrease the flexibility of the LBP diet, which may impact on the breeding success.

An understanding of the marine and terrestrial environments is required when considering factors associated with LBP breeding success and survival. The aim of this study is to identify the most influential factors associated with the North Island subspecies *E.m.iredalei* breeding on Tiritiri Matangi Island. This is achieved through moderate to intensive monitoring of the breeding stages (nesting through to fledging), analysis of the foraging ecology based on current diet and isotopic changes, and examination of annual mortality events.

1.2.8 Significance of this Study

The first study of LBP on Tiritiri Matangi Island (Tiri) was initiated by Jones (1978) who looked at the breeding success of LBP (*E .m. ireledai*) during 1974-1976. This study sampled a large part of the island and provided a good representation of population demographics. Since then no other study has quantified basic breeding data of this population. Miazayki and Waas (2003b) studied penguin vocalisations and the affect of body size on nest selection in 2000. These studies are informative however their findings were from a smaller sample size compared to this study, and came from only one location on Tiri. The only other LBP study on Tiri was by Chen (2004) who looked at breeding success and population demographics in 2003. During 2003 there were a series of big storms resulting in 100% nest desertion, but nest monitoring was restricted to a small area of the island (Chen 2004). There is a growing need to establish sound baseline data for future research, establish the state of LBP on Tiri 30 years after

the last major study, and to gain an understanding of the factors that influence productivity.

1.3 Thesis Aims and Objectives

The major aim of this thesis was to gain a better understanding of the North Island sub-species of Little Blue Penguin through establishing strong baseline data for a population which will assist future monitoring. Chapters were written in a format similar to that of individual papers. Each chapter can be considered as a stand alone topic, however for a greater understanding of Little Blue Penguins each chapter topic should be considered in light of the previous and following chapters.

Chapter 1

Chapter 1 provides a brief introduction to penguin species and establishes a general overview of Little Blue Penguin ecology and biology relevant to the following chapters.

Chapter 2

The aims of this thesis were, in Chapter 2, to provide baseline data on breeding ecology of Little Blue Penguins found on Tiritiri Matangi Island. General study aspects required for in-field monitoring included individual banding and sexing for future identification. Therefore the methods and results of this chapter include banding of individuals and the development of a discriminant function analysis for in-field sexing. This individual information was used to assist the methods for monitoring of breeding success, which aimed to identify the most influential parameters on breeding success and adult survival. Information such as this can be used together with future monitoring to identify a standard performance measure for this population.

Chapter 3

Chapter 3 can be considered as an extension of Chapter 2 by taking a more in-depth approach to understanding breeding success and failure. The aims of this Chapter were to identify the causes of egg and chick mortality and consider factors that may influence success at both nesting stages. The overall thesis aim of establishing baseline data was targeted through identifying embryonic developmental stages and quantifying parameters influencing eggs and chicks. If this is achieved for a population then future monitoring can be achieved through inter-annual comparisons.

Chapter 4

Chapter 4 dealt primarily with the feeding ecology of Little Blue Penguins found on Tiritiri Matangi Island and within the Hauraki Gulf. The primary aims of this Chapter were to a) determine prey types of LBP feeding within the Hauraki Gulf, b) identify which trophic level Little Blue Penguins belong to and whether this has changed through time through $\delta^{15}\text{N}$ analysis, c) identify whether Little Blue Penguins are conforming to inshore feeding and whether productivity levels have changed since 1886 from $\delta^{13}\text{C}$ analysis, and d) consider potential factors that could influence changes within the marine environment that could influence Little Blue Penguin diet. Overall aims and results are related back to impact that diet may have on the productivity and survival of Little Blue Penguins.

Chapter 5

Chapter 5 dealt with mortality of Little Blue Penguins from the Hauraki Gulf and from around New Zealand. The aims of this chapter were to identify a) the cause of death for

Little Blue Penguins found during the study period, b) provide baseline data on the numbers found dead during 2005 and 2006, and c) quantify underlying patterns associated with long-term mass mortalities associated with Little Blue Penguins from around New Zealand. Causes of mortalities and patterns of mortality events were considered on short and long-term scales.

Chapter 6

Finally, in Chapter 6, I aimed to determine the overall link between the feeding, breeding and survival of this penguin species. This study considers the use of Little Blue Penguins as a biological indicator to assess the health of the marine environment since they are greatly influenced by both the land and the sea. Furthermore, this thesis aims to formulate management recommendations for the enhancement of LBP populations and to identify key questions for future monitoring.

This is the first study designed to provide a comprehensive investigation into the feeding and breeding ecology of Little Blue Penguins comparing old and new techniques to gather baseline data. This study has the ultimate goal of providing a starting block for future monitoring to build upon to enhance our understanding of the links between marine and terrestrial environments over the long term. This is particularly important for species that rely heavily on both media, but also in terms of island systems since the land is directly and indirectly influenced by the immediate surrounding environment.

CHAPTER 2 Breeding ecology of Little Blue Penguins



Plate 2.1. Little Blue Penguin chick. Photo by J.Geurts 2005.

2.1 ABSTRACT

To understand population demographics of a species detailed baseline data are required, preferably over long-term monitoring periods. The breeding success of Little Blue Penguins (LBP) on Tiritiri Matangi Island (Tiri), Hauraki Gulf, New Zealand was monitored during the 2005/06 breeding season. Parameters such as lay date, nest distance, and nest type were measured and correlated with nesting success. Results showed that LBP breeding on Tiri during 2005 had a very poor breeding year with only 0.2 chicks fledging per pair (10% breeding success) All fledged chicks came from early clutches (9th September and 31st October). The lay date was the only significant parameter associated with breeding success, with early laying nests having the highest probability of fledging a chick. The timing of initiation of breeding for six years of known lay dates coincided with sea surface temperature (SST) of 14.68°C. This low temperature for the region may have been associated with higher levels of prey availability and better body condition, but requires further investigation. No pairs were found to double brood (DB) however ten pairs attempted replacement clutches. Nest type may not be a major influence of breeding success however this cannot be ruled out due to lack of long-term monitoring.

2.2 INTRODUCTION

Marine birds are increasingly being used as indicators to determine the health of the marine environment since parameters of seabird behaviour and reproduction change with the level of marine resources and food supply (Boersma 1978; Dann 1992; Cairns 1987). These parameters include monitoring of the lay date (Monaghan 1996), body condition (Cairns 1987; Monaghan 1996), and reproductive success (Cairns 1987). LBP are top predators in the marine environment and as such they forage at high trophic levels within the marine environment, therefore monitoring of their productivity should provide information on prey availability and abundance.

Avian studies have found many factors that influence the success of any breeding attempt. Crucial parameters include biotic factors (foraging behaviour and diet, lay date, age, and fidelity of breeding pairs), and abiotic factors (sea surface temperature: SST, climate, and microhabitat) (Schreiber and Burger 2002). For example, breeding success of many species will decrease with lay date as a result of many different ecological and physiological reasons: European Starling (*Sturnus vulgaris*: Dawson 2003), Wren (*Troglodytes troglodytes*: Evans and Goldsmith 2000), Black Kite (*Mulrus migrans*: Sergio 2003), Sparrow Hawk (*Acipiter nisus*: Newton *et al.* 1981; Newton and Rothery 1998), Black-legged Kittawakes (*Rissa tridactyla*: Gill and Hatch 2002) Snow Geese (*Anser caerulescens*: Hamann and Cooke 1987), King Penguin (*Aptenodyptes patagonicis*: Van Heezik *et al.* 1994), Common Tern (*Sterna hirundo*: Arnold *et al.* 2004), Common Murres (*Uria aalge*: Benowitz-Fredericks and Kitaysky 2005) and LBP (Knight and Rogers 2004).

The onset of breeding in LBP is hypothesised to coincide with low sea temperatures as productivity of the marine environment inversely relates to temperature.

Increase in primary productivity will therefore increase prey species through the food chain. Studies of LBP breeding on Phillip Island, Australia have shown a clear dependency of breeding in relation to food supply (Michelson *et al.* 1992). Increases in food abundance will influence adult body condition and food provisioning in chicks as it enables adults to meet the energetic demands of breeding. If sea temperatures do affect food availability the variation in annual onsets of breeding should correlate with variations in SST (Perriman *et al.* 2000). For example, the Galapagos Penguin *Spheniscus mendiculus* has been found to time the onset of breeding with lower SST (Boersma 1978; Robinson *et al.* 2005). Penguins spend at least half of their lifetime at sea and rely entirely on the marine environment for their foraging requirements, therefore it is likely that they synchronise their breeding with the time of year that will ensure the best chance for chick survival (Monaghan *et al.* 1992; Robinson *et al.* 2005).

2.2.1 Breeding in Little Blue Penguin

Productivity of breeding pairs is defined here as breeding success and is determined by the number of clutches laid, the number of eggs per clutch, hatching rate, and fledging rate. LBP typically lay two eggs and incubate for 35 - 36 days (Miyazaki and Waas 2003). Once chicks hatch they are guarded by at least one parent for up to three weeks. Following the guard phase the chicks enter a post-guard stage where both parents will forage at sea, returning regularly to feed the chicks. Fledging of the chick/s occurs around 50 days after hatching with an average fledged weight of 1000 g (Perriman *et al.* 2000). Chicks fledging around December will return to molt within a year. Mating and breeding occurs between two and three years of age (Marchant and Higgins 1990). The mortality rate is highest during the first year after fledging but if chicks survive they can live for 25 years (Perriman *et al.* 2000). The timing of the LBP breeding season varies

from year to year, but generally the egg laying period occurs between July to November with variations occurring between conspecifics (Jones 1978). Although most pairs usually produce just one clutch, southern populations of LBP can successfully fledge chicks from two clutches within a single breeding season (Williams 1995; Johannesen *et al.* 2002). For example in good years LBP in Otago begin egg-laying in May raising two clutches (Renner 1998). This has not been observed anywhere else in New Zealand (Perriman *et al.* 2000).

Two types of breeders have been classified; double breeders (DB) and single breeders (SB). DB are defined as those that lay and raise two clutches. Some lay a second clutch to replace the first because of failure. SB only lay a single clutch during the breeding season regardless of the fate of the nesting attempt. Those that manage to successfully raise two clutches are thought to increase their fitness by increasing their current reproductive effort (Johannesen *et al.* 2002). These DB birds tend to be older and more experienced therefore it is not surprising that past studies have found that double breeders are more successful at fledging chicks than single breeders (Johannesen *et al.* 2002). To double brood LBP must lay very early in the breeding season as breeding must be completed before the moult period (late December to early March) (Perriman *et al.* 2000). On Tiri the majority of LBP start moulting in January and complete their moult by February (Jones 1978; Chen 2004). Past studies have found nest quality and age of the breeding pair to also influence LBP breeding success (Pielotti and Annette 1990; Perriman *et al.* 2000). For example Pielotti and Annette (1990) found that the number of chicks that fledged per pair was more dependent on the quality of each individual nest site than large scale climatic variation (Perriman *et al.* 2000).

2.2.2 Significance of Current Study

Despite increasing numbers of studies on LBP (*Eudyptula minor*) knowledge of the North Island sub-species (*E. m. iredalie*) is still limited, especially in the northern areas of the North Island. Studies of the North Island sub-species have been conducted on Matiu-Somes Island, Wellington and Tiritiri Matangi Island, Auckland. Three studies have come from Tiri (Jones 1978; Miyazaki and Waas 2003; Chen 2004) spanning 30 years. This timescale covers the period of restoration and predator control on the island. Knowledge of basic breeding parameters such as population estimates and breeding success for different populations of a species is useful for accounting for variation. However, the factors that may influence the productivity of any particular population within a species can vary spatially and temporally. Therefore, there is a need to estimate and identify the key breeding parameters that influence the viability of a given population. Identification of parameters for each population can enhance conservation efforts by targeting the actual requirements of any one population. Therefore baseline data are very important for management schemes but also provide a building block for more informative research.

2.3 Aims

1. To band individuals and establish an in-field method of gender determination for the population of Little Blue Penguins from Tiritiri Matangi Island population.
2. Establish baseline data on the breeding ecology for the LBP population on Tiri.
3. Identify the major drivers of nest success.
4. Compare between years and between other populations.

2.4 METHODS

This chapter is a general chapter written to include methods required for all chapters. Initial set up for analysis of LBP include banding, nest location, and measurements of gender differences.

2.4.1 Study Site

This study was carried out on Tiritiri Matangi Island (Tiri) (36°36'S, 174°53'E), New Zealand (Figure 2.1), during the 2005/ 2006 breeding season (Austral summer). Tiri is a 220 hectare island located 32km north of Auckland, and approximately 4 km from the Whangaparaoa coastline. Tiri is a refuge to some of New Zealand's most endangered birds and is recently free from introduced predators such as cats, dogs, Kiore (*Rattus exulans*), and mustelids. The island coastline is rugged and steep, composed of large grey-wake boulders, shingle beaches, and rocky headlands (Jones 1978). The mainland is composed of regenerating forest, grassland, and dense bracken bush (Jones 1978).

2.4.1.1 Sampling Area

Surveys of penguin nests were carried out around the accessible coast of the island. This included coastlines, bush, and grass land searches. Areas sampled were similar to those covered by Jones (1978), who classified areas of low to high accessibility. The island's coast was scanned for nests while searches through bush areas were time consuming and less profitable as also found by Jones (1978). Some areas were accessible during both high and low tide, while others were only accessible during low tide or not at all.

Monitored sites on the island were divided into six areas relating to the location on the island: East Coast (Fisherman's Bay to Lighthouse), North East coast (Pohutakawa Cove up to North East Bay), West (from North Point down to Tiritiri

Matangi Pa Point, including bush 1), Southwest (from Pa Point to Hobbs Beach, including bush 2 and 3), South (Wharf and Hobbs Beach), Southeast (below Wattle Valley)(Figure 2.2).

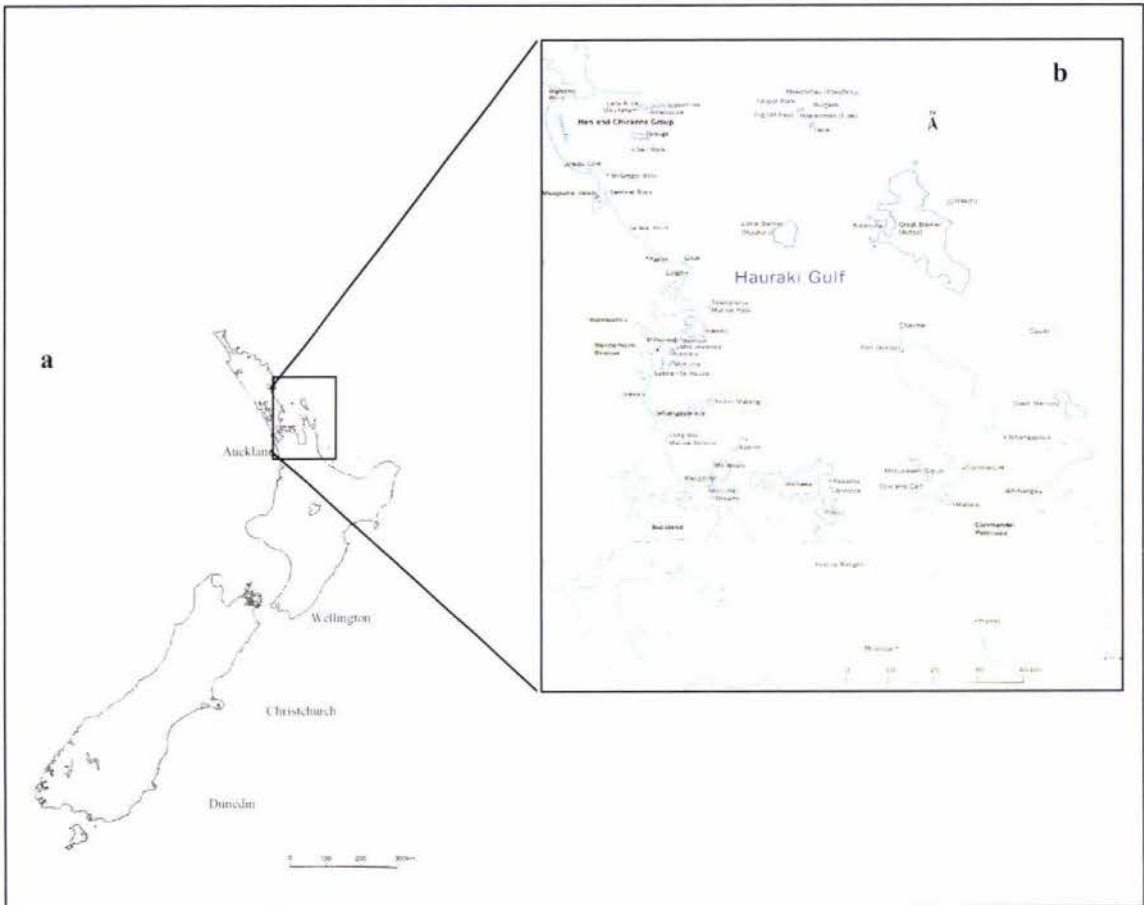


Figure 2.1.a) Map of New Zealand outlining the Hauraki Gulf, b) enlarged area of the Hauraki Gulf with Tiritiri Matangi Island situated of Whangaparaoa Peninsula. (Altered from Rimmer 2004, p. 12).

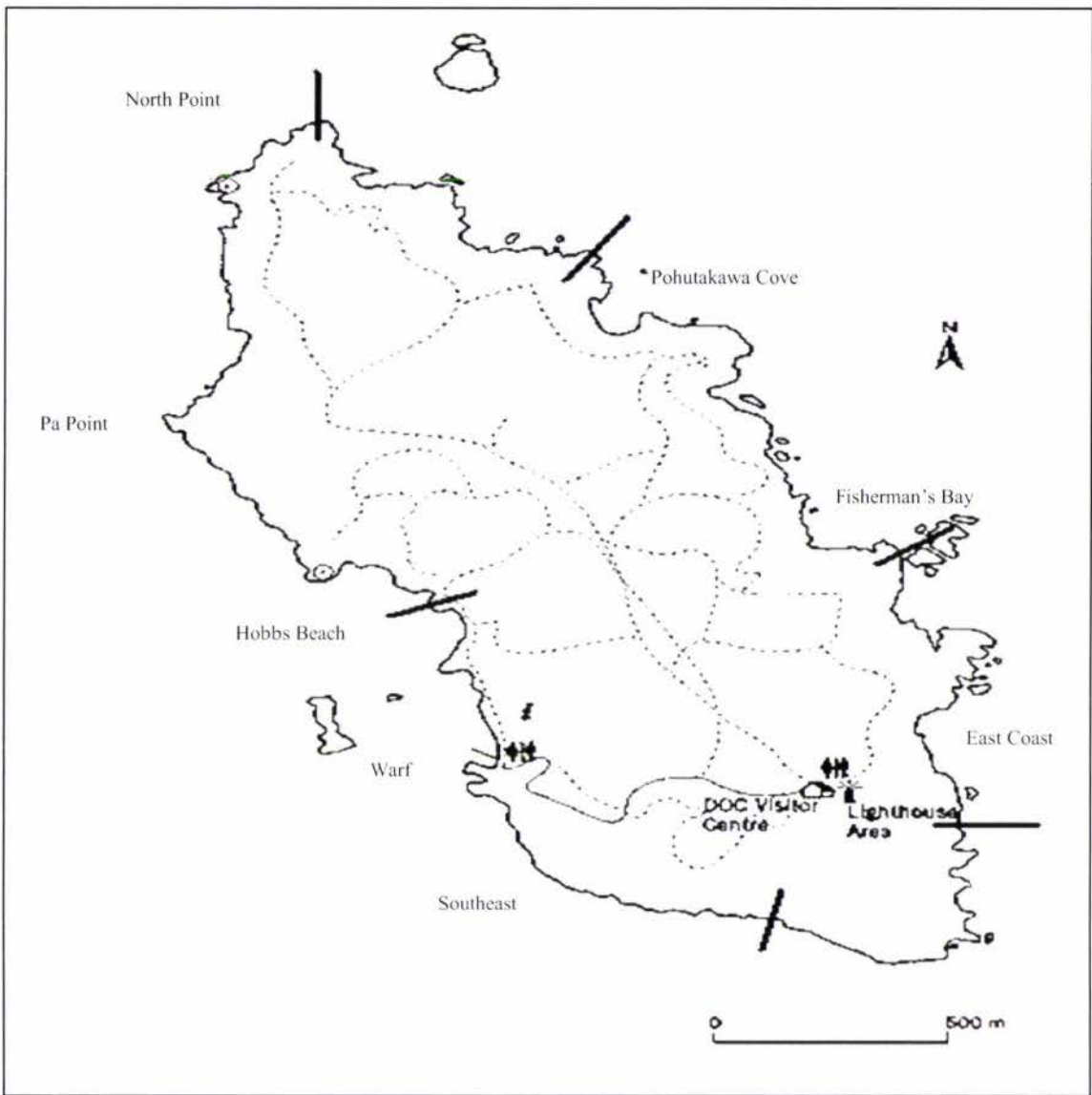


Figure 2.2. Locations monitored on Tiritiri Matangi Island. Lines represent the boundaries of the six sections which relate to the location on the island.

2.4.2 Survey Methods

2.4.2.1 Locating Nests

Survey trips to Tiri started at the end of June 2005. A total of 24 trips were made during the course of the study and were determined based on weather conditions, usually lasting for 3-4 days each week. Nest searches were initially conducted at night by listening for penguin vocalisations to locate any potential breeding sites (Miyazaki and Waas 2003b). Nest searches were then undertaken during the day which allowed for tide

changes and increased the sampling area. Daytime searches could be conducted in the more inaccessible areas of the island such as the East Coast and the North West Point. Two types of nests were identified; Potential Nests (PN) and Breeding Nests (BN) (Mattern 2001). Potential nests were identified as burrows that contained penguin faeces, nest material, or the presence of one or two adult birds. Breeding nests were defined as burrows that showed signs of breeding activity (eggs and/ or chicks). Each nest was individually marked with flagging tape (*Geosystems Ltd*) and identified according to the island location. A number was also allocated to each nest according to the order in which they were found. A global positioning point was recorded with a *Garmin 60c* positioning device (GPS) (*Geosystems Ltd*); with 4-15 m accuracy.

The distance that each nest was from the spring high tide mark was also measured with a 100 meter measuring tape. Nest to sea distance was then categorised as; close (<10m), medium (10m-100m) or far (>100m). Vegetation was also associated with each distance. For example, close was near the coastline with surrounding rocks, grassland, or flax, medium extended past this into dense bracken and bush, and far was usually associated with inland areas of bush.

Playback calls were then trialled to help locate breeding pairs. Calls were recorded from three pairs located on the island. The presence of other breeding pairs was identified by vocal responses to the playback. This technique did obtain responses, however a solitary penguin appears to stay quiet so as not to advertise its presence (Waas 2001; Miyazaki and Waas 2003b). Nests that were hard to observe directly were checked with a burrow scope (*Provision 636, Technical Solutions Corp.*).

2.4.2.2 Nest Types

Each nest was assigned to one of four nest types based on the main substrate type; rock, earth, tree, and artificial (

Figure 2.3). These nest types were similar to that of Renner (1998). Rock types are associated with rock crevices, under boulders, and rock caves. Earth nests are usually nest holes that are built into the dirt bank, or within the ground. Tree type nests are holes that are found under any vegetation such as Flax (*Phormium tenax*), dense Bracken (*Pteridium esculentum*), and tree roots. In 1988 four stone nests were built near the Wharf area (Chen 2004) and in 1999 36 wooden nests were placed around the island to aid breeding. These are identified as the artificial breeding nest types.



Figure 2.3. The different Little Blue Penguin nest type classifications used: a) 'dirt' nest, b) 'tree' nest, c) 'rock' nest, and d) 'artificial' nest. Photos by J.Geurts 2005.

2.4.2.3 Banding Procedure

During the 2005 breeding season 69 breeding pairs were identified. Adult pairs were marked with individually numbered stainless steel flipper bands (unless previously banded), which allowed for future identification (Plate 2.2). Since 1974 LBP have been banded using similar bands. Each band was attached using three pairs of pliers. The large pair allowed for closure of the band, while two needle pliers worked together to pull the band apart in order to get the band ends to meet. Pressure was then applied to push the ends together to force the band face (part with the numbers) to become flattened.



Plate 2.2. Stainless steel band on Little Blue Penguin. Photo by J. Geurts 2005.

2.4.2.4 Sexing

Where possible, sex was determined using molecular sexing of feather samples (Allan Wilson Centre, Massey University).

2.4.3 Data Collection

2.4.3.1 Nest Monitoring

Nests were checked as often as possible throughout the breeding season (between 3 - 4 days a week). Nest checks occurred once during the day depending on the nest and involved visual sighting with a torch. Monitoring allowed identification of any nest changes such as adult change over and nest contents. Data collection from each nest check included: presence/ absence of adults, contents (eggs/ chicks), weather data, and measuring of chicks. Additionally, any newly found adults were banded.

At the time of banding each adult was weighed in a breathable cotton bag of known weight with a five kg pesola scale. A sample of two feathers were taken from the lower back of each bird and put into a labelled plastic bag. Adult birds were placed back on the nest according to their weight size with smaller individuals being put back first to avoid aggression between birds. If the gender of the birds were known through behavioural monitoring (aggression, stance or vocalisation) then females would be placed on the nest first (Houston *pers. comm*). This was done to limit the aggression that could occur between the different sexes when the natural entrance to a nest has not been established. This may mean that under usual circumstances, the individuals may call or make their presence known to the other one before entering the burrow.

Monitoring of each nest allowed for identification of the different breeding stages. This generally started with nest building, followed by egg laying, incubation,

hatching, brooding, guarding, post guarding, and finally fledging. If possible the nest was checked every day until an outcome was achieved, such as success or failure. Since nests were found at different stages, monitoring varied. Monitoring of nests during the incubation phase occurred as often as possible; averaging around three to four checks a week. The presence of eggs or nestlings was checked for by using the flattened end of kitchen tongs to gently lift the back end of the adult up. Nests with a known lay date could be visually checked until close to hatching (~26 days), after which the incubating adult would be lifted once a week. Adults on nests found during incubation with lay date unknown were lifted once a week. When nests were close to hatching, visitations included listening for chick calls. This was a good method of identifying chick presence without nest intrusion. Physical manipulation was limited where possible. Adult pairs that recently laid their eggs or hatched chicks were left undisturbed for three days to avoid nest desertion. If at any stage during incubation both adults were away from the nest, the duration of absence was noted.

If chicks were still in the brooding and guard stages, the removal of the chicks required adult birds to be taken off the nest first. Adults were held in a cotton bag during chick measurements. Chicks were removed from the nest by hand. If there were two chicks on the nest the youngest or lightest was marked with non-toxic correction fluid. Marking occurred on the back of the neck, tail, under the right flipper, and foot. Other studies have marked chicks by punching a hole in the foot webbing, or clipping a toenail (Jones 1978). These alternatives were not considered ideal and correction fluid was effective but required re-marking at each visit. Chicks were always placed back on the nest before release of the adult. Chicks were weighed twice a week from hatching, or from the date of finding. Each chick was placed in a micro fleece bag and weighed on 400g electronic scale. The bag weight was also taken afterwards. Eight morphological

measurements (after Jones (1978) were taken once a week at the time of the first weekly weigh using 200 mm digital calipers (*Kincrome*): 1) Head length (HL), 2) Head width (HW), 3) Beak length (BL), 4) Nostril to beak tip (NT), 5) Beak depth 1 (BD1), 6) Beak depth 2 (BD2), 7) Wing length (WL), and 8) Tarsus (T) (Table 2.4). An overall beak depth was calculated as used by Jones (1978) (Appendix 2.7.1). Each measurement was double-checked, always taken from the right side of the bird, and at the same time of day for accuracy and consistency. The weight of the bird could vary with the time of day due to feeding and sweating (Miyazaki and Waas 2003a).

When the chicks were near fledging (usually around six weeks), they were banded with a flipper band and a feather sample taken. For nests found during the chick stage, chick age could be estimated based on comparisons with other active nests and through morphological measurements. Age could be estimated to an accuracy of around five days by comparing growth measurements with chick growth curves associated with LBP on Tiri (Jones 1978).

Table 2.1. Description of the morphological measurements that were taken from Little Blue Penguin adults and chicks throughout the course of the 2005/06 study period at Tiritiri Matangi Island. Measurements with asterisk were also used by Jones (1978).

| Measurements | Description |
|-------------------------------|-----------------------------------------------------------------------|
| 1. Head Length (HL)* | From the tip of the bill to the back of the skull |
| 2. Head Width (HW)* | From behind the eye at the widest part of the skull |
| 3. Bill length (BL)* | From the tip of the bill to the integument of the forehead |
| 4. Nostril to Tip (NT)* | From the tip of the bill to the posterior end of the nostril |
| 5. Depth at the Gonys (BD1)* | From the base tip of the gonys |
| 6. Depth at the Culmen (BD2)* | From the base of the culmen to the lower edge of the mandibular ramus |
| 7. Wing Length (WL) | From the wrist to the wing tip |
| 8. Tarsus (T) | From the heel of the right foot to the mid of the foot pad |
| 9. Overall Beak depth* | $BD2 \times BD1 / 100$ |

No chicks were taken off the nest during rain to minimise any chicks getting wet. New nests may be found deserted and could be defined as one that has not been occupied by an adult for a week. Potentially nests could be occupied but the eggs pushed out which was considered as failed. If a nest was found deserted or failed, any eggs or dead chicks were removed for later analysis. Eggs that were found alone on a nest were left for a week before removal to ensure that they were actually abandoned and not just unattended for a day or two. Removal of eggs was done either by hand, or where needed, with long steel tongs. Eggs were put into plastic re-sealable bags and frozen at -20°C for later necropsies. Dead chicks were removed on the day they were found dead, put into labelled bags, and frozen for later necropsies. Once a nest was classified as abandoned a sample of nesting material was taken, bagged and frozen for parasite analysis.

2.4.3.2 Definitions

Lay date was taken from the first egg laid within the season. If nests were found with a complete clutch the lay date could be estimated at time of hatching using the average incubation length of 36 days (Marchant and Higgins 1990). If nests were found at the

chick stage then lay date could be estimated from chick age and average incubation length.

Breeding pairs were classified as replacement double brooders (RDB) if they were found to lay a replacement clutch during the monitored breeding season. Single brooders (SB) were those breeding pairs that only laid one clutch. RDB is distinct from actual double brooders where birds lay and raises two clutches. Each egg was assigned 1 of 5 classifications of failure (Renner 1998).

- a. *Nest desertion* was defined as eggs that were abandoned by the adults and were not being incubated for longer than a week.
- b. *Weather effects* were based on how wet the burrow was at the time the egg was removed.
- c. *Egg out of nest* was when the egg was cold and off the nest as a result of adults pushing it out as they left, or due to other adults that may have come in to the nest.
- d. *Egg broken* was when the egg was broken and empty, or had cracks.
- e. *Unknown* was when the egg disappeared or if the other classifications could not be applied. When failed the eggs were removed, the condition of the nest (wet/ dry) was also recorded.

Chick failure was assigned one of five classifications based on the probable cause of failure and used by Renner (1998):

- f. *Nest desertion* was used when a dead chick was found on a nest with no adults. The reasons for actual death from this vary depending on the age of the chick. If the chick is within the first two weeks of hatching nest desertion could cause hypothermia and /or starvation. Older than two weeks and nest desertion would cause starvation due to lack of provisioning (Renner 1998).

- g. *Starvation* was assigned when the weights of the chicks were found to be decreasing or if necropsies show evidence of empty stomach and crop.
- h. *Weather effects* were assigned when the nest and plumage of the chicks were soaking wet.
- i. *Disappearance* was assigned when the chick was found to be absent from the nest, which could happen without complete failure of the nest (siblings). Disappearance could occur when the chick is older and more mobile. If deserted they may leave the nest in search of food.
- j. *Unknown* was assigned when a chick was found dead and the cause unable to be ascertained.

2.4.4 Weather Data

During the breeding season, weather parameters such as daily wind direction, speed, and total rainfall were obtained from two meteorological stations, including Tiri and Whangaparaoa Peninsula obtained from the National Institute of Water and Atmospheric Science (NIWA) for 2005/ 06.

Monthly averages in sea surface temperature (SST) for the Hauraki Gulf were obtained through Leigh Marine Laboratories (Auckland University) for the period covering the study season. This was also obtained for previous years that lay dates were known: 1974-1976 and 2000-2003. SST was measured at 0900 daily with a bucket of seawater and a calibrated mercury thermometer within 30sec of obtaining the sample.

2.4.5 Data Analyses

2.4.5.1 Sexing

A binary logistic regression was used to test the differences of each morphological measurement between the sexes using *SPSS version 0.8*. A discriminant function analysis (DFA) was also conducted however this assumes normal distribution whereas the binomial test does not. Student's *t*-tests (*SAS v.8*, 2002) were used to test the differences between the average weights between the two sexes (male versus female), dead birds (1970 versus 2005/06), and fledged chicks (male versus female) in. Birds of unknown gender were not used in any analysis.

2.4.5.2 Breeding success

The affects of lay date, nest type, nest distance, on hatching and fledging successes were analysed separately using categorical data modelling (*CATMOD* in *SAS v.8*). This method provided a chi-square approximation of expected probabilities and standard errors on estimates. Nest distance required all medium classifications to be grouped into close with far representing distances of over 100 m from the spring high tide mark. Grouping distances this way allowed for greater combinations and hence sample sizes for all combinations to run the test.

Lay date was classified as early or late based on the relationship to the median lay date (31st October). Nest type was either rock, artificial, earth, or tree, distance was either close, or far with all medium nests being classified as far to increase sample size with all combinations. Nests classified as 'medium' distance were combined with 'far' nests since anything further than 10 meters could be considered a long way to travel for an animal less adapted to walking on land. Fisher's exact test was used to test the difference between lay dates of early and late versus the outcome (success or fail). A

Fisher's exact test was used to test the difference between the lay date (early or late) and stage of failure (egg or chick).

Reproductive success was calculated as the number of eggs that fledged chicks (Table 2.2) and was compared to other studies. Breeding success was also calculated using Mayfield's method estimated by the number of nest days monitored (Mayfield 1975). This also estimates the probability of survival at incubation and nestling period and at hatching.

Table 2.2 Definitions used for calculating the different stages of success for Little Blue Penguin on Tiritiri Matangi Island, New Zealand.

| Term | Definitions |
|---------------------------|----------------------------------------------------------|
| Nests | A nest or burrow that contains at least one egg or chick |
| Hatching success | % of eggs that hatched |
| Fledging success | % of eggs that hatched and fledged chicks |
| Breeding success | % of laid eggs that fledged chicks |
| Number of chicks per nest | The number of chicks that fledged per nest |
| Nest success | % of nests that fledged at least one chick |

2.4.5.3 Weather Effects

The laying dates from previous years (1974 - 1976, 1999, 2003) and the current breeding season were graphed against SST averaged over the corresponding month that eggs were laid. The average SST (\pm SE) for the lay month was compared against the average SST for the month prior to and after the initial laying month, and against the maximum and minimums for the corresponding year.

2.4.5.4 Nest Material

Nest material was analysed in the laboratory to look for any nesting parasites. Any parasites found were subsequently analysed at AgResearch for species identification to consider effects that they may have on chick and adult survival.

2.5 Results

2.5.1 Banding

Since 1974 a total of 999 birds have been banded on Tiri (Table 2.3). In this study a total of 103 were banded. Banding of LBP occurred at different times: Night time capture on the beach, on nests, as chicks, and during regurgitation sampling. A total of 22 previously banded birds were re-sighted during the course of this study (Appendix 2.7.2). A total of four birds newly banded were found dead within the Hauraki Gulf region and two previously banded were found dead on Tiri during 2005-06.

Table 2.3. Number of Little Blue Penguin banded in previous years on Tiritiri Matangi Island, New Zealand. Table altered from existing table in Chen 2004, p. 15.

| Year | # Banded | Researcher |
|-------|----------|------------------|
| 1974 | 172 | Jones (1979) |
| 1975 | 111 | |
| 1976 | 148 | |
| 1977 | 58 | |
| 1981 | 4 | Waas (2002) |
| 1994 | 59 | |
| 1999 | 192 | |
| 2000 | 40 | Geurts (2005/06) |
| 2001 | 112 | |
| 2005 | 103 | |
| Total | 999 | |

Several of the nests were found at abandonment and therefore the adult was unidentifiable ($n = 12$). Other nests were able to be visually monitored, but the adults were not able to be banded. Finally some nests were abandoned or went to unguarded stages of nesting, before adults could be banded. This meant that even though one bird of the pair was identified, the other was not. Banding at the nest occurred during the day as many nests were too dangerous to access during the night.

2.5.2 Sexing

2.5.2.1 Molecular sexing

Although the sex of some individuals was not able to be identified through molecular DNA analysis, gender ratios of male, female adults and chicks were calculated for the population.

Averages of morphological measurements for both males and females were calculated (Table 2.4). Results of the binary logistic regression showed that the most discriminating measurement was beak depth 1 at the gonys giving >80% accuracy of classifying a bird as either male or female for both adults and sub adults. This only misclassified 1/7 males as female, and 1/15 females as male (Table 2.5). The calculation to identify an individual bird as either male or female is as follows;

$$Y = 9.438 - 0.717 \times \text{BD1}$$

Gender classification: Female when $Y > 0$ and a male when $Y < 0$. Where BDI was bill depth 1 (see Methods Section 2.4.3.1).

Table 2.4. Morphological measurements of Little Blue Penguin on Tiritiri Matangi island, New Zealand, during 2005/06 monitoring period. Highlighted boxes indicate the sample that was misclassified as the opposite sex.

| | HL | HW | BL | NT | BD1 | BD2 | WL | T | BD | n |
|---------------|-------|-------|-------|-------|-------|-------|-------|-------|-------|----|
| Male | 87.35 | 35.33 | 35.22 | 22.48 | 13.28 | 15.54 | 61.52 | 33.02 | 2.14 | 10 |
| | ±2.52 | ±0.92 | ±0.89 | ±0.81 | ±0.63 | ±0.48 | ±0.84 | ±0.61 | 0.13 | |
| Female | 86.21 | 32.60 | 33.20 | 21.77 | 11.51 | 14.55 | 59.10 | 31.80 | 1.77 | 16 |
| | ±0.95 | ±0.53 | ±0.85 | ±0.25 | ±0.36 | ±1.40 | ±0.77 | ±0.29 | ±0.16 | |

Table 2.5. Measurements for male and female Little Blue Penguin from Tiritiri Matangi Island. The discriminant function (DF) estimates male (DF <0) and female (DF > 0). Highlighted values represent incorrect classification of gender. Age classification: U/K = unknown and Sub = sub-adult.

| Sex | Age | HL | HW | BL | NT | BD1 | BD2 | WL | T | BD | DF |
|--------|-----------|-------|--------|--------|--------|--------|--------|--------|-------|------|-------|
| Female | Adult | 90.4 | 34.3 | 43.7 | 23 | 12.1 | 14 | 57.4 | 31.3 | 1.69 | 0.76 |
| Female | U/K | 80 | 30 | 30 | 21 | 9 | 11.1 | 61.3 | 32.4 | 1.00 | 2.99 |
| Female | sub-adult | 88.06 | 32.73 | 34.65 | 23.88 | 11.11 | 13.22 | 59.49 | 32.96 | 1.47 | 1.47 |
| Female | Adult | 91.13 | 37.07 | 35.99 | 22.82 | 15.66 | 13.57 | 59.29 | 33.28 | 2.13 | -1.79 |
| Female | Adult | 81.83 | 31.52 | 30.36 | 19.68 | 9.47 | 11.05 | 57.37 | 29.61 | 1.05 | 2.65 |
| Female | Adult | 89.36 | 34.09 | 32.4 | 21.83 | 11.84 | 13.8 | 63.46 | 32.52 | 1.63 | 0.95 |
| Female | Adult | 84.54 | 32.19 | 31.95 | 22 | 11.62 | 13.86 | 63.33 | 30 | 1.61 | 1.11 |
| Female | Adult | 85.7 | 34.82 | 31.78 | 20.99 | 11.58 | 13.56 | 56.06 | 30.92 | 1.57 | 1.14 |
| Female | U/K | 87.06 | 32.94 | 33.71 | 22.43 | 11.68 | 13.42 | 61.03 | 32.1 | 1.57 | 1.06 |
| Female | Adult | 77.75 | 30.49 | 32.92 | 21.67 | 11.62 | 13.38 | 60.21 | 31.68 | 1.55 | 1.11 |
| Female | Adult | 87.31 | 32.54 | 30.89 | 21.75 | 10.84 | 14.22 | 63.99 | 32.24 | 1.54 | 1.67 |
| Female | Adult | 87.85 | 31.78 | 33.11 | 21.16 | 11.87 | 13.93 | 56.33 | 33.34 | 1.65 | 0.93 |
| Female | Adult | 84.95 | 30.23 | 32.63 | 21.55 | 12.08 | 13.17 | 54.97 | 30.63 | 1.59 | 0.78 |
| Female | Adult | 87.05 | 35.15 | 32.72 | 21.81 | 11.34 | 13.02 | 59.59 | 31.53 | 1.48 | 1.31 |
| Female | Adult | 89.12 | 30.22 | 34.73 | 22.02 | 10.785 | 34.625 | 57.02 | 31.66 | 3.73 | 1.71 |
| Female | Adult | 87.27 | 31.51 | 29.58 | 20.79 | 11.64 | 12.8 | 54.75 | 32.63 | 1.49 | 1.09 |
| Male | Adult | 94.34 | 39.04 | 39.27 | 24.69 | 13.68 | 15.36 | 63.7 | 32.14 | 2.10 | -0.37 |
| Male | Adult | 94.05 | 36.48 | 38.27 | 26.27 | 14.38 | 17.21 | 65.35 | 34.03 | 2.47 | -0.87 |
| Male | Adult | 91.17 | 35.78 | 34.81 | 21.73 | 14.47 | 15.49 | 63.66 | 34.28 | 2.24 | -0.94 |
| Male | Adult | 71.09 | 35.07 | 37.31 | 23.28 | 14.83 | 16.54 | 59.275 | 33.97 | 2.45 | -1.20 |
| Male | Adult | 91.27 | 38.33 | 34.99 | 20.65 | 13.72 | 16.18 | 62.94 | 33.99 | 2.22 | -0.40 |
| Male | Adult | 89.04 | 32.5 | 33.86 | 23.81 | 13.95 | 15.36 | 59.48 | 33.44 | 2.14 | -0.56 |
| Male | Adult | 84.77 | 35.4 | 33.54 | 22.31 | 14.08 | 15.63 | 59.28 | 33.53 | 2.20 | -0.66 |
| Male | Sub | 85.05 | 30.685 | 32.395 | 20.535 | 10.415 | 12.57 | 60.005 | 28.79 | 1.31 | 1.97 |

2.5.2.2 Weight differences

The mean (\pm SE) weights of all known adult males was 907.64 ± 19.40 g ($n = 49$, range 731.25 – 1125 g), for adult females was 853.71 ± 24.62 g ($n = 40$, range 507 – 1229 g) and for adult birds of unknown sex 857.47 ± 66.60 g ($n = 7$, range 660-1000 g). No significant differences were found between male and female mean weights ($t = 1.77$, $df = 86$, $p = 0.432$) (Figure 2.4). The mean weight of dead birds associated with Jones's study (unpubl. data, 1978) was 515 g ($n = 44$), whereas the mean dead weight of birds in this study was 465.82 ± 10.10 g ($n = 39$, range 325-565 g). The range of fledging weights was 557 to 1122.71 g. The mean (\pm SE) weight of male fledged chicks was 878.19 ± 34.14 g ($n = 7$, range 557-1097 g), females fledged chicks was 890.73 ± 68.95 g ($n = 7$, range 732-1104 g), and birds of unknown sex 961.53 ± 157.35 g ($n = 3$, range 707-1122 g). No significant difference was found between the fledging weights of males and females ($t = -0.13$, $df = 10$, $p = 0.9004$).

