농학석사 학위논문

Relationship between body condition and breeding performance in Varied Tits (*Sittiparus varius*)

곤줄박이(*Sittiparus varius*)의
건강 상태와 번식의 관계

2017 년 2 월

서울대학교 대학원
산림과학부 산림환경학 전공
탁 성 준

Relationship between body condition and breeding performance in Varied Tits (*Sittiparus varius*)

곤줄박이(*Sittiparus varius*)의

건강 상태와 번식의 관계

지도 교수 이 우 신

이 논문을 농학석사 학위논문으로 제출함

2017 년 1 월

서울대학교 대학원

산림과학부 산림환경학전공

탁 성 준

탁성준의 농학석사 학위논문을 인준함

2017 년 1 월

위 원 장 ______ 강 규 석 ______ (인)

부위원장 ______ 이 우 신 ______ (인)

위 원 ______ 정 옥 식 ______ (인)

Abstract

Reproduction in birds is an immensely energy–consuming process. Like many other bird species, body condition in tits affects breeding performance throughout many different stages. The purpose of this study was to investigate how this relationship between body condition and breeding performance functions in the Varied Tit (*Sittiparus varius*), and artificial nextboxes were used in Mt. Baekwoon, the Republic of Korea. The body condition of parents was measured before and at the end of the breeding period of 2016 using both the Body Condition Index (BCI) and the level of glycated hemoglobin (HbA1c). Breeding performance was comprehensively derived from the laying date, clutch size, breeding success, and chick condition and growth rate.

The HbA1c level of females before the breeding period was positively correlated to chick weight, which seems to be a result of higher energy reserve for chick rearing and possibly better foraging ability. The relationship between the BCI of parents and breeding performance was different by sex at the end of the breeding period, and this is thought to be the result of different breeding strategies between the sexes. The BCI of females showed negative correlation with chick condition at the late stage, while chicks of males in better condition grew faster. While the decrease in BCI of females seems to represent direct investment of energy for chicks, the BCI of males seems to represent the resource holding potential which influences the growth rate of chicks. Early start of breeding resulted in larger chicks at first, but the gap among nests was mitigated possibly due to intensive parental care of late breeders. No significant relationship was found between the HbA1c and the BCI, and the HbA1c of parents measured at the end of the period was not significantly correlated to any of the breeding performance variables.

In this study, the body condition of Varied Tits showed various correlations with breeding performance. HbA1c could be used as an index to represent a bird's body condition before or at the early stage of breeding, while at later stages of breeding, conventional measurements such as the BCI seem to be more reliable in representing the body condition of parents and reflecting breeding performance.

Keywords: body condition, Body Condition Index, breeding performance, glycated hemoglobin, Varied Tit

Student Number: 2015-21502

Table of Contents

List of Tables

List of Figures

1. Introduction

For many birds, reproduction is an immensely energy-consuming process. To achieve a successful outcome, individual birds consume a great amount of energy to build nests, lay eggs, and take care of chicks; the process is often a trade-off between an individual bird's fitness and reproductive success. Excessive investment in reproduction can affect the body condition, future reproduction and even survival of parents (Linden 1988, De Laet and Dhondt 1989, Visser and Lessells 2001). Some parents even abandon their clutch when this trade-off does not seem to result in their benefit (Dufva 1996, Dubiec 2011).

This concept applies to tits (Family Paridae, including the genus *Sittiparus*, *Parus*, *Poecile* and many more) as well as many other birds. Although each individual invests differently in its offspring, parent tits with better body conditions tend to achieve higher success in reproduction. Being in better condition can simply mean that one contains more energy to deliver to its chicks (Sanz and Tinbergen 1999), which implies that chicks of fit parents are more likely to have higher fitness before fledging and survive (Smith et al.

1989, Tinbergen and Boerlijst 1990, Naef−Daenzer and Keller 1999). But in a way, higher investment in breeding could negatively influence an individual's body condition and act as a determinant of whether the parents will continue to raise the nestlings or abandon them, depending on the environment (Hõrak et al. 1999) or if disturbance occurs (Dubiec 2011).

Similarly, the body condition of tits before or during the early stages of breeding can affect the reproductive outcome in many ways. For tits that feed mainly on caterpillars during the breeding season, it is advantageous to lay eggs early and control the breeding stages based on food supply (Perrins and McCleery 1989, Perrins 1991, Verhulst and Tinbergen 1991, Van Noordwijk et al. 1995, Hinks et al. 2015). Females with better body conditions generally lay eggs earlier (Norte et al. 2010) and are known to have a bigger clutch size (Slagsvold and Lifjeld 1990, Norte et al. 2010).

Body condition in birds is measured in many different ways. Most commonly, some of physical measurements such as the tarsus, weight, and alula are used together to create an index, such as the Body Condition Index (BCI), to represent an individual's physical condition (Johnson et al. 1985, Labocha and Hayes 2012, Guindre−

Parker et al. 2013). In recent studies, hematological parameters such as hematocrit, the heterophile/lymphocyte ratio (Kilgas et al. 2006, Norte et al. 2010), the hemoglobin level (Kaliński et al. 2015) and parasites (Rooyen et al. 2013, Bailly et al. 2016) have been considered as well.

Glycated hemoglobin (HbA1c) is another measure that has recently been used as an indicator of avian body condition (Andersson and Gustafsson 1995, Ardia 2006). Although it has not been fully certified as an appropriate indicator of body condition, it can reflect the relative long-term condition of birds and can indicate the nutritional status of breeding birds (Andersson and Gustafsson 1995).

The Varied Tit (*Sittiparus varius*) is a socially monogamous species (Yamaguchi et al. 2004) that breeds in tree cavities (Yamaguchi and Kawano 2001) and artificial nestboxes. With their limited distribution in East Asia, their breeding ecology is studied relatively less compared to more common species that are widely distributed, such as the Great Tit (*Parus major*).

In this study, the correlation between the body condition and breeding performance of the Varied Tit was investigated. The purpose of this study was to identify how pre-breeding and breeding body condition is related to breeding performance, using nutritional as well as physical measurements of body condition. The hypothesis of the study was that the BCI and HbA1c would be indicators that adequately reflect the body condition of breeding Varied Tits and thus could be utilized to predict their breeding performance.

2. Literature Review

2.1 Body condition and reproduction of tits

Since reproduction is a highly energy and time−consuming process for many birds, the body condition of parents before and during the breeding period is a crucial factor that affects the time of breeding, clutch size, clutch desertion, condition of chicks, breeding success and many more throughout the stages of breeding.

