

Diet of yellow-billed loons (*Gavia adamsii*) in Arctic lakes during the nesting season inferred from fatty acid analysis

Trevor B. Haynes¹ · Joel A. Schmutz² · Jeffrey F. Bromaghin² · Sara J. Iverson³ · Veronica M. Padula¹ · Amanda E. Rosenberger⁴

Received: 20 November 2014/Revised: 30 March 2015/Accepted: 1 April 2015/Published online: 11 April 2015
© US Government 2015

Abstract Understanding the dietary habits of yellow-billed loons (*Gavia adamsii*) can give important insights into their ecology, however, studying the diet of loons is difficult when direct observation or specimen collection is impractical. We investigate the diet of yellow-billed loons nesting on the Arctic Coastal Plain of Alaska using quantitative fatty acid signature analysis. Tissue analysis from 26 yellow-billed loons and eleven prey groups (nine fish species and two invertebrate groups) from Arctic lakes suggests that yellow-billed loons are eating high proportions of Alaska blackfish (*Dallia pectoralis*), broad whitefish (*Coregonus nasus*) and three-spined stickleback (*Gasterosteus aculeatus*) during late spring and early summer. The prominence of blackfish in diets highlights the widespread availability of blackfish during the early stages of loon nesting, soon after spring thaw. The high proportions of broad whitefish and three-spined stickleback may reflect a residual signal from the coastal staging period prior to establishing nesting territories on lakes, when loons

are more likely to encounter these species. Our analyses were sensitive to the choice of calibration coefficient based on data from three different species, indicating the need for development of loon-specific coefficients for future study and confirmation of our results. Regardless, fish that are coastally distributed and that successfully overwinter in lakes are likely key food items for yellow-billed loons early in the nesting season.

Keywords Arctic fish · Diet estimation · Quantitative fatty acid signature analysis · Yellow-billed loons

Introduction

The yellow-billed loon (*Gavia adamsii*) is a large waterbird that winters in the marine coastal waters of North America and Eurasia and nests and raises young on Arctic lakes in Alaska, Canada and Russia. Given its large body size and strong territorial behavior, this species functions as an apex predator in large lake ecosystems in circumpolar regions (North 1994; Schmutz et al. 2014). Unlike other loon species, such as the common loon (*Gavia immer*), relatively little is known about the ecology of yellow-billed loons, including diet (North 1994). Food limitation can produce strong bottom-up effects on a population's distribution and abundance (White 1978) and is therefore key to understanding many aspects of a species' ecology including distribution, population dynamics and ecological niche (Wiens and Rotenberry 1979; Sih and Christensen 2001).

The limited information available on the diet of yellow-billed loons is based on a few anecdotal records of stomach contents collected from loons in marine waters. Based on these collections (summarized by North 1994), yellow-billed loons forage on marine fish and invertebrates.

Electronic supplementary material The online version of this article (doi:10.1007/s00300-015-1690-3) contains supplementary material, which is available to authorized users.

✉ Trevor B. Haynes
thaynes9@alaska.edu; t.haynes@alaska.edu

¹ School of Fish and Ocean Sciences, University of Alaska Fairbanks, Fairbanks, AK 99775, USA

² U.S. Geological Survey, Alaska Science Center, Anchorage, AK 99508, USA

³ Department of Biology, Dalhousie University, 1355 Oxford Street, Halifax, NS B3H 4J1, Canada

⁴ Missouri Cooperative Fish and Wildlife Research Unit, U.S. Geological Survey, University of Missouri, Columbia, MO 65211, USA

Stomach contents from birds collected off the Alaskan coast contained tomcod (*Microgadus proximus*), sculpin species (family Cottidae; Cottam and Knappen 1939), rockfish (likely *Sebastes* sp.; Bailey 1922) and a small amount of invertebrate prey (amphipods, isopods, shrimp and crab; Cottam and Knappen 1939). Other diet items included Pacific sanddab (*Citharichthys sordidus*) from a bird near Baja, California (Jehl 1970), and sculpin from birds near Norway (Collett 1894). Stickleback and salmonids were found in a bird in Russia, but it was unclear whether this loon was captured in a marine or freshwater system (Uspenskii 1969; cited in North 1994).

To our knowledge, no published information documents the diet of yellow-billed loons during the nesting season. In western North America, loons migrate from marine wintering areas to the Chukchi and Beaufort Sea coasts of Alaska in late May/early June and stage in coastal waters until ice on inland freshwater lakes begins to break up. Loons move onto lakes and begin nesting soon after open water is available. During the breeding season, yellow-billed loons forage primarily on their nesting lake (North 1994, 2008; J.A. Schmutz unpublished data); therefore, nesting pairs must select lakes that meet habitat requirements and contain adequate food resources for successful reproduction. The availability of suitable prey is likely a major constraint on reproductive success for yellow-billed loons, given the extra energetic demands of the breeding season, including defending a territory, brooding eggs, and feeding and caring for chicks (Barr 1996).

It is difficult to infer diet of yellow-billed loons based on our current understanding of other loon species. The common loon forages primarily on fishes and also on a variety of crustaceans and other aquatic animals (Barr 1996). However, common and yellow-billed loon distributions do not overlap during the breeding season; each species inhabits environments with different available prey, limiting the parallels that can be drawn regarding diet. On the Arctic Coastal Plain of Alaska (hereafter, Arctic Coastal Plain), the distribution of yellow-billed loons is sympatric with red-throated (*Gavia stellata*) and Pacific (*Gavia pacifica*) loons; however, these two species are much smaller in body size and have different ecological niches (e.g., Haynes et al. 2014b, c). Red-throated loons forage almost entirely on coastal marine fishes, whereas Pacific loons principally consume invertebrates from small tundra lakes (Barr et al. 2000; Russell 2002; Rizzolo and Schmutz 2009).