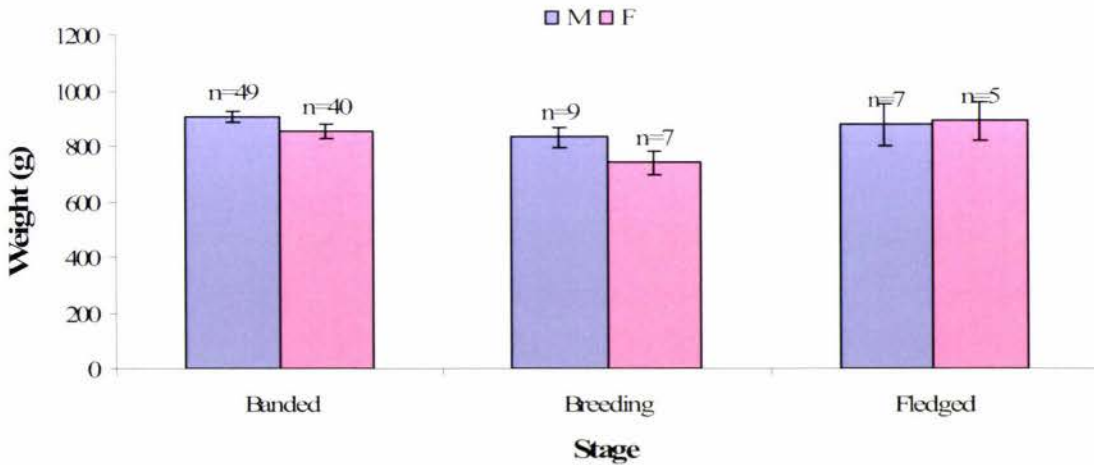


Figure 2.4. Mean weights (\pm SE) of the different Little Blue Penguin sexes at banding (all birds weighed), during breeding (on the nest), and for chicks that fledged. Weights of unknowns were not included.

2.5.3 Nesting Attempts

A total of 109 nesting burrows were found on Tiri and the breeding success of 87 nesting attempts was monitored over the course of the 2005/06 breeding season. Of the 69 different nesting territories, 56 nests were found on the coast (from mean high tide mark to 100 meters inland and 13 were found in the bush or at high coastal elevation (further than 100m inland). Nests were found in a range of locations on the Island (Figure 2.5): 15 nests on the east coast (Fisherman's Bay to light house), three on North East coast (North of Fisherman's Bay to Pohutakawa Cove), 12 were on the West Coast (from North Point to The start of Hobbs Beach), 27 were on the South Side (The Wharf to Hobbs beach), and four on the South East side (East of the Wharf and ferry landing).

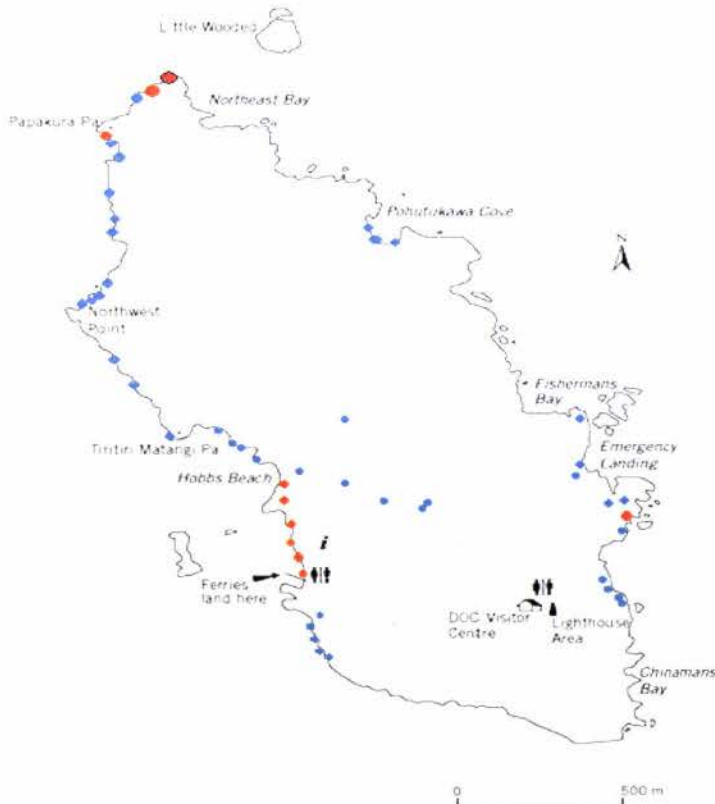


Figure 2.5. Diagram of Little Blue Penguin nest sites found on Tiritiri Matangi Island, New Zealand during 2005/06 breeding season. Blue dots represent single nests while red dots represent sites with ≥ 4 nests within the area.

2.5.4 Egg Laying

Egg laying began on the 9th of September 2005 and continued until late December (Figure 2.6). The median lay date was calculated as the 31st of October. The month of laying for any year was taken as the month that laying was initiated. The mean SST (\pm SE) for the month of laying for each year ($n = 6$) is 14.68 ± 0.22 °C compared to the minimum 14.3 ± 0.21 °C of the corresponding year ($n = 6$) and maximum 21.22 ± 0.5 °C ($n = 6$) SST for the same year (Figure 2.7). The mean SST for the month of laying (14.68 ± 0.22 °C) for any year is on average lower than the months before laying (14.85 ± 0.49 °C) and after laying (15 ± 0.24 °C) (Figure 2.8).

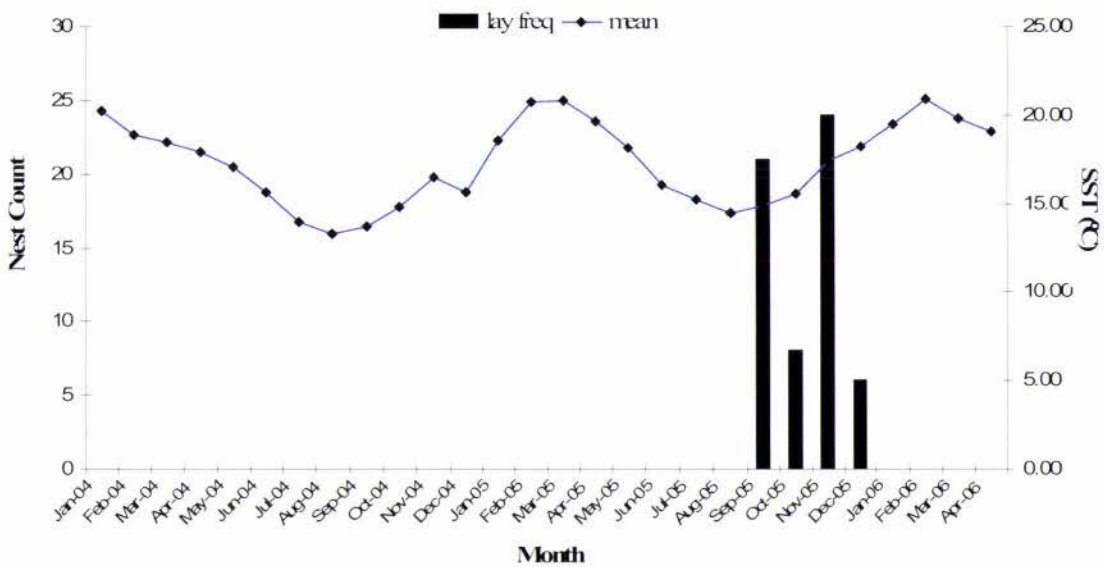


Figure 2.6. Little Blue Penguin nest counts on Tiritiri Matangi Island, New Zealand, according to the monthly lay date and the corresponding sea surface temperature (SST) averaged per month for 2004 and 2005.

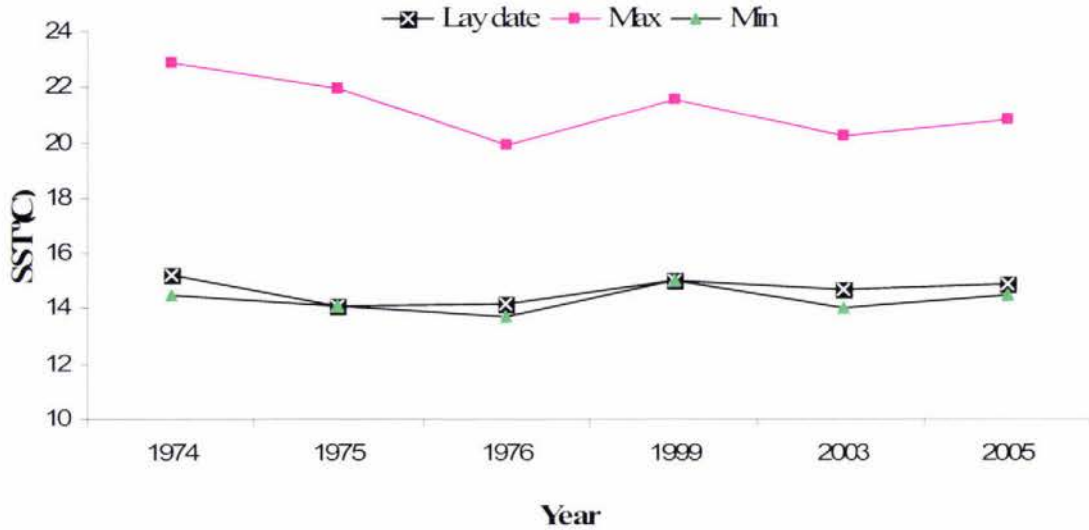


Figure 2.7. Little Blue Penguin lay date on Tiritiri Matangi Island, New Zealand, for six years in relation to the maximum and minimum monthly average sea surface temperature (SST) for the corresponding year. Data for months of laying are from 1974 – 1976 Jones (1978), 1999 Miyazaki and Waas (2003), 2003 Chen (2004) and this study 2005/6.

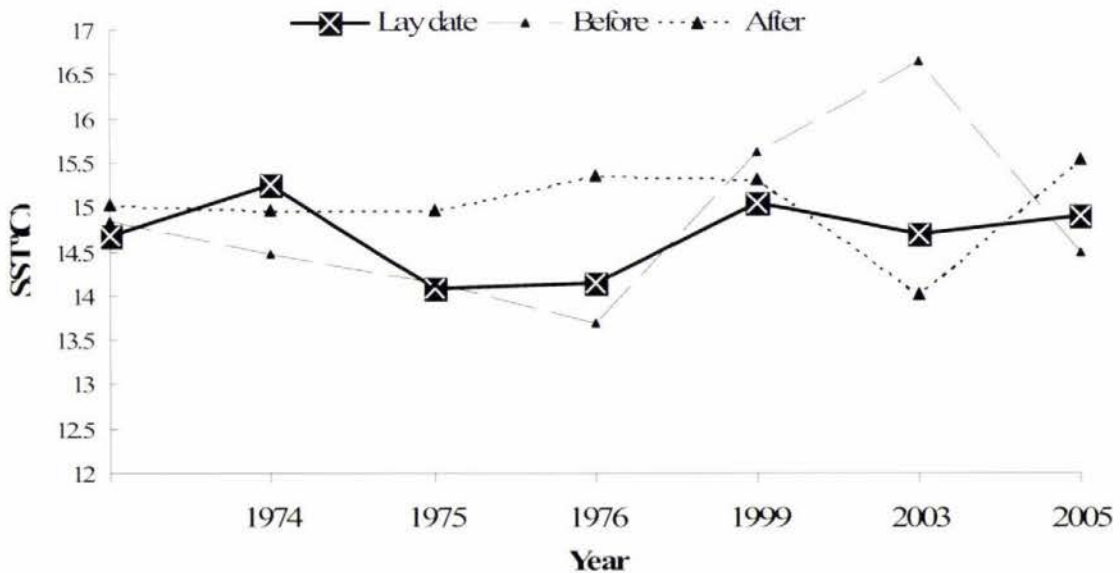


Figure 2.8. The monthly average sea surface temperature (SST) corresponding to the month of Little Blue Penguin egg laying for each year on Tiritiri Matangi Island, New Zealand, compared to the monthly average before and after the laying month.

The lay dates showed two peaks (Figure 2.9) Based on the median lay date the periods correspond to early (1st September to 31st October), and late (1st November to 31st December). Forty of the 74 nests with known lay dates occurred early and 34 occurred late. Of the 87 breeding attempts, ten pairs of individually marked birds laid two clutches as replacement clutches and another six were re-layed were thought to be the same individuals however this couldn't be confirmed. These were identified as 'Replacement Double Brooders' in analysis. Breeding attempts were found at different stages; Nest building n = 56 (64.37%), incubation n = 17 (19.54%), guard stage n = 8 (9.2%) and abandoned n = 12 (13.79%).

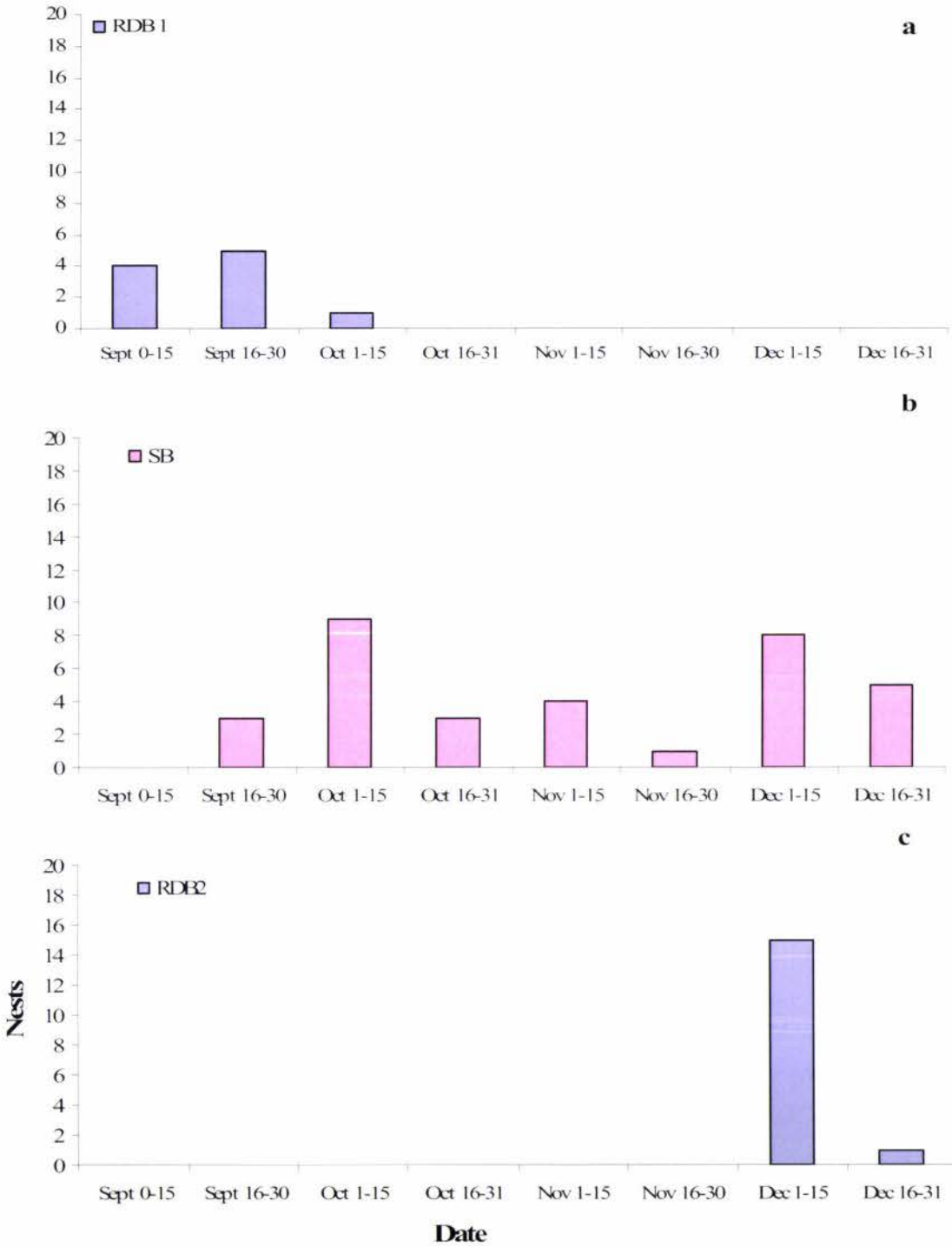


Figure 2.9. Little Blue Penguin lay dates associated with a) replacement double brooding (RDB) LBP and b) single brooding (SB), with replacement clutches of the c) double brooding birds (RDB) being laid during the second peak of breeding, on Tiritiri Matangi Island, New Zealand.

2.5.5 General nest types

The proportions of nest types used during the 2005/06 breeding season showed that the most common nest substrate was; rock 55% (n = 46), then earth 30% (n = 25), artificial (nesting boxes) 7% (n = 6) and tree 7% (n = 6). More rock nests were occupied during the early breeding period these were the most common nest type used overall (Figure 2.10).

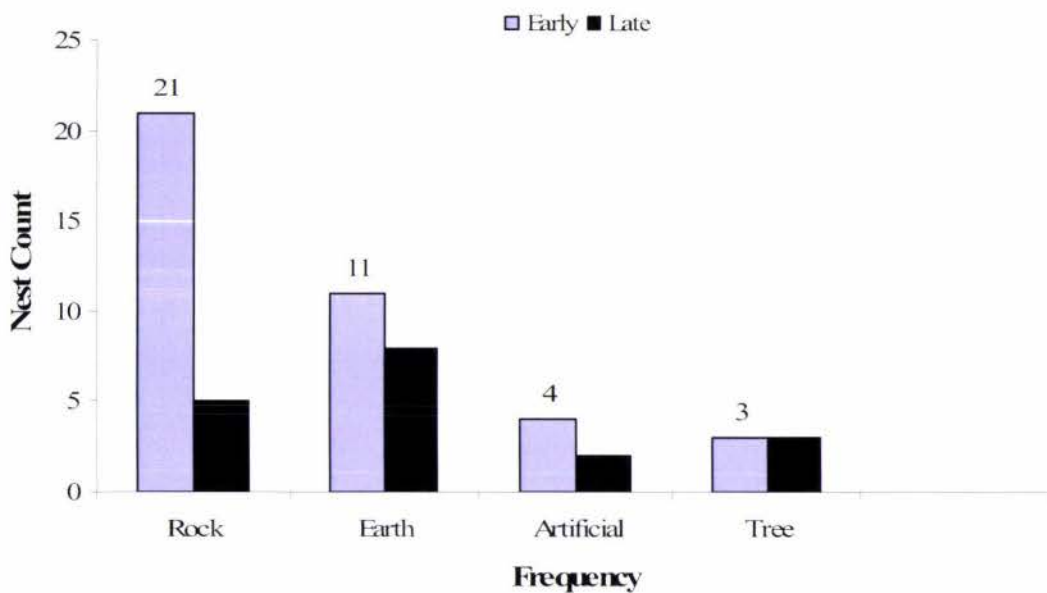


Figure 2.10. Comparison of the nest types used by Little Blue Penguin on Tiritiri Matangi Island, New Zealand, during the early and late periods of 2005/06 breeding season.

2.5.6 Breeding Success

A total of 2685 nest days (incubation days = 1888, nestling days = 797) were included in the analysis using Mayfield's method. This resulted in a chance of survival at incubation of 35% (1 - 53/ 1888), and at nestling stage of 0.4% (1 - 14/ 797). Hatching rate (eggs hatched) was 0.35, meaning 65% of eggs failed to hatch. A modified version of this, which excluded nests that were not followed from the start of egg laying, found 68 % of eggs failed to hatch. The total success rate is 8%. For comparison, calculations of reproductive success were 10%, with a 28% chance of fledging a chick (Table 2.6).

Table 2.6. Figures associated with the calculations of Little Blue Penguin breeding success. Where possible figures were compared with past years of Little Blue Penguin breeding on Tiritiri Matangi Island, New Zealand, Jones (1979) is marked with an asterisk, Miyazaki and Waas (2003) marked with a ^Δ and (Chen 2004) marked with double asterisks.

| | Years | | | | | |
|-----------------------------|-----------|-------|-----------|----------|-------------------|----------|
| | 2005 | 1974* | 1975* | 1976* | 1999 ^Δ | 2003** |
| Initiation of laying | 9-Sep-05 | | 17-Aug-75 | 2-Sep-76 | 25-Aug-99 | 4-Jul-03 |
| Median laying date | 31-Oct-05 | | | | | |
| Nests | 87 | | 34 | 34 | 33 | 7 |
| Nests laid early | 43 | | | | | |
| Nests laid late | 38 | | | | | |
| Eggs failed to hatch | 102 | | | | | 12 |
| Eggs hatched | 60 | | | | | 2 |
| Hatching success | 27% | | | | | 21% |
| Number of chicks per nest | 17 | 0 | 0 | 0 | 0 | 0 |
| Fledging success | 28% | | | | 0% | 0% |
| Breeding success | 10% | 0% | 0% | 0% | 0% | 0% |
| Nest success | 19% | | | | | |
| Nests failed at chick stage | 14% | | | 15 | | |
| Nests failed at egg stage | 53% | | | 19 | | |

The only parameter influencing the outcome of hatching and fledging was lay date (Table 3.1). Early and late lay dates had significantly different success rates (Fisher's exact test, $p = 0.0013$, $n = 81$). A chick was more likely to survive if it was laid early within the breeding season (September to end of October) than if it was laid after this time. All 17 chicks that fledged came from nests that were laid early. The stage that failure occurred was also significantly different for eggs and chicks (Fisher's exact test, $p = 0.046$, $n = 71$).

Table 2.7. Categorical data modelling was conducted for the probability (%) of Little Blue Penguin success for hatching or fledging responses on Tiritiri Matangi Island, New Zealand. *P*-values marked with an asterisk is significant. Nest type is classified as artificial (A), earth (E), rock (R), and tree (T).

| Variable | | n | Response | | | | | | | |
|----------------------|-------|----|----------|--------|----|--------|----------|--------|----|---------|
| | | | Hatching | | | | Fledging | | | |
| | | | % | Chi-sq | df | P | % | Chi-sq | df | p |
| Lay Date | Early | 46 | 37 | | | | 22 | | | |
| | Late | 40 | 40 | 0.08 | 1 | 0.7723 | 1 | 4.47 | 1 | 0.0346* |
| Nest Type | A | 8 | 13 | | | | 6 | | | |
| | E | 25 | 44 | | | | 4 | | | |
| | R | 45 | 44 | 4.68 | 3 | 0.1966 | 18 | 2.76 | 3 | 0.4303 |
| | T | 8 | 13 | | | | 13 | | | |
| Nest Distance | Close | 51 | 37 | | | | 12 | | | |
| | Far | 35 | 40 | 0.07 | 1 | 0.7971 | 11 | 0 | 1 | 0.9619 |

2.6 Discussion

During 2005/06 on Tiri, LBP had a very low breeding success of only 10% (0.2 chicks per pair) and 28% fledging success. Jones (1978) also found a high number of nest desertions (56%) during 1974-1976 with no broods being successful. Chen (2004) found 100% nest desertion in 2002/03 although this was from a small sample size. Levels of breeding success have been identified by Dann *et al.* (2000) as high (> 1.2), average (0.7 to 1.2), and low (≤ 0.7) chicks per pair for Australian populations of LBP. Using this ranking the North Island sub-species has a particularly low breeding success compared to LBP in Otago, New Zealand and Australia which fledge 1.07 chicks in average years (Robinson *et al.* 2005). The reason for the low success rate on Tiri is unknown.

There is an increasing awareness of the large variability of breeding success in LBP within and between populations. LBP in Victoria, Australia, and Otago, New Zealand have some of the highest LBP productivity rates compared to other regions. To achieve a stable population, parameters such fledging success and recruitment into the population must balance mortality and emigration. Currently it is unknown whether 2005/06 on Tiri was a normal year due to the lack of comparisons with other years. Compared to other studies conducted on Tiritiri Matangi this was better than past years however this could be a function of sample size and sampling area. Only with longer term monitoring of this population can such trends be identified.

Within the population on Tiri the most influential parameter for 2005/06 breeding season was the initiation of breeding (lay date). All successful chicks came from single brooding pairs laying early within the season with breeding success decreasing as the breeding season progressed. This pattern has also been found in many

other bird species (Price *et al.* 1988; Perrins 1996; Arnold *et al.* 2004; Benowitz-Fredericks and Kitaysky 2005).

Two hypotheses have been proposed for why reproductive success declines with lay date: 1) the timing hypothesis, and 2) parental quality hypothesis (Arnold *et al.* 2004). The timing hypothesis suggests that reproductive success declines due to factors associated with the date of laying such as a change in environmental conditions reduced breeding synchrony, and/ or parental restraint with late hatched chicks (growth) (Hatchwell 1991; Monero 1998; Arnold *et al.* 2004). The parental quality hypothesis suggests that it is due to the parental quality and age of the birds that there is a progressive decline in success as younger less experienced birds will tend to lay late within the season (Parsons 1975; Hatchwell 1991; Brinkhof *et al.* 1993; Arnold *et al.* 2004).

Black Kites (*Milvus migrans*) lay earlier within the season as a response to an increase in the spring SST (Sergio 2003). If food availability is high, then the body condition of the adults will increase and the result may be earlier laying (Newton 1979, 1998; Drent and Dann 1980; Meijer *et al.* 1989; Sergio 2003). Indeed, better individual body condition has also been associated with earlier mating (Miyazaki and Waas 2003b) and laying dates (Robinson *et al.* 2005) in LBP. Chicks that hatch early within the season may experience better growing conditions if food abundance is high (Schekkerman *et al.* 2003; Tulp and Schekkerman 2006) and when adult body condition is good more energy can be directed towards provisioning and caring for chicks.

Earlier laying by older individuals may also be a function of intrinsic physiological differences. Older European Starlings (Dawson 2003), Snow Geese (Hamann and Cooke 1987), and Sparrow Hawks (Newton *et al.* 1981; Newton and Rothery 1998) all lay earlier than younger birds regardless of condition (Newton *et al.*

1981; Hamann and Cooke 1987; Dawson 2003). Nonetheless, both physiological and behavioural (experience) differences can occur between younger and older birds. It has been found that testicle size, testosterone concentrations, and cloacal protuberance increases with age (Evans and Goldsmith 2000). Male Wrens lay earlier due to larger testicle size (Evans and Goldsmith 2000). European Starlings were also found to lay early due to testicular maturation being more advanced earlier in the breeding season than younger birds (second year breeding) (Dawson 2003). Lastly, studies on the Common Tern have found that chicks are able to be raised successfully when hatched during late breeding periods if they are cared for by quality parents that are able to forage better and provide better parental care (Arnold *et al.* 2004). This is only possible if food availability is high during the course of the breeding season (Arnold *et al.* 2004).

Distinguishing between the timing and parental quality hypothesis requires known aged individuals and longer term breeding information. In this study it is unlikely that both hypotheses are true (discussed further in Section 2.6.3).

2.6.1 Lay Date and Environmental Conditions

The 2005/06 breeding season covered a period of considerable climatic fluctuations. During the early phase of breeding the weather was stormy and cold, with periods of high wind speeds and rainfall. Later in the breeding season the weather was less variable with less rain fall and warmer air temperatures due to summer. During the early period major storms were associated with adult mass mortalities. Three nests were found to have one of the known breeding pairs dead outside the nest. These nests were deserted shortly after. During summer, the warmer water can cause a reduction in the phytoplankton and therefore a decrease in planktivorous feeding species, something that is likely to influence higher trophic levels.

The first lay date for 2005 occurred during periods of low SST for the year. SST has been considered an important determinant of food availability since it is associated with increases in primary productivity with implications for higher levels of the food web. Information from other years shows that lay date is associated with annual minimums in SST (mean 14.5 - 15°C). In contrast, hatching success of LBP breeding in South East Australia over a 30 year period is not associated with either SST or SOI but lay date is still a factor associated with success (Kemp and Dann 2001). However, other species (e.g. Sooty Shearwaters *Puffinus griseus*) have been found to be influenced by SST and SOI since it is thought that these climatic perturbations influence food availability (Lyver *et al.* 1999).

For LBP on Tiri the variability in the lay dates between years suggests lay dates are a response not to an increase in light levels (longer days) but to some more variable factor. Evidence for this is that LBP shifted the initiation of breeding as much as two months between years and this coincided with the period of low annual SST. The onset of the 2005 breeding season was the 9th of September. The earliest lay date in the sample available occurred in July (Chen 2004; Jones 1978) and the latest in September (Jones 1978). The actual minimum temperatures for those years had little variability however the month when this minimum occurred differed between years. The plasticity of lay dates within LBP has been documented for both Australian and New Zealand populations (Stahel and Gales 1987; Robinson *et al.* 2005). Clearly given the relationship between increased productivity and low SST, LBP are timing their onset of breeding with peak productivity levels for a given year. Thick-billed Murres (*Uria lomvia*) are also found to have high inter-annual variability of life-history traits such as median lay dates and egg size which is thought to be a response of phenotypic plasticity to variable environmental conditions (Hipfner *et al.* 2005). There is a need for a greater

understanding on the biology of prey species and the direct effect that climate has on the availability and abundance of prey species.

2.6.2 Food Availability and Body Condition

The variability in lay dates may be due to LBP attempting to breed when the conditions are most favourable. Penguins feed exclusively in the marine environment (Lack 1968; Robinson *et al.* 2005). If there is an increase in prey availability then a short-term increase in body condition could initiate breeding (Robinson *et al.* 2005). However if this body condition is unable to be maintained then the adults will struggle at the chick stage. Foraging is linked to breeding success but requires a balance between chick provisioning and adult self maintenance. The ability to obtain enough energy will be affected by individual quality (body weight, sex and age) and food availability. Food availability will vary with SST, climate changes including local weather conditions and events, and prey type and levels of competition (including human take of marine resources).

LBP and other seabird species (e.g. Wandering Albatross *Diodmedea exulans*, Lesser Black-backed Gull *Larus fuscus*: Nagar *et al.* 2003) are central place foragers during the breeding season returning to the nest to provision chicks (Green *et al.* 2002). This is problematic if the adults have to forage further distances due to local depletion in food supply. Mattern (2001) found that the longer the foraging trip of LBP, the increased likelihood that the nest would fail. Therefore the foraging ground and the availability of prey items will influence the ability of adults to raise their chicks (Tremblay and Cherel 2005). This was compared for two regions which were found to vary depending on the productivity levels at each site (Mattern 2001). Michelson *et al.*

(1992) found that the increased fish abundance in Bass Strait, Australia, was associated with an earlier breeding season and heavier chick body weight (Chen 2004).

LBP are known to be generalist feeders changing preferences with availability of prey (Montague and Cullen 1988). Pilchard (*Sardinops sagax*) and Anchovy (*Engraulis australis*) are major prey sources, but these species are also associated with large commercial catches and die-off events (Norman *et al.* 1992). A study on Rockhopper Penguins (*Eudyptes chrysocome*) (the second smallest penguin species) found that they require 211 Kjd-1 during the first week of breeding with 5.5 times more halfway through, with a total of 59, 000 kj (Brown 1987; Tremblay and Cherel 2005). Pilchards are considered a high-energy source (Batchelor and Ross 1982; Kirkham *et al.* 1985; Berruti *et al.* 1993; Bunce and Norman 2000), and fish in general are considered better quality than cephalopods and crustaceans. Therefore a change in diet could impact on nutritional requirements and foraging effort.

Prey abundances can fluctuate within and between seasons and may explain the annual differences in LBP success. LBP may be breeding earlier as a response to environmental conditions and an increase in food availability. Nonetheless in any year some individuals do breed later. Johannesen *et al.* (2002) found a positive correlation of reproductive output and resources for LBP suggesting that individuals vary in access to more or better quality resources. The ability to dive and access prey species is a function of predator size. Larger animals may exhibit greater individual fitness and while size is a hereditary component (Miyazaki and Waas 2003), it can also be related to age.

2.6.3 Age, Quality and Reproductive Success

Breeding experience has been linked to early laying and higher reproductive success (Agnew and Perriman 2004). Johannesen *et al.* (2002) found evidence that the

likelihood of double breeding in LBP increases with age. Dann and Cullen (1990) found that penguins breeding earlier had greater fledging success than those breeding late. This was also found by Miyazaki and Waas (2003) and for LBP in this study. Previous studies have identified the LBP on Tiri as single breeders, only laying one clutch per year (Jones 1978; Miyazaki and Waas 2003). This was generally found during 2005/06, however ten known pairs were identified as attempting double breeding (replacement clutches after first clutch failed). This finding was likely due to this study sampling a much larger portion of the population than previous work. Though actual DB (raising two clutches) has only been witnessed in New Zealand for the Otago population (Perriman and Steen 2000). To achieve successful DB individual pairs need to lay very early within the year, since raising two clutches to fledging requires seven months (Johannesen *et al.* 2002).

The age of LBP on Tiri is generally unknown and so age related analysis could not be incorporated into the breeding success of this study. All breeding pairs associated with nests that fledged were SB and early. Due to the level of monitoring it is unlikely that these nests were replacement clutches of DB when the first clutch was missed in the monitoring. Monitoring began at the end of June, and ongoing monitoring of some banded SB pairs showed that they didn't breed until late. In addition, at least three of the nests that fledged chicks were from previously banded birds from other burrows monitored before nesting occurred.

Life history theory predicts that time and energy given to current reproductive effort versus future varies with age. In a clutch manipulation study on Lesser Black-backed Gulls results found evidence for interbrood trade-off between current egg production effort and future fitness (Nagar *et al.* 2001). A study on common Goldeneye Ducks (*Bucephala clangula*) controlled for age and breeding experience and found that

females will lay later and smaller clutches in the following year if they reared a brood in the previous year (Milonoff *et al.* 2004). Potentially those DB pairs could be older birds that are increasing their current reproductive effort due to low chances of surviving to breed the next year. An alternative to this could be that those fledging the chicks are from older parents that are more experienced, or from younger birds that are of better quality. Laying replacement clutches is costly to parents and may result in lower quality eggs than the first laid eggs (Arnold *et al.* 2004).

LBP laying replacement clutches, suggests that 1) either food was available later in the season, 2) birds were older with a less chance to lay in future years, or 3) of better quality and could obtain enough food to relay. Any inference of success and laying as a function of age and reproductive output can only be speculated for the Tiri population and has not been quantified for this study. This shows the importance of long-term studies and effective sampling to ensure overall extrapolation of the ecological parameters that can influence breeding success.

Furthermore, double breeding is a weak inference of individual quality as it can be confounded with environmental effects (Johannesen *et al.* 2002). The number of double brooding birds in Oamaru decreases as a response to a delay in the onset of breeding and results in a lower number of chicks fledging (Lalas *et al.* 2004). The shift in breeding was found to be a response to an increase in warmer-water associated with *La Niña* event (Lalas *et al.* 2004).

2.6.4 Nest Type and Site Fidelity

Like Yellow-eyed Penguins *Megadyptes antipodes* (Ratz *et al.* 2004), LBP return to the same natal site to breed, hence potentially limiting nest site and mate choice availability. A less important predictor of breeding success on Tiri is nest type although more chicks

were found to fledge from rock and earth nests than artificial and tree. Miyazaki and Waas (2003) found that larger male penguins on Tiri were associated with the low to mid altitude nests and suggested that these were potentially better breeding sites. These sites are relatively close to the sea and food resources (Miyazaki and Waas 2003). The current study did not find any significant effect of nest distance or nest type on breeding success in LBP. Though nest types and nest microhabitats have been found to influence breeding success in other species and populations of penguins (Chapter 3).

Other bird species have been reported to give up a nest site if they were unsuccessful in past breeding attempts (see Nager *et al.* 1996, 2003). For example, Lesser Black-backed Gull females have been found to emigrate from a nesting location causing mate infidelity (Nager *et al.* 2003). Nest fidelity in LBP is also considered to be more likely to increase if individuals have been successful in previous attempts (Bull 2000). Jones (1978) found low mate fidelity in LBP on Tiri and suggests this is due to the high mortality in this population, but found breeding site tenacity is strong. In contrast, Agnew and Perriman (2004) found that mate fidelity in LBP is more important than nest fidelity. Pair infidelity due to dispersal will result in new pair bond formation, which causes an initial decrease in breeding success in several species (Coulson 1966; Davies 1976; Ollason and Dunnet 1978; Greenwood and Harvey 1982; Bradely *et al.* 1990; Johnson and Gaines 1990; Pärt 1991; Mills *et al.* 1996; Nagar *et al.* 2001). If LBP on Tiri show high levels of dispersal from the natal site rather than the expected pattern of philopatry, then they could experience a reduction in breeding success due to new breeding pairs.

2.6.5 Baseline Data

2.6.5.1 Body Condition

On average, male LBP on Tiri were approximately 50 g larger than females, a smaller difference than previous studies that found a difference of around 100 g. This could be explained by sub-species differences (Kinsky and Falla 1960; Renner 1998). Body size can vary during the seasons and care needs to be taken since juvenile males can be misclassified as females based on small size. For example the differences in body condition between males and females can be associated with the differences in metabolic demands due to females requiring increased levels of fat for egg production (Johnson *et al.* 1985; Hocken 2000). The demands of breeding in conjunction with food availability (Chapter 4) on LBP body mass were likely to have resulted in the lower body condition of LBP during 2005/06 breeding season. Body condition will influence survival and could be a useful tool in monitoring marine productivity if gender and individual differences are taken into account.

The fledging weight of chicks from other studies is approximately 1000 g, (Perriman *et al.* 2000) though in this study chicks fledged at weights close to half this value (minimum 557 g). Because penguin species have been found to have differences in growth rate, fledging weights may be a good predictor of food availability. Since the weights are highly variable this could be related to parental provisioning differences and/or the timing of hatching (Chapter 2).

Given that weights overlapped between the sexes, weight alone is a poor field measure of sex. An alternative simple measurement of beak depth (BD) and depth at the gonys (BD2) can be taken within the field is needed and should be verified by molecular analysis. Molecular analysis is commonly used since it has high accuracy and

is easily obtainable by taking a feather from each bird upon handling (e.g. banding). This will further decrease the physiological stress on the bird as handling is minimised.

Body size has also been a part of sub-species classification within LBP (Kinsky and Falla 1960). However classification of sub-species requires a combined approach incorporating biological and ecological factors as well as genetic analysis. A combined approach would enhance management efforts since different populations of sub-species will vary spatially and temporally depending on the environmental factors that they experience within any area.

2.6.5.2 Banding and Baseline Data

Within this study only 35 LBP were found previously banded and two were recovered dead during this study, with four newly banded birds being recovered in other regions. Jones (1978) found that dead banded birds were discovered up to 290 km away from where they were originally banded. This meant that penguins from Tiri could potentially reach areas such as Whangarei, Cape Karikari, and North Cape. Jones (1978) also found that there was a high loss of banded birds from Tiri within the first year with only 26% coming ashore two years from banding. Unless birds are banded LBP found dead along the Tiri coast cannot be assumed to all have originated from Tiri population.

Longer monitoring of banded birds is required to assess survival rates, population size, dispersal, recruitment, mortality levels and mate and site fidelity. These characteristics are important to quantify when considering the effects that mass mortalities may have on a species, especially a monogamous and highly philopatric species such as LBP. Furthermore, these data will aid conservation management since threat classifications are based around population numbers.

An aim of this study was to provide strong baseline data with minimum disturbance to each nest. This meant that there were some limitations to banding and disturbance of breeding pairs. Banding on the nest was conducted during chick rearing stages to reduce stress and minimise abandonment unless the adults were banded when they were coming ashore. Birds were removed from nests for identification or banding only when visual checks were not productive.

Initial monitoring of survival requires general baseline data, which includes information on ecological life history traits, population recruitment, mortality, and movement. To achieve this goal, parameters such as age and gender must be recorded. In the field there is no way of aging a live penguin. Banding is the best form of individually assigning a number to an individual which will allow for re-sighting over long and short-term monitoring periods. Where and when you re-sight an individual can help with identifying a species short-term movements over seasonal life stages (e.g. breeding) to compare within and between years. Mortality differences can relate to age and sex (Jones 1978) therefore banding chicks near fledging can help with aging individuals and enhance investigations on the cause of death.

2.6.6 Considerations

Banding and individual identification has been an important component for effective monitoring during this breeding season since it allowed a more comprehensive understanding of nesting activities. Identifying an individual allows for a detailed approach on individual behaviours, but more importantly it meant limiting disturbance and handling of each LBP. Consideration and care needs to be given to external attachment of tags (such as stainless steel flipper bands) as initial handling will incur a cost of stress to the bird. No attempt was made to identify the potential impacts that the

bands had on the LBP. Upon handling of birds no sightings of harmful side effects of bands were found. Recommendations for future monitoring such as mark/recapture counts of the population could also monitor band condition and quantify the effect that they could have. This is important since flipper bands can cause feather abrasion and serious injuries (i.e. death) if they are not applied well or are of poor design (Sallaberry and Valencia 1985; Renner 1998). Within this study there was no evidence to suggest that a bird was harmed by a band, however old bands were found to open which could potentially cut the bird. Furthermore, due to the loose fit of the tag there is the risk of attachment to external objects which could also harm the bird and threaten survival. Potentially, external banding tags could cause an increase in energy expenditure by the bird since tags have been reported to cause up to 24% additional drag when tested for swimming in an artificial channel (Culik *et al.* 1993; Renner 1998). Where possible future identification methods should consider calculating the impact of bands and consider devices such as transponders which are implanted under the skin. Due to financial reasons this was not an option within this study.

2.6.7 Conclusions

There are many factors involved in the breeding success of LBP, and only considering a few of them does not allow for consideration of interactions between factors (Knight and Rogers 2004). To understand the fitness consequences of breeding at different times requires an understanding of the environmental conditions experienced at the time chick's hatch and the optimal times of breeding (Arnold *et al.* 2004). LBP display large variations in breeding success within and between populations. This variation could be due to colony specific factors (weather and burrow substrate) (Perriman and McKinlay

1995), and other factors such as food availability (Cullen *et al.* 1992) and climate changes.