Like several other families of birds, a number of studies have been conducted in family Paridae to verify the relationship between body condition and reproduction.

Firstly, the pre−breeding condition of tits was proven to be correlated to some of the early reproductive decisions. In the Great Tit, females in good condition could mobilize a large amount of nutrients for egg formation early in the season when food is scarce and are able to start breeding early (Norte et al. 2010). Likewise, as clutch size in altricial birds is generally thought to be limited by the ability of parents (Lack 1947), females with lower fitness were seen to lay smaller number of eggs (Slagsvold and Lifjeld 1990, Sanz and Tinbergen 1999).

Secondly, many studies have proven that condition of parents during the breeding period influences the outcome. Dubiec (2011) suggested that individuals with worse body condition were commonly found to desert their clutches, while Hõrak et al. (1999) reported that birds giving up on their nests were heavier in weight. This is still controversial because desertion could occur when (1) in better body condition, because they have invested less, or (2) in worse body condition, because they cannot physically continue to invest in their chicks (Hõrak et al. 1999). Nevertheless, it is quite clear that body condition can affect an individual's decision on clutch desertion and consequentially the reproductive outcome. And it shows that the body condition of parents also varies based on the level of investment they made on breeding, as it is proven in some studies where indicators showed that the body condition of females deteriorated while breeding (Verhulst 1998, Dubiec and Cichoñ 2001). Likewise, Naef-Daenzer and others have identified that the time of fledging and fledging mass were related to the survival rate of post-fledging chicks (Naef-Daenzer et al. 2001) and these factors are highly dependent on their parents' breeding performance(Garnett 1981).

In summary, the body condition of tits before and during breeding is known to affect breeding performance in many different stages.

2.2 Ways to measure the condition of birds and HbA1c

To represent a bird's condition using a single index is challenging and there are many ways to measure the condition of birds. Among those, physical measurements are used most widely. In various studies, some of the tarsus, weight, beak and wing measurements have been used together to represent the physical condition of birds (Johnson et al. 1985, Labocha and Hayes 2012, Guindre−Parker et al. 2013). Parasites are also related to the condition of birds and have been used to evaluate the condition of tits (Rooyen et al. 2013, Bailly et al. 2016).

Recently, hematological parameters are used more frequently. Hematocrit, albumin, globulin and triglyceride concentrations, and the heterophile/lymphocyte ratio are some of the hematological parameters that are used as indicators for physiological condition (Ots and Horak 1996, Ots et al. 1998, Kilgas et al. 2006, Norte et al. 2010).

Unlike many other hematological parameters used widely in avian studies, glycated hemoglobin (HbA1c) has only recently been applied to birds to measure their condition. It is thought to be able to reflect the average blood glucose level during the 3–5 weeks before the blood sampling in birds (Andersson and Gustafsson 1995). Andersson and Gustafsson (1995) have found that the level of HbA1c in the Collared Flycatcher (*Ficedula albicollis*) was significantly correlated to arrival date, clutch size, and the number of fledged young and suggested that HbA1c be used as an indicator of body condition for birds. Similarly, Beuchat and Chong (1998) proved that in hummingbirds (*Calypte anna*, *C. costae*, and *Archilochus colubris*), high blood glucose concentrations were accompanied by high HbA1c levels. Also in American Kestrels (*Falco sparverius*), HbA1c was found to reflect nestling growth and condition along with the albumin level (Ardia 2006). Furthermore, it was discovered to be correlated to the blood glucose level of the Mute Swan (*Cygnus olor*) and Rook (*Corvus frugilegus*) (Mikšik and Hodný 1992).

Although there were a few studies where HbA1c was used as a measurement of avian body condition, HbA1c in non-human animals is yet at the early stage of research and its mechanism has not been fully identified (Ardia 2006).

2.3 Study species: Varied Tit

The Varied Tit (*Sittiparus varius*) is a small tit species of family Paridae. Four subspecies are limitedly distributed in East Asian countries; *S. v. varius* in Korea, Japan and China, *S. v. sunsunpi* off south Japan, *S. v. namiyei* and *S. v. orii* in Izu and Ryukyu island of Japan respectively (Gill and Donsker 2016).

It is a socially monogamous species (Yamaguchi et al. 2004) and one of the most common tit species within Korea, together with the Great Tit (*Parus major*), Marsh Tit (*Poecile palustris*) and Coal Tit (*Periparus ater*). They are cavity-nesters that nest in tree cavities made by woodpeckers (Yamaguchi and Kawano 2001) and often use artificial nestboxes for breeding (Lee et al. 2000).

Unlike the closely-related Great Tit, which is one of the most widely studied tit species, the Varied Tit has not been the subject of many studies. In particular, their breeding ecology has not been investigated thoroughly; therefore, this study is meaningful in that it specifically looks into the breeding patterns of the Varied Tit.

In Korea, its general breeding ecology has been studied using nestboxes (Rhim et al. 2011) and in Mt. Jiri, parental provisioning of nestlings was studied at different altitudes (Lee et al. 2011). In the study, provisioning rate of parents changed with altitude, different stages of breeding, and also differed by sex. Another study done at the same sites concluded that an increase in the provisioning rate at higher elevation is associated with the decrease in prey size (Lee et al. 2016).

The Varied Tit has been a species of study interest in Japan and China as well. The effect of body size on the resource holding potential (Yamaguchi and Kawano 2001), cheek-patch coloration (Yamaguchi 2005) and life history of three different subspecies (Yamaguchi and Higuchi 2005) were studied in Japan. It has been found that supplemental food changed group structures (Kubota and Nakamura 2000) and the pair bond of Varied Tits (Nakamura 1998), while the sex ratio of chicks in a brood was significantly related to the father's tarsus length (Yamaguchi et al. 2004). In China, the mating system, especially the extra-pair paternity of Varied Tits has recently been studied (Ju et al. 2014, Ma et al. 2015).

Nevertheless, studies on the Varied Tit are still limited compared to many other species of family Paridae.

