

CHAPTER FOUR

SILVER GULL DIET ANALYSIS

4.1 Introduction

There are many methods that can be utilised to understand the diet of a bird species. The previous chapter used the observational technique, whereby birds were observed feeding at SBT farms with the food source and amount consumed noted. Similar methods have been used successfully on aquaculture farms and fishing boats to quantify aquaculture stock loss and discard scavenging rates (Carss, 1993; Glahn *et al.*, 1999; Skov & Durinck, 2001; Barrett *et al.*, 2002; Leukona, 2002; Valeiras, 2003; Hodgens *et al.*, 2004; Svane, 2005; Werner *et al.*, 2005; Werner & Dorr, 2006). Although observational studies coupled with experiments are relatively non-invasive, reasonably easy to undertake and provide useful data, they are not 'ground truthed' and only include birds observed at these food sources, which may not represent the diet of the entire population and therefore have limitations. Whilst food source exploitation can be estimated by combining the data obtained in the field with that from the literature, the actual diet of a seabird population can only be understood with specific dietary analyses. Methods range from low to high stress activities and require more training, skill and knowledge than observations alone. These methods include the following: observations of mate or chick feeding from a hide, which although not invasive, requires long periods of observation for very small sample sizes and poor levels of food identification (Spaans, 1971; Moore *et al.*, 2000; Votier *et al.*, 2003; Taylor & Roe, 2004); pellet and prey collection which produces large sample sizes, but may underestimate soft bodied prey (Spaans 1971; Pierotti & Annett, 1991; Blaber *et al.*, 1995; Oro, 1996; Oro *et al.*, 1996; Watt *et al.*, 1997; Green *et al.*, 1998; Annett & Pierotti, 1999; Huppopp & Wurm, 2000; Johnson *et al.*,

2000; Votier *et al.*, 2003; Rome & Ellis, 2004; Schwemmer & Garthe, 2005; Votier *et al.*, 2007; Lindsay & Meathrel, 2008); faecal collection which can produce large sample sizes but which may poorly represent the diet (Spaans, 1971; Ford *et al.*, 1982), spontaneous regurgitations of chicks or adults which may produce large sample sizes, depending on the species, but samples may not represent the entire diet (Mudge & Fern, 1982; Smith *et al.*, 1991; Smith & Carlile, 1993; Annett & Pierotti, 1999; Cherel *et al.*, 2002; Pedrocchi *et al.*, 2002; Gonzalez-Solis, 2003; Votier *et al.*, 2003; Phillips, 2006), collection of blood for stable isotope analysis which although useful and accurate, requires time, skill and funding to perform the analyses (Yves *et al.*, 2005); administering emetics which can cause mortalities in birds and can take a reasonably long time to take effect (Ford *et al.*, 1982; Montague & Cullen, 1988), stomach flushing which can produce quick, accurate results, although it is very invasive (Wilson, 1984; Weimerskirch & Cherel, 1988; Green *et al.*, 1998; Votier *et al.*, 2003; Page, pers. comm.) to culling birds which can provide an accurate representation of the diet but can be unethical if performed during the breeding season as parent birds may have chicks that would starve to death (Ford *et al.*, 1982; Jahncke *et al.*, 2005; Werner *et al.*, 2005).

Although these methods can be used alone for dietary analysis, they do have their limitations, so they can be coupled with other methods to gain more accurate data. For example, many aquaculture stock loss studies and discard feed rate studies have coupled observations with gut analyses of culled birds (Werner *et al.*, 2005) or collection of pellets and regurgitations from nesting colonies to better understand the diet of each bird species (Blaber *et al.*, 1995; Huppopp & Wurm, 2000).

For the Port Lincoln Silver Gull population study, observations of feed loss at the tuna farms needed to be linked or 'ground truthed' with diet analyses of gulls in the area. Breeding gulls were chosen for this analysis as a large proportion of the population breeds during the tuna season, meaning a large sample size of gulls that would return to the colony each day was available for whatever dietary analysis method was chosen. Due to unpredictable weather, distance, time and financial constraints, we did not have the option of staying for long periods of time on the islands and so the methods used had to fit in with the collection of other data on the breeding colonies. Thus, the collection of pellets and prey and spontaneous regurgitations was coupled with stomach flushing to ensure a larger sample size to gain more accurate results.

Pellet Collection/Prey Remains

Pellets consist of indigestible components of prey that are covered in mucus and regurgitated by birds (Votier *et al.*, 2003). Prey remains consist of whole prey items or animal carcasses carried to the colony (Votier *et al.*, 2003). Pellets have been widely used in dietary studies of birds because they are convenient, cast in large numbers (usually once a day, depending on diet type), provide large sample sizes, can be collected with limited disturbance and are generally easy to classify into prey types (on the basis of non-vertebrae bone structure and otoliths) providing a relatively accurate method for assessing diet composition (Spaans 1971; Pierotti & Annett, 1991; Blaber *et al.*, 1995; Oro, 1996; Oro *et al.*, 1996; Watt *et al.*, 1997; Green *et al.*, 1998; Annett & Pierotti, 1999; Huppopp & Wurm, 2000; Johnson *et al.*, 2000; Votier *et al.*, 2003; Rome & Ellis, 2004; Schwemmer & Garthe, 2005; Votier *et al.*, 2007). Prey remains such as fish skeletons are also attractive as a dietary analysis technique because of the ease of collection and identification (Votier *et al.*,

2003). While some authors state that pellets and prey remains accurately and closely reflect the diet of seabirds (Spaans 1971; Pierotti & Annett, 1991; Oro *et al.*, 1996; Annett & Pierotti, 1999; Rome & Ellis, 2004), others suggest they may overestimate prey with identifiable hard parts and underestimate soft bodied prey, such as mackerel and herring that have easily digested otoliths and indistinct bone structure (Spaans, 1971; Votier *et al.*, 2003; Schwemmer & Garthe, 2005; Votier *et al.*, 2007; Lindsay & Meathrel, 2008). However, although they might not provide the best absolute measure of diet composition or the ratio of species within the diet, they can provide the best index of relative importance of prey types among time periods or localities (Spaans, 1971; Votier *et al.*, 2003).