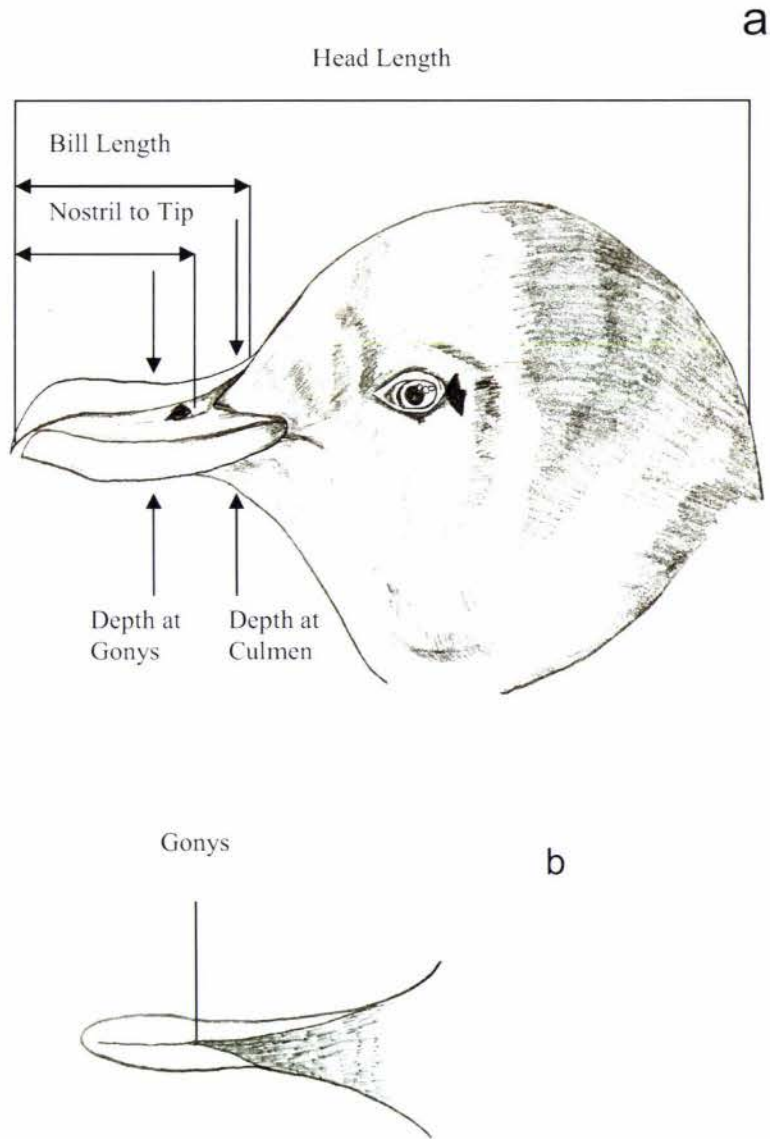
Breeding variation between years on Tiri is extreme with large numbers of nest desertions and low breeding success occurring in some years. The major correlate of this is the lay date with more successful nests laid early in the season. Initiation of breeding was associated with low yearly SST (based on six years of known lay dates). This suggests that future monitoring should take into account connection between the SST and lay date. Monitoring could include several aspects such as food availability and biology, LBP adult body condition and the effect of this on chick weight and survival.

The delay in the onset of breeding for 2005 meant that the breeding season overlapped with the moulting period (5th January 2006) (see Chapter 5). Moulting initiation appears to be less flexible in the timing of onset. At least five nests were found to be active when the adults tending them began their moult. The overlap of these two behaviours has extreme consequences as the adults were not able to head out to sea to forage to provision chicks due to their plumage not being water proof and poorly insulated (see Chapter 4). Failure of these nests is therefore not surprising since both chick rearing and moult are energetically demanding behaviours for penguins and not compatible.

Future research should investigate questions such as 1) why breeding success on Tiri is so low compared with other Little Blue Penguin populations?, 2) is SST related to productivity and early breeders being more successful?, and 3) whether early breeders are in better body condition (weight, size, age) than later breeders?. Lastly, further investigation should encompass the dispersal and distribution of Little Blue Penguins within the Hauraki Gulf and whether emigration and immigration has an impact on breeding.

2.7 Appendix

2.7.1 Morphological measurements from Little Blue Penguin



Schematic diagram of the morphological measurements taken from LBP a) left side view of the head and b) ventral view of the beak. Redrawn from Jones (1978).

2.7.2 Little Blue Penguin band numbers

Band numbers allocated to individual penguins or old bands re-sighted during the 2005/06 monitoring period. All birds were banded as adults or sub-adults unless marked with chick. Those with a * are known to be dead.

| Band ID | Weight | Gender | Age | Old bands | Weight | Gender |
|----------|---------|---------|-------|-----------|--------|----------|
| P38 361 | 1000 | Female | | | | |
| P38 362 | 975 | Female | | P30 594 | | Male |
| P38 363 | 960 | Male | | P30 611 | | |
| P38 365 | 1075 | Female | | P30 615 | | |
| P38 368 | 1125 | Male | | P30 619 | | |
| P38 369* | 1350 | Male | | P30 624* | | 29/10/05 |
| P38 370 | 950 | Male | | P30 655 | | |
| P38 371 | 850 | Female | | P30 678 | | |
| P38 372 | | Female | | P30 684 | 850 | |
| P38 373 | 732.64 | Female | | P30 685 | | Unknown |
| P38 374 | 657.64 | Female | | P30 692 | | |
| P38 375 | 807.64 | Male | | P30 721 | 832.64 | Unknown |
| P38 375 | 807.64 | Male | | P30 724 | | |
| P38 376 | | Female | | P30 745 | | Unknown |
| P38 378 | 507.64 | Female | Chick | P30 752 | | |
| P38 379 | 857.64 | Male | | P30 756 | | |
| P38 380 | 557.64 | Male | Chick | P32 383 | | |
| P38 535 | 775 | Male | | P32 422 | | |
| P38 803 | | Male | | P32 427 | | |
| P38 804 | | Female | | P32 453 | | |
| P38 805 | 822.71 | Female | | P32 458 | | Unknown |
| P38 806* | 679.24 | Female | | P32 474 | 1400 | Male |
| P38 807 | 1022.71 | Male | | P32 498 | | Unknown |
| P38 808 | 729.24 | Male | | P32 529 | 750 | Male |
| P38 809 | 972.71 | Female | | P32 530 | | Unknown |
| P38 810 | 832.64 | Male | Chick | P32 531 | | Unknown |
| P38 812 | 628.55 | Female | | P32 533 | | |
| P38 813 | 857.64 | unknown | | P32 535 | | |
| P38 814 | 957.64 | Male | | P32 538 | 850 | Female |
| P38 815 | 857.64 | Male | | P32 540 | | |
| P38 816 | 857.64 | Male | | P32 576 | 738.65 | Female |
| P38 817 | 907.64 | Female | | P32 583 | | |
| P38 818 | 1032.64 | Male | | P32 593 | 850 | Unknown |
| P38 820 | 847.71 | Female | | P32 594 | | |
| P38 821 | 829.24 | Male | | P32 613 | | |
| P38 822* | 822.71 | Female | | P32 656 | | |
| P38 823 | 879.24 | Male | | | | |
| P38 824 | 997.71 | Male | | | | |
| P38 825 | 1029.24 | Male | | | | |
| P38 826 | 1033.4 | Male | | | | |
| P38 827 | 883.4 | Female | | | | |
| P38 829 | 933.4 | Male | | | | |
| P38 831 | 829.3 | Female | | | | |
| P38 834 | 779.3 | Unknown | | | | |
| P38 835 | 1029.3 | Male | | | | |
| P38 837 | 733.4 | Male | | | | |

| | | | |
|----------|---------|---------|-------|
| P38 838 | 829.3 | Male | |
| P38 841 | 879.24 | Male | |
| P38 842 | 829.24 | Female | |
| P38 843 | 779.24 | Male | |
| P38 843 | 779.24 | Male | |
| P38 845* | 729.24 | Female | |
| P38 847 | 779.24 | Male | |
| P38 848 | 731.25 | Male | |
| P38 849 | 1029.24 | Male | |
| P38 850 | 1125 | unknown | Chick |
| P38 851 | 864.83 | Female | Chick |
| P38 852 | 779.24 | Female | |
| P38 853 | 989.83 | Male | Chick |
| P38 854 | 972.71 | Male | |
| P38 855 | | Female | Chick |
| P38 856 | 1000 | unknown | Chick |
| P38 857 | | Male | Chick |
| P38 858 | 885.55 | Female | |
| P38 860 | 1022.71 | Male | |
| P38 861 | 822.71 | Female | |
| P38 862 | 1079.24 | Male | |
| P38 863 | 1029.24 | Male | |
| P38 864 | 979.24 | Female | |
| P38 865 | 972.71 | Female | |
| P38 866 | 829.24 | Female | Chick |
| P38 867 | 929.24 | Male | Chick |
| P38 868 | 907.64 | Male | Chick |
| P38 869 | 857.64 | Male | Chick |
| P38 870 | 957.64 | Female | |
| P38 870 | 950 | Female | |
| P38 871 | 929.24 | Male | Chick |
| P38 873 | 707.64 | unknown | Chick |
| P38 874 | 1104.24 | Female | Chick |
| P38 875 | 872.71 | Unknown | |
| P38 877 | 1122.71 | Male | |
| P38 878 | 829.24 | Male | |
| P38 879 | 844.24 | Male | |
| P38 880 | 822.71 | Female | |
| P38 883 | 740.9 | Male | |
| P38 884 | 772.71 | Female | |
| P38 885 | 972.71 | Male | |
| P38 886 | 754.24 | Male | |
| P38 887 | 772.71 | Female | |
| P38 888 | 849.24 | Female | |
| P38 889 | 1229.24 | Female | |
| P38 890 | 972.71 | Male | |
| P38 891 | 872.71 | Male | |
| P38 892 | 979.24 | Female | |
| P38 895 | 1097.71 | Female | |
| P38 896 | 1054.24 | Female | |
| P38 897 | 722.71 | Female | |
| P38 898 | 804.24 | Male | |
| P38 900 | 897.71 | Male | |
| P38 965 | 765 | Female | |

| | | |
|----------|-----|---------|
| P38 967 | 706 | Female |
| P38 969 | 660 | unknown |
| P38 972 | 585 | Female |
| P38 973 | | Unknown |
| P38 974 | 780 | Male |
| P38 979* | | |

CHAPTER 3 Causes of egg and chick mortality in Little Blue Penguins on Tiritiri Matangi Island



Plate 3.1. Little Blue Penguin chick on Tiritiri Matangi Island. Photo by J.Geurts.

3.1 ABSTRACT

Little Blue Penguin (*Eudyptula minor*) hatching success, fledging success and nest failure on Tiritiri Matangi Island, New Zealand were studied over the 2005/06 breeding season. Eighty-seven nesting attempts were intensively monitored to establish the likely cause of death at both the egg and chick stage. Laboratory analysis of egg fertility and necropsies of dead nestlings were conducted to identify the likely cause of death of 77 failed nests. Nest desertion was the main cause of nest failure and was associated with poor timing of parental care change of duty during the egg stage and adequate provisioning of chicks by parents during the chick stage. Poor weather conditions were directly and indirectly associated with egg failure early in the breeding season. Low food availability later in the breeding season was found to be the most influential determinant of success at the chick stage. Nest desertion resulted in failure at the egg stage via death of the embryo due to sub-optimal incubation temperatures. Although early embryo death was the primary cause of egg failure, adults often continued to incubate these dead eggs. During early chick rearing (< 10 days) desertion caused death by hypothermia and starvation, but the death of older chicks (> 10 days) was associated with starvation of chicks. Regardless of chick age, adults appeared to struggle with chick provisioning and many chicks left the nest early, presumably to search for food. Results suggest that 2005/06 was a very poor breeding season and nest failure was exacerbated by late egg laying and interrelating factors such as climate and food resources.

3.2 INTRODUCTION

Intensive monitoring of nests provides valuable insights into the parameters associated with nest failure and nest success. The cause of failure is likely to vary with nesting stage since eggs and chicks are vulnerable to different risks. Factors that can influence the viability of an egg include initial fertility of the egg, microhabitat (Stohleson and Beissinger 1999), egg properties (Massaro and Davis 2004a), and nest activities (Boersma *et al.* 2004). If a chick survives through incubation (initial development) and hatching, other risks need to be considered. Chicks are prone to direct mortality due to hypothermia (within first few weeks), starvation, chick competition, predation and parasites.

3.2.1 Risks during Incubation

Temperature is considered an important factor associated with egg survival for all avian species due to oviparity. The normal development of avian eggs requires an average temperature of 36°C to 38°C. In captive situations embryos are prone to cooling (Rol'nik 1970; White and Kinney 1974; Wilson 1991; Stohleson and Bessinger 1999) and overheating causing death or abnormal development (Romanoff and Romanoff 1972; Wilson 1991; Deeming and Ferguson 1992). The average temperature requirement is a narrow range to be maintained and it is possible that this range may be lower in the wild (Drent 1975; Webb 1987; Rahn 1991; Stoleson and Beissinger 1999). Eggs are potentially more susceptible to overheating than cooling (Webb 1987; Stoleson and Beissinger 1999) therefore incubating birds maintain optimal and steady temperatures through sensitivity and warmth of their brood patches, as well as by egg turning, and egg position (Massaro and Davis 2004b).

The effect of microhabitat and hatchability is also a function of the properties associated with each individual egg. Properties include pore density and size, egg size (length and width), and egg shell thickness. For example egg development requires respiratory gases and water vapour to move across the shell (Ar *et al.* 1974; Massaro and Davis 2005). In fact an increase in gas exchange is thought to lead to faster embryo development and a decrease in incubation time (Massaro and Davis 2004a).

Within phylogenetic constraints egg size will influence the health of chicks since large eggs contain more nutrients to supply the growing embryo (Nisbet 1978; Arnold *et al.* 2004). More nutrients will mean a larger chick, which has also been associated with faster subsequent growth rates (Williams 1990; 1995; Massaro and Davis 2005). Differences in egg size vary within and between penguin species, for example: LBP lay on average two eggs of equal size while Rockhopper (*Eudyptes chrysocome*), Fiordland (*E. pachyrhynchus*), and Snares Crested Penguins (*E. robustus*) usually lay a second egg larger than the first (Wharham 1974a, 1974b, 1975; Lamey 1990; Massaro and Davis 2005). Egg size is thought to increase with either female age or individual quality (Massaro *et al.* 2002). Older females may be better able to adapt to seasons and different resource availability (Birkhead and Nettleship 1982; Massaro *et al.* 2002), while individual quality (i.e. health, size, behavioural parameters) may influence the input of the female to egg production. This may be associated with food intake (Coulson 2002) and also additional intake of required nutrients for egg formation (Massaro *et al.* 2002). Eggs are made up of calcium carbonate and require an increase in uptake of calcium for egg formation hence the effort taken to do this may vary with each individual. The more calcium that is ingested the thicker the egg shell. This may help decrease the chance of breakage (Massaro *et al.* 2002). Broken eggs can be the result of nesting conditions, adult fights, or rain fall which will decrease reproductive success

(Stokes and Boersma 2000; Renison *et al.* 2002; Boersma *et al.* 2004). Interestingly several seabirds (e.g. Common Murres (*Uria aalge*): Birkhead 1975; and Thick-billed Murres (*Uria lomvia*): Gaston and Hipfner 2000) have been found to reduce their aggression within the nest during periods of incubation as opposed to non-incubating times (Boersma *et al.* 2004).

Nest material has also been found to influence the breakage of eggs as well as the length of incubation. For species that nest in areas with little soft nesting material, eggs can be prone to breakage resulting in failure of the egg. Penguins nesting in areas of hard rock, ice, or hard dirt with little or no nest material have increased incubation lengths due to the lack of insulation (Boersma *et al.* 2004).

3.2.2 Risks during the Chick Stage

Once a chick hatches and grows the risks to survival change. The presence of the adult at hatching may be required to assist the chick out of the shell but once the chick has hatched brooding is required until the chick is able to thermoregulate its body temperature. If at this time brooding is not provided by the parent the chicks are likely to suffer from hypothermia. Once the chicks reach the intermediate (> 21 days) and post-guard stages the risk of death by hypothermia is reduced, however as the feeding demands of the chicks increase with age so do the risks of starvation.

During periods where food availability is low or variable within-nest competition may occur between siblings. Brood reduction is often the result of inter-sibling rivalry since the stronger chick will be more likely to receive more food at the cost of the smaller chick. Asynchronous hatching in bird species usually results in size asymmetries of chicks however the effects of this vary per species. For instance, Chinstrap Penguins (*Pygoscelis Antarctica*) exhibit size asymmetry however by the post

guard stage this was found to diminish and not cause any difference in growth and survival of the chicks (Monero *et al.* 1994; Massaro and Davis 2004).

LBP are highly philopatric returning to the same nest site to breed (Dann 1992). On average they lay two similar-sized eggs two to three days apart. Incubation is by both parents and will take on average 35 - 36 days. The male will incubate first while the female forages to replenish food stores. Due to the energetic cost of egg-laying they will exchange roles every one to two days. Hatching occurs asynchronously, approximately around 36 hours apart, after which the chick/s requires brooding for around 15 to 21 days by one parent at a time (Marchant and Higgins 1990). LBP are capable of laying three eggs however, based on an egg manipulation study by Dann (1988) LBP will tend to push eggs out of the nest when clutch size is large ($n = 6$ eggs), suggesting that the ability to raise large clutches is a function of brood patch size, method of incubation (Loyd 1977; Dann 1988) and egg size (Norman and Gottsch 1969; Dann 1988). Regardless of this, LBP have been known for their high rate of nest abandonment at the egg stage and LBP on Tiri have been found to have high rates of nest abandonment with associated/subsequent low breeding success in past years (Jones 1978; Chen 2004). Moreover, more work is needed to determine whether this low breeding success is typical for this population. Also to determine at what stage nests are most likely to fail and the cause of failure. This study examines the causes of egg and chick mortality in an effort to identify what key factor is impacting on survival of LBP clutches. This is the first study of this population to consider factors associated with different times of breeding which is important when considering optimal breeding times and survival risks (Arnold *et al.* 2004).

3.3 Aims

1. Identify causes of egg and chick mortality for LBP clutches.
2. Consider factors influencing egg success.
3. Consider factors influencing chick success.

3.4 METHODS

Note to the reader

A summary of methods relevant to this chapter are included here, however for more detail on the study site and nest locations refer to Chapter 2 (Sections 2.4.1 to 2.4.4).

3.4.1 Monitoring

Monitoring of each nest allowed the identification of the different breeding stages. This generally started with nest building, followed by egg laying, incubation, hatching, brooding, guarding, post guarding, and finally fledging. If at any stage during incubation both adults were away from the nest, the duration of absence was noted. The length of each nesting stage was also identified.

If chicks were still in the brooding and guard stages, the removal of the chicks required adult birds to be taken off the nest first. Adults were held in a cotton bag during chick measurements. Chicks were removed from the nest by hand. If there were two chicks on the nest, the smaller or younger of the two was marked with non-toxic correction fluid. Marking occurred on the back of the neck, tail, under the right flipper, and foot. This method was less invasive than other known methods for marking chicks such as punching a hole in the foot webbing, or clipping a toenail. Marking with fluid was just as effective but required re-marking at each visit. Chicks were always placed back on the nest before release of the adult. Chicks were weighed twice a week from hatching date, or from the date of finding. Each chick was placed in a micro fleece bag and weighed on electronic scales. The bag weight was also taken afterwards. The weight of the bird could vary with the time of day due to feeding, and evaporative water loss. Therefore to ensure that all chicks were treated equally they were weighed at

approximately the same time of day (Miyazaki and Waas 2003). Using the same side of the birds for all measurements meant that growth changes were more consistent.

When the chicks were near fledging, they were banded with a flipper band (usually around six weeks), and a feather sample was taken. For nests found during the chick stage, chick age could be estimated based on comparisons with other active nests and through morphological measurements. Age estimates were taken from Jones (1978) chick growth curves associated with LBP on Tiritiri Matangi. No chicks were taken off the nest during raining periods to avoid any chicks getting wet.

Newly located nests were sometimes found after desertion. A deserted nest is defined as one that has not been occupied by an adult for a week. In addition nests could be occupied but if the contents had been pushed out this was considered as failed. If a nest was found deserted or failed, any eggs or dead chicks were removed from inside/ outside the nest and kept. Eggs that were found alone on a nest were left for a week before removal. This was to ensure that they were actually abandoned and not just unattended. Removal of eggs was done either by hand, or where needed, with long steel tongs. Eggs were put into plastic re-sealable bags, and frozen at -20°C for later analysis. Chicks were removed on the day they were found dead, put into labelled bags, and frozen for later necropsies.

Once a nest was classified as abandoned, a sample of nesting material was also taken, bagged and frozen. Nest material was sorted through for presence of parasites, which were identified by AgResearch an independent laboratory. All nests were classified as one of four types based on location and substrate as follows: rock, earth, artificial, and tree.

3.4.2 Egg Analysis

Individual eggs were assigned a classification of failure after (Renner 1998) these included;

1. Nest desertion was defined as eggs that were abandoned by the adults and had not being incubated for at least one week.
2. Weather effects were based on how wet the burrow was at the time the egg was discovered.
3. Egg out of nest was when the egg was cold and off the nest.
4. Egg broken was when the egg was broken and empty, or cracked.
5. Unknown was when the egg disappeared or if the other classifications could not be applied. When failed the eggs were removed. The condition of the nest (wet/ dry) was also recorded.

3.4.2.1 Embryo Development

Each egg was necropsied to establish the developmental stage reached before failure. The length and width of each egg was measured with 200 mm digital callipers (*Kincrome*). Each egg was opened by removing the shell. The stage of egg failure was classified in terms of the level of chick development reached by comparing each embryo to photographs of domestic fowl (*Gallus gallus*), (Freeman and Vince 1974). The incubation length for the domestic fowl is 20 - 21 days, while the LBP is on average 36 days, therefore ratios were adjusted for penguins. The levels of chick development were (Plate 3.2);

1. Primary (P) – Little or no visible development. (1 – 3 days in fowl; 1 - 4 days in LBP).

2. Intermediate (I) – Eye pigmentation, allantoic bud and hindgut visible, soft limbs. (4 – 10 days in fowl; 5 - 16 days in LBP).
3. Tertiary (T) – Visible feather coverage, flexed limbs resting on body or over top of the head. (Day \geq 11 in domestic fowl; day \geq 17 in LBP).
4. Eggs that were in very early primary stages or infertile were unable to be identified due to the time between collection and death of the egg. Therefore rotten eggs were classified as unknown.

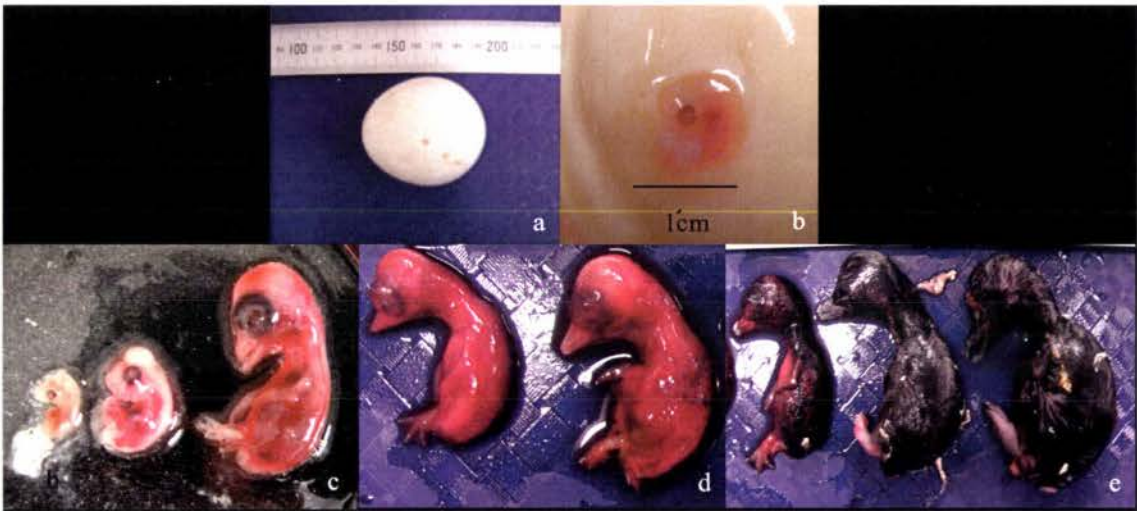


Plate 3.2. Photos of the embryonic developmental stages of the Little Blue Penguin, which include; a) LBP egg, b) end of primary development with a 4 day old foetus followed by c) & d) intermediate stages (5 - 16 days) and e) tertiary stage (\geq 17 days). Photo by J.Geurts 2006.

3.4.3 Chick Necropsies

The cause of death was based on known nest attendance and activity of the adults.

Chick failure was assigned a classification as follows;

1. Nest desertion was used when a dead chick was found on a nest with no adults. The reasons for actual death from this varied on the age of the chick. If the chick was within two weeks of hatching, then nest desertion could cause hypothermia and/ or starvation. Chicks older than two weeks in deserted nests were assumed to die from starvation.

2. Starvation was assigned when chick weight was found to be decreasing during the period prior to death.
3. Weather effects were assigned when the nest and plumage of the chicks were saturated with water.
4. Disappearance was assigned when the chick was absent from the nest. This could happen without complete failure of the nest due to successful siblings. Disappearance could occur when the chick was older and mobile and if deserted they may leave the nest in search of food.
5. Unknown was assigned when none of the other categories could be assigned (Renner 1998)

Any LBP chicks found dead were necropsied to identify the cause of death. The level of subcutaneous fat was noted and the stomach and crop checked for contents. Chicks that were able to be necropsied were also sexed from tissue using molecular techniques.

3.4.4 Data Analysis

3.4.4.1 Nest failure

The numbers of nest failures were tallied over each five day period and weather parameters were averaged over the same time period. The number of failed nests was correlated against weather parameters such as wind speed (km/h) and rainfall (mm) (Spearman's rank correlation).

3.4.4.2 Egg and chick failure

Cause of failure at the egg stage was compared with the lay date. Eggs that had a known lay date were used with eggs being classified individually. Using the median lay date

(31st October 2005) as the division the periods of laying were identified as either early or late.

3.5 Results

3.5.1 Nest Desertion

Egg and chick failure occurred at all stages of the nesting cycle while chicks only fledged at the end of the season (December 2005). The largest cause of failure associated with nesting attempts was nest desertion. Nest failure was not significantly correlated with wind speed or rainfall (Spearman's wind $r_s = -0.148$, $p = 0.435$, rain $r_s = -0.251$, $p = 0.181$) (Figure 3.2).

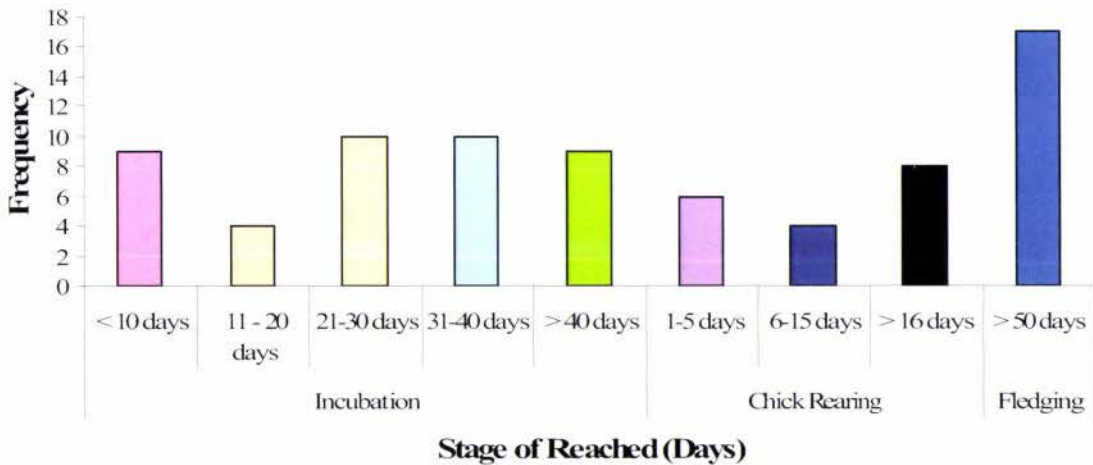


Figure 3.1. Nest desertions occurred throughout the nesting season regardless of the stage (egg or chick) that the nest reached. The frequency of chicks failed at both the guard and post-guard stage (> 16 days) due to hypothermia and starvation as also found for past studies of Little Blue Penguin on Tiritiri Matangi Island, New Zealand.

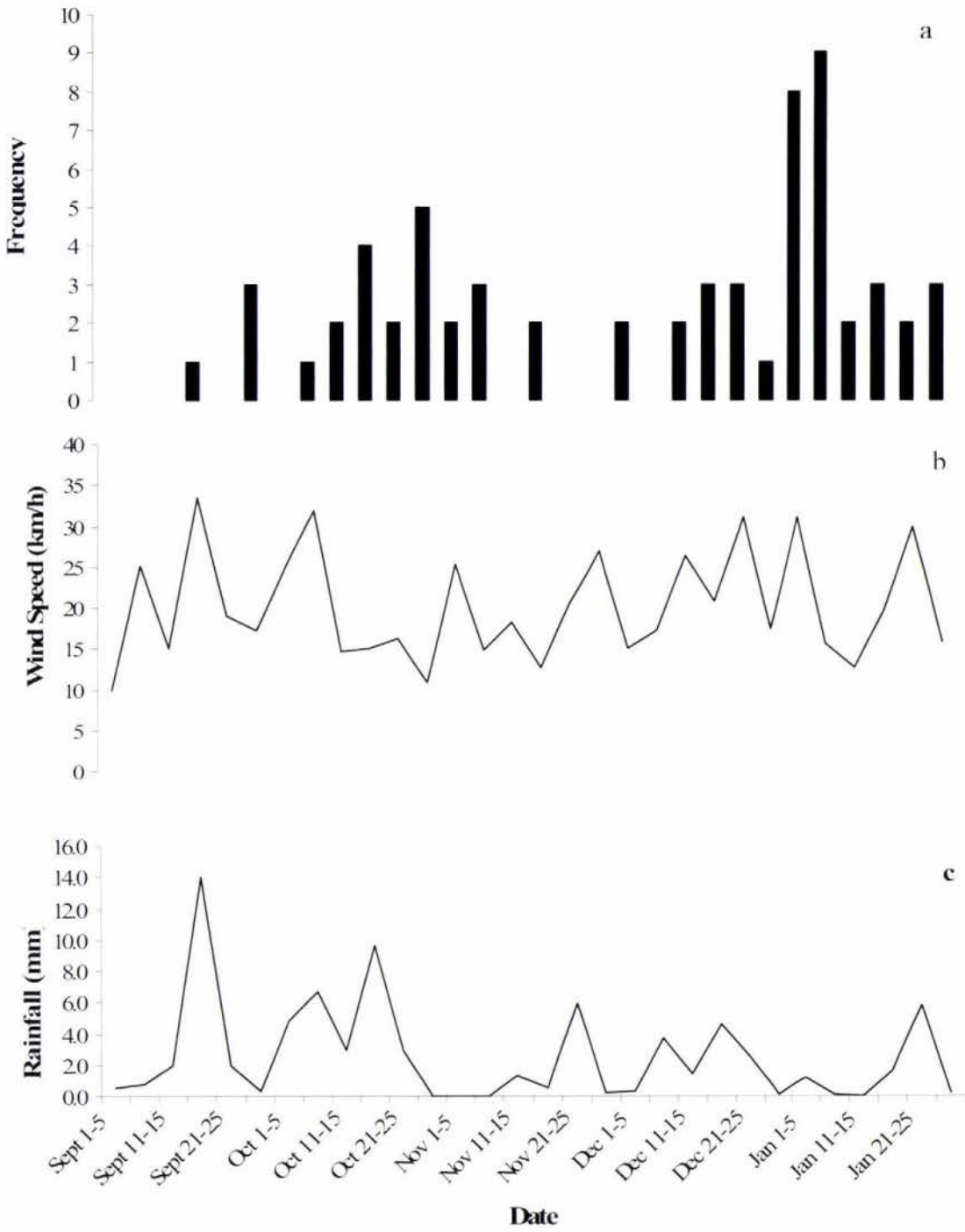


Figure 3.2. a) The frequency of nest desertions of Little Blue Penguins (bars) in relation to the b) wind speed (line) and c) rain fall (line).

The main cause of failure at the egg stage for both lay periods (early and late) was nest desertion for both eggs and chicks (Table 3.1). Similar numbers of eggs were deserted at early ($n = 43$) and late ($n = 41$) lay periods. Weather events resulting in failure were more common for the early lay period ($n = 10$) compared to later ($n = 2$). Nest desertion was also largest for the chick stage followed by starvation and disappearance. Weather did not factor as a direct cause of death for chicks.

Table 3.1. Comparison of the failure outcomes of Little Blue Penguin eggs and chicks with relation to the laying period (early or late) on Tiritiri Matangi Island, New Zealand.

| Stage | Period | n | Failure Classification | | | | |
|--------|--------|----|------------------------|---------|------------|---------------|---------|
| | | | Nest Desertion | Weather | Egg Out | Egg Broken | Unknown |
| Eggs | Early | 43 | 22 | 10 | 8 | 3 | 0 |
| | Late | 41 | 27 | 2 | 4 | 2 | 6 |
| | | | Nest Desertion | Weather | Starvation | Disappearance | Unknown |
| Chicks | Early | 11 | 5 | 0 | 3 | 2 | 1 |
| | Late | 28 | 10 | 0 | 4 | 9 | 5 |

3.5.2 Egg Analysis

The total number of eggs monitored during 2005/06 breeding season was 162. Of those 99 eggs failed before hatching and five failed at hatching.

Table 3.2. Comparison of sample number of nests, average incubation, and failure stage for early versus late laying periods. Non-significant test (^{ns}).

| | | Early | | Late | | <i>t-test</i> (<i>p</i> =) |
|----------------------|--------|--------------|-------|------|-------|--------------------------------|
| | | n | Days | n | Days | |
| Eggs Length | | 5.2.4 ± 0.27 | | | | |
| Egg Width | | 4.09 ± 0.02 | | | | |
| Incubation | Eggs | 14 | 32.85 | 17 | 32.35 | 0.47 ^{ns} |
| | Chicks | 8 | 35.5 | 13 | 35.95 | 0.43 ^{ns} |
| Failure Stage | Eggs | 43 | | 41 | | |
| | Chick | 11 | | 27 | | |

3.5.2.1 Egg Properties

There was no significant difference between the average length and width of LBP eggs within the population since 1975 (Table 3.3).

Table 3.3. Comparison of LBP egg dimensions (\pm SE) between years on Tiritiri Matangi island (Tiri) and between subspecies from Matiu-Somes Island (Matiu-Some), Wellington and Phillip Island (Phillip), Australia.

| | 2005/06 | Tiri 1975 | 1976 | Matiu-Somes | Phillip |
|-------------|-----------------|----------------|----------|-------------|---------|
| Length (mm) | 52.4 \pm 0.27 | 52.4 \pm 0.7 | 56 \pm | 54.9 | 56.06 |
| Width (mm) | 40.9 \pm 0.13 | 40.8 \pm 0.5 | 43.2 | 41.9 | 43.15 |

Of the 71 eggs that were necropsied 52 were suitable for classification in terms of their developmental stage of failure while 13 remained unknown and six were infertile. The unknown eggs were either fertile and failed at the primary stage, or infertile. Failure of the egg during development was largest at the primary stage 31% (n = 22) and equally at the intermediate 21% (n = 15) and tertiary stages 21% (n = 15) while unknown was 27% (n = 19).

3.5.2.2 Incubation Length

The incubation length (mean \pm SE) for each stage were; Primary 22 \pm 4.39 days (n = 9, range 3-34 days), Intermediate 23.5 \pm 2.38 days (n = 4, range 18-28days), Tertiary 32 \pm 2.5 days (n = 7, range 24 - 41days), Infertile eggs 51.5 \pm 6.63 days (n = 6, range 41 - 80).

On average eggs were incubated longer in tree nest types as opposed to other nest types (rock, earth, and artificial) although the variability was also the greatest (Figure 3.3).

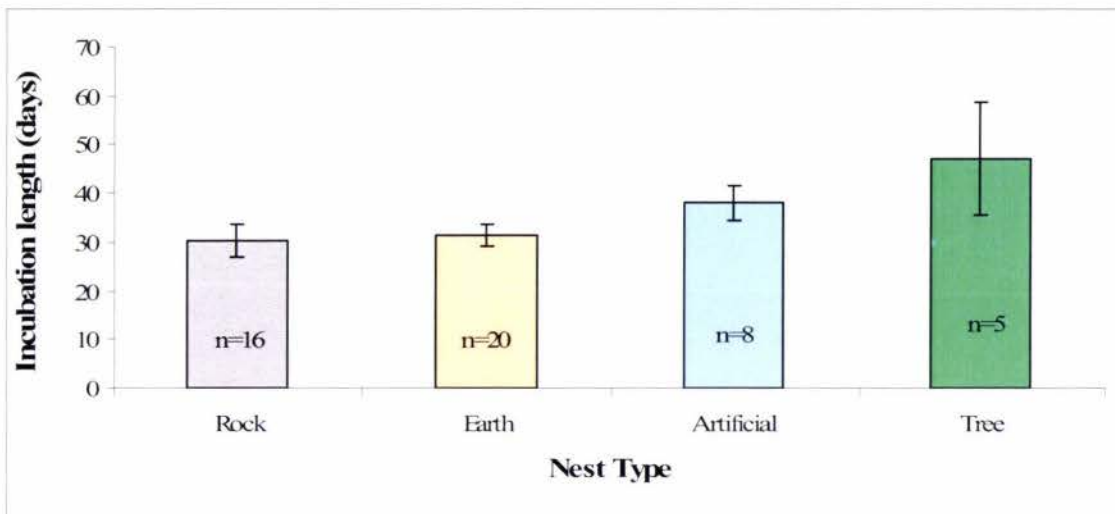


Figure 3.3. Comparison of the different incubation lengths (\pm SE) for each nest type used by LBP during the 2005/06 breeding season.

3.5.3 Chick Analysis

The largest cause of failure at the chick stage was due to nest desertion. Greater desertions of chicks occurred during the late ($n = 27$) period of the breeding season compared to the early ($n = 11$). Weather was not a factor associated with death of chicks. Starvation was the known cause of death for seven chicks but may also have played a larger role influencing chicks to leave the nest, and for chick deaths where cause was unknown.

Only five chicks were necropsied out of the 13 that were collected due to the poor state of seven chicks. Results showed that all five chicks had no fat stores and four had no stomach contents. The chick that did have the stomach material was the oldest and from a nest of two chicks and the younger chick had disappeared from the nest. Cause of death for the older chick is unknown.

Thirty-four nests hatched chicks but only ten nests fledged chicks. From the 34 nests that failed, 18 nests hatched two chicks (twin brood). Twin broods failed if the first chick died after the second chick ($n = 11$), or if both chicks died at the same time (n

= 7). Those twin broods where chicks died at different times were all found to be in the post-guard stage (100%), while twin broods found dead at the same time were still within the guard stage (100%).

DNA results of necropsied chicks had a sex ratio of female (n = 6), male (n = 5), and unknown (n = 2). Compared to fledged chicks female (n = 4), male (n = 8), and unknown (n = 2). Therefore from the 62 chicks that hatched females (n = 10), males (n = 13), and unknown (n = 39). For twin broods with known sex of both chicks, the first chicks were female (n = 6) or male (n = 7). These sample sizes did not allow for statistical comparisons.

3.6 Discussion

During the 2005/06 breeding season nest failures occurred across the whole season but varied with the lay period (early or late). Nest desertion was high in LBP on Tiri but was similar to studies by Jones (1978) and for other LBP populations (Bull 2000). The cause of nest failures did not correlate with wind and rain measures however this may not best explain the link between climate and nest failure. Further analysis of the different nesting stages gives greater understanding to the factors associated with breeding.

Nest attendance patterns have been found to provide interesting insights into the reason for nest desertions (Numata *et al.* 2000) since the cause of nest failure can vary temporally and spatially depending on each population of LBP. Jones (1978) found high nest abandonment's (56%) during 1975-1977. The reason for this was unknown however a large number were found to be pushed out of the nest or broken. The major cause of death at both the egg and chick stages during 2005/06 was nest desertion. This is also the main cause of failure of eggs in Adélie Penguins *Pygoscelis adeliae* (Spurr 1975; Davis 1982; Davis and McCaffrey 1986; Kemp and Dann 2001). LBP rely heavily on both adults to successfully raise a clutch (Numata *et al.* 2000) therefore nest desertion is the result of one adult failing to relieve its partner from nest duty (incubation and chick rearing). The causes associated with the inability for adults to change over duty (food, injury/ death) will have a direct consequence on the clutch (incubation temperature, hypothermia and starvation of chicks).

Although weather was not correlated with nest failures the underlying effect that wind speed can have may not identified by the correlation or could have been associated with a delayed effect. The potential cause of delayed nest relief during the early lay

period could be a series of storm events. This may be evident from the larger number of eggs that were affected by weather during the earlier period than later, however chicks were not affected by weather. However, more eggs were present during the September/October period which occurred during the early months of the breeding season and is associated with increased wind speed and rainfall. Similar weather conditions were found to have affected LBP breeding attempts on Tiri in 2003 (Chen 2004).

Storms can result in adults being unable to forage and build up stores for incubation and can also cause adult death at sea. These factors were the main case for the death of one incubating bird which didn't desert the nest but died of starvation (based on necropsy results, see Chapter 5). Later in the season, nest relief and ultimate abandonment was thought to be associated with a lack of food associated with either a decrease in food abundance or inability of adults to forage (Chapter 4). If an adult does not get enough food then they will not have the body condition to continue incubation duty or provision chicks, therefore nest attendance patterns will reflect the way that LBP are able to balance the demands of breeding with that of self maintenance (Numata *et al.* 2000). Differences in the causes of nest desertion during the breeding season have also been found for LBP on Matiu-Somes Island, Wellington (Bull 2000).

The results of nest failure and success emphasise the importance of laying earlier within the season since all chicks that fledged came from birds that laid early within the season (Chapter 2). Although the proximate mechanisms are not well understood, the lay date of eggs within a breeding season have been found to influence incubation length of several bird species (e.g. Herring Gull, Parsons 1972; Rockhopper Penguins, St Clair 1996; (see Massaro and Davis 2004). This is thought to be a function of ambient temperature, egg size, and nesting activities (Massaro and Davis 2004).

3.6.1 Survival at the egg stage

3.6.1.1 Incubation

Numata *et al.* (2000) has reported that the main reason for poor reproductive success of LBP is failure at the incubation phase. During the egg stage survival is related to factors that impact on the hatchability of the eggs and which will ultimately reduce breeding success (Darby and Seddon 1990). These include incubation length, fertility, and the stage of embryonic development. The incubation lengths of eggs that fledged chicks were 36, 39, and 41 days while other clutches ranged from 32 to 44 days. The incubation length for Tiri birds is highly variable (Kinsky 1960; Jones 1978) which could relate to the number of days that nests are left unattended during the incubation period. In the current study, nests with eggs were left one to four days and up to three times during incubation. Irregular incubation of a clutch could be due to younger, inexperienced birds nesting (Richdale 1957) or the need for any particular pair to forage longer. This could force the incubating bird off the nest to replace lost energy stores (Bull 2000).