3. Materials and Methods

3.1 Study sites and nestboxes

The research was conducted at four study sites of different altitudes in Mt. Baekwoon (35° 07' 01.18” N, 127° 36' 10.43” E), Gwangyang, Korea, from March to July in 2016 (Figure 1). Mt. Baekwoon is located on the southern end of the Korean peninsula and 9.74 km^2 of the mountain area was designated by the Korean Ministry of Environment as an Ecological and Landscape Conservation Area in 1993. Most of the area is covered with natural cool-temperate forest which consists of deciduous broadleaf species such as *Quercus serrata*, *Q. mongolica*, *Carpinus laxiflora*, *Cornus controversa*, *Fraxinus mandshurica* and *Acer pictum* var. *mono*. Annual rainfall is around 1300-1500 mm and average monthly temperature was highest (27.9℃) in August and lowest (2.2℃) in January (Unpublished data from the Korea Meteorological Administration).

Artificial nestboxes were set up at four sites in Mt. Baekwoon before the Varied Tit's breeding period of 2016; one at an elevation of 1,000 m, two at 800 m (Hanjae and Dosolbong) and one at 400 m.

A total of 184 nestboxes (each with an entrance diameter of 35 mm) were installed: 40 nestboxes per site at 400 m, and 48 per site at 800 m and 1,000 m.

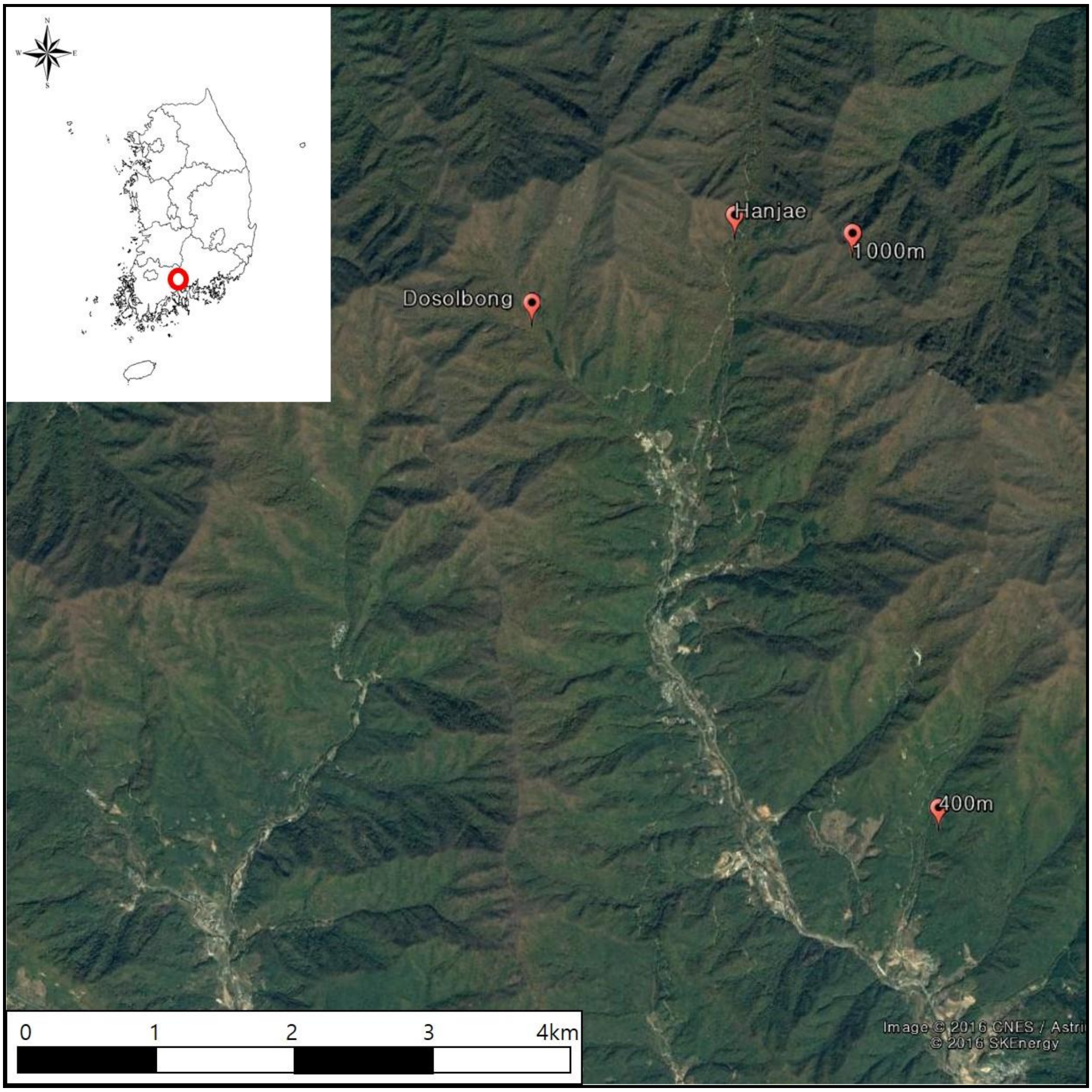

Figure 1. Location of the four study sites at different altitudes in Mt. Baekwoon; 400 m, 800 m (Hanjae and Dosolbong) and 1,000 m a. s. l.

3.2 Nestbox monitoring

From mid-April to early July, the nestboxes were visited 2-3 times every week to check the breeding species, laying date, clutch size, and breeding success of each nest. As tits are known to lay one egg every day, the laying date of the first egg was deduced from the number of eggs found on the day of visitation. The breeding success of a nest is usually calculated as the ratio of the number of chicks fledged to the clutch size (Verhulst and Tinbergen 1991), but to prevent premature fledging of chicks by disturbance, nests were finally checked on day 13 after hatching and the number of chicks survived on this day was used to calculate the breeding success. Monitoring was minimized during incubation to reduce possible disturbance and from the 10^{th} day of incubation, nests were checked every day to verify the hatching date. After hatching, nests were checked on days 3, 8 and 13 after hatching, to check the condition of the chicks (see 3.3).

3.3 Condition of parents and chicks

The condition of the parents was checked before and at the end of the breeding period. From late March to early April, birds were captured at four study sites using mist nets and conspecific audio lures. Nets were checked at 20–30 minute intervals, and birds were removed immediately when noticed. Varied Tits were ringed, measured, blood sampled and released immediately to minimize stress. When more than two birds were caught simultaneously, birds were kept in cotton bags hung above the ground before being processed and none of the birds were kept for more than 30 minutes. On the other hand, birds were also captured from their nestboxes at the end of the breeding period. On day 13 after hatching, traps were set up above the entrance and when a parent entered the nest, the entrance was blocked using the trap (Figure 2). Then a net was wrapped around the nestbox to capture the bird. The male and female were captured separately and they were both ringed (in case they did not have a ring around the tarsus), measured, and blood sampled. None of the birds captured abandoned the nest on the day of capture.