Spontaneous Regurgitation of Adults and Chicks

The chicks and adults of some species of seabird spontaneously regurgitate as a defence mechanism and this occurs when they are handled for ringing and measuring (Mudge & Fern, 1982; Smith *et al.*, 1991; Smith & Carlile, 1993; Annett & Pierotti, 1999; Cherel *et al.*, 2002; Pedrocchi *et al.*, 2002; Gonzalez-Solis, 2003; Votier *et al.*, 2003; Phillips, 2006). Most of the regurgitated food is largely undigested and can be collected and used to analyse the diet of these birds (Mudge & Fern, 1982; Oro *et al.*, 1995). This method has been used for dietary analysis of Silver Gulls where spontaneous regurgitations have been obtained from chicks during handling for weighing and banding (Smith *et al.*, 1991; Smith and Carlile; 1993). Although relatively easy to collect, the number of samples collected can be small as some species are more likely to regurgitate than others (Wilson, 1984). In addition, if researchers handle the birds with great care and limit disturbance, they will not regurgitate as often (longer handling time often prompts regurgitation) (Votier *et al.*, 2003). For example, Votier *et al.*, (2003) handled 250 Great Skua chicks, but only

obtained nine samples and Phillips (2006) stated that regurgitates could only be obtained from 20% of gull chicks selected at random within a colony.

This method is non-invasive, very rapid and requires minimal training (although enough to ensure that the risk of plumage soiling is eliminated) and has no apparent long term deleterious effects on fledging, survival of chicks and reproductive output of adults (Votier *et al.*, 2003; Phillips, 2006). Its use on chicks alone can also eliminate the risk of chick and nest desertion, as may occur with handling adults.

Most food is largely undigested and can provide an indication of the food items collected by adults (Mudge & Fern, 1982), however, young chicks may be selectively fed small items, with larger chicks having a diet almost identical to adults which needs to be taken into account (Pedrocchi *et al.*, 2002; Votier *et al.*, 2003).

There is also a difference in samples obtained from birds with full stomachs and those such as older chicks or adults with empty stomachs. Full stomach samples are likely to be biased towards soft bodied prey as they are more easily regurgitated (Votier *et al.*, 2003) whilst those from empty stomachs could be under represented in fish and other soft-bodied prey that are digested relatively quickly with the sample likely to be coming from the previous meal (Phillips, 2006).

Stomach Flushing

Stomach flushing (water off-loading) involves pumping water into the stomach of a seabird, via a tube and syringe, until full and then encouraging the bird to regurgitate, in essence flushing the stomach contents from them (Wilson, 1984). Both adult birds and chicks can be stomach flushed and it provides good information on the types of prey consumed, but is invasive and time consuming (Wilson, 1984; Votier *et al.*, 2003). The method widely used now was devised by Wilson (1984) and requires only one person, although two are better (Page, pers. comm.). Wilson (1984) trialled

this method by flushing a group of penguins stomachs, feeding them a known quantity of food and then flushing them again. He found that 100% of the stomach contents were regurgitated. However, recovery rates are also dependent on meal size, ingestion time or prey type and may result in birds being sampled with empty stomachs (Votier *et al.*, 2003).

This method has been used on many seabird species including penguins, albatrosses, petrels, shearwaters and terns suggesting that the method can be used on a variety of sizes, and types of seabirds, thus obviating the need to destroy individuals to study their diet (Wilson, 1984; Weimerskirch & Cherel, 1988; Green *et al.*, 1998; Votier *et al.*, 2003). This method can also be calibrated in regards to recovery rates and can provide useful information on feeding frequency, meal sizes, as well as occurrence of prey species and can be performed systematically apparently providing a non-biased sample (Votier *et al.*, 2003).

Although stomach flushing obtains good results, a licence is required, it is time consuming, requiring considerable effort and it can cause stress to adults and chicks (Votier *et al.*, 2003). Using this method on parent birds of some species can result in desertion of the nest and chick, so in these cases chicks should be flushed instead (Weimerskirch & Cherel, 1988). However, on studies with Great Skuas, Votier *et al.*, (2003) found that stomach flushing chicks could result in them running from their territories and being predated by conspecifics. In contrast, studies on several species of penguin and the Westland Petrel have found no such desertion or long lasting effects on reproductive output (Wilson, 1984; Robertson *et al.*, 1994; Freeman, 1998; Phillips, 2006). This method is likely to be too stressful to use with sensitive or

endangered species at the risk of impacting on long term reproductive output (Votier *et al.*, 2003).