The lack of a steady incubation pattern has not hindered the hatching process for some eggs which may be due to the stage of egg development since eggs within the tertiary stage have been found to hatch due to their own metabolic heat of the developing embryo (Reilly and Balmford 1975; Jones 1978). Although a manipulation study on incubation temperatures found that maintenance of a constant temperature in Nazca Boobies (*Sula granti*) may be critical for development and survival during the last third quarter of incubation (Morgan *et al.* 2004). The development of a brood patch in Yellow-eyed penguins (*Megadyptes antipodes*) has also been found to reach full vascularisation for incubation after 15 to 42 days of the incubation period (Farner 1958;

Morgan *et al.* 2004). Regardless, failure to maintain optimal temperatures can either cause retardation in the egg (Romanoff and Romannoff 1972; Wilson 1991; Deeming and Ferguson 1992), lengthening of the incubation period (Tulp and Schekkerman 2006) or ultimately lead to nest desertion. Time allocation of birds requires a trade-off between factors that affect cooling rates, parental requirements, and feeding intake (Tulp and Schekkerman 2006).

Incubation temperatures are energetically expensive to maintain since they require transfer of heat from parents to the egg and physical manipulation of the eggs, therefore a trade-off exists between maintaining optimal temperatures while minimizing the cost to parents (Morgan *et al.* 2004). Being absent from a nest will also require partial reheating of the egg which is more costly than maintaining a constant temperature (Drent 1973; Biebach 1986; Hainsworth & Voss 2001; Turner 2001; Tulp and Schekkerman 2006). In bi-parental systems where both parents participate in incubation regular exchanges without absent periods can provide optimal temperatures and limit potential cooling. Foraging to maintain energy reserves is also energetically costly (Piersma *et al.* 2003; Tulp and Schekkerman 2006), hence leaving the nest to forage would require a net energy gain from foraging to outweigh the costs of energy expenditure (Tulp and Schekkerman 2006).

The majority of eggs were found to fail at the stage of primary development regardless of incubation length. One nesting pair was found to incubate an egg for 80 days consistently without the egg hatching and the egg was therefore classified as infertile. This suggests that the parents cannot detect early embryo death.

3.6.1.2 Nest type

Incubation length can also be affected by the nest type and can influence egg success and breeding success (Jones 1978; Bull 2000). Nest type was not found to be associated with breeding success for 2005/06 breeding season (Chapter 2) however it has been found to influence the success of fledging chicks for other populations of LBP (Knight and Rogers 2004), Magellanic Penguins *Speniscus magellanicus* (Stokes and Boersma 1991; Bull 2000), Black-footed Penguins *Speniscus demersus* (La Cock 1988; Bull 2000), and LBP on Matiu-Somes Island (Bull 2000).

The lack of breeding observed in artificial nest boxes could be due to the lack of thermal insulation and the ambient temperature was very similar to the external nest box temperature. This appears not to be ideal nesting conditions when compared to other nest types (rock and flax bush) that maintain a relatively constant inside humidity and temperature (R:20.5, T:23/26 respectively) regardless of the outside temperature fluctuations (R:27/25, T:23/34 respectively) (from Jones 1978). A steady temperature within a burrow is thought to be a contributing factor to nest success and has been an important factor within the Jackass Penguin *Speniscus demersus* (Frost *et al.* 1976; Jones 1978). There does not appear to be a shortage of nesting sites on Tiritiri Matangi Island; however, it has been suggested that there is a low number of quality nest sites (Jones 1978).

Low humidity levels have been considered to make incubation difficult since nest types such as caves, bushes, and nest boxes may not provide much relief from extreme temperatures (Klomp *et al.* 2001). Nest temperatures vary with breeding stage and with the number of individuals within the nest. Chicks release a lot of heat especially when covered in down. The temperatures in the artificial concrete nest boxes reached very high levels, although not as high as those found in Australia (Rouper-

Coudert *et al.* 2004). High temperature may force the adults and hatchlings to leave the nest for relief during the peak summer months.

Nest microhabitat may not be the only factor limiting nest selection or reuse of old nest sites. Properties of protection such as sound structure and susceptibility to flooding will be affected by weather, substrate, predation, and nest cover. Depending on the location changes to nesting conditions could be associated with flooding during high periods of precipitation, and collapse after storm periods. Several nests were found to be prone to flooding and collapse, especially nests that were low in the ground, or made from loose dirt and plant substrate.

3.6.2 Chick Survival

3.6.2.1 *Potential influences of egg dimensions on chick development*

Egg volume was not calculated within this study, however the size of eggs is thought to increase with latitude and/ or age of the laying adult (Richdale 1955). Egg length and width dimensions were found by Jones (1978) to be slightly larger than the most recent breeding season, however smaller than other LBP populations (Reilly and Cullen 1975; Jones 1978). Population differences between sub-species of LBP, however differences in time on Tiri may be due to a change in demography to a younger population.

LBP within this study were not found to show any large differences in intra-clutch egg dimorphism and therefore potential effects of egg size on chick success are unlikely to be large.

Differences in the egg size of the first and second laid eggs may be an attempt at increasing the chance of survival of the second laid egg. Little has been done on the impacts of egg size on post-hatching success, however manipulation of eggs in Thick

billed Murres *Uria lomvia* and Razor Bills *Alca torda* have shown that larger eggs were associated with earlier development of wing feathers (Hipfner 2000). In seabirds this would be important for early chick fledging and hence earlier foraging. Chinstrap and Magellanic Penguins produce eggs of variable size however their effect on chick survival and sibling size is unknown (Monero *et al* 1994; Boersma and Stokes 1995; Kemp and Dann 2001). Furthermore, Crested Penguins are associated with a brood reduction strategy and the smaller egg of two has a lower chance of survival (Williams 1980, 1990; St Clair 1992; Kemp and Dann 2001). This is unknown at this stage for LBP.

Penguins are known for their asynchronous egg laying and hatching, hence the first hatched chick will have a head start at feeding. Weights of chicks within this study were slightly different between chicks, a finding similar to Jones (1978). This suggests that there is intra-brood competition. During periods of low prey availability such sibling competition is likely to be highest (Green and Gales 1990). At these times the stronger chick would be more likely to survive at the expense of the younger chick. This is likely to have been the case during the 2005/06 breeding period on Tiri.

Nest desertions overall may be driven by the high energy demands associated with chick provisioning. Miyazaki and Waas (2003) found that no chicks died within the guard stage (n = 18 chicks) in 2001 on Tiri in contrast to Jones's study in 1974-1976 (1978) where all chicks died within days of hatching. Such discrepancies could be due to adult body condition. Although adults will desert the nest at the chick stage if they are in poor condition (Chiaradia *et al.* unpubl. data).

3.6.2.2 *Effects of parasites*

Parasites associated with LBP nests on Tiri include louse, mites, and fleas. The louse species found in this study was the *Austrogoniodes watersoni*. This has also been found on LBP from Coromandel Peninsula, Wellington Harbour, Chatham Island, Banks Peninsula, Otago Harbour, and Phillip Island Australia (Banks *et al.* 2006). A mite species (Acarina, Veigaiidae, *Veigaia* sp.) was also found from nesting material but this has not been identified or associated with LBP before. The flea species found *Parapasyllus longicornis* was also found by Jones (1978).

Nests are often the site of large numbers of ectoparasites since it is an environment that is associated with high rates of host occupancy and these ectoparasite levels have the potential to cause nesting failure in birds and influence the growth of young (Duffy 1983). *Ixodus* species have caused mortality of young (Zolotov and Buker 1976; Duffy 1983), through transmission of viruses (Duncan *et al.* 1978; Duffy 1983) and undernourished young (Johnstone *et al.* 1975; Duffy 1983). Nest material samples and analysis of parasites showed that ticks of the Acari: *Ixodus eudyptis* species were present in LBP nests and these were also found previously by Jones (1978). Chicks were found to have large number of ticks feeding under the feet, in the corner of the mouth, and throughout the plumage. Adults on the nest were also found to have large numbers of ticks within the ears.

The Aragasid tick is capable of fasting for long periods of time (Duffy 1983) which means that they can persist in a nest from one breeding season to another making transmission of diseases a real possibility. These ticks require several days to gorge on blood from a host after which they will drop off. Adult female ticks require several different hosts to obtain their full requirements (Oliver 1989), therefore associating themselves with nesting species will be beneficial. The effects of ticks are not well

known and require more research. However, there was no evidence in this study that chicks died as an effect of ectoparasite infestation, this finding is consistent with that found by Jones (1978). Parasites may have a greater effect on an individual when stress levels are high (Ranum and Wharton 1996). Young chicks left for the day due to unsynchronised partner changeovers during brooding appeared to result in hatching of fly eggs on live chicks.

This is the first study to identify the range of parasites associated with the North Island population of LBP. However due to the lack of prior knowledge, few conclusions can be made about their effect on chick survival. This suggests future research within this area is needed.

3.6.3 Conclusions

It is apparent that regardless of the nesting stage nest desertion in LBP was a major factor associated with mortality of eggs and chicks. This has also been found in other bird species and for other populations of LBP. The factors that affect survival at each stage appear to be different and may incorporate a number of complex relationships that may not always be picked up by simple statistical analysis. Regardless, lay date was the most influential parameter associated with breeding success in LBP (Chapter 2) and analysis of the different nesting stages allowed for further investigation into the key factors associated with this. During the early period of breeding, nesting attempts were affected more by weather conditions which hindered adults from obtaining food thus influencing partner nest relief (ref Chapter 4). During the later stages of the breeding season, low food availability and chick provisioning resulted in starvation of chicks (Chapter 4). Furthermore there were a large number of LBP mortalities within the Hauraki Gulf which was found to be related to starvation (see Chapter 5). Both the lack

of food and the struggle for adult survival will have a direct impact on population productivity and deserves further investigation of the long-term impacts.

Due to the lack of long-term data it was difficult to determine what an average breeding season on Tiri was and what the most common cause of failure was across years. Hopefully this study will provoke future research into the risks associated with the egg and chick stages. Monitoring of breeding success and chick survival is an important part of using birds as biological indicators and understanding population demographics (Boersma 1978; Cairns 1987; Monaghan *et al.* 1989; Stenzel *et al.* 1995; Erwin *et al.* 1996; Harris and Wanless 1997; Renner 1998).

CHAPTER 4 Feeding ecology



Plate 4.1. A Little Blue Penguin from Tiritiri Matangi Island. Photo by J. Geurts 2006.

4.1 Abstract

The foraging ecology of Little Blue Penguins (LBP) on Tiritiri Matangi Island (Tiri), New Zealand, during 2006 was assessed based on stomach regurgitations and using stable isotope ratios from feather samples from 1886 to 2006. Regurgitations covered the feeding period from January to April 2006. Stable isotope data were obtained from historical specimens (Auckland Museum) as well as live birds and spanned a period of 120 years (1886 – 2006). Stable isotopes were used to examine trophic levels of penguins as well as other terrestrial birds occupying Tiritiri Matangi Island. Since little work has been done with stable isotope analysis within New Zealand and in particular the North Island marine environment, comparisons were limited to studies of other species of sea birds in different locations. Major prey types were fish (85.71%) followed by cephalopods (42.86%) in stomach regurgitations ($n = 14$). Target species included Anchovy (*Engralis australis*), Yellow-eyed Mullet (*Aldrichetta forsteri*), Goby, Red Cod (*Pseudophycis bachus*), and Arrow Squid (*Nototodardus sloanii*), with findings of myctophids and several other unidentifiable fish species. The $\delta^{13}\text{C}$ levels indicated that LBP were foraging within inshore regions associated with juvenile fish stages. This was supported by the size ranges of prey species. Both $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values were higher than terrestrial bird species which was consistent with a high protein diet. However, the trophic levels of LBP were low for a top predator (fluctuating from 3.3 to 4) and were similar to marine birds feeding on planktivorous rather than piscivorous species. While stable isotopes varied from the earliest record of 1886, these increases and decreases in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ showed no particular trend and were not correlated with Southern Oscillations. Isotope analysis of feathers from 2004 ($n = 22$) and 2005 ($n = 23$) were significantly different in the levels of $\delta^{13}\text{C}$ but not in $\delta^{15}\text{N}$. Chicks from the 2005

breeding season had similar isotope values to the 2005 adults. Observed individual variation and a lack of correlation with large scale climatic events such as Southern Oscillations suggests that finer scale factors associated with prey abundance and availability may better explain isotope variations. The value of this information is discussed in terms of understanding the nutritional links between secondary and high level consumers and its role in identifying trophic shifts within the marine environment.

4.2 Introduction

Seabirds are increasingly used as biological indicators due to their responses to climate and prey changes that occur within the marine environment (Schreiber and Burger 2002). Specifically, these may include changing sea surface temperatures (Ratz *et al.* 2004), productivity levels (Montague and Cullen 1988), and long-term climate changes (Hilton *et al.* 2006). Seabird responses to these environmental changes vary with species but include breeding success (e.g. Gentoo Penguins *Pygoscelis papua*, Croxall *et al.* 1999), foraging effort (e.g. Wandering Albatross *Diomedea exulans*, Shaffer *et al.* 2003), diet switching (e.g. Rockhopper Penguin *Eudyptes chrysocome* Hilton *et al.* 2006), and survival (e.g. Common Guillemot *Uria aalge*, Sandvik *et al.* 2005).

Seabirds rely primarily on the marine environment for their food requirements therefore variations in breeding success, survival and foraging behaviour often reflect food supply (Monaghan 1996) or the ability to obtain food. To cope with changing food availability seabirds may; alter foraging behaviours (e.g. travelling distance, feeding depth, or time spent foraging), or life-history parameters (e.g. clutch size, breeding success). For example free-ranging Wandering Albatrosses vary their foraging effort depending on prey availability thereby maximizing foraging efficiency when food supply is low (Shaffer *et al.* 2003). Furthermore Australasian Gannets (*Morus serrator*) in New Zealand have variable breeding success between years and populations which is thought to be related to weather and food supply (Wingham 1982).

Penguin species have also been documented to respond to changes within the marine environment, making them suitable biological indicators. For instance long term monitoring of Yellow-eyed Penguins (*Megadyptes antipodes*) has shown that population sizes fluctuate with food supply (attributed to changes in sea temperature)

(Ratz *et al.* 2004). Variations in krill abundance have been found to influence the breeding success of Gentoo Penguins (Williams 1991; Williams *et al.* 1992a, b; Croxall and Prince 1999). However under the same environmental pressure Macaroni Penguins (*Eudyptula chrysolophus*) will tend to switch prey types (Croxal *et al.* 1999). LBP have also been shown to switch prey types when main prey species are low (Montague and Cullen 1988). For example poor breeding years for LBP in Southern Australia were associated with mass die-offs of Pilchard (*Sardinops sagax*) during 1995 and 1998 (Cullen *et al.* 1992; Hobday 1992; Chiaradia *et al.* 2003) but LBP numbers subsequently improved due to a shift in their diet to other prey species (Chiaradia *et al.* 2004).

Knowledge of diet is therefore a prerequisite to understanding the role of top predators within the marine environment (Croxall *et al.* 1984; Gales 1985). To date knowledge of foraging behaviours is poor (Schreiber and Burger 2002), however the use of new and improving technologies are increasing our understanding. These technical advances have included boat surveys (at sea distributions), satellite imagery (habitats) (see Schreiber and Burger 2002), satellite and telemetry tracking (individual foraging ranges) (Mattern 2001; Chiaradia *et al.* unpub), tracking with time depth recorders (diving depths and foraging patterns) (Mattern 2001; Chiaradia *et al.* unpubl. data), and stable isotope analysis (trophic utilisation and dietary shifts) (Hobson *et al.* 1994; Sydeman *et al.* 1997; Schreiber and Burger 2002; Hilton *et al.* 2006).

Seabirds exploit the ocean's resources in many ways and are found at many trophic levels within marine food webs (Schreiber and Burger 2002). For example storm petrels (*Oceanites* sp.) feed on zooplankton, gannets and some penguins feed on pelagic species of fish and squid, while some albatrosses are scavengers (Schreiber and Burger 2002). Different dietary requirements and morphological differences between seabird

species influence the foraging range (inshore versus offshore and depth), and foraging tactics (pursuit diving e.g. penguins, or diving e.g. petrels) (Schreiber and Burger 2002).

4.2.1 Diet Analysis of Little Blue Penguins

4.2.1.1 Conventional diet analysis of LBP

The LBP is considered to be a generalist feeder foraging on a wide variety of prey types (Cullen *et al.* 1992; Gales and Pemberton 1990; Van Heezik 1990; Montague and Cullen 1998; Williams 1995), such as surface schooling fish, squid, and crustaceans (Perriman *et al.* 2000). Particular prey species have been found to include Pilchard (*Sardinops sagax*), Slender Sprat (*Sprattus anipodum*), Arrow Squid (*Nototododarus sloanii*), juvenile species of Red Cod Snapper (*Pagrus auratus*), Barracuda (*Thyrsites atun*), Blue Wharehou (*Seriolella brama*), and Gem Fish (*Rexea solandri*) (Cullen *et al.* 1992; Gales and Pemberton 1990; Van Heezik 1990; Montague and Cullen 1998; Williams 1995). These prey species change in size and abundance seasonally due to life-cycle traits and climatic changes, respectively. When available, LBP may specialise on optimal prey species such as Pilchard (Chiaradia *et al.* 2004) and Anchovy, otherwise they are likely change to sub-optimal prey species such as Red Cod, Blue Wharehou and Baracouta (Chiaradia *et al.* 2003).

LBP feed exclusively at sea and spend at least 50% of their life doing so (Weavers 1992). However, unlike other seabird species, LBP do not regurgitate their stomach contents upon disturbance. Past techniques used to investigate the diet of penguins have involved killing the animal, emetics (Gales 1985; Montague and Cullen 1985, 1988), and stomach pumping (Gales 1984, 85; Ainley *et al.* 2003). The stomach flushing technique has been applied to at least seven penguin species and has shown to be capable of obtaining around 95% of the full contents (Gales 1985).

Although studies have been conducted on the feeding ecology of the LBP, no attempt has been made to document this in the New Zealand North Island sub-species. Such information would be valuable as only a few species can provide information regarding the links between primary producers and secondary consumers (Cushing 1975; Sydeman *et al.* 1997) like LBP and bait fish. For example commercial bait fisheries operating within New Zealand target prey species which have been found to be preferred prey of LBP from other populations within New Zealand and Australia (i.e. Pilchard and Anchovy: Cullen *et al.* 1992; Williams 1995, Slender Sprat: Fraser 1999; Perriman *et al.* 2000, and Arrow Squid: Montague and Cullen 1988). The largest catches from the bait fisheries are taken from the North Island region (East Northland to Bay of Plenty) annually, and have been increasing (Ministry of Fisheries Plenary Report 2005).

4.2.1.2 Long term diet analysis using stable isotopes

Despite the importance of understanding the feeding ecology of top predators, studies on species do not always extend back far enough to elucidate effects of the marine environment on predator-prey interactions. This is the case for LBP in the North Island. Research into the LBP population on Tiri has been sporadic and for short time periods. However, comparative analysis of stable isotopes found in tissues of historic specimens (Auckland Museum) with that of current tissue of LBP may help identify any major diet changes over time.

Stable isotopes of delta $^{13}\text{C}/^{12}\text{C}$ Carbon and $^{15}\text{N}/^{14}\text{N}$ Nitrogen occur naturally within tissues of organisms but vary depending on assimilated dietary components. As the components are broken down and incorporated into various tissues, the lighter isotopes (^{12}C and ^{14}N) drop out meaning that the heavier isotopes (^{13}C and ^{15}N) can be easily measured and distinct isotopic signatures are obtained.

Stable isotopes can be obtained via a variety of tissues such as eggs, blood, breath, and bone and are measured in parts per thousand (‰) (DeNiro and Epstein 1978; 1981; Sydeman *et al.* 1997). Since dietary nutrients are incorporated into these tissues during the period of growth, they will reflect the individual diet over the time period that these tissues were produced. Measures of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ will change depending on individual diet, which will therefore reflect the current foraging environment. For example, if penguins show a shift in their diet to lower primary productivity then the stable-carbon values would be expected to decrease. However, it has also been suggested that stable-carbon values can identify the source of feeding such as inshore or offshore foraging within the marine environment since inshore areas have greater productivity and hence greater $\delta^{13}\text{C}$ values (Hobson *et al.* 1994).

Stable-nitrogen levels increase in a step-wise fashion in marine systems as you move from primary producers to higher secondary consumers (Rau 1982; Wada *et al.* 1987; Fry 1988; Hobson and Welch 1992; Hobson *et al.* 1994). Therefore, the denser the tissue that a bird is feeding on (i.e. fish as opposed to crustaceans) the higher the stable-nitrogen value. This means that the $\delta^{15}\text{N}$ can be used to calculate the trophic level that the birds are feeding at (Wada *et al.* 1987; Rau *et al.* 1992; Hobson *et al.* 1994). Trophic levels can be estimated based on enrichment factors, which is expressed as the difference in stable isotope values between prey and predator tissues. However, different tissues will vary depending on their half-life and the metabolic processes of each bird. To calculate trophic levels the half-life of each tissue needs to be known. For example the half life of feathers has been estimated to be around 4‰ (parts per thousand) (Mizutani *et al.* 1990; Hobson and Clark 1992).

Feathers are ideal for long term analysis since they are moulted every year in penguin species and reflect the diet relative to the month before feather growth (Hobson

and Clark 1992; Bearhop *et al.* 2002; Pearson *et al.* 2003; Hilton *et al.* 2006) and are relative to assimilated dietary nutrients rather than ingested food (Hobson and Clark 1992). Feathers and blood are mostly protein so the $\delta^{13}\text{C}$ of the tissues is similar to the actual dietary protein (Podlesak *et al.* 2005). Therefore, the differences between stable-isotopic signatures can help us to identify differences between individual diets (Podlesak *et al.* 2005).

The ratios of stable carbon and nitrogen also change with response to ecological process (Hilton *et al.* 2006) such as upwelling, coastal advection, and variability of prey (Sydeman *et al.* 1997). Temporal changes in isotopic signatures have been studied in fish (MacAvoy *et al.* 2001; Post 2003; Podlesak *et al.* 2005), birds (Sydeman *et al.* 1997; Cherel *et al.* 2005; Hilton *et al.* 2006) and mammals (Sydeman *et al.* 1997). The ecological history of the Rockhopper Penguin was recreated by isotope analysis of the feathers from 1861 to present, and compared between sites from the sub-Antarctic regions (Hilton *et al.* 2006). The results showed evidence of a decline in the nitrogen levels at some sites with a negative relationship to sea surface temperature. This suggests that there was a shift towards lower trophic levels during warm years. There was also a significant relationship between declines in primary productivity levels (CO_2) and Rockhopper Penguin numbers over time (Hilton *et al.* 2006).

4.2.1.3 Current study

Stable isotope analysis and conventional methods (stomach regurgitations) of diet analysis (Hilton *et al.* 2006) have advantages and disadvantages depending on the question being considered. However, when used together the two methods offer a complimentary approach to understanding diets (Sydeman *et al.* 1997). The number of studies using stable isotope analysis has increased due to their long term applicability and inference of species associations within food webs. These analyses have never been

done on LBP. Furthermore, no published studies have been done within the North Island of New Zealand using stable isotopes to identify nutritional links between predator-prey species or changes associated within the marine environment. This study aims to identify the basic feeding ecology of LBP within the Hauraki Gulf through conventional dietary analysis as well as through stable isotope analysis. This will be considered with reference to the dietary requirements of LBP, predator-prey interactions, the balance associated with terrestrial and marine environments, and the limitations associated with dietary studies.

4.3 Aims

1. Identify likely prey species of the LBP in the Hauraki Gulf, North Island, New Zealand through identifying stomach contents of foraging penguins.
2. Use $\delta^{15}\text{N}$ to establish trophic levels of LBP found within the Hauraki Gulf and consider if changes have occurred since 1886.
3. Use $\delta^{13}\text{C}$ to estimate whether LBP are conforming to inshore feeding and if productivity levels have changed between 2004 and 2005 and since 1886.
4. Consider the potential factors (Southern Oscillation, sea surface temperatures) that could influence changes within the marine environment and the influence that this may have in the diet of LBP.
5. Relate findings back to implications that foraging and diet has on the productivity and survival of LBP from Tiri.

4.4 Methods

4.4.1 General Application

Note to reader, for information on the study site see Chapter 2 (Sections 2.4.1).

4.4.2 Overview

The stomach flushing technique was conducted at night on incoming penguins during the end of the breeding season (January and February) and again after the breeding season had finished (March and April). Adult birds were caught as they first emerged from the water to minimise digestion of the food (Montague and Cullen 1988). Birds were weighed in a cotton bag of known weight with a 5 kg pesola scale. Each bird was banded (unless already banded) so that they were not re-sampled in the future. A feather sample was taken for genetic assignment and isotope analysis. The stomach flushing method followed that of Wilson (1984). Each individual was re-weighed and held for around 20 – 30 minutes until it proved to be active and in good condition. Homogenized pilchard was then supplemented to the birds to re-hydrate, aid recovery, and provide the adult with a meal to feed any chicks.

Each sampled penguin was returned to the spot that they were collected from. All samples were stored under refrigeration for weighing the next day, after which they were frozen (-20°C) for later analysis. No birds were found to die as a result of this method in this study or in previous studies (Gales 1987).

4.4.3 Data Collection

4.4.3.1 Stomach Flushing

A 5mm catheter tube was used for stomach flushing. The tube had holes at the inserted end for the water to flow out of and this end was also firmer to prevent folding back of the tube ends as it was inserted (Plate 4.2). The catheter was inserted down the oesophagus of each LBP until it reached the bottom of the stomach. Ambient sea water was slowly pumped into the stomach by using a 100 ml syringe attached to the catheter. This was repeated until the mouth of the penguin started to fill up with water and overflow. The total amount of water required to regurgitate each bird was recorded.



Plate 4.2. Regurgitation procedure for Little Blue Penguin. Photo by M.Rensburg 2006.

Once the water began to overflow the tube was removed and the penguin was inverted over an empty 10 L bucket. As the stomach contents flowed out, gentle pressure was applied to the bottom of the stomach to help push out the contents. The neck was massaged with one hand to prevent any bolus becoming stuck. If the regurgitated stomach contents were small and not consistent with the amount of water that was pumped in, then the procedure was repeated. The collected samples were filtered through a sieve covered with a fine mesh stocking and held over a plastic container. The mesh was able to be pulled off and be put into a labelled snap lock bag. A new mesh was used for each sample. The water that was caught in the container was measured with the syringe to see how much water had come out of the bird. Any contents left in the bucket were washed out with more seawater and sieved through the mesh.

4.4.3.2 *Quantification of Food Samples*

After each sample was obtained they were taken back to the laboratory for analysis. The total wet weight (g) was measured with an electronic weighing scale. The sample was then sorted into the different food types: fish (teleosts), squid (cephalopods), crustaceans (decapods) and other items such as shells, stones and feathers were also noted but not included in any later analyses. Each food type was individually weighed.

4.4.3.2.1 *Fish*

Fish species were identified by body morphology and hard parts. Classification of the individual fish species can be difficult since most of the samples were a homogenous paste. However hard parts such as otoliths, mandibles, opercula, and pre-opercula are often in tact and allow for some identification. Any fish that were found complete (whole column and hard parts) were used as reference for identifying individual fish

parts. Fish that were found whole were also measured to gain an idea of prey size. Fish length was from the snout to tail base (Montague and Cullen 1988). Otoliths were sorted into the left and right sides, the length and width were taken, and each one was rated (0 - 1) according to condition (good and bad) respectively. Otolith length was used as an estimate of fish size and fish mass taken from growth curves generated from published equations for South African species of anchovy *Lampanyctodes hectori*, and myctophid *Engraulis japonicus* (Smale *et al.* 1995) (Appendix 4.7.1 & 4.7.2).

4.4.3.2.2 Cephalopods

Identification of squid species was aided by a reference animal that was found intact. This allowed identification of beaks found within the stomach contents. The length of the reference squid body was obtained by measuring from the top of the head to the end of the tentacle as well as measuring the dorsal mantle length and the rostral length of the lower beak.

4.4.3.2.3 Crustaceans

Although crustaceans were found within two stomach samples this was only two legs and these were unable to be identified to species level.

4.4.3.3 **Bait Fisheries**

Annual catch data from the bait fisheries (Pilchard, Anchovy, sprat and squid) were obtained through National Institute of Water and Atmospheric Sciences (NIWA) Plenary Reports 2005.

4.4.4 Isotope Sampling

4.4.4.1 General

Feathers were obtained from penguins that were found during the entire 2005/06 survey period as well as historical samples taken from Auckland's Museum collection. To limit biases museum samples were only used if the sex was known, they were all adults, and were from the Hauraki Gulf area.

Since LBP moult their entire plumage each year during a short period of time, the feathers collected after that time correspond to the diet for the previous year and for the month prior to moult. Samples obtained from fledged chicks represented the period of feather growth, since hatching. Sampling of pre- and post-moult feathers from the same individual provided information about two foraging years. The old feathers relate to diet in 2004 and the new feathers to diet in 2005 (Table 4.1).

Table 4.1. The number (n) of analysed Little Blue Penguin feather samples and stomach regurgitations their corresponding years. Moulting samples were obtained from the same individual (n = 9) and therefore provided two feathers (n = 18). All birds were known to be from the Hauraki Gulf.

| | N | Years |
|-----------------------|-------------------|------------------------------------------|
| Non-penguin | 5 | 2003 – 2005 |
| Historical | 12 | 1886, 1940, 1961, 1994, 2002, 2004, 2005 |
| Moulting | 18 (9 pre & post) | 2004 and 2005 |
| Chicks | 13 | 2005 |
| Regurgitations | 14 | 2005 |

As a comparison to LBP, five terrestrial species of birds found on Tiri were also sampled. Each of these birds related to a different avian trophic level and general foraging diet; Paradise Duck *Tadorna variegata* (herbivore), Tui *Prothemadera novaeseelandiae* (frugivore/ nectivore), Bellbird *Anthornis melanara* (frugivore/ nectivore), Robin *Petroica australis* (insectivorous), and Pukeko *Porphyrio porphyrio* (omnivore).

Before analysis, feather samples were cleaned with ethanol and dried at room temperature. Feathers were then cut up into pieces with steel scissors, weighed and analysed using mass spectrometry. In collaboration with NIWA feather samples were analysed at the NIWA laboratory.

4.4.4.2 Methods used by NIWA

All stable isotope analyses were carried out on a Delta^{Plus} (Thermo-Finnigan, Bremen, Germany) continuous flow, isotope ratio mass spectrometer at NIWA stable isotope laboratory in Wellington. Solid samples were prepared in tin boats and combusted in an NA 1500N (Fisons Instruments, Rodano, Italy) elemental analyser combustion furnace at 1020°C in a flow of oxygen and Helium carrier gas. Oxides of nitrogen were converted to N₂ gas in a reduction furnace at 640°C. N₂ and CO₂ gases were separated on a Porapak Q gas chromatograph column before being introduced to the mass spectrometer detector via an open split ConFlo II interface (Thermo-Finnigan, Bremen, Germany). CO₂ and N₂ reference gas standards were introduced to the mass spectrometer with every sample analysis. ISODAT (Thermo-Finnigan) software was used to calculate $\delta^{15}\text{N}$ values against atmospheric air, and $\delta^{13}\text{C}$ values against the CO₂ reference gas relative to PDB, correcting for ¹⁷O. Percent C and % N values were calculated relative to a solid laboratory reference standard of urea (*Elemental*

Microanalysis, U.K.) at the beginning of each run. Internal standards were routinely checked against National Institute of Standards and Technology (NIST) standards.

4.4.5 Data Analysis

4.4.5.1 Food Samples

The weight of all the food samples were averaged. The total amount of water required to stomach pump each sex was also averaged and the before and after weights of individual LBP sampled were compared with a *t*-test. Unfortunately the total amount of water that was regurgitated was unable to be calculated due to scribing inconsistencies.

All food samples were quantified by a) individual counts of each prey species, b) frequency of occurrence (FOO) (Ashmole and Ashmole 1967; Montague and Cullen 1988;), and c) relative occurrence (RO) (Hasson 1970; Montague and Cullen 1988). The FOO was also applied to the individual fish species relative to the number of times a species was present in the total number of food records containing fish. Details of each calculation are presented below:

1. *Individual counts* were obtained from counts of columns and hard parts. This could also be calculated as a proportion of the total of all individual counts of all species.
2. *Frequency of Occurrence*: Proportion of a particular food taxon (e.g. Anchovy) within samples that only contain the taxon in question (presence or absence) rather than all samples. Since this considers each taxon separately the total proportions of all taxa do not add to 100%. This can also be applied to a single food type (i.e. fish, cephalopods, decapods).
3. *Relative Occurrence*: Is the calculated FOO of a particular taxon divided by the sum of all the FOO's. For example FOO's of species A, B, C are 90, 60 and 40%

respectively, then the RO of A is $90 / (90 + 60 + 40) = 47\%$ (Montague and Cullen 1988).

4.4.5.2 *Species identification*

The anchovy was identified by the spinal column, otoliths and dentary structures. Some fish types were unidentified but classified as different types A, B, C, and D. A fifth classification 'unknown' was used for other types since the columns appeared different from all others and there were no other distinguishing hard parts due to small sample size or digestion.

Analysis of the available otoliths (Figure 4.1) identified fish to 'type' but not down to the fish species. Based on knowledge of fish species in the Hauraki Gulf several species classifications were used. Type A was considered to be either Yellow-eyed Mullet (based on otolith shape) or myctophid species (Lantern Fish) (based on a lower dentary structure). Type B was thought to be goby (species unknown) based on otoliths (distinct square shape, small size, and footprint marks on the ventral surface). Type C and D remained unknown. There was only one species of cephalopod found within all of the samples. All the beaks appeared to be of the same type and were identified as Order Ommastrephidae based on the transparent band that runs vertical from the base of the lower rostrum into the wing. *Nototodarus* sp. (Arrow Squid) is found within the Hauraki Gulf and has been found within the stomachs of LBP in other studies (Montague and Cullen 1988).

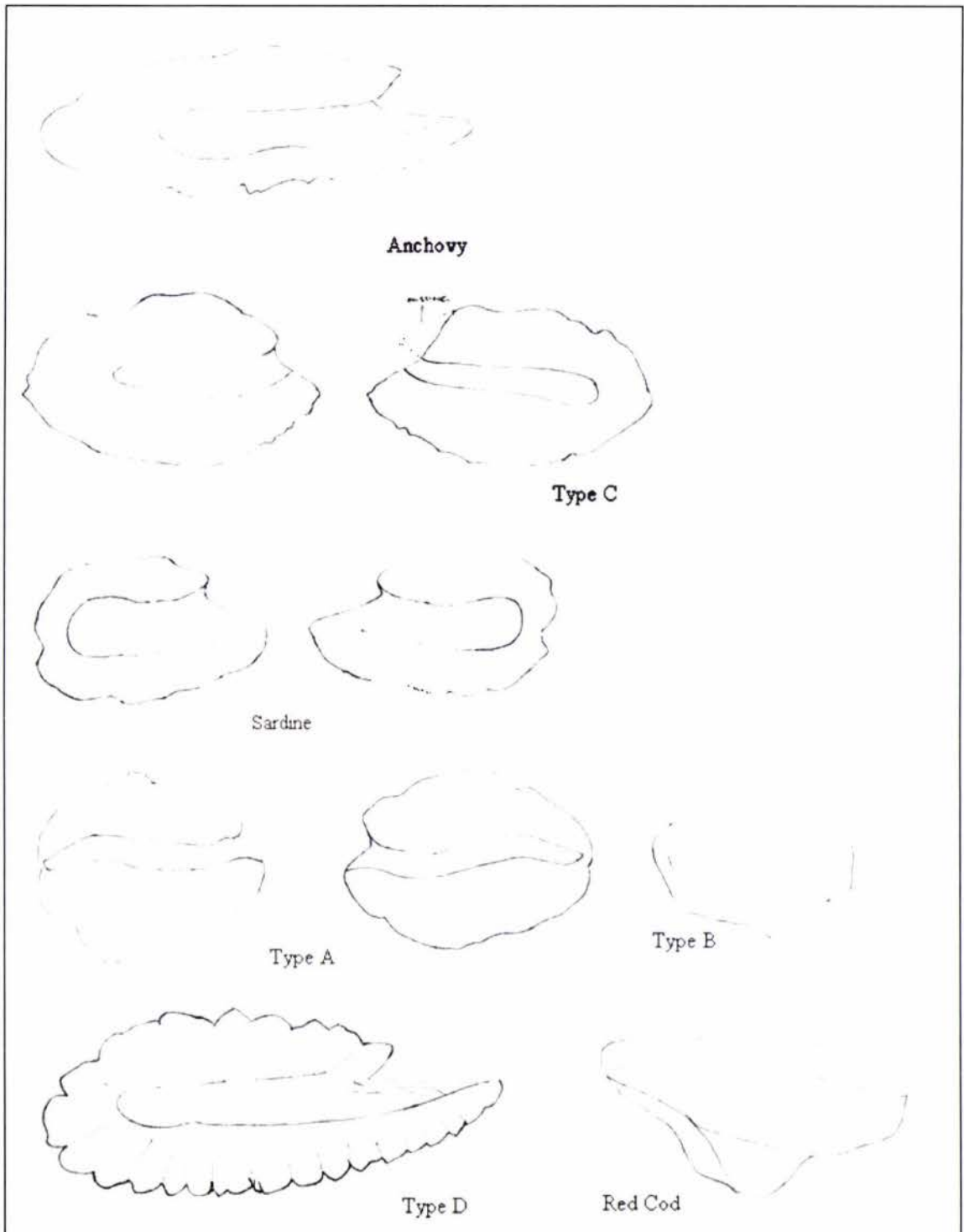


Figure 4.1. Drawings of left and right otoliths from fish species obtained from Little Blue Penguin on Tiritiri Matangi Island during 2006. Lengths of otolith lengths (mm) were; Anchovy (3), Type C (1), sardine (1), Type A (1 to 2), Type B (0.5), Type D (3), and Red Cod (1).

4.4.6 Isotope Analysis

4.4.6.1.1 *Accuracy*

Accuracy and precision data for NIST standard analyses are given in Appendix 4.7.3. The precision on repeat analyses of urea standards during batch analysis of data presented in this paper are given in Appendix 4.7.4.

Repeat analysis of NIST standards produces data accurate to within 0.1 - 0.5‰ for $\delta^{15}\text{N}$ and 0.3 - 0.4 ‰ for $\delta^{13}\text{C}$ and a precision of better than 0.5‰ for N and 0.25‰ for C. For % N and C content data are accurate to within 0.4%, with a precision usually better than 0.3% for N and 0.2% for C.

4.4.6.1.2 *Isotope Analysis*

Isotope values for each year were pooled to avoid pseudo-replication (Hilton *et al.* 2006) therefore means were used in analyses. Values for all years were graphed against other terrestrial bird species from Tiri, as well as other seabird species and sympatrically similar prey types (fish, squid, and crustaceans) found in stomach samples of LBP. These stable isotope values were taken from another study sampled by Sydeman *et al.* (1997) since this information is not available for New Zealand species. It must be noted that krill was included in analysis as a reference for the stable isotope values expected for crustaceans however krill per se were not found within the stomach sample.

The difference between values of moulting adults from 2004 and 2005 were compared using a one-way ANOVA, which was also used to test the difference between values obtained for all 2004 adults and chicks, as well as 2005 adults and chicks.

Trophic levels (TL) for each year were calculated using a fractionation factor of 4‰ for the change expected between diet and feathers (Mizutani *et al.* 1990; Hobson and Clark 1992) and using the equation:

$$TL = 2.5 + (\delta^{15}N - 11.2) / 4 \text{‰}$$

Where TL equals the 2.5 trophic level calculated for krill (Sanger 1987; Sydeman *et al.* 1997), the delta value of nitrogen measured in the mean of feathers for each year, and the fractionation factor of feathers. To compare trophic levels across all species and altered version of the fractionation enrichment factor was also used to take out any discrepancies associated with different tissue sampling and species differences. The altered enrichment factor used was 2.54 ‰ ($\pm 0.11 \text{‰ SE}$) as calculated by Vanderklift and Ponsard (2003).

Differences in the stable isotopes means were calculated for each year. The difference in stable isotope values for each year was also calculated as a proportion relative to the oldest record of 1886 (Auckland Museum). These proportional differences were correlated with the corresponding SOI values for the year by using Spearman's rank in *SAS v.8*. Yearly values for 2004 and 2005 were plotted against other years to identify potential variation of historical samples since sample size was low.

4.5 Results

4.5.1 Regurgitations

A total of 20 birds had their stomach flushed. Three of these were not included in the analysis and were for ensuring a standard practice of the technique and three regurgitated green watery bile which meant that their stomachs were empty. Therefore a total of 14 samples were used for content analyses (seven males, 10 females, and three of unknown sex). The mean weights of birds after regurgitations were larger than that of the initial weights due to hand feeding (Table 4.2).

Table 4.2. Mean (\pm SD) wet weight (g) of the samples obtained from Little Blue Penguin regurgitated and the differences in application of the procedure with each sex. Sample sizes are in parenthesis.

| | Max | Min | Mean |
|---------------------------|----------------------------|-----------------------------|-------------------------|
| Sample weights (g) | 24.84 | 0.06 | 6.23 \pm 2.58 |
| | Male | Female | Unknown |
| Before (g) | (6) 861.78 \pm 156.40 | (9) 1094.17 \pm 243.89 | (2) 755 \pm 134.35 |
| After (g) | 1120 \pm 282.14 | 929 \pm 65.80 | 660 |
| Water in (ml) | 637.5 \pm 93.22 | 276.33 \pm 170.11 | 375 |

A total of 12 marine species were found within the stomach samples of LBP and were similar for both sexes (Figure 4.2). The most common food type was fish which was found in 85.71% (12/ 14) of samples. The second most common food type was cephalopods 42.86% (6/ 14). While crustaceans were present in some of the samples 14.29% (2/ 14) these were only represented by two legs found which were unidentifiable. Copepods were found in 35.71% (5/ 14) of the samples (Table 4.3).

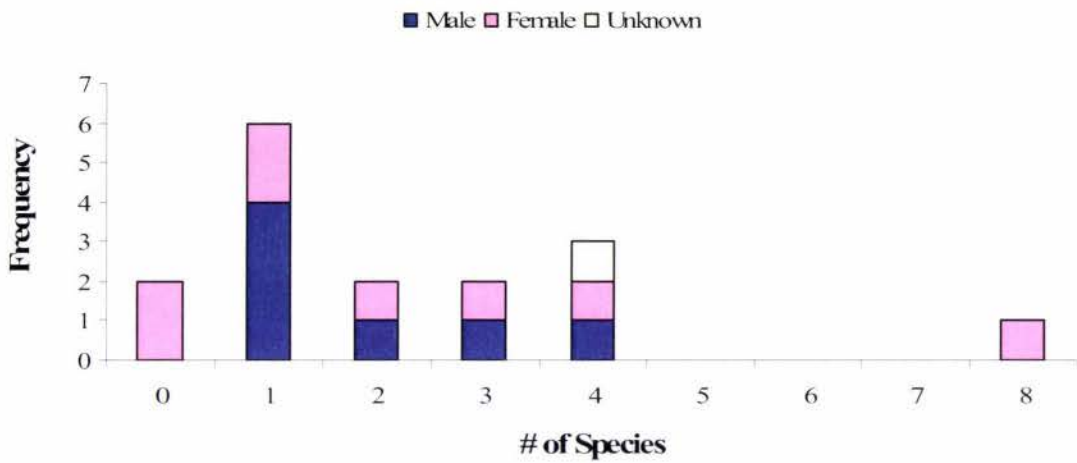


Figure 4.2. Number of prey species in stomach samples of males and females Little Blue Penguin during 2006 on Tiritiri Matangi Island, New Zealand.

