Dietary Ecology of the Glaucous-winged Gull (Larus Glaucescens) in the Pacific North-West: Conventional and Stable Isotope Techniques and Implications for Eco-toxicology Monitoring

by
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B.Sc. (Marine Biology), University of British Columbia, 2007

Thesis Submitted In Partial Fulfillment of the Requirements for the Degree of Master of Science

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Abstract

Effective use of seabirds in ecotoxicology monitoring programs (e.g. Canada’s Chemical Management Plan) requires detailed knowledge of their ecology. I examined the dietary ecology of Glaucous-winged Gulls (*Larus glaucescens*) in British Columbia, using conventional diet analysis and stable isotope analysis. Conventional analysis suggests that gulls forage in an opportunistic manner, with a variety of prey types consumed at a colony closest to urban development, but that marine sources (fish, invertebrates) were the predominant dietary component at all colonies. However, variation in chick diet between 2009 and 2010 indicates that diet can vary considerably on a short time scale. Compared with historical records, gulls currently consume less food from anthropogenic sources and more fish in the Salish Sea, whereas at Cleland Island diet has remained marine-based over time. Stable isotope analysis confirmed that gulls at all three monitored colonies fed primarily on near-shore marine prey at a high trophic level.

**Keywords:** Glaucous-winged Gull; *Larus glaucescens*; dietary ecology; stable isotope analysis, conventional diet analysis; contaminant monitoring
I dedicate my thesis to my parents, who have always encouraged me to explore, dream, and discover; and who supported me even when chasing ‘seagulls’ for a Master’s degree was a confusing choice.
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1. General Introduction

Human activities have impacted marine ecosystems for centuries through actions such as over exploitation of fish stocks (Pauly et al. 1998, Jackson et al. 2001, Myers et al. 2003), modification of coastal habitats (Burke et al. 2001), dumping of wastes, and pollutant run-off from terrestrial activities into coastal waters (Elliott et al. 1992, Burke et al. 2001). Since marine ecosystems act as contaminant sinks, a large body of research has been devoted to characterizing contaminant levels and sources, and understanding contaminant flow through these ecosystems. Most concerning are volatile contaminant classes with lipophilic properties which persist in the environment, leading to global distribution and the bioaccumulation in marine food webs (Jones and De Voogt 1999, Tanabe 2004). For example, research has demonstrated the global distribution of legacy organochlorine chemicals (e.g. DDT, PCB, HCH) in marine mammals (Tanabe et al. 1994), and the bioaccumulation and persistence of PBDE flame retardants in both the North Sea and Canadian Arctic food webs (Boon et al. 2002, Kelly et al. 2008). These persistent organic pollutants (POPs) occur at greater concentrations in high trophic level species and/or in species with increased marine diet input (e.g. Jarman et al. 1996, Elliott 2005, Elliott et al. 2009, Hebert et al. 2009).

Seabirds often represent avian top predators in marine food webs and are frequently used as biomonitors of contaminants in the marine environment (Furness and Camphuysen 1997). In particular, seabird eggs have widely been used as a medium in contaminant monitoring programs because of their ease of collection, collecting eggs has minimal impact on the population, their eggs are rich in lipids and have lipophilic contaminant concentrations that closely correlate with those in the blood of laying females, and because the within-sample variation in contaminant concentrations in seabird eggs is low compared with other taxa of marine organisms (Gilbertson et al. 1987, Becker 1989, 2003, Furness and Camphuysen 1997). Although a variety of resource allocation strategies exist for egg production (capital-income continuum) (Meijer and Drent 1999), research has demonstrated that seabird eggs often reflect local
pollutant contamination as a result of laying females foraging close to the breeding colony in the days prior to egg-lay (Becker 1989), or in the case where species are non-migratory. However, in order to successfully utilize a seabird species as a toxicology biomonitor it is necessary to have detailed knowledge of that species’ ecology, behavior, and dietary composition (Pearce et al. 1989, Furness and Camphuysen 1997). Since the pathway of contaminant acquisition in birds occurs through dietary uptake, an adult female’s foraging habits and pre-breeding dietary composition will affect the classes and concentrations of contaminants deposited in eggs (e.g. Morrissey et al. 2004, Hebert et al. 2009).

Since 1974, Herring Gull (*Larus argentatus*) populations throughout the Laurentian Great Lakes have been monitored by Environment Canada, under the Great Lakes Herring Gull Monitoring Program (GLHGMP), in order to assess contaminants in the Great Lake’s ecosystem. The Herring Gull was selected as the primary monitoring species because it is a top predator in the Great Lakes food web, is the only piscivorous bird species present in large numbers year round, it breeds at accessible nesting colonies, and has a holoarctic distribution allowing for geographic comparisons (Hebert et al. 1999). Initially the GLHGMP was aimed at monitoring halogenated hydrocarbons in eggs as a cause of poor reproductive success in gulls; however, it became clear that Herring Gull eggs could also be used to monitor spatial and temporal trends in organochlorine and PCB contaminant levels (Norstrom et al. 1995, Hebert et al. 1999). Many studies have also used the extensive long-term GLHGMP data set to focus on how dietary ecology informs contaminant flow and uptake. For example, Weseloh et al. (1995) concluded that egg contaminant levels were reflective of dietary preferences, and that piscivorous birds, such as the Herring Gull, consistently had the highest levels of accumulated contaminants. Hebert et al. (1997, 2000) found that changes in food web structure in Lake Erie and Lake Ontario explained PCB fluctuations in Herring Gull eggs, with a higher fish consumption leading to elevated PCB concentrations. In both these cases diet explained the observed patterns in egg contaminant burdens.

In 2006 Environment Canada launched the Chemical Management Plan (CMP), under which the Environmental Monitoring and Surveillance Program was established to identify and track legacy and emerging POPs of concern at the national level in air, sediment, water, landfills, wastewater treatment, and biota (fish and avian wildlife).
(Environment Canada 2011). Due to the successes of the GLHGMP, gulls were selected as a national aquatic wildlife monitor. However, since no single gull species has a breeding distribution that spans Canada, colonies from four different gull species were selected, including some previously used in the GLHGMP. Three CMP-monitored colonies were chosen on the west coast of Canada, all occupied by Glaucomic-winged Gulls (*L. glaucescens*), as they are the sole Larid gull species breeding in British Columbia’s coastal waters (Vermeer and Devito 1987).

The Glaucomic-winged Gull resides on the west coast of North America, with a breeding range extending along the coast from Alaska to Washington State. Generally considered an inshore species, this gull is found nesting on smaller offshore islands (Hayward and Verbeek 2008). Most breeding adults arrive at the colony by late April, with nest initiation occurring in late April and May (Vermeer 1963, Hayward and Verbeek 2008). Clutches of 2-3 eggs are usually laid in British Columbia between May and early July (Vermeer 1963, B.C. Conservation Data Centre 2011). Incubation lasts approximately 26-29 days, with young departing the colony at approximately 57 days old (Vermeer 1963). Fledging success can be defined as survival of chicks past four weeks or 31 days, as the vast majority of chick mortality occurs prior to this time (Vermeer 1963, Reid 1987, 1988). Like populations in Northern Washington, Glaucomic-winged Gulls in the Canadian Pacific region are considered relatively non-migratory (Reid 1988, Hatch et al. 2011) (Elliott unpublished data).

Although Glaucomic-winged Gulls are preferentially piscivorous, they are opportunistic feeders whose diet has been found to reflect the food items that are abundant and accessible in their area (Vermeer 1982). Research conducted on the diet of BC’s breeding Glaucomic-winged Gull populations during the early 1970s and 1980s found that adult gulls in the Strait of Georgia fed mostly upon anthropogenic sources before their chicks hatched but mainly fed fish to their chicks. In contrast, on the west coast of Vancouver Island, both pre-hatch adults and chicks were almost exclusively marine (Henderson 1972, Ward 1973, Vermeer 1982). As these birds’ diet has not been examined since 1982, and given that availability of human refuse and forage fish has changed during this time (Hayward and Verbeek 2008, Therriault et al. 2009), it is important, in the context of recent developments in contaminant monitoring programs, to
re-examine and characterize the dietary ecology of Glaucous-winged Gulls throughout BC to gain a more detailed understanding of dietary variation.

This study was conducted during the 2009 and 2010 breeding seasons at the three CMP-monitored Glaucous-winged Gull colonies located on the Pacific coast of Canada. Mandarte Island and Mitlenatch Islands are located in the Georgia Strait region of the Salish Sea, with Cleland Island located off the west coast of Vancouver Island. Specifically, I sought to establish an updated baseline for dietary ecology at various breeding stages, in an attempt to determine whether the gulls’ emphasis is on marine, terrestrial, or anthropogenic dietary sources. In the following chapters I describe my research on the current dietary ecology of the Glaucous-winged Gull using two approaches to dietary analysis. In my first data chapter, Chapter 2, I examine dietary ecology using a more traditional approach to diet analysis. The direct sampling of pellets and regurgitations allowed for the identification of specific food items in recent meals from adult and chick stages during the breeding period. Comparisons with historical data collected 30 years ago allowed for the investigation of temporal variation in diet between sampling periods. In Chapter 3, I utilized the more recent technique of stable isotope analysis to investigate diet assimilated over a longer time frame (weeks) at all breeding stages. Here, isotopes are used as a proxy for diet, with emphasis on identifying foraging habitats and trophic positions. My concluding chapter synthesizes major findings from my two data chapters, addresses unresolved questions, and provides recommendations for future dietary research in light of interpreting contaminant-monitoring data.
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2. Spatial and temporal variation in the dietary ecology of the Glaucous-winged Gull (Larus glaucescens) in the Pacific Northwest, as inferred from conventional diet techniques

2.1. Introduction

Contaminants of most concern in the marine environment, such as persistent organic pollutants (POPs), do not readily degrade and are relatively volatile, leading to their global distribution (Jones and De Voogt 1999, Tanabe 2004). As lipophilic compounds, POPs accumulate in fatty tissues of organisms, and thus bioaccumulate and biomagnify in marine food chains (Jones and De Voogt 1999, Tanabe 2004), with particularly high concentrations in piscivorous and predatory seabirds (Gilbertson et al. 1987, Furness and Camphuysen 1997, Gochfeld and Burger 2001, Becker 2003). For this reason, seabirds are integral components of several monitoring programs, and provide data on trends, exposure pathways, and effects of persistent contaminants (e.g. Newton et al. 1990, Elliott et al. 1992, 2005, Bignert et al. 1998, Hebert et al. 1999a, Becker et al. 2001, Braune 2007, Verreault et al. 2010).

As part of a program to assess the state of environmental health of the Laurentian Great Lakes, Environment Canada established the Great Lakes Herring Gull Monitoring Program (GLHGMP) in the early 1970s, which has successfully utilized the Herring Gull (Larus argentatus) to track spatial and temporal trends of many POPs (e.g. Hebert et al. 1994, 1999b, Norstrom et al. 1995, Gauthier et al. 2008, 2009). Building on the GLHGMP, the Environment Canada Chemical Management Plan (CMP) expanded annual monitoring to the national level in 2006 (under the Environment Monitoring and Surveillance program), with the intention of tracking emerging contaminant trends across Canada (Gebbink et al. 2011, Chen et al. 2012). Since no single species of Larus gull has a breeding range that extends across Canada, four different gull species are
included in the monitoring program [Glaucous-winged (L. glaucescens), California (L. californicus), Ring-billed (L. delawarensis) and Herring Gulls]. As the sole Larid species breeding in British Columbia’s coastal waters (Vermeer and Devito 1987), the Glaucous-winged Gull is the only species utilized on the Pacific coast as part of the national contaminant-monitoring program.