Analysis of Dietary Samples

Pellets, prey remains and stomach contents are identified to the lowest possible taxon and the number of each species obtained using jaws, otoliths, crustacean remains, cephalopod beaks, eye lens, shells and other identifiable hard parts through reference collections and identifying terrestrial items including garbage and other birds (Blaber *et al.*, 1995; Oro *et al.*, 1995; Huppopp & Wurm, 2000; Votier *et al.*, 2003). Remains can also be sorted into categories i.e. fish, human refuse, terrestrial vertebrates, crab and other marine prey (Smith *et al.*, 1991; Smith & Carlile, 1993; Rome & Ellis, 2004) and foraging habitat (Oro *et al.*, 1996). Length mass relationships of cephalopod beaks and otoliths can be used to estimate the size of the prey item (Blaber *et al.*, 1995).

The diet data can be coupled with reproductive output and population dynamics data to estimate the quantity of food required throughout the breeding season by the entire population (Johnson *et al.*, 2000). In addition, by using several methods that complement each other, the limitations and bias of each method can be decreased and thus a more accurate result of the diet can be obtained (Pierotti & Annett, 1991; Oro *et al.*, 1995; Oro *et al.*, 1996; Annett & Pierotti, 1999; Gonzalez-Solis, 2003; Rudstam *et al.*, 2004; Asseld *et al.*, 2006; Phillips, 2006). Diet analyses can also be coupled with other technologies such as time-depth recorders, radio telemetry, GPS and satellite tags to further understand foraging ecology and this can be useful when the diet of instrumented birds are required (Weimerskirch & Cherel, 1988; Green *et al.*, 1998; Wilson *et al.*, 2002; Catry *et al.*, 2004; Phillips, 2006; Page, pers. comm.).

The selection of these dietary analysis methods would allow the opportunistic collection of pellets and regurgitated prey from nest sites and spontaneous regurgitations from chicks during handling for weighing and banding. However, as spontaneous regurgitations may only produce a small sample size and pellets can bias towards prey with hard parts, stomach flushing was planned when time allowed complementing the other methods.

This chapter aims to understand the dietary requirements of the breeding Silver Gull population in Port Lincoln.

Aims

The primary aims of this chapter were:

- To compare the diet of the Port Lincoln Silver Gull population to those of reference populations with no access to SBT feed.
- To determine what proportion of the Port Lincoln Silver Gull diet consisted of SBT feed.

Hypotheses

H0: The diet of the nesting Port Lincoln gulls will not be different to that of the reference gulls.

HA: It is predicted that the diet of the nesting Port Lincoln Silver Gulls will be significantly different to that of the reference site gulls.

4.2 Methods

4.2.1 Study Area

Data on the diet of breeding Silver Gulls from the Port Lincoln area and from

reference sites were collected during the 2005 and 2006 breeding seasons as part of the investigation into the reproductive ecology of these populations. The breeding colonies used in this study were Rabbit and Sibsey Islands in the Port Lincoln area, and Lipson Island and Venus Bay Island C for the reference sites (Table 4.1).

Samples were only collected on Sibsey Island and Venus Bay Island C during 2005, as these islands were not used as study sites in 2006.

Refer to Chapter Two for further information on these islands.

Table 4.1: Number of diet samples collected each month for individual islands (including regurgitated pellets, spontaneous regurgitations of chicks, prey remains and stomach flush samples).

	Rabbit Island		Sibsey Island	Lipson Island		Venus Bay Island C
	2005	2006	2005	2005	2006	2005
April	-	1	-	-	-	-
May	-	1	1	2	2	-
June	-	2	3	4	5	-
July	-	5	8	10	16	-
August	1	25	2	4	-	-
September	-	-	-	2	20	-
October	-	-	-	-	-	-
November	-	-	-	-	-	7

4.2.2 Sample Collection

Data on the composition of the diet of these populations was gained by collecting regurgitated pellets and prey remains from around the breeding colony, both from nests that were used as part of the reproductive output analysis and randomly as they were encountered on the islands. The pellets consisted of undigested portions of food and were assumed to be from the adult birds. Pellets were placed into a plastic bag which was marked with island, nest number (if applicable) and date collected. Once back at the laboratory, the pellets were then placed in a -20°C freezer until they were

sorted and analysed.



Figure 4.1: An example of a regurgitated pellet in a Silver Gull nest.

Chick spontaneous regurgitations were placed into a plastic bag, labelled and frozen. Stomach flushing was found to be too time consuming and so only five adult Silver Gulls were stomach flushed on Rabbit Island (4) and Lipson Island (1) in late July 2006. Adult birds were caught on the nest using a nest trap and weighed, and then stomach flushed. This was undertaken by restraining the bird between the legs and holding the beak open with the first and second fingers of one hand. The other hand was used to place a 5mm catheter tube down the seabird's oesophagus until it reached the base of the stomach (Figure 4.2). Ambient temperature seawater/freshwater was then pumped into the stomach using a syringe, until it began to flow back out around the sides of the catheter (in this case 30ml of freshwater). The catheter was then removed, and the bird inverted over a bucket. The stomach was then gently massaged with a hand until regurgitation occurred (Wilson, 1984;

Votier *et al.*, 2003, Page, pers. comm.). Any excess food in the oesophagus was removed by gently massaging the neck (Wilson, 1984; Votier *et al.*, 2003, Page, pers. comm.). The stomach sample was then placed into a sealed container, labelled and frozen until sorted. The bird's stomach contents were then replaced with 30ml of a processed baitfish and vitamin mix via another tube and syringe. This method was only performed on the one day on each island, as it took over an hour to catch each bird. The parent gulls were reluctant to go back to the nest, presumably as it was nearing the end of their breeding season (the end of July), and parents that breed later are usually considered to be inexperienced birds which are not as fit parents compared to birds that breed earlier (Smith, 1995).