Table 4.3. Proportion of the main prey types within the total number of samples ($n = 14$). Note a fish record refers to the number of times that fish is found with a total of samples.

| Number of samples containing: | n | % |
|-------------------------------|----|-------|
| Teleosts | 12 | 85.71 |
| Cephalopods | 6 | 42.86 |
| Crustaceans | 2 | 14.29 |
| Copepods | 5 | 35.71 |
| Number of fish records | 23 | |

Individual counts of prey items found fish type B (27%) was the most abundant followed by anchovy (24.32%) and fish type A (18.92%) (Table 4.4). However, based on FOO and RO values anchovy (21.88%, 19.44%) and cephalopods (18.75%, 16.67%) make up the largest portion of the LBP diet, followed by fish type A (15.63%, 13.89%), copepods (15.63%, 13.89%), and fish type B (12.50%, 11.11%) (Table 4.5). The FOO proportions (calculated from fish records only) found anchovy (30%), type A (21.7%) and type B (21.7%) occurring most frequently (Table 4.6).

Table 4.4. Individual counts and their proportions of the total number of samples. The otolith length and mass could be used to calculate the actual prey total body length and mass for some species (marked with an *), those that are not marked represents the Otolith length therefore no mass, and those marked with ** represents species length based on columns since no otoliths were found. Means for Anchovy based on (n = 23) otoliths and type A using myctophid values (n = 2).

| | Species | Length (mm) | mass (g) | N | % |
|--------------------|-------------|-------------|----------|-----|-------|
| Fish | Type B | 0.8 | | 50 | 27.03 |
| | Anchovy | 56.6* | 27.4* | 45 | 24.32 |
| | Type A | 0.07* | 24.5* | 35 | 18.92 |
| | Unknown | 25** | | 15 | 8.11 |
| | Type C | 2.5 | | 2 | 1.08 |
| | Type D | 4.5 | | 1 | 0.54 |
| | Red Cod | 1.5 | | 1 | 0.54 |
| | Sardine | 1 | | 1 | 0.54 |
| Cephalopods | Arrow Squid | 38 | | 13 | 7.03 |
| Copepods | | | | 15 | 8.11 |
| Crustaceans | | | | 2 | 1.08 |
| Flies | | | | 5 | 2.7 |
| Total | | | | 185 | |

Table 4.5. Calculations of the FOO (%) and RO (%) of all species found within the samples. Calculated from the total number of records (n = 32) that all items are present within the total number of samples (n = 14). For example FOO Anchovy = $7/32 \times 100$, and RO $21.88/112.50 \times 100$.

| | # samples | FOO (%) | RO (%) |
|-------------|-----------|---------|--------|
| Anchovy | 7 | 21.88 | 19.44 |
| Type A | 5 | 15.63 | 13.89 |
| Type B | 4 | 12.50 | 11.11 |
| Unknown | 2 | 6.25 | 5.56 |
| Type C | 1 | 3.13 | 2.78 |
| Type D | 1 | 3.13 | 2.78 |
| Red cod | 1 | 3.13 | 2.78 |
| Sardine | 1 | 3.13 | 2.78 |
| Cephalopods | 6 | 18.75 | 16.67 |
| Copepods | 5 | 15.63 | 13.89 |
| Crustaceans | 2 | 6.25 | 5.56 |
| Flies | 1 | 3.13 | 2.78 |
| | | 112.50 | 100 |

Table 4.6. The number of stomach samples (n) containing each fish type and the proportion (%) that each one makes up from the total number of samples (food records) that contain fish (n = 23).

| Fish | N | % fish diet |
|-------------|----------|--------------------|
| Anchovy | 7 | 30 |
| Type A | 5 | 21.7 |
| Type B | 5 | 21.7 |
| Unknown* | 2 | 8.7 |
| Sardines | 1 | 4.3 |
| Type C | 1 | 4.3 |
| Type D | 1 | 4.3 |
| Red Cod | 1 | 4.3 |
| Total | 23 | 100% |

4.5.2 Isotope Sampling

4.5.2.1 Diet Assessment

The ratios of $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ for the terrestrial bird species were considerably lower than those for LBP. The isotopic values for LBP are larger than potential prey species, although there is overlap of both $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values of LBP with that of the Anchovy and Sardine.

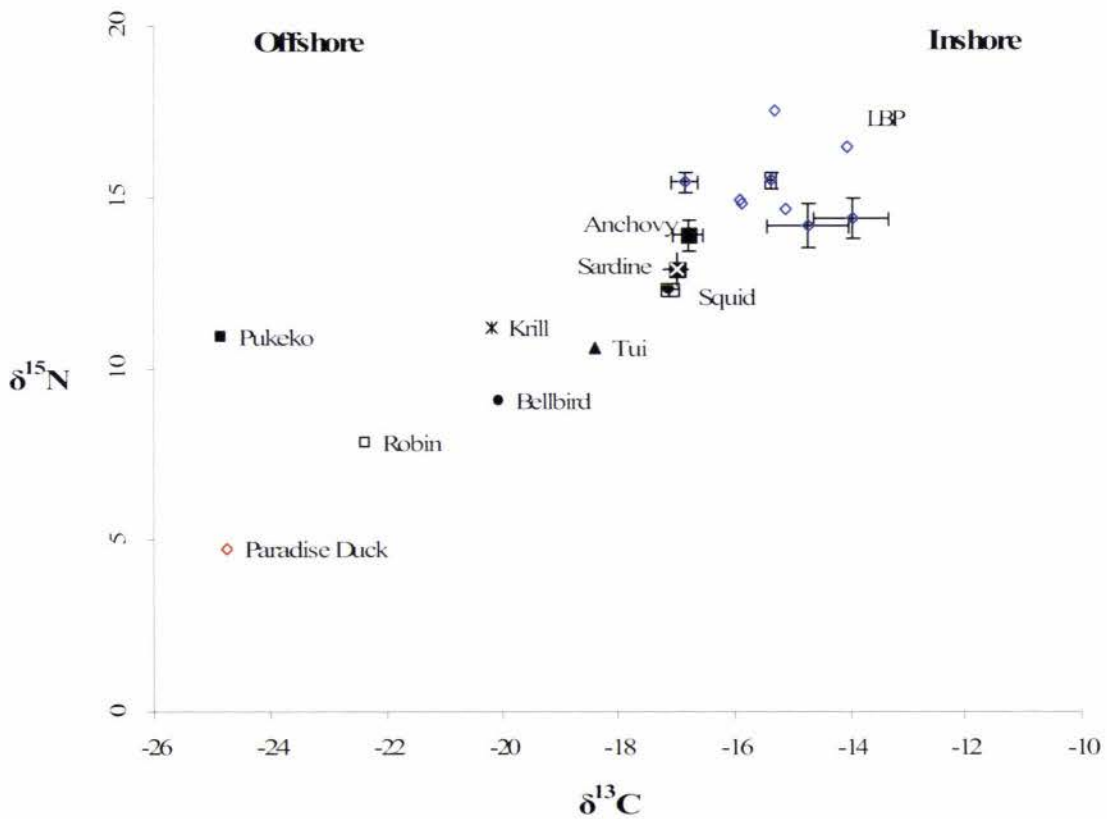


Figure 4.3. Comparison of the Isotope means for LBP (\diamond) for 1886-2005 ($n = 62$), Anchovy (\blacksquare), sardine (\blacksquare with an x in centre), squid (\bullet), krill ($*$), Tui (\blacktriangle), Bellbird (\bullet), Robin (\square), Pukeko (\bullet), and Paradise Duck (\diamond) (\pm SE).

4.5.2.2 Individual Diet Assessment

The pre-moult (2004, $n = 20$) was significantly different from the post-moult (2005, $n = 14$) for $\delta^{13}\text{C}$ ($F_{1,31} = 118.63$ $p = <0.0001$) but not $\delta^{15}\text{N}$ levels ($F_{1,31} = 0.06$ $p = 0.8147$). There was no difference in the mean (\pm SE) $\delta^{13}\text{C}$ levels between 2005 adults ($n = 14$) $\bar{x} = -16.85 \pm 0.67$ compared to 2005 chicks ($n = 13$) $\bar{x} = -16.88 \pm 0.17$, ($t = 1.725$, $df = 20$, $p = 0.898$), or for the $\delta^{15}\text{N}$ between chicks $\bar{x} = 14.939 \pm 0.13$ ($n = 13$) and 2005 adults $\bar{x} = 15.454 \pm 1.21$ ($n = 23$) ($t = 1.745$, $df = 20$, $p = 0.116$) (Figure 4.4).

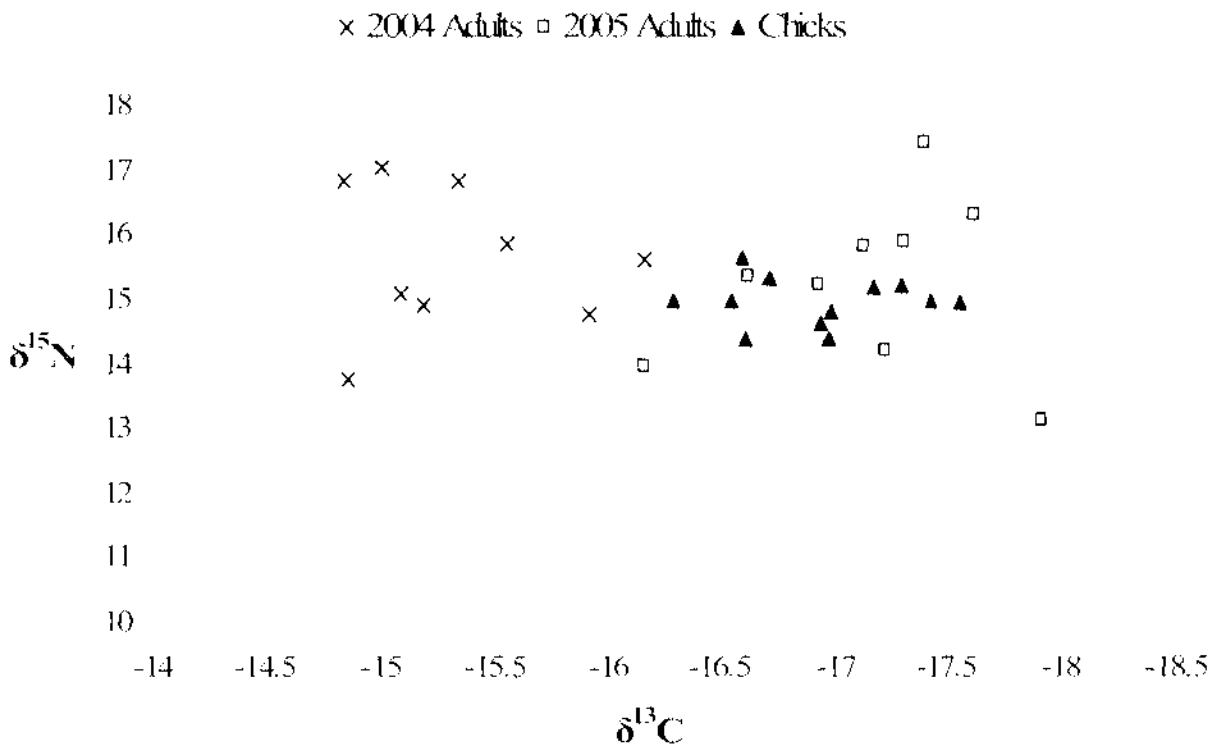


Figure 4.4. Stable isotope signatures from adult Little Blue Penguin feeding during 2004 and 2005 and chicks that fledged from 2005 breeding season.

The regurgitated birds were separated by time into two sampling periods due to the separation in sampling times. January and February ($n = 10$) had higher stable-carbon (-14 to -16%) levels than the birds sampled in April and May (-16.5 - -17.5) ($n = 5$) (Figure 4.5). These were assigned to 2004 (January & February) and 2005 (April and May).

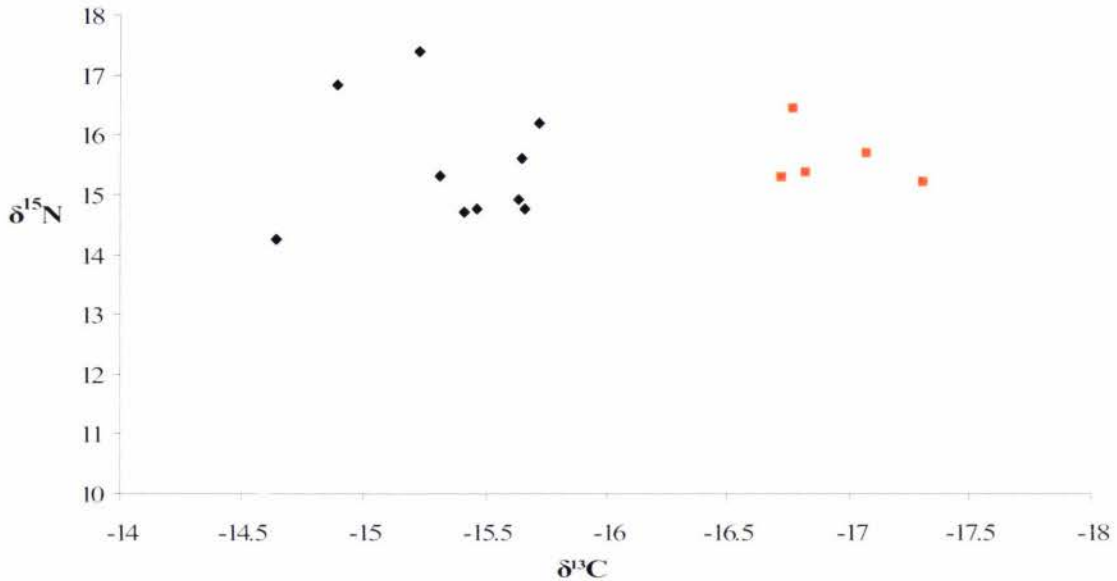


Figure 4.5. Stable isotope measurements from regurgitated Little Blue Penguin showing clear separation of sampling seasons with birds sampled during January and February (black \diamond) separated from sampled birds during April and May (red \square).

4.5.2.3 Trophic levels

The stable isotopes values for LBP were compared with a range of species of different trophic levels (Table 4.7). Comparisons of the trophic levels (TL) reveals Sealions to have the largest (TL = 5.3, altered TL = 5.9) with Krill (2.5) and Squid (2.9) being the lowest. LBP have atrophic level within these two (TL = 3.3 to 4, altered TL = 4.1).

Calculations of TL for all the years showed that there were fluctuations from 3.29 TL to 4. The lowest TL (3.29), ($n = 3$) was found in 1994 and the highest TL (4.08), ($n = 1$) was in 2002 (Appendix 4.7.5).

Table 4.7. Stable $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ isotope abundance ($\bar{x} \pm 1 \text{ SE}$) with associated trophic levels. Those marked with an * are from the Gulf of Farallones, California food web (see Sydeman *et al.* 1997). Trophic levels for Little Blue Penguin are calculated from all years (1886 to 2005). Altered trophic level as suggested by Vanderklift and Ponsard 2003.

| Species | N | $\delta^{13}\text{C}$ | $\delta^{15}\text{N}$ | Trophic Level | Altered TL |
|----------------------------|----|-----------------------|-----------------------|---------------|------------|
| <i>Crustaceans</i> | | | | | |
| Krill* | 5 | -20.2 \pm 0.2 | 11.2 \pm 0.3 | 2.5 | 2.5 |
| <i>Cephalopods</i> | | | | | |
| Market Squid* | 5 | -17.1 \pm 0.1 | 12.3 \pm 0.2 | 2.9 | 2.9 |
| <i>Fishes</i> | | | | | |
| Pacific Sardine* | 3 | -17 \pm 0.3 | 12.9 \pm 0.5 | 3.1 | 3.2 |
| Northern Anchovy* | 4 | -16.84 \pm 0.3 | 13.9 \pm 0.5 | 3.4 | 3.6 |
| <i>Herbivorous Birds</i> | | | | | |
| Paradise Duck | 1 | -24.7 | 4.7 | | 0.1 |
| <i>Omnivorous Birds</i> | | | | | |
| Pukeko | 1 | -24.82 | 10.93 | | 2.4 |
| <i>Insectivorous</i> | | | | | |
| Robin | 1 | -22.38 | 7.85 | | 1.2 |
| <i>Nectarivorous</i> | | | | | |
| Bell Bird | 1 | 20.07 | 9.07 | | 1.7 |
| Tui | 1 | -18.38 | 10.64 | | 2.3 |
| <i>Planktivorous Birds</i> | | | | | |
| Cassin's Auklet* | 7 | -18.2 \pm 0.5 | 13.9 \pm 0.5 | 3.4 | 3.6 |
| Common Murre* | 3 | -16.9 \pm 0.5 | 14.8 \pm 0.5 | 3.7 | 3.9 |
| Little Blue Penguins | 63 | -15.27 \pm 0.3 | 15.30 \pm 0.4 | 3.3 - 4 | 4.1 |
| <i>Piscivorous Birds</i> | | | | | |
| Rhinoceros Auklet* | 6 | -17.7 \pm 0.7 | 16.9 \pm 0.5 | 4.3 | 4.7 |
| Brandt's Cormorant* | 7 | -15.9 \pm 0.3 | 17.3 \pm 0.2 | 4.5 | 4.9 |
| <i>Mammals</i> | | | | | |
| Northern Sealion* | 5 | -15.2 \pm 0.5 | 19.8 \pm 0.6 | 5.3 | 5.9 |

4.5.2.4 Long term diet change

The SI values of LBP from 2005 were higher than all other years in $\delta^{13}\text{C}$ levels (Appendix 4.7.6) whereas 2002 was higher than that of all other years in the $\delta^{15}\text{N}$ values (Appendix 4.7.7). The largest deviations from 2002 occurred between 1947, 1949, and 1994, whereas the largest deviations from 2002 occurred between 1949, 1994, and 1887.

The year 2005 had the largest proportional deviation from 1886 in $\delta^{15}\text{N}$ (Figure 4.6a) while 2002 for $\delta^{13}\text{C}$ (Figure 4.6b). Levels of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ fluctuate over the 120 years. Mean levels of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ for each year were correlated with corresponding yearly Southern Oscillation (SOI) values and not significant (Spearman's rank, $\delta^{15}\text{N}$ $n = 9$, $r_s = -0.083$, $p = 0.831$, $\delta^{13}\text{C}$ $n = 9$, $r_s = 0.250$, $p = 0.517$).

Comparisons of all years against the 2005 and 2004 samples showed that the trophic levels ($\delta^{15}\text{N}$) of LBP over 120 years had not changed, but 2005 was a lot lower than 2004. $\delta^{13}\text{C}$ levels also dropped from 2004 to 2005 (Figure 4.7).

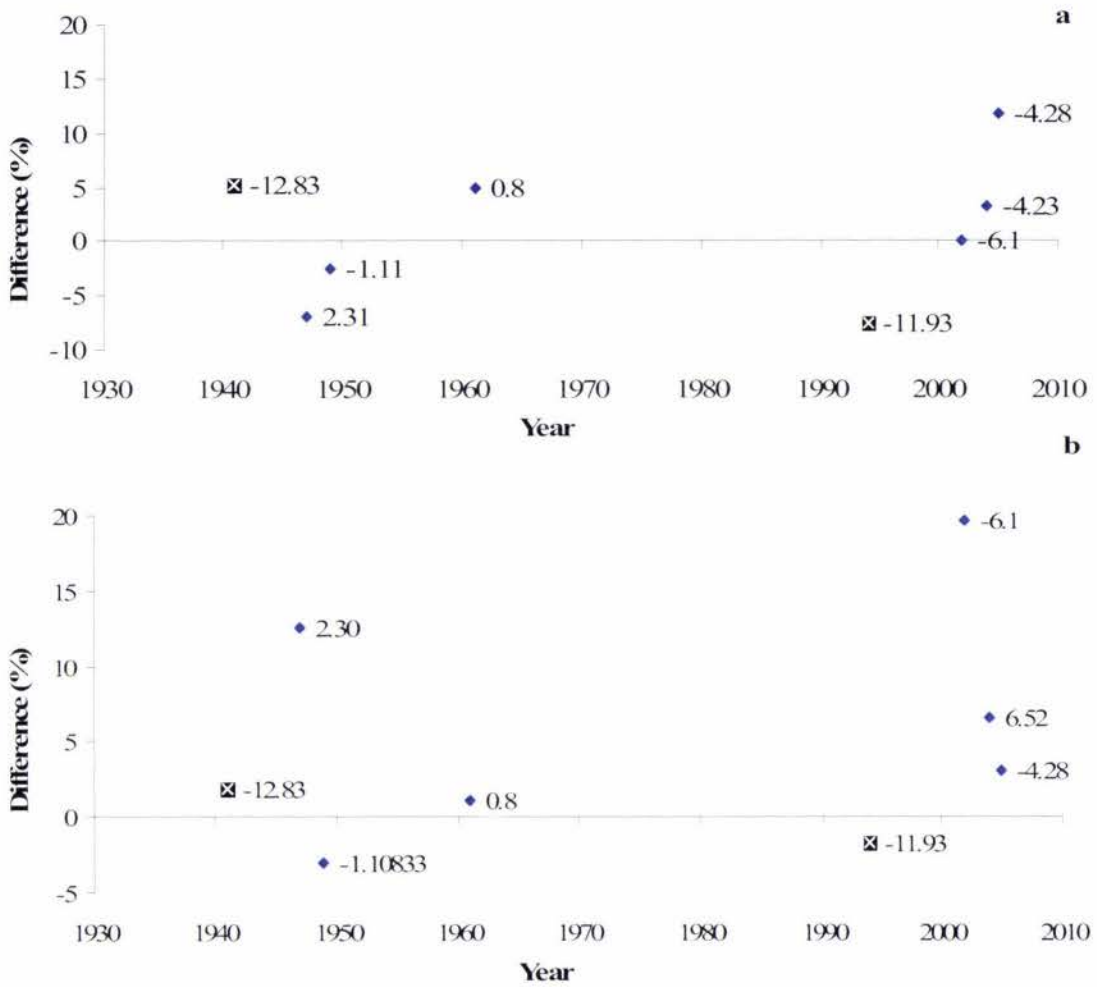


Figure 4.6. a) $\delta^{15}\text{Nitrogen}$ & b) $\delta^{13}\text{Carbon}$: The percentage (%) difference (increase or decrease) in nitrogen and carbon signatures for each year from 1886 has fluctuated. The SOI values are next to the data point and those that reached *El Niño* conditions for any year is represented by a black box with a white square on the inside as opposed to years of normal SOI conditions which is represented with a dark diamond shape (\diamond).

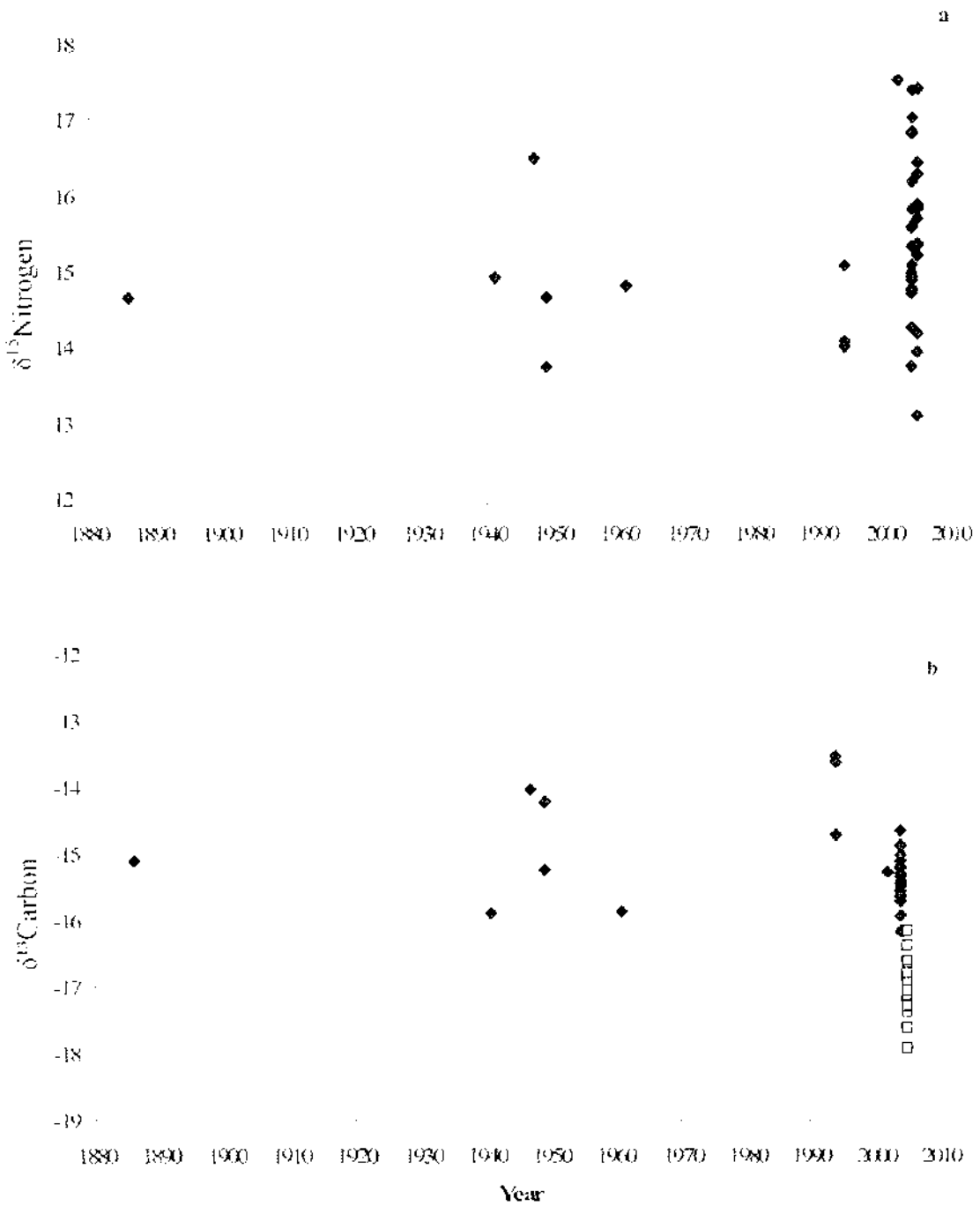


Figure 4.7. Comparison of the different yearly stable isotope values. Figure a) shows that the $\delta^{15}\text{N}$ values for historical samples fall within 2004 and 2005, whereas b) shows that the $\delta^{13}\text{C}$ levels of 2005 (•) is much lower than all other years (black \diamond).

4.6 Discussion

4.6.1 The Diet of LBP

The stomach contents of Little Blue Penguins contained a diverse array of prey types similar to those in other studies of LBP subspecies (Montague and Cullen 1988). The generalist feeding pattern of LBP could result from factors such as prey availability (daily, seasonal, and annually), the individual ability to obtain food (age, experience), competition with other marine species and human catch efforts and anthropogenic influences (eg: runoff, dredging and sewage) and restrictions (time, foraging area). Each of these factors varies in spatial and temporal scales and contributes to variability in the LBP diet.

Isotope signatures of $\delta^{15}\text{N}$ within the feathers show that the LBP within the Hauraki Gulf feed at trophic levels similar to seabirds feeding on high planktivorous diets. A planktivorous diet means that birds are foraging on prey items (larval fish, copepods, and crustaceans) that are feeding on planktivore. This is lower than expected for a top predator which should have a more piscivorous diet. The trophic level of the LBP based on the enrichment factor for feathers (4 ‰) as identified by Mizutani *et al.* (1990) (Sydeman *et al.* 1997) suggests LBP are feeding at a trophic level of 3.3-4 (lower than certain fish types). This is similar to the altered trophic level (TL = 4.1) which should account for different tissues tested for each species and also differences that could exist in C/N levels across the different comparable species. In contrast, fish was found to be the largest prey type within regurgitated food samples (85.71%) with the most common species being Anchovy, potentially Yellow-eyed Mullet (type A) and Goby sp (type B). These are all species that occur throughout New Zealand's coastlines

and harbours (Thompson 1981). Surprisingly no pilchard was found within the diet and will be discussed further in section 4.5.3.3).

Several explanations for this contradiction are possible. Firstly, there are biases associated with the stomach regurgitation procedure, secondly the sample sizes in both sampling methods, and thirdly the enrichment measures for prey species and the sampling times may vary.

4.6.1.1 Biases associated with the conventional method

There are several biases associated with the conventional method of dietary analysis. Firstly large prey items can be over represented in the samples due to digestion of the smaller prey items and secondly, hard parts aid the identification of the larger teleost fish species. Lastly, there were biases in retrieving the whole stomach sample.

4.6.1.1.1 Prey species identification

Several fish species within this study were initially unidentified since the otoliths and other hard parts were unknown. However further investigation and identification from experienced personal helped with the most likely species identification from the otolith drawings.

The use of otoliths and other hard parts as diagnostic features of teleost fish species is important (Gales 1988) as they are species-specific, and can be used to identify prey age, total length and mass. Certain biases such as digestion rate and ontogenetic differences need to be taken into account when using them in dietary studies (Grenfell 1982 unpubl. data). Otolith condition decreases with increased retention time in the stomach (Gales 1988) hence identification and quantification of prey types can be difficult to determine. The absence of otoliths within faeces of LBP suggests that they are completely digested but this may vary for other penguin species

(Gales 1988). Variations in digestibility of different prey types and species also need consideration since cephalopod beaks are considered to be less digestible than fish (Jackson and Ryan 1986; Furness *et al.* 1984; Van Heezik and Seddon 1989). Because fish tissue and otoliths are digested faster than squid beaks, fish are often assumed less important within a particular diet while over estimating cephalopods. Furthermore, smaller prey items such as copepods and other planktivorous species that do not have identifiable hard parts could go undetected such as the crustaceans that were found within some of the samples within this study.

Studies on other seabird diets have found variation in residence times of beaks versus otoliths (e.g. Laysan Albatross *D. immutabilis*: Furness *et al.* 1984), with otoliths only lasting 24 hours to a couple of days (e.g. Thickbilled Murre *Uria lomvia*: Upenski 1956; Duffy and Laurenson 1983; Furness *et al.* 1984, and Cape Cormorants *Phalacrocorax capensis* Furness *et al.* 1984). Fish length and mass are often underestimated by otoliths even after only 1-2 hours retention time, but this can be overcome by only using otoliths that appear in good condition (Gales 1988). In general care needs to be taken when analysing stomach contents based on hard parts while acknowledging the differences that can occur between otoliths (ontogenetic differences), digestibility, and between species.

The individual counts and the FOO can be misleading (Montague and Cullen 1988) however some measure of prey items is useful (Hyslop 1980). The use of the conventional approach for diet analysis is often difficult to obtain and quantification is never free from bias or error (Hyslop 1980). This may implicate some of the results however this is the first documentation of what species LBP from Tiritiri Matangi population are feeding on.

4.6.1.1.2 *Extraction of stomach contents*

The extraction of stomach contents with only one flush has been found to cause bias in sampling since LBP may not regurgitate the entire stomach contents on the first flush. If contents are obtained, it is often the more recently consumed prey items that are regurgitated first. Therefore to collect an unbiased sample a LBP may require flushing multiple times (Gales 1985). However, in this study handling time was minimised by only flushing individual birds up to two times to limit stress. Regardless of this, samples still provided an estimate of actual prey species that LBP have been foraging on during the months that they were sampled.

4.6.1.2 *Sample sizes*

Sample sizes of historic samples were very small and it is possible that isotope values could reflect individual preferences however unlikely that 63 samples would have low stable isotope values. Further evidence of yearly differences rather than individual differences comes from the fluctuations of stable isotope values compared to the 2004 and 2005 years. If the sampled feathers of historical studies were potentially similar to 2004 or 2005 we would expect the values to fall within the confidence intervals. This was the case for the $\delta^{15}\text{N}$ values of all years reinforcing the trophic levels of LBP however this was not the case for the of $\delta^{13}\text{C}$ which suggests a drop in productivity of the Hauraki Gulf during 2005. This may further explain the die-off that occurred during 2005 suggesting low food availability.

Furthermore, the chicks correlated with adults from 2005, however the adults were not associated with the chicks (were not parents). The 2005 LBP values were significantly different than the 2004 LBP and therefore reflected an annual similarity of diet within the LBP diet rather than individual differences.

If LBP within the Hauraki Gulf were feeding on the same dietary components over the past 120 years then the stable-isotope values in the feathers would be estimated as being consistently similar. Tissues from Mouse (*Mus musculus*) (Tieszen and Fagre 1993), and birds (Hobson and Clark 1992; Bearhop *et al.* 2002; Poldesak *et al.* 2005) have shown that they reflect the signatures of the diet when fed isotopically consistent diets. Feather isotopes from LBP over a time-scale of 120 years, suggests that catches of similar prey types have not been consistent.

Also, if LBP were feeding primarily on anchovy, then the $\delta^{15}\text{N}$ should be expected to be a trophic level higher as $\delta^{15}\text{N}$ values increase in a stepwise fashion as you move up the food chain. For example the Rhinoceros Auklet which fed primarily on Anchovy, have a trophic level of 4.3 (or altered 4.7). Similar trophic levels do not necessarily mean the same diet, but can assess trends. A positive relationship of increasing $\delta^{15}\text{N}$ values have also been found between prey quality and the stable-nitrogen values for Magellanic Penguins (*Spheniscus magellanicus*) (Hilton *et al.* 2006).

Fluctuations in the $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ levels within feathers of LBP since the oldest record (1886) showed that the type of prey LBP are taking has varied over 120 years and could be associated with changes in the proportional composition of different prey types consumed (fish %, cephalopods %, and crustaceans %). Studies have found that individuals feeding on a mixed diet will have higher stable-isotope signatures (Podlesak *et al.* 2005) Furthermore, results show that the $\delta^{15}\text{N}$ overlap with anchovy and sardine values suggesting that LBP have been feeding on a diet lower than these fish.

4.6.1.3 Isotope signatures and sampling period

Isotope comparisons made on bird species were used to estimate trophic level for LBP. Similarities within isotope values between species does not necessarily imply the same

diet, however, it can be used to indicate trophic level association (Hobson *et al.* 1994) and may also help identify productivity level changes. Comparisons of LBP data with other terrestrial bird species showed that the more terrestrial bird species exhibited lower stable isotope values consist with low density dietary material such as grass, insects, honey, as opposed to fish. In addition, the carbon levels of the terrestrial bird species show similar levels to that of birds feeding on low protein diets. For example, Japanese Quails (*Coturnix japonica*) and the American Crow (*Corvus brachyrhynchos*) fed wheat and corn have $\delta^{13}\text{C}$ values of; Quail (-24.1, -19.5) and Crow (-23.7, -20.1) respectively (Hobson and Clark 1992). This comparison with other terrestrial birds is a good comparison of the different diets and enrichment levels.

Using the $\delta^{15}\text{N}$, the LBP are most similar to seabird species that have a planktivorous diet (of fish or squid) such as the Cassin's Auklet and Common Murre. Seabirds with this diet are expected to have stable-nitrogen values of around 16.4 and -17.9‰ (Hobson *et al.* 1994); levels considerably higher than the LBP values for all years (15.30 and -15.37‰). The Cassin's Auklet has a primary diet of crustaceans and the Common Murre has a primary diet of Rockfish (*Sebastes jordani*) and Northern Anchovy (*Engraulis mordax*) (Sydeman *et al.* 1997). Seabirds with a more piscivorous diet have trophic levels of 4.3 (altered TL = 4.7) and 4.5 (altered TL = 4.9), again higher than those found for LBP during this study. Piscivorous seabirds include the Rhinoceros Auklet which has a principal diet of Northern Anchovy, Rockfish, and sardines (*Sardinops sajax*) (Sydeman *et al.* 1997). Further support for a planktivorous fish diet in LBP is from a study on Magellanic Penguins from the Argentinean Patagonian coast where measures of $\delta^{15}\text{N}$ (17.8 to 20.00) and $\delta^{13}\text{C}$ (-18.1 to -14.8) were reported due to the high proportion of anchovy (69% male and 67% female) in adult diets (Forero *et al.* 2002).

The major prey species found within the stomach regurgitations of LBP was anchovy which has been given a $\delta^{15}\text{N}$ value of 13.09 ± 0.8 (Sydeman *et al.* 1997) (altered TL = 3.6), however values for other fish species found within the regurgitations (Table 4.4) were not obtainable from the literature. One of the limitations to comparisons between conventional methods and stable isotope values is that prey signatures are required as well as the enrichment levels between tissues. Isotope values from comparable bird species included tissues from eggs as opposed to feathers. This may not affect the trophic level assessment since the altered trophic level comparisons calculated from a metaanalysis approach (Vanderklift and Ponsard 2003), takes into consideration differences of tissues and species within results between the two trophic level calculations being similar (Table 4.4). The isotopic signatures for fish species and fractionation values used for the different types of tissues have also found to be similar for the same species and tissue types. Furthermore trophic enrichment values used here came from Barkley Sound and Alaskan waters since they have not been measured for New Zealand. Comparisons could potentially be different between these locations due to different environmental enrichments (upwelling and coastal advection). However the trophic enrichment values found within Alaska and Barkley Sound are similar for those reported in other food webs (Schoeninger and DeNiro 1984; Wada *et al.* 1987; Sholto-Douglas *et al.* 1992; Hobson and Welch 1992; Hobson *et al.* 1994).

4.6.1.4 Application of enrichment values

The precision of estimates of trophic level using enrichment values can be influenced by differences between organisms that include: the biochemical method of nitrogen excretion for different organisms (e.g. ureotelic, uricotelic, ammonotelic, and guanotelic), diets, taxonomic classifications, environment (e.g. freshwater, marine or terrestrial), tissue analysis, % nitrogen content in food source, and C/N ratio of food

source (Vanderklift and Ponsard 2003). However, for the comparisons made in this study only the C/N ratio of the food source is expected to differ significantly. Seabirds are considered *terrestrial* due to breeding occurring on land and all species compared use uricotelic excretion. Furthermore trophic level assessment was calculated two ways which accounts for tissue differences and species differences. Environmental differences could not be controlled in the current study and there is clearly a need for a greater understanding of enrichment values for local prey species for particular regions as currently all values come from Northern hemisphere sources with potentially varying enrichment levels.

A meta-analysis approach to understanding sources of variation in consumer-diets based on $\delta^{15}\text{N}$ calculated an overall enrichment value of 2.54‰ ($\pm 0.11\%$ SE) for all organisms (i.e. mammals, birds, crustaceans, insects, and fishes) (Vanderklift and Ponsard 2003). However, Vanderklift and Ponsard's value (2003) was used in this study as a comparison to the more refined estimates that were available for just seabirds and feather fractionation as used in all LBP samples. This approach covered potential differences suggested earlier. However, for future general investigations into LBP and their trophic level changes over small and large scales, it is recommended that the Vanderklift and Ponsard (2003) version of the $\delta^{15}\text{N}$ is applied or specific enrichment values calculated for the species, tissue and prey being tested. Further trophic level comparisons need to control for potential variation that could occur as proposed by Vanderklift and Ponsard (2003).

4.6.1.5 Sampling period

Penguins are hard study subjects for diet analysis since they do not frequently come ashore during the non-breeding season. Although seasonal variations have been

associated with LBP (Montague and Cullen 1988) most work is often associated with breeding birds which is often biased towards high trophic levels (Hobson 1993; Hobson *et al.* 1994).

Stomach sampling within this study occurred near the end of the breeding season (January-February) and within the moulting period (March-April), however due to the small sample size no comparisons were made between the different months. Sampling times may explain the large diet of fish and the birds with empty stomachs due to seasonal changes in prey availability or due to individual ages of birds. But whether this was due to seasonal prey availability or individual ability to obtain food is unknown. For example LBP feeding in Australia during the breeding season have been found to feed mainly on fish but outside the breeding season (March and July) penguins were mainly foraging on cephalopods and crustaceans (Chiaradia *et al.* 2003). Individual birds could vary in their diet depending on whether they are recently fledged and therefore less experienced at feeding, or if they are foraging to feed chicks or store for moult. Younger birds do not moult till the following year and therefore may not be pressured to forage everyday.

The regurgitations were performed within the months that LBP would have foraged before moulting and should therefore represent the time period associated with feather growth. Therefore differences between the stomach samples and isotopes may not be best explained by the sampling season. Although, the sample size of the regurgitations was much lower than that of past studies investigating the diet of LBP (i.e. Montague and Cullen 1988) and therefore the birds captured may not have been representative. The contradictory results in LBP diet between the stomach samples and stable isotope measures is more likely a function of sample size of the stomach samples.

4.6.2 Stomach Regurgitations

4.6.2.1 Generalist feeding pattern in LBP

Despite biases there are advantages to using the conventional method. The conventional method can identify prey species which when linked with known behaviours of predators may allow for a greater understanding of the predator-prey interactions that occur within the marine environment. Although this may be based on assumptions it will still give leeway into areas of possible future research.

Based on the stomach samples the preferred prey of LBP in this study and other studies is shoaling fish species which is efficient to obtain in large amounts (Montague and Cullen 1988). However, other items were found which suggests a generalist diet. The generalist feeding pattern of the LBP is thought to be part of the reason that the populations within Otago are successful (Periman *et al.* 2000) as this enables them to take the prey items based on abundance. Prey switching may be why LBP and other bird species (e.g. Brandt's Cormorant, *Phalacrocorax penicillatus*, Pelagic Cormorant *Stictocarbo pelagicus* and Pigeon Guillemots *Cephus Columba*, Ainley *et al.* 1995; Sydeman *et al.* 1997) are able to attempt breeding within years of poor prey availability (Chiaradia *et al.* 2004). This is often associated with variable breeding success, but the question still remains as to how these species and LBP can switch between trophic levels (Sydeman *et al.* 1997). Prey availability can vary as a function of the prey lifecycle, and the foraging potential of LBP to obtain food.

4.6.2.2 Effect of prey life-cycle

The size range of prey taken is, generally between, 3 – 12 cm for LBP (Cullen *et al.* 1992) but was found to be the smaller range 2-3 cm for this study. This suggests they have been feeding on younger fish (in Northern New Zealand 2-5 year old fish are 8-14

cm) (Plenary Report Appendix 4.7.8). For example the size of juvenile Pilchard found within Leigh Marine Reserve is 45 mm, Yellow-eyed Mullet is 15-30 mm (Thompson 1981). The size ranges obtained in samples were complimentary to the $\delta^{13}\text{C}$ values which together suggest inshore foraging of the LBP.

The composition of prey items taken by LBP could be due to the life-cycle of particular prey items. Anchovy was found to be abundant within the samples which occur around much of New Zealand and Australian coasts, inshore bays, gulfs, and harbours (Thompson 1981). The life-cycle of Anchovy involves movement to estuaries to spawn over the summer months, with highest abundances in surface waters during January and March (Thompson 1981). Most other shoaling fish (e.g. yellow eyed mullet and pilchard) move seaward to deep waters to spawn followed by a return to shallow water (Peterson *et al.* 2006).

Yellow-eyed Mullet are also thought to be abundant around the river mouths and coasts during December and March (Thompson 1981). Adult pilchards are taken by LBP in Australia between August and December and post-larval Anchovies between January and June (Montague and Cullen 1988). No pilchard was found within the diet of LBP within this study and this could be due to the sampling period (January to April) which would be when pilchards may be spawning (November to February).