Loons are visual pursuit divers that capture and, most often, consume prey underwater. This behavior makes direct observation of prey consumed by adult yellow-billed loons logistically infeasible. Observation of parents feeding their young is also difficult because adults and chicks avoid human presence and are highly mobile on large lakes. Given

these limitations, indirect methods provide the most tractable way to estimate loon diet given the impracticality of collecting the entire animal. Recent advances in molecular methods have made it possible for researchers to investigate the diet of species for which direct observation of feeding or lethal collection of stomach contents is not possible (Barrett et al. 2007). Indirect molecular methods may require capture of individual predators and potential prey for collection of tissue samples, but are generally non-lethal for the predators (Dunsha 2009). Here, we estimate the diet of yellow-billed loons breeding on the Arctic Coastal Plain using Quantitative Fatty Acid Signature Analysis (QFASA; Iverson et al. 2004). QFASA models estimate the proportion of potential prey items in an individual predator's diet using the fatty acid (FA) signatures from the predator and the potential prey tissues, while accounting for FA metabolism by the predator (e.g., Iverson et al. 2004, 2007; Beck et al. 2007; Wang et al. 2010; Bromaghin et al. 2013). By estimating diet, this study provides context for yellow-billed loon breeding ecology, including how the spatial distributions of nesting loons may be influenced by prey distributions across the landscape.

Methods

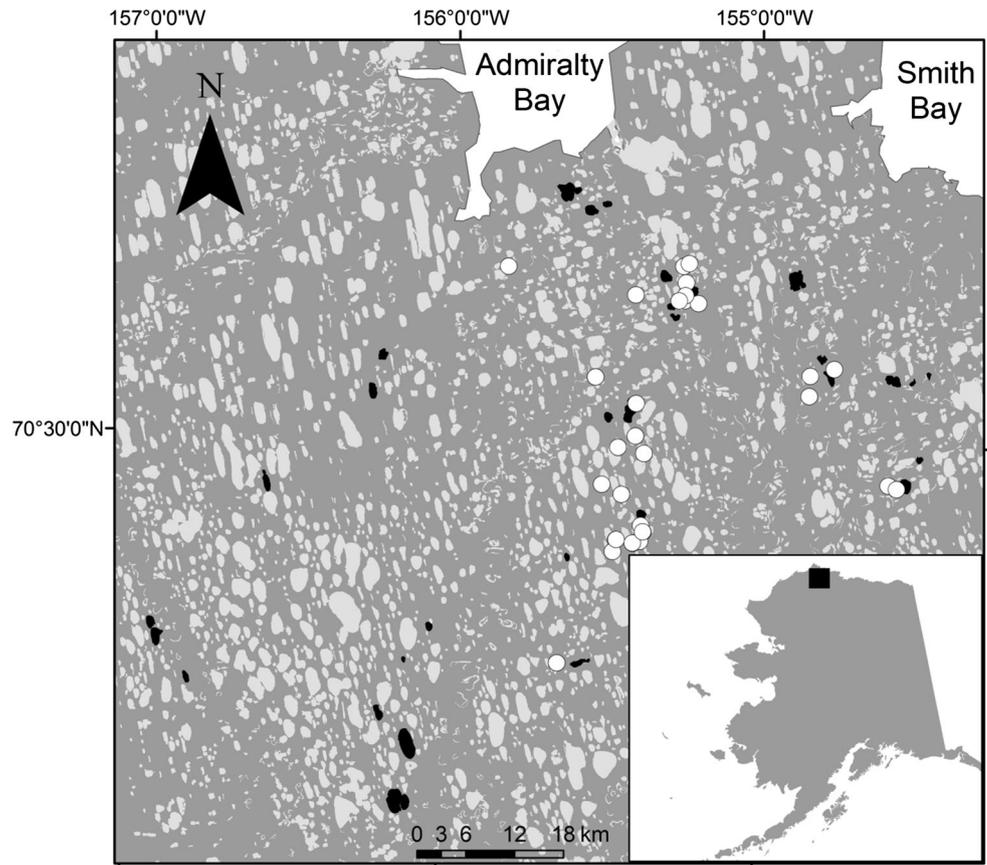
Prey tissue collection

We collected potential prey (fish and invertebrates) at 32 lakes over an area of the Arctic Coastal Plain covering 9000 km² (Fig. 1). We sampled prey over three summer seasons (2009–2011) using multiple sampling techniques, including fyke nets, minnow traps, gill nets, dip nets and beach seines (see Haynes et al. 2013 for details). We euthanized fish by placing them in 2 L of lake water with carbon dioxide tablets for approximately 10 min, and then pithed individuals to ensure they would not revive. We wrapped prey in aluminum foil, placed the samples in vacuum pump plastic bags (Ziploc[®] brand Vacuum Freezer System) and used the vacuum pump to reduce the air volume in the sample bag. We placed samples in a cooler and transported them from the remote field location to a –20 °C freezer (within 24 h).

Loon tissue collection

We captured 26 loons from 23 lakes for tissue collection (Fig. 1). Twenty-two loons were captured with a bow-net (Salyer 1962), while they incubated eggs. Four loons were captured during the nesting period with a lift-net (Kenow et al. 2009) or suspended dive net (Uher-Koch et al. 2015) set away from the nest. A decoy loon and broadcasted

Fig. 1 Locations where yellow-billed loons (*Gavia adamsii*) were captured for tissue sampling (*white circles*) and lakes sampled for prey (*black polygons*) on the Arctic Coastal Plain, Alaska. *Inset* shows the study region (*black square*) relative to the state of Alaska



recordings of loon calls lured the loon to the lift-net or suspended dive net area. For 19 of these captured loons, we used a local anesthetic, scalpel and forceps to remove a subcutaneous fat sample near and lateral from the uropygial gland, a common approach for avian species (Owen et al. 2010). The other seven samples were obtained from loons that underwent a surgery to implant a satellite transmitter (Korschgen et al. 1996). For these seven loons, a sample of subcutaneous fat was obtained from the abdominal area after the abdominal wall was cut open in preparation for implanting the transmitter. Previous studies have shown that FA composition across various adipose depots within birds is uniform (Iverson et al. 2007). All fat samples were placed in vials containing chloroform, in accordance with Budge et al. (2006), transported to a laboratory and stored at -80°C until analysis.