Figure 2. A trap covering the entrance of a nestbox

For each captured parent, the following measurements were taken: wing length (maximum and flattened length) and tail length to the closest 1 mm with a wing ruler, tarsus length, bill length to skull, bill depth and bill width to the closest 0.01 mm with calipers and weight with a digital scale in 0.01 g units (Figure 3).

A small amount (approximately 30−50 μl) of whole blood was collected from the brachial vein using a 30G needle and a micro capillary tube. I used 70% isopropyl ethanol swabs to clear the feathers and make the brachial vein visible. The lancing area was dried completely before the puncture. Directly after sampling, the extracted blood was used to measure the % level of glycated hemoglobin using an HbA1c test kit ($A1cNOW^{+}$; Polymer Technology Systems, Indianapolis, USA; Figure 4). According to the manufacturer, the range of the detectable percentage level of HbA1c was 4.0−13.0 and it was certified by the National Glycohemoglobin Standardization Program with the average of 99% accuracy.

Figure 3. Measuring a Varied Tit in pre-breeding period

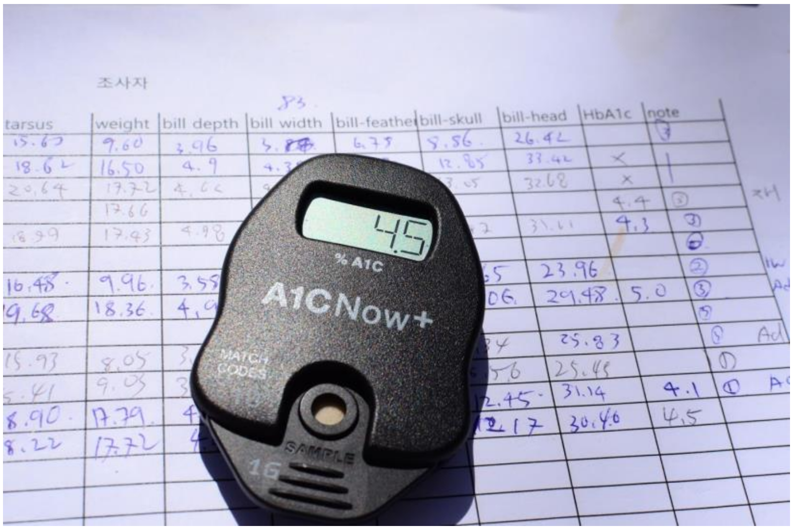

Figure 4. HbA1c test kit (A1cNow+) showing a value of 4.5% HbA1c level

On the other hand, chicks were captured from the nest and gently placed in a cotton bag. Each bird was measured three times to check its body condition at the early, middle, and late stage (day 3, 8 and 13 after hatching, respectively). They were ringed on day 13 and before then, their tarsi were colored for identification, using innocuous, water-based paint. The measurement of the tarsus and weight was done in the same way as their parents.

All the handling and sampling were done with caution and followed guidelines from reliable references (Ralph et al. 1993, Owen 2011).

3.4 Data analysis

3.4.1 Body Condition Index and HbA1c

Of all the measurements done in parents, tarsus length and weight were used for the Body Condition Index (BCI) to represent their body condition. Weight can reflect an individual tit's fat reserve, and tarsus length is known to be an inheritable trait in tits (Yamaguchi et al. 2004). Many different measurements including tarsus length, weight, alula size and bill size (such as depth, width) could be considered for an index (Johnson et al. 1985, Labocha and Hayes 2012). However, this study utilized one of the most widely used indicators of body condition, the residuals from a linear regression of body mass against tarsus length (Norte et al. 2010, Dubiec 2011, Labocha and Hayes 2012).

The percentage level of HbA1c was measured from the sampled blood, but the kit used in this study could detect only the percentage level between 4.0 and 13.0; therefore, HbA1c levels outside of this range were unattainable leaving them unidentified. Because of this problem, the measurements of HbA1c were first sorted by sex and period of sampling (pre-breeding and breeding;

measured before and at the end of the breeding period), and then divided into two groups for analysis. Measurements lower than 4.0% (written as "<4.0" on the kit) were categorized as "low" and measurements of 4.1 or higher were categorized as "high". None of the birds captured in this study showed an HbA1c percentage level higher than 13.0.

3.4.2 Statistical analysis

To analyze the correlation between the body condition of birds and their breeding performance, the BCI and percentage level of HbA1c were statistically analyzed with breeding performance variables.

The HbA1c level, which was divided into two groups (low and high), was compared with breeding performance using the Mann-Whitney U test. The breeding performance included clutch size, laying date, breeding success, average chick tarsus length and weight (of days 3, 8 and 13 after hatching), and average tarsus and weight growth rate (of days 3-8, 8-13 and 3-13). HbA1c was also compared with the BCI to see if they were correlated. Birds captured in the three sites of high altitudes (800 and 1,000 m a. s. l.) were analyzed together since these sites showed relatively consistent habitat conditions. Only the females were analyzed for the pre-breeding period, as not enough males were captured, and both males and females were analyzed for the breeding period.

For the analysis of the relationship between the BCI, laying date, and breeding performance, the Generalized Linear Mixed Model (GLMM) was performed using the *lme4* package (Bates et al. 2012) in the R software. P-values were obtained by likelihood ratio tests of the full model with the effect in question against the model without the effect in question. For breeding performance variables, breeding success, average chick tarsus length and weight (of days 3, 8 and 13 after hatching), and average tarsus and weight growth rate (of days 3-8, 8-13 and 3-13) were analyzed. As fixed effects, the BCI and laying date were used for females, and for males, only the BCI was used because the laying date is mostly dependent on females' decision. For random effects, site and clutch size were used as the altitude (Rooyen et al. 2013, Lee et al. 2016) and clutch size (Nilsson and Svensson 1993, Verhulst 1998) are known to affect the outcome of breeding and the condition of parents. Only the body condition during the breeding period was analyzed since not enough samples were collected during the pre-breeding period.

All analyses were performed with R software version 3.2.5 (R Core Team, 2016).

4. Results

4.1 Breeding status of Varied Tits

During the breeding period of 2016 in the four study sites, a total of 69 nestboxes were used and 52 of these were used by Varied Tits for breeding. Great Tits (*Parus major*) were found to breed using nestboxes and Siberian Flying Squirrels (*Pteromys volans aluco*) were also observed to use them.