Furthermore, some fish species have temperature-dependent gonadal development (Ware and Tanasichuk 1989; Sims *et al.* 2004) and will spawn and migrate earlier in warmer years (Sims *et al.* 2004). Although, changes in temperature between seasons is not always large a 1-2°C difference can cause shifts in migration and spawning. This sort of synchronisation in aquatic ectotherms is important due to the seasonal changes in food availability (Cushing 1982; Cushing 1990; Sims *et al.* 2004) but may cause a shift in behaviours of secondary consumers. For example, during the

winter Macaroni Penguins *Eudyptes chrysolophus* (Brandt 1837; Green *et al.* 2005) have been found to dive deeper and for greater durations than in summer which could be related to the vertical movements and prey availability during those months (Green *et al.* 2005). Temperature may also impact on the survival of fish species rather than just changing their spawning pattern (Jutila *et al.* 2005).

Changes in prey abundance and availability can vary seasonally due to small scale sea surface temperature (seasonal) which can influence temporal availability since spawning in many fish species involves migration to pelagic waters which is often initiated by temperature (Thompson 1981). The spawning of Pilchard has been found to relate to temperatures of 14°C below this they will not breed (Thompson 1981). Once spawning occurs and the eggs hatch juveniles return to inshore regions (e.g. Yellow Eyed Mullet, Thompson 1981). Seasonal variation in diet has also been recognised in other seabird species as a result of different factors (Schreiber and Burger 2002). Three different species of penguins feeding within a similar area during the breeding season were found to show a general diet with spatial and temporal variation (Clausen and Pütz 2002). Therefore spatial and temporal variation needs to be taken into consideration when interpreting diets.

The ability for LBP to catch prey could vary during the year depending on the small scale climate changes with the different seasons. The LBP are visual foragers and require day light hours to target prey species, therefore they have less time to forage during the shorter winter days (Bull 2000).

4.6.3 Stable Isotope Analysis

4.6.3.1 Annual differences

Despite potential biases, application of stable isotope analysis is also advantageous to dietary studies. In contrast to the conventional method stable isotopes analysis of diet is less intrusive on an individual, easy to apply, and can assess long-term dietary changes depending on available specimens. Furthermore, stable isotope analysis can be used to describe food webs and identify trophic levels within an ecosystem.

If seasonal changes occur within any year then this can influence the timing of food availability and abundance. Annual abundances of fish species such as slender sprat have been found to increase with a decrease in SST (Lalas *et al.* 2004). On a larger scale SOI values were not found to relate to the trophic level fluctuations and potential diet changes for LBP feeding within the Hauraki Gulf. The lack of correlation with SOI values, which represent *El Niño* and *La Niña* periods, does not preclude the possibility of small scale variables influencing foraging ability and prey availability in any year. Warmer waters associated with *La Niña* years should lower prey abundances, although the exact relationship between *El Niño* and fish abundance is not well known in New Zealand. Climate events such as *La Niña* (warming) can cause an increase in chlorophyll levels, therefore the next step in the analysis would be to see if the chlorophyll levels within the Hauraki Gulf has changed since 1886.

This study found that the nitrogen and carbon values in LBP have fluctuated over long time scales independently of SOI. This suggests shorter term variations in food supply influence the LBP foraging. There has been much concern over 'system shifts' within the Southern Hemisphere due to climate change (Reid and Croxall 2001; Croxall *et al.* 2002; Fraser and Hoffman 2003; Jenouvrier *et al.* 2003; Weimerskirch *et*

al. 2003; Hilton *et al.* 2006) but overall effects can be hard to obtain. For example, Rockhopper Penguins (*Eudyptes chrysocome*) have experienced marked declines in population numbers but causes could not be established due to the lack of long term data (Hilton *et al.* 2006). This means that any potential variability occurring on an annual basis is often missed. LBP within Australia have been found to shift their diet according to the season (Montague and Cullen 1988), however, it is also in terms of species survival to know of any long term changes in their trophic level.

4.6.3.2 Foraging range

The stable isotope values for $\delta^{13}\text{C}$ for LBP show that they are feeding within the inshore regions of the Hauraki Gulf but the levels have fluctuated over time. However, what constitutes inshore within the Hauraki Gulf (surrounded by mainland and other Islands) is problematic. The depth of the gulf is relatively consistent reaching around 30 metres. This implies that care has to be taken when considering LBP foraging range from isotope levels since anywhere in the Gulf could be considered inshore. To identify where the LBP are going within the Gulf and how long they are foraging for would need direct measures such as the use of tracking devices. It would also require direct assessment of the $\delta^{13}\text{C}$ values from different areas.

LBP like other seabirds (e.g. Weimerskirch 1998) are central place foragers having to return to their breeding site regularly to provision chicks and to share duties (incubation and chick rearing), which places restrictions on their foraging range. However, unlike other seabirds, LBP cannot fly which limits their ability to track the most abundant and energetically profitable prey. Travelling long distances to forage may mean that birds will be able to find optimal prey which could cover the energetic costs of searching. However, it also means that chicks are provisioned less frequently.

If LBP are feeding inshore then the reason for their high failure rate at fledging chicks as well as their inability to lay down enough stores for moult (Chapter 2) would imply that they are not finding enough prey. A recent study by Chiaradia *et al.* (unpubl. data) compared populations of LBP from two islands within Australia (Phillip Island and Penguin Island) and New Zealand (Motuara Island and Omaru) and results showed that higher fledging success occurred for individuals making shallower dives and less diving effort in water < 50. Conclusions of this study were that the bathymetry of the seafloor was one parameter influencing breeding success (Chiaradia *et al.* unpubl. data) therefore vertical and horizontal restrictions are associated with foraging areas. Prey availability is thought to be influenced by the sea depth. Clupeiformes are able to migrate to depths of 200m (Kailola *et al.* 1993; Chiaradia *et al.* unpubl. data), therefore shallower waters would mean less room to hide from potential predators by vertical migration (Takahashi *et al.* 2003; Ropert-Coudert *et al.* 2006; Chiaradia *et al.* unpubl. data). This suggests that the morphology of the feeding habitat (water depth, geographical features) as well as behavioural aspects of LBP and prey will influence foraging strategies by LBP (Chiaradia *et al.* unpubl. data).

Although prey behaviour could be influenced by sea floor bathymetry it could also be influenced by the high level of boat disturbance and recreational fishing within the waters surrounding Tiri. LBP have been found to feed in higher densities around Leigh Marine Reserve than Tiri which could have been associated with the different levels of disturbance (Chen 2004). Leigh is also a Marine Reserve which protects marine life acting as a spawning ground for fish. Therefore, the size and abundance levels of fish species may be larger within these areas, which may influence LBP foraging within these areas. Little has been done to investigate the effects that a Marine Reserve could have on top predators. A tracking study on King Penguins *Aptenodytes*

patagonicus from Maquarie island have found these penguins to forage within the boundaries of a proposed marine protected area during the incubation period (Weinecke and Robertson 2002).

4.6.3.3 Competition

This study did not directly quantify the effect that natural and anthropogenic competition has on the prey abundances and foraging in LBP. Competition for optimal prey types may exist between other natural prey species such as birds and mammals, and anthropogenic catches (recreational and commercial fisheries). A study on the diets of three penguin species within the Falkland Islands (Gentoo Penguin *Pygoscelis papua*, Magellanic Penguin *Spheniscus magellanicus*, and Rockhopper Penguin) found overlap with the commercial fishery for Patagonian Squid *Loligo gahi* and some competition may exist with the Patagonian Toothfish (*Dissostichus eleginoides*), Hake (*Merluccius sp*) and Southern Blue Whiting (*Micromesistius australis*) (Clausen and Pütz 2002).

Pilchard is considered an important component for Australian LBP diet while slender spat has been identified as the most important part of the LBP diet found in Oamaru (Fraser 1999; Perriman *et al.* 2000). Other seabird species such as Lesser Black-backed Gulls (*Larus fuscus*), Australasian Gannets (*Morus serrator*, Wingham 1985; Robertson 1992), and marine mammals such as the Common Dolphin (*Delphinus delphinus*) (Stockin, unpubl. data) have found Pilchard to be an important prey species within the Hauraki Gulf. A study on the Australasian gannet in New Zealand found that preferred prey items were Pilchard, Anchovy, Saury, and Jack Mackerel, and other less important prey species of Garfish, Yellow-eyed Mullet, squid, and Red Cod (Robertson 1992). This is similar to the diet of LBP within this study.

If there are a large number of foragers within an area this could cause localised prey depletion or reduced prey availability due to disturbance and competition causing individuals to work harder to obtain their requirements (Lewis *et al.* 2001). Competition with other larger more efficient foragers may limit the ability of LBP to find Pilchard and may explain the lack of Pilchard in the stomach samples. Slender Sprat (*Sprattus antipodum*), Pilchard, Anchovy, Yellow-eyed Mullet, and Barrocouta (*Thyrsites atun*) are also caught by commercial operations, with the largest takes occurring within the Northern East Coast waters of New Zealand (Appendix 4.7.8). Therefore LBP may have major competition with the fishing industry, especially since annual catch rates have increased overtime. Largest takes for Pilchard have occurred in 2000-01 (1,290t) and the second largest taken 2003-04 (1,284) from the Northeast Northland area alone (MFA Plenary Report 2005). For Anchovy most of the landings have occurred in 2000-01(10), 2002-03(8t), and 2003-04 (4.3t) from the Northeast coast. Both species are generally caught together so the lower levels of Anchovy may be due to over reporting of pilchards. The main prey of LBP within this study was anchovy as opposed to pilchard which could suggest that either, seasonal differences in prey availability are occurring or there is potential overlap with fisheries. However, more direct studies to measure the competition effects between commercial fisheries, food availability and its effect on penguin species in general is required (Cairns 1992).

4.6.4 Conservation Management Recommendations

Diet analysis is an important aspect of ecology needed to gain an understanding of the link that a species has with the marine environment and to assess how behaviours and ecological aspects of their life-history are determined by climate and prey changes. The marine environment is a dynamic system and is always changing therefore species that

spend a large amount of time associated with this system and rely on it for food resources will be affected in some way. Understanding these links is important to conservation management practices in order to conserve populations of a species.

Application of research procedures depend on the questions that require answering. For diet analysis the application of the conventional method may be best suited to short-term analysis of diet where the question requires consideration of species-specific answers applicable to research on energetics or seasonal changes. This does not seem a viable option for long-term studies that target questions based on population diet shifts, the effect of commercial fisheries, or the long term stability of the population due to food availability. Furthermore, it may not be applicable when targeting large sample sizes to gather a good representation of the population.

The implementation of conventional methods is time consuming since it requires application of the procedure and further analysis of the samples. Furthermore specialist training is involved to utilise the method safely and gain accurate samples and to obtain accurate identification of particular species. More importantly there is also the need to limit stress to the animals where possible and even though no LBP died as a result of the regurgitation procedure stress risks are not a viable option when dealing with a species that is limited in population numbers and critically threatened.

A better method for obtaining both short and long-term data would be via stable-isotope analysis. In contrast to the conventional method it is easy to apply and can utilise old specimens to create 'ecological histories'. Since different tissues can be sampled a range of dietary time periods can be targeted. Daily diets can be assessed based on blood and breath, feathers will provide monthly diet, and growth diet of chicks and adults can be analysed from toenails and bone. Feathers are an ideal tissue to sample as they can be obtained from occupied sites without actual handling of animals,

which makes sampling even less intrusive. Stable isotope analysis may be more enlightening when used as a complimentary procedure to the conventional method. Although, for a more direct comparison with stable isotope analysis, stomach samples should be taken at the same time that tissues are sampled for stable isotopes so that a measure of prey nutrients assimilated could be quantified. A better tissue comparison with stomach samples would be blood since it is relevant to a short period of time.

It would also be practical to sample the Hauraki Gulf area for bait fish and take stable isotope measures from tissue samples for each prey type to create a trophic food web based on the different signatures. However, this was outside the scope for this current study since the limitation to stable-isotope analysis is that they are expensive. Therefore, sampling of other LBP tissues and prey tissues were not a viable option within this study. Regardless, once prey signatures are obtained this is highly applicable at an ecosystem level and can therefore be used for other species.

To enhance dietary studies stable isotope analysis could be coupled with global positioning devices and time depth recorders which will provide information about the foraging ranges of a species. Information about movement can be directly linked to the terrestrial behaviour of species such as marine birds that depend on land for breeding. Acknowledging that marine birds rely on both media is important to management since there are very different limitations and stresses associated with each and are highly interchangeable.

LBP may not be the best top predator to use as a biological indicator of productivity within the marine environment since it appears that they will switch diets according to what is available within their limited foraging environment. A better species may be the Australasian Gannet (*Morus serrator*) since it is a group forager and can be more easily monitored feeding at sea and gathering of prey samples at any

particular point in the breeding season more easily obtainable. Therefore a more direct link could be obtained regardless of whether the bird diet switches. LBP may be better used as indicators of Marine Reserves and the effect that they can have on higher levels within the food chain. This would be appropriate since Marine Reserves often monitor abundances and size ranges of lower trophic levels and require long term monitoring to gather this information. The influence that Marine Reserves have on predator-prey interactions would be of interest as a community approach to conservation. Long-term monitoring of these links will provide a more dimensional approach which is often lacking within conservation management. This is usually the case since research and conservation efforts often target single-species and for a short period without the knowledge of the future impacts that this can have on other interacting species. In the meantime monitoring of LBP (and other bird species) is valuable from an indicator perspective because of the availability of museum specimens, and the relative abundance of species.

In a broader context understanding the diet of LBP and its effect on their breeding success and survival is limited by the lack of knowledge in the biology and actual abundances of many prey species. For example it is unknown as to whether there are separate fish stocks within areas or if populations are stable or variable, however the latter is suspected more likely (MFA Plenary Reports 2005). Plenary reports from the Ministry of Fisheries (MFA 2005) state that there is potential for localized depletion if there is limited migration between stocks. The lack of knowledge in the biological aspects of prey species can cause difficulties in trying to understand the influence that they have on the LBP (predator-prey interactions). Indeed the breeding success of LBP

populations has been associated with changes in the abundance of major prey items in Australia (Jones *et al.* 1997) and requires further investigation in New Zealand.

4.6.5 Conclusions

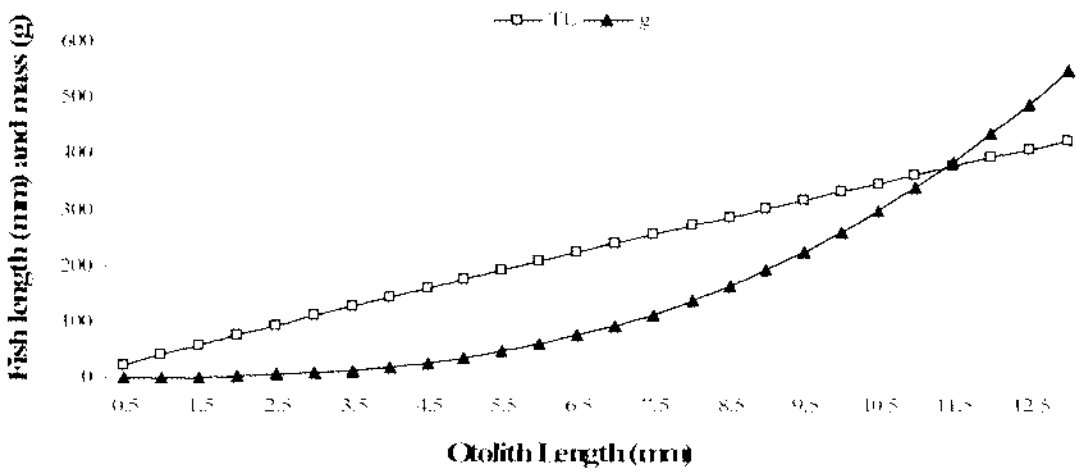
Many transformations within marine ecosystems has been associated with pollution, removal of top predators, over-harvesting, and climate change (Jackson *et al.* 2001) which has resulted in removal of prey species, changes in predator-prey interactions (Jackson 2001), fishing through the food webs (Essington *et al.* 2006), and alterations in life-history behaviours. The impacts that these can have on other prey species is often not well documented but can be achieved by monitoring species that rely on the marine environment for their foraging requirements.

Fluctuations in prey availability has been associated with predator productivity and requires long-term monitoring (Croxall and Prince 1979; Cairns 1987; Croxall *et al.* 1988; Montervecchi *et al.* 1988; Monaghan *et al.* 1989; Montevecchi and Berruti 1995; Furness and Greenwood 1993; Hammer *et al.* 1993; Ainley *et al.* 1995; Monaghan 1996; Croxall *et al.* 1999). It is unknown what roles of prey availability, opportunism, nutrition, and the constraint of attachment to breeding sites have on LBP, and the potential for them to switch between diets. Understanding the different responses of predators to climate change is important to understand the consequences of this on trophic dynamics (Murawski 1993; Sims *et al.* 2004). Since the natural and anthropogenic influences on marine resource availability can influence ecological aspects of top marine predators. The potential competition that could exist between the commercial fisheries and LBP has been identified (Norman *et al.* 1992) and deserves further investigation.

Knowledge of predator-prey relationships is crucial when considering seabirds as indicators of the marine environment. Diet studies will increase the understanding of these links and over the long term help elucidate the possible competition that exists between natural and anthropogenic competitors (Croxall *et al.* 1999). Potential implications can arise for conservation efforts since little is known of the consequences that a widespread decline of fish eating seabird populations would have. Or the effects that this could have on top-down, bottom-up patterns of trophic cascades within marine systems.

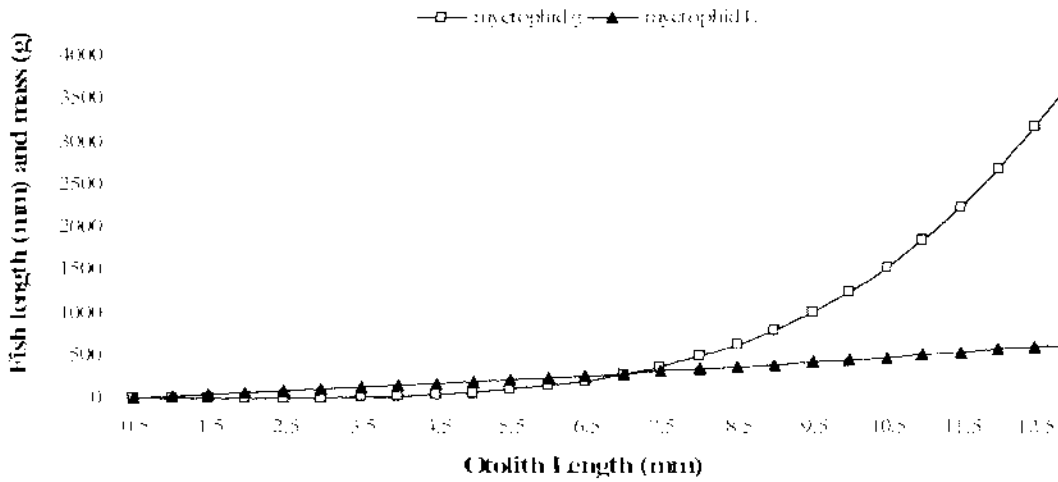
4.7 Appendix

4.7.1 Total length and weight regression of a South African Anchovy



Total length (TL) and weight (g) regression of the South African species of Anchovy (*Engraulis japonicus*) from otolith length (OL) based on the equation: $M = -1.0158 + 2.8541 OL$; $TL = 3.7039 + 0.9137 OL$ (Smale *et al.* 1995).

4.7.2 Total length and weight regression of a South African Myctophid



Total length (TL) and weight (g) regression of the South African species of Myctophid (*Lampanyctodes hectoris*) from otolith length: (OL) based on the equation: $M = 2,5907 + 4,2197 OL$; $TL = 3,1988 + 1,2676 OL$. (Smale *et al.* 1995).

4.7.3 Comparison of $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ values

Comparison of $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ values analysed on the NIWA *Thermo-Finnigan Delta^{plus}* mass spectrometer compared to reported NIST values. The +/- values represent 1 standard deviation.

| NIST standard | NIST $\delta^{15}\text{N}$ ‰ reported values (n=) | NIWA measured $\delta^{15}\text{N}$ ‰ values (n=) | NIST $\delta^{13}\text{C}$ ‰ reported values | NIWA $\delta^{13}\text{C}$ ‰ values measured (n=) |
|------------------------------|---------------------------------------------------------|---------------------------------------------------------|----------------------------------------------------|---------------------------------------------------------|
| 1577b Bovine Liver* | +7.78 +/- 0.22 (61) | +7.88 +/- 0.26 (8) | - | - |
| 1547 Peach Leaves* | +2.08 +/- 0.18 (32) | +2.26 +/- 0.31 (8) | - | - |
| 2685a Coal Sub-bituminous* | +2.79 +/- 0.74 (12) | +2.33 +/- 0.27 (3) | - | - |
| 2704 Buffalo River Sediment* | +3.80 +/- 0.39 (59) | +3.88 +/- 0.47 (5) | - | - |
| 2682a Coal Bituminous* | +3.38 +/- 0.75 (15) | +2.27 +/- 0.10 (3) | - | - |
| 8547 N1 Ammonium sulphate | +0.40 +/- 0.20 | +0.56 +/- 0.23 (17) | - | - |
| 8548 N2 Ammonium sulphate | +20.3 +/- 0.20 | +20.4 +/- 0.21 (17) | - | - |
| 8549 N3 Potassium nitrate | +2 to +5 | +4.61 +/- 0.51 (10) | - | - |
| 8541 Graphite | - | - | -15.90 +/- 0.25 | -15.48 +/- 0.11 (10) |
| 8542 Sucrose | - | - | -10.47 +/- 0.13 | -10.78 +/- 0.38 (10) |
| 1577b Bovine Liver* | 10.2 +/- 0.29 (61) | 9.76 +/- 0.47 | - | - |
| 1547 Peach Leaves* | 2.83 +/- 0.11 (32) | 2.63 +/- 0.09 | - | - |
| 2685a Coal Sub-bituminous* | 0.96 +/- 0.04 (12) | 0.88 +/- 0.04 | - | - |
| 2704 Buffalo River Sediment* | 0.20 +/- 0.01 (59) | 0.18 +/- 0.00 | - | - |
| 2682a Coal Bituminous* | 1.11 +/- 0.06 (15) | 0.83 +/- 0.11 | - | - |
| 8547 N1 Ammonium sulphate | 21.21 | 20.83 +/- 0.66 | - | - |
| 8548 N2 Ammonium sulphate | 21.21 | 21.01 +/- 0.18 | - | - |
| 8549 N3 Potassium nitrate | 13.86 | 13.33 +/- 0.28 | - | - |
| 8541 Graphite | - | - | - | - |
| 8542 Sucrose | - | - | 42.11 | 43.84 +/- 0.61 (9) |

* reported values were co-ordinated by Environmental Isotope Laboratory, University of Waterloo

4.7.4 Precision data for repeat analysis of urea standards

Precision data for repeat analysis of urea standards during sample batch analyses. The +/- values represent 1 standard deviation.

| Internal Urea Standard | Wt% N (n=8) | $\delta^{15}\text{N}$ (n=10) | Wt % C (n=3) | $\delta^{13}\text{C}$ (n=5) |
|------------------------|---------------|------------------------------|----------------|-----------------------------|
| Known value | 46.64 | | 20 | |
| Measured value | 46.30 +/- 0.8 | -0.82 +/- 0.09 | 19.64 +/- 0.14 | -46.73 +/- 0.14 |

4.7.5 Trophic levels for all years

Calculated trophic levels for all years

| | Year | | | | | | | | |
|-----------------------|--------|--------|--------|--------|--------|--------|--------|--------|--------|
| | 1887 | 1941 | 1947 | 1949 | 1961 | 1994 | 2002 | 2004 | 2005 |
| $\delta^{13}\text{C}$ | -15.11 | -15.89 | -14.05 | -14.73 | -15.86 | -13.97 | -15.29 | -15.60 | -16.89 |
| $\delta^{15}\text{N}$ | 14.65 | 14.92 | 16.50 | 14.20 | 14.81 | 14.39 | 17.53 | 15.61 | 15.10 |
| TL | 3.36 | 3.43 | 3.82 | 3.25 | 3.40 | 3.30 | 4.08 | 3.60 | 3.47 |

4.7.6 Yearly mean differences for $\delta^{13}\text{C}$ Carbon values

Differences between each yearly mean for $\delta^{13}\text{C}$ Carbon values.

| | Year | | | | | | | | |
|------|-------|------|-------|-------|-------|-------|-------|-------|------|
| | 1887 | 1941 | 1947 | 1949 | 1961 | 1994 | 2002 | 2004 | 2005 |
| 1941 | -0.78 | | | | | | | | |
| 1947 | 1.06 | 1.84 | | | | | | | |
| 1949 | 0.38 | 1.16 | -0.68 | | | | | | |
| 1961 | -0.75 | 0.03 | -1.81 | -1.13 | | | | | |
| 1994 | 1.14 | 1.92 | 0.08 | 0.76 | 1.89 | | | | |
| 2002 | -0.18 | 0.6 | -1.24 | -0.56 | 0.57 | -1.32 | | | |
| 2004 | -0.5 | 0.29 | -1.55 | -0.87 | 0.26 | -1.63 | -0.31 | | |
| 2005 | -1.78 | -1 | -2.84 | -2.16 | -1.03 | -2.92 | -1.59 | -1.28 | |

4.7.7 Yearly mean differences for $\delta^{15}\text{N}$ Nitrogen values

Differences between each yearly mean for $\delta^{15}\text{N}$ Nitrogen values.

| | Year | | | | | | | | |
|------|-------|-------|-------|------|-------|------|-------|-------|------|
| | 1887 | 1941 | 1947 | 1949 | 1961 | 1994 | 2002 | 2004 | 2005 |
| 1941 | 0.27 | | | | | | | | |
| 1947 | 1.85 | 1.58 | | | | | | | |
| 1949 | -0.45 | -0.72 | -2.3 | | | | | | |
| 1961 | 0.16 | -0.11 | -1.69 | 0.61 | | | | | |
| 1994 | -0.26 | -0.53 | -2.11 | 0.19 | -0.42 | | | | |
| 2002 | 2.88 | 2.61 | 1.03 | 3.33 | 2.72 | 3.14 | | | |
| 2004 | 0.96 | 0.68 | -0.89 | 1.41 | 0.79 | 1.21 | -1.92 | | |
| 2005 | 0.45 | 0.17 | -1.4 | 0.9 | 0.28 | 0.7 | -2.43 | -0.51 | |

4.7.8 Total catches for commercial fisheries from North East Coast areas of New Zealand

Total catches (tonnes) for commercial fisheries taken from the North East Coast areas of New Zealand and known relevant biology of each species. (Ministry of Fisheries Plenary Reports 2005).

| | Species | | | | | |
|----------------|---------|-----------|------------|-----------|----------|----------|
| | Sprat | Red cod | Barracouta | Y. mullet | Anchovy | Pilchard |
| 1983-84 | | 12 | 7 805 | 2 | | |
| 1984-85 | | 9 | 5 442 | 12 | | |
| 1985-86 | | 6 | 5 395 | 24 | | |
| 1986-87 | | 5 | 8 887 | 14 | | |
| 1987-88 | | 8 | 9 256 | 11 | | |
| 1988-89 | | 9 | 5 838 | 3 | | |
| 1989-90 | | 8 | 9 209 | 1 | | |
| 1990-91 | 3 | 12 | 9 401 | 21 | <1 | 15 |
| 1991-92 | 1 | 26 | 6 733 | 15 | 1 | 59 |
| 1992-93 | <1 | 46 | 9 032 | 32 | 21 | 163 |
| 1993-94 | <1 | 44 | 7 299 | 53 | <1 | 258 |
| 1994-95 | <1 | 63 | 10 023 | 32 | <1 | 317 |
| 1995-96 | <1 | 28 | 11 252 | 19 | 1 | 168 |
| 1996-97 | <1 | 42 | 11 873 | 32 | 2 | 419 |
| 1997-98 | <1 | 22 | 11 543 | 10 | 1 | 440 |
| 1998-99 | 2 | 10 | 9 229 | 16 | 4 | 785 |
| 1999-20 | <1 | 3 | 10 032 | 10 | 3 | 1 227 |
| 2000-01 | <1 | 5 | 7 118 | 9 | 10 | 1 290 |
| 2001-02 | <1 | 6 | 6 900 | 6 | 7 | 574 |
| 2002-03 | <1 | 8 | 7 595 | 9 | 8.3 | 792 |
| 2003-04 | <1 | 11 | 5 948 | 4 | 4.3 | 1 284 |
| Biology | | | | | | |
| Size | | | | | | |
| Juvenile | | 25cm | | | | |
| Adult | | 52cm | 50-60 | | 16 cm | 10 to 20 |
| Spawn | J - N | A - O | A -S | D -M | Sp - S | Sp - S |
| Eggs | P | P | DW | P | P | P |
| Range | | DW | | 1m-32km | | In - P |
| Depth | | >300-750m | <100-400 | | | |
| Growth | | Fast | | | | Fast |
| Life | | <6 years | | 7 years | 6 years* | 9 years |

* Note: Spawning months are July (J), August, (A), October (O), November (N), and December (D), with spawning seasons spring (Sp) and summer (S). Eggs are either pelagic (P) or deep water (DW). Range that they travel is deep water (DW) inshore (IN), pelagic (P) or in meters.

CHAPTER 5 Survival of Little Blue Penguins



Plate 5.1. Little Blue Penguins found dead on Tiritiri Matangi Island. Photo by J.Geurts 2006

5.1 ABSTRACT

The cause of Little Blue Penguin (LBP: *Eudyptula minor*) deaths during periods of mass mortalities within the Hauraki Gulf during 2005 and 2006 was explored through the necropsy of birds found dead on beaches. A total of 96 birds were found dead on Tiritiri Matangi Island (Tiri) during September 2005 and April 2006 and a further 13 from Tawharanui and other Hauraki Gulf regions. Necropsies were performed on 53 of the birds with results suggesting death by starvation. A total of 11 birds were found to have low levels (< 30) of intestinal cestodes (*Tetrabothriidae* sp) and histology tests also found evidence of liver flukes (unknown species) and kidney trematodes (unknown sp), thought to be uncommon in LBP.

Analysis of 2005-06 deaths showed no significant correlation with weather data (wind speed and rainfall). Analysis of long term data of mass mortalities obtained through the Ornithological Society of New Zealand (OSNZ) found no significant correlation with large scale climatic events such as Southern Oscillations Index (SOI). Annual occurrences of mortalities over a 33 year period showed no general pattern however, when counts of LBP numbers found dead on all New Zealand beaches were totalled over all years by month, particular months consistently had peaks. Starvation was thought to be a factor associated with post-breeding, moulting, and winter. These periods are energetically expensive and food intake due to prey availability or individual ability to obtain food. Analysing the cause of death through necropsies and histology tests is important for conservation management especially since the largest counts of dead LBP are occurring within the North Island region of New Zealand.

5.2 INTRODUCTION

5.2.1 Mortality in Seabirds

Seabird populations were thought to be relatively stable through time (Furness and Monaghan 1987) with large mortalities of adults being rare (Cairns 1987). However a recent mass mortality of a range of species within New Zealand has been documented (Taylor 1996, 1997, 1999) without conclusive evidence of death. Consideration of factors that may be causing mass death in seabirds, and LBP in particular, will help assess the susceptibility of populations. Understanding the long-term effects of such events will be directly applicable for conservation efforts.

Past records by the Ornithological Society New Zealand (OSNZ) began in 1959 mass mortalities of LBP have been occurring within both Australia and New Zealand (Norman *et al.* 1992). This is considered to be a 3 to 4 year occurrence within New Zealand (Taylor *pers. comm.*) but is not well understood. Hypothesis associated with these mass beach wrecks include storm events causing injury and fatigue (Norman *et al.* 1992; Dann 1992; Taylor 1999), response to changes in food supply (Norman *et al.* 1992; Dann *et al.* 2000), and disease or toxicity (Norman *et al.* 1992; Hocken 2002). Classifications for deaths of LBP have been grouped into major causes including: starvation, predation (e.g mustelids, dog, sharks or killer whale), drowning, disease, rail/road injury, and natural death (e.g aspergillosis), and unknown (Hocken 2002).

Factors associated with death may not be the same for each region for example Obendorf and McColl (1980) found that LBP deaths in Victoria, Australia was due to poor body condition and moderate to high parasite load (Harrigan 1992). In 1995 mass mortalities of LBP were correlated to a mass die-off of a major prey species Pilchards

(*Sardinops sagax*) within Australia (Dann *et al.* 2000). The reason for the fish die-off was due to a herpes-like virus (Jones *et al.* 1997). Moreover Dann (1992) suggests predation and rough weather may have a significant impact on mortalities. According to Mickleson *et al.* (1992) oceanographic events have had significant effects on the breeding success of LBP and mortalities have also correlated with weather events such as El Niño years. The SOI drives global scale climate variation which influences trade wind strength, direction, and trade winds (Trenberth and Shea 1987; Quinn *et al.* 1992; Bunce *et al.* 2002). Since penguins spend so much time at sea it seems reasonable to assume that the weather conditions will have some effect, whether indirectly affecting food availability or directly through injury or inability to forage.

Introduced predators are considered a major negative factor for much of New Zealand's fauna and LBP are no exception. Predation of adults, chicks and eggs in other penguin species has also contributed to declining population numbers. For example, a comparison between the numbers of Yellow-eyed penguins on Stewart Island (predator island) and the adjacent Codfish Island (predator free) found that significantly higher penguin numbers were recorded for the former (Ratz 2000). Ratz 2000 suggests that feral cats and ferrets (*Mustela furo*) are responsible for the declining numbers.

5.2.2 Determining Cause of Death

Mortality of sea birds can be divided into two types: 1) mortality at sea and 2) mortality on land (Dann 1992). Juveniles tend to be more susceptible to mortality at sea as they tend to spend most of their time at sea and only head ashore the following year to moult and later to breed (Norman *et al.* 1992; Dann 1992). In addition juveniles may starve due to inexperience at foraging (Norman *et al.* 1992) and have higher endoparasitic infections (Harrigan 1992; Dann 1992). Parasitic infection could also restrict digestion

and cause starvation or cause paralysis in birds causing drowning. The major cause of adult mortality is starvation (Harrigan 1992), but whether this is due to food shortages or weather changes is unknown.

Ascertaining and correlating causes for mass penguin die-offs can be difficult due to the unpredictable nature of these events. It is difficult to identify the primary cause of death as opposed to secondary effects since many factors are often interrelated. More often than not there may be more than one possible cause of death. For example, it is often hard to ascertain whether a bird died 'of' starvation, or 'with' starvation (Hocken 2002). Parasites can lower body condition due to stress or energetic constraints and this may inhibit foraging hence resulting in death with starvation (Harrigan 1992). Moreover, starvation may also be due to unavailability of prey species due to seasonal changes in abundance, competition, individual ability to obtain food, and factors that may cause foraging restrictions (weather, foraging range, time) (Chapter 4).

The time of year that mortalities occur can help with diagnoses. For example breeding is the one of the most demanding behaviours associated with the life-cycles of birds (Gales and Green 1990). High food resources allow birds to fast during incubation of eggs, to provision growing chicks, and maintain their own body condition. Moulting is also demanding since birds are required to fast ashore for 15 days (Marchant and Higgins 1990). Plumage becomes mottled brown and white (Williams 1995) losing its condition during the year, LBP and other penguin species (exceptions being Galapagos Penguin *Spheniscus mendiculus*, King Penguin *Aptenodytes patagonicus*, and African *S. demersus*) need to renew their whole body plumage annually (Hull *et al.* 2001). During this time LBP are not able to go to sea to forage since they do not have the body condition to deal with the marine environment (Hull *et al.* 2001). Moulting occurs immediately after the breeding season (Reilly and Cullen 1983; Johannesen *et al.* 2002)

and relies on fat stores and protein reserves (Groscolas and Cherel 1992; Cherel *et al.* 1994; Hull *et al.* 2000). It is between breeding and moulting that the birds head to sea to build up these reserves. If this is not achieved for some reason they may attempt to leave for sea early or may die of starvation (Cherel *et al.* 1994; Hull *et al.* 2001). Penguins in general are threatened with mortality during the moult fast period. For example mass mortalities of Rockhopper Penguins (*Eudyptes chrysocome*) within the Falkland Islands occurred as a result of starvation during the 1987 moult period due to a lack of energy stores (Keymer *et al.* 2001).

A second highly demanding period is winter. Gales and Green (1990) found that it was during the winter months (July in Southern hemisphere) that there was a negative energy balance for LBP (Johannesen *et al.* 2002). Lower temperatures and productivity at sea during this time may cause an increase in mortality (Johannesen *et al.* 2002). Stormy periods during winter have also been a factor associated with many seabird mortalities (Hudson 1985; Sanvik *et al.* 2005) since this may prevent birds from heading to sea during rough periods.

5.2.3 Significance of This Study

Although environmental factors are pertinent, examination of less obvious factors also need consideration. Ascribing any particular cause of death to a corpse can be difficult without further information from necropsies, histological tests, and toxicology tests. One of the best methods for determining and ascertaining cause of death is through post-mortem analysis (Hocken 2002). This has been achieved in LBP populations within Australia (Obendorf & McColl 1980; Harrigan 1992) and New Zealand (Hocken 2000a, 2000b, 2003). However, post-mortems have not been carried out on the North

Island subspecies of LBP (*E.m.irelidae*) since 1975 (Crocket & Kearns 1977, Jones unpubl data 1978).

The cause of LBP mortalities throughout New Zealand remains elusive and hypothetical since there have been no prior research to establish this. To understand the frequency and timing of these occurrences and more importantly why they are occurring requires investigation of long-term data. One source of long-term data is beach count records, which have shown that the largest counts of seabird deaths come from the North Island regions of New Zealand (Taylor 1997).

Six species of the penguin Genus *Eudyptes* have been classified as threatened, including LBP (*Eudyptula minor*). Northern and Southern populations of LBP in New Zealand are in gradual decline (Hitchmough 2002). Therefore the need to identify the cause of death for these marine birds is important since these impacts can potentially affect future population numbers.

5.3 Aims

1. To identify likely cause of death for samples of dead penguin found during mass mortality periods.
2. Provide data on penguin numbers during the study period of 2005 and 2006 to provide baseline data for future monitoring.
3. Quantify patterns of long-term beach count records of LBP mass-mortality occurrences.
4. Discuss potential causes of mortality patterns of both short term and long term LBP.

5.4 Methods

5.4.1 Dead Bird Collection

Accessible coastline of Tiritiri Matangi Island (Chapter 1) was scanned for dead penguins from the start of October 2005 through to April 2006. Before this period dead penguins were collected by Tiritiri Matangi Island (Tiri) volunteers and the Department of Conservation workers on the Island. Public reports from other Hauraki Gulf areas also resulted in dead birds for the study. Dead birds were also retrieved by dolphin research vessels operating within the Hauraki Gulf and were often received as frozen carcasses.

5.4.2 Beach Counts from Previous Years

Numbers of dead penguins found during beach patrol counts by the OSNZ from 1966 to 1999 were used as the basis for long-term analysis. For further information on this scheme and the beach patrol methods see Powlesland and Imber (1988).

The beach patrol cards keep track of the different seabirds found along fixed stretches of beach. The New Zealand coastline is divided into 18 patrolled regions (Figure 5.1). Only the Northland East (NE), Auckland East (AE), and Bay of Plenty (BP) regions were used in. A classification of “freshness” was used for all bird samples (Table 5.1).

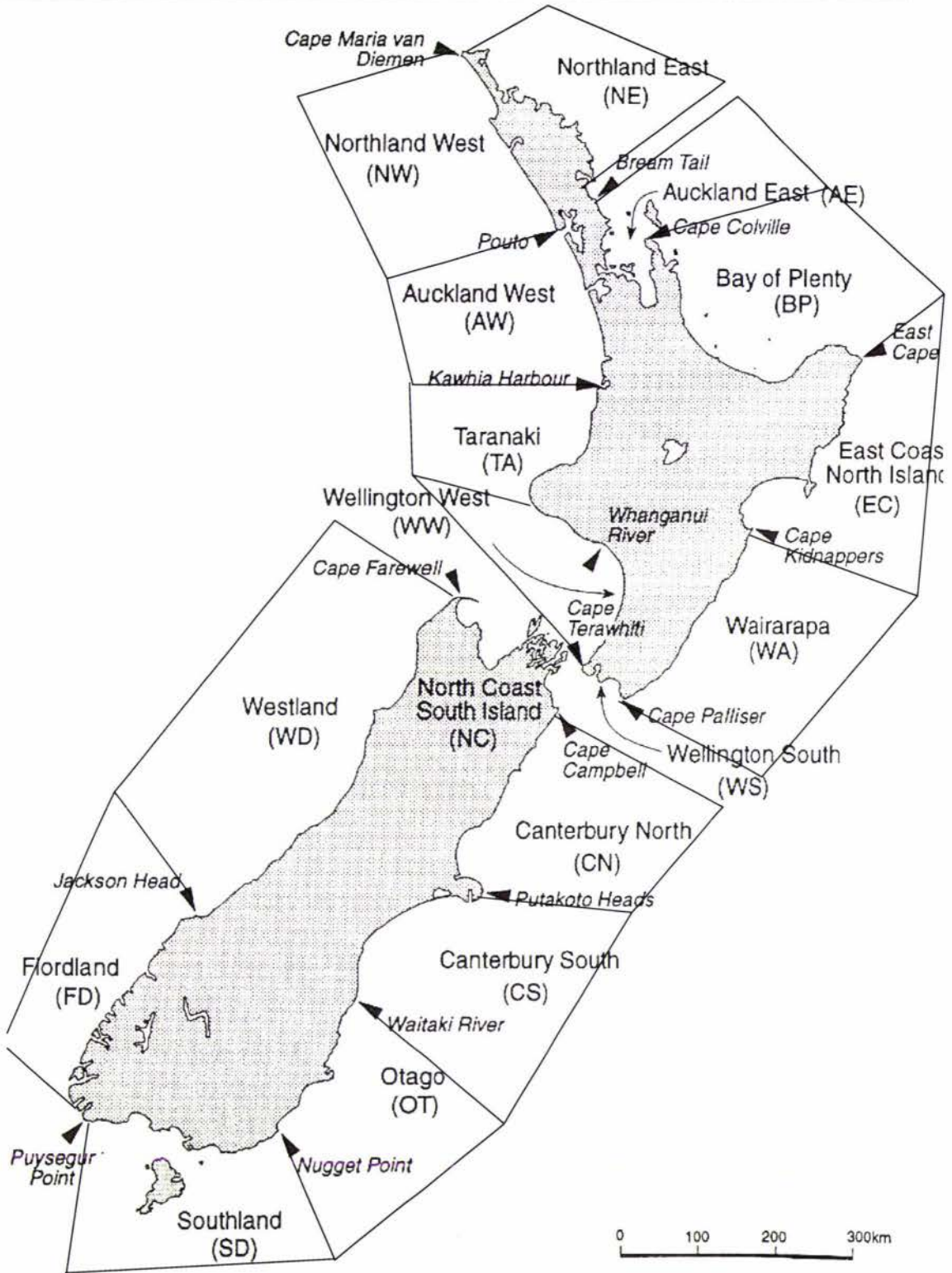


Figure 5.1 Different sections of New Zealand used by the Ornithological Society of New Zealand (OSNZ) for beach patrols. Abbreviations: North East (NE), North West (NW), Auckland East (AE), Auckland West (AW), Bay of Plenty (BP), East Coast (EC), Taranaki (TA), Wellington West (WW), Wairarapa (WA), Wellington South (WS), North Coast South Island (NC), Canterbury North (CN), Westland (WD), Canterbury South (CS), Otago (OT), Fiordland (FD), Southland (SE). The arrows represent the boundaries. (Altered from Taylor 1997 .p 202).