The effective use of a seabird species as a contaminants biomonitor requires detailed knowledge of its ecology, diet composition and trophic level, and migratory behaviour (Butler et al. 1971, Furness and Camphuysen 1997, Becker 2003, Burger and Gochfeld 2004), without which toxicological monitoring data can be misconstrued. In particular, the use of omnivorous and opportunistic gull species requires a comprehensive understanding of their dietary variation. Whilst the biology of the Herring Gull at the Great Lakes colonies has been well researched (e.g. reproductive success: Teeple 1977; distribution: Moore 1976; energetics: Norstrom et al. 1986; foraging and diet: Fox et al. 1990; Ewins et al. 1994; Hebert et al. 1999; Hebert et al. 2008; distribution and abundance: Morris et al. 2003), a recent study by Gebbink et al. (2011) examining perfluorinated compound trends at all fifteen monitored colonies concluded that further knowledge of dietary structure and foraging ecology of the newly added gull species is required to properly interpret contaminant-monitoring data.

Studies of seabird diets have employed a range of sampling methods (Duffy and Jackson 1986, Shealer 2001, Barrett et al. 2007). Conventional methods involve direct sampling of diet through collections of regurgitations (undigested or partially digested food regurgitated prior to or during capture), pellets (indigestible prey items regurgitated after feeding), and feces, or by direct observations of feeding behaviour. Conventional methods are known to both over- and under-represent particular prey groups, and are reflective of recent meals rather than assimilated diet (Duffy and Jackson 1986, Brown and Ewins 1996, González-Solís et al. 1997, Barrett et al. 2007, Weiser and Powell 2011). Nonetheless, those methods have the advantage of identifying specific prey items and have been widely used in gull diet research to determine dietary composition (Vermeer 1982, Fox et al. 1990, Ewins et al. 1994, Kubetzki and Garthe 2003, Herrera et al. 2005, Ramos et al. 2009, Weiser and Powell 2010). More recently developed indirect techniques, such as stable isotope and fatty acid signature analysis, are frequently regarded as advantageous since sampling methods are often less invasive and allow for
time-integrated estimates of diet (Barrett et al. 2007). While stable isotope analysis has been used extensively to infer seabird trophic level (based on $\delta^{15}$N), as well as inform on sources of dietary items (e.g. marine vs terrestrial, based on $\delta^{13}$C, see chapter 3) (Hobson 1987, Hobson et al. 1994), prior knowledge of specific prey groups and dietary variation from direct sampling are also essential to a comprehensive interpretation of contaminants data.

The utility of the Glaucous-winged Gull as a marine contaminant monitoring species on the Pacific coast of Canada hinges on the extent to which it relies on a marine-based diet. However, there is a lack of recent and reliable data on foraging behaviour and dietary plasticity of this species, and it is important to re-examine the diet in order to interpret trends in toxicological monitoring data accurately. In this study we characterize the current feeding ecology of the Glaucous-winged Gull at two monitored breeding colonies on the Pacific Coast of Canada. One located in close proximity to urbanized areas, and the other occurring in a more remote location on the West coast of Vancouver Island. Using conventional diet methods, we sampled at different stages of the breeding season in order to: (1) elucidate intra-colonial dietary shifts over the course of the breeding season, (2) examine inter-colony spatial variation in diet, and (3) compare our findings with those of historic studies conducted 30-40 years prior (Henderson 1972, Ward 1973, Vermeer 1982) at both colonies, in order to investigate temporal variation.

2.2. Methods

2.2.1. Study Sites

Two Glaucous-winged Gull colonies were selected for sampling based on their use in current CMP toxicological monitoring, the existence of historical information on diet (Henderson 1972, Ward 1973, Vermeer 1982) and proximity to (or distance from) potential anthropogenic diet sources. Both colonies occur on small to medium sized, mostly treeless, offshore islands in close proximity to Vancouver Island (Vermeer and Devito 1987, Hayward and Verbeek 2008, B.C. Conservation Data Centre 2011).
We conducted the majority of our fieldwork at Mandarte Island (Georgia Strait, BC; 48.633°N, 123.283°W), currently the largest Glaucous-winged Gull colony in British Columbia (>1800 active nests in 2009) (Blight 2012). There, gulls nest predominantly in meadow areas with grass cover (Henderson 1972, Vermeer and Devito 1987). Mandarte Island was chosen for its relative ease of access and its proximity to urbanized areas and landfills (Vancouver 55km, Victoria 23km, Sidney 9km) where the gulls may acquire anthropogenic food sources. Cleland Island, BC (49.167°N, 126.083°W) is located off the West Coast of Vancouver Island, and had approximately 1400 active nests in 2010 (pers. comm. Peter Clarkson, Resource Conservation Supervisor, Pacific Rim National Park Reserve of Canada). On Cleland Island gulls are restricted to nesting on the bare rock margin encompassing the island (Henderson 1972, Vermeer and Devito 1987). The colony represents a more exposed (Henderson 1972), remote site where diet has historically consisted of marine sources (Henderson 1972, Ward 1973, Vermeer 1982). Designated as an Ecological reserve, Cleland Island provides sensitive habitat for several seabird and marine species; accordingly, collection trips were limited to minimize disturbance.

2.2.2. Sample Collection

To determine adult Glaucous-winged Gull diet prior to egg laying (during nest initiation and construction) and during incubation, nest areas of the colonies were surveyed for regurgitated pellets. Fresh pellets were collected from nest surroundings and stored individually in small Ziploc bags (see sample size summary in Table 2.1). To ensure that the pellets were reflective of diet during that sampling period, methods similar to Weiser and Powell (2010) were utilized: if pellets appeared old, bleached, or had fallen apart they were not collected. Adult gulls occasionally regurgitate a mass of food (sometimes partially digested) in reaction to disturbance. These regurgitations were opportunistically collected during trapping and handling for blood sampling (see chapter 3), and stored in Ziplocs with dry ice until they could be frozen and returned to the lab for processing (see Table 2.1 for sample sizes).

To characterize chick diet, regurgitated food samples were collected in July and August. Trips to colonies were timed to sample chicks at approximately 2 weeks (early chick diet) and 4 weeks of age (late chick diet, near fledgling), and were based on our
knowledge of mean lay or hatching dates for the colonies. Researchers captured chicks by hand (chicks are usually incapable of flight until 37-53 days of age; Vermeer 1963), with only one chick sampled every few meters to avoid pseudoreplication by multiple samples from chicks fed by the same parents (Vermeer 1963, B.C. Conservation Data Centre 2011). Chicks were weighed and developmental characteristics were noted to ensure they were approximately 2 or 4 weeks, with average chick weight-ranges taken from a previously-developed arithmetic growth curve (Vermeer 1963). During handling chicks often regurgitate spontaneously; however, it was occasionally necessary to gently stroke the throat to encourage regurgitation. Regurgitations were collected in individual Ziploc bags and kept on dry ice until they could be transported to the lab for analysis (see sample size summary in Table 2.1).

The existence of technique-dependent biases in conventional diet studies has been well documented. Research has demonstrated that pellet analysis is biased towards indigestible hard prey items (Duffy and Jackson 1986, Brown and Ewins 1996, González-Solís et al. 1997, Weiser and Powell 2011) and spontaneous regurgitations may not provide complete proventriculus contents (Duffy and Jackson 1986); however, the conventional sampling methods we employed are considered suitable for determining a current baseline of specific prey items (Karnovsky et al. 2012) and providing insight into broad dietary trends (Furness and Monaghan 1987).

2.2.3. Laboratory Analysis

Pellets and regurgitations were weighed, carefully broken apart, and separated. Prey items were identified to the lowest possible taxonomic level, using a dissecting microscope and local marine invertebrate and fish guides (Lamb and Edgell 1986, Kozloff 1987, Harbo 1999). Along with taxonomic ranking, samples were also scored for the presence/absence of items assigned to one of the following broad diet categories: human refuse, fish, marine invertebrate, terrestrial invertebrate, terrestrial animal, and plant matter. The frequency of occurrence (%) of each category was then calculated as a measure of dietary composition, indicating the percentage of total samples that contained that particular food category. Frequency of occurrence data allowed for direct comparison of adult pellet and chick regurgitation data, along with statistical comparison to historical results obtained by Vermeer (1982). As historical results obtained by
Henderson (1972) and Ward (1973) were calculated differently; these were only used for qualitative comparisons. Although often included in seabird dietary results, percent biomass (wet weight) and numerical abundance (percent number of items in a diet category out of the total number of diet items) were not calculated due to an advanced state of digestion in some regurgitation samples.

2.2.4. Data and Statistical Analysis

With each sample being scored for the presence/absence of the various dietary categories, diet becomes a multiple response categorical variable, since individual samples containing more than one type of diet category are represented more than once in a contingency table. Thus, the assumption of independence applied to Pearson’s chi-squared tests is violated. We therefore tested for differences in diet using a statistical method developed for multiple categorical choices (see Agresti and Liu 1999). Data were broken down into multiple Pearson’s chi-squared tests for each comparison (e.g. adult pre-lay vs. incubation stages, chick early vs. late stages within colonies, and our 2009 and 2010 data vs. historical data) using counts of presence/absence data (0 for absence, 1 for presence) and analysed using JMP (version 8.0.2). An adjusted p-value was then calculated for each individual test using the Bonferroni method to account for multiple comparisons:

\[ \hat{p}_i = \min(cP_i, 1) \]

where \( c \) is the number of food categories (or tests), and \( P_i \) is the p-value of the \( i \)th test. The null hypothesis \( (H_0) \) is rejected when \( \hat{p}_i \leq \alpha \) (\( \alpha = 0.05 \)). In order to facilitate comparisons between diets of adults and chicks, we pooled adult pre-laying and incubation breeding stages and early- and late chick stages; however, these were not analysed statistically since pellets and regurgitations represent different sample types.
2.3. Results

2.3.1. Intra-colony variation within adult and chick breeding stages

Mandarte Island

Adults – Diet of adults did not differ significantly between the pre-laying and incubation stages after Bonferroni correction (p > 0.0593 and ŕ > 0.3558 in all tests; Fig. 2.1). At the broad classification level, no significant difference was found in percentage occurrence of marine invertebrates among breeding stages. Pellets from incubating adults contained a greater percentage occurrence of errant polychaetes (47.1%) than pellets from pre-laying adults (19.7%), but the presence of other identifiable invertebrate taxa was similar (Table 2.2). Prey content of adult regurgitations from the incubation stage was generally consistent with the pellet data, with a slightly higher occurrence of fish (57.1%) and slightly lower occurrence of marine invertebrates (42.9%) and plant matter (14.3%) than pellets (Fig. 2.1).

