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농학석사 학위논문

Food-niche Partition and Sexual Dimorphism of
Northern Boobooks (*Ninox japonica*) and
Oriental Scops Owls (*Otus sunia*) in Korea
한국에서 서식하는 소쩍새 (*Otus sunia*) 와
솔부엉이 (*Ninox japonica*) 의
성적 이형성과 먹이-니치(niche) 연구

2015 년 8 월

서울대학교 대학원
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이 논문을 농학석사 학위논문으로 제출함
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Abstract

Two small forest owls, the Oriental Scops Owl (*Otus sunia*) and the Northern Boobook (*Ninox japonica*), are among the common breeding raptors in the Republic of Korea. However, very limited ecological information is available on these nocturnal raptors. In this study, I focused on two aspects of these owls' ecology: sexual dimorphism and food-niche partitioning during their early breeding period. The morphometric study was aimed to understand sexual dimorphism in these two small owls through external measurements, and to develop sex-discriminant functions for both species. The study of food-niche partitioning of two sympatric owls used stable isotope analysis (SIA) to determine trophic levels and estimate prey type selection. From both morphometric information and individual SIA results, I questioned how body size can effect individual prey use, and further its relationship with the evolution of reversed sexual dimorphism in raptors. From April to May in 2014 and 2015, 30 Oriental Scops Owls and 10 Northern Boobooks were captured using mist nets after sunset between 19:30 to 00:30 at Gwang-reung Forest and Gwan-ak Arboretum in Gyeonggi-do province. Blood samples and measurements were taken from captured owls. Moreover, 16 scops owls and 22 boobooks admitted to or kept in four wildlife rescue centers were measured and sampled for the morphometric study. For diet reconstruction, potential prey items (ground insects, moths, mice, and birds) were collected and analyzed to estimate the diet composition of each owl species through multi-source mixing model. In terms of morphology, Oriental Scops Owls showed significant but subtle reversed sexual dimorphism whereas, Northern Boobooks had no clear evidence of sexual dimorphism. Bill depth, bill width, tail length and tarsus length were selected for the stepwise discriminant function in Oriental Scops Owls, and they sexed of 79% the birds correctly by re-substitution. The stepwise discriminant function for Northern Boobooks using head length and bill length to skull, correctly classified the sexes with about 70% accuracy. Given the low power of morphometric sex discrimination as well as non-obvious sexual size dimorphism, additional techniques such as examination of brood patch or even molecular methods, are required for reliable sex determination. SIA showed

significant distinctions between the two species, both in carbon and nitrogen isotope ratios. Northern Boobooks had higher values in nitrogen and carbon isotope ratios, and greater variance in carbon isotope ratios than Oriental Scops Owls. According to the multi-source mixing models, Northern Boobooks mainly consumed vertebrate prey, especially birds, but Oriental Scops Owls utilized mainly ground insects followed by birds, mice, and moths. Each individual owl's overall body size index was extracted from principal component analysis, and compared with individual trophic level (nitrogen isotope ratio). Only Oriental Scops Owls showed significant correlation with smaller individuals consuming higher trophic level diets. In conclusion, Northern Boobooks and Oriental Scops Owls demonstrated distinct food niche separation in a sympatric habitat. In particular, the larger bodied Northern Boobooks occupied a higher trophic level with more flexible foraging habits, and they selected diverse prey sources from forests to open environments, whereas the smaller Oriental Scops Owl belonged to a lower trophic level and was a more specialized forager relying on forest ecosystems. Furthermore, individual trophic level was found to be related to the body size only for the sexually dimorphic Oriental Scops Owls, but not in the monomorphic Northern Boobooks. These findings partially support the 'small male hypothesis', in Oriental Scops Owls that smaller males are efficient foragers which is beneficial in reproductive success and thus more adaptive in raptors with reversed sexual dimorphism.

Keywords: diet reconstruction, discriminant function analysis, reversed sexual dimorphism, small male hypothesis, stable isotope analysis, trophic level

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Table of Contents

Abstract.....	i
Table of Contents.....	iii
List of Tables	v
List of Figures.....	vi
Chapter 1. Introduction.....	1
Chapter 2. Literature Review.....	4
2.1 Study species	4
2.2 Reversed sexual size dimorphism in raptors	6
2.3 Food-niche and diet studies in raptors.....	8
Chapter 3. Materials and Methods.....	11
3.1 Study sites and period.....	11
3.2 Sample collection.....	13
3.3 Data analysis	16
3.3.1 Molecular sex identification.....	16
3.3.2 Stable isotope analysis	17
3.3.3 Statistical analysis	19
Chapter 4. Results.....	22
4.1 Sampling and molecular sex identification.....	22
4.2 Morphology and sexual size dimorphism.....	23
4.2.1 Discriminant analysis	28
4.3 Stable isotope ecology	32
4.3.1 Stable isotope ratio analysis	32
4.3.2 Diet reconstruction of owls	35
4.3.3 Body size and trophic level	37

Chapter 5. Discussion	40
5.1 Sexual size dimorphism in owls	40
5.2 Trophic ecology of owls.....	43
5.3 Body size and trophic ecology	46
Chapter 6. Conclusions.....	48
References	50
Appendix	59
국문 초록	60

List of Tables

Table 1. Morphometric parameters of Oriental Scops Owls in the Republic of Korea. Asterisks indicate significant differences (* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$). CV and DI represent the coefficient of variance and the dimorphic index, respectively.	24
Table 2. Morphometric parameters of Oriental Scops Owls in the Republic of Korea. Asterisks indicate significant differences (* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$). CV and DI represent the coefficient of variance and the dimorphic index, respectively.	25
Table 3. Morphometric characteristics of Northern Boobooks in the Republic of Korea. Asterisks indicate significant differences (* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$). CV and DI represent the coefficient of variance and the dimorphic index, respectively.....	26
Table 4. Morphometric characteristics of Northern Boobooks in the Republic of Korea. Asterisks indicate significant differences (* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$). CV and DI represent the coefficient of variance and the dimorphic index, respectively.....	27
Table 5. Nitrogen and carbon stable isotope values for Oriental Scops Owls and Northern Boobooks.....	33
Table 6. Stable isotope values of potential prey items of Oriental Scops Owls and Northern Boobooks in the Republic of Korea.	34
Table 7. Results of principal component analysis of Oriental Scops Owls and Northern Boobooks (HC: hind claw length, BN: bill length to nostril, BS: bill length to skull, BW: bill width, BD: bill depth).....	38

List of Figures

Figure 1. The location of wildlife rescue centers and trapping sites. Numbers indicate following locations. 1: Gwang-reung Forest, 2: Gwanak Arboretum, 3: Gyeonggi Wildlife Rescue Center, 4: Ulsan Wildlife Rescue Center, 5: Gyungseong University Bird Museum, 6: Busan Wildlife Rescue Center.	12
Figure 2. Sample image of gel electrophoresis showing females (two bars) and males (single bar).	22
Figure 3. Histogram of discriminant scores (D) for Oriental Scops Owls.	28
Figure 4. Relationship between discriminant scores and posterior probability of being a male of Oriental Scops Owls. Dashed lines indicate 95%, 75%, 25%, and 5% probability cut offs, and the solid line indicates a 50% cut-off of discriminant score to be a male.	29
Figure 5. Histogram of discriminant scores of Northern Boobooks.	30
Figure 6. Relationship between discriminant score and posterior probability of being a male in Northern Boobooks. Dashed lines indicate 95%, 75%, 25%, and 5% probability cut offs, and the solid line indicates a 50% cut-off of discriminant score to be a male.	31
Figure 7. Distribution of carbon and nitrogen stable isotopes in Oriental Scops Owls, Northern Boobooks, and potential prey items.	33
Figure 8. Proportional diets of Oriental Scops Owls estimated by a multi-source mixing model. Line thickness indicates 50%, 75%, and 95% probability.	36
Figure 9. Proportional diets of Northern Boobooks estimated by a multi-source mixing model. Line thickness indicates 50%, 75%, and 95% probability.	36
Figure 10. Scatterplot of overall body size index (PC1) and individual trophic level ($\delta^{15}\text{N}$) in Northern Boobooks (<i>Ninox japonica</i>).....	39
Figure 11. Relationship between overall body size index (PC1) and individual trophic level ($\delta^{15}\text{N}$) in Oriental Scops Owls (<i>Otus sunia</i>).....	39

Chapter 1. Introduction

Raptors are high-level consumers in trophic webs and play important ecological roles in ecosystems (Ferguson-Lees and Christie 2001). However, their position as high-level consumers in ecosystems makes them prone to threats such as bioaccumulation of toxic elements, decline of food sources and habitat fragmentation. In addition, most raptors species in the world (66%) are migratory, which increases their vulnerability during their migratory life cycle (Bildstein 2006).

Raptors are generally hard to detect and observe, since they usually have low densities due to their habitat requirements and territoriality, elusive behavior, and often inaccessible of remote habitats (Bildstein and Bird 2007). These features make raptor research difficult, especially obtaining enough a large sample size, in spite of the necessity of understanding ecology of raptors. On the other hand, their relatively large body size and high trophic level, with relatively simple intra-guild communities often make raptors a good model for studying ecological aspects of competitive interaction and resource use within intra-guild communities (Jaksic and Carothers 1985, Burton and Olsen 1997, Bildstein and bird 2007). Besides, reversed sexual size dimorphism, with females having a larger body size than males, is a common phenomenon in diurnal and nocturnal raptors. The ecology and evolution of reversed sexual dimorphism in raptors intrigued numerous researcher to study raptors as a model species, but it never reached to a consensus so far.

Including all the falcons (Falconiformes), hawks (Accipitriformes) and owls (Strigiformes), total 546 raptors species are known to the world currently, and 221 species of owls, are mainly nocturnal birds of prey. Owls are difficult to observe because of their nocturnal behavior, and most of them use densely covered roosts during the daytime. However, their habit of casting pellets of indigestible materials, enabled us to study their diet more efficiently than other raptor species (Marks *et al.* 1999).

Oriental Scops Owls (*Otus sunia*) and Northern Boobooks (*Ninox japonica*) are two small, primarily insectivorous owls that breed in Korea during summer (Marks *et al.* 1999, Lee *et al.* 2000, Brazil 2009). These two migratory owls are the most

common owls in forests and forested patches during summer in Korea. In spite of their relative abundance in comparison to other Korean raptors, there is very limited information available on the ecology of these two small forest owls, including the extent of their sexual dimorphism and the quantitative study on diet are available in Korea, to our current knowledge. Because Oriental Scops Owls and Northern Boobooks are both sympatric insectivorous owls in Korean forests during summer, one would expect that they shall have some separate their niche to some extent to avoid unnecessary competition on resources. Niche partitioning, by separating their foraging microhabitat and/or food is a common mechanism (Schoener 1974, Reynolds and Meslow 1984, Burton and Olsen 1997, Ricklefs 2012) in the community of coexisting organisms. In owls, the separation or the overlap of diets on coexisting species are well documented over the world (Earhart and Johnson 1970, Herrera and Hiraldo 1976, Kavanagh 2002, Ibarra *et al.* 2014), thanks to their pellet-regurgitating behavior.

However, in studying the diet of these insectivorous owls, it is difficult to use their pellet castings since they decompose easily (Bildstein and Bird 2007, Olsen 2011) during their breeding seasons which include heavy raining period in Korea, and are hence difficult to find in dense closed-canopy forests. Thus, to understand the diet of adult Northern Boobooks and Oriental Scops Owls while they forage for themselves, conventional methods are not applicable, or are extremely time consuming. The stable isotope approach in trophic ecology and food web studies has become more common in recent years (Hobson and Wassenaar 1999, Kelly 2000, Inger and Bearhop 2008, Carreon-Martinez and Heath 2010, Boecklen *et al.* 2011), because using blood or feathers is relatively less destructive than conventional stomach content analysis. This approach can also help to understand time-integrated diet information of individual birds by characterizing turnover rates of isotope ratios in tissues. Whole blood, for example, is known to represent the diet composition of birds for 3-5 weeks. Carbon and nitrogen isotopes are most commonly used in food web studies. Therefore, the stable isotope approach is suitable for studies of the diet of Northern Boobooks and Oriental Scops Owls.

In multiple species of raptors, which have reversed sexual dimorphism, one of the

most argued factor that influences the degree of sexual dimorphism is their diet (Krüger 2005). Smaller males are suggested to be more efficient in flight and hence better at foraging prey for chicks, as well as better in prey delivery during courtship feedings (Hakkarinen *et al.* 1996). Because owl can take only one prey at a time by their talons, it will be energetically beneficial to be able to hunt few high quality prey at one foraging trip than multiple mediocre quality preys during multiple foraging trip for hungry chicks. Most female raptors invest their effort in incubation and brooding, whereas males are the ones who dedicate their effort on foraging for incubating females and chicks (Storer 1966, Marks *et al.* 1999). So, the male's ability to be a good forager would be an important factor in the reproductive fitness of a pair. This hunting efficiency may work in the same manner to the individual diet, because individual birds also require certain amount of energy for survival and reproduction. Thus, I expect that there would be relationships between body size and individual diets of Oriental Scops Owls and Northern Boobooks.

In summary, the first objective of this study is to examine morphological characteristics of Northern Boobooks and Oriental Scops Owls in relation to their sex, to develop sex-discriminant functions applicable in field conditions, and describe the degree of sexual dimorphism of the two owl species.

Second, I assessed blood stable isotope ratio to compare the food-niche partitioning of Northern Boobooks and Oriental Scops Owls. In addition, the dietary composition of bloods of Northern Boobooks and Oriental Scops Owls were reconstructed by using multi-source mixing model based on potential prey samples.

And finally, I considered how individual trophic level is related to the body size and to the degree of sexual dimorphism in Oriental Scops Owls and Northern Boobooks.

Chapter 2. Literature Review

2.1 Study species

Oriental Scops Owls (*Otus sunia*) and Northern Boobooks (*Ninox japonica*) are small migratory owls that breed in Korea. These insectivorous owls are summer visitors that arrive in Korea from early April and start to breed in the mid of May (Seo and Park 2008, Noh 2013). The other four regularly breeding owl species in Korea, the Japanese Scops Owl (*Otus semitorques*), Ural Owl (*Strix uralensis*), Tawny Owl (*Strix aluco*), and Eurasian Eagle Owl (*Bubo bubo*), are resident and are scarcer (Lee *et al.* 2000, Seo and Park 2008, Brazil 2009), and all are known to prey primarily on vertebrates. Moreover, these four resident owls are known to breed earlier than the migratory Oriental Scops Owls and Northern Boobooks.

Oriental Scops Owls have a wide distribution range, and 7–9 subspecies are recognized currently. Northern populations, including *O. s. japonicus* that breed in Japan and *O. s. stictonotus* that breeds from Korea to Far Eastern Russia, are migratory. The Korean breeding subspecies, *O. s. stictonotus* is known to be the largest and heaviest subspecies (Brazil 2009, Mikkola 2013). There are two color morphs and a gradient of intermediate forms in Oriental Scops Owls. Between the gray-brown and rufous morphs, the gray or grayish-brown morph are more common in Korea (Seo and Park 2008).

Northern Boobooks, together with Chocolate (*Ninox randi*) and Brown Boobooks (*Ninox scutulata*), recently separated from the Brown Hawk Owl (King 2002), Northern Boobooks in South Korea and Japan belong to the race *N. j. japonica*, and while the race *N. j. florensis* ranges from North Korea to SE Siberia. The resident population in Taiwan, *N. j. totogo*, occupies the southern limit of this species. Northern Boobooks are sexually monomorphic in plumage (Mark *et al.* 1999, Brazil 2009, Mikkola 2013).

Neither of Northern Boobooks or Oriental Scops Owls has been well studied, but there are a few reports on the diet and breeding in Japan (Noguchi 2002, Taniguchi 1983), and diet and prey handling in Taiwan (Lin and Yeh 2004) of Northern Boobooks, although these studies were based on small sample sizes. Few

more studies on Northern Boobooks are done in Japan and Taiwan, including a study of incubation behavior of breeding owls using an automatic temperature logging system (Masuda 1974), and another study of breeding ecology and sexual dimorphism (Lin *et al.* 2012). Additionally, a cryptic separate lineage of sedentary population of Northern Boobooks among the migratory population were found in Taiwan (Lin *et al.* 2013). Calling behaviors were also recorded by King and Icarangal (2008) at wintering grounds and by Oba (1996) in breeding period

Oriental Scops Owls have been the subject of even fewer previous studies. Noh (2013) studied their breeding biology for his master's thesis and described their natural nests, eggs, and prey delivery during the chick rearing period. More studies have been done on Eurasian Scops Owls and Elegant Scops Owls, in Italy (Marchesi and Sergio 2005, Martínez *et al.* 2007) and in Japan (Toyama and Saitoh 2011), respectively.

2.2 Reversed sexual size dimorphism in raptors

Unlike most other birds and mammals, where males have larger body sizes than females, most nocturnal and diurnal raptors show reversed sexual size dimorphism, with females that are larger and heavier than males (Mueller 1986). This phenomenon raises the question of why female raptors have larger body size than males, a question that has been asked many times by numerous ornithologists in the past (Selander 1966, Storer 1966, Reynolds 1972, Shine 1989, Krüger 2005).

The difference in the roles of each sex during breeding, that with the females investing most of their time on incubation and chick guarding (Storer 1966, Marks *et al.* 1999, Eldegard and Sonerud 2012 Steen *et al.* 2012), and the males are delivering food for chicks and females, is believed to have an important effect on the evolution and maintenance of reversed sexual size dimorphism (Storer 1966, Reynolds 1972).

The functional advantage of members of a breeding pair having different body sizes has been examined with respect to both of the advantage of smaller males and the advantage of larger females. Larger females are expected to have an advantage in having a larger body that can act as an energy reserve for breeding (Lundberg 1986) and/or as a weapon for defending the nest against predators (Storer 1966, Andersson and Norberg 1981). Smaller males, are considered to have higher efficiency in foraging and thus in providing food for chicks and females, and thus, smaller males must also be advantageous in the reproduction of offspring (Koprimaki 1986, Sunde *et al.* 2003).

In Kruger's comparative review (2005), the strength of reversed sexual dimorphism was strongly related to prey-related and foraging-related factors in owls (Strigiformes) and hawks (Accipitriformes).

Raptors are single-loaded central place foragers while breeding, a strategy that makes it more efficient for them to carry one large prey item when they forage further. By contrast, it is more energetically efficient to catch smaller items more frequently close to the nest (Olsen 2013), which females must do as they guard their chicks.

Some owl species, like the Great Gray Owl (*Strix nebulosa*), Boreal Owl (*Aegolius funereus*), and Snowy Owl (*Bubo scandiacus*) show high level of sexual size

dimorphism, with females can weigh about 40% more than males (Josephson 1980).

In addition to the ecological and evolutionary aspects of sexual size dimorphism, researchers have been developed morphometric sex discriminant functions for diverse taxonomic group of birds (in owls: Eurasian Eagle Owl, del Mar and Penteriani 2004, Boreal Owl, Hayward and Hayward 1991, Eastern Screech Owl, Smith and Wiemeyer 1992), that do not have sexual plumage dimorphism and thus are difficult to identify the sex of the birds in the field. In such cases, size differences enable researchers to identify the sex of the live birds when caught, without additional molecular methods that are costly and often impossible in field studies at remote locations (del Mar Delgado and Penteriani 2004, Leppert *et al.* 2006).

In both Oriental Scops Owls and Northern Boobooks, the amount of sexual dimorphism is not well studied, and no discriminant function analysis has been done to best of my knowledge. There are several published notes on the sexual size differences of Oriental Scops Owls that show slight reversed size dimorphism (Marks *et al.* 1999, Seo and Park 2009, Mikkola 2013). However, the dimorphism of Northern Boobooks is disputed.

Won (1981) reported slightly larger wing lengths in males (212–213mm) than in females (209–211mm); and even though the data of Lin and Yeh (2004) do not show statistical significance, DI indicates slightly large average values for males.

2.3 Food-niche and diet studies in raptors

In studies of the diets of owls, there are several conventional methods, including direct observation, analysis of prey remains and stomach contents, pellet analysis, fecal analysis, DNA analysis, and combinations of these methods (Rosenberg and Cooper 1990, Lee and Severinghaus 2004). However, pellet analysis and fecal analysis can be biased toward certain prey types (Redpath *et al.* 2001, Sergio 2002), and DNA analysis for prey composition may characterize only the number of species fed upon, not their relative quantities. Direct observation and analysis of prey remains can be biased by the observer's and the bird's behaviors, allowing the observer to see only what can be easily seen.

In recent studies of avian trophic levels, stable isotope analysis (SIA) using carbon and nitrogen becoming more common. The stable isotope ratio of a consumer's tissue will reflect the diet's stable isotope ratio according to the rate at which each isotope is assimilated from the diet into the tissue. The ratio of nitrogen isotopes increases, about 3.4 ‰ per trophic level, and thus represents consumer-diet relationship well; and the carbon stable isotopes are trace elements of carbon sources from the primary producer level. It is known that C3 plants and C4 plants assimilate carbon isotopes differently, and thus have markedly different carbon isotope signatures (Fry 2007). Consequently, stable isotope ratios can be used to quantify the relative proportion of diet sources in consumer tissue, by using isotopic linear mixing models (Post 2002). However, even though stable isotope analysis to assess trophic ecology and resource partitioning has proven to be a powerful tool, it has rarely been used to assess the trophic ecology of terrestrial avian top predators (Resano-Mayor *et al.* 2014a).

As mentioned above, the diets of Northern Boobooks and Oriental Scops Owls are not well studied, but all reports emphasize the insectivorous diet of these owls. Noguchi (2002) reported only one vertebrate among 1442 prey items identified, from a breeding pair of Northern Boobooks (most prey items were insects), and Taniguchi (1983) reported 64 vertebrate prey remains (62 birds, a lizard, and a bat) along with 5968 insect prey remains from a pair of breeding Northern Boobooks during three

consecutive years (1979–1981). In Taiwan, Lin and Yeh (2004) reported 2269 discarded insect remains from Northern Boobooks around the streetlights over 2 months (November–December 1999). This difference between studies might be caused by seasonal availability of prey, which was also mentioned by Taniguchi (1983) who noted that birds were more often captured in the early part of the breeding season. Food brought to the nest of a Korean breeding pair of Oriental Scops Owls assessed by Noh (2013), also composed of 96% of invertebrate prey, 3.5% unidentified prey, and less than 1% vertebrate prey.

However, as Taniguchi (1983) mentioned, prey use may change greatly depending upon seasonal prey availability of study period. A study of an Italian population of Eurasian Scops Owls (*Otus scops*), which were once considered conspecific with Oriental Scops Owls, reported one lizard among 59 identified prey items from pellets collected during breeding period, while the rest of the items were invertebrates (Panzeri *et al.* 2014). During the wintering period, when the invertebrate prey resources decrease, a total of seven Shrews and mice were found in the pellets, whereas only six invertebrates were identified (Panzeri *et al.* 2014). As seen in this study in Italy, the Eurasian Scops Owl, the European counterpart of the Oriental Scops Owls, may also feed on vertebrate prey when invertebrate prey abundance is slow.

According to the competitive exclusion principle (Hardin 1960, den Boer 1986), species in the same habitat may not occupy exactly the same niches, in order to minimize competition (Jankowski *et al.* 2012). Accordingly, most studies on sympatric owl's diets were done with pellets (Marti 1974): Burrowing Owls (*Glucidium gnoma*), Saw-whet Owls (*A. acadicus*), vs. Western Screech Owls (*O. kennicotti*) (Hayward and Garton 1988); Little Owls (*Athene noctua*) vs. Long-eared Owls (*Asio otus*) (Zhao *et al.* 2011) Tawny Owls (*Strix aluco*) vs. Ural Owls (*S. uralensis*) (Pavon-jordan *et al.* 2013) and direct Observations Elegant Scops Owls (*Otus elegans*) vs. Japanese Scops Owls (*O. semitorques*) (Toyama and Saitoh 2011). In these studies, general pattern of niche separation was that the larger bodied owls utilized larger sized prey, but smaller owls preyed on smaller preys like small birds and invertebrates.

Stable isotope analysis has been relatively little used in owl studies. Two studies of Saw-whet Owls (*Aegolius acadicus*) and Snowy Owls (*Bubo scandiacus*) used stable isotopes to reveal the marine-origin of their prey (Hobson and Sealy 1991, Therrien *et al.* 2011b) and only one study revealed diet-tissue discrimination factor of Snowy Owl (Therrien *et al.* 2011a),

Chapter 3. Materials and Methods

3.1 Study sites and period

Field sampling was done at Gwang-reung Forest (37°44'42.55" N, 127° 9'35.90" E) and Seoul National University Gwanak Arboretum at Gwanak Mountain (37°25'12.12" N, 126°56'14.73" E), at Gyeonggi-do province, Korea, from May to June in 2014 and April to May in 2015 (Figure 1). Gwang-reung Forest is an old-growth temperate broadleaf forest which is under management of Korea National Arboretum, Korea Forest Service. Study area in Gwang-reung Forest consists of natural deciduous broadleaf forest and plantations located at an elevation of around 100-600m. Annual rainfall is around 1500mm, and average monthly temperatures are highest in August (29.1°C), and lowest in January (-13°C). Major tree species of the natural deciduous broadleaf stands in Gwang-reung Forest are *Carpinus laxiflora*, *C. cordata*, *Quercus serrata*, *Q. mongolica* and *Q. variabilis* (Korea National Arboretum 2011). Gwanak Arboretum and adjacent forests have annual rainfall of 1260mm, and annual average temperature is 12.4°C, highest in August (29.5°C) and lowest in January (-6.4°C). Common tree species in natural stands of Gwanak Arboretum and adjacent mountain forests are *Q. serrata*, *Q. mongolica*, *Q. variabilis*, and *Pinus densiflora* (Jang *et al.* 2013).

For the sexual dimorphism study, additional birds were sampled from Seoul National University (SNU)'s Gwanak Arboretum, and Gwanak Mountain forests adjacent to the SNU campus. Also, measurements of birds and DNA samples were collected from dead specimens and live individuals under rehabilitation at four wildlife rescue centers (Gyeonggi Wildlife Rescue Center, Busan Wildlife Rescue Center, Ulsan Wildlife Rescue Center and Kyungshung University Bird Museum; Figure 1). For trapping and sampling birds for trophic ecology study using stable isotope ratio analysis, birds captured at Gwang-reung Forest and National Arboretum from 22 May to 5 June 2014 were only included in the analysis.