Eggs of Varied Tits were found in 8, 18, 13 and 13 nestboxes at 400 m, Hanjae and Dosolbong at 800 m, and at 1,000 m respectively (Table 1). The first laying date of each site was different: the site at 400 m was the earliest on the 11th of April and Dosolbong at 800 m was the latest on the 18th of April (Table 1).

Table 1. Breeding status of nestboxes at the four study sites in Mt. Baekwoon in 2016

	400 m	800 m		1,000 m	Total
		Hanjae	Dosolbong		
First laying date	Apr. 11th	Apr. 14th	Apr. 18th	Apr. 14th	
No. of nestboxes used	13	23	17	16	69
No. of nestboxes used by Varied Tits	8	18	13	13	52
Clutch size	6.38 ±0.52	6.12 ±1.32	6.00 ±1.28	6.54 ±0.52	

4.2 HbA1c and breeding performance

4.2.1 Pre-breeding HbA1c and breeding performance

The percentage level of HbA1c in females before breeding was shown to have had a significant influence on the average chick weight of day 3 and 8 after hatching. Females with higher percentage levels of HbA1c in the pre-breeding period had heavier chicks on day 3 ($U=3$, $p=0.015$; Figure 5) and day 8 ($U=5$, $p=0.041$; Figure 6). But clutch size, laying date, breeding success, chick tarsus and growth rate were not significantly correlated to the pre-breeding HbA1c level of females (Table 2).

4.2.2 Breeding HbA1c and breeding performance

The HbA1c percentage level in both females and males during the breeding period did not have a significant correlation to clutch size, laying date, breeding success, chick tarsus and weight, nor chick growth rate (Table 3-4).

Table 2. Pre-breeding HbA1c of females and breeding performance in Varied Tits. Significant relationships are marked in bold.

	HbA1c	Median	Range	U	p
Clutch size	high (n=6)	6.5	6-8	15.5	0.739
	low (n=6)	6.5	5-10		
Laying date	high (n=6)	113.5	105-119	15.5	0.748
	low (n=6)	114	107-136		
Breeding success	high (n=6)	91.7	33.3-100.0	16.0	0.803
	low (n=6)	82.9	30.0-100.0		
Tarsus on day 3	high (n=6)	6.98	5.96-8.34	9.0	0.180
	low (n=6)	6.23	5.16-6.63		
Tarsus on day 8	high (n=6)	15.31	13.86-16.62	7.0	0.093
	low (n=6)	13.63	11.96-16.87		
Tarsus on day 13	high (n=6)	18.3	17.87-19.19	12.0	0.394
	low (n=6)	18.08	16.83-18.80		
Weight on day 3	high (n=6)	3.27	2.69-4.52	3.0	**0.015**
	low (n=6)	2.44	1.84-3.14		
Weight on day 8	high (n=6)	11.59	10.42-13.23	5.0	**0.041**
	low (n=6)	9.51	8.08-13.15		
Weight on day 13	high (n=6)	16.29	15.70-16.71	6.0	0.065
	low (n=6)	14.85	13.87-16.86		
Tarsus growth between day 3-8	high (n=6)	1.16	0.83-1.63	16.0	0.818
	low (n=6)	1.22	0.98-1.55		
Tarsus growth between day 8-13	high (n=6)	0.19	0.15-0.31	7.0	0.093
	low (n=6)	0.33	0.12-0.50		
Tarsus growth between day 3-13	high (n=6)	1.7	1.15-2.03	13.0	0.485
	low (n=6)	1.87	1.76-2.28		
Weight growth between day 3-8	high (n=6)	2.42	1.52-3.31	6.0	0.065
	low (n=6)	3.26	2.64-3.72		
Weight growth between day 8-13	high (n=6)	0.38	0.28-0.59	9.5	0.180
	low (n=6)	0.55	0.19-1.24		
Weight growth between day 3-13	high (n=6)	4.06	2.48-5.19	10.0	0.240
	low (n=6)	4.94	4.05-8.60		

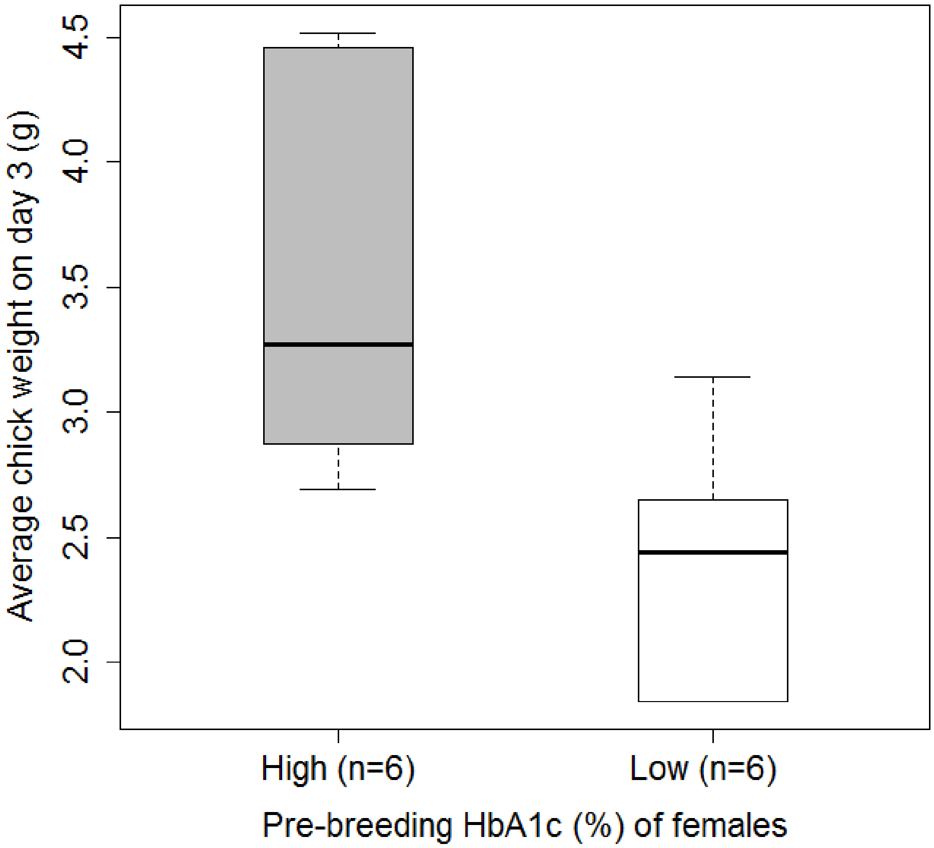

Figure 5. A Mann–Whitney U test boxplot of the average chick weight on day 3 after hatching in different groups of pre–breeding HbA1c level in female Varied Tits