Chicks – During both early and late chick-rearing stages in 2009 and 2010, chicks were fed mainly fish and plant matter, with a small fraction of regurgitations containing human refuse and marine invertebrates, and a few incidences of terrestrial invertebrates or other animal matter (Tables 2.2, 2.3, Fig. 2.1). In 2009, there were no significant differences in percentage occurrence of the main prey categories comparing early and late chick-rearing stages, with the exception of plant matter (82.6% early, 54.7% late; χ² = 9.347, d.f. = 1, p = 0.0022, ŕ = 0.0132). Similarly, in 2010 percent occurrence of most prey did not differ by chick stage with the exception of marine invertebrates (0.0% early, 14.3% late; χ² = 8.110, d.f. = 1, p = 0.0044, ŕ = 0.0264). Late-stage chicks at Mandarte Island were fed bivalve/gastropod molluscs (3.6%), crab/shrimp crustaceans (7.1%), and errant polychaetes (10.7%) whereas early stage chick samples did not contain marine invertebrates (Table 2.2). Occurrences of various fish species were similar between early- and late chick-rearing stages, within years (Table 2.2).
Cleland Island

Adults – No pellets were obtained for the incubation period at Cleland Island, precluding a comparison with the breeding stage. During the pre-laying period 100% of the pellets contained marine invertebrates, primarily Gooseneck barnacles (*Pollicipes polymerus*) (93.6%) (Table 2.2). Regurgitations were only obtained during the incubation period and with 100% occurrence of fish, almost exclusively Pacific Sand Lance (*Ammodytes hexapterus*) (Fig. 2.1).

Chicks – There were no significant differences in chick diet between the early and late stages of chick rearing (p > 0.0583 and \( \hat{p} > 0.1749 \) in all tests). Regurgitations from both chick stages contained a high occurrence of fish (early 100%; late 94.7%), with late stage chicks containing a greater occurrence of Pacific Herring (*Clupea pallasi*) (0.0% early vs 27.8% late) and similar occurrences of Pacific Sand Lance (50.0% for both). Late-stage chick regurgitations also contained a small percent occurrence of marine invertebrates compared with early-stage samples (Fig. 2.1).

2.3.2. **Intra-colony variation:**

**Comparison of adult and chick breeding stages**

Mandarte Island

Overall, adult pre-hatch diet (pre-laying and incubation stages combined) had a higher percentage occurrence of human refuse (+37.3%), digested terrestrial animal matter (+22.0%), and marine invertebrate items (+52.8%), and lower frequency of fish (-59.1%) and plant matter (-24.1%) than chicks (Table 2.3). Adult regurgitations collected during the pre-hatch adult stages also reflect this trend.

Cleland Island

Diet assessed from chick samples differed considerably from that in adult pre-laying samples (no pellet samples collected during adult incubation-stage), with chick samples having a higher percentage occurrence of fish species (+79.6%) and a lower percentage occurrence of marine invertebrates (-91.3%; Table 2.3). Chick regurgitations also contained more plant matter than adult pellets (+15.3%).
2.3.3. **Inter-colony spatial variation:**

*Comparison of Mandarte and Cleland Islands*

Adults – Diet in the Georgia Strait was much more variable than at Cleland Island. While marine prey commonly occurred (fish 37.2%, marine invertebrates 57.7%), human refuse (41.0%), terrestrial invertebrate (3.8%) and animal matter (24.4%) were also present in pellets. At Cleland Island diet was almost exclusively marine (fish or marine invertebrates, depending on breeding stage), with a smaller occurrence of plant matter (West coast; Fig. 2.1, Table 2.3). No incidences of human refuse were found in diet samples or observed at the colony; offal (fish scraps from fishing vessels and fish packing plants) appears to be the only possibility of human influence in diet.

Chicks – Only chick samples from 2010 were used for comparison since no samples were collected on Cleland Island in 2009. Overall, percentage occurrence of different prey categories was similar between the two colonies (in contrast to adult diets), with chicks being fed primarily fish and plant matter (Fig. 2.1). At Cleland Island, percentage occurrence of plant matter was lower than at Mandarte (-65.8%), and there was no occurrence of human refuse, terrestrial insect or animal matter items. Both colonies had comparable percentage occurrences of marine invertebrates (Table 2.3). Although fish occurrence was similar between colonies, the occurrence of specific fish taxa did differ: chicks at Mandarte Island consumed less Herring (-20.2%) but similar amounts of Pacific Sand Lance as those at Cleland Island.

2.3.4. **Historical changes in diet**

To compare 2009 and 2010 diet data with historical results from Vermeer (1982), pooled data were again utilized. Data presented by Vermeer did not lend themselves to statistical comparison. Although Vermeer’s (1982) Georgia Strait and West coast locations consisted of several pooled colonies, Vermeer’s (1982) pooled data due to a low number of intra-regional differences in broad diet categories, and therefore allowed us to make spatial comparisons.
Georgia Strait/Salish Sea

Adults – There was a significantly lower percentage occurrence of human refuse in pellets in 2010 compared with 1980 (-28.3%; $\chi^2 = 18.172$, d.f = 1, $p < 0.0001$, $\tilde{p} = 0.0006$), and a higher percentage occurrence of fish (+20.4%; $\chi^2 = 12.807$, d.f = 1, $p = 0.0003$, $\tilde{p} = 0.0018$; Table 2.3). The format of Vermeer’s (1982) data does not allow for a detailed comparison of marine invertebrate occurrence; however, the presence of bivalve/gastropod molluscs and sea stars in the adult diet appears equivalent between studies; whilst the presence of crab/shrimp crustaceans and errant polychaetes was increased in 2010 compared to 1980 (+20.0% and +25.6% respectively). The occurrence of digested animal matter in the adult diet was also higher in 2010 than in 1980 (+23.8%; $\chi^2 = 42.881$, d.f = 1, $p < 0.0001$, $\tilde{p} = 0.0006$; Table 2.3).

Chicks – The chief component of chick diet remains fish; however, our sampling demonstrates an increase in the prevalence of this diet category (+24.0% 2009 and +35.8% 2010), with fish significantly higher in 2010 samples than 1980 and 2009 ($\chi^2 = 14.134$, d.f = 1, $p = 0.0002$, $\tilde{p} = 0.0012$; and; $\chi^2 = 7.005$, d.f = 1, $p = 0.0081$, $\tilde{p} = 0.0486$; Table 2.3). Similarly, chicks in both 2009 and 2010 consumed less human refuse than 1980 but only chicks in 2010 were fed significantly less than 1980 and 2009 ($\chi^2 = 14.630$, d.f = 1, $p < 0.0001$, $\tilde{p} = 0.0006$; and $\chi^2 = 7.785$, d.f = 1, $p = 0.0053$, $\tilde{p} = 0.0318$).

As a broad diet category, marine invertebrates cannot be compared, as Vermeer did not calculate this. However, examination of more specific invertebrate taxa shows that a higher frequency of bivalves was consumed in 1980 than 2009 and 2010 (Table 2.3). Plant matter was also significantly more common in recent years than 1980 (+57.6% 2009; $\chi^2 = 126.336$, d.f = 1, $p < 0.0001$, $\tilde{p} = 0.006$; and +71.7% 2010; $\chi^2 = 158.224$, d.f = 1, $p < 0.0001$, $\tilde{p} = 0.0006$). Data suggests that the types of fish fed to chicks have shifted from a Herring-dominant diet in 1980 (-45.9% 2009 and -53.1% 2010) to more of a Pacific Sand Lance-dominated diet in 2009 and 2010 (Table 2.4).

West Coast

Adults – The presence of fish in the diet of adults on the West coast of Vancouver Island in 2010 significantly declined in comparison to 1980 ($\chi^2 = 8.100$, d.f = 1, $p = 0.0044$, $\tilde{p} = 0.0264$). As with the Georgia Strait discussion, direct comparison of a
broad marine invertebrates category is not possible; however, a breakdown of invertebrates by taxa shows similar patterns of consumption (Table 2.3).

Chicks – No significant differences were found in percentage occurrence of the main diet categories between data collected by Vermeer and our sampling efforts ($p = 0.3026$ and $\bar{p} = 1.0$ in all cases), with the exception of plant matter ($\chi^2 = 24.008$, d.f. = 1, $p < 0.0001$, $\bar{p} = 0.0006$; Table 2.3). Fish occurred at similar frequencies in 1980 and 2010; however, when regurgitations containing fish are broken down by taxa, it appears that a higher frequency of Pacific Sand Lance was consumed in 2010 and a lower frequency of Pacific Saury (Table 2.4).

2.4. Discussion

In this study we examined the spatio-temporal variation in the diet of the Glaucous-winged Gull, in order to assess its utility as a marine contaminant monitoring species on the Pacific coast of Canada. Our results show that Glaucous-winged Gulls forage in an opportunistic manner, based on the wide variety of prey types consumed at the colony in closest proximity to urban development (Mandarte Island). Nevertheless, marine sources (fish, invertebrates) form a prominent dietary component at both our study colonies, Mandarte and Cleland Islands. We documented several sources of variation in diet which might need to be considered in interpreting contaminants monitoring data: a) within-colonies, and within-years, there was little short-term temporal variation in diet either in adults (comparing pre-lay and incubation diets), or in chicks (comparing early versus late chick-rearing stages); b) in contrast, adult pre-hatch diet and chick diet within colonies were very different: chick diets at both colonies comprised mainly fish (>80%), whereas adult diets comprised <40% fish; c) adult diets at Cleland were predominantly marine, with 100% occurrence of marine invertebrates, but were much more diverse, and more terrestrial, at Mandarte, with significant contributions of human refuse and digested terrestrial animal matter; and d) differences from historical dietary studies were also present at both colonies, with our adult and chick samples at Mandarte Island exhibiting a higher percent occurrence of fish and lower occurrence of human refuse than 1980 (Vermeer 1982), and adult diet samples at Cleland Island containing less occurrence of fish than 1980 (Vermeer 1982).
We found only minor short-term temporal variation in diet, both in adults (comparing pre-laying and incubation diets) and in chicks (comparing early versus late chick-rearing stages). There was a significantly higher occurrence of plant matter in the diet of early stage chicks (compared with late stage) at Mandarte Island in 2009, although this was not found in 2010. We speculate that the occurrence of plant matter in chick diets is largely a by-product of provisioning, likely acquired by chicks when adults drop crop contents on the ground for chicks to feed upon, but may also be a sampling artifact. A significantly higher occurrence of marine invertebrates in chick diet was also noted at Mandarte Island in the 2010 breeding season, but not 2009, and may relate to the tidal sequences during the two chick stages. Irons et al. (1986) demonstrated that foraging effort of Alaskan Glaucous-winged Gulls in rocky intertidal areas was greatest at maximum low tide. However, marine invertebrates are known to exhibit a diel vertical migration, becoming less accessible to foraging predators during peak daylight hours. While both chick stages had similar low tide levels in 2010, the lows occurred during hours of lower light intensity (dawn to early morning) during the late chick stage, making marine invertebrates more accessible to foraging gulls than in the early chick period.

Our results are consistent with those of Ramos et al. (2009) who found only minor variation in chick diet composition with age in Yellow-legged Gulls (*L. michahellis*) breeding on the western Mediterranean coast of Spain. However, our results are contrary to those of Nogales et al. (1995), who reported that Herring Gull chick diet in south-west Scotland varies considerably between chick age classes, with a significant decrease in fish and increase in human refuse (chiefly poultry and pork) with age. However, both aforementioned studies were conducted in European ecosystems where human influence has significantly altered food web dynamics, and fish were acquired as offal from commercial fishery vessels rather than directly foraged from the marine environment. Variation in diet with chick age was also observed at Mandarte Island in the early 1970s, with a decrease in occurrence of fish and an increase in garbage and intertidal prey as chicks aged (Henderson 1972). Since the timing of refuse increase coincided with peak chick hatch, Henderson (1972) postulated that the demand for forage fish peaked, depleting available Herring and forcing gulls to make opportunistic use of refuse sources. In our study, a slight, non-significant, increase in human refuse
and a decrease in fish were observed between the two chick stages in 2009, while no similar shift was observed in 2010.