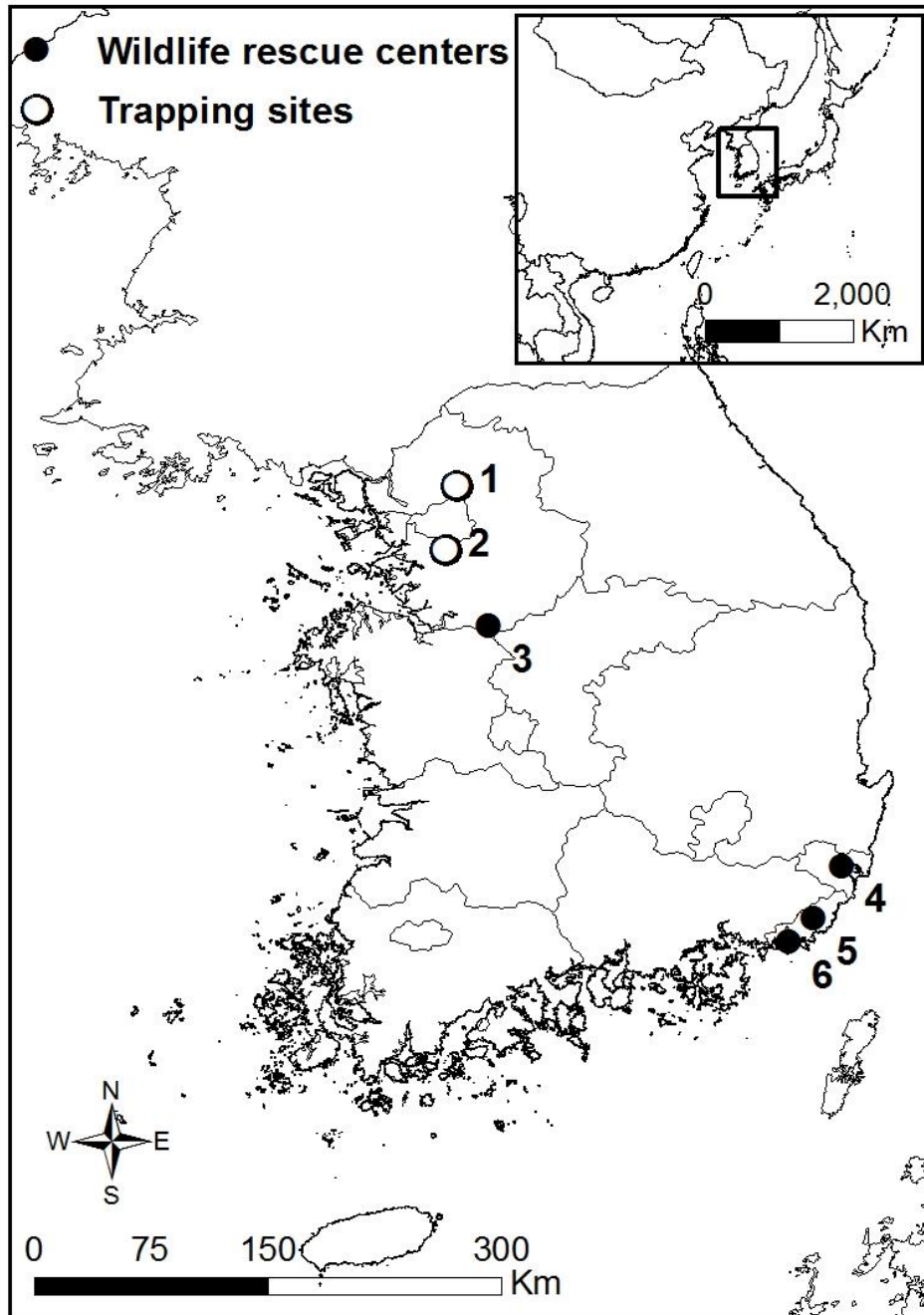


Figure 1. The location of wildlife rescue centers and trapping sites. Numbers indicate following locations. 1: Gwang-reung Forest, 2: Gwanak Arboretum, 3: Gyeonggi Wildlife Rescue Center, 4: Ulsan Wildlife Rescue Center, 5: Gyungsung University Bird Museum, 6: Busan Wildlife Rescue Center.

3.2 Sample collection

Mist nets (12×2.5 m, 5 shelves, 16×16 mm mesh), conspecific audio lures, and stuffed decoys were used to trap the owls (Bildstein and Bird 2007). Mist nets were placed near locations where the calls of Oriental Scops Owls and Northern Boobooks had been previously detected. Nets were checked at 30 minute intervals until capture. Each mist net station and audio lure was operated for no more than three hours if the capture was unsuccessful, to minimize disturbance to the birds. Locations of unsuccessful trapping attempts were re-visited within three days. Birds were extracted from mist nets immediately and ringed, measured, blood and feather sampled, and released after being photographed. Each bird was handled within approximately five minutes to minimize stress. When more than two birds were caught simultaneously, birds were kept in a cotton bag hung from the ground before being processed, and none of the birds were kept more than 30 minutes.

For each captured birds the following morphometric measurements were taken: wing length (maximum flattened length) and tail length to closest 1 mm with the wing ruler, tarsus length, hind claw length, bill length (to skull and to nostril), bill depth, bill width and head length to the closest 0.1 mm with calipers; weight was measured with a Pesola® spring scale or CAS digital scale to 0.1 g. Trapping, sampling and banding of birds were permitted by the Cultural Heritage Administration (2014.5.22), because Northern Boobooks and Oriental Scops Owls are designated as a natural monument and they are legally protected in Korea. All procedures including handling of birds were based on Redfern and Clark (2001) and Hardey *et al.* (2013) to sustain minimum disturbance and stress of individual birds.

Blood collection, containment and preservation methods were followed as Owen (2011)'s review. Small amount (approximately 75–100 μ l) of whole blood was collected by venipuncture from the brachial vein using a 26G needle and micro-capillary tube. 70% isopropyl ethanol swabs were used to clear the feathers, making the brachial vein visible before puncture. The lancing area was dried completely before bleeding to avoid mixing ethanol with the blood sample. Collected whole blood was directly moved to a 1.5 ml micro-tube and kept frozen at -20°C until it could be analyzed (Hobson *et al.* 1997). 3–5 body feathers were also collected from right or left flank for further analysis.

In addition to the data collected from the birds captured in the wild at Gwang-reung Forest and Gwanak Arboretum. Wildlife rescue centers were visited for measuring and collecting tissue samples from additional birds. Under the supervision of a rehabilitator or veterinarian, rescued birds were measured quickly, following the methods described above. Small amounts of blood or feathers were sampled also, following the same method used with trapped wild birds. Frozen specimens of Oriental Scops owls and Northern Boobooks were also measured after a few hours of defrosting. Feathers, tips of tongues or small amounts of pectoral muscle (0.3 g) were collected for molecular sex identification. All samples from wildlife rescue centers were kept frozen at -20°C until analysis.

Potential prey items were collected during mist net operations. Potential prey types were chosen based on previous studies reporting the diet of Northern Boobooks from Japan (Taniguchi 1983, Noguchi 2002) and Taiwan (Lin and Yeh 2004), and the diet of Oriental Scops Owls in Korea (Noh 2013) were collected from the field. Information from a diet study of two closely related congeneric owl species on the Daito Island, Japan (Toyama and Saitoh 2011) was also utilized in choosing prey groups.

Several prey species were collected from the study site during the trapping period. Potential arthropod preys were collected after being entangled in the mist net randomly, as well from the ground by hand. Since these owls are known to prey also on larger vertebrate species, birds and mice were included among the potential prey types. In keeping with ethical practices, bird samples were collected from bird-window collisions near the study site and in adjacent areas that have similar deciduous forest environments. Mice samples were obtained from a separate small mammal monitoring study by collecting animals that are died accidentally from the live traps. Vertebrate samples were selected based on the collection date and locale, to take into account their seasonal state and environment which are closest to the study area. All potential prey species used in analysis were identified to the lowest taxonomic level to best knowledge (see Appendix 1 for the species list).

3.3 Data analysis

3.3.1 Molecular sex identification

To determine the sex of the owls, molecular sex identification using the polymerase chain reaction (PCR) was conducted by following methods adjusted from Han *et al.* (2009). Genomic DNA was extracted from air-dried whole blood, feathers, tongue, and pectoral muscle tissue using the Qiagen DNeasy Blood® and Tissue Kit (Qiagen, USA). The PCR amplification was conducted in a total volume of 20 µl, using AccuPower® PCR PreMix, which contains 30mM KCl, 10mM Tris-HCl(pH 9.0), 1.5mM MgCl₂, 250 µl of each dNTP and 1 Unit of Top DNA polymerase. A total of 2 µl (10 pmol/µl) of mixture of 3 primers (P2: 5'-TCTGCATCGCTAAATCCTTT-3', P0: 5'-ACIAGACCAAGGTTGAGTTA-3', P8: 5'-GTYAARGAGTAGGAACCCTC-3') and 2 µl of extracted genomic DNA was added before performing the PCR procedure with a Lifepro Thermal Cycler® (Bioer, China).

Denaturation was started at 94°C for 2 min 30 s and was followed by 30 cycles of 94°C for 30 s, 53°C for 30 s, and 72°C for 45 s. A final run at 72°C for 5 min completed the PCR procedure. The PCR product were visualized and checked by 3% agarose gel electrophoresis at 100V operated for 45 minutes. This method, allowed molecular sex identification of owls and some other avian taxa by applying the P0 primer in addition to the P2 and P8 primers which were described by Griffith *et al.* (1998) successfully (Han *et al.* 2009).

Individual samples with two distinct bands were considered to be females, and individuals with single bands were considered to be males.

3.3.2 Stable isotope analysis

Whole blood and feather samples collected from Gwang-reung Forest were analyzed for carbon and nitrogen stable isotope ratios. Potential prey items were prepared before stable isotope analysis. Invertebrate samples were rinsed with deionized water using an ultrasonic cleaner (SAEHAN digital sonic© model: SHB-1025) and dismembered, as the owls do before consumption, to increase the ease of grinding and further homogenize the sample. Wings and jaws of large beetles were removed and wings of large moths were removed as well. From vertebrate species, about 2g of muscle was sampled from the breasts or legs with a disposable dissecting blade, and any other tissue (e.g. membranes, ligaments, and fat) was removed from the sample. Muscle samples were rinsed with deionized water using the ultrasonic cleaner. Variation in the lipid content of tissue samples is known to affect $\delta^{13}\text{C}$ values (Post *et al.* 2007), thus lipid extraction is a common procedure in preparation for tissue carbon stable isotope analysis. However, Bennett and Hobson (2009) monitored C:N ratios in their study of forest arthropods to avoid the effects of lipid extraction in tissue $\delta^{15}\text{N}$ values. Since a nitrogen stable isotopic signature is also important in predicting diet composition of the two owl species, Bennett and Hobson (2009)'s approach were used to monitor C:N ratios instead of lipid extraction, and considered samples with relatively high C:N ratios to have high lipid content.

Prepared samples were air-dried in a dry oven at 60°C for at least 72 hours and ground into fine particles and homogenized with mortars and pestles.

Stable isotope ratios of carbon and nitrogen were analyzed with a continuous-flow stable isotope ratio mass spectrometer (Isoprime-EA, Micromass, UK) linked with a CN analyzer (NA Series 2, CE Instruments, Italy) at the National Instrumentation Center for Environmental Management (NICEM), Seoul National University, Seoul, Korea. Stable isotopes were expressed in δ notation as per mill, calculated according to the following equation:

$$\delta X(\text{‰}) = [(R_{\text{sample}} - R_{\text{standard}}) - 1] \times 1000$$

where X is the element in the analysis, R_{sample} is the ratio of isotopes from the analyzed sample, and R_{standard} is the ratio of isotopes in international standard materiald, which are Pee Dee Belemnite (PDB) for carbon and atmospheric nitrogen.

3.3.3 Statistical analysis

For analyzing sexual size dimorphism in the two owl species, each measurements of both sexes were compared with Wilcoxon Mann-Whitney rank sum test (PROC NPAR1WAY) to find any significant differences between the sexes. The magnitude of the size difference in each morphological measurements was calculated using the dimorphic index (DI; Storer 1966, Greenwood 2003),

$$DI = \frac{f - m}{(m + f)} \times 100$$

where m is the mean value of male measurements and f is the mean value of female measurements. A positive DI indicates that the morphometric character is larger in females, and so represents reversed sexual dimorphism.

Considering that these two owl species are complete migrants, we did not include body mass in performing a discriminant function analysis (DFA). Since our data did not fit multivariate normality, quadratic discriminant analysis (PROC DISCRIM) was performed with measurement variables selected through a stepwise procedure (PROC STEPDISC). Birds with missing measurements due to the specimen's condition (e.g., damaged claws, bills or feathers, were excluded from the discriminant analysis.

The birds were classified as male or female based on the discriminant score (D) from the discriminant analysis. The accuracy of the discriminant functions produced by DFA was evaluated by the re-substitution method and jackknife method using the POSTERR and CROSSVALIDATE options in PROC DISCRIM of SAS software. To estimate the posterior probability from the discriminant scores of individuals, a non-linear regression model was fitted, where k and c were constants calculated from a nonlinear procedure (PROC NLIN).

$$PP = 1/(1 + \exp(kD - c))$$

Wilcoxon Mann-Whitney rank sum test (PROC NPAR1WAY) was used in comparing the difference of stable isotope values of two owl species. Coefficients of variance (CV) were calculated to compare the variation of stable isotope ratios of owl's whole blood, and the Levene's test was also performed to compare isotopic variance between two species.

Potential prey species were categorized into five broad taxonomic groups, and arthropods were separated based on major prey types identified by previous studies. A one way Kruskal-Wallis was first performed to see the difference in the stable isotope ratios of nitrogen and carbon in the prey type groups. Dunn's post hoc test was then used to compare each group. Among these potential prey groups, bush-crickets and beetles did not show a significant difference, and thus, pooled as a single group.

The diet composition of both owl species was reconstructed with the R package SIAR. This package is designed to run mixing models for isotope data based on Bayesian statistics, and it provides means and 95% confidence intervals of sources that contributed to the consumer tissue, and presents posterior probability distributions for each contributions graphically.

This model incorporated a trophic enrichment factor (TEF, or also called fractionation rate or discrimination factor) for stable isotope ratios between consumer tissue and diet source. However, there is no known value of trophic enrichment factor available for Oriental Scops Owls or Northern Boobies in the literature. Thus, I used a trophic enrichment factor following the model of Caut *et al.* (2009) that provides diet-specified model for different tissues of each taxonomic groups. I present the mean contribution of each prey group to the diet of the two owl species. Stable isotopic values were given as means and standard deviations (SD).

To see the effect of body size on the trophic status of individual owls, I first conducted a principal component analysis to extract the principal component (PC1) that best explains multiple morphometric measures (Rising and Somers 1989), rather than using a single morphometric character for the analysis. Then, I used a Spearman correlation analysis with PC1 and the nitrogen stable isotope ratio. Too small sample size hindered analyzing the sexes separately in Northern Bobwhites, thus instead, I pooled both sexes together for the analysis. However, for male Oriental Scops Owls, separate correlation analysis was performed.

Chapter 4. Results

4.1 Sampling and molecular sex identification

During the two spring seasons in 2014 and 2015, a total of 10 Northern Boobooks and 30 Oriental Scops Owls were trapped and measured at Gwang-reung Forest and the forests of Gwanak Arboretum. Measurements of 22 Northern Boobooks and 16 Oriental Scops Owls, dead and alive, which were obtained from wildlife rescue centers, were also used in the sexual dimorphism study.

All sampled birds were successfully sexed, using molecular method.

Among the 46 Oriental Scops Owls, 19 were identified as females and 27 were identified as males. 13 of the 32 Northern Boobooks were identified as females and 19 were identified as males by examining gel electrophoresis results of PCR products (Figure 2).

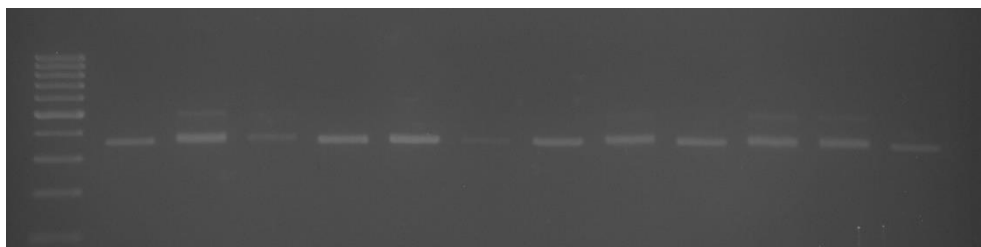


Figure 2. Sample image of gel electrophoresis showing females (two bars) and males (single bar).

4.2 Morphology and sexual size dimorphism

Six external characteristics were found to differ statistically between males and females in Oriental Scops Owls (Table 1). Although the differences were small, all measurement variables showed reversed sexual dimorphism in Oriental Scops Owls. By contrast, Northern Boobooks had a few external characteristics that showed male-biased size dimorphism, but none of those measurement variables were statistically significant (Table 1, 2). In Northern Boobooks, length of bill to nostril, bill to skull and bill width had positive dimorphism index (DI). And seven other external characteristics were found to have negative indices, indicating larger males (Table 3, 4). Although I used only a small sample from the wild birds, the weight of northern Boobooks showed the strongest male-biased sexual size dimorphism, with males that were about 12% heavier than females. In Oriental Scops Owls, the width of the bill and the weight showed the strongest sexual size dimorphism. The coefficient of variance was highest in bill width (BW) in Northern Boobooks and second highest in Oriental Scops Owls, and weight had the highest coefficient of variance in Oriental Scops Owls.

Table 1. Morphometric parameters of Oriental Scops Owls in the Republic of Korea. Asterisks indicate significant differences (* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$). CV and DI represent the coefficient of variance and the dimorphic index, respectively.

Variables	Sex (<i>n</i>)	Mean \pm SD (range)	Median	CV	U	Z	<i>P</i>	DI
Wing* (max)	Total (46)	148.4 \pm 3.7 (138.5 – 153.0)	149.2	2.48				
	Male (27)	147.3 \pm 3.8 (138.5 – 153.0)	147.0	2.59	553	2.382	0.017	0.434
	Female (19)	149.9 \pm 3.0 (142.5 – 153.0)	151.5	1.98				
Tail***	Total (46)	69.3 \pm 3.5 (60.0 - 75.0)	70.0	5.01				
	Male (27)	67.81 \pm 3.15 (60.0 – 72.0)	68.5	4.65	611	3.685	0.000	1.286
	Female (19)	71.4 \pm 2.8 (66.0 - 75.0)	72.0	3.92				
Head	Total (46)	41.17 \pm 0.87 (39.30 - 42.90)	41.15	2.11				
	Male (27)	41.04 \pm 0.87 (39.30 - 42.50)	41.00	2.13	495.5	1.084	0.279	0.200
	Female (19)	41.37 \pm 0.85 (39.90 - 42.90)	41.20	2.06				
Bill length to nostril (BN)*	Total (46)	9.43 \pm 0.63 (7.6 - 10.5)	9.5	6.67				
	Male (27)	9.25 \pm 0.62 (7.6 - 10.42)	9.1	6.71	544	2.179	0.029	1.136
	Female (19)	9.68 \pm 0.56 (8.5 - 10.5)	9.8	5.83				
Bill length to skull (BS)	Total (46)	20.14 \pm 0.76 (18.1 - 21.5)	20.1	3.78				
	Male (27)	20.04 \pm 0.83 (18.1 - 21.5)	20.1	4.16	479	0.716	0.474	0.298
	Female (19)	20.28 \pm 0.64 (19.4 - 21.4)	20.2	3.17				
Bill depth (BD)***	Total (46)	8.57 \pm 0.6 (7.3 - 10.8)	8.5	7.05				
	Male (27)	8.35 \pm 0.47 (7.3 - 9.4)	8.3	5.64	592	3.248	0.001	1.566
	Female (19)	8.89 \pm 0.64 (7.9 - 10.8)	8.9	7.17				
Bill width (BW)**	Total (46)	5.38 \pm 0.58 (4.7 - 7.2)	5.3	10.81				
	Male (27)	5.23 \pm 0.54 (4.7 - 6.5)	5	10.34	545.5	2.203	0.016	1.708
	Female (19)	5.6 \pm 0.58 (4.8 - 7.2)	5.5	10.45				

Table 2. Morphometric parameters of Oriental Scops Owls in the Republic of Korea. Asterisks indicate significant differences (* $P<0.05$, ** $P<0.01$, *** $P<0.001$). CV and DI represent the coefficient of variance and the dimorphic index, respectively.

Variables	Sex (<i>n</i>)	Mean \pm SD (range)	Median	CV	U	Z	<i>P</i>	DI
Tarsus**	Total (46)	25.15 \pm 1.02 (22.37 - 27.6)	25.2	4.05				
	Male (27)	24.79 \pm 0.99 (22.37 - 27)	24.7	3.98	568	2.703	0.007	0.862
	Female (19)	25.66 \pm 0.85 (24.3 - 27.6)	25.3	3.31				
Hindclaw	Total (46)	6.97 \pm 0.39 (6.1 - 7.7)	6.9	5.62				
	Male (27)	6.86 \pm 0.38 (6.1 - 7.5)	6.9	5.59	214	1.885	0.059	1.070
	Female (19)	7.16 \pm 0.34 (6.7 - 7.7)	7.16	4.73				
Weight	Total (30)	81.34 \pm 11.52 (63.3 - 123.4)	78.45	14.16				
	Male (19)	76.56 \pm 6.06 (63.3 - 91.0)	76.70	7.91	238.5	2.907	0.004	3.924
	Female (11)	89.60 \pm 14.15 (75.0 - 123.4)	89.00	15.79				

Table 3. Morphometric characteristics of Northern Boobooks in the Republic of Korea. Asterisks indicate significant differences (* $P<0.05$, ** $P<0.01$, *** $P<0.001$). CV and DI represent the coefficient of variance and the dimorphic index, respectively.

Variables	Sex (<i>n</i>)	Mean \pm SD (range)	Median	CV	U	Z	P	DI
Wing* (max)	Total (31)	226.0 \pm 6.0 (215-237)	225	2.64				
	Male (12)	227.8 \pm 5.8 (220-237)	228.5	2.56	220.0	1.1174	0.2638	-1.2370
	Female (19)	225.0 \pm 6.0 (215-235)	223.5	2.65				
Tail***	Total (31)	120.7 \pm 5.0 (108-130)	121	4.11				
	Male (12)	120.9 \pm 4.8 (113-127.5)	121.5	3.93	195.5	0.1221	0.0904	-0.2485
	Female (19)	120.6 \pm 5.2 (108-130)	121	4.33				
Head	Total (31)	48.68 \pm 0.99 (46.6-50.9)	48.8	2.03				
	Male (12)	49.09 \pm 1.00 (47.3-50.9)	49.18	2.04	236.5	1.7873	0.0840	-1.3742
	Female (19)	48.42 \pm 0.91 (46.6-49.7)	48.7	1.89				
Bill length to nostril (BN)*	Total (31)	12.20 \pm 0.60 (11.1-13.4)	12.2	4.91				
	Male (12)	12.05 \pm 0.66 (11.2-13.4)	12.05	5.51	165.0	-1.0790	0.2892	2.0534
	Female (19)	12.30 \pm 0.55 (11.1-13.4)	12.3	4.49				
Bill length to skull (BS)	Total (31)	25.08 \pm 1.00 (22.4-27.4)	25.2	4				
	Male (12)	24.88 \pm 1.2 (22.4-26.6)	24.9	4.84	180.0	-0.467	0.644	1.318
	Female (19)	25.21 \pm 0.86 (24.2-27.4)	25.2	3.42				
Bill depth (BD)***	Total (31)	10.25 \pm 0.53 (9.1-11.9)	10.2	5.17				
	Male (12)	10.41 \pm 0.57 (9.9-11.9)	10.2	5.47	208.0	0.653	0.519	-2.529
	Female (19)	10.15 \pm 0.49 (9.1-10.8)	10.2	4.85				
Bill width (BW)**	Total (31)	7.01 \pm 0.77 (5.4-8.4)	6.9	11.01				
	Male (12)	6.9 \pm 0.94 (5.4-8.4)	6.85	13.67	181.0	-0.427	0.673	2.575
	Female (19)	7.08 \pm 0.66 (6.2-8.2)	7.1	9.33				

Table 4. Morphometric characteristics of Northern Boobooks in the Republic of Korea. Asterisks indicate significant differences (* $p<0.05$, ** $p<0.01$, *** $p<0.001$). CV and DI represent the coefficient of variance and the dimorphic index, respectively.

Variables	Sex (<i>n</i>)	Mean \pm SD (range)	Median	CV	U	Z	<i>P</i>	DI
Tarsus**	Total (31)	27.73 \pm 1.89 (24-32.3)	27.8	6.82				
	Male (12)	28.35 \pm 2.15 (24-32.3)	28	7.6	224.0	1.279	0.211	-3.627
	Female (19)	27.34 \pm 1.64 (24-29.85)	27.7	6.02				
Hindclaw	Total (10)	10.89 \pm 0.53 (9.95-11.5)	10.98	4.89				
	Male (4)	11.28 \pm 0.39 (10.7-11.5)	11.45	3.43	31.0	1.818	0.103	-5.933
	Female (6)	10.63 \pm 0.47 (9.95-11.1)	10.75	4.45				
Weight	Total (9)	184.11 \pm 14.23 (168-206)	186	7.73				
	Male (4)	196.50 \pm 9.15 (186-206)	197	4.65	29.0	2.091	0.070	-12.031
	Female (5)	174.20 \pm 8.14 (168-188)	170	4.67				

4.2.1 Discriminant analysis

In spite of some overlap in measurements, a stepwise backward selection chose tail, tarsus, bill depth and bill width as variables that best classify male and female Oriental Scops Owls. With those selected values, the following discriminant function analysis formulated below equation ($\chi^2 = 10.004$, $P = 0.440$; Wilk's lambda = 0.592, $F = 7.07$, $P < 0.001$). Discriminant scores calculated from DFA for each individual had certain amount of overlap, but females had generally greater discriminant scores than males (Figure 3)

$$D_o = -56.0873 + 1.18762 * \text{Bill depth} + 0.51508 * \text{Bill width} \\ + 0.34729 * \text{Tail length} + 0.73489 * \text{Tarsus length}$$

Error rates of this discriminant function tested by the re-substitution and leave-one out methods were 20.39% and 20.78%, respectively.