Table 5.1 Freshness levels and definitions used for classifying dead Little Blue Penguin corpses found along New Zealand beaches as developed by the Ornithological Society of New Zealand (OSNZ).

| Category Level | Definition |
|----------------|---------------------------------------------------------------|
| A | Very fresh corpse, less than a few days old |
| B | Decaying bird, rotting, dead a few days to 1-3 weeks |
| C | Dried corpse, been washed up on the beach for weeks or months |
| D | Skeletal remains only, been there for months or years |
| E | No attempt to record freshness |

5.4.3 Definitions

5.4.3.1 Cause of Death

The cause of death was assessed based on necropsies and tissue analysis. No cause of death was assigned until all results were returned. Classifications of death include; starvation, drowning, Aspergillosis, predation, and endoparasitism. To ascertain primary cause of death lab tests such as histology and toxicology were needed. All histology tests were done through Massey University Wildlife Centre, Palmerston North. Toxicology was beyond the scope of this study. Cause of death was assigned one of the following classifications;

1. *Starvation*: Classification based on an empty gut, muscle wasting and no fat stores. If the gut is empty and starvation is the cause, bleeding is usually associated. Blood appears as a dark brown or black (but not dark green) gut content. If blood is absent in the gut and intestines, pathology tests are required to confirm starvation. Without this the cause of death is unknown (Hocken, 2002).
2. *Drowning*: Determined by presence the presence of free fluid in the respiratory tract (Hocken 2005). Factors that are observed on recovery of the corpse can help identify if drowning was the cause of death. If the bird was found at the high tide mark, with

sandy plumage, and pale pink mouth then this suggests death at sea. Necropsy would show that the lung was watery. Red colouration and consolidation suggests pneumonia. The presence of cheesy puss-like white spots in the lung could indicate a chronic infection (Hocken, 2002).

3. *Predation*: Cause of death by predation can be seen through bites, and attack marks (Hocken 2005), as well as internal and external bleeding and hemorrhaging.
 - i) *Mustelid* – Attacks at the back of the neck damaging the spinal cord. Evidence of attack is associated with upper neck muscle and lower truck being eaten.
 - ii) *Dog* – Attacks occur at the back of the bird crushing the rib cage. Associated with bleeding from the mouth and puncture tears on the inside of the skin and pectoral muscle.
 - iii) *Shark* – Associated with flesh injuries that appear similar to a knife cut. (Hocken 2002).
4. *Asperogilosis*: This fungal disease occurs within the lung, and is identifiable through granulomatous tumors, white plaques, and nodules (Hocken 2002).
5. *Endoparasitism*: These can be seen in the stomach, liver, and kidneys when sliced open and investigated for cavities and irregularities. Coccidia can found by looking for off-white spheres on the kidney (Hocken, 2002). Liver flukes and tape worms are also identifiable by sight or through histology tests, and were sent to AgResearch laboratories for identification to the species or genus level.
6. *Unknown*: Birds not starving or with no evidence of disease or physical injury (Hocken 2002, 2005).

5.4.4 Necropsies

External examinations of the birds were conducted within the field repeated again during the actual necropsy to account for potential post-mortem effects. For necropsy procedures in wild birds see van Riper III and van Riper (1980), or for penguins see Hocken (2002).

5.4.4.1 *Preservation*

Carcasses were necropsied fresh, without prior freezing where possible. Carcasses found in good condition were generally stored in a refrigerator (4°C) and necropsied within 48 hours. If a necropsy was not possible within this time, or if carcasses were slightly in decay they were frozen (-20°C) until the necropsy.

5.4.4.2 *External Examination*

Carcasses were given an initial external examination when they were first found and again at the start of each necropsy. Each necropsy started with giving the carcass an individual ID (date that the necropsy was performed and case number).

The following data were recorded at the time of discovery; island location, beach location (high tide, top of the beach, in the bush, on the nest), band number (if present), date of death, or estimate if not fresh. The freshness of the body was categorised using the OSNZ Beach Patrol method (Table 5.1). The coat condition was recorded as wet or dry, sandy or clean, and the presence of external parasites, bleeding, and broken limbs. The eyes were checked for any obvious abnormalities. If birds were in a fresh state signs of sunken eyes were recorded as a sign of dehydration. The mouth was also checked for maggots, and any discolouration. Examinations included; weight, external parasite check, removal of any ticks from the ears, mouth, feet, or plumage, and examination of the overall physical body condition. This included: plumage condition

(pre/ post-moult), and fat stores (how prominent the sternum is). Several feathers were taken from each bird, and stored in a plastic container. Parasite samples were taken and put into tubes with ethanol 70%.

5.4.4.3 Internal Examination

Each bird was placed on its dorsal surface and prepared by wetting the ventral plumage. Feathers were cleared along the midline extending from the beak down to the rectum. The skin was lifted up with a pair of forceps and a horizontal incision was made with a pair of stainless steel scissors. The skin was separated from the pectoral muscle and the underlying body cavity by using a blunt dissection method. This involved inserting the scissors sideways into the chest cavity and opening the blades to separate the skin from the underlying muscle layers. The separated skin was then cut along the vertically exposed midline with the blunt end of the scissors running under the skin. The skin was peeled away from the underlying fat or muscle to expose the full length of the body and organs.

All major tissues and organs were assessed individually as described below.

5.4.4.3.1 Musculoskeletal System

The muscle mass of the chest and hence the pectoral muscles were assessed based on the shape. A flat muscle layer is often evidence for minor wasting, where concave muscle indicates malnutrition. An estimate of muscle thickness was made on a scale of 1 to 5 was given; 1 being very thin with some ribs uncovered, and 5 being a flat thick muscle layer. This quantity (level 1 to 5) and location of any fat stores (e.g. abdominal, subcutaneous, associated organs) were recorded.

5.4.4.3.2 *Body Cavity*

The rib cage was removed and major organs were assessed before removal. This included; the heart, liver, pancreas, spleen, gonads (testes/ovaries), kidneys trachea, esophagus, stomach and intestines (Figure 5.2). Each of these were inspected for abnormalities such as discolouration, lesions, haemorrhage, spotting, raised areas, cheese- like pus, and obvious parasites. If present, any abnormal cavity fluid was also noted, as well as the level of autolysis. Care was taken when analysing each organ structure, since deviation from normality could be due to post-mortem effects, and freezing. Each organ was sampled to obtain a 10mm² sample fixed in 10% formalin for histological analysis.

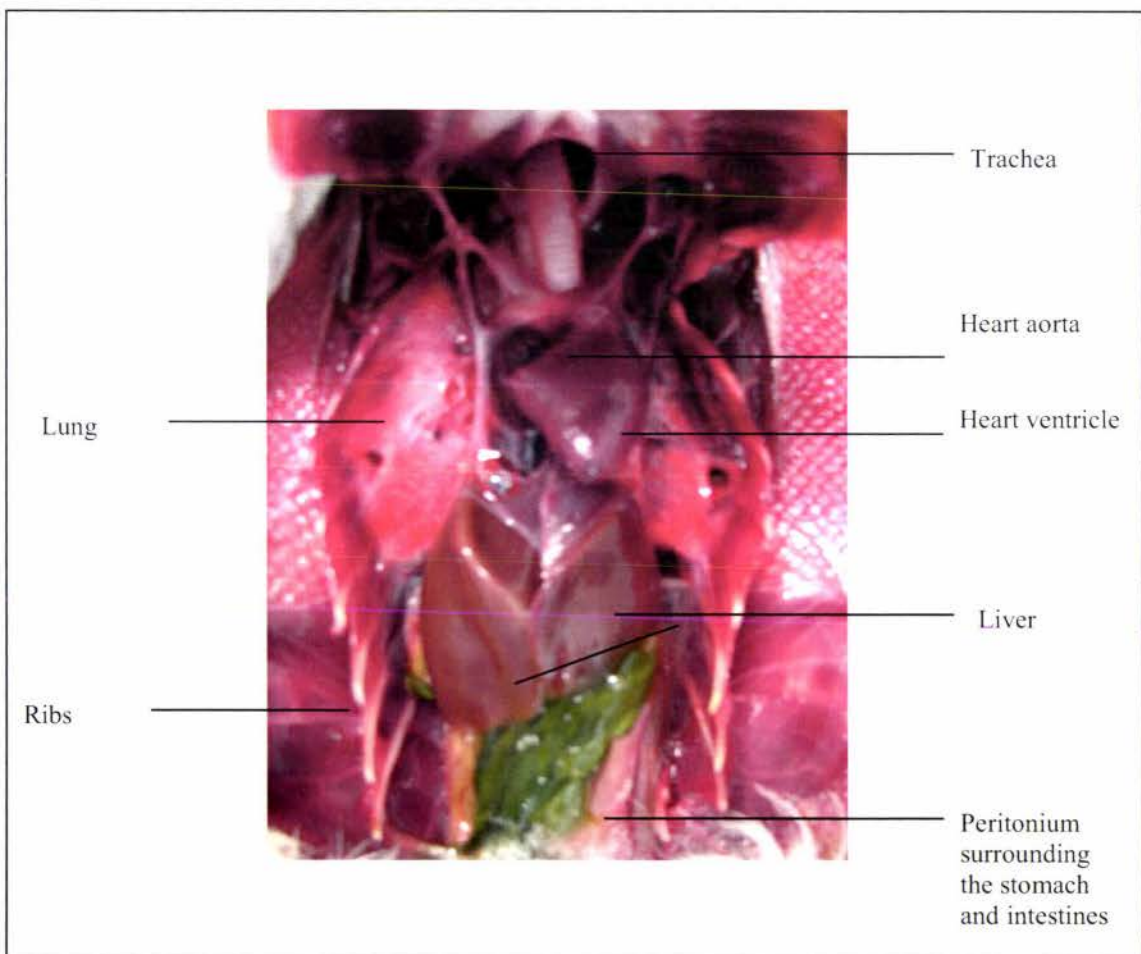


Figure 5.2 Diagram of the internal organs of a Little Blue Penguin considered to be in good condition. Photo by J.Geurts (2006).

5.4.4.3.3 Respiratory System

The lungs were sectioned to reveal the internal regions of the lung. Normal lungs are light pink and spongy in texture. Variations from this included discolouration of dark red or white, consolidation, watery, or a pus texture. The lungs were also checked for the presence/ absence of congestion. The trachea was removed by cutting at the branching of the main bronchi and the anterior region including the tongue. The trachea was then cut open laterally, and a section along the length of it was taken out to prevent collapse.

5.4.4.3.4 Cardiovascular System

The whole heart was removed from the cavity. The apex of the heart was taken as a fixed sample for histology. Each ventricle was cut open to observe the internal region for any abnormalities. The remaining heart tissue was frozen at -20°C.

5.4.4.3.5 Digestive System

The liver was removed and checked for discoloration, parasitic worms, and any lesions. The edges of the lobes were checked for sharpness, and each one was cut through for internal checks.

The first trialled method for sampling the length of the intestines was by squeezing along the length a 10cm section at a time and then cutting at emptied portions for ease of emptying. This was abandoned as it could potentially cut parasites and complete intestines were stored for another study.

The gut, including the oesophagus, was removed and cut open. Food or other items found in either the gut or oesophagus was noted. Any signs of bloody dysentery, and the source such as ulcers, were investigated by washing the gut wall under tap water. Any stomach contents were washed into a container at this stage, and kept for

sorting. Contents were emptied from the length of the intestines. Any cestodes were taken and washed before storing in 70% ethanol.

5.4.4.3.6 *Urinary System*

Both kidneys were removed and dissected to check for obvious signs of tumours or potential cestodes. One kidney was fixed in 70% ethanol and stored at room temperature.

5.4.4.3.7 *Reproductive System*

The gonads (testis or ovaries) were removed (Figure 5.3). One of these was fixed in ethanol. In the first five necropsies the width and length of each testis were measured with digital callipers but this was later abandoned. The stage of development was noted, especially if oocytes or testis were enlarged.

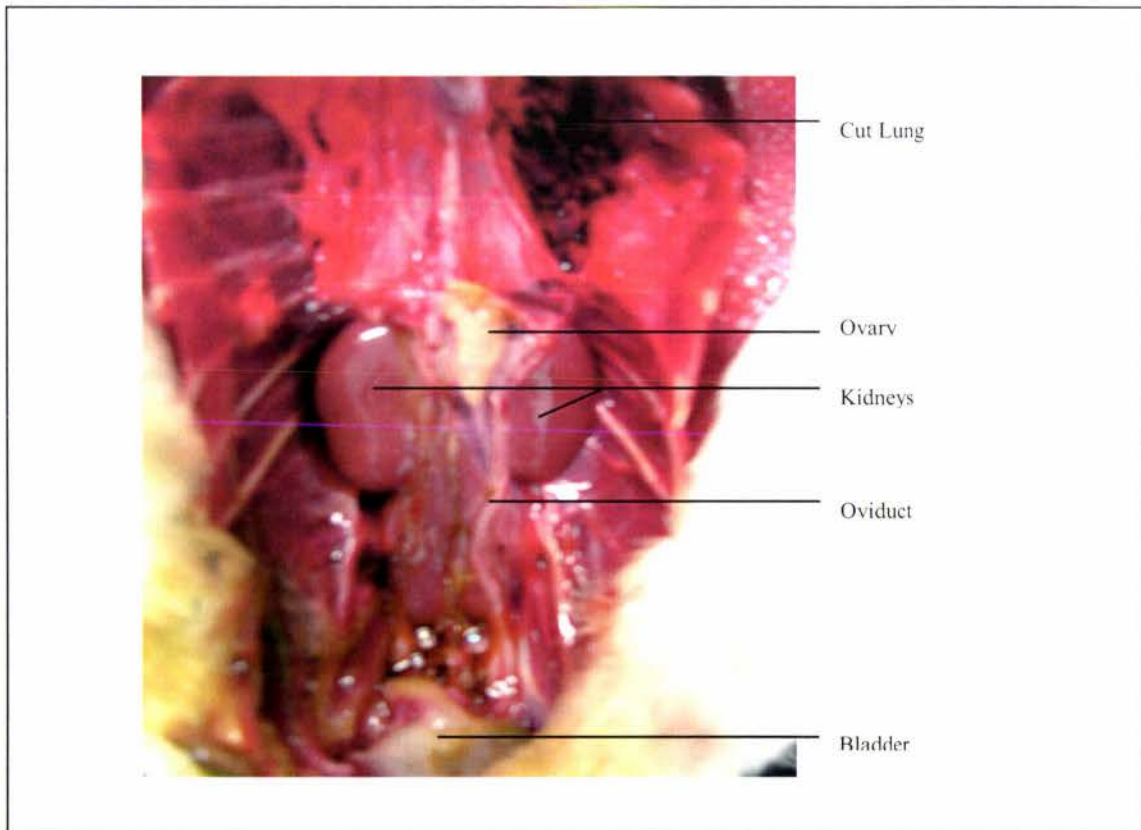


Figure 5.3. Diagram of a female Little Blue Penguin. Photo by J.Geurts (2006).

5.4.4.3.8 Nervous System

The skull was cut into and the top section removed to reveal the brain. The brain was then inspected to reveal any bruising or bleeding in the brain was looked for and noted.

5.4.4.3.9 Bacterial Tests

From seven of the birds fixed organs were sent down to Massey Universities Wildlife Clinic in Palmerston North for histology tests. Tests looked for a range of infections (eg; any sings of asperogillosis, liver flukes, coccidia).

5.4.4.3.10 Parasite Analysis

Any parasite samples were sent to AgResearch for identification.

5.4.5 Necropsy Results

Several carcasses that had also been found during the 2005 North Island beach wrecks (mass mortalities of seabirds), had been sent to the Wildlife Clinic at Massey University Palmerston North. Necropsies were performed on these carcasses and included histology tests. Histology tests were also run on several necropsied birds found on Tiri.

5.4.6 Weather Data

Weather data were obtained from National Institute of Water and Atmospheric Science (NIWA) for the period of 1966 to 2006. Daily values and monthly averages were taken from two weather stations, Tiri and Whangaparaoa. These included daily storm event data, surface temperatures, rainfall, wind speed, and wind direction. Southern Oscillation data for the period of 1966 to 1999 were obtained through the meteorological database.

5.4.7 Data Analysis

5.4.7.1 2005 and 2006 beach counts

Number of dead LBP found on Tiri and other Gulf Harbour regions were correlated with storm data such as wind speed and rainfall. Weather parameters were averaged over 15 days and correlated with counts taken over the same period using Spearman's rank (SAS V.8.0)

5.4.7.2 OSNZ data from 1966 to 1999

From OSNZ beach counts of LBP from 1966 to 1999 were compared by year and region. The search effort in kilometres travelled (KMT) and kilometres travelled per card (KMC) was also totalled for each year and correlated with the total dead penguin counts recorded for each corresponding region (Spearman's rank *SAS v.8*). Counts for all regions were totalled over each year and standardised for search effort (KMT). Standardised counts were then correlated against the Southern Oscillation Index values (SOI) by using Spearman's rank correlation (*SAS v.8*). Correlations were also done on SOI with the following year beach counts, to test for a delayed effect.

Total penguin counts were pooled for all years according to month. Each peak year was assessed based on the month that peaked the largest. Total dead penguin counts were also totalled for three New Zealand east coast regions (Northland East (NE), Auckland East (AE), and Bay of Plenty (BOP)).

5.5 Results

5.5.1 Necropsy Findings

A total of 96 birds were found dead on Tiri and 13 from other Hauraki Gulf regions (Tawharanui, or at sea) from the 15th of October 2005 to 30th May 2006. More dead birds were found during late September and early October, 2006 ($n = 55$) and were recorded ad hoc by Department of Conservation volunteers (Price *pers. comm*).

Of 85 birds found dead from Dec 31st to 31st April 26% ($n = 22$) had not moulted and still had old plumages while 24% ($n = 21$) had only just completed moulting and 9% ($n = 8$) were halfway through moulting.

A total of 53 birds were necropsied, 45 from Tiri and 8 from other Hauraki Gulf areas (Table 5.2). Ten of the chicks were only opened to look for fat and muscle levels as they were too decomposed to necropsy.

The average dead weight of LBP was lower than that of the banding weights for both males and females (Table 5.3). Overall average adult weight at death was $465.82\text{g} \pm 10.10\text{ SE}$ ($n = 39$, range 325 – 560) and chick weight was $280\text{g} \pm 32.10\text{ SE}$ ($n = 4$).

Eight of the birds had ticks taken from the ears. The number of ticks ranged from 1 to 8 per bird. Ectoparasites were found within the plumage of 11/44 birds, and 5/44 birds had lesions on the feet and under the flippers.

The muscle mass of the adults was generally very low. Of all the adults only one (1/30) had signs of subcutaneous fat (level 3).

Table 5.2 Age of Little Blue Penguin found dead on Tiritiri Matangi Island and other Hauraki regions refers to total sample size for each age group.

| Age | N | Male | Female | unknown |
|-----------|----|------|--------|---------|
| Adult | 23 | 11 | 20 | 8 |
| sub-adult | 6 | | | |
| Chicks | 15 | 5 | 5 | 5 |
| Unknown | 9 | 0 | 0 | 0 |

Table 5.3 Weights (grams \pm SE) of male and female Little Blues Penguin found dead compared to weights of LBP found alive on Tiri during 2005/ 06. Number in parentheses refers to the sample size for each group.

| Weight | Male | Female |
|-----------|-------------------------|-------------------------|
| Alive (g) | 907.64 \pm 19.40 (49) | 857.91 \pm 24.62 (40) |
| Range | 731.25 – 1125.00 | 507.00 – 1229.00 |
| Dead (g) | 494.93 \pm 22.07 (11) | 446.30 \pm 13.24 (20) |
| Range | 350.00 -550.00 | 325.00 – 565.00 |

Body cavities of all the birds appeared normal except that 3/ 30 carried excess abdominal fluid. 1/ 30 of the birds also had a broken sternum. Hearts appeared normal in 30/30 birds and all the livers appeared normal apart from post-mortem autolysis and discolouration 2/30. However, 2/30 of the birds appeared to have slightly swollen lobes associated with the right side of the liver. The majority of the birds had empty stomachs and intestines, although (1/ 15) of chicks had a full stomach and (4/ 30) adult stomachs had feathers inside or plant material.

A total of 11/ 43 birds found either on Tiri or Tawharanui (not including the 10 chicks) had intestinal worms occurring within some of the intestine or though out the whole intestine length. Cestode load extended from 1 to more than 30 in an individual. There was bleeding within the stomachs and intestines of 14/44 however no stomach lesions or ulcers were found.

All kidneys appeared normal upon visual inspection. The brains appeared normal apart from one having enlarged blood vessels and one other having a jelly-like appearance and dark red colouration.

5.5.2 Cause of Death

5.5.2.1 Laboratory Tests

Histopathology tests were done on fixed samples of the brain, intestines, spleen, kidney, lung, heart, testicles, and liver taken from an adult male from the 2005 mass beach wreck. All organs were found to exhibit freezing artefact, post-mortem bacterial proliferation, and autolysis. The liver showed possible peracute bacteracemia which could have been a result of poor body condition. The addendum showed a negative gram stain with no bacteria seen in the liver section.

The same tests were performed on six fresh birds collected from Tiri on the 5th February 2006 and two from 2nd April 2006, and showed that all the tissues had variable degrees of autolysis ranging from moderate to severe. All of the eight samples showed signs of moderate haemosiderosis within the liver. This is usually associated with excess iron due to catabolism of muscle tissue associated with starvation or wasting diseases (Gartrell *pers.comm*). From the six fresh birds four of the sampled kidneys showed signs of trematode cross sections and a large number trematode eggs within the collecting ducts and ureters.

There were no other significant findings within any of the other organs tested (heart, ovaries, thyroids, air sacs, or adrenal glands). Diagnosis of birds was primarily starvation and wasting with secondly effects due to renal trematodiasis.

Cestodes found in the intestines and lower stomach of the birds necropsied were later identified as a *Tetrabothriidae* sp. unknown species level.

The ticks that were removed from the ears and plumage of different birds were all the biting tick (*Ixodus eudyptidis*) and the liver trematodes were unidentified (Table 5.4).

Table 5.4 Parasites found associated with Little Blue Penguin. All ectoparasites were taken from chicks, and out of nest material samples. Parasites associated with Little Blue Penguin in Australia and New Zealand that has been found in the literature are compared with this study.

| Parasite Type | | | Current Study | Past Literature | |
|----------------------|--------------|----------------------------------|---------------|-----------------|-----------|
| Common Name | Order | Species | | New Zealand | Australia |
| <i>Ectoparasites</i> | | | | | |
| Flies | Diptera | <i>Asteia tonnoiri</i> | √ | | |
| | | <i>Lucilia sericata</i> | √ | | |
| Flea | Siphonaptera | <i>Parapsyllus longicornis</i> | √ | √ | |
| Louse | Phthiraptera | <i>Austrogoniodes waterstoni</i> | | √ | |
| Mite | Acarina | <i>Veigaia</i> sp. | √ | | |
| Ticks | Acari | <i>Ixodus eudyptidis</i> | √ | √ | |
| <i>Endoparasites</i> | | | | | |
| Coccidia | | <i>Renicola</i> sp. | | √ | √ |
| | | <i>Contracaecum spiculigerum</i> | | | √ |
| Nematodes | Ascarid | <i>Contracaecum eudyptulae</i> | | | √ |
| | | <i>Tetrabothriidae</i> | √ | | √ |
| Worms | Cestoda | | | | |
| | Coleoptera | | | | |
| Liver Flukes | Trematode | <i>Mawsopotrema eudyptulae</i> | | | √ |
| | | Unknown | √ | | |

5.5.3 Analysis of Beach Counts

5.5.3.1 2005-2006 counts

The frequency of dead birds obtained from the Tiri coast and other Hauraki Gulf regions were correlated against wind speed and precipitation (Figure 5.4 a & b) using Spearman's rank correlation. Results showed that there was no significant relationship ($n = 30$, wind $r_s = -0.148$, $p = 0.436$, rain $r_s = -0.250$, $p = 0.812$) between weather and the number of dead birds per 15 days found during 2005.

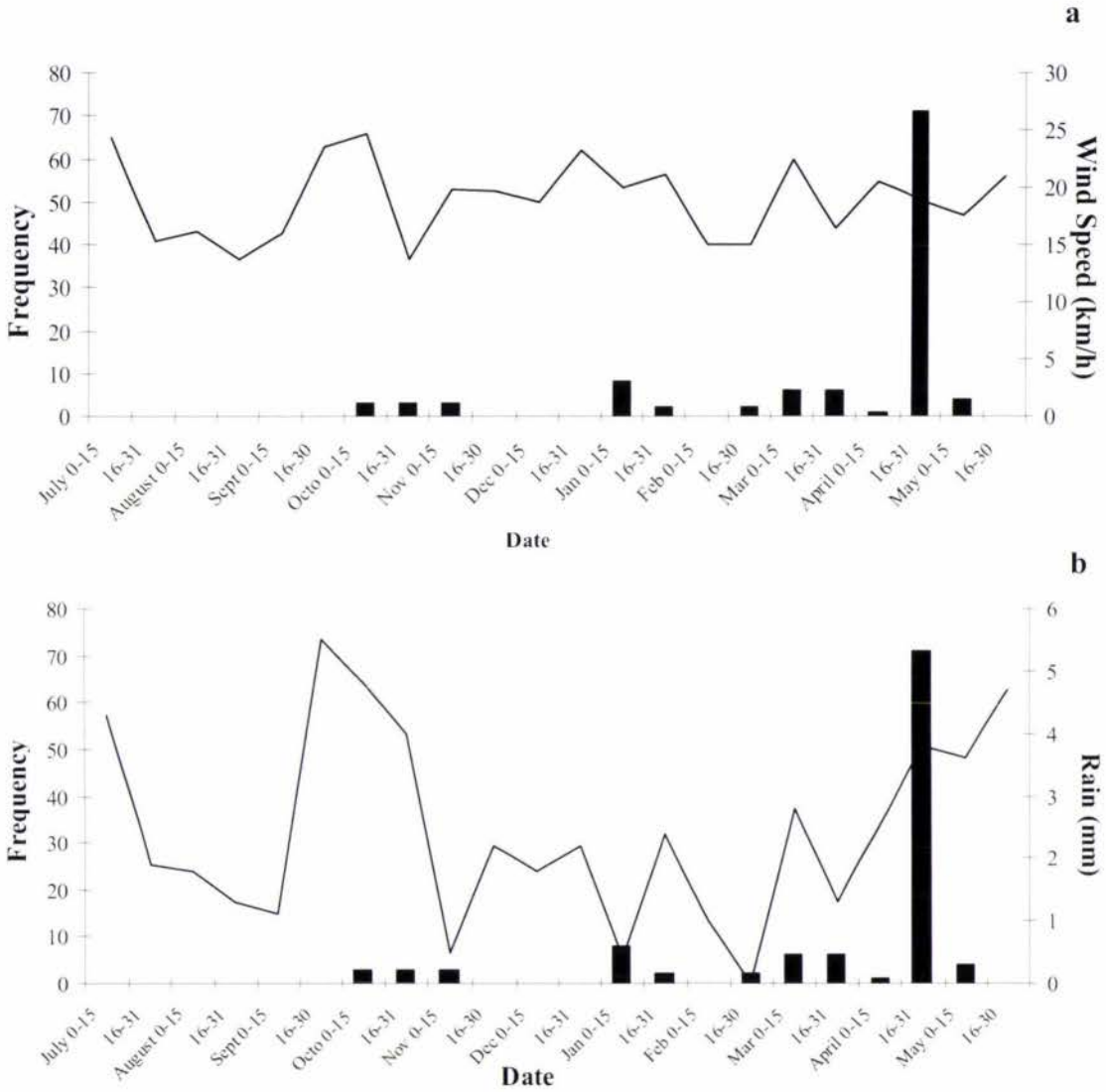


Figure 5.4 Total number of penguins that were found on Tiritiri Matangi Island during 2005 and 2006 against a) wind speed (km/ h) and b) rainfall (mm) averaged for 2 data points a month.

5.5.3.2 OSNZ data 1966 – 1999

5.5.3.2.1 Yearly counts

The correlation (Figure 5.5) between search effort (KMT and KMC) and the total counts of LBP found dead was significantly high (KMT: $n = 34$, $r_s = 0.684$ $p = <0.0001$; KMC: $n = 34$, $r_s = 0.660$ $p = <0.0001$) hence the total number of penguins for each year had to be standardised by dividing each year by the search effort. Results showed that there was no significant correlation between SOI and the number of birds found dead for each year ($n = 34$, $r_s = 0.238$ $p = 0.1757$) (Figure 5.6) or between SOI and the delayed effect ($n = 33$, $r_s = 0.079$ $p = 0.6635$). Only variation between years can be accounted for by SOI.

5.5.3.2.2 Monthly counts

Total counts for all areas from 1966 to 1999 were pooled for monthly comparisons (Figure 5.7). Largest numbers of LBP found dead occurred in January, April, and August. These peaks did not occur in the same way every year. Years tended to have single peaks.

5.5.3.2.3 Regional counts

Analysis of the different New Zealand regions (Figure 5.8) shows that the largest mortalities of LBP ($> 10,000$) were found in the Northern regions Auckland East (13,079) and Auckland West (13,713) of the North Island. The second largest counts (1000 – 10,000) came from other North Island regions Bay of Plenty (2279), North East (2411), North West (2656), and Wellington West (1286). Although, there were no counts in the North East and North West areas until 1995.

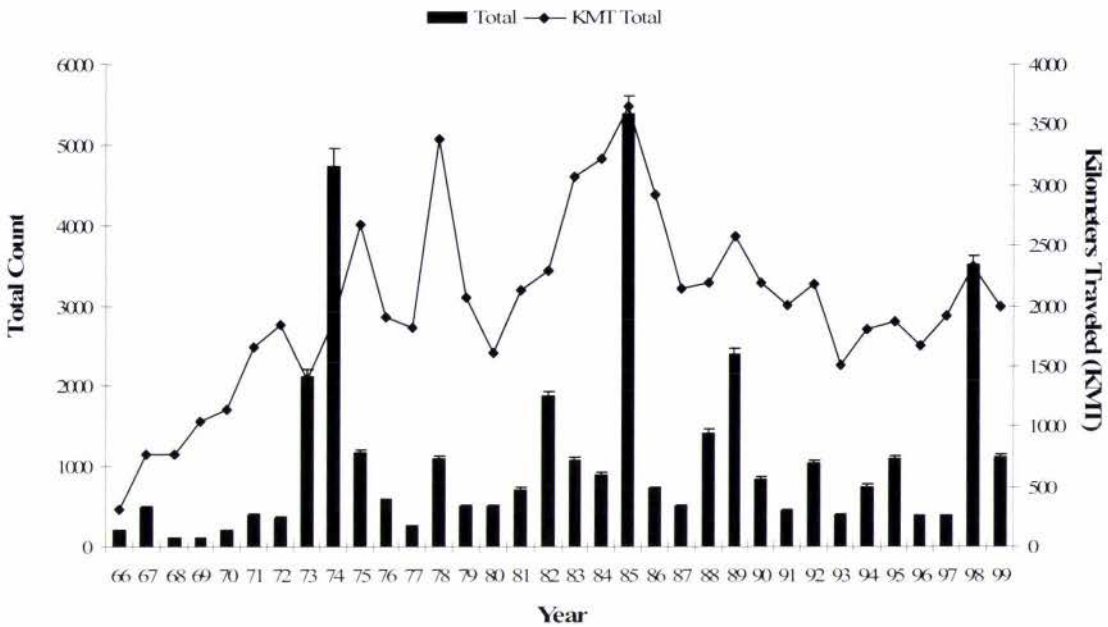


Figure 5.5. Total number (\pm SE) of dead Little Blue Penguins (LBP) found along all New Zealand beaches for each year and the search effort (total kilometers walked) for that year but only relevant to LBP counts. The number of penguins found is significantly related to the total number of kilometers (KMT) walked for each year (r_s 0.684, $n=34$, $p<.0001$).

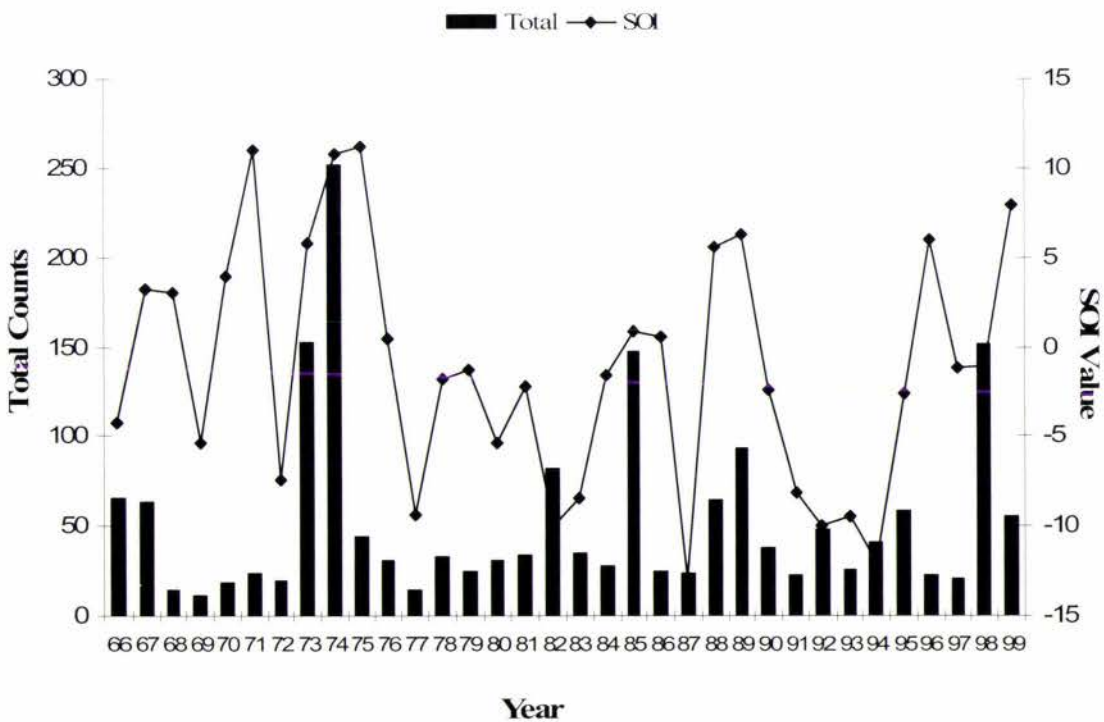


Figure 5.6. Standardised totals of dead penguins found for all years compared with the Southern Oscillation Index (SOI).

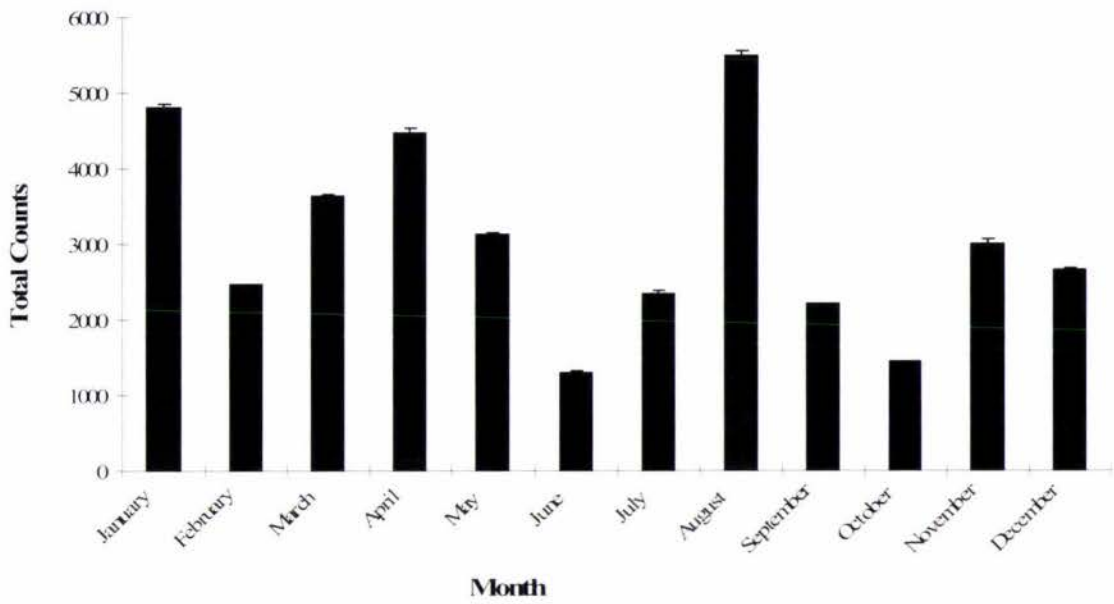


Figure 5.7. Total number (\pm SE) of dead Little Blue Penguins found along all New Zealand beaches covering a 33 year period from 1966 to 1999. Variability is from the differences between years.

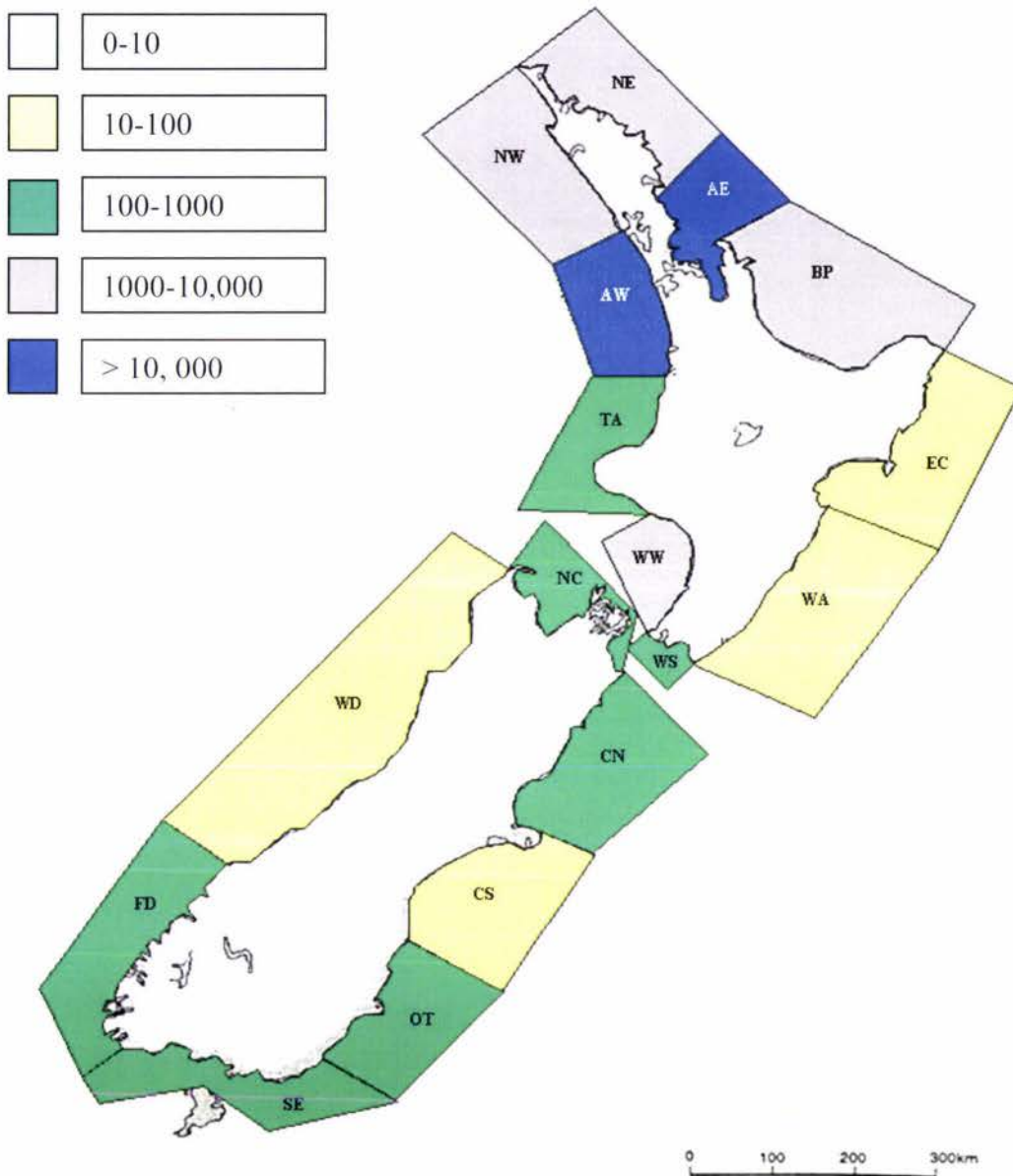


Figure 5.8 Total Little Blue Penguin beach counts for coasts sampled within New Zealand from 1966 to 1999. Abbreviations: North East (NE), North West (NW), Auckland East (AE), Auckland West (AW), Bay of Plenty (BP), East Coast (EC), Taranaki (TA), Wellington West (WW), Wairapa (WA), Wellington South (WS), North Coast South Island (NC), Canterbury North (CN), Westland (WD), Canterbury South (CS), Otago (OT), Fiordland (FD), Southland (SE). The arrows represent the boundaries. (Taylor 1997 modified, pp 202).

5.6 Discussion

5.6.1 Cause of Death in LBP

Large numbers of LBP were found dead during September and October 2005 and again in April 2006 on Tiri and other Hauraki Gulf regions. The majority of birds necropsied were adults however there were a large number found dead that were not able to be identified as adult or juvenile. Starvation was the largest cause of death for LBP found dead on Tiri during the 2005 and 2006 breeding season based on necropsy results. This was evident in the high levels of muscle wastage, no fat stores, and empty stomachs. Poor body condition was also evident in the initial external examination. In a past study dead LBP from Tiri were found to be on average 515g (Jones 1978) and those in this study were around 469 g on average. This suggests a critical threshold of weight for survival around 450 g to 500 g. The weights of dead birds were smaller than those described in other studies such as LBP from Victoria, Australia. Harrigan (1992) found the mean weights for dead penguins from 1984 to range from 524 to 773 g (mean 684 ± 73.1 g) and for 1985 range 564 to 772 g (mean 692 ± 60.0 g). Differences in lower thresholds of weight could be due to the different size morphologies associated with the different sub-species. Australian LBP are heavier than New Zealand LBP (Kinsky and Falla 1960).

Histology tests of the internal tissues confirmed starvation. In particular, the haemosiderosis within the liver is associated with excess iron due to the catabolism of muscle tissue in starvation or wasting diseases, and there was no evidence of other potential causes of death (Gartrell *pers. comm.*).