Tissue preparation

Prey were allowed to partially thaw and then homogenized using a Kinematica GmbH tissue homogenizer (Brinkmann Instruments, Switzerland) for small fish (<250 mm fork length) and a blender with a glass jar (Oster, Sunbeam

Products, Boca Raton, FL, USA) for large fish (>250 mm). Processing the entire fish for FA, including stomach contents, is common in studies of predators that eat whole fish given that it better reflects the FA signature of prey consumed by the predator (Budge et al. 2002; Iverson et al. 2002). We did not remove stomach contents of individual fish because we aimed to characterize their FA signatures as prey for loons. When an individual's mass was <1 g, prey of the same species from the same lake were homogenized together to form composite samples. Approximately 1 g of homogenized prey tissue was subsampled and stored in a glass vial with 4.5 mL of chloroform at -80°C until analysis.

FA analysis

FA methyl esters were prepared using acidic transesterification (Budge et al. 2006; Wang et al. 2010). Thin-layer chromatography indicated the presence of fatty alcohols in diet items resulting from the transesterification of wax esters. Wax ester alcohols were converted to their respective FAs (Budge et al. 2006) to account for wax esters in diets (Budge and Iverson 2003). FA methyl esters were

quantified using temperature-programmed gas liquid chromatography on a Varian Autosystem II Capillary FID gas chromatograph fitted with a 30 m by 0.25 mm (in diameter) column coated with 50 % cyanopropylmethylpolysiloxane (DB-23) and linked to a computerized integration system (Varian Galaxie software; Iverson et al. 2002). Each chromatogram was manually assessed for correct peak identification and integration of peak areas.

We analyzed the diet of yellow-billed loons based on FAs using a QFASA package in program R (Iverson et al. 2004). We used three sets of calibration coefficients determined from feeding trials of captive common murres (*Uria aalge*) fed natural fish diets (Iverson et al. 2007), and spectacled eiders (*Somateria fischeri*) and Steller's eiders (*Polysticta stelleri*) fed primarily a "sea duck formula" supplemented with 5 % marine prey (Wang et al. 2010). We used the extended dietary subset of FAs, which includes 33 FAs that can only be acquired through diet and eight additional FAs that are indicative of diet but can also be biosynthesized by predators (Iverson et al. 2004). We used prey-on-prey simulations to determine the degree to which each prey species can be identified based on their FA signature (Iverson et al. 2004). If a prey item has a FA signature similar to other prey item types (i.e., different species), then QFASA may incorrectly estimate dietary proportions of those prey species in the predator's diet. The prey-on-prey analysis is an iterative process (in our case, performed 100 times; Wang et al. 2010), where the prey data are randomly split into two sets for each prey item type—a set that acts as the prey data and a set that is modeled as the predator data without calibration coefficients (Iverson et al. 2004). Because calibration coefficients are not used, the FA signature of the prey item subset used as the predator diet should most closely reflect the subset of the same prey item type. With overlap in the FA signatures of different prey items, the prey-on-prey analysis can reveal the prey item types which QFASA has difficulty in discriminating among. Because prey samples require splitting to run the simulation, we removed prey item types that had sample sizes that were too low ($N < 3$ samples) to create useful splits. This led to the removal of burbot (*Lota lota*; $N = 2$) and northern pike (*Esox lucius*; $N = 1$) samples. Means are presented with \pm SD.

Results

We collected samples from eleven fish species: Alaska blackfish (*Dallia pectoralis*), arctic flounder (*Liopsetta glacialis*), arctic grayling (*Thymallus arcticus*), broad whitefish (*Coregonus nasus*), burbot, least cisco (*Coregonus sardinella*), ninespine stickleback (*Pungitius pungitius*), northern pike, rainbow smelt (*Osmerus mordax*),

slimy sculpin (*Cottus cognatus*) and three-spined stickleback (*Gasterosteus aculeatus*). We also collected samples from two invertebrate orders: amphipods (order Amphipoda) and fairy shrimp (order Anostraca; Table 1). FA values for individual loons and for pooled prey are provided in Online Resource 1.

FA analysis

Prey-on-prey simulation results suggested that the FA signatures of prey items could be reasonably well distinguished using QFASA (mean classification accuracy = 85 ± 17 %). However, classification accuracy was low for least cisco (61 ± 20 %) and slimy sculpin (65 ± 9 %; Table 2; Online Resource 2). The low classification accuracy for these two species suggests overlap/similarity in FA signatures with other diet items, and thus, difficulty in discriminating least cisco and slimy sculpin in the QFASA model.

Results differed substantially between models using the calibration coefficients from common murres as compared to calibration coefficients from either eider species, which were similar to each other (Meynier et al. 2010; Budge et al. 2012; Fig. 2). The estimate of diet based on FA analysis included 129 prey samples from the 13 prey types (9.9 ± 6.3 samples per type). The model using the common murre calibration coefficients estimated that Alaska blackfish, broad whitefish and three-spined stickleback made up the majority of the yellow-billed loon diet (Fig. 2), although analysis indicated variation among individuals (Table 3). Yellow-billed loons exhibited a high diversity in diet at a population level, but much less diversity at an individual level. The diet patterns of individual loons suggested many individuals fed primarily on a single species such as Alaska blackfish.