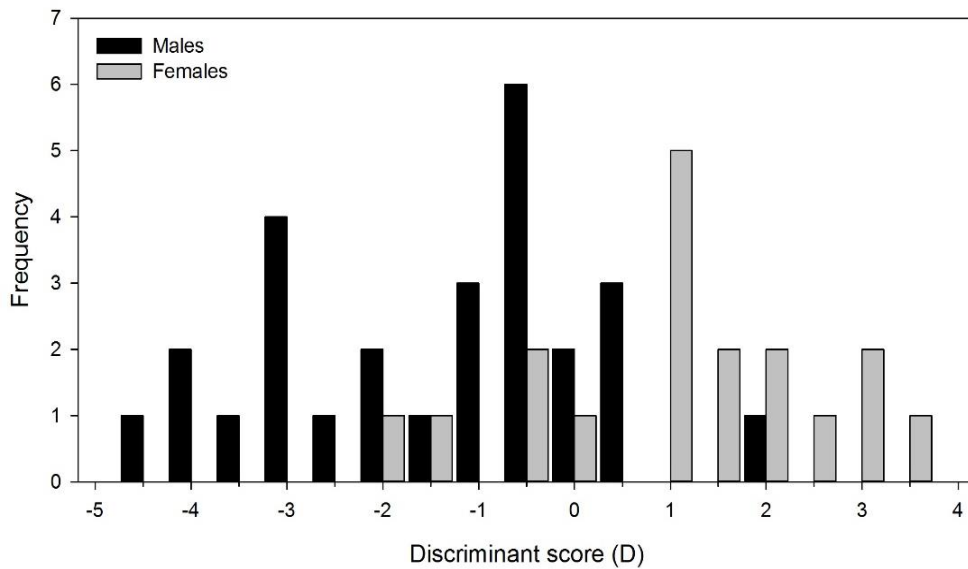


Figure 3. Histogram of discriminant scores (D) for Oriental Scops Owls.

From non-linear regression (Figure 4), the equation to explain posterior probabilities and discriminant scores was calculated as follows ($k = 1.0000$, standard error = 0.0000028472, $\underline{P} < 0.001$; $c = 0.0003$, standard error = 0.00000278, $P < 0.001$)

$$PP_o = 1/[1 + \exp(D - 0.0003)]$$

When the posterior probability is 0.5, this equation calculates cut-off value of being male or female as -0.0003. In addition, birds that have scores of $D \leq -2.941$ or $D \geq 2.947$ can be classified as males and females, respectively, with posterior probability of 0.95.

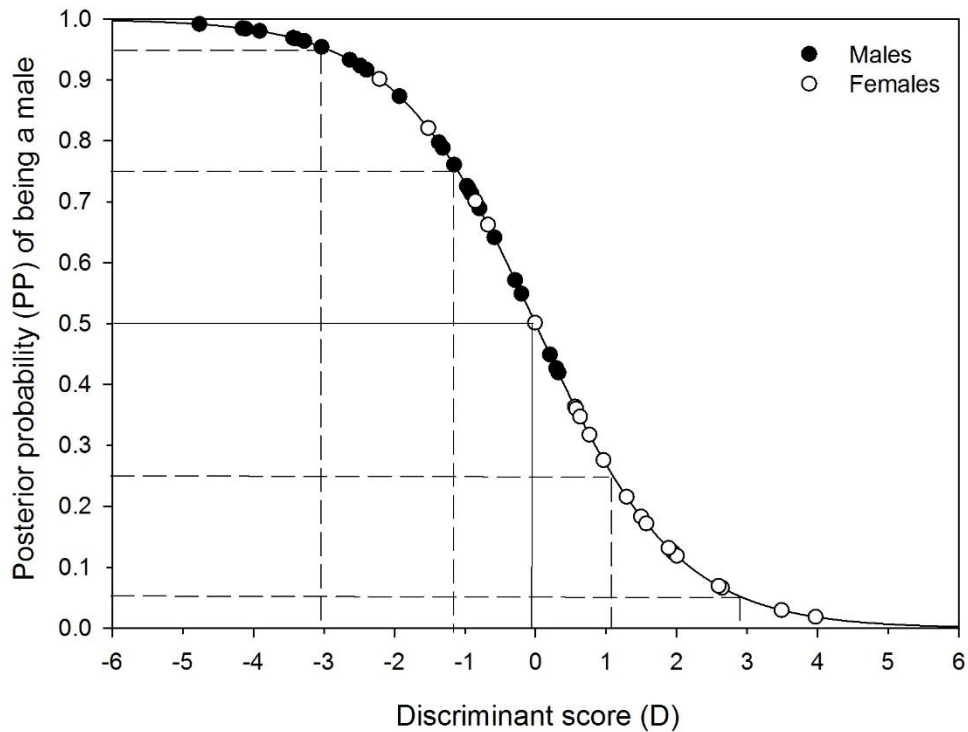


Figure 4. Relationship between discriminant scores and posterior probability of being a male of Oriental Scops Owls. Dashed lines indicate 95%, 75%, 25%, and 5% probability cut offs, and the solid line indicates a 50% cut-off of discriminant score to be a male.

In Northern Boobooks, a stepwise backward selection chose bill length (to skull) and head length as variables that best classify males and females. Discriminant function analysis produced the following linear equation ($\chi^2 = 1.823$, $P = 0.610$; Wilk's Lambda = 0.78434641, $F = 3.99$, $P = 0.030$), however, the overlap between discriminant scores of males and females were greater than in that of Oriental Scops Owls (Figure 5).

$$D_N = 34 - 1.0544 * \text{Head length} + 0.7307 * \text{Bill length (to skull)}$$

The accuracy of this discriminant function was tested by the re-substitution and leave-one-out methods, and the error rates were 30.26% and 30.65%, respectively.

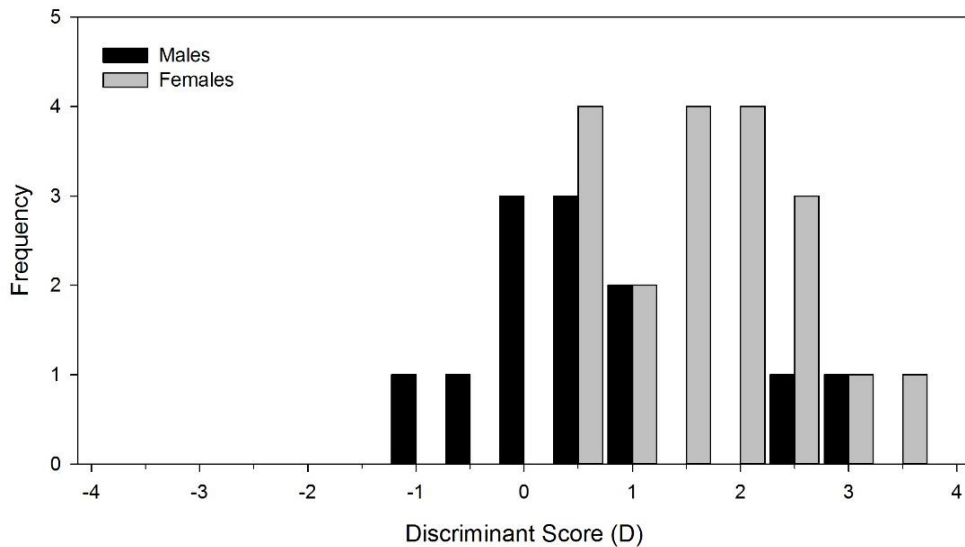


Figure 5. Histogram of discriminant scores of Northern Boobooks.

From non-linear regression (Figure 6), the equation to explain posterior probability and discriminant scores were calculated as follows ($k = -1.0000$, standard error = 0.0000034502 , $P < 0.001$; $c = 0.4364$, standard error = 0.0000027182 , $P < 0.001$).

$$PP_N = 1/[1 + \exp(-D - 0.4364)]$$

When the posterior probability is 0.5, this equation calculates a cut-off value of being male or female as 0.4364. In addition, birds that had scores of $D \leq -3.381$ or $D \geq 2.508$ can be classified as males and females, respectively, with posterior probability of 0.95.

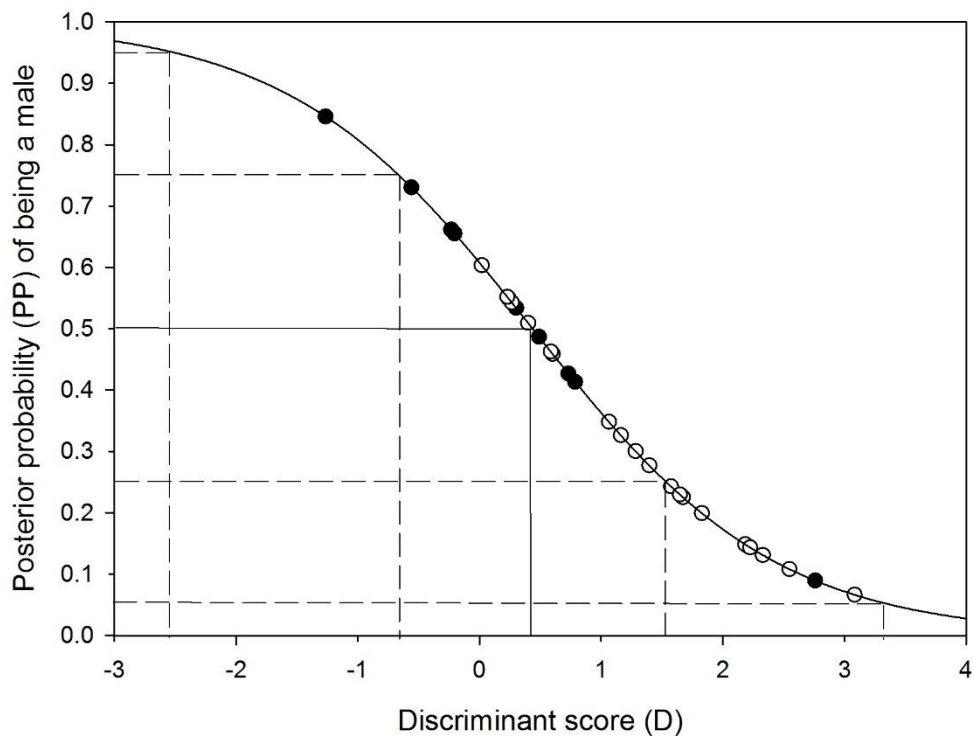


Figure 6. Relationship between discriminant score and posterior probability of being a male in Northern Boobooks. Dashed lines indicate 95%, 75%, 25%, and 5% probability cut offs, and the solid line indicates a 50% cut-off of discriminant score to be a male.

4.3 Stable isotope ecology

4.3.1 Stable isotope ratio analysis

The two owl species showed clear differences in both carbon and nitrogen stable isotope ratios (Figure 7). The nitrogen stable isotope ratios ($\delta^{15}\text{N}$) were significantly higher in Northern Boobooks ($U = 116.0$, $Z = 3.501$, $P < 0.001$) than in Oriental Scops Owls in all samples. Carbon stable isotope ratios ($\delta^{13}\text{C}$) were also significantly higher in Northern Boobooks ($U = 102.5$, $Z = 2.311$, $P = 0.021$), though there was some overlaps between the two species.

In both species, there was some levels of variation in the nitrogen stable isotope ratio. However, the coefficient of variance of the carbon stable isotope ratio was low in Oriental Scops Owls, in contrast to the high coefficient of variance in Northern Boobooks ($\delta^{15}\text{N}$: $F=0.29$, $P=0.597$; $\delta^{13}\text{C}$: $F=10.32$, $P=0.005$; Table 5).

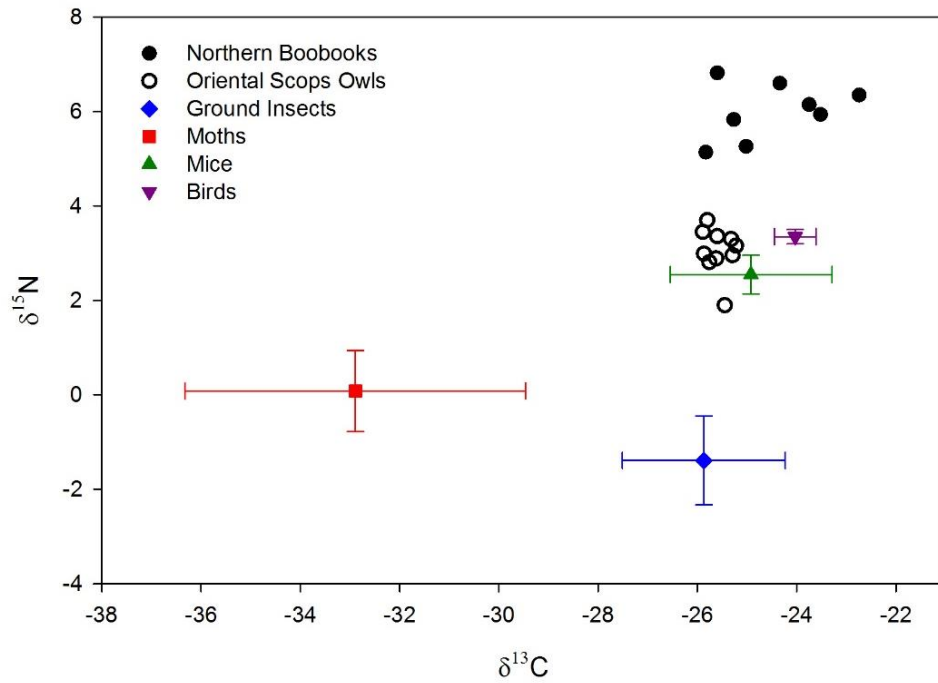


Figure 7. Distribution of carbon and nitrogen stable isotopes in Oriental Scops Owls, Northern Boobooks, and potential prey items.

Table 5. Nitrogen and carbon stable isotope values for Oriental Scops Owls and Northern Boobooks.

	Species	Mean \pm SD	Median	CV (%)	U	Z	P
$\delta^{13}\text{C}$	<i>O. sunia</i> (n=10)	-25.58 ± 0.25	-25.61	0.98	102.5	2.311	0.021
	<i>N. japonica</i> (n=8)	-24.51 ± 1.10	-24.68	4.49			
$\delta^{15}\text{N}$	<i>O. sunia</i> (n=10)	3.05 ± 0.49	3.08	16.09	116	3.510	0.000
	<i>N. japonica</i> (n=8)	6.01 ± 0.60	6.05	9.93			

Among prey groups, there was a significant difference between the carbon ($F = 14.18$, $P < 0.001$) and nitrogen ($F = 33.17$, $P < 0.001$) isotope values of each group, except beetles and bush crickets. Both carbon and nitrogen isotope ratios were indistinguishable between bush crickets and beetles, so I pooled those two groups into one (ground insects) for analytical purposes (Table 6).

Table 6. Stable isotope values of potential prey items of Oriental Scops Owls and Northern Boobooks in the Republic of Korea.

Groups	Sample size (n)	Mean biomass (g)	Elements	Mean	SD
Moths	2	0.8	$\delta^{13}\text{C}$	-32.89	3.43
			$\delta^{15}\text{N}$	0.08	0.86
Mice	3	24.2	$\delta^{13}\text{C}$	-24.92	1.63
			$\delta^{15}\text{N}$	2.55	0.41
Ground insects	12	0.9	$\delta^{13}\text{C}$	-25.87	1.64
			$\delta^{15}\text{N}$	-1.39	0.94
Bush crickets	4	0.9	$\delta^{13}\text{C}$	-27.27	0.65
			$\delta^{15}\text{N}$	-1.63	0.43
Beetles	8	0.9	$\delta^{13}\text{C}$	-25.17	1.54
			$\delta^{15}\text{N}$	-1.27	1.12
Birds	4	45.6	$\delta^{13}\text{C}$	-24.03	0.42
			$\delta^{15}\text{N}$	3.35	0.15

4.3.2 Diet reconstruction of owls

Results from a Bayesian multi-source mixing model showed that the two owl species utilized different proportions of prey groups. Northern Boobooks (Figure 9) relied on more avian prey (Mean = 49%, 95% CI = 19–85%), followed by mice (Mean = 33%, 95% CI = 0–62%), moths (Mean = 10%, 95% CI = 0–20%), and ground insects (Mean = 6%, 95% CI = 0–20%). Oriental Scops Owls (Figure 8) consumed ground insects the most (Mean = 34%, 95% CI = 24–44%), followed by birds (Mean = 23%, 95% CI = 4–40%), moths (Mean = 22%, 95% CI = 12–32%), and mice (Mean = 20%, 95% CI = 0.2–40%). Overall, Oriental Scops Owls selected more arthropod prey than Northern Boobooks, while Northern Boobooks were highly dependent on avian prey sources in this study.

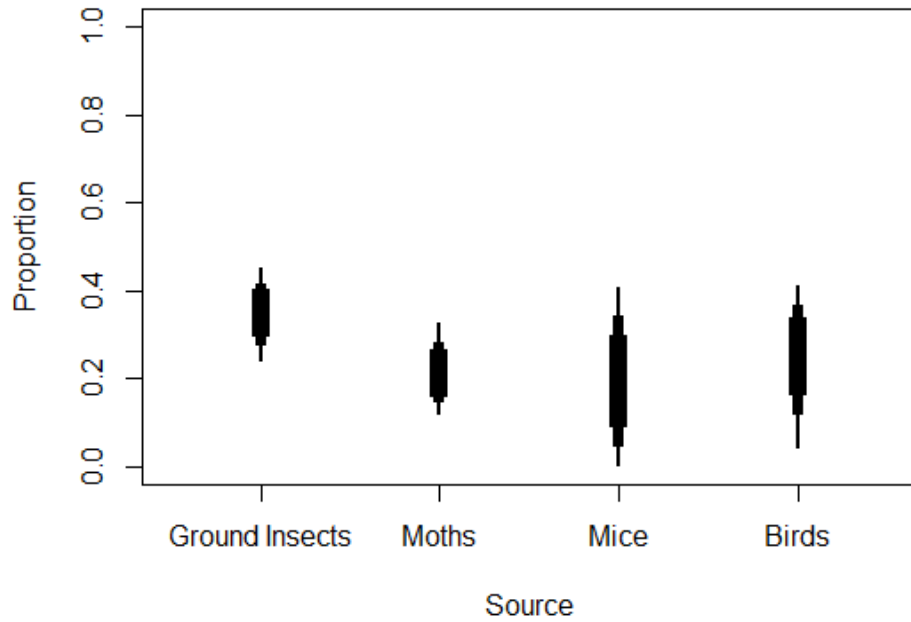


Figure 8. Proportional diets of Oriental Scops Owls estimated by a multi-source mixing model. Line thickness indicates 50%, 75%, and 95% probability.

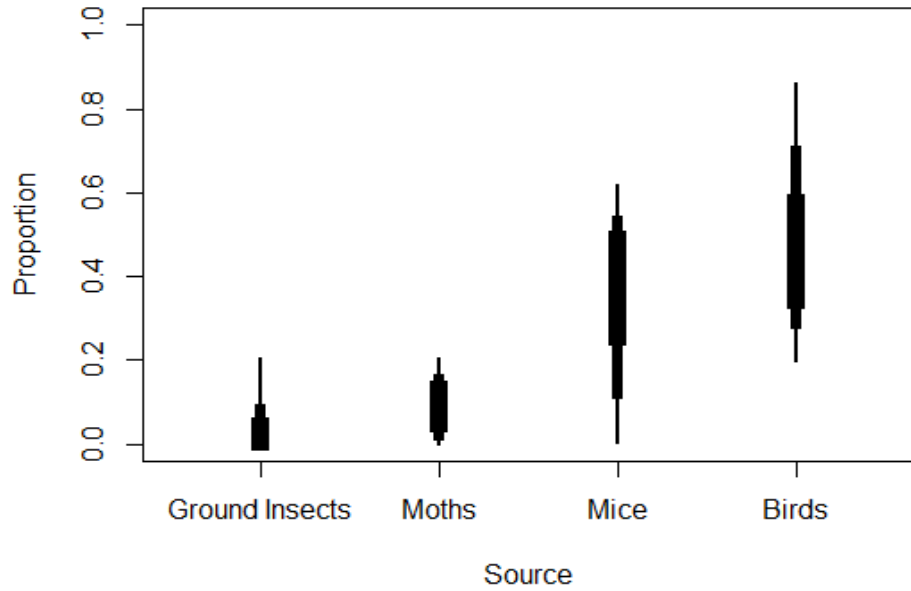


Figure 9. Proportional diets of Northern Boobooks estimated by a multi-source mixing model. Line thickness indicates 50%, 75%, and 95% probability.

4.3.3 Body size and trophic level

The first principal component (PC1) of each species was calculated as an index of overall body size for the subsequent correlation analysis with nitrogen stable isotope ratios. PC1 of Oriental Scops Owls explained 82% of the variation, with wing length having highest positive loading, followed by tail length. Northern Boobooks also had wing length with the highest positive loading, and tail length with the second highest positive loading in PC1, with 71% of variation explained (Table 7).

In Oriental Scops Owls, and especially in 10 individuals from Gwang-reung Forest (male: $n=8$, female: $n=2$), a negative relationship between body size and trophic level was detected ($n = 10$, $r = -0.709$, $P = 0.019$), suggesting that smaller scops owls have a higher trophic level (Figure 11); and while it was statistically not significant, males also had negative correlation coefficients ($n = 8$, $r = -0.476$, $P = 0.207$).

On the other hand, eight Northern Boobooks sampled from the same study area (Figure 10) exhibited no relationship between trophic level and body size ($n = 8$, $r = -0.143$, $P = 0.720$). The sexes were pooled in the analysis due to the small sample size (male: $n = 2$, female: $n = 6$).

Table 7. Results of principal component analysis of Oriental Scops Owls and Northern Boobooks (HC: hind claw length, BN: bill length to nostril, BS: bill length to skull, BW: bill width, BD: bill depth).

	Oriental Scops Owls (n=10)		Northern Boobooks (n=8)	
	PC1	PC2	PC1	PC2
Eigenvectors				
Wing (Max)	0.865258	0.385766	0.912194	-0.33996
Tail	0.461345	-0.79025	0.354196	0.767705
Tarsus	0.0543	0.470741	0.173533	0.33377
Hindclaw	0.041091	-0.03119	0.014598	0.038715
Bill length to nostril	0.079433	-0.01224	0.029698	0.106587
Bill length to skull	0.107248	0.027186	-0.00444	-0.05105
Bill width	0.069028	0.011313	0.059146	0.039099
Bill depths	0.071537	0.05564	0.021543	0.200122
Head	0.078477	-0.00245	-0.08519	0.355829
Eigenvalue	34.02604	5.045944	16.50558	3.956695
Variance explained				
Proportion	0.8236	0.1221	0.7101	0.1702
Cumulative	0.8236	0.9457	0.7101	0.8803

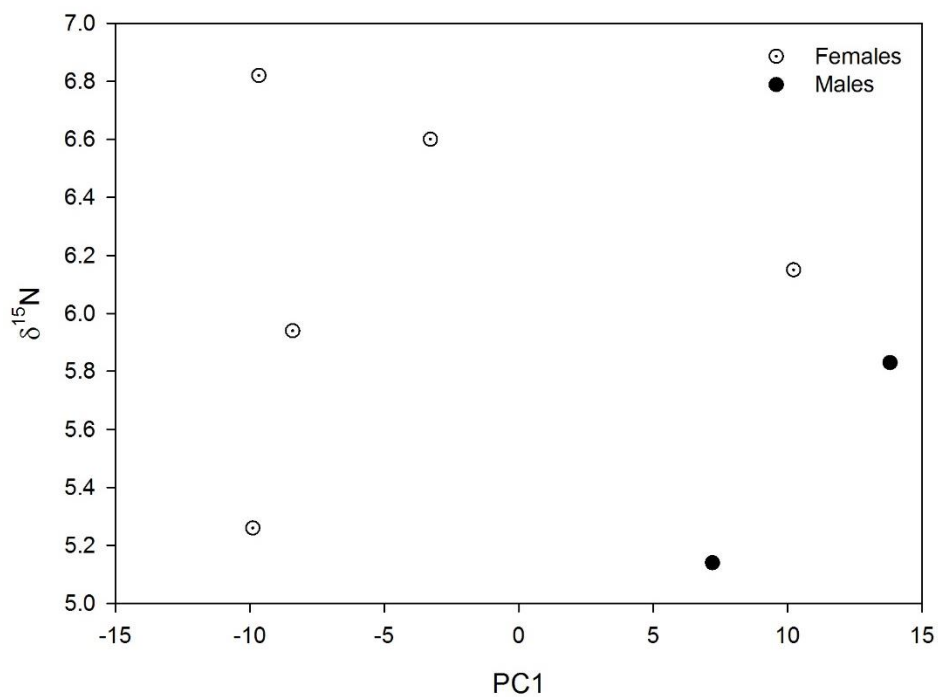


Figure 10. Scatterplot of overall body size index (PC1) and individual trophic level ($\delta^{15}\text{N}$) in Northern Boobooks (*Ninox japonica*)

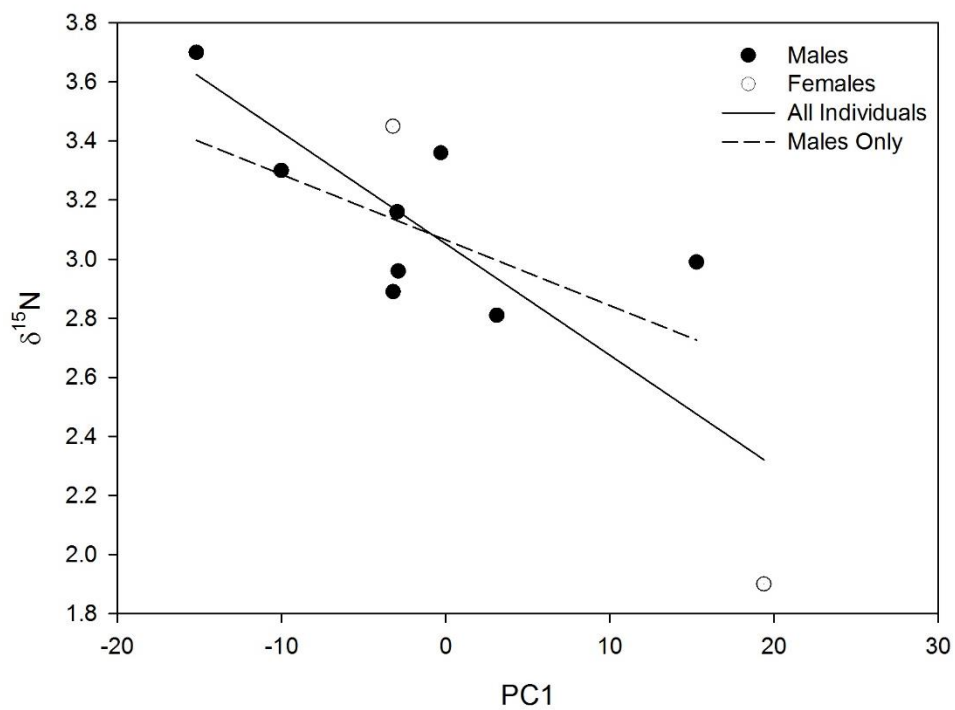


Figure 11. Relationship between overall body size index (PC1) and individual trophic level ($\delta^{15}\text{N}$) in Oriental Scops Owls (*Otus sunia*).