There was a major difference between adult pre-hatch diet and chick diet at both colonies in 2010, with chick diets having a much higher occurrence of fish (>90% occurrence) compared with pre-hatch adults diets (<40% occurrence). At Mandarte Island pre-hatch adult gulls consumed a mixture of marine invertebrates, human refuse, and fish but provisioned chicks predominantly with fish. Some researchers caution against the use of separate sampling techniques (pellets vs. regurgitations) when inferring dietary differences (Brown and Ewins 1996), due to aforementioned pellet biases, while others concluded that pellets accurately predict differences in diet composition (Spaans 1971, Annett and Pierotti 1989). We acknowledge that conclusions on dietary differences should be tentative, as suggested by Ewins et al. (1994); however, the comparison of pre-hatch adult and chick diet still conveys important baseline information. While the disparity in diet composition between pre-hatch adults and provisioned chicks may be reflective of a seasonal change in prey availability, work by Annett and Pierotti (Pierotti and Annett 1987, Annett and Pierotti 1989) on both Western (L. occidentalis) and Herring Gulls concluded that a switch between pre-hatch and provisioning adult diet was likely triggered by nutritional requirements of their nestlings rather than seasonal increases in fish abundance. Due to our sample design we are not able to make inferences about provisioning adult diet during the chick rearing stages, particularly since some seabird studies have revealed that provisioning adults may feed themselves different prey from their nestlings. For example, Hodum and Hobson (2000) revealed trophic segregation between adult and chick diet during the chick-rearing stage in Antarctic Fulmarine petrel species, and Davies et al. (2009) also found differences in Canadian Pacific Alcid species (with the exception of Tufted puffins).

At the more pristine Cleland Island colony diet also differed between pre-hatch adults and provisioned chicks in 2010, adults primarily consumed marine invertebrates while chicks were fed almost an exclusively fish-based diet (although a lack of samples from incubating adults leaves the pre-hatch picture incomplete). Similar to our findings, Henderson (1972) documented the consumption of Goose-neck barnacles (Pollicipes polymerus) during courtship feedings in the adult pre-lay period at Cleland Island; however, a switch to a diet completely dominated by Pacific Sand Lance was noted in
incubating adults. Adult Kelp and Herring Gulls have been found to switch their diet from marine invertebrate-dominated during pre-lay and incubation stages to fish after nestlings hatch (Spaans 1971, Pierotti and Annett 1987, Bertellotti and Yorio 1999). While this seems likely in our study, we did not sample provisioning adult diet and therefore cannot make a direct comparison. The consumption of marine invertebrates, particularly molluscs, prior to egg-lay is thought to provide gulls with the calcium carbonate and manganese resources necessary to egg formation. Gulls that supplement diet with marine invertebrates are thought to deplete their nutrient stores less, exhibit a more rapid recovery from egg-lay stress and, have an increased hatching/fledging success (Pierotti and Annett 1987). Pierotti and Annett (1991) have further demonstrated that pre-lay Herring Gulls that specialized on intertidal prey laid heavier eggs and exhibited an increased hatching success when compared to other diets.

Our study colonies were chosen for monitoring because they represent ecologically distinct sites, owing to differences in site characteristics (e.g. contaminant exposure), geographic location, and proximity to urban development, and were expected to exhibit relatively different baseline diets. As predicted, adult pre-hatch diet differed markedly between the colonies, with adults at Mandarte Island incorporating both natural prey and human refuse and Cleland adults foraging almost exclusively on marine prey. As with Glaucous-winged Gulls in other studies (Vermeer 1992), and findings in the closely-related Herring Gull (Fox et al. 1990), adults at both colonies foraged in an opportunistic manner, with diet reflecting locally abundant items and the association of colonies with urban areas. In contrast, few spatial differences were observed in the occurrence of broad diet categories in chicks, aside from the low occurrence of human refuse at Mandarte Island being absent at Cleland. The importance of forage fish during the chick-rearing stage is clearly exhibited and, given what is known about diet switching in gulls, appears to be a preferential choice in breeding Glaucous-winged Gulls.

Contrary to our predictions, the prevalence of human refuse in the diet of Mandarte Island Glaucous-winged Gulls is lower than results from historical studies conducted 30-40 years ago (Henderson 1972, Ward 1973, Vermeer 1982). Although our data is based only on a single year for comparison, results among historical studies for consumption of human refuse were consistent over a ten-year period, indicating that a change in pattern between 1980 and 2010 is plausible. In the early 1980s human refuse
predominated in the adult diet in the Georgia strait (Vermeer 1982), whereas our results suggest that adults (pooled pre-lay and incubation stages) are currently consuming a higher percentage of fish and significantly less refuse (Table 2.3). After Vermeer’s 1982 study, Pacific Herring total abundance rose to peak levels in 2003 and subsequently declined, leaving Herring less available to foraging gulls at the time of our study (Therriault et al. 2009). While no clear explanation exists for our result, a possible contributing factor is the decline of the Georgia Strait Glaucous-winged Gull population in the last 30 years (Blight 2012), easing the foraging pressure on available marine prey. Another important factor to consider is the abundance of other forage fish species. Pacific Sand Lance is a key forage fish for many seabird species; however, very little is known about its abundance and distribution in the Strait of Georgia (Therriault et al. 2009). Data from chick diet indicates that while the occurrence of Herring has decreased compared with 1980, Sand Lance subsequently increased, possibly compensating.

Both our study and historical studies (Henderson 1972, Ward 1973, Vermeer 1982) found that chicks at Mandarte Island were provisioned primarily with fish. In our study, chick diets also contained a lower occurrence of human refuse than reported by Vermeer (1982); however, considerable inter-annual variation exists between our sampling years (2009 and 2010), possibly indicating short-term fluctuation in forage fish prey availability due to climatic variation (e.g., El Niño-Southern Oscillation). The gulls at Mandarte Island demonstrated the ability to switch prey type during times of lower marine prey abundance, although this appears to affect the breeding success of the population. There was significantly lower fledging success in 2009 (LK Blight unpublished data), a year with lower prevalence of fish prey and elevated anthropogenic input than 2010 data. When comparing reproductive success between Cleland and Mandarte Islands, Ward (1973) determined that fledging success was higher at Cleland, where chicks were provisioned with more natural marine prey (primarily Pacific Sand Lance and Herring) and no human refuse. An increased breeding success with nestling fish consumption has also been previously reported in Glaucous-winged Gulls (Murphy et al. 1984) in Alaska, and Herring Gulls in Newfoundland (Pierotti and Annett 1987) and the Great Lakes region (Fox et al. 1990). Annett and Pierotti (1989) also concluded that early chick survival in Californian Western Gulls is not increased by the consumption of human refuse.
On the west coast of Vancouver Island, adults in our study appear to have consumed less fish than reported by Vermeer (1982) for west coast colonies; however, an absence of adult samples from the incubation period renders this comparison incomplete. Results from chick stages do not indicate any temporal dietary differences when compared with Vermeer (1982), with the exception of plant matter, thought to be an artifact of sampling. The occurrence of fish, as a broad category, remains equivalent between years, although late-stage chicks in 2010 were provisioned with a higher percentage of Pacific Sand Lance and a lower percentage of Pacific Saury (*Cololabis saira*) than 1982 (Vermeer 1982). Overall, when compared with dietary findings for Cleland in the early 70s and west coast colonies in the early 80s, diet on Cleland Island appears to have remained unaffected by human influences, such as urbanization and commercial fishing pressures, in the last 30 to 40 years. This reflects conclusions from Chen et al. (2012), who found a neighbouring colony, Florencia Island, to be the closest to a pristine CMP site in Canada, after showing it to have the lowest levels of flame retardant compounds in Canada.

Despite the smaller scale nature of this study, our assessment of Glaucous-winged Gull diet confirms that they are generalists, exhibiting a preference for natural marine prey types but opportunistically supplementing their diets with anthropogenic and terrestrial items when necessary. Also, availability of preferred marine prey may fluctuate at various temporal scales as a result of anthropogenic disturbances (e.g. fisheries) or of climatic variation (e.g. El Niño-Southern Oscillation). Since trophic level and proportion of marine prey significantly influence the concentrations of persistent organic pollutants accumulating in avian species (Jarman et al. 1996, Hebert et al. 2000, Elliott et al. 2009), knowledge of female Glaucous-winged Gull dietary ecology prior to and during egg production are critical to interpreting the kinetics of contaminant deposition in eggs. Glaucous-winged gulls breeding in the Canadian Pacific region are thought to be local migrants, overwintering locally and arriving at the breeding colonies a month in advance of egg laying (Vermeer 1963, Hatch et al. 2011) (Elliott unpublished data). Therefore, resources allocated to egg production should reflect adult female diet and contaminants acquired within the region. Our results support the utilization of the Glaucous-winged Gull egg as a medium for monitoring marine contaminants on the west coast of Vancouver Island since adult pre-lay diet consisted almost exclusively of marine sources.
(mixture of forage fish and marine invertebrates). However, some caution should be extended to the interpretation of egg contaminant results from colonies in close proximity to urban locations, as adult gulls foraged on a mix of dietary sources prior to egg production, and relied more heavily on anthropogenic sources in some years. Later studies may extend their parameters to include investigations into contaminant effects on nestlings, therefore, our data on chick diet and exposure are important for overall assessment. We recommend any further monitoring be paired with stable isotope analysis, to incorporate an assimilated dietary signature and reflect year-to-year baseline fluctuations. Results from this work, along with stable isotope data (see chapter 3), will be applied to the interpretation of regional contaminant-monitoring data collected under Environment Canada’s CMP Environmental Monitoring and Surveillance Program.
References


Henderson, B. A. 1972. The control and organization of parental feeding and its relationships to the food supply for the glaucous-winged gull, Larus glaucescens. MSc, University of British Columbia, Vancouver, BC.