Discussion

Given the lack of loon-specific calibration coefficients, we recognize that our results are sensitive to the choice of calibration coefficients (Meynier et al. 2010). Ideally, we would have used calibration coefficients determined for yellow-billed loons, but loon-specific coefficients have not yet been measured. Species-specific coefficients require controlled feeding studies on captive animals to generate, and thus, calibration coefficients are often limited in availability (Iverson et al. 2004, 2007; Meynier et al. 2010). We chose the common murre calibration coefficient model to interpret results—in part because of evolutionary and ecological similarities—loons are taxonomically more closely related to murres than eiders and both yellow-billed loons and murres are piscivores. But more importantly, the

Table 1 Prey samples collected from lakes on the Arctic Coastal Plain, Alaska, for dietary study of yellow-billed loons (*Gavia adamsii*)

Common names	Scientific names	Abbreviations	Sample sizes
Alaska blackfish	<i>Dallia pectoralis</i>	ALBL	20
Arctic flounder	<i>Liopsetta glacialis</i>	ARFL	5
Arctic grayling	<i>Thymallus arcticus</i>	ARGR	15
Broad whitefish	<i>Coregonus nasus</i>	BRWH	5
Burbot	<i>Lota lota</i>	BURB	2
Least cisco	<i>Coregonus sardinella</i>	LECI	17
Ninespine stickleback	<i>Pungitius pungitius</i>	NIST	15
Northern pike	<i>Esox lucius</i>	NOPI	1
Rainbow smelt	<i>Osmerus mordax</i>	RASM	14
Slimy sculpin	<i>Cottus cognatus</i>	SLSC	7
Three-spined stickleback	<i>Gasterosteus aculeatus</i>	THST	3
Amphipods	Order Amphipoda	AMPH	20
Fairy shrimp	Order Anostraca	FAIR	5

Table 2 Results from prey-on-prey simulations using Quantitative Fatty Acid Statistical Analysis (QFASA) methods

Species	ALBL	AMPH	ARFL	ARGR	BRWH	FAIR	LECI	NIST	RASM	SLSC	THST
ALBL	85.7	0.1	0.7	1.8	3.4	2.1	1.6	1.4	2.7	0.3	0.2
AMPH	0.2	81.7	1.9	0.1	2.9	9.1	0.0	0.1	0.6	3.4	0.0
ARFL	0.0	0.0	100	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
ARGR	0.3	0.3	0.0	82.6	0.7	4.0	7.1	0.8	1.7	2.3	0.0
BRWH	0.0	0.0	0.0	0.0	100	0.0	0.0	0.0	0.0	0.0	0.0
FAIR	0.0	0.0	0.0	0.0	0.0	100	0.0	0.0	0.0	0.0	0.0
LECI	6.7	0.2	0.0	9.6	0.6	1.3	60.6	1.1	19.9	0.1	0.0
NIST	6.4	1.9	0.4	2.2	1.1	2.9	1.3	79.1	1.7	2.9	0.1
RASM	0.1	0.3	0.3	0.1	0.1	0.3	4.6	0.3	93.7	0.2	0.2
SLSC	3.2	8.9	2.7	3.1	1.6	8.4	0.0	2.7	4.2	64.9	0.2
THST	0.1	0.3	1.9	0.0	1.3	0.1	0.1	0.0	8.8	0.1	87.3

The prey-on-prey analysis is an iterative process (performed 100 times in our case), where the prey data are randomly split into two sets for each prey type—a set that acts as the prey data and a set that is modeled as the predator data without calibration coefficients (Iverson et al. 2004). The simulation output can be used to determine how well QFASA can distinguish among prey item types based on how well QFASA correctly classifies the FA signatures of each prey type. The diagonal of the table represents the probability of correctly classifying a species

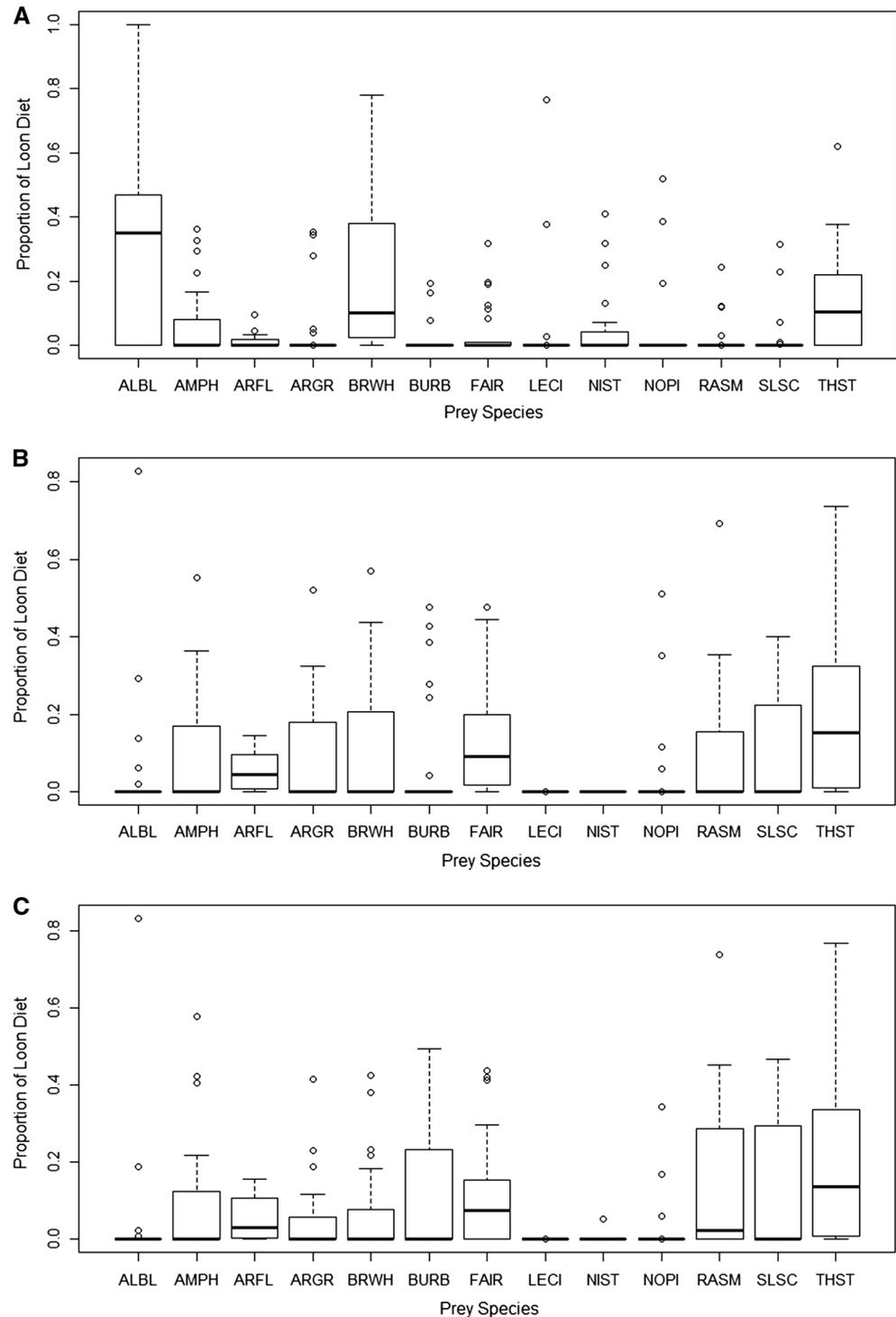
ALBL Alaska blackfish (*Dallia pectoralis*), *AMPH* amphipod species, *ARFL* Arctic flounder (*Liopsetta glacialis*), *ARGR* Arctic grayling (*Thymallus arcticus*), *BRWH* broad whitefish (*Coregonus nasus*), *FAIR* fairy shrimp species, *LECI* least cisco (*Coregonus sardinella*), *NIST* ninespine stickleback (*Pungitius pungitius*), *RASM* rainbow smelt (*Osmerus mordax*), *SLSC* slimy sculpin (*Cottus cognatus*), *THST* three-spined stickleback (*Gasterosteus aculeatus*)