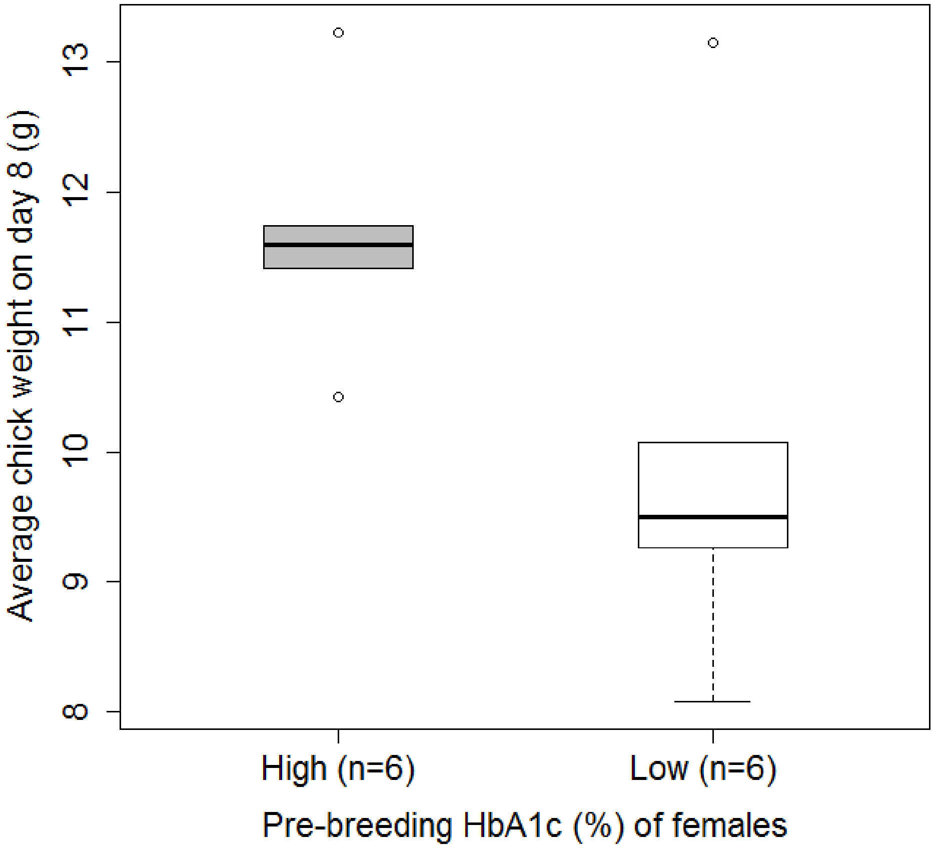

Figure 6. A Mann−Whitney U test boxplot of the average chick weight on day 8 after hatching in different groups of pre−breeding HbA1c level in female Varied Tits

Table 3. HbA1c of breeding females and breeding performance in Varied Tits

	HbA1c	Median	Range	U	p
Clutch size	high (n=13)	7	5−10	36.5	0.064
	low (n=10)	6	5−7		
Laying date	high (n=13)	118	105−141	54.5	0.534
	low (n=10)	118	110−152		
Breeding success	high (n=13)	85.7	30.0−100.0	53.0	0.461
	low (n=10)	92.9	33.3−100.0		
Tarsus on day 3	high (n=13)	6.55	5.46−7.97	48.0	0.313
	low (n=10)	6.69	5.95−8.34		
Tarsus on day 8	high (n=13)	14.74	11.96−16.66	56.0	0.605
	low (n=10)	14.84	13.64−16.87		
Tarsus on day 13	high (n=13)	18.25	16.83−18.98	56.5	0.648
	low (n=10)	18.54	17.87−18.80		
Weight on day 3	high (n=13)	2.65	1.84−4.29	34.5	0.057
	low (n=10)	3.08	2.37−4.52		
Weight on day 8	high (n=13)	10.48	8.08−12.71	47.0	0.284
	low (n=10)	10.69	9.38−13.15		
Weight on day 13	high (n=13)	15.53	11.11−16.86	55.0	0.563
	low (n=10)	15.78	12.85−17.23		
Tarsus growth between day 3−8	high (n=13)	1.24	0.98−1.63	63.0	0.976
	low (n=10)	1.18	0.83−1.55		
Tarsus growth between day 8−13	high (n=13)	0.25	0.12−0.50	58.5	0.780
	low (n=10)	0.25	0.12−0.33		
Tarsus growth between day 3−13	high (n=13)	1.87	1.33−2.28	56.0	0.605
	low (n=10)	1.81	1.15−2.18		
Weight growth between day 3−8	high (n=13)	3.11	2.02−3.72	43.5	0.186
	low (n=10)	2.67	1.52−3.45		
Weight growth between day 8−13	high (n=13)	0.46	(−0.07)−1.24	51.0	0.410
	low (n=10)	0.43	0.19−0.64		
Weight growth between day 3−13	high (n=13)	5.08	2.50−8.60	41.0	0.148
	low (n=10)	4.27	2.49−6.38		