Chapter 5. Discussion

5.1 Sexual size dimorphism in owls

Size dimorphism in birds has been of interest to many ecologists for a long period, for many reasons. Not only is size dimorphism of interest in evolution and functional ecology, but it is also a useful method for field researchers seeking to identify the sex of monomorphic plumaged birds directly with morphometric information.

I assessed sexual size dimorphism in Oriental Scops Owls and Northern Boobooks in Korea, and attempted to generate discriminant functions for morphometric sex identification in field situations. Contrary to the general conception that owls, like diurnal raptors have reversed sexual size dimorphism (RSD), the Northern Boobooks in this study did not show clear sex-related size dimorphism. However, Oriental Scops Owls showed slight RSD as expected from the literature (Seo and Park 2008, Mikkola 2013). Although I failed to detect statistically significant size dimorphism in Northern Boobooks at the typical statistical significance level of 5%, some characters such as tail ($DI = -0.25$, $P = 0.090$), head length ($D = -1.37$, $P = 0.084$) and weight ($DI = -12.03$, $P = 0.07$) likely showed the normal size dimorphism (larger male) having negative dimorphic indices, which was not reported in previous studies (Seo and Park 2008, Mikkola 2013).

This unexpected similarity between the sexes in Northern Boobooks may be related to their taxonomic group, the genus *Ninox*, which includes at least nine species with normal size dimorphism which males are larger or heavier. Three large *Ninox* species show great amount of male-biased sexual size dimorphism: Rufous Owls (*Ninox rufa*; weight range: 700–1300 g, males up to 300 g heavier and larger than females), Powerful Owls (*N. strenua*; 1050–1700 g, males larger and 250 g heavier than females), and Barking Owls (*N. connivens*; 425–510 g; males 100 g heavier than females; Mikkola 2013), and six other smaller species are believed to show normal sexual size dimorphism, though there is no strong quantitative evidence. However, there are also two small species in this genus that shows female-biased sexual size

dimorphism, which are Southern Boobooks (*N. boobook*; 146–360 g, females 65g heavier and larger than males) and Moreporks (*N. novaeseelandiae*; 150–216 g, females 15 g heavier than males Mikkola 2013). The inconsistency in the dimorphism of *Ninox* species leave us in perplexing situation of the common conception on reversed sexual dimorphism in owls. In spite of the diverse forms of sexual size dimorphism in the genus, Northern Boobooks (*N. japonica*) have little evidence of any type of sexual size dimorphism. Resident breeding Northern Boobooks in Taiwan (*Ninox japonica totogo*) were found to be sexually monomorphic in size (10 pairs, Lin *et al.* 2012), as in this study. Recent studies also revealed that there is no morphometric difference between resident populations in Taiwan (n = 9) and migratory populations (*Ninox japonica japonica*, n = 7) in spite of their different genetic lineages (Lin *et al.* 2013), suggesting that the size difference between subspecies is not clear in my study species, Northern Boobooks. Thus, based on this study using birds in Korea, as well as previous studies, I suggest that there is no robust evidence of reversed sexual size dimorphism in Northern Boobooks, at least at the species level, but males still have the potential of having slightly bigger body parts in terms of weight and length of head, wings, and tails.

The sexual dimorphism of Oriental Scops Owls, by contrast, is less controversial than that of Northern Boobooks. Mikkola (2013) suggests that females are on average 6g heavier than males in Oriental Scops Owls (generally ranging from 75g to 95g). In this study, the weight difference between sexes was greater, averaging 13 g difference in average.

These two owl species do not possess any plumage difference between the sexes, and thus, this study of morphometric sex identification may be useful in field applications. Furthermore, except tail length, all other selected external characters are hard, fixed structures which change less during specimen preparation and storage, and thus this discriminant formula can be used with museum skins also. During the trapping sessions, I was able to observe females that had developed brood patches. Since the sexual dimorphism is not strong enough to classify the birds into males or females with certainty, the application of additional clues such as the development of brood patches can support sex identification in the field. However, given the high

error rates of the developed discriminant functions for the two forest owls, it is generally recommended to consider using molecular tools for reliable sex determination in both species.

5.2 Trophic ecology of owls

Clear inter-specific differences in isotopic values confirmed in this study indicate a separation in foraging niche of these two sympatric forest owls. In particular, Northern Boobooks showed higher nitrogen isotopic values, than Oriental Scops Owls. Typically, $\delta^{15}\text{N}$ enrichment of 1.4‰ to 3.3‰ is observed from diet to consumer along increasing food chains, allowing determination of an organism's trophic level and food chain length (Perkins *et al.* 2014). In this study, Northern Boobooks had *circa* 3‰ higher value for nitrogen than Oriental Scops Owls, suggesting that the boobooks to occupy a higher trophic level than the scops owls, and have a longer food chain length by one or two steps.

Similarly, dietary reconstruction models based on stable isotope analysis results explained that the Northern Boobooks used more vertebrate prey (birds and rodents together; 82%) than invertebrates, especially birds. However, Oriental Scops Owls used only 43% of vertebrates prey in diets. Although these proportion of vertebrate prey consumption in both species seems to be higher than the known proportion in previous studies (in % number of items, Northern Boobooks: 0.7-1.9 % in Taniguchi 1983, and 0.06%; Noguchi 2002; Oriental Scops Owls: 0.4% in Noh 2013), it is impossible to have a direct comparison between my study and previous ones, due to the different measures that were used in the study.

Panzeri *et al.* (2014) found significant difference in diet composition of Eurasian Scops Owls in their wintering range, that winter diets have higher percentage of vertebrates. Seasonal differences in diet of Northern Boobooks in Japan were also reported by Taniguchi (1983) that may occurred by seasonal difference in prey availability along season. For instance, larger beetles and moths may not emerge until after than my study period (Bae 1998, Kim 1998, Lee and Kim 2010), causing the limited proportion in this study but the increased amount in the previous studies.

Still, we need to consider the difference between methods used in this study and the previous ones. In general, diet analysis from SIA indicates assimilated contributions

of prey, which is related to the mass of diet consumed, while diet analysis from counting prey items from pellets, prey remains, or field observations are direct numbers caught by the birds (Resano-Mayor *et al.* 2014b). Vertebrate prey may contribute more biomass to the consumer after a single hunt than smaller invertebrates with indigestible exoskeletons, and even the smaller number of vertebrate sources can contribute more to blood stable isotope ratios than a large number of insects. Therefore, this study evaluated the relative dietary significance of different groups in terms of overall diet assimilation, whereas the previous studies noted only the occurrence and frequency of used prey items in diets, resulting in different estimates of vertebrate use in these species. Further studies, comparing two different methods and their results are required to estimate the effects of each diet item on assimilated isotopic values. In other comparative studies of sympatric owl diets, larger species consumed larger vertebrate preys more often, and smaller species relied on more invertebrate prey sources (Toyama and Saitoh 2011, Zhao *et al.* 2011), which was in concordance of estimation in this research .

Northern Boobooks showed greater carbon isotope variation with a higher CV (4.49%), which implies that they consumed foods from diverse carbon sources. On the other hand, Oriental Scops Owls showed smaller variation in carbon stable isotope ratio (CV=0.98%) among individuals. In general, plants growing under dense canopy with lower light intensity and more recycling of biogenic CO₂ have lower ¹³C/¹²C ratios than plants in open environments (Drucker and Bocherens 2009). Also, Trees as carbon sources in temperate forests are mostly C3 plants, whereas all C4 plants recorded in Korea are herbaceous species that prevail in open habitats rather than closed-canopy forests (Kim *et al.* 2011). These two different photosynthetic pathways, Calvin cycle (C3) and Hatch-Slack (C4) cycle, have profound difference (*circa.* 11.0‰ higher in C4 plants) in isotopic discrimination of ¹³C during photosynthesis (Marshall *et al.* 2007), and perhaps greatest source of variation within the study area. In addition, leaf δ¹³C along a vertical profile of the forest increases by canopy height as assimilation rate increases (Hanba *et al.* 1997). In the context of these vertical and horizontal profiles of primary producer carbon sources in forest food webs, the difference in carbon isotope ratios between Northern

Boobooks and Oriental Scops Owls could be explained by the Northern Boobook's utilization of prey from more diverse trophic webs, ranging from closed-canopy forests to carbon enriched open habitats. On the contrary, Oriental Scops Owls in this study area mainly relied on carbon sources from a very confined ecosystem in forests, not extending to open habitats.

5.3 Body size and trophic ecology

Most raptors show reversed sexual size dimorphism (Mueller 1986). Among multiple hypotheses to explain the RSD in raptors, Kruger (2005) suggested that diet might have a strong influence on the evolution of reversed sexual dimorphism: females prefer to select smaller, more agile, and more efficient males for breeding. This implies the avoidance of inter-sexual competition for limited prey resources in their territories, and it also means that the males have to be more efficient in foraging to supply food for chicks and brooding females.

Because of the efficient flight of smaller bodied birds by that have lower weight and hence lower wing loading, being small while foraging may be beneficial to individual adaptation in terms of energy efficiency in foraging (Andersson and Norberg 1981). In Northern Goshawks (*Accipiter gentilis*), smaller body size allowed birds to take an abundance of smaller and more agile prey (small birds and mammals) in Finland (Tomberg *et al.* 1999). In vole-eating raptors, Eurasian Kestrels (*Falco tinnunculus*) (Hakkarainen *et al.* 1996, Massemin *et al.* 2000) and Tengmalm's Owls (Hakkarainen and Korpimäki 1991), pairs with smaller males had more productive breeding results in low vole years when prey availability decreases. Because smaller male kestrels handed more voles to females during courtship during low vole years, small size in males can be more sexually selective trait in pair formation (Hakkarainen *et al.* 1996). In Tengmalm's Owls in Finland, it is suggested that smaller males may be preferred by females, since they are more efficient in foraging and territorial defense, hence can provide better prey items to the females (Korpimäki 1986). All these examples support sexual selection hypothesis that larger dominant females prefer to select smaller energy-efficient males.

In this study, smaller Oriental Scops Owls had higher nitrogen isotopic signatures, suggesting that the smaller birds consumed prey from higher trophic levels than larger individuals. Although the correlation was not statistically significant in the males only, due to the small sample size, if the smaller body size itself is advantageous in capturing larger and more vigilant vertebrate prey, it may

work as same in both males and females. On the contrary, no relationship between trophic level and body size (PC1) in Northern Boobooks was found, not supporting the hypothesis that smaller individuals may prey on higher trophic level preys. This lack of relationship may be linked with their sexual size monomorphism.

These findings support the general hypothesis that reversed sexual size dimorphism is related to foraging or diets (Kruger 2005). Individual trophic level was related to body size only in sexually dimorphic Oriental Scops Owls, but not in monomorphic Northern Boobooks. My study on the size dimorphism and trophic level did not include any aspects of breeding success or sexual selection, but did successfully find the trophic level of each individual via stable isotope analysis method. However, the results of this study may help to explain why female Oriental Scops Owls select smaller males, which are at higher trophic levels (a potential index of higher hunting skills and better chick provisioning) causing reversed sexual dimorphism. Similarly, no difference in trophic condition by size may responsible for the sexual size monomorphism of boobooks. However, given the small sample sizes and the strongly sex-biased samples (in particular, females in boobooks), the relationship between sexual dimorphism and individual trophic condition by body size still remains unanswered. Nevertheless, the findings in this study provide a fundamental clue to understand the relationship between foraging and size dimorphism in small, sympatric nocturnal raptors in East Asia.

Chapter 6. Conclusions

Of the two examined species of small forest owls, only Oriental Scops Owls showed significant, but subtle, reversed sexual dimorphism. In contrast, no clear evidence of sexual dimorphism was found in Northern Boobooks, though there were subtle sexual differences in some morphometric parameters. I developed a sex discriminant function based on external measurements for each species that excludes sex-related plumage differences. Based on re-substitution and leave-one-out tests, error rates for discriminant functions of 20.39–20.78% and 30.26–30.65% were estimated for Oriental Scops Owls and Northern Boobooks, respectively. Given the high error rates, I suggest using this formula as a supplementary method to identify the sex of a bird correctly in the field.

Stable isotope analysis showed a clear separation between the two species, both in carbon and nitrogen isotope ratios. Northern Boobooks had a higher trophic level (3‰, one level) than Oriental Scops Owls, and they also utilized prey sources from more diverse ecosystems. The diet of Northern Boobooks consisted of more vertebrate prey, especially birds, while Oriental Scops Owls utilized ground insects the most followed by birds, mice and moths. These results suggest that these two sympatric owls in Korean forests occupy clearly different ecological niches in terms of foraging, and that Oriental Scops Owls could be considered as forest specialists while Northern Boobooks are generalist predators utilizing both forest ecosystems and open habitat ecosystems when hunting for prey. High proportion of vertebrate preys estimated from the blood stable isotope ratio infer the significance of vertebrate prey to their diet, although their number may be much lower than invertebrate preys.

Finally, the negative relationship with the body size and trophic level of individual Oriental Scops Owls supported the previous arguments of smaller and more efficient-hunting males that can hunt for larger, and thus higher trophic level prey. But this was not the case in Northern Boobooks, which individual body size did not correlated with individual trophic level. This study does not include sufficient sample size to generalize the results, but it may partially support the hypothesis of

smaller and better males in raptors with sexual dimorphism.

In this study, stable isotope analysis in terrestrial high-lever consumer successfully identified different food niche between species that are known to have similar food habit. And I suggest that the utility of stable isotope analysis as a measure of individual trophic level can be a useful tool for studying the relationship between morphometric and individual diet.

This study is limits its scope on the food of the birds to explain the evolutionary fitness of reversed sexual dimorphism with only a handful of individuals. I suggest that the presence and absence of sexual dimorphism in sympatric two owl species with separated, but relatively close food niche, make them excellent models to study the inter-species interaction and driving force of the sexual dimorphism. Therefore, further study with more comprehensive approach, in example, study of sex roles during breeding, foraging mode and behavior, and comparison with species would be needed to increase our knowledge of these two owl species and general concepts of sexual dimorphism and intra-guild interaction between nocturnal raptors.

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Appendix

Appendix 1

List of Potential prey items collected.

Prey type	Common name	Scientific name	# of samples
Mice	Striped Field Mice	<i>Apodemus agrarius</i>	3
Moths	Hawk moth	<i>Phillosphingia dissimilis</i>	1
	Noctuid moth	<i>Xestia</i> sp.	1
Bush crickets	Ussur Brown Katydids	<i>Phillosphingia dissimilis</i>	3
	Katydid sp.	<i>Eobiana engelhardti</i>	1
Beetles	Adams stag-horned beetles	<i>Dicranocephalus adamsi</i>	3
	Carabus beetle	<i>Carabus smaragdinus</i>	1
	Stag beetles	<i>Dorcus rectus</i>	2
	Chafer beetles	<i>Melolonthinae</i>	2
Birds	Vinous-throated Parrotbill	<i>Sinosuthora webbiana</i>	1
	Eastern Great Tit	<i>Parus minor</i>	1
	Tristram's Bunting	<i>Emberiza tristramii</i>	1
	Pale Thrush	<i>Turdus palidus</i>	1

국문 초록

한국에서 서식하는 소쩍새(*Otus sunia*)와

솔부엉이(*Ninox japonica*)의

성적 이형성과 먹이-니치 연구

소쩍새와 솔부엉이는 한국에서 번식하는 비교적 흔한 소형 산림성 맹금류임에도 불구하고 두 종의 형태적 특징 및 생태에 대한 정보는 매우 부족한 실정이다. 본 연구에서는 두 종의 성적 이형성을 파악하고, 안정성동위원소 분석을 통하여 초기 번식기 동안의 먹이-니치 분할에 대하여 파악하고자 하였다. 그리고 이를 바탕으로 개체별 몸 크기와 먹이이용 관계를 비교하여, 맹금류의 역전된 성적 이형성을 진화적 관점에서 고찰하였다. 2014년과 2015년 두 조사기간 동안 광릉숲과 서울대학교 관악수목원에서 30개체의 소쩍새와 10개체의 솔부엉이를 포획하여 외부 형태를 측정하고, 성식별과 안정성동위원소 분석을 위한 혈액과 깃털을 수집 후 방사하였다. 추가적으로 4곳의 야생동물 구조센터에서 폐사하거나 구조 후 계류중인 솔부엉이 22마리와 소쩍새 16마리에서도 외부 형태를 측정하고 성식별을 위한 조직을 수집하였다. 또한 다원혼합모형분석을 위한 조류, 설치류, 나방류, 및 육상 곤충류 등 잠재적인 먹이원도 수집하여 분석하였다. 두 종의 외부형태를 분석한 결과 소쩍새는 미미하나 통계적으로 유의한 역전된 성적 이형성이 나타난 반면, 솔부엉이에서는 성적 이형성이 나타나지 않았다. 측정치를 통한 판별식 산출 결과, 소쩍새의 경우, 부리높이, 부리 폭, 꼬리길이, 부척길이가 유의한 단계적 판별분석의 변수로 선정되었으며, 판별분석을 통해 동일 표본에서 79%의 성이 올바르게 판별되었다. 솔부엉이는 머리길기와 두개골부터의 부리길기가 단계적 성판별분석에 선정되었으며, 동일표본의 70%만 올바르게 판별되었다. 두 종 모두 판별분석의 정확도가 낮고, 성적이형성이 없거나 차이가 크지 않으므로, 성 식별 시 포란반 등 추가적인 정보를 참고해야 하

며, 가능하다면 분자생물학적 방법이 이용되어야 할 것으로 보인다. 혈액 안정성동위원소 분석 결과, 질소 안정성동위원소비가 솔부엉이가 소쩍새에 비해 높게 나타나 솔부엉이가 소쩍새보다 높은 영양단계의 먹이를 이용하는 것으로 나타났다. 탄소 동위원소비의 경우, 탄소 동위원소비 뿐만 아니라, 동위원소비의 분산도 솔부엉이가 높게 나타나, 솔부엉이가 산림 내부에서부터 외부까지 더 다양한 생태계로부터 먹이원을 이용하는 유연한 섭식 행동을 하는 것으로 추정되었다. 이러한 명확한 안정성동위원소의 차이는 동일 지역에서 서식하는 소쩍새와 솔부엉이간 명확한 먹이-니치 분할이 이뤄진 결과로 판단된다. 다원혼합모형결과, 솔부엉이는 특히 조류를 포함한 척추동물 먹이를 주로 소비한 것으로 추정되었으며, 소쩍새는 육상 곤충을 가장 많이 소비하였고 조류, 설치류, 나방류를 다음으로 많이 소비하는 것으로 추정되었다. 이는 기존연구에 비해 척추동물의 비율이 매우 높은 결과로서, 조사 시기 및 연구 방법에 의한 것으로 판단된다. 개체 별 크기와 영양단계간 비교를 위해, 주성분분석을 이용하여 추출한 개체의 몸 크기를 대표할 수 있는 제 1 주성분을 개체의 몸 크기를 대표하는 지수로 삼아 개체의 질소 안정성동위원소비와 상관분석을 실시하였다. 분석결과, 작은 개체가 높은 영양단계의 먹이를 소비한 것으로 분석된 소쩍새와 달리 솔부엉이는 상관성을 나타내지 않았다. 성적이형성이 나타난 소쩍새에서만 개체의 영양단계와 몸 크기의 상관성이 나타났다는 점에서, 이는 소쩍새에서 작은 크기의 수컷이 더 효율적인 사냥을 통해 진화적으로 역전된 성적이형성을 강화한다는 ‘작은 수컷 가설’을 부분적으로 지지한다고 볼 수 있다.

주요어: 먹이원 재구성, 안정성 동위원소 분석, 역전된 성적이형성, 영양 단계, 작은 수컷 가설, 판별 분석

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농학석사 학위논문

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2015 년 8 월

서울대학교 대학원
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Abstract

Two small forest owls, the Oriental Scops Owl (*Otus sunia*) and the Northern Boobook (*Ninox japonica*), are among the common breeding raptors in the Republic of Korea. However, very limited ecological information is available on these nocturnal raptors. In this study, I focused on two aspects of these owls' ecology: sexual dimorphism and food-niche partitioning during their early breeding period. The morphometric study was aimed to understand sexual dimorphism in these two small owls through external measurements, and to develop sex-discriminant functions for both species. The study of food-niche partitioning of two sympatric owls used stable isotope analysis (SIA) to determine trophic levels and estimate prey type selection. From both morphometric information and individual SIA results, I questioned how body size can effect individual prey use, and further its relationship with the evolution of reversed sexual dimorphism in raptors. From April to May in 2014 and 2015, 30 Oriental Scops Owls and 10 Northern Boobooks were captured using mist nets after sunset between 19:30 to 00:30 at Gwang-reung Forest and Gwan-ak Arboretum in Gyeonggi-do province. Blood samples and measurements were taken from captured owls. Moreover, 16 scops owls and 22 boobooks admitted to or kept in four wildlife rescue centers were measured and sampled for the morphometric study. For diet reconstruction, potential prey items (ground insects, moths, mice, and birds) were collected and analyzed to estimate the diet composition of each owl species through multi-source mixing model. In terms of morphology, Oriental Scops Owls showed significant but subtle reversed sexual dimorphism whereas, Northern Boobooks had no clear evidence of sexual dimorphism. Bill depth, bill width, tail length and tarsus length were selected for the stepwise discriminant function in Oriental Scops Owls, and they sexed of 79% the birds correctly by re-substitution. The stepwise discriminant function for Northern Boobooks using head length and bill length to skull, correctly classified the sexes with about 70% accuracy. Given the low power of morphometric sex discrimination as well as non-obvious sexual size dimorphism, additional techniques such as examination of brood patch or even molecular methods, are required for reliable sex determination. SIA showed

significant distinctions between the two species, both in carbon and nitrogen isotope ratios. Northern Boobooks had higher values in nitrogen and carbon isotope ratios, and greater variance in carbon isotope ratios than Oriental Scops Owls. According to the multi-source mixing models, Northern Boobooks mainly consumed vertebrate prey, especially birds, but Oriental Scops Owls utilized mainly ground insects followed by birds, mice, and moths. Each individual owl's overall body size index was extracted from principal component analysis, and compared with individual trophic level (nitrogen isotope ratio). Only Oriental Scops Owls showed significant correlation with smaller individuals consuming higher trophic level diets. In conclusion, Northern Boobooks and Oriental Scops Owls demonstrated distinct food niche separation in a sympatric habitat. In particular, the larger bodied Northern Boobooks occupied a higher trophic level with more flexible foraging habits, and they selected diverse prey sources from forests to open environments, whereas the smaller Oriental Scops Owl belonged to a lower trophic level and was a more specialized forager relying on forest ecosystems. Furthermore, individual trophic level was found to be related to the body size only for the sexually dimorphic Oriental Scops Owls, but not in the monomorphic Northern Boobooks. These findings partially support the 'small male hypothesis', in Oriental Scops Owls that smaller males are efficient foragers which is beneficial in reproductive success and thus more adaptive in raptors with reversed sexual dimorphism.