“sea duck formula” which comprised the majority of diets fed to eiders was actually corn based and thus high in carbohydrates and low in important common marine FAs (Wang et al. 2010). Consumption of carbohydrates is extremely uncommon in piscivores or zooplanktivores and would lead to significantly greater deposition of FAs arising from biosynthesis and thus result in incomparable/un-natural calibration coefficients. Although the sensitivity of our analysis to the calibration coefficients used was likely due primarily to the unusual carbohydrate formula fed in the two captive eider experiments, it also suggests that diet

estimates would be improved with loon-specific calibration coefficients.

Understanding the diet of yellow-billed loons has been hampered by the paucity of studies that have examined loon diet and by their large potential prey base; depending on the time of year, yellow-billed loon diet could potentially include both marine and fresh water species. Based on the FA model using the common murre calibration coefficient, the most prominent prey items in yellow-billed loon diets were Alaska blackfish, three-spined stickleback and broad whitefish. Alaska blackfish

Fig. 2 Mean diet estimate of 26 yellow-billed loons (*Gavia adamsii*) from the Arctic Coastal Plain, Alaska, based on fatty acid analysis of yellow-billed loon and prey tissues using common murre (*Uria aalge*; **a**), Steller's eider (*Polysticta stelleri*; **b**) and spectacled eider (*Somateria fischeri*; **c**) calibration coefficients. Boxes represent first and third quartiles around the mean (thick black line), whiskers represent the range (the interquartile range multiplied by 1.5) and the dots represent outliers (falling outside the range). Estimates of the proportions of 13 prey items (sources) in the loon diet were derived using a Quantitative Fatty Acid Signature Analysis (QFASA; Iverson et al. 2004). See Table 1 for species abbreviations



is one of the most common freshwater species on the Arctic Coastal Plain, occupying about 75 % of lakes >7 ha in surface area (Haynes et al. 2014a). Also, blackfish are tolerant to harsh overwintering conditions (Scholander et al. 1953) and persist over winter months (Haynes et al. 2014a). Thus, blackfish are likely available at the beginning of the season when other species are

either still moving into summering lakes (e.g., least cisco) or recovering from winter die-offs (e.g., ninespine stickleback; Haynes et al. 2014a). Although blackfish are generally not locally abundant, their availability soon after breakup may make them an important food source during the early stages of breeding for yellow-billed loons, when other prey species are less available.

Table 3 Diet estimates of individual yellow-billed loons (*Gavia adamsii*) based on a Quantitative Fatty Acid Statistical Analysis using calibration coefficients from common murre (*Uria aalge*; Iverson et al. 2007)