Tables

Table 2.1  Sample sizes collected during different breeding stages at Glaucous-winged Gull colonies. P for pellet and R for regurgitation

<table>
<thead>
<tr>
<th>Colony</th>
<th>2009</th>
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<th>2010</th>
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<tbody>
<tr>
<td></td>
<td>Chick Early</td>
<td>Chick Late</td>
<td>Adult Pre-laying</td>
<td>Adult Incubation</td>
<td>Chick Early</td>
<td>Chick Late</td>
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<tr>
<td>Mandarte Is.</td>
<td>46R</td>
<td>64R</td>
<td>61P</td>
<td>17P:6R</td>
<td>54R</td>
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<td>Cleland Is.</td>
<td>-</td>
<td>-</td>
<td>47P</td>
<td>1P:5R</td>
<td>4R</td>
<td>19R</td>
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Table 2.2  Frequency of occurrence (%) of marine invertebrate and fish prey taxa consumed by Glaucous-winged Gulls at Mandarte Island and Cleland Island during different breeding stages. Invertebrate taxa represented as occurrence in total samples, fish taxa represented as occurrence in samples containing fish. Total sample size in parentheses. * indicates fish species were not identified in pellets.
<table>
<thead>
<tr>
<th>Food categories</th>
<th>Pellets from adults</th>
<th>Pellets from adults</th>
<th>Regurgitations from chicks</th>
<th>Regurgitations from chicks</th>
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<tr>
<td></td>
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<td>Cleland Island</td>
<td>Mandarte Island</td>
<td>Cleland Island</td>
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<td>Prelay 2010 (61)</td>
<td>Incubation 2010 (17)</td>
<td>Prelay 2010 (46)</td>
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<td></td>
<td></td>
<td>96.4</td>
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</tr>
<tr>
<td># samples containing fish</td>
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<tr>
<td></td>
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<td></td>
<td></td>
<td>18</td>
</tr>
<tr>
<td>Herring</td>
<td>*</td>
<td>*</td>
<td>*</td>
<td>7.3</td>
</tr>
<tr>
<td></td>
<td></td>
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</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>27.8</td>
</tr>
<tr>
<td>Pacific Sand lance</td>
<td>*</td>
<td>*</td>
<td>*</td>
<td>26.8</td>
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<tr>
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<td>50.0</td>
</tr>
<tr>
<td>Salmon</td>
<td>*</td>
<td>*</td>
<td>*</td>
<td>-</td>
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<td></td>
<td></td>
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</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>-</td>
</tr>
<tr>
<td>Pricklebacks/gunnels</td>
<td>*</td>
<td>*</td>
<td>*</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>7.4</td>
</tr>
<tr>
<td>Midshipman</td>
<td>*</td>
<td>*</td>
<td>*</td>
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</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>5.6</td>
</tr>
<tr>
<td>Unidentified/digested</td>
<td>*</td>
<td>*</td>
<td>*</td>
<td>64.4</td>
</tr>
<tr>
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<td>65.4</td>
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<td>50.0</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>27.8</td>
</tr>
</tbody>
</table>
Table 2.3  *Glaucous-winged Gull diet composition by gull age (adult vs chick), year, and colony location. Data are expressed as frequency of occurrence (%) of each food category, sample size in parentheses. Historical data taken from Vermeer (1982). * indicates a difference in the way diet categories were compiled between Vermeer (1982) and current data.*

<table>
<thead>
<tr>
<th>Food categories</th>
<th>Pellets from adults</th>
<th>Regurgitations from chicks</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Georgia Strait</td>
<td>West Coast</td>
</tr>
<tr>
<td></td>
<td>1980 (179)</td>
<td>2010 (78)</td>
</tr>
<tr>
<td></td>
<td>2010 (31)</td>
<td>2010 (47)</td>
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<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Human refuse</td>
<td>69.3 41.0</td>
<td>22.2 16.4 3.7 0.7</td>
</tr>
<tr>
<td>Fish</td>
<td>16.8 37.2</td>
<td>60.5 84.5 96.3</td>
</tr>
<tr>
<td>Marine invertebrate</td>
<td>* 57.7 * 100</td>
<td>* 5.5 * 5.9 *</td>
</tr>
<tr>
<td>Bivalves</td>
<td>23.5 * 12.9 *</td>
<td>11.3 * * *</td>
</tr>
<tr>
<td>Gastropods</td>
<td>0.6 * 6.5 *</td>
<td>0.4 * * *</td>
</tr>
<tr>
<td>Bivalve/gastropod</td>
<td>* 25.6 * 14.9</td>
<td>* 1.8 * 1.2 *</td>
</tr>
<tr>
<td>Chitons</td>
<td>2.8 2.6</td>
<td>0.4 - -</td>
</tr>
<tr>
<td>Squid</td>
<td>- -</td>
<td>- -</td>
</tr>
<tr>
<td>Crabs</td>
<td>9.5 * 3.2 *</td>
<td>0.8 * * *</td>
</tr>
<tr>
<td>Shrimp</td>
<td>0.6 * - *</td>
<td>0.4 * * *</td>
</tr>
<tr>
<td>Crabs/Shrimp</td>
<td>* 29.5 * 4.3</td>
<td>* 1.8 2.4 * *</td>
</tr>
<tr>
<td>Isopods</td>
<td>0.6 - -</td>
<td>- -</td>
</tr>
<tr>
<td>Euphausiids</td>
<td>- -</td>
<td>0.9 - -</td>
</tr>
<tr>
<td>Gooseneck</td>
<td>barnacles</td>
<td>1.7 - 87.1 93.6</td>
</tr>
<tr>
<td>Errant polychaete</td>
<td>- 25.6 - -</td>
<td>0.4 0.9 3.7</td>
</tr>
<tr>
<td>Sea star</td>
<td>1.7 1.3 - 2.1</td>
<td>5.0 2.7 -</td>
</tr>
<tr>
<td>Terr invertebrates</td>
<td>3.0 3.8 - -</td>
<td>0.4 - 1.2 -</td>
</tr>
<tr>
<td>Mice</td>
<td>0.6 - 3.2 -</td>
<td>- - - -</td>
</tr>
<tr>
<td>Digested animal</td>
<td>matter</td>
<td>0.6 24.4 - -</td>
</tr>
<tr>
<td>Plant material</td>
<td>62.6 56.4 12.9 2.1</td>
<td>8.8 66.4 80.5 - 17.4</td>
</tr>
</tbody>
</table>
Table 2.4  **Composition of fish species found in Glaucoous-winged Gull chick fish regurgitations, by colony location and year. Represented as frequency of occurrence (%). Sample size in parentheses. Historical data taken from Vermeer (1982).**

<table>
<thead>
<tr>
<th>Fish</th>
<th>Georgia Strait</th>
<th></th>
<th>West Coast</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1980 (144)</td>
<td>2009 (93)</td>
<td>2010 (79)</td>
<td>1980 (129)</td>
</tr>
<tr>
<td>Herring (<em>Clupea pallasii</em>)</td>
<td>55.6</td>
<td>9.7</td>
<td>2.5</td>
<td>24.8</td>
</tr>
<tr>
<td>Pacific sand lance (<em>Ammodytes hexapterus</em>)</td>
<td>1.4</td>
<td>25.8</td>
<td>53.2</td>
<td>21.7</td>
</tr>
<tr>
<td>Pacific saury (<em>Cololabis saira</em>)</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>20.2</td>
</tr>
<tr>
<td>Salmon (<em>Oncorhynchus sp</em>)</td>
<td>4.2</td>
<td>1.1</td>
<td>1.3</td>
<td>2.3</td>
</tr>
<tr>
<td>Pricklebacks/gunnels (suborder Zoarcoidei)</td>
<td>3.5</td>
<td>-</td>
<td>2.5</td>
<td>-</td>
</tr>
<tr>
<td>River lamprey (<em>Lampetra ayresi</em>)</td>
<td>1.4</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Midshipman (<em>Porichthys notatus</em>)</td>
<td>-</td>
<td>1.1</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Sculpin (Superfamily Cottoidea)</td>
<td>2.1</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Unidentified and digested</td>
<td>43.1</td>
<td>64.5</td>
<td>43.1</td>
<td>34.9</td>
</tr>
</tbody>
</table>
Figure 2.1  Composition of Glaucous-winged Gull diet by breeding stage at Mandarte Island and Cleland Island. Data are expressed as frequency of occurrence (%) of six broad food categories: HR – human refuse, Fish – fish, MI – marine invertebrate, TI – terrestrial invertebrate, TA – terrestrial animal, PM – plant matter.
3. **Dietary ecology of the Glaucous-winged Gull (Larus glaucescens) in the Pacific Northwest, as inferred from stable isotope analysis**

3.1. **Introduction**

Marine systems have been affected by human activities for centuries, from over exploitation of fish stocks (Pauly et al. 1998, Jackson et al. 2001, Myers et al. 2003), and modification of coastal habitats (Burke et al. 2001), to dumping of wastes and run-off of pollutants and nutrients from terrestrial activities (Elliott et al. 1992, Burke et al. 2001). In relation to contaminants, persistent organic pollutants (POPs) are of particular concern due to their global distribution, and persistent and lipophilic properties that lead to bioaccumulation in marine food webs (Jones and De Voogt 1999). In 2006 Environment Canada launched the Chemical Management Plan (CMP), under which the Environmental Monitoring and Surveillance program was established to track legacy and emerging POPs of concern at the national level in air, sediment, water, landfills, wastewater treatment and biota (fish and birds). Building on the existing Great Lakes Herring Gull Egg Monitoring Project, several additional gull colonies (Laridae family) were added to monitor contaminant levels and trends in aquatic wildlife across Canada (e.g. Gebbink et al. 2011, Chen et al. 2012). As the sole Larid species breeding in British Columbia’s coastal waters (Vermeer and Devito 1987), the Glaucous-winged Gull (*Larus glaucescens*) is the only species utilized to monitor contaminants in the marine environment at three Pacific coast CMP-funded sites.

Larid gull species, like many seabird species, often act as avian top predators in marine ecosystems (Furness and Camphuysen 1997) and numerous studies have demonstrated that many persistent organic pollutants bio-accumulate with increasing trophic level and/or marine dietary input (e.g. Jarman et al. 1996, Elliott 2005, Elliott et al. 2009, Hebert et al. 2009). Additionally, lipophilic contaminants accumulate and persist
in tissues with higher lipid content, such as eggs (Jones and De Voogt 1999, Dauwe et al. 2006). Seabird eggs have been widely used in monitoring programs because they are easy to collect, collection has minimal impact on the population, their eggs are rich in lipid composition and lipid-soluble contaminant concentrations in eggs reflect female blood concentrations (Gilbertson et al. 1987, Becker 1989, 2003, Furness and Camphuysen 1997). Various Larid gull species can exhibit different resource allocation strategies for egg production (capital-income continuum) (Houston et al. 1983, Meijer and Drent 1999, Ramirez et al. 2011). Capital-leaning breeders mobilize more endogenous nutrients and lipids from dietary intake outside the breeding season, while income breeders use more exogenous nutrients and lipids acquired at the breeding grounds in a short time prior to egg production. Although the resource allocation strategy of Glaucous-winged Gulls on the Pacific coast of Canada has not been investigated, they are considered largely non-migratory (Elliott unpublished) (Vermeer 1963, Hatch et al. 2011), and contaminant levels in their eggs are assumed to reflect contaminants accumulated locally within the Pacific Coast region of British Columbia. To fully understand contaminant intake and egg contaminant burdens, researchers require information on current dietary information and seasonal ranges of the population in question (Pearce et al. 1989).