Figure 4.2: Stomach flushing a shearwater (Photo courtesy of Dr Brad Page, SARDI Aquatic Sciences).

4.2.3 Pellet and Prey Sorting

Pellets, regurgitates and prey items were taken out of the freezer and left to thaw for an hour. Each sample was weighed and then the pellets were placed into a petri dish

with water and left to soak for a few minutes. Tweezers were then used to flatten the pellet into the water. Once the pellet contents were teased apart, the petri dish was placed under a dissecting microscope. Otoliths found in the sample were counted, recorded and placed in a vial and left to dry. Other identifying hard parts such as squid beaks, eye lens, leatherjacket spines, cockle shells, fish jaw bones etc were counted, recorded and placed into a container of 75% ethanol. These were used to assess how many of each species were in the sample. Other aquatic items such as shells, fish bones, fish teeth and some jaws were identified if possible and recorded as a single specimen of that species, or of an unidentified type of prey (e.g. fish) as there was no way to accurately quantify individuals from these parts. The numbers of terrestrial items (grain, plants, insects, chop/chicken bones etc) were also counted and recorded.

Prey items, which were mainly fish backbones were identified to species level if possible. Whole fish skeletons or backbones with the skull intact were identified by removing the otoliths. The otoliths were stored in a separate dry container for future identification and the skeleton was placed into container of 75% ethanol. If no skull was attached, but the backbone was entire, it was recorded as one unidentified fish species. If the backbone was disarticulated, it was also recorded as one unidentified fish species as there was no way of knowing if the pieces were from the same or separate fish.

Chick regurgitations were weighed and then placed under a dissecting microscope to identify the contents. Stomach flushing samples were weighed and then filtered through a fine sieve to retain the hard items. The contents of the sieve were then washed into a petri dish and placed under a dissecting microscope for identification.

One stomach flush sample contained an entire fish in it which was identified, weighed and had the otoliths removed and placed into a vial to dry.



Figure 4.3: Sorting through pellet samples.

Otoliths, cephalopod beaks, leatherjacket spines and some jawbones were later identified down to species level by Dr Brad Page from SARDI Aquatic Sciences.

If some prey items could not be recognised, such as fish bones, eroded otoliths, fish backbones and crustacean exoskeleton, they were recorded as one specimen of either unidentified fish or unidentified crustacean.



Figure 4.4: Contents of a pellet sample 7x magnification (leatherjacket teeth, spine and lock. Spine = 14.02 mm, tooth = 3.7mm).

4.2.4 Data Analysis

The data obtained from all sampling techniques were combined for the data analysis. The Numerical Abundance and Frequency of Occurrence of each prey type was compared to test for overall differences in prey type between the two populations of gulls (Port Lincoln and reference sites) using similarity of percentage analyses, SIMPER, using the PRIMER statistical program (Primer-e, PML, Plymouth, UK) (Field *et al.*, 2007). The frequency of occurrence of a prey species is an expression of its presence, or the number of times a given species occurs in the diet of a population. In any given data set, FOO % of any prey taxon defines the proportion of samples in that data set that contained a minimum of one individual of that prey taxon. Numerical prey abundance describes, for each prey taxon identified in all the samples, the proportion of the total number of prey items that is made up by that prey taxon.

Ordination multi-dimensional scale plots (MDS) were created to assess the intra-specific variation in Silver Gull diet. The Bray and Curtis association measure was used for the analysis (Beals, 1984). The stress value (how distorted or scattered the data are after being confined to a limited number of vectors) gives an indication of how well the data were represented during ordination (Page *et al.*, 2005). The lower the stress value, the better the MDS representation of the data (with a value <0.10 considered a good representation of the data) (Page *et al.*, 2005).

4.3 Results

4.3.1 Port Lincoln Gulls vs Reference Gulls

Forty nine diet samples (from all methods) containing 22 different taxa were obtained and examined from the Port Lincoln gulls, and 72 diet samples containing 27 different taxa were obtained from the reference site gulls. Fish, in particular, leatherjackets, were the most abundant prey group for gulls at both sites, however, the reference site gulls had a larger number of fish species in their diet. Grain was very important for the reference gulls, but was also found in the diet of the Port Lincoln gulls. The numerical abundance (NA) and frequency of occurrence (FOO) of each prey type was analysed for both the Port Lincoln and reference site gulls and a SIMPER analysis used to test for differences in diet between the two sites (Table 4.2). A 2D multi-dimensional scale plot (MDS) was created to determine any variation or groupings within the diets.

Numerical Abundance

The diets of the Port Lincoln gulls and the reference site gulls were significantly different in terms of numerical abundance of prey types ($R = 0.143$, $p < 0.001$) with an average dissimilarity of 84.87%. The top nine prey species made up 85% of the

numerical abundance dissimilarity between the Port Lincoln and reference gulls. The remaining 15% was comprised of 20 prey types. The top nine prey types which together comprised 85% of the diet dissimilarity between the two sites included leatherjackets (29% diet dissimilarity) which were higher in the diet of Port Lincoln gulls, grain (19.6%), parrotfish (12%) and unidentified fish (8.6%) which were higher in diet of the reference site gulls, marine plants (3.4%, higher in Port Lincoln gulls), snails (3.3%, higher in reference gulls), insects (3.2%, higher in Port Lincoln gulls) wrasse/trumpeter (3.1%, higher in reference gulls) and unidentified crustaceans (3.02%), which were higher in the diet of Port Lincoln gulls (Table 4.2).