Prey species														
Individual loon	ALBL	AMPH	ARFL	ARGR	BRWH	BURB	FAIR	LECI	NIST	NOPI	RASM	SLSC	THST	Number of prey types in diet (>5 %)
1	0.03	0.00	0.00	0.00	0.59	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.38	2
2	0.00	0.29	0.00	0.00	0.39	0.00	0.01	0.00	0.00	0.00	0.00	0.00	0.30	3
3	0.53	0.07	0.02	0.00	0.22	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.16	4
4	0.40	0.17	0.02	0.00	0.33	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.09	4
5	0.00	0.00	0.01	0.35	0.03	0.00	0.20	0.38	0.04	0.00	0.00	0.00	0.00	3
6	0.00	0.01	0.00	0.34	0.10	0.00	0.32	0.00	0.00	0.00	0.00	0.23	0.00	4
7	0.00	0.00	0.00	0.00	0.00	0.00	0.11	0.77	0.00	0.00	0.12	0.00	0.00	3
8	0.00	0.00	0.00	0.28	0.00	0.00	0.19	0.00	0.41	0.00	0.00	0.00	0.12	4
9	0.59	0.36	0.00	0.00	0.02	0.00	0.00	0.00	0.03	0.00	0.00	0.00	0.00	2
10	0.50	0.00	0.00	0.00	0.00	0.08	0.00	0.03	0.13	0.19	0.03	0.00	0.04	4
11	0.00	0.00	0.00	0.00	0.67	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.33	2
12	0.40	0.00	0.02	0.00	0.00	0.00	0.00	0.00	0.00	0.52	0.03	0.00	0.03	2
13	0.14	0.07	0.00	0.00	0.65	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.15	4
14	0.00	0.00	0.00	0.00	0.78	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.22	2
15	0.36	0.22	0.01	0.00	0.14	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.27	4
16	0.72	0.08	0.00	0.05	0.10	0.00	0.00	0.00	0.04	0.00	0.00	0.00	0.00	3
17	0.34	0.00	0.00	0.04	0.07	0.00	0.01	0.00	0.32	0.00	0.00	0.07	0.15	5
18	0.47	0.00	0.00	0.00	0.05	0.16	0.00	0.00	0.07	0.00	0.24	0.00	0.00	4
19	0.37	0.00	0.03	0.00	0.04	0.00	0.00	0.00	0.00	0.38	0.12	0.00	0.06	4
20	0.41	0.08	0.05	0.00	0.18	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.29	4
21	0.00	0.00	0.09	0.00	0.02	0.19	0.13	0.00	0.25	0.00	0.00	0.31	0.00	5
22	0.19	0.33	0.01	0.00	0.29	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.18	4
23	0.00	0.00	0.00	0.00	0.38	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.62	2
24	0.68	0.08	0.03	0.00	0.06	0.00	0.00	0.00	0.00	0.00	0.00	0.01	0.14	4
25	1.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	1
26	0.38	0.07	0.00	0.00	0.46	0.00	0.08	0.00	0.00	0.00	0.00	0.00	0.00	4
Frequency in diet (>5 %)	0.62	0.42	0.04	0.12	0.65	0.12	0.23	0.08	0.19	0.12	0.12	0.12	0.62	

Frequency in diet is the frequency that the prey species were found to make up >5 % of the diet of individuals in the sample loon population ($N = 26$). Number of prey types in diet was calculated as the total number of prey that made up >5 % of an individual's diet

ALBL Alaska blackfish (*Dallia pectoralis*), AMPH amphipod species, ARFL Arctic flounder (*Liopsetta glacialis*), ARGR Arctic grayling (*Thymallus arcticus*), BRWH broad whitefish (*Coregonus nasus*), BURB burbot (*Lota lota*), FAIR fairy shrimp species, LECI least cisco (*Coregonus sardinella*), NIST ninespine stickleback (*Pungitius pungitius*), NOPI northern pike (*Esox lucius*), RASM rainbow smelt (*Osmerus mordax*), SLSC slimy sculpin (*Cottus cognatus*), THST three-spined stickleback (*Gasterosteus aculeatus*)

We initially found it surprising that broad whitefish and three-spined stickleback were identified as dominant prey items given they are not very common in lakes on the Arctic Coastal Plain. Broad whitefish have a relatively low occupancy probability in lakes (found in about a third of lakes large enough for breeding yellow-billed loons), and three-spined stickleback have a distribution restricted to brackish lakes and coastal waters, with an extremely low

occupancy probability further inland (Craig 1984; Haynes et al. 2014a). However, because we collected tissues from yellow-billed loons two to four weeks after they first arrived on the breeding lakes, the dominance of broad whitefish and three-spined stickleback in the diet may reflect a residual signal in the loon tissue from marine or brackish waters before they moved inland to choose nest sites. During spring, before ice cover has melted and inland

lakes are available for nesting, loons feed along the coast or on coastal brackish lakes and ponds. Both broad whitefish and three-spined stickleback inhabit coastal waters and are more commonly found in lakes closer to the coastline (Haynes et al. 2014a) and in estuarine or brackish pond habitat occupied by staging loons prior to the breeding (Craig 1984; Reist and Bond 1988). When lakes have thawed to the point that they are accessible to loons (a ring of unfrozen water forms around the lake perimeter; North 1994) loons will migrate from staging areas to breeding lakes, behaviorally establish ownership of a lake, and commence mating and nest building. Because of the gradual turnover of tissues (Wang et al. 2010), the FA signature during this late staging period in coastal areas may persist in fat samples gathered a few weeks later during mid-incubation.

We were also surprised that, based on the FA diet estimates, ninespine stickleback and least cisco were not important prey items. Ninespine stickleback is the most widespread fish species in the region (94 % lake occupancy rate; Haynes et al. 2014a), and yellow-billed loons have been noted to feed stickleback to their young (J.A. Schmutz, unpublished data). However, during spring freshet, shortly after breakup, ninespine stickleback may be less available to loons compared to later in the season. During early spring, ninespine stickleback may have restricted distributions due to winter die-off, be more dispersed because of increased movement into flooded waterways, and less abundant early in the season because the first cohort of young-of-the-year stickleback is not present (Haynes et al. 2014a).

Least cisco is the most widely distributed large-bodied fish species in the region (occupancy probability >50 % of lakes <7 ha; Haynes et al. 2014a) and are abundant in various size classes. Despite its apparent suitability as a prey item, least cisco was only estimated to be consumed for two individual loons (Table 3). However, least cisco had the highest misclassification rate based on the prey-on-prey simulations, so it is possible that the proportion of least cisco in yellow-billed loon diet may be underestimated due to overlap with other species FA signatures. Alternatively, a low prevalence in diet may again be explained by the timing of the loon sampling. Early in the season, least cisco migrate from overwintering habitat to summer feeding lakes (Haynes et al. 2014a), and thus, likely have not arrived en masse before we sampled the loon tissues. Because of their high energy content (Ball et al. 2007) and high occupancy probability and abundance in lakes after June, we expect that least cisco plays a more important role in diet of loons later in the season.