Numerous approaches can be taken to investigating diet of marine seabird species, including "traditional" analysis of pellets and regurgitates (see Chapter 2). However an alternative technique, stable isotope analysis, has become a widely used tool in avian dietary and foraging research (Inger and Bearhop 2008). Unlike conventional diet sampling, which measures recent meals, stable isotope analysis allows researchers to investigate assimilated diet over longer time periods, by using tissues with a known metabolic turnover rate (e.g. blood for days to weeks, bone collagen for months) (Hobson and Clark 1992, 1993). Although specific taxonomic information on different prey items cannot be inferred, stable isotope analysis is advantageous as it avoids the biases in identifying soft-bodied prey types inherent in conventional sampling (Barrett et al. 2007, Karnovsky et al. 2012). Due to the predictable nature with which the nitrogen and carbon isotopes move from prey to consumer these isotopes provide a measure of trophic level and dietary carbon source, respectively. In marine food webs, $\delta^{15}N$ signatures increase by 3-5‰ from one trophic level to the next, due to excretion of
the light amine group (Minagawa and Wada 1984, Hobson and Welch 1992, Michener and Schell 1994). Since carbon fractionates at a rate of less than 1‰ from one trophic level to the next, and the δ¹³C signature varies depending on the photosynthetic pathway, it can be utilized to determine foraging habitat (Inger and Bearhop 2008). Terrestrial and freshwater systems are depleted in ¹³C relative to marine systems, with pelagic/offshore regions depleted relative to benthic/inshore regions (Michener and Schell 1994). Therefore, stable isotope analysis provides a valuable tool for the assessment of integrated diet over known time periods, the identification of foraging habitat type, and determination of trophic level, which can compliment more traditional approaches to dietary analysis (Hobson et al. 1994).

In the present study we investigated the diet of the Glaucous-winged Gull over a two-year period at the three Pacific coast CMP monitored colonies, Mandarte and Cleland Islands. Specifically, we used δ¹³C and δ¹⁵N stable isotope analysis of egg homogenate, adult cellular blood, and chick blood plasma to elucidate: a) within-site differences in breeding stages, b) within-site differences between years, and c) geographic differences among colonies. Based on prior results from conventional sampling (chapter 2), we predicted that all isotope signatures would reflect a predominant marine prey influence. Adult signatures from Mandarte Island were expected to show the most variation, with the strongest marine signal predicted in all chick stages and in Cleland Island adults. Our results are pertinent to future CMP egg-monitoring research, as knowledge of adult foraging and diet prior to egg-laying are essential to understanding kinetics of contaminant accumulation in Glaucous-winged Gull eggs. Data on diet from various breeding stages, including chicks, are also important for overall assessment of seabird health.

3.2. Methods

3.2.1. Study sites

Glaucous-winged gulls were sampled at all three Pacific coast CMP-monitored colonies during the breeding seasons of 2009 and 2010. Two of these sites, Mandarte Island (48.63° N, 123.28° W) and Mitlenatch Island Nature Provincial Park (49.95° N,
125.00° W) are located within the Georgia Strait/Salish Sea Ecosystem, BC, and represent sites in close proximity to urbanized areas. Previous conventional diet analysis at Mandarte Island reflects the opportunistic nature of their foraging, with a preference for marine prey sources (Chapter 2). Both are among the largest colonies in the Strait (LK Blight pers. com.), with gulls nesting predominantly in grassy meadows (Henderson 1972, Vermeer and Devito 1987). Located on the exposed west coast of Vancouver Island, Cleland Island Ecological Reserve (49.16°N, 126.08°W) represents a more remote site, with nesting habitat restricted to the rocky margin surrounding the island (Henderson 1972, Vermeer and Devito 1987). The colony supports ca. 1400 breeding pairs (pers. comm. Peter Clarkson, Resource Conservation Supervisor, Pacific Rim National Park Reserve of Canada), with historical and recent conventional diet studies reflecting a nearly exclusive marine diet (chapter 2) (Henderson 1972, Ward 1973).

### 3.2.2. Sample collection

Adult gulls were trapped on Mandarte Island during nest initiation prior to laying (May 7-10, 2010, Table 3.1 sample sizes) by placing fish-baited noose-mats in nest territories. Once an adult was caught the trap was relocated to avoid trapping adults from the same breeding pair. Morphometric measurements (weight, bill length and depth, and wing and tarsus length) were taken, and a maximum of 5ml of blood was drawn from the brachial vein using a sodium-heparinized syringe. Blood was centrifuged into plasma and red blood cell (cellular) fractions, and stored in liquid nitrogen for transport to the lab.

During the incubation stage, adult gulls were captured using drop traps placed over active nests containing three egg clutches [as described by Mills and Ryder (1979)]; on Mandarte Island between June 7th-9th 2010, on Cleland June 19th 2009 and July 1-2nd 2010, and on Mitlenatch Island on June 17th 2009 and June 11th 2010. To minimize disturbance and avoid prolonged exposure of eggs, the traps were relocated after an adult was caught, or after 20 minutes without incubation, whichever occurred first. Morphometric measurements and blood were taken as mentioned above. When an adult was successfully trapped, one egg from the nest was also collected, after a float-test to determine embryonic development stage. Since in ovo chick development can significantly alter the isotopic signatures of egg components, eggs were only collected if
the float-test indicated they were freshly-laid (Liebezeit et al. 2007, Sharp et al. 2009). No trapping was attempted on Mandarte Island in 2009 to avoid having the adults becoming trap shy. Instead, eggs were randomly collected from three egg clutches (see Table 3.1 for sample sizes).

In order to examine how isotopic signatures changed across the 2009 and 2010 breeding seasons, chick blood plasma samples were also collected for isotope analysis at approximately 2 weeks (Mandarte Island: July 22\textsuperscript{nd}-31\textsuperscript{st} 2009 and July24-26\textsuperscript{th} 2010, Cleland: August 5\textsuperscript{th} 2010) and 4 weeks of age (Mandarte Island: August 4-11\textsuperscript{th} 2009 and August 14-16\textsuperscript{th} 2010, Cleland Island: August 26\textsuperscript{th} 2010; sample sizes Table 3.1). Blood plasma exhibits a higher turnover rate than the red blood cell fraction (for example, δ\textsuperscript{13}C has a turnover \(\frac{1}{2}\) life of 2.9 days for plasma versus 29.8 days for cellular blood in the American crow (Hobson and Clark 1993) and was therefore not expected to contain maternal influences from egg-lay, and more likely to reflect diet provisioned by parents. Methods of capture were identical to those described in Chapter 2 as this work was conducted simultaneously with conventional sampling. Blood was drawn from the brachial vein using a sodium heparinized syringe, to a maximum of 1.5 ml, and processed as described above (see table 1 for sample sizes).

Marine invertebrate filter feeder and grazer samples were collected from intertidal zones at each colony. These reflect primary consumer values at each site and are used to determine regional differences in base food web stable isotope signatures. Whole samples of Pacific Herring (\textit{Clupea pallasii}) and Pacific Sand Lance (\textit{Ammodytes hexapterus}) were collected from Mandarte Island nestling regurgitations. Regionally relevant isotopic values for offshore, zooplanktivorous (Leach’s Storm Petrel) and near-shore, piscivorous seabird (Pelagic and Double-crested Cormorant) eggs, human refuse, and terrestrial invertebrates were obtained from unpublished data (John Elliott unpublished) and literature (Hebert et al. 1999, Evans Ogden et al. 2005).

\textbf{3.2.3. Isotope analysis}

All samples were dried in an oven at 60°C to a constant mass and ground to a powder using a mortar and pestle. Lipids were extracted from ca. 20 mg of dried, powdered egg homogenate and fish prey muscle tissue using 2 ml of
chloroform/methanol solvent (2:1). Samples were placed on ice and homogenized using a Glas-Col Stirrer (Glas-Col, LLC) with teflon pestle, then centrifuged at 4°C and 3200 rpm for 10 minutes, after which the supernatant containing lipids was discarded. Rinsing process was repeated three times before solvent was evaporated and samples were allowed to dry in the fumehood overnight. Lipids were not extracted from blood samples or marine invertebrate muscle tissues since a recent study by Bearhop et al. (2000) on Great Skuas demonstrated no effect of lipid extraction on isotopic values of whole blood, and animal muscle tissue has also been shown to contain minimal lipid content (Tieszen et al. 1983). Lipid extraction on Mytilus californianus muscle tissue has also been shown to have no effect on δ13C or δ15N isotopic signatures (Gilbane 2006). Sub-samples of 1-2mg were analysed for nitrogen (15N/14N) and carbon (13C/12C) isotope ratios at the University of California Davis Stable Isotope Facility. Analyses were performed on a continuous flow system using a PDZ Europa ANCA-GSL elemental analyser and a PDZ Europa 20-20 isotope ratio mass spectrometer (Sercon Ltd., Cheshire, UK). Final sample values are expressed in delta notation, parts-per-mil (‰), relative to international standards (Vienna PeeDee Belemnite (V-PDB) for carbon and atmospheric air for nitrogen), according to the following equation:

\[
\delta X = \left( \frac{r_{\text{sample}}}{r_{\text{standard}}} - 1 \right) \times 1000
\]

Where \(X\) is 15N or 13C and \(r\) is the corresponding ratio (15N/14N or 13C/12C). Long-term facility measurement precision is 0.2‰ for δ13C and 0.3‰ for δ15N.

3.2.4. **Statistical analysis**

To determine any dietary differences in a) within-colony breeding stages (adult pre-lay/incubation or chick early/late), b) years (adult incubation, egg homogenate, or chick stage) or c) colony locations (chicks), we compared mean isotopic values (δ13C and δ15N) in two-sample t-tests. Adults and chicks were not compared statistically as different blood fractions were tested for each. When unequal variances between groups occurred we used Welch’s T-test. Statistical analysis was performed in JMP v9.0.2 (SAS Institute Inc. 2010). We also ran a General Linear Model (GLM) with Games-Howell post-hoc test using SPSS Statistics v.19 (SPSS Inc.) to determine whether marine invertebrate prey signatures (δ13C and δ15N) differed among colonies, and investigate
effects of year and colony location on incubating adults and egg homogenate isotopic signatures ($\delta^{13}C$ and $\delta^{15}N$). For all tests significance was assumed at $p < 0.05$. Figures were created using Graphpad Prism v.4.0 (Graphpad Software Inc. 2005).

3.3. Results

3.3.1. Within-site inter-individual variation at each CMP colony

Individual isotopic signatures at Mandarte Island and Mitlenatch Island showed a greater degree of variation in both $\delta^{13}C$ and $\delta^{15}N$ than Cleland Island (Fig. 3.1). At Mandarte Island this was particularly noticeable with the adult prelay stage (2010 $\delta^{13}C$ -15.38 to -22.48‰ and $\delta^{15}N$ 7.26 to 14.62‰) and incubation stage (2010 $\delta^{13}C$ -14.28 to -20.85‰ and $\delta^{15}N$ 9.70 to 16.99‰) (Fig.1). The incubating adult stage was most variable at Mitlenatch Island in both years (2009 $\delta^{13}C$ -16.21 to -18.33‰ and $\delta^{15}N$ 13.21 to 14.86‰; 2010 $\delta^{13}C$ -13.72 to -19.02‰ and $\delta^{15}N$ 11.81 to 14.34‰) (Fig. 3.1).