Table 4. HbA1c of breeding males and breeding performance in Varied Tits

	HbA1c	Median	Range	U	p
Clutch size	high (n=16)	6	4−9	40.0	0.271
	low (n=7)	6	5−7		
Laying date	high (n=16)	117	105−153	41.5	0.347
	low (n=7)	115	106−120		
Breeding success	high (n=16)	84.5	33.3−100.0	53.5	0.892
	low (n=7)	83.3	20.0−100.0		
Tarsus on day 3	high (n=16)	6.78	5.16−7.97	52.0	0.820
	low (n=7)	6.63	6.09−8.34		
Tarsus on day 8	high (n=16)	15.03	11.96−16.66	52.0	0.820
	low (n=7)	14.44	12.84−16.87		
Tarsus on day 13	high (n=16)	18.49	16.83−19.19	44.0	0.452
	low (n=7)	18.06	16.18−18.80		
Weight on day 3	high (n=16)	2.98	1.84−4.46	47.0	0.579
	low (n=7)	3.14	2.37−4.52		
Weight on day 8	high (n=16)	1.85	8.08−13.23	53.0	0.871
	low (n=7)	11.22	9.44−13.15		
Weight on day 13	high (n=16)	15.76	11.11−17.23	46.0	0.535
	low (n=7)	15.59	13.67−15.99		
Tarsus growth between day 3−8	high (n=16)	1.21	0.90−1.56	55.5	1.000
	low (n=7)	1.32	0.83−1.55		
Tarsus growth between day 8−13	high (n=16)	0.24	0.12−0.42	49.0	0.664
	low (n=7)	0.25	0.12−0.33		
Tarsus growth between day 3−13	high (n=16)	1.77	1.33−2.28	54.0	0.922
	low (n=7)	1.78	1.15−1.99		
Weight growth between day 3−8	high (n=16)	2.63	2.02−4.08	53.0	0.871
	low (n=7)	2.71	1.53−3.35		
Weight growth between day 8−13	high (n=16)	0.44	(−0.07)−0.78	52.0	0.820
	low (n=7)	0.44	0.19−0.62		
Weight growth between day 3−13	high (n=16)	4.20	2.50−7.21	50.5	0.738
	low (n=7)	4.05	2.48−5.49		

4.3 BCI, laying date and breeding performance

Analyses using generalized linear mixed models showed some correlation between the BCI measured at the end of the breeding period, laying date, and chick condition.

In females, the BCI was correlated to the average tarsus length of chicks on day 13 after hatching (χ^2=3.841, p=0.050), shortening it by about 0.085±0.130 mm. Except for the weight growth rate between day 8 and 13 showing positive marginal insignificance (χ^2=3.750, p=0.053), the breeding success and other measurements of chicks did not have significant correlations to the BCI of females (Table 5).

On the other hand, in males, the BCI was correlated to the growth rate of early-mid stage chick brooding. Average chick tarsus growth (χ^2=5.874, p=0.015) and weight growth (χ^2=4.270, p=0.039) between day 3 and 8 after hatching were significantly related to the BCI of fathers, increasing by 0.175±0.068 and 0.445±0.207 respectively (Table 6).

The laying date of females also influenced the chick tarsus length and chick growth (Table 7). Females that laid eggs earlier had chicks with longer tarsi (χ^2=4.054, p=0.044) on day 3 after hatching, estimated by 0.004±0.012 mm. In contrast, when the laying date was earlier, average chick tarsus growth (χ^2=5.298, p=0.021) and weight growth (χ^2=5.030, p=0.025) between days 3-13, and average chick weight growth between days 3-8 (χ^2=4.239, p=0.040) after hatching were lower, by 0.003±0.004, 0.038±0.022, and 0.009±0.010, respectively.

Table 5. Generalized linear mixed models (GLMMs) on breeding performance and BCI in breeding females. Significant relationships are marked in bold.

	Estimate	Std. error	χ^2	p
Breeding success	−9.678	5.870	0.622	0.431
Tarsus on day 3	−0.485	0.229	1.233	0.267
Tarsus on day 8	−0.213	0.375	0.175	0.676
Tarsus on day 13	−0.085	0.130	3.841	**0.050**
Weight on day 3	−0.325	0.229	0.951	0.329
Weight on day 8	−0.082	0.387	0.637	0.425
Weight on day 13	−0.888	0.421	2.476	0.116
Tarsus growth between day 3−8	0.116	0.060	0.010	0.922
Tarsus growth between day 8−13	0.019	0.030	1.210	0.271
Tarsus growth between day 3−13	0.197	0.078	0.454	0.500
Weight growth between day 3−8	0.396	0.185	0.907	0.341
Weight growth between day 8−13	0.124	0.070	3.750	0.053
Weight growth between day 3−13	1.042	0.431	2.730	0.098

Table 6. Generalized linear mixed models (GLMMs) of breeding performance and BCI in breeding males. Significant relationships are marked in bold.

	Estimate	Std. error	χ^2	p
Breeding success	−4.051	6.172	0.428	0.513
Tarsus on day 3	−0.415	0.245	2.655	0.103
Tarsus on day 8	0.359	0.353	0.983	0.322
Tarsus on day 13	0.172	0.156	0.973	0.324
Weight on day 3	−0.265	0.239	1.204	0.273
Weight on day 8	0.712	0.350	3.693	0.055
Weight on day 13	0.086	0.411	0.044	0.835
Tarsus growth between day 3−8	0.175	0.068	5.874	**0.015**
Tarsus growth between day 8−13	−0.021	0.027	0.575	0.448
Tarsus growth between day 3−13	0.155	0.091	2.759	0.097
Weight growth between day 3−8	0.445	0.207	4.270	**0.039**
Weight growth between day 8−13	−0.068	0.056	1.240	0.265
Weight growth between day 3−13	0.379	0.388	0.921	0.337

Table 7. Generalized linear mixed models (GLMMs) of breeding performance and laying date in breeding females. Significant relationships are marked in bold.

	Estimate	Std. Error	χ^2	p
Breeding success	−0.261	0.314	2.556	0.110
Tarsus on day 3	−0.004	0.012	4.054	**0.044**
Tarsus on day 8	−0.008	0.019	0.318	0.573
Tarsus on day 13	0.017	0.007	0.416	0.519
Weight on day 3	−0.011	0.012	1.933	0.164
Weight on day 8	−0.016	0.019	0.044	0.833
Weight on day 13	0.037	0.022	3.541	0.060
Tarsus growth between day 3−8	0.000	0.003	3.536	0.060
Tarsus growth between day 8−13	0.002	0.002	0.335	0.563
Tarsus growth between day 3−13	0.003	0.004	5.298	**0.021**
Weight growth between day 3−8	0.009	0.010	4.239	**0.040**
Weight growth between day 8−13	0.007	0.004	2.700	0.100
Weight growth between day 3−13	0.038	0.022	5.030	**0.025**

4.4 HbA1c and BCI

When analyzed using the Mann−Whitney U test, no significant relationship between the percentage level of HbA1c and the BCI was found. None of the breeding females, breeding males and pre−breeding females showed differences in the BCI between groups of the high and low HbA1c level (Table 8).