Yellow-billed loons may be opportunistic foragers, feeding on the prey available at a given location at that point of the season. This is demonstrated by the variation in diets which included prey species associated with marine

or brackish waters and strictly freshwater species. Individual variation in diet is common in aquatic predators and is only partially related to the sex or age of the individual (e.g., Bolnick et al. 2003; Woo et al. 2008). In this study, individual variation in diet likely explains the variation in QFASA signatures (Bromaghin et al. 2013). For example, if yellow-billed loons nest on a lake with only Alaska blackfish in high availability, these breeders would be required to feed on blackfish. The ability of yellow-billed loons to opportunistically feed on available prey likely buffers individuals to temporal variation in prey availability (e.g., feeding on coastal species before lakes become ice-free or feeding on blackfish before migratory prey species become available in lakes).

It is clear from this study that a strong understanding of the prey ecology is important when investigating the diet of a predator. Knowledge of seasonal and spatial variation in both loon (Schmutz et al. 2014) and fish distributions (Haynes et al. 2014a) was the key in interpreting diet estimates. This information not only allowed us to determine what species are important, but also why they are important. Alaska blackfish may not be as abundant or calorie rich as other fish prey throughout the season; however, its widespread distribution in spring is likely what makes it a main prey item for loons arriving on lakes. Broad whitefish and three-spined stickleback are geographically restricted and have a low occupancy in inland lakes; however, these species are likely targeted in coastal staging areas before lake territories are formed. This finding highlights that understanding diet not only requires dietary models, but also an understanding of prey ecology and distributional patterns.

Acknowledgments Laboratory preparation and analysis were possible thanks to the assistance of J. Morehouse, B. Baker, S. Merrill, the Causey lab, S. Al-Shaghay and the Iverson lab. Field data were collected by T. Buckley, E. Dykstra, S. Furusawa, I. Isler, L. Koloski, C. Margolin, J. McCarter, K. Overdjuin, B. Price, T. Shoemaker, S. Stortz, A. Tate, B. Uher-Koch and K. Wright. This project was funded by a State Wildlife Grant awarded through Alaska Department of Fish & Game and the U.S. Geological Survey's Changing Arctic Ecosystem Initiative, supported, in part, by the Wildlife Program of the USGS Ecosystem Mission Area. S. Wang made helpful suggestions during our analysis stage. J. Pearce, M. Lindberg, K. Titus, and two anonymous reviewers provided useful reviews that improved the manuscript. TBH was funded by a postgraduate scholarship provided by the Natural Sciences and Engineering Research Council of Canada (NSERC) and a Changing Alaska Science Education graduate fellowship provided by the National Science Foundation. Additional support was provided by NSERC research and equipment grants to SJI. Any use of trade, firm or product names is for descriptive purposes only and does not imply endorsement by the U.S. Government.