Keywords: diet reconstruction, discriminant function analysis, reversed sexual dimorphism, small male hypothesis, stable isotope analysis, trophic level

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Table of Contents

Abstract.....	i
Table of Contents.....	iii
List of Tables	v
List of Figures.....	vi
Chapter 1. Introduction.....	1
Chapter 2. Literature Review.....	4
2.1 Study species	4
2.2 Reversed sexual size dimorphism in raptors	6
2.3 Food-niche and diet studies in raptors.....	8
Chapter 3. Materials and Methods.....	11
3.1 Study sites and period.....	11
3.2 Sample collection.....	13
3.3 Data analysis	16
3.3.1 Molecular sex identification.....	16
3.3.2 Stable isotope analysis	17
3.3.3 Statistical analysis	19
Chapter 4. Results.....	22
4.1 Sampling and molecular sex identification.....	22
4.2 Morphology and sexual size dimorphism.....	23
4.2.1 Discriminant analysis	28
4.3 Stable isotope ecology	32
4.3.1 Stable isotope ratio analysis	32
4.3.2 Diet reconstruction of owls	35
4.3.3 Body size and trophic level	37

Chapter 5. Discussion	40
5.1 Sexual size dimorphism in owls	40
5.2 Trophic ecology of owls.....	43
5.3 Body size and trophic ecology	46
Chapter 6. Conclusions.....	48
References	50
Appendix	59
국문 초록	60

List of Tables

Table 1. Morphometric parameters of Oriental Scops Owls in the Republic of Korea. Asterisks indicate significant differences (* $P<0.05$, ** $P<0.01$, *** $P<0.001$). CV and DI represent the coefficient of variance and the dimorphic index, respectively.	24
Table 2. Morphometric parameters of Oriental Scops Owls in the Republic of Korea. Asterisks indicate significant differences (* $P<0.05$, ** $P<0.01$, *** $P<0.001$). CV and DI represent the coefficient of variance and the dimorphic index, respectively.	25
Table 3. Morphometric characteristics of Northern Boobooks in the Republic of Korea. Asterisks indicate significant differences (* $P<0.05$, ** $P<0.01$, *** $P<0.001$). CV and DI represent the coefficient of variance and the dimorphic index, respectively.....	26
Table 4. Morphometric characteristics of Northern Boobooks in the Republic of Korea. Asterisks indicate significant differences (* $p<0.05$, ** $p<0.01$, *** $p<0.001$). CV and DI represent the coefficient of variance and the dimorphic index, respectively.....	27
Table 5. Nitrogen and carbon stable isotope values for Oriental Scops Owls and Northern Boobooks.....	33
Table 6. Stable isotope values of potential prey items of Oriental Scops Owls and Northern Boobooks in the Republic of Korea.	34
Table 7. Results of principal component analysis of Oriental Scops Owls and Northern Boobooks (HC: hind claw length, BN: bill length to nostril, BS: bill length to skull, BW: bill width, BD: bill depth).....	38

List of Figures

Figure 1. The location of wildlife rescue centers and trapping sites. Numbers indicate following locations. 1: Gwang-reung Forest, 2: Gwanak Arboretum, 3: Gyeonggi Wildlife Rescue Center, 4: Ulsan Wildlife Rescue Center, 5: Gyungseong University Bird Museum, 6: Busan Wildlife Rescue Center.	12
Figure 2. Sample image of gel electrophoresis showing females (two bars) and males (single bar).	22
Figure 3. Histogram of discriminant scores (D) for Oriental Scops Owls.	28
Figure 4. Relationship between discriminant scores and posterior probability of being a male of Oriental Scops Owls. Dashed lines indicate 95%, 75%, 25%, and 5% probability cut offs, and the solid line indicates a 50% cut-off of discriminant score to be a male.	29
Figure 5. Histogram of discriminant scores of Northern Boobooks.	30
Figure 6. Relationship between discriminant score and posterior probability of being a male in Northern Boobooks. Dashed lines indicate 95%, 75%, 25%, and 5% probability cut offs, and the solid line indicates a 50% cut-off of discriminant score to be a male.	31
Figure 7. Distribution of carbon and nitrogen stable isotopes in Oriental Scops Owls, Northern Boobooks, and potential prey items.	33
Figure 8. Proportional diets of Oriental Scops Owls estimated by a multi-source mixing model. Line thickness indicates 50%, 75%, and 95% probability.	36
Figure 9. Proportional diets of Northern Boobooks estimated by a multi-source mixing model. Line thickness indicates 50%, 75%, and 95% probability.	36
Figure 10. Scatterplot of overall body size index (PC1) and individual trophic level ($\delta^{15}\text{N}$) in Northern Boobooks (<i>Ninox japonica</i>)	39
Figure 11. Relationship between overall body size index (PC1) and individual trophic level ($\delta^{15}\text{N}$) in Oriental Scops Owls (<i>Otus sunia</i>)	39

Chapter 1. Introduction

Raptors are high-level consumers in trophic webs and play important ecological roles in ecosystems (Ferguson-Lees and Christie 2001). However, their position as high-level consumers in ecosystems makes them prone to threats such as bioaccumulation of toxic elements, decline of food sources and habitat fragmentation. In addition, most raptors species in the world (66%) are migratory, which increases their vulnerability during their migratory life cycle (Bildstein 2006).

Raptors are generally hard to detect and observe, since they usually have low densities due to their habitat requirements and territoriality, elusive behavior, and often inaccessible of remote habitats (Bildstein and Bird 2007). These features make raptor research difficult, especially obtaining enough a large sample size, in spite of the necessity of understanding ecology of raptors. On the other hand, their relatively large body size and high trophic level, with relatively simple intra-guild communities often make raptors a good model for studying ecological aspects of competitive interaction and resource use within intra-guild communities (Jaksic and Carothers 1985, Burton and Olsen 1997, Bildstein and bird 2007). Besides, reversed sexual size dimorphism, with females having a larger body size than males, is a common phenomenon in diurnal and nocturnal raptors. The ecology and evolution of reversed sexual dimorphism in raptors intrigued numerous researcher to study raptors as a model species, but it never reached to a consensus so far.

Including all the falcons (Falconiformes), hawks (Accipitriformes) and owls (Strigiformes), total 546 raptors species are known to the world currently, and 221 species of owls, are mainly nocturnal birds of prey. Owls are difficult to observe because of their nocturnal behavior, and most of them use densely covered roosts during the daytime. However, their habit of casting pellets of indigestible materials, enabled us to study their diet more efficiently than other raptor species (Marks *et al.* 1999).

Oriental Scops Owls (*Otus sunia*) and Northern Boobooks (*Ninox japonica*) are two small, primarily insectivorous owls that breed in Korea during summer (Marks *et al.* 1999, Lee *et al.* 2000, Brazil 2009). These two migratory owls are the most

common owls in forests and forested patches during summer in Korea. In spite of their relative abundance in comparison to other Korean raptors, there is very limited information available on the ecology of these two small forest owls, including the extent of their sexual dimorphism and the quantitative study on diet are available in Korea, to our current knowledge. Because Oriental Scops Owls and Northern Boobooks are both sympatric insectivorous owls in Korean forests during summer, one would expect that they shall have some separate their niche to some extent to avoid unnecessary competition on resources. Niche partitioning, by separating their foraging microhabitat and/or food is a common mechanism (Schoener 1974, Reynolds and Meslow 1984, Burton and Olsen 1997, Ricklefs 2012) in the community of coexisting organisms. In owls, the separation or the overlap of diets on coexisting species are well documented over the world (Earhart and Johnson 1970, Herrera and Hiraldo 1976, Kavanagh 2002, Ibarra *et al.* 2014), thanks to their pellet-regurgitating behavior.

However, in studying the diet of these insectivorous owls, it is difficult to use their pellet castings since they decompose easily (Bildstein and Bird 2007, Olsen 2011) during their breeding seasons which include heavy raining period in Korea, and are hence difficult to find in dense closed-canopy forests. Thus, to understand the diet of adult Northern Boobooks and Oriental Scops Owls while they forage for themselves, conventional methods are not applicable, or are extremely time consuming. The stable isotope approach in trophic ecology and food web studies has become more common in recent years (Hobson and Wassenaar 1999, Kelly 2000, Inger and Bearhop 2008, Carreon-Martinez and Heath 2010, Boecklen *et al.* 2011), because using blood or feathers is relatively less destructive than conventional stomach content analysis. This approach can also help to understand time-integrated diet information of individual birds by characterizing turnover rates of isotope ratios in tissues. Whole blood, for example, is known to represent the diet composition of birds for 3-5 weeks. Carbon and nitrogen isotopes are most commonly used in food web studies. Therefore, the stable isotope approach is suitable for studies of the diet of Northern Boobooks and Oriental Scops Owls.

In multiple species of raptors, which have reversed sexual dimorphism, one of the

most argued factor that influences the degree of sexual dimorphism is their diet (Krüger 2005). Smaller males are suggested to be more efficient in flight and hence better at foraging prey for chicks, as well as better in prey delivery during courtship feedings (Hakkarinen *et al.* 1996). Because owl can take only one prey at a time by their talons, it will be energetically beneficial to be able to hunt few high quality prey at one foraging trip than multiple mediocre quality preys during multiple foraging trip for hungry chicks. Most female raptors invest their effort in incubation and brooding, whereas males are the ones who dedicate their effort on foraging for incubating females and chicks (Storer 1966, Marks *et al.* 1999). So, the male's ability to be a good forager would be an important factor in the reproductive fitness of a pair. This hunting efficiency may work in the same manner to the individual diet, because individual birds also require certain amount of energy for survival and reproduction. Thus, I expect that there would be relationships between body size and individual diets of Oriental Scops Owls and Northern Boobooks.

In summary, the first objective of this study is to examine morphological characteristics of Northern Boobooks and Oriental Scops Owls in relation to their sex, to develop sex-discriminant functions applicable in field conditions, and describe the degree of sexual dimorphism of the two owl species.

Second, I assessed blood stable isotope ratio to compare the food-niche partitioning of Northern Boobooks and Oriental Scops Owls. In addition, the dietary composition of bloods of Northern Boobooks and Oriental Scops Owls were reconstructed by using multi-source mixing model based on potential prey samples.

And finally, I considered how individual trophic level is related to the body size and to the degree of sexual dimorphism in Oriental Scops Owls and Northern Boobooks.

Chapter 2. Literature Review

2.1 Study species

Oriental Scops Owls (*Otus sunia*) and Northern Boobooks (*Ninox japonica*) are small migratory owls that breed in Korea. These insectivorous owls are summer visitors that arrive in Korea from early April and start to breed in the mid of May (Seo and Park 2008, Noh 2013). The other four regularly breeding owl species in Korea, the Japanese Scops Owl (*Otus semitorques*), Ural Owl (*Strix uralensis*), Tawny Owl (*Strix aluco*), and Eurasian Eagle Owl (*Bubo bubo*), are resident and are scarcer (Lee *et al.* 2000, Seo and Park 2008, Brazil 2009), and all are known to prey primarily on vertebrates. Moreover, these four resident owls are known to breed earlier than the migratory Oriental Scops Owls and Northern Boobooks.

Oriental Scops Owls have a wide distribution range, and 7–9 subspecies are recognized currently. Northern populations, including *O. s. japonicus* that breed in Japan and *O. s. stictonotus* that breeds from Korea to Far Eastern Russia, are migratory. The Korean breeding subspecies, *O. s. stictonotus* is known to be the largest and heaviest subspecies (Brazil 2009, Mikkola 2013). There are two color morphs and a gradient of intermediate forms in Oriental Scops Owls. Between the gray-brown and rufous morphs, the gray or grayish-brown morph are more common in Korea (Seo and Park 2008).

Northern Boobooks, together with Chocolate (*Ninox randi*) and Brown Boobooks (*Ninox scutulata*), recently separated from the Brown Hawk Owl (King 2002), Northern Boobooks in South Korea and Japan belong to the race *N. j. japonica*, and while the race *N. j. florensis* ranges from North Korea to SE Siberia. The resident population in Taiwan, *N. j. totogo*, occupies the southern limit of this species. Northern Boobooks are sexually monomorphic in plumage (Mark *et al.* 1999, Brazil 2009, Mikkola 2013).

Neither of Northern Boobooks or Oriental Scops Owls has been well studied, but there are a few reports on the diet and breeding in Japan (Noguchi 2002, Taniguchi 1983), and diet and prey handling in Taiwan (Lin and Yeh 2004) of Northern Boobooks, although these studies were based on small sample sizes. Few

more studies on Northern Boobooks are done in Japan and Taiwan, including a study of incubation behavior of breeding owls using an automatic temperature logging system (Masuda 1974), and another study of breeding ecology and sexual dimorphism (Lin *et al.* 2012). Additionally, a cryptic separate lineage of sedentary population of Northern Boobooks among the migratory population were found in Taiwan (Lin *et al.* 2013). Calling behaviors were also recorded by King and Icarangal (2008) at wintering grounds and by Oba (1996) in breeding period

Oriental Scops Owls have been the subject of even fewer previous studies. Noh (2013) studied their breeding biology for his master's thesis and described their natural nests, eggs, and prey delivery during the chick rearing period. More studies have been done on Eurasian Scops Owls and Elegant Scops Owls, in Italy (Marchesi and Sergio 2005, Martínez *et al.* 2007) and in Japan (Toyama and Saitoh 2011), respectively.

2.2 Reversed sexual size dimorphism in raptors

Unlike most other birds and mammals, where males have larger body sizes than females, most nocturnal and diurnal raptors show reversed sexual size dimorphism, with females that are larger and heavier than males (Mueller 1986). This phenomenon raises the question of why female raptors have larger body size than males, a question that has been asked many times by numerous ornithologists in the past (Selander 1966, Storer 1966, Reynolds 1972, Shine 1989, Krüger 2005).

The difference in the roles of each sex during breeding, that with the females investing most of their time on incubation and chick guarding (Storer 1966, Marks *et al.* 1999, Eldegard and Sonerud 2012 Steen *et al.* 2012), and the males are delivering food for chicks and females, is believed to have an important effect on the evolution and maintenance of reversed sexual size dimorphism (Storer 1966, Reynolds 1972).

The functional advantage of members of a breeding pair having different body sizes has been examined with respect to both of the advantage of smaller males and the advantage of larger females. Larger females are expected to have an advantage in having a larger body that can act as an energy reserve for breeding (Lundberg 1986) and/or as a weapon for defending the nest against predators (Storer 1966, Andersson and Norberg 1981). Smaller males, are considered to have higher efficiency in foraging and thus in providing food for chicks and females, and thus, smaller males must also be advantageous in the reproduction of offspring (Koprimaki 1986, Sunde *et al.* 2003).

In Kruger's comparative review (2005), the strength of reversed sexual dimorphism was strongly related to prey-related and foraging-related factors in owls (Strigiformes) and hawks (Accipitriformes).

Raptors are single-loaded central place foragers while breeding, a strategy that makes it more efficient for them to carry one large prey item when they forage further. By contrast, it is more energetically efficient to catch smaller items more frequently close to the nest (Olsen 2013), which females must do as they guard their chicks.

Some owl species, like the Great Gray Owl (*Strix nebulosa*), Boreal Owl (*Aegolius funereus*), and Snowy Owl (*Bubo scandiacus*) show high level of sexual size

dimorphism, with females can weigh about 40% more than males (Josephson 1980).

In addition to the ecological and evolutionary aspects of sexual size dimorphism, researchers have been developed morphometric sex discriminant functions for diverse taxonomic group of birds (in owls: Eurasian Eagle Owl, del Mar and Penteriani 2004, Boreal Owl, Hayward and Hayward 1991, Eastern Screech Owl, Smith and Wiemeyer 1992), that do not have sexual plumage dimorphism and thus are difficult to identify the sex of the birds in the field. In such cases, size differences enable researchers to identify the sex of the live birds when caught, without additional molecular methods that are costly and often impossible in field studies at remote locations (del Mar Delgado and Penteriani 2004, Leppert *et al.* 2006).

In both Oriental Scops Owls and Northern Boobooks, the amount of sexual dimorphism is not well studied, and no discriminant function analysis has been done to best of my knowledge. There are several published notes on the sexual size differences of Oriental Scops Owls that show slight reversed size dimorphism (Marks *et al.* 1999, Seo and Park 2009, Mikkola 2013). However, the dimorphism of Northern Boobooks is disputed.

Won (1981) reported slightly larger wing lengths in males (212–213mm) than in females (209–211mm); and even though the data of Lin and Yeh (2004) do not show statistical significance, DI indicates slightly large average values for males.

2.3 Food-niche and diet studies in raptors

In studies of the diets of owls, there are several conventional methods, including direct observation, analysis of prey remains and stomach contents, pellet analysis, fecal analysis, DNA analysis, and combinations of these methods (Rosenberg and Cooper 1990, Lee and Severinghaus 2004). However, pellet analysis and fecal analysis can be biased toward certain prey types (Redpath *et al.* 2001, Sergio 2002), and DNA analysis for prey composition may characterize only the number of species fed upon, not their relative quantities. Direct observation and analysis of prey remains can be biased by the observer's and the bird's behaviors, allowing the observer to see only what can be easily seen.

In recent studies of avian trophic levels, stable isotope analysis (SIA) using carbon and nitrogen becoming more common. The stable isotope ratio of a consumer's tissue will reflect the diet's stable isotope ratio according to the rate at which each isotope is assimilated from the diet into the tissue. The ratio of nitrogen isotopes increases, about 3.4 ‰ per trophic level, and thus represents consumer-diet relationship well; and the carbon stable isotopes are trace elements of carbon sources from the primary producer level. It is known that C3 plants and C4 plants assimilate carbon isotopes differently, and thus have markedly different carbon isotope signatures (Fry 2007). Consequently, stable isotope ratios can be used to quantify the relative proportion of diet sources in consumer tissue, by using isotopic linear mixing models (Post 2002). However, even though stable isotope analysis to assess trophic ecology and resource partitioning has proven to be a powerful tool, it has rarely been used to assess the trophic ecology of terrestrial avian top predators (Resano-Mayor *et al.* 2014a).

As mentioned above, the diets of Northern Boobooks and Oriental Scops Owls are not well studied, but all reports emphasize the insectivorous diet of these owls. Noguchi (2002) reported only one vertebrate among 1442 prey items identified, from a breeding pair of Northern Boobooks (most prey items were insects), and Taniguchi (1983) reported 64 vertebrate prey remains (62 birds, a lizard, and a bat) along with 5968 insect prey remains from a pair of breeding Northern Boobooks during three

consecutive years (1979–1981). In Taiwan, Lin and Yeh (2004) reported 2269 discarded insect remains from Northern Boobooks around the streetlights over 2 months (November–December 1999). This difference between studies might be caused by seasonal availability of prey, which was also mentioned by Taniguchi (1983) who noted that birds were more often captured in the early part of the breeding season. Food brought to the nest of a Korean breeding pair of Oriental Scops Owls assessed by Noh (2013), also composed of 96% of invertebrate prey, 3.5% unidentified prey, and less than 1% vertebrate prey.

However, as Taniguchi (1983) mentioned, prey use may change greatly depending upon seasonal prey availability of study period. A study of an Italian population of Eurasian Scops Owls (*Otus scops*), which were once considered conspecific with Oriental Scops Owls, reported one lizard among 59 identified prey items from pellets collected during breeding period, while the rest of the items were invertebrates (Panzeri *et al.* 2014). During the wintering period, when the invertebrate prey resources decrease, a total of seven Shrews and mice were found in the pellets, whereas only six invertebrates were identified (Panzeri *et al.* 2014). As seen in this study in Italy, the Eurasian Scops Owl, the European counterpart of the Oriental Scops Owls, may also feed on vertebrate prey when invertebrate prey abundance is slow.

According to the competitive exclusion principle (Hardin 1960, den Boer 1986), species in the same habitat may not occupy exactly the same niches, in order to minimize competition (Jankowski *et al.* 2012). Accordingly, most studies on sympatric owl's diets were done with pellets (Marti 1974): Burrowing Owls (*Glucidium gnoma*), Saw-whet Owls (*A. acadicus*), vs. Western Screech Owls (*O. kennicotti*) (Hayward and Garton 1988); Little Owls (*Athene noctua*) vs. Long-eared Owls (*Asio otus*) (Zhao *et al.* 2011) Tawny Owls (*Strix aluco*) vs. Ural Owls (*S. uralensis*) (Pavon-jordan *et al.* 2013) and direct Observations Elegant Scops Owls (*Otus elegans*) vs. Japanese Scops Owls (*O. semitorques*) (Toyama and Saitoh 2011). In these studies, general pattern of niche separation was that the larger bodied owls utilized larger sized prey, but smaller owls preyed on smaller preys like small birds and invertebrates.

Stable isotope analysis has been relatively little used in owl studies. Two studies of Saw-whet Owls (*Aegolius acadicus*) and Snowy Owls (*Bubo scandiacus*) used stable isotopes to reveal the marine-origin of their prey (Hobson and Sealy 1991, Therrien *et al.* 2011b) and only one study revealed diet-tissue discrimination factor of Snowy Owl (Therrien *et al.* 2011a),

Chapter 3. Materials and Methods

3.1 Study sites and period

Field sampling was done at Gwang-reung Forest (37°44'42.55" N, 127° 9'35.90" E) and Seoul National University Gwanak Arboretum at Gwanak Mountain (37°25'12.12" N, 126°56'14.73" E), at Gyeonggi-do province, Korea, from May to June in 2014 and April to May in 2015 (Figure 1). Gwang-reung Forest is an old-growth temperate broadleaf forest which is under management of Korea National Arboretum, Korea Forest Service. Study area in Gwang-reung Forest consists of natural deciduous broadleaf forest and plantations located at an elevation of around 100-600m. Annual rainfall is around 1500mm, and average monthly temperatures are highest in August (29.1°C), and lowest in January (-13°C). Major tree species of the natural deciduous broadleaf stands in Gwang-reung Forest are *Carpinus laxiflora*, *C. cordata*, *Quercus serrata*, *Q. mongolica* and *Q. variabilis* (Korea National Arboretum 2011). Gwanak Arboretum and adjacent forests have annual rainfall of 1260mm, and annual average temperature is 12.4°C, highest in August (29.5°C) and lowest in January (-6.4°C). Common tree species in natural stands of Gwanak Arboretum and adjacent mountain forests are *Q. serrata*, *Q. mongolica*, *Q. variabilis*, and *Pinus densiflora* (Jang *et al.* 2013).

For the sexual dimorphism study, additional birds were sampled from Seoul National University (SNU)'s Gwanak Arboretum, and Gwanak Mountain forests adjacent to the SNU campus. Also, measurements of birds and DNA samples were collected from dead specimens and live individuals under rehabilitation at four wildlife rescue centers (Gyeonggi Wildlife Rescue Center, Busan Wildlife Rescue Center, Ulsan Wildlife Rescue Center and Kyungsoo University Bird Museum; Figure 1). For trapping and sampling birds for trophic ecology study using stable isotope ratio analysis, birds captured at Gwang-reung Forest and National Arboretum from 22 May to 5 June 2014 were only included in the analysis.

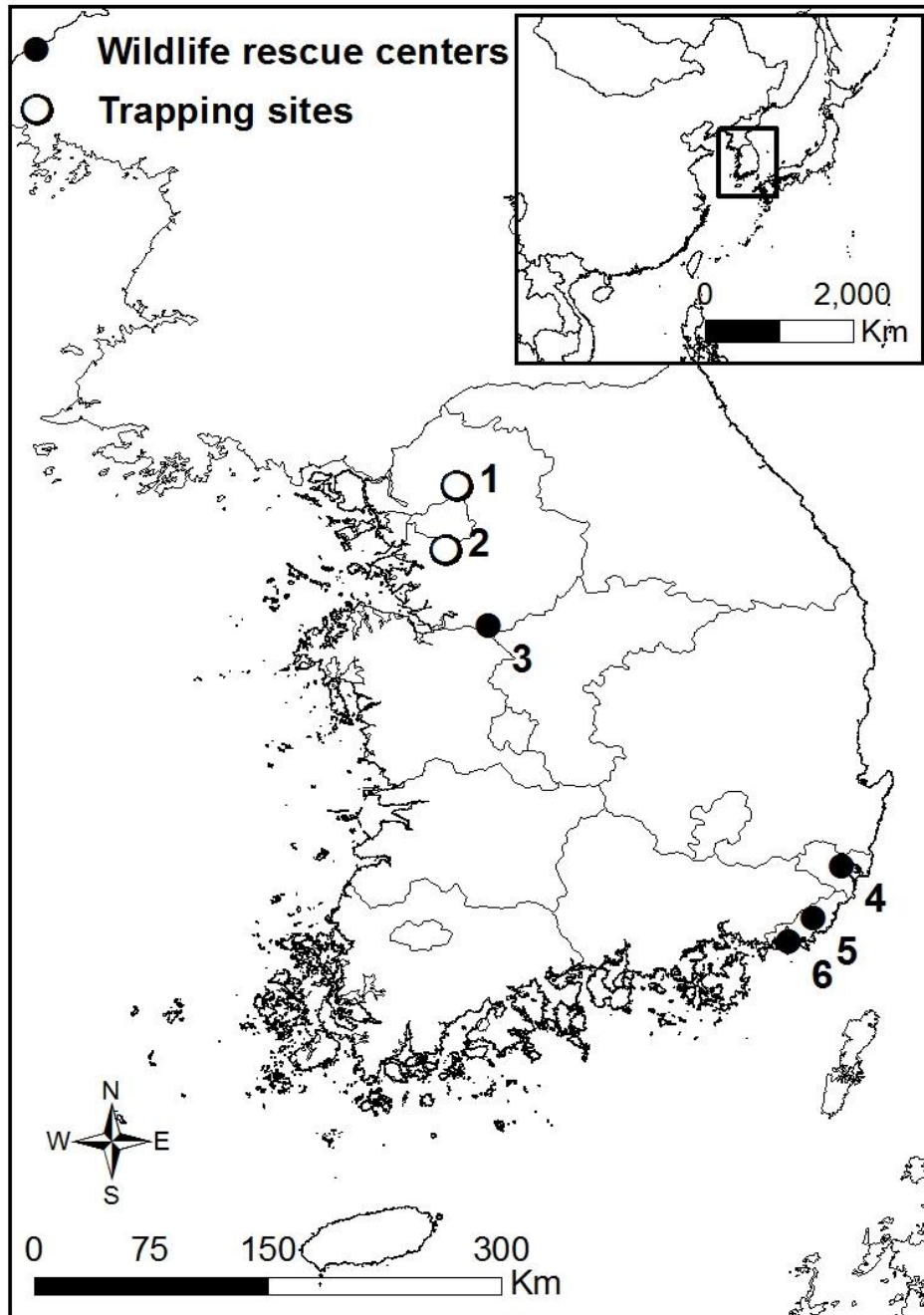


Figure 1. The location of wildlife rescue centers and trapping sites. Numbers indicate following locations. 1: Gwang-reung Forest, 2: Gwanak Arboretum, 3: Gyeonggi Wildlife Rescue Center, 4: Ulsan Wildlife Rescue Center, 5: Gyungsung University Bird Museum, 6: Busan Wildlife Rescue Center.

3.2 Sample collection

Mist nets (12×2.5 m, 5 shelves, 16×16 mm mesh), conspecific audio lures, and stuffed decoys were used to trap the owls (Bildstein and Bird 2007). Mist nets were placed near locations where the calls of Oriental Scops Owls and Northern Boobooks had been previously detected. Nets were checked at 30 minute intervals until capture. Each mist net station and audio lure was operated for no more than three hours if the capture was unsuccessful, to minimize disturbance to the birds. Locations of unsuccessful trapping attempts were re-visited within three days. Birds were extracted from mist nets immediately and ringed, measured, blood and feather sampled, and released after being photographed. Each bird was handled within approximately five minutes to minimize stress. When more than two birds were caught simultaneously, birds were kept in a cotton bag hung from the ground before being processed, and none of the birds were kept more than 30 minutes.