Frequency of Occurrence

The diets of the Port Lincoln gulls and the reference site gulls were also significantly different in terms of frequency of occurrence of prey types ($R = 0.165$, $p < 0.001$) and had an average dissimilarity of 72.9%. Nine prey types made up more than 69% of the frequency of occurrence dissimilarity between the Port Lincoln and reference gulls. The remaining 31% was made up of 20 prey types. The top nine prey types which together comprised 69% of the diet dissimilarity included leatherjackets (11.3% of the diet dissimilarity), with more diet samples in the reference group containing this prey taxon; unidentified fish (10.1%) and marine plants (10%) with more diet samples at the Port Lincoln site containing these prey taxon, parrotfish (9%, higher in reference gulls), terrestrial plants (8%), insects (6.3%) and grain (5.5%) which were higher in the diet of Port Lincoln gulls, snails (4.9%, higher in reference gulls) and unidentified crustacean (4.8%, higher in Port Lincoln gulls).

Table 4.2: Silver Gull diet analysis data for the Port Lincoln (n=49) and reference (n=72) sites. Data includes the sum of each prey type collected, numerical abundance (NA) and frequency of occurrence (FOO). “Unid” means unidentified.

Diet Analysis	Port Lincoln (n=49)			Reference sites (n=72)		
	Raw Data (sum)	NA (%)	FOO (%)	Raw Data (sum)	NA (%)	FOO (%)
Fish						
Leatherjacket	346	55.4	10.9	345	20.9	13.2
Parrotfish	23	3.7	3.6	181	10.9	13
Unid Fish	32	5.1	18.1	128	7.8	12.3
Wrasse/trumpeter	0	0	0	56	3.4	4.9
Red Mullet	1	0.2	0.7	42	2.5	3.3
Silverbelly	4	0.6	2.9	20	1.1	3.3
Flathead	5	0.8	2.9	13	0.8	2.3
Silver Bullseye	0	0	0	7	0.4	0.8
Tommy Ruff	0	0	0	3	0.2	0.5
Garfish	1	0.2	0.7	2	0.1	0.5
Common Bullseye	1	0.2	0.7	2	0.1	0.5
Sprat	0	0	0	1	0.1	0.3
Sardine	5	0.8	2.9	0	0	0
Cephalopod						
Cuttlefish	3	0.5	2.2	15	0.9	2.8
Octopus	2	0.3	1.4	6	0.4	1.3
Unidcephalopod	2	0.3	1.4	4	0.2	1
Ommastrephid squid	0	0	0	3	0.2	0.8
Calamary Squid	1	0.2	0.7	2	0.1	0.5
Other Marine						
Marine plant	25	4	18.1	50	3	12.7
Unidcrustacean	12	1.9	5.7	19	1.1	3.4
Cockle	0	0	0	26	1.6	2
Anemone	0	0	0	1	0.1	0.3
Terrestrial						
Grain	117	18.7	5.1	634	38.3	3.3
Snail	10	1.6	2.2	38	2.3	4.6
Terrestrial plant	12	1.9	8.7	29	1.8	7.4
Insect	12	1.9	6.5	25	1.5	4.8
Chop/chicken bones	7	1.1	1.4	1	0.1	0.3
Bird remains	2	0.3	1.4	1	0.1	0.3
Plastic/paper rubbish	2	0.3	1.4	0	0	0

Although there was a significant difference between the diet of the Port Lincoln and the reference site gulls, there was no clear grouping or separation of diet shown in

the MDS plot (Figure 4.5). Although, there were intra-specific groupings within each classification (Port Lincoln and reference site), which indicates variability in the diet of the same population.