References

- Bailey AM (1922) Notes on the yellow-billed loon. *Condor* 24:204–205

- Ball J, Esler D, Schmutz J (2007) Proximate composition, energetic value, and relative abundance of prey fish from the inshore eastern Bering Sea: implications for piscivorous predators. *Polar Biol* 30:699–708
- Barr JF (1996) Aspects of common loon (*Gavia immer*) feeding biology on its breeding ground. *Hydrobiologia* 321:119–144
- Barr JF, Eberl C, McIntyre JW (2000) Red-throated Loon (*Gavia stellata*). The Birds of North America Online (A. Poole, Ed.). Ithaca: Cornell Lab of Ornithology. Retrieved from the Birds of North America Online: <http://bna.birds.cornell.edu/bna/species/513>
- Barrett RT, Camphuysen K, Anker-Nilssen T, Chardine JW, Furness RW, Garthe S, Huppoo O, Leopold MF, Montevecchi WA, Veit RR (2007) Diet studies of seabirds: a review and recommendations. *ICES J Mar Sci* 64:1675–1691
- Beck CA, Iverson SJ, Bowen WD, Blanchard W (2007) Sex differences in grey seal diet reflect seasonal variation in foraging behaviour and reproductive expenditure: evidence from quantitative fatty acid signature analysis. *J Anim Ecol* 76:490–502
- Bolnick DI, Svanbäck R, Fordyce JA, Yang LH, Davis JM, Hulsey CD, Forister ML (2003) The ecology of individuals: incidence and implications of individual specialization. *Am Nat* 161:1–28
- Bromaghin JF, Lance MM, Elliott EW, Jeffries SJ, Acevedo-Gutiérrez A, Kennish JM (2013) New insights into the diets of harbor seals (*Phoca vitulina*) in the Salish Sea revealed by analysis of fatty acid signatures. *Fish Bull* 111:13–26
- Budge SM, Iverson SJ (2003) Quantitative analysis of fatty acid precursors in marine samples direct conversion of wax ester alcohols and dimethylacetals to FAMES. *J Lipid Res* 44:1802–1807
- Budge SM, Iverson SJ, Bowen WD, Ackman RG (2002) Among- and within-species variability in fatty acid signatures of marine fish and invertebrates on the Scotian Shelf, Georges Bank, and southern Gulf of St. Lawrence. *Can J Fish Aquat Sci* 59:886–898
- Budge SM, Iverson SJ, Koopman HN (2006) Studying trophic ecology in marine ecosystems using fatty acids: a primer on analysis and interpretation. *Mar Mamm Sci* 22:759–801
- Budge SM, Penney SN, Lall SP (2012) Estimating diets of Atlantic salmon (*Salmo salar*) using fatty acid signature analyses: validation with controlled feeding studies. *Can J Fish Aquat Sci* 69:1033–1046
- Collett P (1894) On the occurrence of *Colymbus adamsi* in Norway. *Ibis* 36:269–283
- Cottam C, Knappen P (1939) Food of some uncommon North American birds. *Auk* 56:138–169
- Craig PC (1984) Fish use of coastal waters of the Alaskan Beaufort Sea: a review. *Trans Am Fish Soc* 113:265–282
- Dunshie G (2009) DNA-based diet analysis for any predator. *PLoS ONE* 4:e5252
- Haynes TB, Rosenberger AE, Lindberg MS, Whitman M, Schmutz JA (2013) Method- and species-specific detection probabilities of fish occupancy in Arctic lakes: implications for design and management. *Can J Fish Aquat Sci* 70:1055–1062
- Haynes TB, Rosenberger AE, Lindberg MS, Whitman M, Schmutz JA (2014a) Patterns of lake occupancy by fish indicate different adaptations to life in a harsh Arctic environment. *Freshw Biol* 59:1884–1896
- Haynes TB, Schmutz JA, Lindberg MS, Rosenberger AE (2014b) Risk of predation and weather events affect nest site selection by sympatric Pacific (*Gavia pacifica*) and yellow-billed (*Gavia adamsii*) loons in Arctic habitats. *Waterbirds* 37:16–25
- Haynes TB, Schmutz JA, Lindberg MS, Wright KG, Uher-Koch BD, Rosenberger AE (2014c) Occupancy of yellow-billed and Pacific loons: evidence for interspecific competition and habitat mediated co-occurrence. *J Avian Biol* 45:296–304
- Iverson SJ, Frost KJ, Lang S (2002) Fat content and fatty acid composition of forage fish and invertebrates in Prince William Sound, Alaska: factors contributing to among and within species variability. *Mar Ecol Prog Ser* 241:161–181
- Iverson SJ, Field C, Bowen WD, Blanchard W (2004) Quantitative fatty acid signature analysis: a new method of estimating predator diets. *Ecol Monogr* 74:211–235
- Iverson SJ, Springer AM, Kitaysky AS (2007) Seabirds as indicators of food web structure and ecosystem variability: qualitative and quantitative diet analyses using fatty acids. *Mar Ecol Prog Ser* 352:235
- Jehl JR Jr (1970) A Mexican specimen of the yellow-billed loon. *Condor* 72:376
- Kenow KP, Wilson JM, Meyer MW (2009) Capturing common loons during prenesting and nesting periods. *J Field Ornithol* 80:427–432
- Korschgen CE, Kenow KP, Gendron-Fitzpatrick A, Green WL, Dein FJ (1996) Implanting intra-abdominal radiotransmitters with external whip antennas in ducks. *J Wildl Manag* 60:132–137
- Meynier L, Morel PCH, Chilvers BL, Mackenzie DDS, Duignan PJ (2010) Quantitative fatty acid signature analysis on New Zealand sea lions: model sensitivity and diet estimates. *J Mamm* 91:1484–1495
- North MR (1994) Yellow-billed loon (*Gavia adamsii*). The Birds of North America Online (A. Poole, Ed.). Ithaca: Cornell Lab of Ornithology; Retrieved from the Birds of North America Online: <http://bna.birds.cornell.edu/bna/species/121>
- North MR (2008) Activity budgets and habitat use by yellow-billed loons in Alaska. Big Deep Lake Publishing, Pillager
- Owen E, Daunt F, Wanless S (2010) Sampling avian adipose tissue: assessing a nondestructive biopsy technique. *J Field Ornithol* 81:92–98
- Reist JD, Bond WA (1988) Life history characteristics of migratory coregonids of the lower Mackenzie River, Northwest Territories, Canada. *Finn Fish Res* 9:133–144
- Rizzolo DJ, Schmutz JA (2009) Monitoring marine birds of concern in the Eastern Chukchi Nearshore Area (Loons). Annual Report for 2009 to Minerals Management Service, Alaska Region, p 55
- Russell RW (2002) Pacific Loon: *Gavia pacifica*: Arctic Loon: *Gavia arctica*. The Birds of North America Online (A. Poole, Ed.). Ithaca: Cornell Lab of Ornithology; Retrieved from the Birds of North America Online: <http://bna.birds.cornell.edu/bna/species/657a>
- Salyer JW (1962) A bow-net trap for ducks. *J Wildl Manag*:219–221
- Schmutz JA, Wright KG, DeSorbo CR, Fair J, Evers DC, Uher-Koch BD, Mulcahy DM (2014) Size and retention of breeding territories of yellow-billed loons (*Gavia adamsii*) in Alaska and Canada. *Waterbirds* 37:53–63
- Scholander PF, Flagg W, Hock RJ, Irving L (1953) Studies on the frozen plants and animals in the Arctic. *J Cell Comp Physiol* 42:1–56
- Sih A, Christensen B (2001) Optimal diet theory: when does it work, and when and why does it fail? *Anim Behav* 61:379–390
- Uher-Koch BD, Schmutz JA, Wright KG (2015) Nest visits and capture events affect breeding success of Yellow-billed and Pacific loons. *Condor* 11:121–129
- Uspenskii SS (1969) Life in high latitudes, a study of bird life. Transl. from Russ. by Amerind Publ. Co. Pvt. Ltd., New Delhi, for U.S. Dept. Int. and Natl. Sci. Found. Washington, D.C
- Wang S, Hollmén T, Iverson S (2010) Validating quantitative fatty acid signature analysis to estimate diets of spectacled and Steller's eiders (*Somateria fischeri* and *Polysticta stelleri*). *J Comp Physiol B* 180:125–139
- White TCR (1978) The importance of a relative shortage of food in animal ecology. *Oecologia* 33:71–86
- Wiens JA, Rotenberry JT (1979) Diet niche relationships among North American grassland and shrubsteppe birds. *Oecologia* 42:253–292
- Woo KJ, Elliott KH, Davidson M, Gaston AJ, Davoren GK (2008) Individual specialization in diet by a generalist marine predator reflects specialization in foraging behaviour. *J Anim Ecol* 77:1082–1091