For each captured birds the following morphometric measurements were taken: wing length (maximum flattened length) and tail length to closest 1 mm with the wing ruler, tarsus length, hind claw length, bill length (to skull and to nostril), bill depth, bill width and head length to the closest 0.1 mm with calipers; weight was measured with a Pesola® spring scale or CAS digital scale to 0.1 g. Trapping, sampling and banding of birds were permitted by the Cultural Heritage Administration (2014.5.22), because Northern Boobooks and Oriental Scops Owls are designated as a natural monument and they are legally protected in Korea. All procedures including handling of birds were based on Redfern and Clark (2001) and Hardey *et al.* (2013) to sustain minimum disturbance and stress of individual birds.

Blood collection, containment and preservation methods were followed as Owen (2011)'s review. Small amount (approximately 75–100 μ l) of whole blood was collected by venipuncture from the brachial vein using a 26G needle and micro-capillary tube. 70% isopropyl ethanol swabs were used to clear the feathers, making the brachial vein visible before puncture. The lancing area was dried completely before bleeding to avoid mixing ethanol with the blood sample. Collected whole blood was directly moved to a 1.5 ml micro-tube and kept frozen at -20°C until it could be analyzed (Hobson *et al.* 1997). 3–5 body feathers were also collected from right or left flank for further analysis.

In addition to the data collected from the birds captured in the wild at Gwang-reung Forest and Gwanak Arboretum. Wildlife rescue centers were visited for measuring and collecting tissue samples from additional birds. Under the supervision of a rehabilitator or veterinarian, rescued birds were measured quickly, following the methods described above. Small amounts of blood or feathers were sampled also, following the same method used with trapped wild birds. Frozen specimens of Oriental Scops owls and Northern Boobooks were also measured after a few hours of defrosting. Feathers, tips of tongues or small amounts of pectoral muscle (0.3 g) were collected for molecular sex identification. All samples from wildlife rescue centers were kept frozen at -20°C until analysis.

Potential prey items were collected during mist net operations. Potential prey types were chosen based on previous studies reporting the diet of Northern Boobooks from Japan (Taniguchi 1983, Noguchi 2002) and Taiwan (Lin and Yeh 2004), and the diet of Oriental Scops Owls in Korea (Noh 2013) were collected from the field. Information from a diet study of two closely related congeneric owl species on the Daito Island, Japan (Toyama and Saitoh 2011) was also utilized in choosing prey groups.

Several prey species were collected from the study site during the trapping period. Potential arthropod preys were collected after being entangled in the mist net randomly, as well from the ground by hand. Since these owls are known to prey also on larger vertebrate species, birds and mice were included among the potential prey types. In keeping with ethical practices, bird samples were collected from bird-window collisions near the study site and in adjacent areas that have similar deciduous forest environments. Mice samples were obtained from a separate small mammal monitoring study by collecting animals that are died accidentally from the live traps. Vertebrate samples were selected based on the collection date and locale, to take into account their seasonal state and environment which are closest to the study area. All potential prey species used in analysis were identified to the lowest taxonomic level to best knowledge (see Appendix 1 for the species list).

3.3 Data analysis

3.3.1 Molecular sex identification

To determine the sex of the owls, molecular sex identification using the polymerase chain reaction (PCR) was conducted by following methods adjusted from Han *et al.* (2009). Genomic DNA was extracted from air-dried whole blood, feathers, tongue, and pectoral muscle tissue using the Qiagen DNeasy Blood® and Tissue Kit (Qiagen, USA). The PCR amplification was conducted in a total volume of 20 µl, using AccuPower® PCR PreMix, which contains 30mM KCl, 10mM Tris-HCl(pH 9.0), 1.5mM MgCl₂, 250 µl of each dNTP and 1 Unit of Top DNA polymerase. A total of 2 µl (10 pmol/µl) of mixture of 3 primers (P2: 5'-TCTGCATCGCTAAATCCTTT-3', P0: 5'-ACIAGACCAAGGTTGAGTTA-3', P8: 5'-GTYAARGAGTAGGAACCCTC-3') and 2 µl of extracted genomic DNA was added before performing the PCR procedure with a Lifepro Thermal Cycler® (Bioer, China).

Denaturation was started at 94°C for 2 min 30 s and was followed by 30 cycles of 94°C for 30 s, 53°C for 30 s, and 72°C for 45 s. A final run at 72°C for 5 min completed the PCR procedure. The PCR product were visualized and checked by 3% agarose gel electrophoresis at 100V operated for 45 minutes. This method, allowed molecular sex identification of owls and some other avian taxa by applying the P0 primer in addition to the P2 and P8 primers which were described by Griffith *et al.* (1998) successfully (Han *et al.* 2009).

Individual samples with two distinct bands were considered to be females, and individuals with single bands were considered to be males.

3.3.2 Stable isotope analysis

Whole blood and feather samples collected from Gwang-reung Forest were analyzed for carbon and nitrogen stable isotope ratios. Potential prey items were prepared before stable isotope analysis. Invertebrate samples were rinsed with deionized water using an ultrasonic cleaner (SAEHAN digital sonic© model: SHB-1025) and dismembered, as the owls do before consumption, to increase the ease of grinding and further homogenize the sample. Wings and jaws of large beetles were removed and wings of large moths were removed as well. From vertebrate species, about 2g of muscle was sampled from the breasts or legs with a disposable dissecting blade, and any other tissue (e.g. membranes, ligaments, and fat) was removed from the sample. Muscle samples were rinsed with deionized water using the ultrasonic cleaner. Variation in the lipid content of tissue samples is known to affect $\delta^{13}\text{C}$ values (Post *et al.* 2007), thus lipid extraction is a common procedure in preparation for tissue carbon stable isotope analysis. However, Bennett and Hobson (2009) monitored C:N ratios in their study of forest arthropods to avoid the effects of lipid extraction in tissue $\delta^{15}\text{N}$ values. Since a nitrogen stable isotopic signature is also important in predicting diet composition of the two owl species, Bennett and Hobson (2009)'s approach were used to monitor C:N ratios instead of lipid extraction, and considered samples with relatively high C:N ratios to have high lipid content.

Prepared samples were air-dried in a dry oven at 60°C for at least 72 hours and ground into fine particles and homogenized with mortars and pestles.

Stable isotope ratios of carbon and nitrogen were analyzed with a continuous-flow stable isotope ratio mass spectrometer (Isoprime-EA, Micromass, UK) linked with a CN analyzer (NA Series 2, CE Instruments, Italy) at the National Instrumentation Center for Environmental Management (NICEM), Seoul National University, Seoul, Korea. Stable isotopes were expressed in δ notation as per mill, calculated according to the following equation:

$$\delta X(\text{‰}) = [(R_{\text{sample}} - R_{\text{standard}}) - 1] \times 1000$$

where X is the element in the analysis, R_{sample} is the ratio of isotopes from the analyzed sample, and R_{standard} is the ratio of isotopes in international standard materiald, which are Pee Dee Belemnite (PDB) for carbon and atmospheric nitrogen.

3.3.3 Statistical analysis

For analyzing sexual size dimorphism in the two owl species, each measurements of both sexes were compared with Wilcoxon Mann-Whitney rank sum test (PROC NPAR1WAY) to find any significant differences between the sexes. The magnitude of the size difference in each morphological measurements was calculated using the dimorphic index (DI; Storer 1966, Greenwood 2003),

$$DI = \frac{f - m}{(m + f)} \times 100$$

where m is the mean value of male measurements and f is the mean value of female measurements. A positive DI indicates that the morphometric character is larger in females, and so represents reversed sexual dimorphism.

Considering that these two owl species are complete migrants, we did not include body mass in performing a discriminant function analysis (DFA). Since our data did not fit multivariate normality, quadratic discriminant analysis (PROC DISCRIM) was performed with measurement variables selected through a stepwise procedure (PROC STEPDISC). Birds with missing measurements due to the specimen's condition (e.g., damaged claws, bills or feathers, were excluded from the discriminant analysis.

The birds were classified as male or female based on the discriminant score (D) from the discriminant analysis. The accuracy of the discriminant functions produced by DFA was evaluated by the re-substitution method and jackknife method using the POSTERR and CROSSVALIDATE options in PROC DISCRIM of SAS software. To estimate the posterior probability from the discriminant scores of individuals, a non-linear regression model was fitted, where k and c were constants calculated from a nonlinear procedure (PROC NLIN).

$$PP = 1/(1 + \exp(kD - c))$$

Wilcoxon Mann-Whitney rank sum test (PROC NPAR1WAY) was used in comparing the difference of stable isotope values of two owl species. Coefficients of variance (CV) were calculated to compare the variation of stable isotope ratios of owl's whole blood, and the Levene's test was also performed to compare isotopic variance between two species.

Potential prey species were categorized into five broad taxonomic groups, and arthropods were separated based on major prey types identified by previous studies. A one way Kruskal-Wallis was first performed to see the difference in the stable isotope ratios of nitrogen and carbon in the prey type groups. Dunn's post hoc test was then used to compare each group. Among these potential prey groups, bush-crickets and beetles did not show a significant difference, and thus, pooled as a single group.

The diet composition of both owl species was reconstructed with the R package SIAR. This package is designed to run mixing models for isotope data based on Bayesian statistics, and it provides means and 95% confidence intervals of sources that contributed to the consumer tissue, and presents posterior probability distributions for each contributions graphically.

This model incorporated a trophic enrichment factor (TEF, or also called fractionation rate or discrimination factor) for stable isotope ratios between consumer tissue and diet source. However, there is no known value of trophic enrichment factor available for Oriental Scops Owls or Northern Boobies in the literature. Thus, I used a trophic enrichment factor following the model of Caut *et al.* (2009) that provides diet-specified model for different tissues of each taxonomic groups. I present the mean contribution of each prey group to the diet of the two owl species. Stable isotopic values were given as means and standard deviations (SD).

To see the effect of body size on the trophic status of individual owls, I first conducted a principal component analysis to extract the principal component (PC1) that best explains multiple morphometric measures (Rising and Somers 1989), rather than using a single morphometric character for the analysis. Then, I used a Spearman correlation analysis with PC1 and the nitrogen stable isotope ratio. Too small sample size hindered analyzing the sexes separately in Northern Bobwhites, thus instead, I pooled both sexes together for the analysis. However, for male Oriental Scops Owls, separate correlation analysis was performed.

Chapter 4. Results

4.1 Sampling and molecular sex identification

During the two spring seasons in 2014 and 2015, a total of 10 Northern Boobooks and 30 Oriental Scops Owls were trapped and measured at Gwang-reung Forest and the forests of Gwanak Arboretum. Measurements of 22 Northern Boobooks and 16 Oriental Scops Owls, dead and alive, which were obtained from wildlife rescue centers, were also used in the sexual dimorphism study.

All sampled birds were successfully sexed, using molecular method.

Among the 46 Oriental Scops Owls, 19 were identified as females and 27 were identified as males. 13 of the 32 Northern Boobooks were identified as females and 19 were identified as males by examining gel electrophoresis results of PCR products (Figure 2).

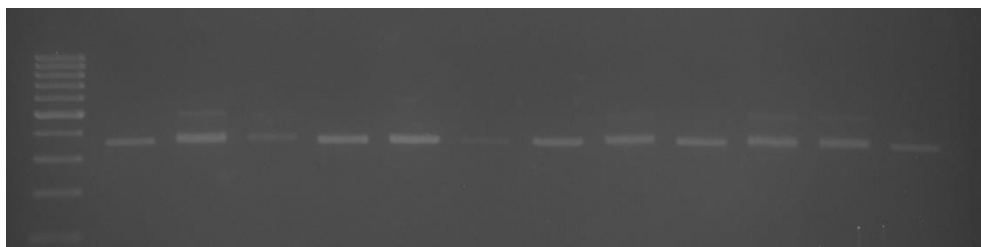


Figure 2. Sample image of gel electrophoresis showing females (two bars) and males (single bar).

4.2 Morphology and sexual size dimorphism

Six external characteristics were found to differ statistically between males and females in Oriental Scops Owls (Table 1). Although the differences were small, all measurement variables showed reversed sexual dimorphism in Oriental Scops Owls. By contrast, Northern Boobooks had a few external characteristics that showed male-biased size dimorphism, but none of those measurement variables were statistically significant (Table 1, 2). In Northern Boobooks, length of bill to nostril, bill to skull and bill width had positive dimorphism index (DI). And seven other external characteristics were found to have negative indices, indicating larger males (Table 3, 4). Although I used only a small sample from the wild birds, the weight of northern Boobooks showed the strongest male-biased sexual size dimorphism, with males that were about 12% heavier than females. In Oriental Scops Owls, the width of the bill and the weight showed the strongest sexual size dimorphism. The coefficient of variance was highest in bill width (BW) in Northern Boobooks and second highest in Oriental Scops Owls, and weight had the highest coefficient of variance in Oriental Scops Owls.

Table 1. Morphometric parameters of Oriental Scops Owls in the Republic of Korea. Asterisks indicate significant differences (* $P<0.05$, ** $P<0.01$, *** $P<0.001$). CV and DI represent the coefficient of variance and the dimorphic index, respectively.

Variables	Sex (<i>n</i>)	Mean \pm SD (range)	Median	CV	U	Z	<i>P</i>	DI
Wing* (max)	Total (46)	148.4 \pm 3.7 (138.5 – 153.0)	149.2	2.48				
	Male (27)	147.3 \pm 3.8 (138.5 – 153.0)	147.0	2.59	553	2.382	0.017	0.434
	Female (19)	149.9 \pm 3.0 (142.5 – 153.0)	151.5	1.98				
Tail***	Total (46)	69.3 \pm 3.5 (60.0 - 75.0)	70.0	5.01				
	Male (27)	67.81 \pm 3.15 (60.0 – 72.0)	68.5	4.65	611	3.685	0.000	1.286
	Female (19)	71.4 \pm 2.8 (66.0 - 75.0)	72.0	3.92				
Head	Total (46)	41.17 \pm 0.87 (39.30 - 42.90)	41.15	2.11				
	Male (27)	41.04 \pm 0.87 (39.30 - 42.50)	41.00	2.13	495.5	1.084	0.279	0.200
	Female (19)	41.37 \pm 0.85 (39.90 - 42.90)	41.20	2.06				
Bill length to nostril (BN)*	Total (46)	9.43 \pm 0.63 (7.6 - 10.5)	9.5	6.67				
	Male (27)	9.25 \pm 0.62 (7.6 - 10.42)	9.1	6.71	544	2.179	0.029	1.136
	Female (19)	9.68 \pm 0.56 (8.5 - 10.5)	9.8	5.83				
Bill length to skull (BS)	Total (46)	20.14 \pm 0.76 (18.1 - 21.5)	20.1	3.78				
	Male (27)	20.04 \pm 0.83 (18.1 - 21.5)	20.1	4.16	479	0.716	0.474	0.298
	Female (19)	20.28 \pm 0.64 (19.4 - 21.4)	20.2	3.17				
Bill depth (BD)***	Total (46)	8.57 \pm 0.6 (7.3 - 10.8)	8.5	7.05				
	Male (27)	8.35 \pm 0.47 (7.3 - 9.4)	8.3	5.64	592	3.248	0.001	1.566
	Female (19)	8.89 \pm 0.64 (7.9 - 10.8)	8.9	7.17				
Bill width (BW)**	Total (46)	5.38 \pm 0.58 (4.7 - 7.2)	5.3	10.81				
	Male (27)	5.23 \pm 0.54 (4.7 - 6.5)	5	10.34	545.5	2.203	0.016	1.708
	Female (19)	5.6 \pm 0.58 (4.8 - 7.2)	5.5	10.45				

Table 2. Morphometric parameters of Oriental Scops Owls in the Republic of Korea. Asterisks indicate significant differences (* $P<0.05$, ** $P<0.01$, *** $P<0.001$). CV and DI represent the coefficient of variance and the dimorphic index, respectively.

Variables	Sex (<i>n</i>)	Mean \pm SD (range)	Median	CV	U	Z	<i>P</i>	DI
Tarsus**	Total (46)	25.15 \pm 1.02 (22.37 - 27.6)	25.2	4.05				
	Male (27)	24.79 \pm 0.99 (22.37 - 27)	24.7	3.98	568	2.703	0.007	0.862
	Female (19)	25.66 \pm 0.85 (24.3 - 27.6)	25.3	3.31				
Hindclaw	Total (46)	6.97 \pm 0.39 (6.1 - 7.7)	6.9	5.62				
	Male (27)	6.86 \pm 0.38 (6.1 - 7.5)	6.9	5.59	214	1.885	0.059	1.070
	Female (19)	7.16 \pm 0.34 (6.7 - 7.7)	7.16	4.73				
Weight	Total (30)	81.34 \pm 11.52 (63.3 - 123.4)	78.45	14.16				
	Male (19)	76.56 \pm 6.06 (63.3 - 91.0)	76.70	7.91	238.5	2.907	0.004	3.924
	Female (11)	89.60 \pm 14.15 (75.0 - 123.4)	89.00	15.79				

Table 3. Morphometric characteristics of Northern Boobooks in the Republic of Korea. Asterisks indicate significant differences (* $P<0.05$, ** $P<0.01$, *** $P<0.001$). CV and DI represent the coefficient of variance and the dimorphic index, respectively.

Variables	Sex (<i>n</i>)	Mean \pm SD (range)	Median	CV	U	Z	P	DI
Wing* (max)	Total (31)	226.0 \pm 6.0 (215-237)	225	2.64				
	Male (12)	227.8 \pm 5.8 (220-237)	228.5	2.56	220.0	1.1174	0.2638	-1.2370
	Female (19)	225.0 \pm 6.0 (215-235)	223.5	2.65				
Tail***	Total (31)	120.7 \pm 5.0 (108-130)	121	4.11				
	Male (12)	120.9 \pm 4.8 (113-127.5)	121.5	3.93	195.5	0.1221	0.0904	-0.2485
	Female (19)	120.6 \pm 5.2 (108-130)	121	4.33				
Head	Total (31)	48.68 \pm 0.99 (46.6-50.9)	48.8	2.03				
	Male (12)	49.09 \pm 1.00 (47.3-50.9)	49.18	2.04	236.5	1.7873	0.0840	-1.3742
	Female (19)	48.42 \pm 0.91 (46.6-49.7)	48.7	1.89				
Bill length to nostril (BN)*	Total (31)	12.20 \pm 0.60 (11.1-13.4)	12.2	4.91				
	Male (12)	12.05 \pm 0.66 (11.2-13.4)	12.05	5.51	165.0	-	0.2892	2.0534
	Female (19)	12.30 \pm 0.55 (11.1-13.4)	12.3	4.49		1.0790		
Bill length to skull (BS)	Total (31)	25.08 \pm 1.00 (22.4-27.4)	25.2	4				
	Male (12)	24.88 \pm 1.2 (22.4-26.6)	24.9	4.84	180.0	-0.467	0.644	1.318
	Female (19)	25.21 \pm 0.86 (24.2-27.4)	25.2	3.42				
Bill depth (BD)***	Total (31)	10.25 \pm 0.53 (9.1-11.9)	10.2	5.17				
	Male (12)	10.41 \pm 0.57 (9.9-11.9)	10.2	5.47	208.0	0.653	0.519	-2.529
	Female (19)	10.15 \pm 0.49 (9.1-10.8)	10.2	4.85				
Bill width (BW)**	Total (31)	7.01 \pm 0.77 (5.4-8.4)	6.9	11.01				
	Male (12)	6.9 \pm 0.94 (5.4-8.4)	6.85	13.67	181.0	-0.427	0.673	2.575
	Female (19)	7.08 \pm 0.66 (6.2-8.2)	7.1	9.33				

Table 4. Morphometric characteristics of Northern Boobooks in the Republic of Korea. Asterisks indicate significant differences (* $p<0.05$, ** $p<0.01$, *** $p<0.001$). CV and DI represent the coefficient of variance and the dimorphic index, respectively.

Variables	Sex (<i>n</i>)	Mean \pm SD (range)	Median	CV	U	Z	<i>P</i>	DI
Tarsus**	Total (31)	27.73 \pm 1.89 (24-32.3)	27.8	6.82				
	Male (12)	28.35 \pm 2.15 (24-32.3)	28	7.6	224.0	1.279	0.211	-3.627
	Female (19)	27.34 \pm 1.64 (24-29.85)	27.7	6.02				
Hindclaw	Total (10)	10.89 \pm 0.53 (9.95-11.5)	10.98	4.89				
	Male (4)	11.28 \pm 0.39 (10.7-11.5)	11.45	3.43	31.0	1.818	0.103	-5.933
	Female (6)	10.63 \pm 0.47 (9.95-11.1)	10.75	4.45				
Weight	Total (9)	184.11 \pm 14.23 (168-206)	186	7.73				
	Male (4)	196.50 \pm 9.15 (186-206)	197	4.65	29.0	2.091	0.070	-12.031
	Female (5)	174.20 \pm 8.14 (168-188)	170	4.67				

4.2.1 Discriminant analysis

In spite of some overlap in measurements, a stepwise backward selection chose tail, tarsus, bill depth and bill width as variables that best classify male and female Oriental Scops Owls. With those selected values, the following discriminant function analysis formulated below equation ($\chi^2 = 10.004$, $P = 0.440$; Wilk's lambda = 0.592, $F = 7.07$, $P < 0.001$). Discriminant scores calculated from DFA for each individual had certain amount of overlap, but females had generally greater discriminant scores than males (Figure 3)

$$D_o = -56.0873 + 1.18762 * \text{Bill depth} + 0.51508 * \text{Bill width} \\ + 0.34729 * \text{Tail length} + 0.73489 * \text{Tarsus length}$$

Error rates of this discriminant function tested by the re-substitution and leave-one out methods were 20.39% and 20.78%, respectively.

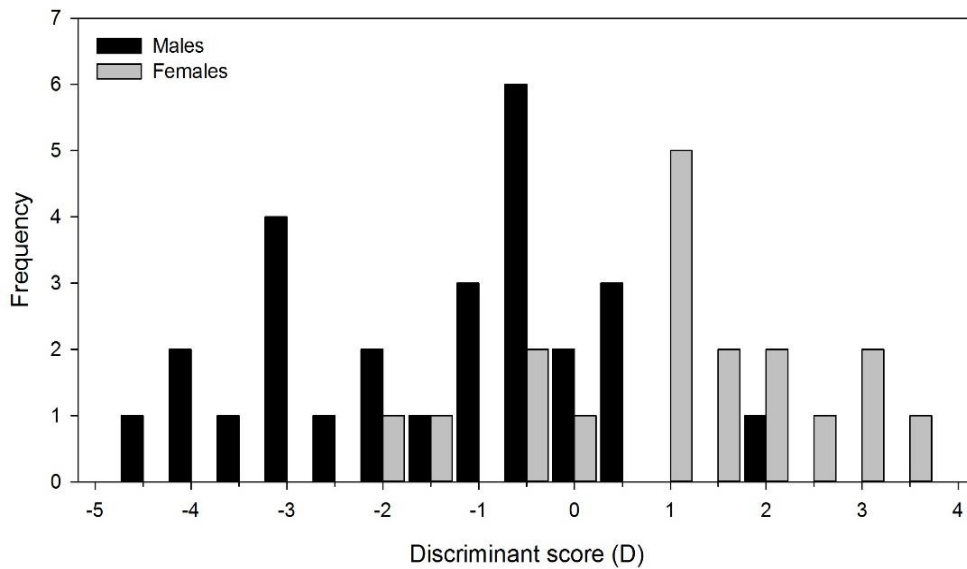


Figure 3. Histogram of discriminant scores (D) for Oriental Scops Owls.

From non-linear regression (Figure 4), the equation to explain posterior probabilities and discriminant scores was calculated as follows ($k = 1.0000$, standard error = 0.0000028472, $\underline{P} < 0.001$; $c = 0.0003$, standard error = 0.00000278, $P < 0.001$)

$$PP_o = 1/[1 + \exp(D - 0.0003)]$$

When the posterior probability is 0.5, this equation calculates cut-off value of being male or female as -0.0003. In addition, birds that have scores of $D \leq -2.941$ or $D \geq 2.947$ can be classified as males and females, respectively, with posterior probability of 0.95.

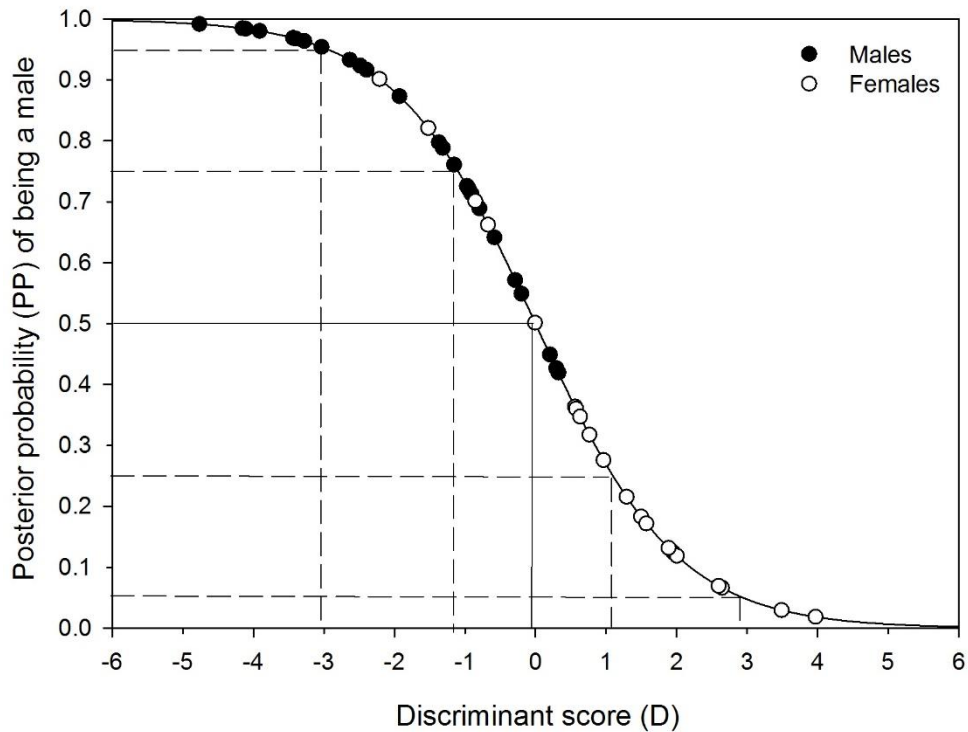


Figure 4. Relationship between discriminant scores and posterior probability of being a male of Oriental Scops Owls. Dashed lines indicate 95%, 75%, 25%, and 5% probability cut offs, and the solid line indicates a 50% cut-off of discriminant score to be a male.