3.3.2. Within-site breeding stage differences at each CMP colony

Mandarte Island

The 2010 mean cellular blood $\delta^{13}C$ signatures for both pre-lay and incubating adults fell within the marine range; however, the two differed significantly (pre-lay values $+0.53‰$) (Table 3.1, Fig. 3.2; $t_{27} = -2.58, p = 0.0155$). Incubating adults fed at a higher trophic level ($+3.04‰$) than pre-lay adults as indicated by the significant difference in $\delta^{15}N$ cellular blood signatures (Table 3.1, Fig. 3.2; $t_{27} = -3.28, p = 0.0029$). For chicks in 2009, there were no differences in $\delta^{13}C$ or $\delta^{15}N$ blood plasma signatures between early and late stages (Table 3.1, Fig. 3.2; t-test for $\delta^{13}C$: $t_{18} = -1.38, p = 0.1855$; t-test for $\delta^{15}N$: $t_{18} = 0.97, p = 0.3446$). In contrast, early and late stage chicks in 2010 differed significantly in both $\delta^{13}C$ and $\delta^{15}N$ blood plasma signatures, with early chicks having a significantly higher $\delta^{13}C$ ($+0.94‰$) and $\delta^{15}N$ ($+0.68‰$) (Table 3.1, Fig. 3.2; t-test for $\delta^{13}C$: $t_{58} = -6.77, p < 0.0001$; t-test for $\delta^{15}N$: $t_{58} = -4.77, p < 0.0001$).
Cleland Island

In 2010, chick stages did not differ in $\delta^{13}$C, with both stages feeding within the marine range (Table 3.1, Fig. 3.2; $t_{48} = 0.25$, $p = 0.8041$); however, later stage chicks were fed at a significantly higher trophic level ($+0.21\%$) (Table 3.1, Fig. 3.2; $t^{48} = 2.76$, $p = 0.0081$).

3.3.3. Inter-year dietary differences at CMP colonies

Mandarte Island

Mean $\delta^{13}$C and $\delta^{15}$N egg homogenate values did not differ significantly between 2009 and 2010 (Table 3.1, Fig. 3.2; t-test for $\delta^{13}$C: $t_{12} = 0.84$, $p = 0.4200$; t-test for $\delta^{15}$N: $t_{12} = -0.72$, $p = 0.4844$). While early stage chicks in both 2009 and 2010 were fed marine-based prey, 2010 chicks had a higher ($+1.02\%$) mean $\delta^{13}$C blood plasma signature (Table 3.1, Fig. 3.2; $t_{38} = 3.25$, $p = 0.0024$), and a higher ($+1.52\%$) trophic level (Table 3.1, Fig. 3.2; $t_{38} = 6.69$, $p < 0.0001$). Later stage chicks from 2009 and 2010 did not differ in their mean $\delta^{13}$C blood plasma signatures (Table 3.1, Fig. 3.2; $t_{38} = 0.34$, $p = 0.7373$) but the later stage chicks in 2010 were fed at a significantly higher ($+0.52\%$) trophic level (Table 3.1, Fig. 3.2; $t_{38} = 2.44$, $p = 0.0193$).

Mitlenatch Island

Mean $\delta^{13}$C and $\delta^{15}$N egg homogenate values did not differ significantly between 2009 and 2010 (Table 3.1, Fig. 3.2; t-test for $\delta^{13}$C: $t_{9} = 0.76$, $p = 0.4681$; t-test for $\delta^{15}$N: $t_{9} = -0.84$, $p = 0.4233$). Incubating adults did not differ between years in their mean $\delta^{13}$C cellular blood signatures (Table 3.1, Fig. 3.2; Welch’s $t_{19.899} = 0.81$, $p = 0.4269$), however, 2009 adults had a higher ($+0.84\%$) mean $\delta^{15}$N signature (Table 3.1, Fig. 3.2; Welch’s $t_{21.457} = 2.77$, $p = 0.0113$).

Cleland Island

Mean egg homogenate values did not differ in $\delta^{13}$C (Table 3.1, Fig. 3.2; $t_{10} = -2.17$, $p = 0.0556$), whereas mean $\delta^{15}$N egg homogenate in 2010 was higher ($+0.45\%$) than 2009 (Table 3.1, Fig. 3.2; $t_{10} = 3.99$, $p = 0.0026$). The same trend existed between years for incubating adult cellular blood, with $\delta^{13}$C reflecting a marine signature, and no
difference between years (Table 3.1, Fig. 3.2; $t_{23} = 0.59$, $p = 0.5603$), and trophic level higher in 2010 (+0.53‰) than 2009 (Table 3.1, Fig. 3.2; $t_{23} = 5.42$, $p < 0.0001$).

### 3.3.4. Spatial variation in diet among the three CMP colonies

**Marine Invertebrate prey**

No significant difference was found in mean $\delta^{13}$C marine invertebrate signatures among sites, indicating that baseline marine isotopic signature did not vary with geographic location (GLM $F_{2,47} = 2.713$, $p = 0.077$). However, mean $\delta^{15}$N marine invertebrate signatures differed significantly among colony locations (GLM $F_{2,47} = 17.406$, $p = 0.000$), marine invertebrates from Cleland Island occupied a significantly higher trophic level than both Mandarte Island (Games Howell $p = 0.001$) and Mitlenatch Island (Games Howell $p = 0.000$), with no significant difference between Mandarte and Mitlenatch Islands (Games Howell $p = 0.433$).

**Egg Homogenate samples**

GLM analysis investigating the effects of year and colony location on mean $\delta^{13}$C egg homogenate values showed no differences between years ($F_{1,37} = 0.33$, $p = 0.719$), locations ($F_{2,37} = 0.30$, $p = 0.590$) or year*site interaction ($F_{2,37} = 1.16$, $p = 0.328$). The GLM for mean $\delta^{15}$N egg homogenate values showed no effect of year or year*site interaction, so these were removed and the model re-run. A significant difference in mean $\delta^{15}$N egg homogenate values among colonies was found (GLM $F_{2,37} = 4.24$, $p = 0.023$), a Games-Howell Post-hoc test revealed no significant difference between Mandarte and Mitlenatch Islands ($p = 0.852$) and Mitlenatch and Cleland Islands ($p = 0.090$), and a significant difference between Cleland and Mandarte Islands ($p = 0.013$).

**Adult Incubation samples**

When the effects of year and colony location on adult incubation were investigated using GLM, no effect of year or year*site interaction were found on mean $\delta^{13}$C cellular blood values, year was removed and the model rerun, with no significant difference among colony locations ($F_{2,66} = 0.12$, $p = 0.885$). The GLM to examine mean $\delta^{15}$N adult incubation values yielded no effect of year, but a year*site interaction warranted splitting the analysis to analyze years separately. In 2009, a significant
difference was found among colonies, with Cleland adults higher (+1.09‰) in $\delta^{15}$N than Mitlenatch adults ($F_{1,23} = 8.81$, $p = 0.007$). In 2010 a significant difference ($F_{2,43} = 6.99$, $p = 0.002$) was also revealed between Cleland and Mitlenatch Islands (Games Howell $p = 0.000$) but not between Cleland and Mandarte (Games Howell $p = 0.068$) or Mandarte and Mitlenatch (Games Howell $p = 0.561$).

### 3.4. Discussion

In the present study, we used stable isotope analysis to examine the dietary ecology of the Glaucous-winged Gull at three Pacific Canadian colonies during various stages of the breeding season. As predicted, analysis of $\delta^{13}$C isotope signatures from a variety of tissue types demonstrated that Glaucous-winged Gull diet at all three colonies, fell within a near-shore marine range when assessed against near-shore invertebrate prey, regardless of breeding stage or year. With the exception of samples from pre-laying adults at Mandarte Island in 2010, $\delta^{15}$N signatures indicated that diet during all breeding stages occupied a high trophic position, which was similar to that of near-shore piscivorous seabirds sampled in the same area (Double-crested Cormorant and Pelagic Cormorant) (Elliott unpublished). The range of potential isotopic signatures for human refuse is varied which could lead to mis-assignment of marine/terrestrial foraging habitat and/or trophic level of stable isotope signatures. However, the majority of meat products in human refuse are assumed to originate from herbivorous domesticated animals (poultry, cattle, swine, and sheep) (Hebert et al. 1999). Since commercial feeds in Canada are limited to a very small proportion of animal protein (usually <5%, Darin Bennett pers. comm.), $\delta^{15}$N values of human refuse protein are much lower than $\delta^{15}$N signatures of marine invertebrate prey from colonies in the Canadian Pacific waters, but similar to local terrestrial invertebrate $\delta^{15}$N signatures (Fig. 3.2). As a result, gulls consuming greater proportions of terrestrial prey or human refuse would be expected to occupy lower trophic positions, the opposite of what we observed. Although our study confirms that Glaucous-winged Gulls can be considered primarily a ‘marine’ species in the context of the Chemical Management Plan (CMP), or other, environmental toxicology monitoring, our study highlights several sources of variation in diet that should be considered in the interpretation of egg contaminants data: a) within-colonies, and within years there may be considerable individual variation in a breeding stage that is masked.
if the mean breeding stage value is only examined; b) variation in adults (comparing pre-lay and incubation stages) and chicks (comparing early and late chick-rearing stages); c) inter-year differences within-colonies; d) inter-colony differences may relate to food web base-line differences or absolute differences in dietary ecology.

Inter-individual variation in both δ¹³C and δ¹⁵N signatures was greater at Mandarte Island compared with other sites, particularly for diets during adult pre-lay and incubation stages. Incubating adults at Mitenatch Island also exhibited a substantial degree of amount of δ¹³C and δ¹⁵N variation (Fig. 3.1). Combined, that suggests diet composition was more varied at colonies located in the Salish Sea, in closer proximity to urbanized areas. Findings from conventional analysis support this idea, as occurrences of marine prey, human refuse, and terrestrial prey were found in adult samples at Mandarte Island; whereas samples from Cleland Island were almost entirely marine prey (Chapter 2).

At Mandarte Island, adult diet during the incubation period in 2010 was estimated to be at a significantly higher trophic level than during the adult pre-lay period at the same site and year. Analysis of regurgitated pellets did not reveal any significant differences in the species composition of diets, although samples from incubating adults had slightly lower occurrences of human refuse and fish, and higher occurrences of marine invertebrate prey (Chapter 2). A similar pattern, of incubating adults occupying a higher trophic level than pre-lay adult stages, was found in Tasmanian Silver Gulls (Larus novaehollandiae) breeding near urbanized areas, where authors concluded that it indicated a change in available refuse (Auman et al. 2011). Our results are also consistent with those of Blight (2012), who examined isotopic trends (over 150 years) in adult Glaucous-winged Gulls in the Salish Sea region and found pre-breeding adults fed at a somewhat decreased trophic level. This could be driven by the need to acquire specific nutrients prior to egg-lay, since work by Pierotti and Annett (1987, 1991) suggested that gulls which specialized on marine invertebrate prey (therefore feeding at a lower trophic level) had earlier lay dates, produced larger and heavier clutches, and exhibited higher rates of hatching/fledging success than generalists and other prey specialists.
Between chick rearing stages at Mandarte Island, early and late stage chicks in 2009 did not differ in isotopic signatures, as supported by conventional regurgitation analysis (chapter 2). However, in 2010 early stage chicks had significantly higher $\delta^{15}\text{N}$ signatures than late stage chicks. Conventional analysis in 2010 found the diet of late stage chicks had a significantly higher occurrence of marine invertebrate prey, but contained similar occurrences of fish, explaining the slight dip in $\delta^{15}\text{N}$ signatures. The opposite was found for chick isotopic signatures at the more remote Cleland Island, with later stage chick tissues reflecting a higher $\delta^{15}\text{N}$ value than early stage chicks, although no significant difference was found with conventional diet analysis. Isotope studies have recently begun to address how nutritional stress and growth can affect isotopic signatures. In particular, chicks with faster growth rates displayed depleted $\delta^{15}\text{N}$ values (Sears et al. 2009). Earlier work by Henderson (1972) and Ward (1973) found that on Mandarte Island growth efficiency decreases with number of days post-hatch (after the first 10 days), and that average growth rates for chicks at Cleland Island were higher than Mandarte Island. Assuming Cleland chicks still exhibit a faster growth rate that may explain the observed difference in $\delta^{15}\text{N}$ signatures.