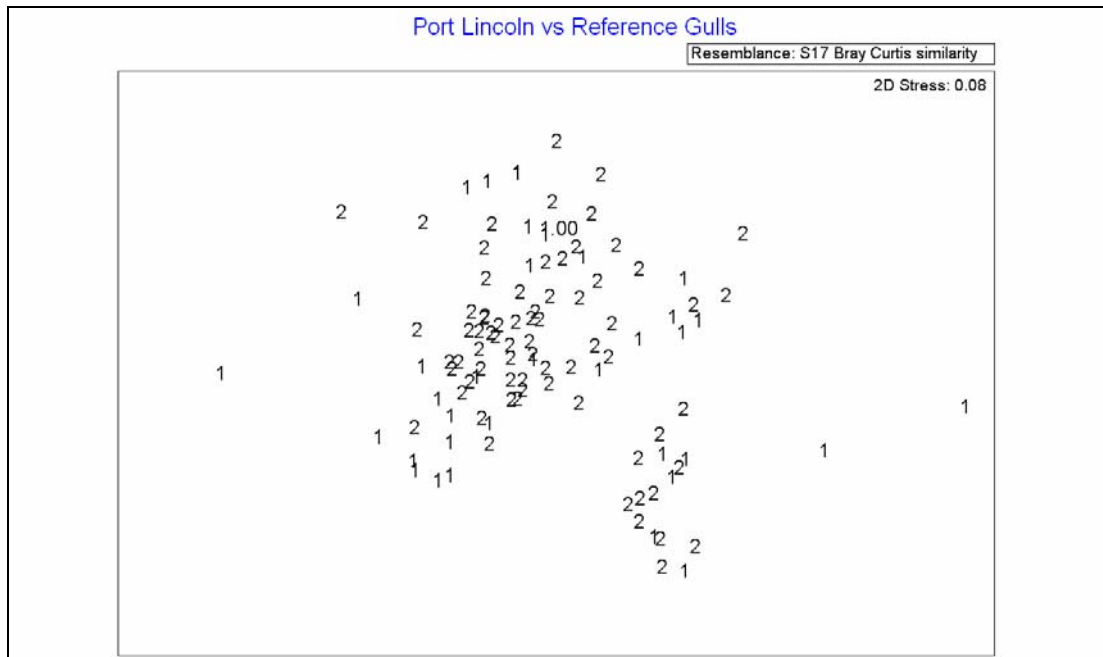


Figure 4.5: Intra-specific variation in the diet of the Silver Gull (Port Lincoln=1, reference site=2).

No sardine or human refuse was found in the diet of the reference gulls, however, these prey items were found in the diet of the Port Lincoln gulls (Table 4.3). There were also a total of seven chop/chicken bones found in the Port Lincoln gulls diet, with only one for the reference site gulls. This shows the true niche of the Port Lincoln Silver Gull population gulls as scavengers.

Ten of the 13 pelagic fish backbones found in the Port Lincoln samples did not have a head attached, so they were classified as unidentified fish, however, three very similar backbones did have heads attached and when otoliths were removed, they were identified as sardines. Thus it seems highly likely that the other ten backbones

were also sardines, but as they had no otoliths they could not be classified as such for the analysis. However, if it is assumed that these fish were sardines then 14 out of the 49 (i.e. 28%) of the diet samples collected from the Port Lincoln gulls had sardine in them.

Table 4.3: Numbers of items of interest in the diet of Silver Gulls. No. is the number of diet samples this prey item was found in.

Prey Items of Interest	Port Lincoln (n=49)		Reference (n=72)	
	No.	%	No.	%
Local Sardines	3	6.1	0	0
Pelagic Backbones	10	20.4	2	2.8
Imported Sardine	1	2	0	0
Plastic/Rubbish	2	4.1	0	0
Chop/chicken bones	7	14.3	1	1.4

4.3.2 Port Lincoln Gull Diet Comparison – during vs end of tuna season

The diet of the Port Lincoln gulls became more variable as the tuna season progressed, with fish, particularly leatherjackets, becoming more dominant. Whereas nearing the end of the tuna season, grain, unidentified fish, parrotfish and items such as human refuse, chop/chicken bones and bird remains which were not found in the diet during the season became more important (Table 4.4).

Numerical Abundance

The diet of the Port Lincoln gulls during the main tuna farming season and near the end of the season was significantly different in terms of numerical abundance of prey ($R = 0.079$, $p = 0.015$) and had an average dissimilarity of 85.9% (Table 4.4). The top nine prey species made up almost 88% of the numerical abundance dissimilarity between the two groups and consisted of leatherjackets (40%), which were higher during the season, grain (13.8%), unidentified fish (8.9%), marine plant (5.9%),

unidentified crustacean (4.6%) and parrotfish (4.1%) which were all higher nearing the end of the season, sardine (4%, higher during), terrestrial plants (3.6%) and chop and chicken bones (2.6%) which were both higher near the end of the season (Table 4.4).

Interestingly, the diet of the Port Lincoln gulls near the end of the season was also significantly different to the reference gulls ($R = 0.24$, $p = 0.001\%$) with an average dissimilarity of 85.4%. The top nine prey species made up almost 85% of the numerical abundance dissimilarity between the two groups and consisted of grain (24.1%), leatherjackets (21%), parrotfish (12.9%), unidentified fish (9.2%), marine plant (3.6%), snail (3.6%), insect (3.5%) and wrasse/trumpeter (3.2%) which were all higher in the reference site gulls diet than the Port Lincoln gulls at the end of the tuna season (Tables 4.2 and 4.4). In addition, no cockles or wrasse/trumpeter were found in the Port Lincoln gulls diet near the end of the season, but these were found in the reference gulls diet and there were more chop/chicken bones found in the diet of the Port Lincoln gulls near the end of the tuna farming season.

Frequency of Occurrence

The diet of the Port Lincoln gulls during and at the end of the tuna season was not significantly different in terms of frequency of occurrence of prey items ($R = 0.045$, $p = 0.098$) but had an average dissimilarity of 75.8% (Table 4.4).

Table 4.4: Numerical abundance and frequency of occurrence of each prey type in the Port Lincoln Silver Gull population’s diet during and at the end of the tuna season. “Unid” means unidentified.