In Northern Boobooks, a stepwise backward selection chose bill length (to skull) and head length as variables that best classify males and females. Discriminant function analysis produced the following linear equation ($\chi^2 = 1.823$, $P = 0.610$; Wilk's Lambda = 0.78434641, $F = 3.99$, $P = 0.030$), however, the overlap between discriminant scores of males and females were greater than in that of Oriental Scops Owls (Figure 5).

$$D_N = 34 - 1.0544 * \text{Head length} + 0.7307 * \text{Bill length (to skull)}$$

The accuracy of this discriminant function was tested by the re-substitution and leave-one-out methods, and the error rates were 30.26% and 30.65%, respectively.

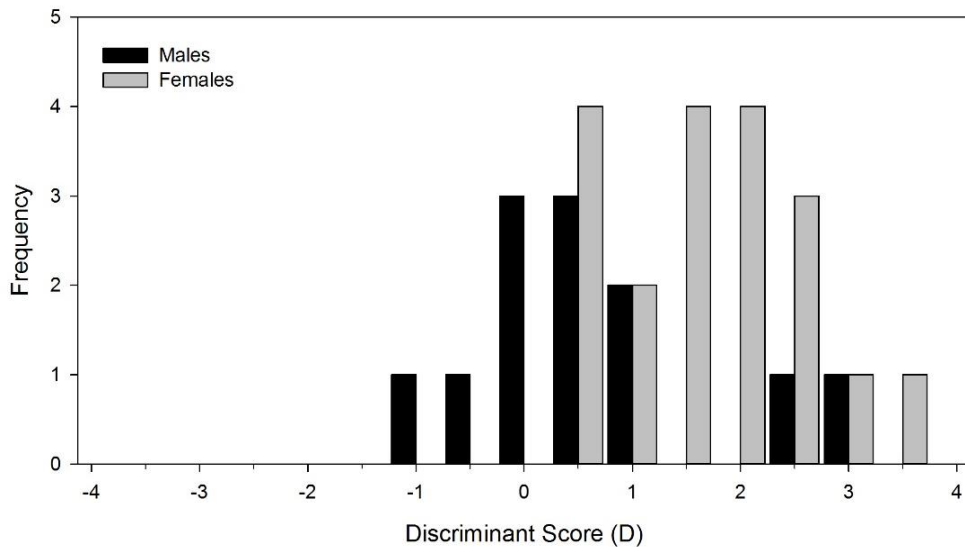


Figure 5. Histogram of discriminant scores of Northern Boobooks.

From non-linear regression (Figure 6), the equation to explain posterior probability and discriminant scores were calculated as follows ($k = -1.0000$, standard error = 0.0000034502 , $P < 0.001$; $c = 0.4364$, standard error = 0.0000027182 , $P < 0.001$).

$$PP_N = 1/[1 + \exp(-D - 0.4364)]$$

When the posterior probability is 0.5, this equation calculates a cut-off value of being male or female as 0.4364. In addition, birds that had scores of $D \leq -3.381$ or $D \geq 2.508$ can be classified as males and females, respectively, with posterior probability of 0.95.

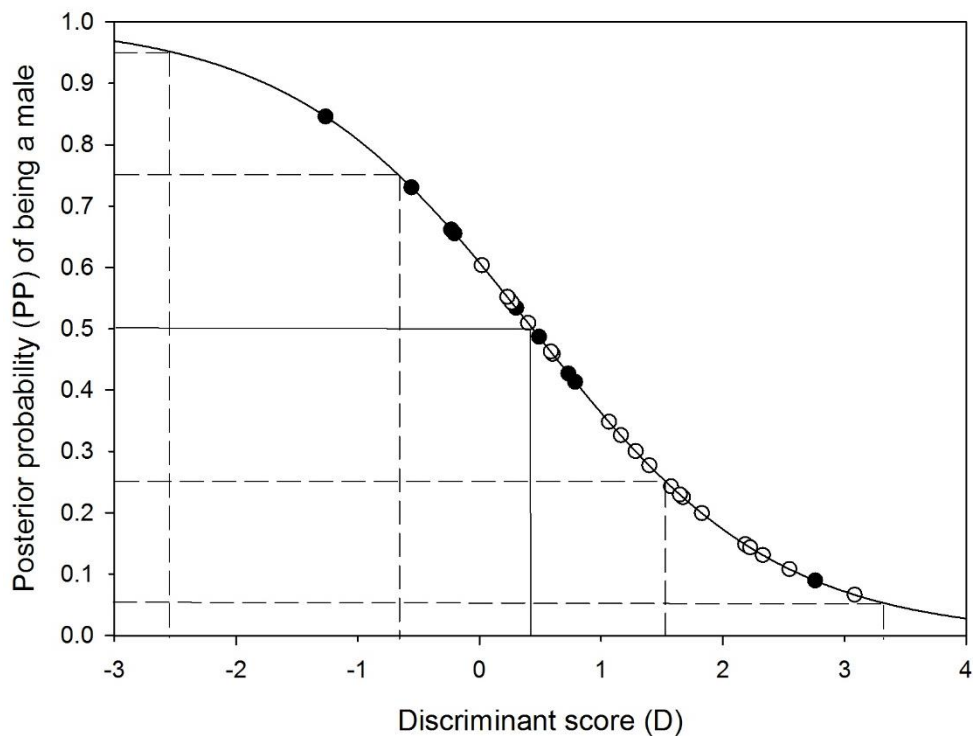


Figure 6. Relationship between discriminant score and posterior probability of being a male in Northern Boobooks. Dashed lines indicate 95%, 75%, 25%, and 5% probability cut offs, and the solid line indicates a 50% cut-off of discriminant score to be a male.

4.3 Stable isotope ecology

4.3.1 Stable isotope ratio analysis

The two owl species showed clear differences in both carbon and nitrogen stable isotope ratios (Figure 7). The nitrogen stable isotope ratios ($\delta^{15}\text{N}$) were significantly higher in Northern Boobooks ($U = 116.0$, $Z = 3.501$, $P < 0.001$) than in Oriental Scops Owls in all samples. Carbon stable isotope ratios ($\delta^{13}\text{C}$) were also significantly higher in Northern Boobooks ($U = 102.5$, $Z = 2.311$, $P = 0.021$), though there was some overlaps between the two species.

In both species, there was some levels of variation in the nitrogen stable isotope ratio. However, the coefficient of variance of the carbon stable isotope ratio was low in Oriental Scops Owls, in contrast to the high coefficient of variance in Northern Boobooks ($\delta^{15}\text{N}$: $F=0.29$, $P=0.597$; $\delta^{13}\text{C}$: $F=10.32$, $P=0.005$; Table 5).

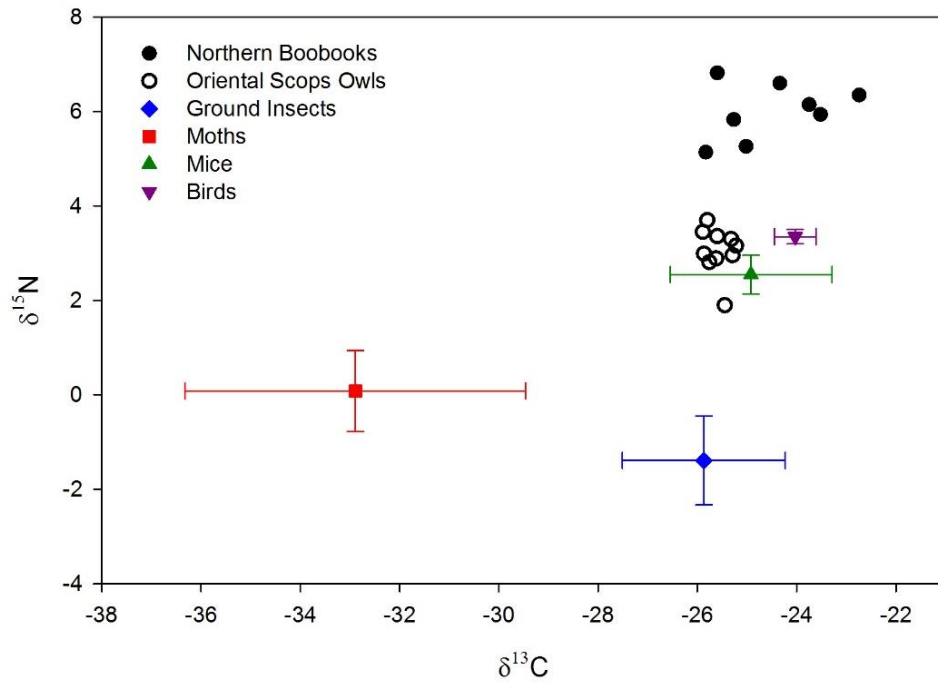


Figure 7. Distribution of carbon and nitrogen stable isotopes in Oriental Scops Owls, Northern Boobooks, and potential prey items.

Table 5. Nitrogen and carbon stable isotope values for Oriental Scops Owls and Northern Boobooks.

	Species	Mean \pm SD	Median	CV (%)	U	Z	P
$\delta^{13}\text{C}$	<i>O. sunia</i> (n=10)	-25.58 ± 0.25	-25.61	0.98	102.5	2.311	0.021
	<i>N. japonica</i> (n=8)	-24.51 ± 1.10	-24.68	4.49			
$\delta^{15}\text{N}$	<i>O. sunia</i> (n=10)	3.05 ± 0.49	3.08	16.09	116	3.510	0.000
	<i>N. japonica</i> (n=8)	6.01 ± 0.60	6.05	9.93			

Among prey groups, there was a significant difference between the carbon ($F = 14.18$, $P < 0.001$) and nitrogen ($F = 33.17$, $P < 0.001$) isotope values of each group, except beetles and bush crickets. Both carbon and nitrogen isotope ratios were indistinguishable between bush crickets and beetles, so I pooled those two groups into one (ground insects) for analytical purposes (Table 6).

Table 6. Stable isotope values of potential prey items of Oriental Scops Owls and Northern Boobooks in the Republic of Korea.

Groups	Sample size (n)	Mean biomass (g)	Elements	Mean	SD
Moths	2	0.8	$\delta^{13}\text{C}$	-32.89	3.43
			$\delta^{15}\text{N}$	0.08	0.86
Mice	3	24.2	$\delta^{13}\text{C}$	-24.92	1.63
			$\delta^{15}\text{N}$	2.55	0.41
Ground insects	12	0.9	$\delta^{13}\text{C}$	-25.87	1.64
			$\delta^{15}\text{N}$	-1.39	0.94
Bush crickets	4	0.9	$\delta^{13}\text{C}$	-27.27	0.65
			$\delta^{15}\text{N}$	-1.63	0.43
Beetles	8	0.9	$\delta^{13}\text{C}$	-25.17	1.54
			$\delta^{15}\text{N}$	-1.27	1.12
Birds	4	45.6	$\delta^{13}\text{C}$	-24.03	0.42
			$\delta^{15}\text{N}$	3.35	0.15

4.3.2 Diet reconstruction of owls

Results from a Bayesian multi-source mixing model showed that the two owl species utilized different proportions of prey groups. Northern Boobooks (Figure 9) relied on more avian prey (Mean = 49%, 95% CI = 19–85%), followed by mice (Mean = 33%, 95% CI = 0–62%), moths (Mean = 10%, 95% CI = 0–20%), and ground insects (Mean = 6%, 95% CI = 0–20%). Oriental Scops Owls (Figure 8) consumed ground insects the most (Mean = 34%, 95% CI = 24–44%), followed by birds (Mean = 23%, 95% CI = 4–40%), moths (Mean = 22%, 95% CI = 12–32%), and mice (Mean = 20%, 95% CI = 0.2–40%). Overall, Oriental Scops Owls selected more arthropod prey than Northern Boobooks, while Northern Boobooks were highly dependent on avian prey sources in this study.

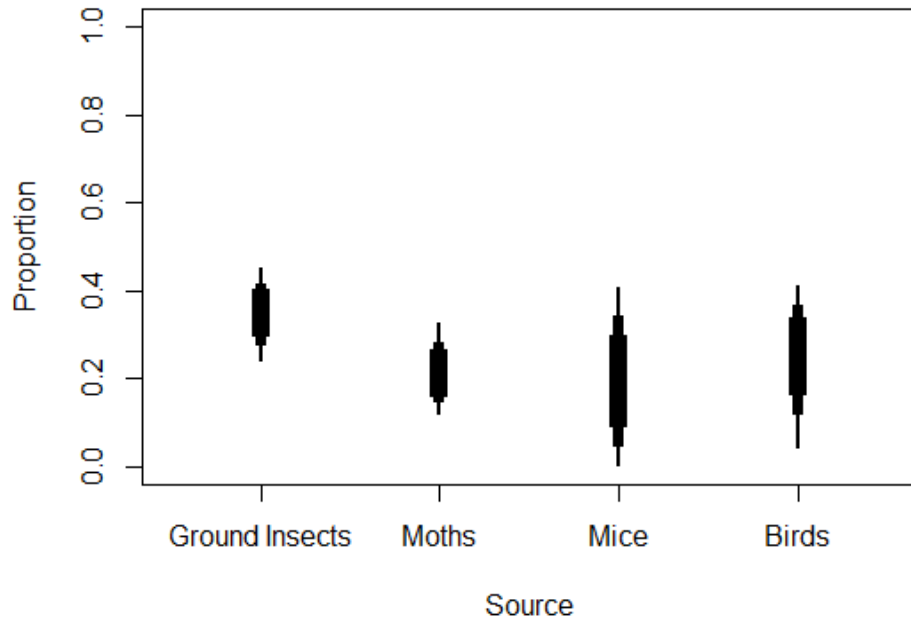


Figure 8. Proportional diets of Oriental Scops Owls estimated by a multi-source mixing model. Line thickness indicates 50%, 75%, and 95% probability.

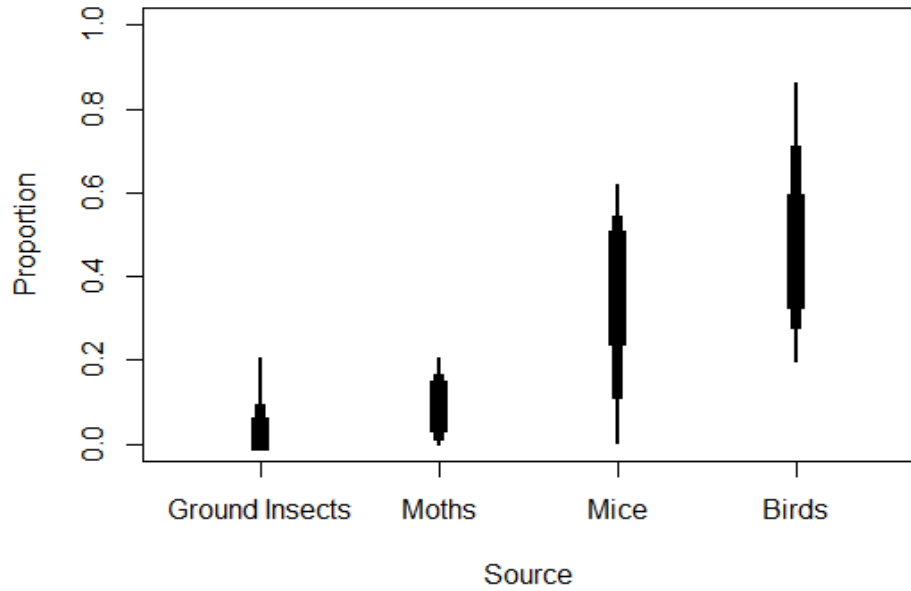


Figure 9. Proportional diets of Northern Boobooks estimated by a multi-source mixing model. Line thickness indicates 50%, 75%, and 95% probability.

4.3.3 Body size and trophic level

The first principal component (PC1) of each species was calculated as an index of overall body size for the subsequent correlation analysis with nitrogen stable isotope ratios. PC1 of Oriental Scops Owls explained 82% of the variation, with wing length having highest positive loading, followed by tail length. Northern Boobooks also had wing length with the highest positive loading, and tail length with the second highest positive loading in PC1, with 71% of variation explained (Table 7).

In Oriental Scops Owls, and especially in 10 individuals from Gwang-reung Forest (male: $n=8$, female: $n=2$), a negative relationship between body size and trophic level was detected ($n = 10$, $r = -0.709$, $P = 0.019$), suggesting that smaller scops owls have a higher trophic level (Figure 11); and while it was statistically not significant, males also had negative correlation coefficients ($n = 8$, $r = -0.476$, $P = 0.207$).

On the other hand, eight Northern Boobooks sampled from the same study area (Figure 10) exhibited no relationship between trophic level and body size ($n = 8$, $r = -0.143$, $P = 0.720$). The sexes were pooled in the analysis due to the small sample size (male: $n = 2$, female: $n = 6$).

Table 7. Results of principal component analysis of Oriental Scops Owls and Northern Boobooks (HC: hind claw length, BN: bill length to nostril, BS: bill length to skull, BW: bill width, BD: bill depth).

	Oriental Scops Owls (n=10)		Northern Boobooks (n=8)	
	PC1	PC2	PC1	PC2
Eigenvectors				
Wing (Max)	0.865258	0.385766	0.912194	-0.33996
Tail	0.461345	-0.79025	0.354196	0.767705
Tarsus	0.0543	0.470741	0.173533	0.33377
Hindclaw	0.041091	-0.03119	0.014598	0.038715
Bill length to nostril	0.079433	-0.01224	0.029698	0.106587
Bill length to skull	0.107248	0.027186	-0.00444	-0.05105
Bill width	0.069028	0.011313	0.059146	0.039099
Bill depths	0.071537	0.05564	0.021543	0.200122
Head	0.078477	-0.00245	-0.08519	0.355829
Eigenvalue	34.02604	5.045944	16.50558	3.956695
Variance explained				
Proportion	0.8236	0.1221	0.7101	0.1702
Cumulative	0.8236	0.9457	0.7101	0.8803

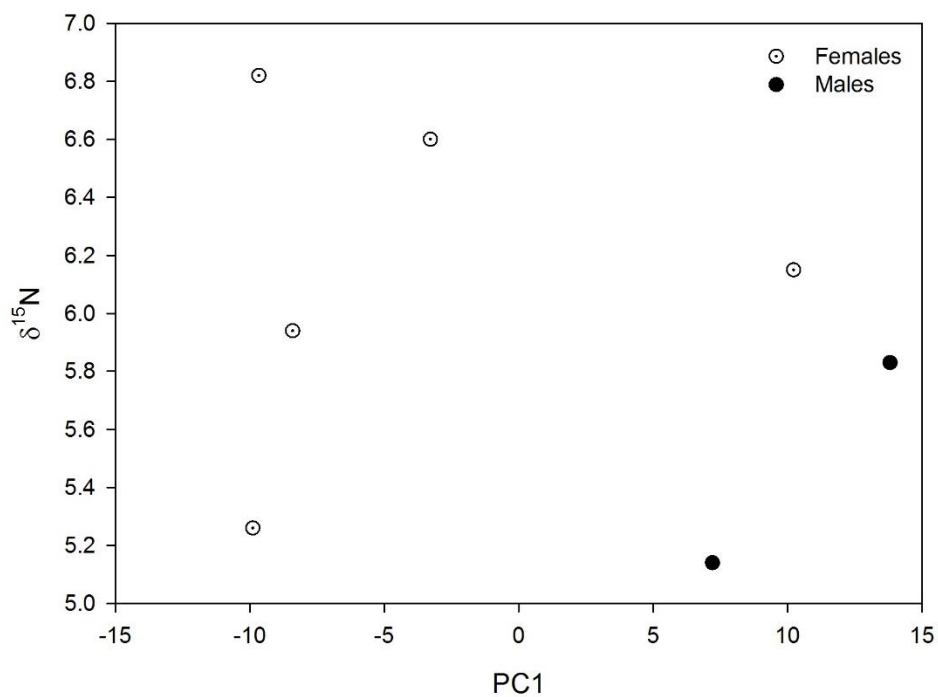


Figure 10. Scatterplot of overall body size index (PC1) and individual trophic level ($\delta^{15}\text{N}$) in Northern Boobooks (*Ninox japonica*)

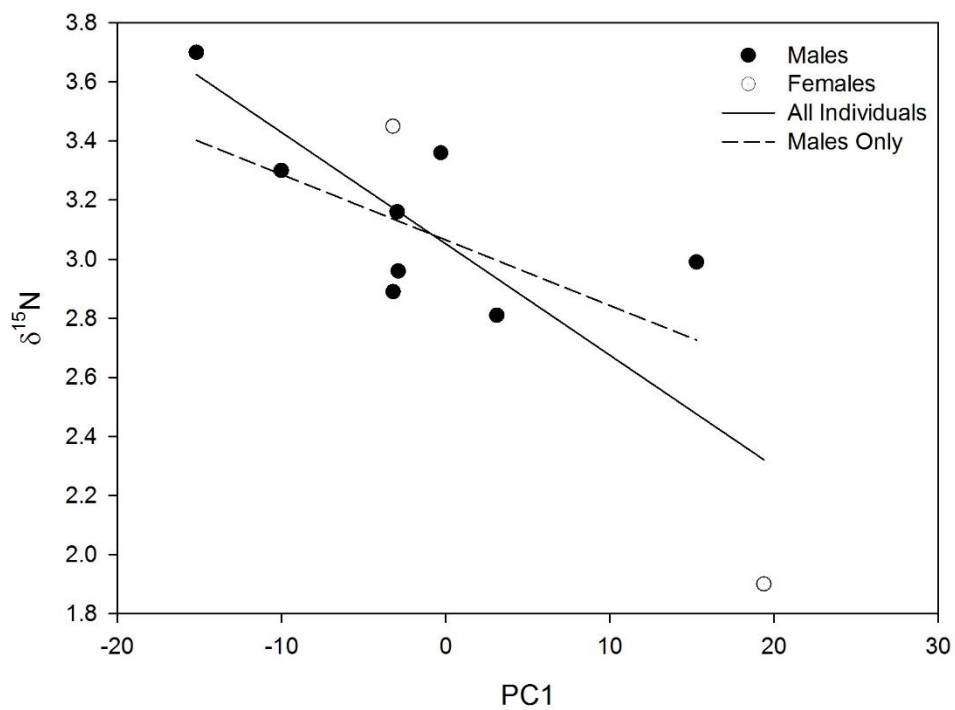


Figure 11. Relationship between overall body size index (PC1) and individual trophic level ($\delta^{15}\text{N}$) in Oriental Scops Owls (*Otus sunia*).

Chapter 5. Discussion

5.1 Sexual size dimorphism in owls

Size dimorphism in birds has been of interest to many ecologists for a long period, for many reasons. Not only is size dimorphism of interest in evolution and functional ecology, but it is also a useful method for field researchers seeking to identify the sex of monomorphic plumaged birds directly with morphometric information.

I assessed sexual size dimorphism in Oriental Scops Owls and Northern Boobooks in Korea, and attempted to generate discriminant functions for morphometric sex identification in field situations. Contrary to the general conception that owls, like diurnal raptors have reversed sexual size dimorphism (RSD), the Northern Boobooks in this study did not show clear sex-related size dimorphism. However, Oriental Scops Owls showed slight RSD as expected from the literature (Seo and Park 2008, Mikkola 2013). Although I failed to detect statistically significant size dimorphism in Northern Boobooks at the typical statistical significance level of 5%, some characters such as tail ($DI = -0.25$, $P = 0.090$), head length ($D = -1.37$, $P = 0.084$) and weight ($DI = -12.03$, $P = 0.07$) likely showed the normal size dimorphism (larger male) having negative dimorphic indices, which was not reported in previous studies (Seo and Park 2008, Mikkola 2013).

This unexpected similarity between the sexes in Northern Boobooks may be related to their taxonomic group, the genus *Ninox*, which includes at least nine species with normal size dimorphism which males are larger or heavier. Three large *Ninox* species show great amount of male-biased sexual size dimorphism: Rufous Owls (*Ninox rufa*; weight range: 700–1300 g, males up to 300 g heavier and larger than females), Powerful Owls (*N. strenua*; 1050–1700 g, males larger and 250 g heavier than females), and Barking Owls (*N. connivens*; 425–510 g; males 100 g heavier than females; Mikkola 2013), and six other smaller species are believed to show normal sexual size dimorphism, though there is no strong quantitative evidence. However, there are also two small species in this genus that shows female-biased sexual size

dimorphism, which are Southern Boobooks (*N. boobook*; 146–360 g, females 65g heavier and larger than males) and Moreporks (*N. novaeseelandiae*; 150–216 g, females 15 g heavier than males Mikkola 2013). The inconsistency in the dimorphism of *Ninox* species leave us in perplexing situation of the common conception on reversed sexual dimorphism in owls. In spite of the diverse forms of sexual size dimorphism in the genus, Northern Boobooks (*N. japonica*) have little evidence of any type of sexual size dimorphism. Resident breeding Northern Boobooks in Taiwan (*Ninox japonica totogo*) were found to be sexually monomorphic in size (10 pairs, Lin *et al.* 2012), as in this study. Recent studies also revealed that there is no morphometric difference between resident populations in Taiwan (n = 9) and migratory populations (*Ninox japonica japonica*, n = 7) in spite of their different genetic lineages (Lin *et al.* 2013), suggesting that the size difference between subspecies is not clear in my study species, Northern Boobooks. Thus, based on this study using birds in Korea, as well as previous studies, I suggest that there is no robust evidence of reversed sexual size dimorphism in Northern Boobooks, at least at the species level, but males still have the potential of having slightly bigger body parts in terms of weight and length of head, wings, and tails.