Larid Gulls employ a variety of resource allocation strategies for egg production that fall along the capital-income continuum (Houston et al. 1983, Meijer and Drent 1999, Ramirez et al. 2011). Glaucous-winged Gulls breeding on the Pacific coast of Canada are considered relatively non-migratory, overwintering locally (Elliott unpublished data) (Vermeer 1963, Hatch et al. 2011), with the majority of birds arriving at the breeding colonies approximately one month in advance of egg-laying (Vermeer 1963). As a result, egg isotope signatures should reflect dietary protein resources acquired locally within the breeding area. Egg signatures from all three colonies indicated that resources allocated to eggs were primarily from near-shore marine dietary sources at higher trophic positions (Fig. 3.2). No inter-year variation was observed at the Salish Sea colonies; however, at Cleland Island egg homogenate had a significantly higher $\delta^{15}\text{N}$ signature in 2010 than 2009. Incubating adults at Cleland also appear to occupy a higher trophic position in 2010 than 2009, possibly indicating that higher quality prey were available to pre-lay and incubating adult stages during 2010.

At Mandarte Island in 2010, diets during both early and late chick stages occupied significantly higher trophic positions than chick diets in the 2009 breeding
season. Blight (2012) reported that fledging success in 2010 was significantly higher than 2009, a year in which chicks were provisioned with more fish and less human refuse (Chapter 2). Increased reproductive success when nestlings were fed fish was reported previously for Alaskan Glaucous-winged Gulls (Murphy et al. 1984), and Herring Gulls in Newfoundland and the Great Lakes (Pierotti and Annett 1987, Fox et al. 1990).

To compare isotopic signatures among geographically separate locations knowledge of local baseline $\delta^{13}C$ and $\delta^{15}N$ signatures should be obtained for each colony, in order to distinguish whether observed differences are due to variances in baseline prey signatures or differences in food web structure (Post 2002). Although Glaucous-winged Gulls do consume anthropogenic and terrestrial items, conventional analysis has demonstrated a preference for marine prey items at all colonies (Chapter 2), and we assume that meat from human refuse has a similar signature at all locations (see discussion above). Marine invertebrate prey $\delta^{13}C$ values from the three CMP colonies were not significantly different, indicating similar baseline carbon sources. However, marine invertebrate prey at Cleland Island had a significantly higher $\delta^{15}N$ signature than the Salish Sea colonies. Located on the west coast of Vancouver Island, Cleland Island is likely under a greater influence of nutrients from upwelling currents than Mandarte and Mitlenatch, which lie in the protected waters of the Salish Sea. A similar pattern was found by researchers investigating biogeographic isotopic trends in South African intertidal communities, where filter-feeders at sites linked more closely to eutrophic, upwelling had more enriched $\delta^{15}N$ signatures (Hill and McQuaid 2008). We found significant differences in the $\delta^{15}N$ signatures of egg homogenate and diets of incubating adults among colonies; however, those did not reflect the results seen with marine invertebrates and yield somewhat ambiguous results. That may relate to differences in dietary composition that were not discernable with the use of two isotopes. The use of sulphur stable isotope ($\delta^{34}S$) analysis (along with $\delta^{13}C$ and $\delta^{15}N$) as a dietary tracer has proven a valuable complement in determining dietary composition of omnivorous species in more complex food webs (Knoff et al. 2002, Hebert et al. 2008, Moreno et al. 2010) and could be considered in future studies.

In conclusion, our study shows that Glaucous-winged Gulls at Pacific CMP colonies are feeding primarily within a marine food web at higher trophic levels.
However, considerable individual variation in adult isotopic signatures from the Salish Sea colonies, supported by data from conventional analysis (Chapter 2), indicates that gulls will supplement their diet with food from anthropogenic sources. Dietary intake of adult gulls prior to egg-laying will have implications for the classes of contaminants sequestered in eggs, since bioaccumulative organic pollutants can increase with trophic level and proportion of marine prey (Jarman et al. 1996, Hebert et al. 2000, Elliott et al. 2009). Stable isotope analysis infers foraging habitat and food web dynamics that, paired with previous conventional diet analysis, provides the current dietary ecology necessary for interpretation of CMP egg contaminants data. We recommend that future contaminants monitoring and isotopic analysis ($\delta^{13}$C and $\delta^{15}$N) be paired with the use of a third isotope ($\delta^{34}$S) and Bayesian mixing models (allows for incorporation of prior diet analysis) as this technique has been shown to provide increased dietary resolution in another omnivorous gull species (Moreno et al. 2010).
References


Henderson, B. A. 1972. The control and organization of parental feeding and its relationships to the food supply for the glaucous-winged gull, Larus glaucescens. MSc, University of British Columbia, Vancouver, BC.


Table 3.1  Glaucous-winged Gull stable isotope signatures (δ^{13}C and δ^{15}N; mean ± SD) for various tissues collected in 2009 and 2010 at three CMP-monitored colonies. Lipids were extracted from egg homogenate samples but not red blood cell or blood plasma fractions.
<table>
<thead>
<tr>
<th>Colony</th>
<th>Year</th>
<th>Breeding Stage</th>
<th>Tissue</th>
<th>N</th>
<th>$\delta^{13}$C (‰)</th>
<th>$\delta^{15}$N (‰)</th>
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</thead>
<tbody>
<tr>
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<td></td>
<td></td>
<td></td>
<td></td>
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<td>SD</td>
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Figure 3.1  *Individual variation in Glaucous-winged Gull stable isotope values (δ\textsuperscript{13}C and δ\textsuperscript{15}N) by breeding stage and year at three CMP-monitored colonies. Cellular blood fraction was used for adult breeding stages and blood plasma for chick breeding stages.*
Figure 3.2  Glaucous-winged Gull mean stable isotope values $\delta^{13}C$ and $\delta^{15}N$; ± SD) by breeding stage and year at three CMP-monitored colonies relative to reference items. Terrestrial invertebrate prey and human refuse prey values were locally relevant values obtained from literature (Hebert et al. 1999, Evans Ogden et al. 2005), seabird egg values (DCCO – Double-crested Cormorant, PECO – Pelagic Cormorant, LSPE – Leach’s Storm Petrel) were obtained from Elliott (unpublished). Cellular blood fraction was used for adult breeding stages and blood plasma for chick breeding stages.
4. General Discussion

Based on both conventional diet analysis (Chapter 2) and stable isotope analysis results (Chapter 3), I concluded that near-shore marine prey (forage fish and marine invertebrates) composed a predominant component of Pacific Canadian Glaucous-winged Gull diet. Both approaches to dietary analysis revealed that the diet composition of pre-lay and incubating adults in the Strait of Georgia (i.e. at colonies in closer proximity to urban areas) was more varied than Cleland Island (Chapter 2, Fig. 2.1, Table 2.3; Chapter 3; Fig. 3.1), reflecting the opportunistic foraging nature of this species in areas where marine prey abundances are known to fluctuate (Therriault et al. 2009). Stable isotope analysis of samples at Mandarte Island indicated that both incubating adults, egg homogenate, and chicks diet signatures fell within a marine range at a fairly high trophic level, while conventional results reflected that pre-hatch adults consumed more anthropogenic sources and less fish than chicks. I did observe that pre-lay adults occupied a lower trophic level, however, incubating adults and egg signatures were similar to those of chicks. This anomaly may be due to the differences reflected from a few recent meals versus an assimilated diet signature over a few weeks. Traditional diet approaches allowed us to compare results with historical dietary work (Vermeer 1982), and indicated that pre-lay and incubating adults in the Georgia Strait consume less human refuse and are more piscivorous than 30 years ago, while occurrences of prey on the West Coast were consistent with Vermeer’s findings of a predominantly marine diet.

This study has increased the current knowledge of breeding Glaucous-winged Gull dietary ecology at three Pacific Canadian colonies however, the scope of this research does not address diet-switching between pre-hatch and provisioning adult breeding stages. Although adult Larid gulls can switch their diet between the pre-hatch and provisioning breeding stages, due to nestling nutritional requirements (Pierotti and Annett 1987, Annett and Pierotti 1989, Bertellotti and Yorio 1999), some seabird studies using stable isotope analysis have found that provisioning adults may consume different prey than their nestlings (Hodum and Hobson 2000, Davies et al. 2009). Any further
investigations into dietary ecology across breeding stages should examine both provisioning adults and chicks during the rearing phase.

Glaucous-winged Gulls in the Canadian Pacific are considered local migrants, overwintering in the region and arriving at the breeding colonies a month in advance of egg laying (Vermeer 1963, Hatch et al. 2011) (Elliott unpublished data); therefore, egg contaminant burdens should reflect contaminants accumulated from the adult female diet foraging within the region. To ensure this is the case, and to investigate contaminant uptake outside of the breeding season, future work could investigate dietary ecology during the winter and early spring (similar to Ewins et al. (1994) with Herring Gulls in the Great Lakes). The application of stable isotopes in dietary ecology could also be extended to identify endogenous and exogenous resources used in egg production, (as was done by Hobson et al. (1997) in the Great Lakes ecosystem) if foraging habitats were thought to differ between breeding and overwintering periods.

The observed difference in Mandarte Island chick diet between 2009 and 2010 indicates that diet can significantly fluctuate on a short-term scale, likely due to climactic variations (e.g. El Niño-Southern Oscillation). I recommend that ongoing contaminant monitoring be paired with stable isotope analysis since inter-year changes in climactic conditions can impact forage fish prey availability and may force gulls to rely more heavily on terrestrial or anthropogenic dietary sources. Also, the use of stable isotopes as a proxy for foraging habitat and trophic level (Minagawa and Wada 1984, Hobson and Welch 1992, Michener and Schell 1994, Inger and Bearhop 2008) has allowed us to gain some insight into assimilated diet over longer periods of time, rather than just recent meals. However, in complex food webs, and with omnivorous species, the use of only two isotopes ($\delta^{13}$C and $\delta^{15}$N) may not provide enough resolution. I recommend the addition of a third isotope ($\delta^{34}$S) to any future isotopic analysis, in combination with the use of a baysian mixing model, in order to determine relative contributions of prey items to consumer tissue isotope values.

Both data chapters combined, my thesis serves as an assessment of breeding Glaucous-winged Gull diet in the Canadian Pacific. Overall, my results indicate that Glaucous-winged Gulls are preferentially piscivorous, foraging at a fairly high trophic level within the near-shore Canadian Pacific region, although they opportunistically
supplement their diets with terrestrial and anthropogenic sources when necessary. I support the use of Glaucous-winged Gull eggs to monitor marine contaminants pathways in the region; however, I recommend some caution with interpretation of contaminant results from colonies in close proximity to urban locations. Results from this work, paired with studies on the gulls’ capacity to metabolize and excrete POPs, are critical to understanding the kinetics of contaminant intake and deposition during egg-laying, and will be used in the interpretation of regional contaminant-monitoring data collected under Environment Canada’s CMP Environmental Monitoring and Surveillance Program.
References