Further evidence of starvation was suggested by the stomach contents of necropsied birds, which in some adult birds included only debris such as stones, vegetation, and feathers. Harrigan (1992) has also found this during other LBP necropsies and suggests that non food items in the stomach result from failed attempts at finding something to eat. Otherwise the feathers could have been associated with the preening during the moulting period. The appearance of stones within the stomach has also been found in two other spheniscidae species and could be linked with gastrolithic processes or from “by catch” due to bottom feeding. According to Hocken (2005) the actual physiological purpose remains unknown.

Starvation has also been recognised for North Island LBP that were necropsied in 1974-1976 (Crocket & Kearns 1975; Jones 1978) and other studies have found starvation to be a factor associated with mass mortalities of LBP within Australia (Norman *et al.* 1992). The question remaining is why are LBP prone to starvation?

5.6.2 Patterns of Mortalities

5.6.2.1 Monthly counts

During the period of July 2005 to May 2006 two major peaks in mass mortalities of LBP occurred within the Northern region of New Zealand. The first period occurred during large storm events during September and October, however these were relatively low compared to previous findings. Peak mortalities in 1973 and 1974 occurred in August with >1500 and >2400 birds found dead, respectively (Jones 1978).

Examining long term trends showed that three periods were found to have the largest numbers of dead birds over the 33 year period; these were winter (July and August), the end of the breeding season (December and January), and the moulting period (February and April). This suggests that the different ecological stresses

experienced with each stage of the life-cycle need to be taken into consideration when trying to elucidate the underlying factors associated with mass-mortalities of LBP.

5.6.2.2 Annual counts

The peak periods of die-offs have not yet been explained. This is the first study to attempt analysis of mass mortalities of LBP within New Zealand using the OSNZ data and to assess the potential long-term trends.

Peak mortalities were associated with August 1974, 1985, and 1998 but there is a significant correlation between the number of dead birds found and the search effort for each year. Nonetheless, when standardised for search effort, SOI values were not significantly correlated with the number of LBP found dead over the 33 year period and did not have a significant effect on the following year (delayed effect). Furthermore, years peaked at different months suggesting inter-annual differences. Therefore annual small-scale factors may explain the mortalities better than large-scale climate fluctuations.

5.6.3 Inter-Annual Differences and LBP Survival

5.6.3.1 Winter

5.6.3.1.1 Storms

The first die-off during August and October of 2005 could have been due to the stormy weather which could have caused fatigue and food deprivation. Winter is known for its low temperatures and increased stormy periods as opposed to other seasons which could directly or indirectly influence the survival of LBP. However, large numbers of LBP found dead in New Zealand during the 2005 winter period could not be explained by the level of wind speed or precipitation. This could be due to the lack of counts for the early

stage of the breeding season within the analysis, or due to the indirect or delayed effects of the storms.

Stormy periods occurred during September (16, 18-22) and October (7-12, 16-19, 26-30) with periods of wind speeds that reached up to 140km per hour. This could mean that if LBP are at sea they could find it hard to surface and potentially drown, or if on land it could prevent them from foraging effectively. Furthermore, since LBP rest on the surface of the water when at sea, storms may cause LBP to expend most of their energy just to stay afloat and dodge waves. Future monitoring of wrecks in relation to stormy periods is recommended since many seabird wrecks have been related to stormy weather (Hudson 1985; Sandvik *et al.* 2005).

5.6.3.1.2 Temperature

Winter periods have been associated with large mortalities of LBP in other studies (Reilly and Cullen 1979; Stahel and Gales 1987; Dann 1992; Harrigan 1992; Norman *et al.* 1992; Dann *et al.* 2000; Johannesen *et al.* 2002) since days are shorter and colder meaning a shorter period to forage (Gales and Green 1990) and an increase in heat production (Baudinette *et al.* 1986). LBP increase their heat production through food intake (Baudinette *et al.* 1986) therefore low food intake would decrease their ability to thermoregulate.

5.6.3.1.3 Food availability

Other indirect effects on mortality can occur through climate change which can influence food availability by decreasing primary productivity (Fromentin and Planque 1996; Heath *et al.* 1999; Sandvik *et al.* 2005) which is the food source bait fish stocks (Alheit and Hagen 1997; Ottersen and Loeng 2000; Hjermann *et al.* 2004; Sanvik *et al.* 2005) (Chapter 4).

Many seabird wrecks have been identified as the result of changing weather conditions such as sea surface temperature, *El Niño* and *La Niña* years (Sanvik *et al.* 2005). The SOI values did not explain the variation between years of LBP found dead along New Zealand coasts since 1966. These mortalities may therefore be related to more indirect factors such as food supply (Sandvik *et al.* 2005). There was a large mass mortality of pilchard in 1995 and 1998 in Australia, and also New Zealand in 1995 however, the effects of this on LBP is unknown. Food availability will also influence the individual ability of LBP to forage and provision chicks which may result in chick death or low body weight at fledging. Studies have found that LBP chicks that have low fledging weight have an increased chance of mortality within the first year of fledging (Dann and Cullen 1983; Dann 1988; Dann 1992).

It has been suspected that mass mortalities of a major prey species for seabirds may have been associated with the number of other dead seabirds (Sooty Shearwaters, Buller's Shearwaters *Puffinus bulleri*, Mottled Petrels *Pterodroma inexpectata*, Grey-faced Petrels *Pterodroma macroptera gouldi*, Blue Petrels *Halobaema caerulea*, Antarctic Petrels *Thalassoica Antarctica*, and Light-mantled Sooty Albatrosses *Phoebastria palpebrata*) in 1995 since they were recovered at higher than normal rates (Taylor 1997). Several studies have shown that there is a significant correlation between prey abundance and seabird reproductive biology. An example of this is the effect that *El Niño* has had on the spatial redistribution of anchovies (*Engraulis spp*) and sardines (*Sardinops sagax*) a major prey item of Sooty Shearwaters (*Puffinus griseus*) (Barber and Chavez 1986, Philander 1990, Lyver *et al.* 1999).

It is unknown what may have caused changes in prey availability for this study but it could be related to seasonal life-cycle of prey species, competition within and between secondary consumers, changes in the thermocline and nutricline, or the ability

to obtain food (i.e. individual ability, foraging restrictions such as time and foraging range) (Chapter 4). For example, during winter the movement of major prey species such as schooling fish (e.g. Anchovy) during winter and the inability to forage large distances (> 20km) from their breeding site will limit the local abundance of prey and the catch of energetically profitable prey (Chiaradia *et al.* unpubl. data).

El Niño conditions (SOI) have caused some of the largest responses in seabirds (e.g Barber and Chavez 1983; Duffy 1990; Chastel *et al.* 1993; and Sanvik *et al.* 2005) and needs to be taken into consideration when looking at mortalities and breeding success.

5.6.3.2 *Breeding season*

Beach counts affect dead penguins within this study were low during the breeding season but were much higher later in the year. Also more females were found compared to males. The 2005/ 06 breeding season started later than other years (Chapter 2) which could be related to a delay in prey availability.

The breeding season has been recognised as a demanding stage of the LBP lifecycle due to the demands of providing enough for chick survival and the fasting periods associated with incubation and chick rearing. For example, 31% of the annual energy budget is required for chick rearing in LBP (Gales and Green 1990). Furthermore, for the females this is increased due to the resources that are required to produce and lay eggs (Gales and Green 1990). For example, egg production in lesser black backed gulls has been found to deplete a females body reserves (Houston *et al.* 1983; Monaghan *et al.* 1998) which may hinder behavioural performance and survival during other later stages of the life-cycle (Nager *et al.* 2001). Herring Gulls have also

been associated with a peak in deaths during the late summer after breeding (Coulson *et al.* 1983; Nager *et al.* 2001).

Furthermore, one chick was found dead on the rocks due to drowning since the chick had not yet developed adult plumage for waterproofing, since it was not old enough to fledge. Starvation is thought to be a factor driving young unfledged chicks off the nests in search of food. Juvenile birds that have recently fledged at the end of the breeding season will have to rely on their fat stores until they have learnt to feed (Dann 1992). A large number of juveniles were not identified within this study however, this may be a factor associated with the number of birds that were able to be necropsied or identifiable as adult/ sub-adult upon necropsy analysis.

5.6.3.3 *Post- breeding season and moulting*

5.6.3.3.1 *Constraints of breeding and moulting*

The die-offs of early 2006 could have been a combined effect of the demands associated with moulting and the delay in the 2005 breeding season. Moulting was considered a potential factor in mortalities as 26% of the birds found dead on Tiri had not moulted and showed evidence of old plumage, 24% had begun moulting and 9% were halfway in advanced moult. Furthermore Bird Rescue New Zealand had the largest number of sick penguins in care during this period (2005/06) and many had failed to moult completely (Durrant *pers comm*). Also during April 2006 large numbers of the birds were found dead while ashore waiting for their moult to finish. Several LBP were also found along the Tiri coast alive but were weak, with low weights and unable to get under cover. Other carcasses were found just above the high tide mark and appeared to have died on the spot.

A delay in the initiation of the breeding season resulted in an overlap of these two very energetically demanding biological processes with no recuperation period between them. For example in the Falkland Islands the Rockhopper Penguin populations suffered a substantial decrease during the 1987 moult period and mass starvation due to the inability to lay down fat reserves (Keymer *et al.* 2001; Hilton *et al.* 2006). The later the breeding period the shorter the delay before moulting and the ability to increase fat stores to take on a 15 day fasting period which could cause fatigue and death in LBP (Reilly and Cullen 1982; Hull *et al.* 2001).

Near the end of the breeding season several of the adult birds had started moult while still rearing chicks, these birds later abandoned their nest. Reilly and Cullen (1979) and Dann *et al.* (1992) have found that dead penguins recovered from beaches are those that are newly moulted and starved suggesting that the body condition prior to, and during the moult was very low. Johannesen *et al.* (2002) found that the cost of moulting in LBP is high with the largest number of mortalities occurring in post-moult months compared to the rest of the year.

5.6.3.3.2 Implications of moulting

The ability of penguins to dive and forage especially within cold waters, is dependent on the quality of their plumage (Dawson *et al.* 1999; Green *et al.* 2004). This may be important for LBP since the sea water within the Hauraki Gulf can reach levels of 9-11°C. According to Stahel and Nicol (1982) a 900g LBP has a lower critical threshold of 10°C for neutral thermoregulation, similar to Adélie Penguins (*Pygoscelis adeliae*) (Chappell *et al.* 1989) and higher than an Emperor Penguins (*Aptenodytes forsteri*) which has a lower critical threshold of -7 °C (Pinshaw *et al.* 1977).

Many penguin species are able to fast during breeding (Croxall 1982) and successfully moult in some of the most difficult environments on the planet (e.g. Emperor penguins, Marchant and Higgins 1990). The energetic cost of moulting is large due to the development of new tissue and reduced thermal insulation and as a consequence, energy is directed to heat production (King 1980; Green *et al.* 2004). For example a study on Macaroni Penguins (*Eudyptes chrysolophus*) found that the metabolic rate increases during the moult since feather loss increases the vascularisation of the skin and heat production must be increased (Green *et al.* 2004). Furthermore, the higher metabolism associated with moulting LBP increases hematological levels, lowers red blood cell counts and an increase packed cell volumes (Sergent *et al.* 2004).

Fasting reserves decrease up to 50% of the body weight and relies upon lipid stores obtained prior to the moult (Williams *et al.* 1997; Cherel *et al.* 1994; Green *et al.* 2004). During moult, birds are not able to forage at sea since body condition is low (Groscolas 1986; Green *et al.* 2004) and waterproofing and insulation is reduced (Erasmus *et al.* 1981; Reilly and Cullen 1982; Green *et al.*). Therefore peak mortalities often occur after moulting (Boersma 1978; Van Heezik and Davis 1990; Dann 1992; Cherel *et al.* 1994; Green *et al.* 2004). Within this study peak mortalities occurred around the end of the breeding season and after moulting. The delay in breeding for 2005 meant that there would be less time between breeding and moulting which would therefore decrease the amount of time available to forage and build up fat stores.

5.6.4 Other Causes of Mortality

5.6.4.1 *Asperogillis*

Although one of the main causes of natural mortality of penguins has been associated with respiratory *Asperogillis* (Hocken 2005) this was not found to be a factor in terms

of mortality within this study. However, it must be noted that the watery lungs often assigned to death by drowning, could be due to this respiratory infection (Hocken 2005), and is hard to discriminate between the two.

5.6.4.2 Predation

No penguins were found to have wounds or symptoms that would suggest death by predation which is not surprising since introduced predators (i.e. cats, dogs, stoats) have been eradicated from the island (Chapter 1). A cause of death by marine mammals, sharks or fur seals (Spellburg 1975; Jones 1978) was not found for any of the penguins necropsied. However the potential for marine based predation could still be a factor as carcasses may not be recovered. Regardless of these results predation by marine or terrestrial predators may still be influencing LBP survival within the Hauraki Gulf.

5.6.4.3 Parasites

Both ecto- and endoparasites were found to be associated with LBP carcasses within this study. Ticks, fleas, and mites were found around the beak, ears, and eyes and within the plumage. Compared to other studies low levels (< 30) of intestinal worms were found within a small number of birds and histology results confirmed evidence of liver flukes (*Mawsopotrema eudyptulae*) and kidney cestodes (unknown species). There has been little work done on the parasite species and loads that are associated with LBP and therefore it was hard to assess impacts or occurrence rates and persistence levels within naturally occurring populations.

5.6.4.4 Ectoparasites

There is also no record of the fly species *Asteia tonnoiri* or *Lucilia sericata* which were found on LBP within this study. The flea species *Parapysyllus longicornis* was also

identified on LBP during 1974-1975 (Jones 1978) though its effects on LBP are unknown.

The tick *Ixodus eudyptidis* is known to be a seabird parasite confined to New Zealand and Australia and is known to have the ability to paralyse host species (Heath, unpubl data). Not a lot is known about their life histories but they are often associated with nesting since they require different hosts and can last until the next breeding season.

Most ectoparasites have the ability of transferring diseases such as avian malaria, Newcastle disease virus and fowl pox (Williams 1995). For instance *Ixodes uriae* is a widespread tick associated with many seabird species such as the king penguin (Murray and Vestjens 1967; Gauthier-Clerc *et al.* 1998; Bergström *et al.* 1999b, Frenot *et al.* 2001; Mangin *et al.* 2003), and is known to be a vector of viruses (Nuttall 1984; Mangin *et al.* 2003) and Lyme disease agent *Borrelia burgdorferi* (Olsen *et al.* 1995; Gauthier-Clerc *et al.* 1999; Mangin *et al.* 2003). Ticks in the king penguin are thought to cause blood loss and anemia in adults and chicks (Chastel *et al.* 1987; Mangin *et al.* 2003), and cause large featherless patches in adults which may influence heat loss (Mangin *et al.* 2003). Furthermore, several adults and chicks of the Rockhopper Penguin (*Eudyptes chrysocome*) have been killed in the past due to the spread of *Pasteurella multocida* by ticks (de Lisle *et al.* 1990; Chen 2004). Lastly, tick outbreaks have occurred in a range of bird species such as Pelicans (King *et al.* 1977), Prairie Falcons *Falco mexicanus* (Bodner 1980), Common Murres *Uria aalge* (Ballard and Ring 1979), Albatross *Diomedea cauta* (Johnstone *et al.* 1975), and Brown Booby *Sula leucogaster* (Reithmuller 1931) resulting in colony desertion, blindness and death of young, and possibly adult death (see Duffy 1983).

Several of the necropsied birds had lesions on the flipper and on the feet which may be due to tick infestations. The Humbolt Penguin in Peru suffer from pruritus and slowly healing blisters as a result of infestation by the tick *Ornithodoros Alectorobius carpensis* (Hoogstraal *et al.* 1985). The influence that associated parasites had on the cause of death for LBP within this study is unknown and no information (apart from several cases of the King Penguin) of death in adults through tick infestation is available (Gauthier-Clere *et al.* 1998). Death of several adults as a result of ticks in the King Penguin *Aptenodyptes patagonicus* differs from eudyptid penguins (Brooke 1985) since they aggregate, stand motionless for two week periods, and lack allopreening to remove ticks (Gauthier-Clere *et al.* 1998).

5.6.4.5 Endoparasites

The cestode *Tetrabothriidae* sp has only been associated with LBP within Australia and has not been recorded for LBP within New Zealand until this study. Even though intestinal cestodes (*Tetrabothriidae* sp) within LBP of 2005/06 were low compared to other studies that have found up to 300 nematodes in LBP stomachs (Obendorf and McColl 1980), it is also uncommon for LBP to have tapeworms or round worms (Hocken, 2002).

Liver flukes are not thought to be common within LBP but the *Mawsonotrema eudyptulae* sp. have been found in another study by Harrigan (1992). This is thought to be the most pathenogenic of the parasites for LBP in Victoria, Australia (Harrigan 1992). Renal coccidiosis has also been found within the same study and is associated with white spots or patches found on the kidney (Harrigan 1992). High infestation of gut and renal parasites have also been recorded in LBP by Crocket and Kearns (1975).

Although blood was found in the gut and intestines of LBP necropsied within this study evidence of ulceration was not found. LBP with ascaidoids, and trematodes have resulted in stomach and gastric ulcers and haemorrhaging (Harrigan 1992). Gastrointestinal bleeding could be mistaken for symptoms of parasitic infection, however it has also just been associated with dying birds with no gastric parasites (Hocken 2001). On the other hand the infestation of ectoparasites may not show visible effects meaning that their presence could often be missed without further tests. Crockett and Kearns (1975) examined beach wrecked birds in Northland New Zealand during the late winter period of 1973 and late autumn of 1974. Results showed heavy loads of renal trematodes which appear to cause little inflammatory reaction (Crockett & Kearns 1975). Furthermore, internal parasites of the Genus *Contracoecum spiculigerum* was found in 67% of penguins in a study by Obendorf and McColl (1980) and were associated with gastric ulceration. The intestinal cestode *Tetrabothrius sp.* (also found within this study) was associated with 83% of LBP and other cases included intestinal coccidian and/ or trematodes (Obendorf and McColl 1980). They concluded that all the parasite types caused gastric ulceration, and associated intra-gastric hemorrhaging which was not apparent within this study.

It is unknown to what extent endoparasites occur naturally within LBP and what effect they may have. Mass mortalities associated with death within LBP due to endoparasites have been associated with very high levels of parasitic loads, however, this was not found within the current study. Regardless of low levels, parasite impacts are likely to increase during periods of high stress and low body condition (Ranum and Wharton 1996), therefore endoparasites may accentuate the primary cause being starvation or low immunity (Obendorf and McColl 1980). Parasitic infection has also been largely associated with juvenile LBP causing debilitation and emaciation (Harrigan

1992). Furthermore, cestodes may stop digestion by blocking the alimentary canal, or depress appetite through blood loss (Obendorf and McColl; Norman *et al.* 1992). Ectoparasites are also thought to have been a large factor in cause of death by causing anorexia and starvation. A wreck of LBP within Western Australia during 1986 was also associated with high levels of parasites (nematodes, trematodes, and cestodes) suggesting that their influence could have been heightened during periods of storms during winter (Norman *et al.* 1992). This suggests that more investigation is required on the potential effects of parasites is required.

The most likely source of intestinal worms has been suggested to come from an intermediate host such as a prey species (Norman *et al.* 1992; Harrigan 1992). Larval nematodes of the genus *Contracaecum* sp. have been identified as Red Cod (Ranum and Wharton 1996). If this prey species occurs within the diet of LBP on a seasonal basis, then this would assume that parasitic infection of this type is not a chronic occurrence within LBP but a seasonal one. A red cod type was found in LBP stomach content analysis (Chapter 4) consistent with the parasitic-host association, however the intake rates of red cod is unknown from this study. If during the summer months preferred prey abundance is decreased due to warm weather and lifecycle traits then diet switching to sub-optimal prey species such as red cod could mean two things, 1) a decrease in obtainable energy and protein stores as well as 2) an increase in parasitic infestation. In comparison, no healthy birds have been necropsied to identify if they are able to survive with these parasites. Therefore, it would be of interest to consider the effects that diet switching will have in terms of secondary parasitic effects (Norman *et al.* 1992).

5.6.5 Other Considerations

Due to financial constraints and extensive autolysis in some of the tissue few histology tests were achieved. More tests are required to appropriately identify the primary cause of death for the LBP species within the North Island region.

Organs were frozen in case there was a need to test for toxins such as PCB's and DDT's however histology tests suggested this was not necessary. Veitch (1975) and Crocket and Kearns (1975) tested for heavy metals and chlorinated carbons in beach wrecked LBP within the North Island but found nothing significant. Toxicology tests are expensive and require reference to particular toxins. A recent toxicology study has revealed the occurrence of various trace elements, PCB's and organochlorine pesticides in Common Dolphins (*Delphinus delphis*) from the Hauraki Gulf, New Zealand, including DDE and its derivatives (Stockin *et al.* in review). Furthermore, Yellow-eyed Penguins *Megadyptes antipodes* have been found to contain heavy metals such as arsenic, cadmium, and mercury within the liver however levels were low and suggested that they had metabolic and elimination pathways (Numata *et al.* 2005). It would be worthwhile to test the PCB and DDE levels in starving LBP as other studies have suggested that starving birds are at a higher risk of toxic shock as DDE is released into the body and brain when adipose reserves were mobilized (Lyver *et al.* 1999).

Disease has also been recognised as a potential factor associated with penguin species (Moore *et al.* 2001; Hocken 2005), and Shumway *et al.* (2003) have argued that algal blooms and marine toxins are a cause of seabird wrecks, in particular LBP and yellow-eyed penguins (Hocken 2005) and potentially in LBP within Port Bay, Australia during 1984 (Norman *et al.* 1992). However, there were no algal blooms during the 2005/ 06 breeding season (Leigh Marine Laboratory, *pers comm.*).

The lack of effect of SOI on annual counts of LBP should still be considered in future analysis of the mass mortalities occurring. Recommendations include taking into account sea surface temperature since this may have more of an effect on prey availability than SOI values (Sandvik *et al.* 2005). Analysis should also take into account SST and SOI values which relate to the periods of breeding, moulting and winter.

5.6.6 Mortalities and Population Demographics

The survivorship of adults and juveniles needs to be established together with the fidelity of individuals to a breeding site since these will have implications for changing the age structure of colonies. Clearly lower survival of adults and juveniles will cause a population to decline. Monitoring of the Fiordland Crested Penguin (*Eudyptes pachyrhynchus*) which is in population decline, has found that low recruitment of juveniles is caused by high very high mortality (Newton and Tansell 2005). Regardless, banding studies of LBP will allow for identification of the dispersal of LBP from an area and aid future monitoring of the population viability.

Furthermore, LBP populations are thought to be able to recover after mass events since they are able to lay two clutches within a breeding season and have the potential to raise two broods a year (Taylor *pers. comm.*). In contrast, LBP on Tiri during the 2005/06 breeding season and in past monitoring (Jones 1978; Chen 2004) have not raised any double broods and have had very low breeding success compared to other populations (Chapter 2). Monitoring of survival for any species, especially long-lived seabirds that are known for their low reproductive output (Sandvik *et al.* 2005), is important in determining population and species stability.

5.6.7 Conclusions

This study is the first attempt at identifying the factors influencing LBP within New Zealand through short-term analysis of mortalities and long-term analysis of beach counts. The large number of incidences involving beach wrecked birds (Veitch 1975; Jones 1978) suggests mortality in LBP is high (Veitch 1975; Crockett and Kearns 1975; Jones 1978) and that starvation is an important factor.

The identification of starvation via necropsies as the major cause of death within this area suggests that a more detailed investigation into the feeding ecology of this top marine predator is required. A greater understanding of the behaviours and abundance levels of major prey species is also necessary to understand the marine factors which are impacting on the LBP survival. Impacts of climate change have been identified in many seabirds and still needs consideration since indirect effects may not be identifiable through direct analysis. Competition may be a factor associated with LBP foraging within the Hauraki Gulf and requires further investigation as to whether this is negative or positive in its effects.

Furthermore, this is also one of the first known studies to identify parasites associated with LBP within the North Island of New Zealand however more research is needed to identify the role that they have in LBP survival.

Conservation objectives associated with ensuring a species survival are only attained when long term monitoring of simple baseline data is achieved. This requires individual identification, to monitor initial population sizes, dispersal patterns, recruitment rates, and survival rates. Once this is obtained occurrences such as mass mortalities can be assessed in terms of the impact that they can have at a population level. Knowledge of simple baseline data for many populations within a species will

then allow for a meta-population approach to conservation. It is obvious that populations can vary in their susceptibility levels and different risks (i.e. predation, food supply, climate changes) are associated with each. Understanding susceptibility is important when applying strategies to mitigate the effects of these threats.

CHAPTER 6 The use of Little Blue Penguins as biological indicators



Plate 6.1. Moulting Little Blue Penguin. Photo by J.Geurts 2006

6.1 General summary

6.1.1 Conclusions

In general, penguin behaviour and ecology has been shown to be influenced by weather (Perriman *et al.* 2000), predators, anthropogenic factors (human disturbance, habitat change) (Dann 1992), and food availability (Furness 1990; Cullen *et al.* 1992; Chiaradia *et al.* 2003). Many LBP colonies in Midland Australia, Tasmania and the North and South Islands of New Zealand have apparently declined (Lawrie *et al.* 2005)). In Otago, New Zealand, LBP no longer breed at seven previously occupied sites (Dann 1994). Most viable populations exist only on offshore islands but due to very limited monitoring it is unknown if these populations are increasing or decreasing (Dann 1994).

Knowledge of the current breeding and foraging biology of this sub-species is lacking. Apart from long term monitoring on Motuora Island, Wellington, only three research efforts have been made on Tiritiri Matangi Island within the North Island. Without future monitoring of the LBP on Tiri estimates such as population stability or decline will not be obtainable. According to the Department of Conservation (DoC), successful monitoring requires an index of the abundance, information about breeding ecology, and an understanding of foraging ecology (Taylor 2000). Now that valuable baseline data has been obtained from the current study and incorporates information from other years (where possible) this can be used to contribute to future conservation efforts.

The extent to which Little Blue Penguin (LBP) can be used as biological indicators was investigated in this study by using basic population monitoring and new techniques (e.g. isotopes, egg development). The key factors associated with the

breeding success, egg and chick success, foraging ecology, and survival were identified over the short-term and where possible over the long-term. The results suggest that LBP are good indicators, however, establishing causal parameters on both scales has been difficult and requires further investigation. Regardless, sound baseline data has been gathered which will act as a building block for future monitoring and of the North Island sub-species of LBP.

6.1.2 Interrelating Factors

The lifecycle of the LBP reflects their dependence on both terrestrial and marine environments. Both biotic (foraging behaviour and diet, lay date, age and experience, and fidelity of breeding pairs) and abiotic (climate, sea surface temperature: SST, and microhabitat) parameters are associated these two different mediums. The breeding success and survival of LBP but are highly interrelated, therefore key factors need to be measured and the potential relationships explored (

Figure 6.1). The links that exist between different factors were the focus of this study and other studies on penguin species and seabirds.

The life-cycle of the LBP can be viewed as an energy budget and LBP must be able to balance energy expenditure and energy gain to survive and maximise reproductive output (Baudinette *et al.* 1986). Energy is associated with food, while energy expenditure varies with activity budgets and physiological costs such as egg production and moulting (Schreiber and Burgess 2002). The ability to balance expenditures will vary with individual abilities and characteristics (age, weight, experience, and sex) and with other factors experienced at any particular time (weather, prey availability, and season).

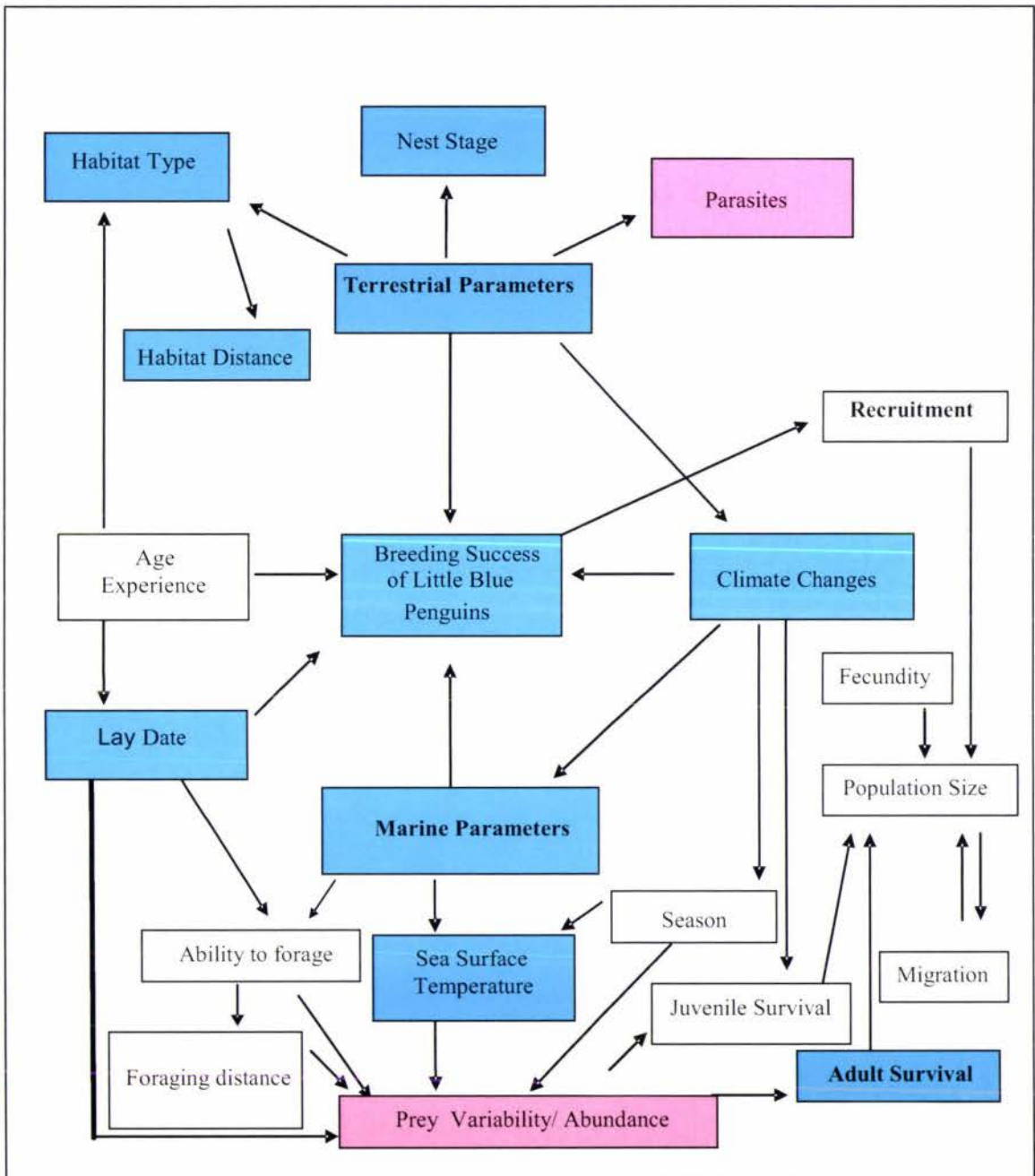


Figure 6.1. Schematic flow diagram of the linkages between marine and terrestrial environmental parameters on breeding success and the influence of these on population demographics (i.e. population size, juvenile and adult survival, fecundity, migration, and recruitment). Highlighted areas indicate factors important to Little Blue Penguin from current study (Blue), less crucial factors (Pink) and factors found to be influential in other seabirds studies (white).

Lay date had the greatest influence on the breeding success and survival of LBP on Tiritiri Matangi Island (Tiri) (driven in part by SST). Other parameters such as habitat type and risks at both the egg and chick stage (temperature and starvation) also influenced breeding success but on a smaller scale compared to studies of other populations.

Laying early within the breeding season was the best determinant of fledging success in chicks and appears to be associated with low SST and hence higher prey abundances. Prey availability was important since LBP depend on finding enough food to meet energetic demands of travelling, breeding, moulting and self maintenance and different times of the cycle. The ability to meet these demands will mean that LBP must make at-sea decisions about foraging distance and depth to maximise their chances of successful breeding while still insuring a high probability of survival.

The survival of chicks is associated with the ability of parents to provision chicks adequately enough to meet the energetic demands of growth since large intervals between chick feedings will slow chick growth (Chiaradia *et al.* 2004). A comparative study on African Penguin *Spheniscus demersus* from different colonies found there is an increase in foraging effort when prey availability is low reducing chick growth (Peterson *et al.* 2006). Therefore, at-sea decisions are required regarding the foraging trip distance and duration. The Wandering Albatross (*Diomedea exulans*) alternates between short and long foraging trips depending on the stage of breeding (incubation/chick rearing) and the costs associated with each (Shaffer *et al.* 2003). This sort of behaviour is found in other seabird species such as Sooty Shearwaters (*Pterodroma griseus*) which undertake short and long trips (dual strategy) based on the body condition of the adult (Weimerskirch 1998). Extended foraging trips may buffer chicks against low food availability, despite reduced growth rates, but will impact on adult

body condition (Zador and Piatt 1999). Therefore, an alternative is to trade-off current reproductive success when prey availability is low (e.g. Chinstrap Penguins *Pygoscelis antarctica*; Croll *et al.* 2006). LBP are thought to act in the same way as chinstrap penguins as they must alternate parental duties on a regular basis, and therefore are constrained in their foraging distance.

During 2005/2006 the breeding season overlapped with the moulting period which meant that the LBP were unable to forage to provision chicks. Furthermore the overlap meant that there was no chance of a recuperation period to increase the fat stores (body condition) required to survive the demands of fasting and reduced thermoregulation during moult. This explained the large number of LBP that died during 2006.

The key selective pressures for laying early are unknown but are likely to be associated with adult age and experience; something that requires individual identification and long-term monitoring to determine. The current study is unique to the North Island sub-species of LBP as the approach used has covered short and long-term analysis of the breeding ecology, feeding ecology, and overall survival. This provides sound baseline data of the Tiri population for future research of both within and between population comparisons. Basic knowledge of this sort is the building block for more focused studies that can target particular parameters (e.g. competition, prey abundances, species interactions); something lacking from many species-specific studies.

Marine ecosystems have been transformed in recent times by pollution, removal of top predators, over-harvesting, and climate change (Jackson *et al.* 2001) and the results are the loss of prey species, changes in predator-prey interactions (Jackson 2001), and alterations of food webs (Essington *et al.* 2006). The impacts that these

transformations can have on other species is often not well documented but can be assessed by monitoring species that rely on the marine environment for their foraging requirements. To achieve this, the right questions need to be asked and an appropriate species targeted. If applied thoughtfully the LBP ecology may be utilised as a biological indicator of the marine environment. This could be done by measuring the direct overlap of fisheries catch takes with LBP foraging, or by simultaneously correlating many parameters associated with marine productivity (e.g. chlorophyll levels, SST, SOI) and relate long term changes to penguin diet (e.g. isotopes). The lack of evidence for the effect of SOI on the survival and breeding of LBP may be superficial and lends itself to further investigation. A study on Sooty Shearwater (*Puffinus griseus*) behaviour in Southern New Zealand is a good example on the use of seabirds as biological indicators. Changes in harvest rates of Sooty Shearwaters were significantly correlated to predict climatic perturbations. This was also suggested to also be associated with food availability, wind characteristics, direct and indirect fishery pressure and PCB/DDE levels (Lyver *et al.* 1999).

Unless long-term baseline data are created for populations of LBP the use of them as biological indicators will not be productive and will be prone to biases.

6.1.3 Future Recommendations

Future research recommendations include the use of individually banded birds for population monitoring by mark recapture analysis. Long-term monitoring using mark recapture data would provide information on recruitment, survival, dispersal, and fecundity; the basics for population demographics and modelling. Future monitoring of the Tiritiri Matangi population of LBP would be benefited through an understanding of the long-term parameters associated with their breeding success. Breeding success could

be associated with emigration or immigration to or from the population and hence age and pair-bond association, or it could be associated with indirect factors such as SST which may influence prey availability, which will in turn influence body condition and/or timing of LBP breeding. Regardless, simple monitoring of the variations in breeding success from year to year will ultimately provide a standard for the population in question which will aid within and between population comparisons.

Studying seabirds is often difficult since the at sea behaviours remain elusive and logistically difficult to quantify and so the direct effect that the marine environment can have on any population at any one time remains unknown. However, linking population demographics with more direct studies such as GPS tracking to identify the at sea behaviour of LBP and isotope analysis of diet would provide a sound basis for understanding LBP ecology.

The lack of information relating to the biology of prey species abundance and distribution makes it hard to draw strong conclusions as to the effect that this can have on the LBP. Therefore future investigations will be best targeted towards direct measures of predator-prey interactions. Investigation into the key parameters that are associated with prey availability, whether a result of natural or anthropogenic influences, will be important when using LBP as biological indicators.

Understanding the mass mortalities associated with LBP could begin with tying prey availability to that of beach wreck variation, SOI fluctuations, SST changes and chlorophyll levels since this appears to be commonly associated with global warming and seabird studies.

Finally, it is important to consider the extent to which restoration needs to be applied when targeting island ecosystems since the immediate surrounding environment also needs consideration.

6.1.4 Conclusions

The use of LBP as biological indicators may be problematic and should be applied with caution. Penguins have been used as oceanographers by carrying devices that measure their surrounding marine environment (Charrassin *et al.* 2002). This is different to using the ecological measurements to measure marine productivity over both the short and long term. Breeding success of LBP as an indicator could be a determinant of prey availability as long as other baseline information is known such as what is a high, low and intermediate level of breeding success. This also needs to be linked with exact prey types taken within the surrounding environment with direct monitoring of prey takes within the local foraging range to the LBP population of concern. This emphasises the need for long term monitoring and a more direct approach at linking potential parameters.

The best approach to utilising LBP as biological indicators is through monitoring the isotope levels within tissues since they are easily obtainable and non-destructive. Understanding of trophic levels is important to food web studies and using species as indicators since trophic levels identify predator-prey interactions. Coupled with stable-carbon levels marine productivity can be monitored over the long-term which may help assess problems faced in the future (e.g. global warming, Sandvik *et al.* 2005). Stable-isotope measures can be used to target different time scales and different measures of diet which appears to be the backbone of seabird life stages. There is a growing need to identify the links between the land and the sea especially when considering island restoration and species inhabiting them. This knowledge would be a great strength to New Zealand conservation and can be applied globally.

6.2 References – Chapter 1

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6.7 References – Chapter 6

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6.8 Permit

9 August, 2005

Our ref:

Assoc. Prof. Dianne Brunton
Conservation & Ecology
Institute of Natural Resources
Massey University, Albany
Building 5, Gate 4
Oteha Rohe Campus
Auckland

Dear Dianne,

I am writing to inform you that your request to extend your research programme on the Little Blue Penguin (*Eudyptula minor*) on Tiritiri Matangi Island and to add Jaqueline Geurts' name to the permit has been granted. The termination date for the research permit issued to you is now the 1st July 2008. As you are the principle researcher named on the permit it is your responsibility to ensure that Jaqueline complies with the conditions of the permit.

All terms and conditions of the original permit described above are still valid and must be adhered to in addition to the following:

1. The study must be carried out as carefully as possible but if significant adverse effects are observed the study must be stopped immediately and the Programme Manager Biodiversity, Islands in the Warkworth Area Office contacted immediately.
2. The permit holder must follow procedures that are advised by Department of Conservation Programme Managers, to prevent the introduction of disease, rodents, insect or weed species to the sites listed in Schedule 1. The Permittee will ensure all field equipment is washed clean and sterilised with anti viral solutions prior to entering the site. Equipment must also be sealed in containers so both the Permittee, DOC and the Motuora Restoration Society (MRS) can be certain it is free of rodents and invertebrates. Footwear and clothing must be free of mud and seeds.
3. The Permit Holder(s) must liaise with and follow advice given by the resident Tiritiri Ranger.
4. The Permittee must not impact on any other absolutely protected wildlife, or other research or management activities at a site.

Accommodation must be booked well in advance through Barbara Walter, the DOC ranger on Tiritiri Matangi (Contact Phone 09 476 0010)

Yours sincerely



Rolien Elliot
Area Manager
Warkworth Area Office
For Auckland Conservator

Warkworth Area Office
P.O. Box 474, S.I.I., Warkworth, Auckland, New Zealand
Telephone 09-425 7812, Fax 09-425 7813



Department of Conservation
Te Papa Atawhai

Research Permit

The Minister of Conservation (the Grantor) **GRANTS** the Permit Holder(s):

Paula Culling & Dianne Brunton

Of: School of Biological Sciences
The University of Auckland
Auckland
New Zealand

A Permit to conduct research specified in the Schedule and on the Site(s) specified in the Schedule for the purposes specified in the Schedule. This permit allows research by the Permit Holder(s) only on land administered by the Department of Conservation in the areas specified in the Schedule. The permit is subject to all the terms and conditions (including special conditions) set out in this Document.

1. The Permit Holder(s) shall pay a fee (GST inclusive) of nil in advance to the Grantor in the manner directed by the Grantor.
2. The Permit does not confer on the Permit Holder(s) any interest in the Site, nor does it derogate in any way from the rights of the public to use and enjoy the whole or any part of the Site.
3. The Permit Holder(s) shall indemnify the Grantor against all claims by any person in respect of any injury, loss or damage (including fire damage) caused by or arising out of any act or omission of the Permit Holder(s), its servants, agents, contractors, clients or invitees, or otherwise caused as a consequence of its use of the Site or as a result of the conduct of the research activity.
4. As required by Section 17W(7) of the Conservation Act 1987 the Permit Holder(s) shall act in accordance with every relevant Conservation Management Strategy and Conservation Management Plan for the time being in force, including any amendments to the Strategy or Plan, whether the Strategy or Plan or amendment was approved before, on or after the date on which the Permit became effective. Any breach or contravention by the Permit Holder(s) of any relevant Conservation Management Strategy or Conservation Management Plan, or both shall be deemed to be a breach of this Permit.
5. The Permit Holder(s) shall not transfer, sublet, assign or otherwise dispose of the rights granted by this Permit.
6. The Grantor may terminate this permit by notice in writing to the Permit Holder(s) if the Permit Holder(s) breaches any of the terms of this Document.

7. The Permit Holder(s) shall comply with all reasonable notices and directions of the Grantor concerning the activities conducted by the Permit Holder(s) on any land administered by the Grantor.
8. This Permit must be produced on demand to Department of Conservation staff and the person(s) collecting shall follow any additional conditions or instructions given by staff regarding the collection of material.
9. Within three (3) months of the date of termination of this permit the Permit Holder(s) shall report to the Conservancy Advisory Scientist, Auckland Conservancy details of research conducted within areas administered by the Department of Conservation. The Permit Holder(s) shall advise the location of study sites within the reserves and also report on any significant finds made.
10. A copy of any published papers, theses or unpublished report(s) arising from the research under this permit shall also be forwarded by the Permit Holder(s) to those specified in condition 9 above.
11. All archival photographs or documents relating to the study shall, if required, be made available to the Department of Conservation for inspection and/or copying on request.

SIGNED by





Rob McCallum
Auckland Conservator

On the 10 of July - 2002.

ACTING FOR AND ON BEHALF OF THE
MINISTER OF CONSERVATION

("the Grantor") pursuant to a written delegation
in the presence of:

Permit not valid until read and signed by
Permit Holder(s):

 DHBurton
 P.M. Culling

By signing this you the Permit Holder(s)
agree to the conditions of the Permit.