The sexual dimorphism of Oriental Scops Owls, by contrast, is less controversial than that of Northern Boobooks. Mikkola (2013) suggests that females are on average 6g heavier than males in Oriental Scops Owls (generally ranging from 75g to 95g). In this study, the weight difference between sexes was greater, averaging 13 g difference in average.

These two owl species do not possess any plumage difference between the sexes, and thus, this study of morphometric sex identification may be useful in field applications. Furthermore, except tail length, all other selected external characters are hard, fixed structures which change less during specimen preparation and storage, and thus this discriminant formula can be used with museum skins also. During the trapping sessions, I was able to observe females that had developed brood patches. Since the sexual dimorphism is not strong enough to classify the birds into males or females with certainty, the application of additional clues such as the development of brood patches can support sex identification in the field. However, given the high

error rates of the developed discriminant functions for the two forest owls, it is generally recommended to consider using molecular tools for reliable sex determination in both species.

5.2 Trophic ecology of owls

Clear inter-specific differences in isotopic values confirmed in this study indicate a separation in foraging niche of these two sympatric forest owls. In particular, Northern Boobooks showed higher nitrogen isotopic values, than Oriental Scops Owls. Typically, $\delta^{15}\text{N}$ enrichment of 1.4‰ to 3.3‰ is observed from diet to consumer along increasing food chains, allowing determination of an organism's trophic level and food chain length (Perkins *et al.* 2014). In this study, Northern Boobooks had *circa* 3‰ higher value for nitrogen than Oriental Scops Owls, suggesting that the boobooks to occupy a higher trophic level than the scops owls, and have a longer food chain length by one or two steps.

Similarly, dietary reconstruction models based on stable isotope analysis results explained that the Northern Boobooks used more vertebrate prey (birds and rodents together; 82%) than invertebrates, especially birds. However, Oriental Scops Owls used only 43% of vertebrates prey in diets. Although these proportion of vertebrate prey consumption in both species seems to be higher than the known proportion in previous studies (in % number of items, Northern Boobooks: 0.7-1.9 % in Taniguchi 1983, and 0.06%; Noguchi 2002; Oriental Scops Owls: 0.4% in Noh 2013), it is impossible to have a direct comparison between my study and previous ones, due to the different measures that were used in the study.

Panzeri *et al.* (2014) found significant difference in diet composition of Eurasian Scops Owls in their wintering range, that winter diets have higher percentage of vertebrates. Seasonal differences in diet of Northern Boobooks in Japan were also reported by Taniguchi (1983) that may occurred by seasonal difference in prey availability along season. For instance, larger beetles and moths may not emerge until after than my study period (Bae 1998, Kim 1998, Lee and Kim 2010), causing the limited proportion in this study but the increased amount in the previous studies.

Still, we need to consider the difference between methods used in this study and the previous ones. In general, diet analysis from SIA indicates assimilated contributions

of prey, which is related to the mass of diet consumed, while diet analysis from counting prey items from pellets, prey remains, or field observations are direct numbers caught by the birds (Resano-Mayor *et al.* 2014b). Vertebrate prey may contribute more biomass to the consumer after a single hunt than smaller invertebrates with indigestible exoskeletons, and even the smaller number of vertebrate sources can contribute more to blood stable isotope ratios than a large number of insects. Therefore, this study evaluated the relative dietary significance of different groups in terms of overall diet assimilation, whereas the previous studies noted only the occurrence and frequency of used prey items in diets, resulting in different estimates of vertebrate use in these species. Further studies, comparing two different methods and their results are required to estimate the effects of each diet item on assimilated isotopic values. In other comparative studies of sympatric owl diets, larger species consumed larger vertebrate preys more often, and smaller species relied on more invertebrate prey sources (Toyama and Saitoh 2011, Zhao *et al.* 2011), which was in concordance of estimation in this research .

Northern Boobooks showed greater carbon isotope variation with a higher CV (4.49%), which implies that they consumed foods from diverse carbon sources. On the other hand, Oriental Scops Owls showed smaller variation in carbon stable isotope ratio (CV=0.98%) among individuals. In general, plants growing under dense canopy with lower light intensity and more recycling of biogenic CO₂ have lower ¹³C/¹²C ratios than plants in open environments (Drucker and Bocherens 2009). Also, Trees as carbon sources in temperate forests are mostly C3 plants, whereas all C4 plants recorded in Korea are herbaceous species that prevail in open habitats rather than closed-canopy forests (Kim *et al.* 2011). These two different photosynthetic pathways, Calvin cycle (C3) and Hatch-Slack (C4) cycle, have profound difference (*circa.* 11.0‰ higher in C4 plants) in isotopic discrimination of ¹³C during photosynthesis (Marshall *et al.* 2007), and perhaps greatest source of variation within the study area. In addition, leaf δ¹³C along a vertical profile of the forest increases by canopy height as assimilation rate increases (Hanba *et al.* 1997). In the context of these vertical and horizontal profiles of primary producer carbon sources in forest food webs, the difference in carbon isotope ratios between Northern

Boobooks and Oriental Scops Owls could be explained by the Northern Boobook's utilization of prey from more diverse trophic webs, ranging from closed-canopy forests to carbon enriched open habitats. On the contrary, Oriental Scops Owls in this study area mainly relied on carbon sources from a very confined ecosystem in forests, not extending to open habitats.

5.3 Body size and trophic ecology

Most raptors show reversed sexual size dimorphism (Mueller 1986). Among multiple hypotheses to explain the RSD in raptors, Kruger (2005) suggested that diet might have a strong influence on the evolution of reversed sexual dimorphism: females prefer to select smaller, more agile, and more efficient males for breeding. This implies the avoidance of inter-sexual competition for limited prey resources in their territories, and it also means that the males have to be more efficient in foraging to supply food for chicks and brooding females.

Because of the efficient flight of smaller bodied birds by that have lower weight and hence lower wing loading, being small while foraging may be beneficial to individual adaptation in terms of energy efficiency in foraging (Andersson and Norberg 1981). In Northern Goshawks (*Accipiter gentilis*), smaller body size allowed birds to take an abundance of smaller and more agile prey (small birds and mammals) in Finland (Tomberg *et al.* 1999). In vole-eating raptors, Eurasian Kestrels (*Falco tinnunculus*) (Hakkarainen *et al.* 1996, Massemin *et al.* 2000) and Tengmalm's Owls (Hakkarainen and Korpimäki 1991), pairs with smaller males had more productive breeding results in low vole years when prey availability decreases. Because smaller male kestrels handed more voles to females during courtship during low vole years, small size in males can be more sexually selective trait in pair formation (Hakkarainen *et al.* 1996). In Tengmalm's Owls in Finland, it is suggested that smaller males may be preferred by females, since they are more efficient in foraging and territorial defense, hence can provide better prey items to the females (Korpimäki 1986). All these examples support sexual selection hypothesis that larger dominant females prefer to select smaller energy-efficient males.

In this study, smaller Oriental Scops Owls had higher nitrogen isotopic signatures, suggesting that the smaller birds consumed prey from higher trophic levels than larger individuals. Although the correlation was not statistically significant in the males only, due to the small sample size, if the smaller body size itself is advantageous in capturing larger and more vigilant vertebrate prey, it may

work as same in both males and females. On the contrary, no relationship between trophic level and body size (PC1) in Northern Boobooks was found, not supporting the hypothesis that smaller individuals may prey on higher trophic level preys. This lack of relationship may be linked with their sexual size monomorphism.

These findings support the general hypothesis that reversed sexual size dimorphism is related to foraging or diets (Kruger 2005). Individual trophic level was related to body size only in sexually dimorphic Oriental Scops Owls, but not in monomorphic Northern Boobooks. My study on the size dimorphism and trophic level did not include any aspects of breeding success or sexual selection, but did successfully find the trophic level of each individual via stable isotope analysis method. However, the results of this study may help to explain why female Oriental Scops Owls select smaller males, which are at higher trophic levels (a potential index of higher hunting skills and better chick provisioning) causing reversed sexual dimorphism. Similarly, no difference in trophic condition by size may responsible for the sexual size monomorphism of boobooks. However, given the small sample sizes and the strongly sex-biased samples (in particular, females in boobooks), the relationship between sexual dimorphism and individual trophic condition by body size still remains unanswered. Nevertheless, the findings in this study provide a fundamental clue to understand the relationship between foraging and size dimorphism in small, sympatric nocturnal raptors in East Asia.

Chapter 6. Conclusions

Of the two examined species of small forest owls, only Oriental Scops Owls showed significant, but subtle, reversed sexual dimorphism. In contrast, no clear evidence of sexual dimorphism was found in Northern Boobooks, though there were subtle sexual differences in some morphometric parameters. I developed a sex discriminant function based on external measurements for each species that excludes sex-related plumage differences. Based on re-substitution and leave-one-out tests, error rates for discriminant functions of 20.39–20.78% and 30.26–30.65% were estimated for Oriental Scops Owls and Northern Boobooks, respectively. Given the high error rates, I suggest using this formula as a supplementary method to identify the sex of a bird correctly in the field.

Stable isotope analysis showed a clear separation between the two species, both in carbon and nitrogen isotope ratios. Northern Boobooks had a higher trophic level (3‰, one level) than Oriental Scops Owls, and they also utilized prey sources from more diverse ecosystems. The diet of Northern Boobooks consisted of more vertebrate prey, especially birds, while Oriental Scops Owls utilized ground insects the most followed by birds, mice and moths. These results suggest that these two sympatric owls in Korean forests occupy clearly different ecological niches in terms of foraging, and that Oriental Scops Owls could be considered as forest specialists while Northern Boobooks are generalist predators utilizing both forest ecosystems and open habitat ecosystems when hunting for prey. High proportion of vertebrate preys estimated from the blood stable isotope ratio infer the significance of vertebrate prey to their diet, although their number may be much lower than invertebrate preys.

Finally, the negative relationship with the body size and trophic level of individual Oriental Scops Owls supported the previous arguments of smaller and more efficient-hunting males that can hunt for larger, and thus higher trophic level prey. But this was not the case in Northern Boobooks, which individual body size did not correlated with individual trophic level. This study does not include sufficient sample size to generalize the results, but it may partially support the hypothesis of

smaller and better males in raptors with sexual dimorphism.

In this study, stable isotope analysis in terrestrial high-lever consumer successfully identified different food niche between species that are known to have similar food habit. And I suggest that the utility of stable isotope analysis as a measure of individual trophic level can be a useful tool for studying the relationship between morphometric and individual diet.

This study is limits its scope on the food of the birds to explain the evolutionary fitness of reversed sexual dimorphism with only a handful of individuals. I suggest that the presence and absence of sexual dimorphism in sympatric two owl species with separated, but relatively close food niche, make them excellent models to study the inter-species interaction and driving force of the sexual dimorphism. Therefore, further study with more comprehensive approach, in example, study of sex roles during breeding, foraging mode and behavior, and comparison with species would be needed to increase our knowledge of these two owl species and general concepts of sexual dimorphism and intra-guild interaction between nocturnal raptors.

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Appendix

Appendix 1

List of Potential prey items collected.

Prey type	Common name	Scientific name	# of samples
Mice	Striped Field Mice	<i>Apodemus agrarius</i>	3
Moths	Hawk moth	<i>Phillosphingia dissimilis</i>	1
	Noctuid moth	<i>Xestia</i> sp.	1
Bush crickets	Ussur Brown Katydids	<i>Phillosphingia dissimilis</i>	3
	Katydid sp.	<i>Eobiana engelhardti</i>	1
Beetles	Adams stag-horned beetles	<i>Dicranocephalus adamsi</i>	3
	Carabus beetle	<i>Carabus smaragdinus</i>	1
	Stag beetles	<i>Dorcus rectus</i>	2
	Chafer beetles	<i>Melolonthinae</i>	2
Birds	Vinous-throated Parrotbill	<i>Sinosuthora webbiana</i>	1
	Eastern Great Tit	<i>Parus minor</i>	1
	Tristram's Bunting	<i>Emberiza tristramii</i>	1
	Pale Thrush	<i>Turdus palidus</i>	1

국문 초록

한국에서 서식하는 소쩍새(*Otus sunia*)와

솔부엉이(*Ninox japonica*)의

성적 이형성과 먹이-니치 연구

소쩍새와 솔부엉이는 한국에서 번식하는 비교적 흔한 소형 산림성 맹금류임에도 불구하고 두 종의 형태적 특징 및 생태에 대한 정보는 매우 부족한 실정이다. 본 연구에서는 두 종의 성적 이형성을 파악하고, 안정성동위원소 분석을 통하여 초기 번식기 동안의 먹이-니치 분할에 대하여 파악하고자 하였다. 그리고 이를 바탕으로 개체별 몸 크기와 먹이이용 관계를 비교하여, 맹금류의 역전된 성적 이형성을 진화적 관점에서 고찰하였다. 2014년과 2015년 두 조사기간 동안 광릉숲과 서울대학교 관악수목원에서 30개체의 소쩍새와 10개체의 솔부엉이를 포획하여 외부 형태를 측정하고, 성식별과 안정성동위원소 분석을 위한 혈액과 깃털을 수집 후 방사하였다. 추가적으로 4곳의 야생동물 구조센터에서 폐사하거나 구조 후 계류중인 솔부엉이 22마리와 소쩍새 16마리에서도 외부 형태를 측정하고 성식별을 위한 조직을 수집하였다. 또한 다원혼합모형분석을 위한 조류, 설치류, 나방류, 및 육상 곤충류 등 잠재적인 먹이원도 수집하여 분석하였다. 두 종의 외부형태를 분석한 결과 소쩍새는 미미하나 통계적으로 유의한 역전된 성적 이형성이 나타난 반면, 솔부엉이에서는 성적 이형성이 나타나지 않았다. 측정치를 통한 판별식 산출 결과, 소쩍새의 경우, 부리높이, 부리 폭, 꼬리길이, 부척길이가 유의한 단계적 판별분석의 변수로 선정되었으며, 판별분석을 통해 동일 표본에서 79%의 성이 올바르게 판별되었다. 솔부엉이는 머리길기와 두개골부터의 부리길기가 단계적 성판별분석에 선정되었으며, 동일표본의 70%만 올바르게 판별되었다. 두 종 모두 판별분석의 정확도가 낮고, 성적이형성이 없거나 차이가 크지 않으므로, 성 식별 시 포란반 등 추가적인 정보를 참고해야 하

며, 가능하다면 분자생물학적 방법이 이용되어야 할 것으로 보인다. 혈액 안정성동위원소 분석 결과, 질소 안정성동위원소비가 솔부엉이가 소쩍새에 비해 높게 나타나 솔부엉이가 소쩍새보다 높은 영양단계의 먹이를 이용하는 것으로 나타났다. 탄소 동위원소비의 경우, 탄소 동위원소비 뿐만 아니라, 동위원소비의 분산도 솔부엉이가 높게 나타나, 솔부엉이가 산림 내부에서부터 외부까지 더 다양한 생태계로부터 먹이원을 이용하는 유연한 섭식 행동을 하는 것으로 추정되었다. 이러한 명확한 안정성동위원소의 차이는 동일 지역에서 서식하는 소쩍새와 솔부엉이간 명확한 먹이-니치 분할이 이뤄진 결과로 판단된다. 다원혼합모형결과, 솔부엉이는 특히 조류를 포함한 척추동물 먹이를 주로 소비한 것으로 추정되었으며, 소쩍새는 육상 곤충을 가장 많이 소비하였고 조류, 설치류, 나방류를 다음으로 많이 소비하는 것으로 추정되었다. 이는 기존연구에 비해 척추동물의 비율이 매우 높은 결과로서, 조사 시기 및 연구 방법에 의한 것으로 판단된다. 개체 별 크기와 영양단계간 비교를 위해, 주성분분석을 이용하여 추출한 개체의 몸 크기를 대표할 수 있는 제 1 주성분을 개체의 몸 크기를 대표하는 지수로 삼아 개체의 질소 안정성동위원소비와 상관분석을 실시하였다. 분석결과, 작은 개체가 높은 영양단계의 먹이를 소비한 것으로 분석된 소쩍새와 달리 솔부엉이는 상관성을 나타내지 않았다. 성적이형성이 나타난 소쩍새에서만 개체의 영양단계와 몸 크기의 상관성이 나타났다는 점에서, 이는 소쩍새에서 작은 크기의 수컷이 더 효율적인 사냥을 통해 진화적으로 역전된 성적이형성을 강화한다는 ‘작은 수컷 가설’을 부분적으로 지지한다고 볼 수 있다.

주요어: 먹이원 재구성, 안정성 동위원소 분석, 역전된 성적이형성, 영양 단계, 작은 수컷 가설, 판별 분석

학번: 2013-23247

감사의 글

처음 새에 대해 알게 된 후부터, 석사과정을 마무리하기까지 수 많은 분들이 제게 도움을 주셨습니다. 철없고 부족한 저를 따뜻한 마음으로 지원해주신 수많은 분들께 감사하다는 이야기를 드리고자 합니다.

먼저 새가 좋다고 나선 대학생이었던 저를 석사 대학원과정까지 저를 이끌어주시고, 연구를 계속할 수 있도록 지도해주신 지도교수 이우신 선생님, 감사합니다. 세계의 다양한 지역과 생태계에서 공부할 수 있도록, 한반도의 새들을 공부할 수 있도록, 선생님께서 주신 배움의 기회들은 제가 조금 더 성장하는데 든든한 초석이 되었습니다. 바쁘신 와중에도 논문 심사위원장과 심사위원을 흔쾌히 맡아주신 김현석 선생님과 박찬열 박사님께도 감사를 드립니다. 정말 부족함이 많았던 초기 원고부터 논문 제출까지, 지도해 주시고 믿고 독려해 주셔서 감사합니다.

숲 생태계와, 그 안에서 살아가는 새들에 대한 관심은 학부과정을 지나며 더 커졌습니다. 학부 과정동안 산림생태계의 면면들을 배울 수 있도록 지도해 주신 이돈구, 한정오, 이경준, 장진성, 박필선 선생님, 산림 환경학의 다양한 분야를 지도해 주신 정주상, 윤여창, 김성일, 임상준, Victor Teplyakov 선생님, 대학 과정 동안 조언과 격려를 아끼지 않고 해 주신 강규석, 박일권 선생님, 학교에서 선생님들께 받은 가르침을 잊지 않겠습니다.

처음 연구실에 나타나, 이것저것 물어보고, 현장 조사에 데려가 달라고 졸랐던 저를 너그럽게 봐 주시고, 야생동물 연구의 다양한 방면을 보여주신 연구실의 허위행 박사님, 정옥식 박사님, 박용수 박사님, 이은재 박사님, 이종구 선배 등 여러 선배들이 아니었다면 지금의 제가 있을 수 없었을 것입니다. 대학원 과정에 입학한 후 함께 고생하며 지낸 연구실 친구들과 선배께 감사의 인사를 드립니다. 늘 고민을 해결해 주는 김종우 선배, 동물 이야기를 늘 할 수 있는 이성민 선배. 열린 마음으로 함께 토론해준 이화수 선배, 삼척, 홍천 등 지방 조사를 많이 함께한 박창득 선배, 그리고 야외조사를 많이 도와 준 정민수, 늘 표본관리에 힘써주는 정지화, 늘 비판적으로 대해줘서 고마운 김준수, 편하게 다가와준 전중훈, 여러 야외조사를 함께한 탁성준, 연구 관련된 의견을 나눌 수 있었던 홍누리에게도 감사의 마음을 표시하고 싶습니다. 또, 연구실에서 함께 생활한 Bounthan Sounyvong 과 Hsu Sandar Aung에게도 고맙다는 말을 전하고 싶습니다. 지금은 철새연구센터에서 근무하는 박세영 선배와 남극 연구도 함께 했던 강화연의 조언도 도움이 되었습니다. 연구실에서 연구과제들을 수행하며 박현정 선생님의 도움이 없었다면 연구비 관리와 관련 행정업무에 큰 어려움이 있었을 것입니다. 정말 감사합니다. 동아리 선배이자 연구실 선배인 최창용 박사님은 현장 조사부터 분석, 논문 작성에 필요한 구체적 기술과 윤리를 알려주시고, 새에 대한 제 지

평을 넓혀주셨습니다.

PCR 기기 사용법 등 첫 유전자분석을 도와준 산림유전 육종학실 박지민과 조류의 DNA 성 식별의 많은 노하우를 이야기해준 자연대 행동생태학실 이기산, 언제 찾아가더라도 산림생태계에 대해 토론할 수 있는 생태학실 정종빈 선배들께도 감사드립니다.

선생님들과 선배, 동료들의 지원과 가르침에도 불구하고 저의 부족으로 학위논문이 여러 기대에 비하여 부끄럽게 느껴집니다. 이러한 부족함을 꾸준한 노력으로 보완할 수 있도록 하겠습니다.

조사지역인 광릉숲이 위치한 국립수목원과 서울대학교 관악수목원 지역은 산림생태계의 보전과 연구를 위해 철저하게 관리되는 곳으로 이 연구를 위한 출입과 연구활동을 지원해 주신 신준환 전 원장님과 이유미 원장님을 비롯한 직원분들의 도움이 없었다면 이 성과를 이루지 못했을 것입니다. 빈번한 야간 시간대 출입에도 불가하고 안양에 위치한 관악수목원 소장님과 직원분들 또한 친절하게 연구활동과 출입을 허가해 주셔서 야외에서 많은 것을 배울 수 있었습니다. 감사합니다. 솔부엉이와 소쩍새는 현재 천연기념물로 허가 없이 포획과 연구가 불가능합니다. 무화재청에서 해당 허가 신청을 검토하고 허가하여 주지 않았다면 이 연구는 불가능했을 것입니다. 다시 한번 감사합니다. 솔부엉이와 소쩍새의 개체 식별을 위한 국가 금속가락지를 보내주시고, 늘 응원해주신 생물자원관 김화정박사님 감사합니다. 그리고 관악수목원과 광릉숲에서 순순히 그물에 걸려, 혈액 시료와 깃털 시료를 제공해주고 바쁜 와중에 상당한 시간을 소비해준 소쩍새와 솔부엉이에게 미안한 마음과 감사한 마음을 꼭 전하고 싶습니다.

각 야생동물 구조센터에서 시료 수집을 할 수 있도록 연락해 주시고, 맹금류와 관련된 소식을 늘 공유해주시는 강승구박사님이 없었다면 추가적인 시료 수집에 어려움이 많았을 것입니다. 경기도 야생동물구조센터의 류시영 센터장님과 김희원 주무관님, 울산야생동물 고영진 센터장님, 오세영 주임님 부산 야생동물치료센터 정명규 팀장님의 도움으로 시료 수집을 할 수 있었습니다. 감사합니다. 부산 치료센터에서 한현진 선배께서는 시료 수집을 도와주시고, 맹금류를 다루는 연습을 도와주셨습니다. 경성대학교 조류관 우동석 선생님은 함께 맹금류 연구에 대한 이야기를 많이 해 주셨습니다. 감사합니다. 제주도에서 강창완 선생님과 김은미 선생님도 연구를 위한 시료 수집을 도와주셨습니다. 감사합니다.

연구로 이어진 새에 대한 관심은 대학교 동아리 야생조류연구회로부터 시작되었습니다. 동아리에서 함께 새를 보며 제게 많은 것들을 알려준 선배, 친구, 후배들은 늘 마음속의 기둥이 되어 저를 지지해 주었습니다. 동아리 새내기였던 저를 따뜻하게 받아주신 최서희, 김윤희, 명판도, 권민우, 이동욱, 이민재 선배님과 부족한 저를 늘 지원해준 박재찬, 강희철, 길보경, 장지웅, 손영식, 신광원, 이창훈, 이광문 선배님, 그리고 정말 울고 웃으며 함께 지냈던 동기 임록근, 이강희, 권아란, 김태영과 김시연,

조민제, 그리고 08, 09 후배들에게 진심으로 감사합니다.

최근 3년간 남극 세종기지를 방문하며 만나게 된 선배 연구자들과 박사님들과 다양한 분야에서 교류할 수 있었던 것은 제게 큰 행운이었습니다. 특히 야조새 선배이자 펭귄을 연구하는 정진우 선배님께 많은 신세를 졌습니다, 같이 새를 연구하는 한영덕과 이원영박사님께도 많은 빛을 졌습니다. 남극에서 만난 김준태, 최성준 선배, 윤영준 박사님, 김석철 선배, 송관영 강사님, 그리고 김상희박사님과 김정훈박사님께도 많은 도움을 받았습니다. 정말 감사합니다.

소쩍새와 솔부엉이를 비롯한 맹금류 연구에 대한 첫 발걸음은 미국 Hawk Mountain Sanctuary에서의 인턴쉽 기간 동안 이루어졌습니다. Keith Bildstein 박사님, Laurie Goodrich 박사님, J-F Therrein 박사님. David Barber 박사님과의 대화와 실습에서 맹금류 연구에 대한 많은 도움을 받았습니다. 또 인턴 동기생이었던 Chong Leong Puan 박사님은 논문 연구에 대해서 조언해 주셨습니다. 호주 캔버라 대학의 Jerry Olsen 박사님과 Sue Trost 선생님은 지난 2015년 4-5월 한국에 방문하여 소쩍새 포획 작업을 도와주셨고, 맹금류의 역전된 성적이형성과 예외적인 호주의 올빼미류에 대하여 많은 조언을 해 주셨습니다.

새를 공부하며 짧은 지면에 다 쓸 수 없을 만큼 많은 분들을 만났고, 많은 것을 배울 수 있었습니다. 특히 국립습지센터의 박진영 박사님, 국립공원연구원의 박종길 선배님의 식지 않는 새에 대한 열정과 생태원 김영준 수의사님, 충남 구조센터 김희중 수의사님의 야생동물에 대한 헌신은 늘 저를 반성하고 되돌아보게 합니다.

마지막으로, 어떤 상황에서도 저를 포기하지 않고 믿고 지지해주신, 사랑하는 어머니, 아버지, 할머니, 동생에게 이 논문을 바칩니다.

2015년 7월 관악캠퍼스에서

김한규