Diet Analysis	During Tuna Season (n=21)		End of Tuna Season (n=28)	
	NA (%)	FOO (%)	NA (%)	FOO (%)
Fish				
Leatherjacket	84.4	15.9	10.2	6.8
Unid Fish	2.6	14.3	9	21
Parrotfish	1.3	1.6	7.8	5.4
Sardine	1	4.8	0.4	1.4
Flathead	1	4.8	0.4	1.4
Silverbelly	1	6.3	0	0
Garfish	0.3	1.6	0	0
Red Mullet	0	0	0.4	1.4
Common Bullseye	0	0	0.4	1.4
Cephalopod				
Cuttlefish	0.5	3.2	0.4	1.4
Octopus	0.3	1.6	0.4	1.4
Unidcephalopod	0.3	1.6	0.4	1.4
Calamary Squid	0	0	0.4	1.4
Other Marine				
Marine Plant	3.7	22.1	4.5	14.9
UnidCrustacean	0.8	4.8	3.7	5.4
Terrestrial				
Terrestrial Plant	1.3	22.1	2.9	9.5
Insect	1	6.3	3.3	6.8
Snail	0.5	3.2	3.3	1.4
Chop/chicken bone	0	0	2.9	2.7
Bird Remains	0	0	0.8	2.7
Grain	0	0	47.6	9.5
Rubbish	0	0	0.8	2.7

Although there was a significant difference between the diet of the Port Lincoln (during and nearing the end of the tuna season) and the reference site gulls (in terms of numerical abundance), there was no clear grouping or separation of diet shown in the MDS plot (Figure 4.6). Although, there were intra-specific groupings within each classification (Port Lincoln (during and end) and reference site)), which indicates variability in the diet of the same population.

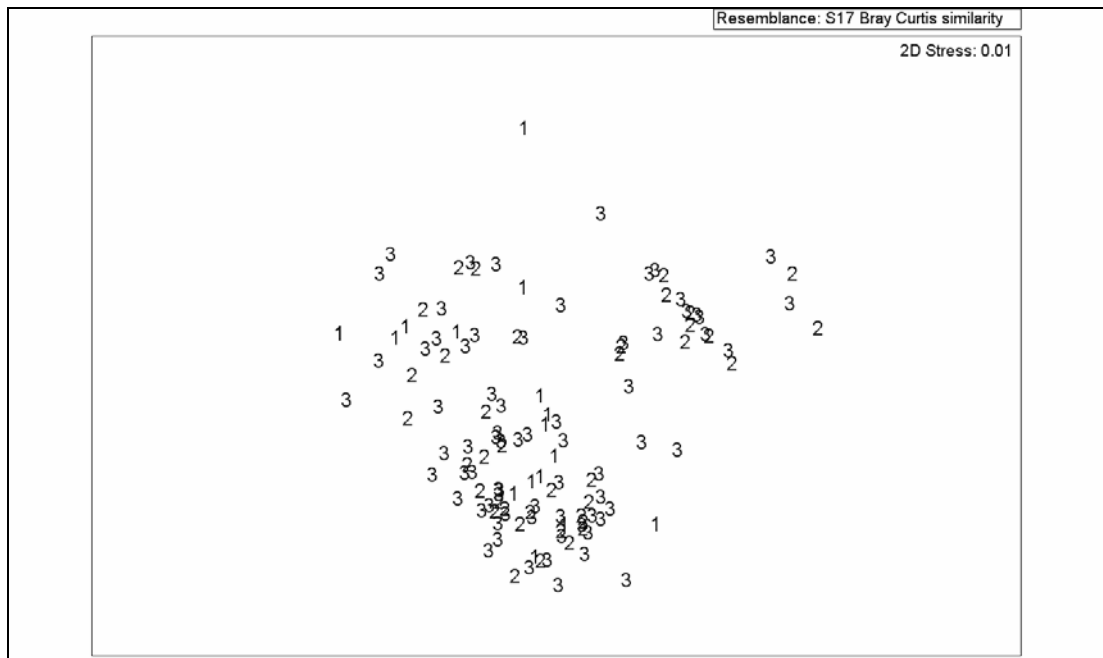


Figure 4.6: Intra-specific variation in the diet of Port Lincoln and reference site Silver Gulls (during the tuna season=1, nearing the end of the tuna season=2, reference site=3).

4.3.3 Stomach Flush Data Only

Of the five stomach flush samples collected, four were from Rabbit Island (Port Lincoln) and one was from Lipson Island (reference). One of the Rabbit Island samples contained an entire Californian Sardine (imported tuna feed). The three remaining samples were from birds that had relatively empty stomachs which only contained remnants of prey including fish bones (unidentified species but pelagic), fish scales, marine plants and unidentified crustaceans. The Lipson Island gull also had an almost empty stomach with only the jaws and teeth of a parrot fish and a terrestrial snail found within it.

4.3.4 Spontaneous Chick Regurgitation Data Only

Of the six spontaneous regurgitations obtained from chicks, two were from Sibsey Island (Port Lincoln) and four were from Lipson Island (reference site). The Sibsey

Island samples included unidentified fish flesh and bones, leatherjacket remains and an insect exoskeleton. The Lipson Island samples included unidentified fish flesh and bones, an unidentified cephalopod, a sea anemone, marine plant, insects, unidentified crustaceans and a terrestrial snail.

4.4 Discussion

A specific dietary analysis was undertaken on the breeding Port Lincoln Silver Gull population and this was compared to two reference Silver Gull populations. This dietary analysis was undertaken to ‘cross reference’ the results obtained from the observations on the SBT farms on the amount of SBT baitfish feed estimated to be consumed by Silver Gulls.

Four methods were used to collect the dietary data from these populations and these included pellet and prey collection, spontaneous regurgitations and stomach flushing analyses which provide a good overview of the diet (Pierotti & Annett, 1991; Oro *et al.*, 1995; Oro *et al.*, 1996; Annett & Pierotti, 1999; Gonzalez-Solis, 2003; Rudstam *et al.*, 2004; Asseld *et al.*, 2006; Phillips, 2006). However, it must be acknowledged that sample sizes in this study were very small (only 49 samples from the Port Lincoln gulls) due to the time consuming and opportunistic nature of the task. Even so, statistical analyses of the data were possible and this provided fairly clear and interesting results.

Reference site birds had a mix of mostly natural, marine and terrestrial prey. The Port Lincoln gulls also had a similar range of natural, marine and terrestrial items, but they also had some additional prey items present. There were a significant

proportion of pelagic backbones in the diet of the Port Lincoln gulls that were almost certainly from the tuna industry, which were not found at all in the reference gulls diet. The Port Lincoln gulls also had a larger proportion of human refuse in their diet than the reference gulls. This is not unlike other Silver Gull diet studies which have shown a range of prey types, both terrestrial and marine (Wood 1991; Smith *et al.*, 1991; Smith & Carlile, 1992; Higgins & Davies, 1996; Svane, 2005), but even though a large proportion of the diet may be made up of a certain feed type, for example 82% human refuse, 18% natural derivation for Big Island gulls (NSW) (Smith *et al.*, 1991), they are opportunistic feeders and will not generally rely 100% on one food type.

Interestingly, the diet of the Port Lincoln gulls became more variable as the tuna season progressed, with fish, particularly leatherjackets, becoming more dominant. Prey items that were not found in the diet during the first part of the season such as grain, unidentified fish, parrotfish and items such as human refuse, chop/chicken bones and bird remains became important prey items nearing the end of the tuna season. This may be as there is gradually less tuna feed over the course of the season as tuna are progressively harvested throughout the season. Therefore, there is more competition for tuna feed as the season progresses and these birds must find other feed sources to substitute their diet. It may also be influenced by the fact that younger, more inexperienced birds have been shown to breed later in the season (Smith & Carlile, 1992; Smith, 1995), and these gulls may have a more varied diet, as they have not yet refined their diet. This has been shown in Western Gulls (*Larus occidentalis*), where younger, inexperienced birds have been shown to have a diet higher in human refuse during the breeding season than the older more experienced

birds which switch to a diet of fish presumably as it provides a better source of nutrients to both the breeding birds and their eggs/chicks (Annett & Pierotti, 1989; 1999). Although the Port Lincoln gulls' diet became more variable nearing the end of the season, it was still significantly different to the reference gulls and therefore, at no time was the diet of the Port Lincoln gulls ever similar to that of the reference gulls.

As mentioned previously, the tuna farm feed loss observations estimate that seabirds consumed ~790 t of SBT baitfish feed per annum, with Silver Gulls consuming ~570 t, whilst the abundance data suggests 37,050 gulls were at the tuna farms each day, and they would consume ~534 t. However, the dietary analysis suggests that 28% of the Port Lincoln gulls diet contained tuna feed. If we assume that this means 28% of the population utilised this as a food source, we can calculate the amount of tuna feed that would be scavenged by these gulls per season. Using the largest number of gulls observed, which was 27,000 nests during 2005, this equates to a total of 87,750 birds (54,000 adults = (total nests*2) + 33,750 chicks (1.25 chicks*total nests)). 28% or 24,570 gulls would consume ~246 t of tuna feed per annum¹ (calculation explained in the Appendix). Conventionally, tuna are farmed or fed for a maximum of 180 days or six months, however, as some companies may catch fish as early as December, whilst others may catch in February, not all companies are feeding for the same period of time and hence tuna feed is available to birds from as early as January to possibly the end of September/start of October each year (270 days). However, as there are likely to be very few pontoons with fish in them during September, this month was not included in the calculations (although seabirds would still be

¹ Adult Silver Gulls consume 60g/day (Kotega, 1991), chicks ~12g/day (chick regurgitations average weight 2g (personal observation) and chicks fed every 90 minutes (Smith, 1995) *240 feeding days.

scavenging a small amount of food) and therefore we assume the tuna baitfish feed is readily available for 8 months or 240 days.

As sardines were likely to have been underestimated in the diet using this method, we can compare 28% of sardine in the diet (~246 tonnes) to 100% (~787 tonnes - See Appendix for calculations) and use this as a comparison to the proportion calculated using the feed loss estimation method on the farm, which was 570 tonnes. Therefore, the tuna farm observation suggest that ~72% (570 of 787 = 68% or 72%) of the breeding Silver Gull population utilised tuna feed as a food source and therefore 28% would have been comprised of items such as leatherjackets, grain and other marine species which were found in the diet analysis.

Although these data indicate that not all Silver Gulls eat sardines, it does show that at least some of the Port Lincoln population do. This may indicate that a few birds routinely target the tuna pens (as indicated by the MDS plots), rather than all birds all of the time, or all birds some of the time. To ascertain this, we purchased GPS trackers in 2006, in the hope that we could obtain enough data to calculate what proportion of the population utilised the tuna farms. However, as they were a new technology with technical glitches, they did not arrive until October 2006, which was too late to use them.

In conclusion, this dietary analysis has shown that the diet of the Port Lincoln gulls was significantly different to those of reference populations with no access to tuna feed. The data from the dietary analysis and the observation of scavenging on the tuna farms indicate that tuna feed represents 28-72% of the diet of this population.