Table 8. Body Condition Index (BCI) of breeding females, breeding males, and pre−breeding females in two different HbA1c groups

Groups	HbA1c	Median	Range	U	p
Breeding females	high (n=13)	−0.030	(−0.760)−0.850	58.0	0.687
	low (n=10)	−0.121	(−1.701)−1.279		
Breeding males	high (n=16)	−0.108	(−0.924)−1.321	53.0	0.871
	low (n=7)	0.118	(−0.600)−0.953		
Pre−breeding females	high (n=6)	−0.187	(−1.152)−0.202	12.0	0.662
	low (n=6)	0.069	(−1.270)−0.971		

5. Discussion

In tits as well as other birds, the condition of parents is known to have a significant impact on breeding. But the relationship between the body condition of parents and their breeding performance differ among species or individuals, and also depending on environmental conditions. Each individual shows distinct breeding behavior, makes different decisions and invests different amounts of energy in breeding. In this study, the relationship between the body condition of Varied Tits and their breeding performance was examined, along with how they are correlated.

The pre-breeding condition of females, which was measured by the percentage level of HbA1c, had an influence on chick weight during the early and middle stages of breeding. Females with higher levels of HbA1c had heavier chicks on day 3 and 8 after hatching. The relationship between the pre-breeding condition and breeding performance of parents has been studied in other species, where Collared Flycatchers (*Ficedula albicollis*) of better conditions had larger clutches (Hargitai et al. 2014) and the energy reserve of Blue Tits (*Cyanistes caeruleus*) during the winter led to higher breeding success (Robb et al. 2008). Similarly, in Varied Tits, higher energy reserve before breeding would have made it possible for them to deliver more energy during the early stages of chick rearing. Although the HbA1c level of females were significantly correlated to only two variables in this particular study, but not other variables including the breeding success, nutritional status of the pre-breeding females seems to influence the condition of the chicks. As blood was sampled before the breeding period when food had been scarce over winter, higher levels of HbA1c could possibly mean better foraging ability during that period. As Varied Tits feed their chicks at a very high frequency (more than six times per hour) when chicks' food demand is at its peak (Lee et al. 2011, observation in survey), this ability could have acted as a crucial element in raising heavier chicks.

HbA1c during the breeding period, however, did not show significant correlation with any of the breeding performance variables. While Varied Tits consume most of the food they have found for themselves before breeding, much of the foraging behavior is performed so as to provide food for the chicks. Even when two birds ingested exactly the same amount of prey, it is unclear whether they have done the same amount of provisioning, because each individual is known to invest different amounts of energy in its chicks (Linden 1988, Ots and Horak 1996). For this reason, HbA1c level measured on day 13 after hatching does not seem to be a good predictor of chick condition nor breeding success.

The BCI of breeding females and males, measured at the end of the breeding period, was correlated to some of breeding performance variables but showed different patterns between sexes.

In females, the BCI was negatively correlated to the average tarsus length of chicks on day 13. This may be the result of excessive parental care leading to a decrease in the body condition of mothers, but larger chicks right before fledging. In tits, females tend to invest more in breeding than males do (Sanz and Tinbergen 1999, Visser and Lessells 2001, Dubiec 2011), which can explain the negative relationship. A chick's condition at fledging is known to determine its future survival (Naef-Daenzer and Keller 1999) and resource holding potential (Yamaguchi et al. 2004), and raising larger chicks could have been the strategy of female tits for successful reproduction. Weight growth rate of chicks between day 8 and 13, on the other hand, showed the opposite result where chicks of females with a higher BCI grew relatively faster. Although it is clear that a female's body condition during the breeding period is a crucial factor that determines the body condition of chicks, in which way the body condition of females is related to that of their chicks is inconclusive from the results of this study.

Males, on the other hand, showed positive correlation between the BCI and chick growth rate. As only females are responsible for egg-laying and incubation, the body condition of males is less affected by the breeding process. In Great Tits, one of the main roles of males in reproduction is to defend their territories (Ydenberg 1984, Lambrechts and Dhondt 1988). As it is thought to be similar in Varied Tits and since a male's condition is related to its resource holding potential (Yamaguchi et al. 2004), males of better condition may have defended their territories better, or had superiority in foraging and fed their chicks more effectively in cooperation with their partners.

The laying date was significantly related to the tarsus length of chicks on day 3 and the growth rate of tarsus and weight. In tits, an early start of breeding is commonly thought to be beneficial in regard to food supply (Perrins 1991, Van Noordwijk et al. 1995), and this seems to explain larger chicks on day 3 of early breeders. Weight growth between day 3-8 along with tarsus and weight growth between day 3-13, on the other hand, showed reversed results where chicks of late breeders grew faster. As late breeders had relatively smaller chicks, they would have fed the chicks intensively to catch up with their early competitors. Compared to Great Tits, the Varied Tit seems to have higher fitness for minimizing the effect of the laying date on the breeding outcome, and this could partially be because of its smaller clutch size, implying that Varied Tits have extra energy to invest in each chick.

In terms of HbA1c, no relationship between HbA1c and the BCI of breeding Varied Tits was found in this study. One of the possible reasons behind it could be that the two might indicate different perspectives of body condition. While HbA1c reflects the average glucose level in blood of 3–5 weeks (Andersson and Gustafsson 1995), thus the average energy intake of parents from egg laying or incubation period to day 13, the BCI rather represents body condition at capture, although it is also a result of the entire breeding performance. There might have been a certain level of correlation between HbA1c and the BCI if pre-breeding measurements had been compared, with considerable influence of breeding removed. In any manner, the mechanism of glycation in breeding birds still needs to be studied in depth for further studies to use HbA1c as an indicator of avian body condition.

Additionally, while many studies of other tit species have discovered that breeding patterns differ as the elevation changes, in this study, no significant difference in breeding patterns of Varied Tits was observed among the four sites ranging from 400 to 1,000 m a. s. l. Contrary to many previous studies in birds, where smaller clutch sizes were observed at higher altitudes (Fargallo 2004, Dillon and Conway 2015), no significant difference in clutch size nor the body condition of both parents and chicks was found in this study. This is thought to be the result of relatively consistent environmental conditions, as altitudinal variation was relatively small (400 to 1,000 m a. s. l.) and vegetation showed similar composition among study sites.

6. Conclusion

The relationship between the body condition, measured by the BCI and the percentage level of HbA1c, and breeding performance of Varied Tits showed different patterns among indicators, times of measurement, and sexes.

Higher levels of HbA1c in pre-breeding females showed to have a positive relationship with the weight of chicks in early and mid-stages. The HbA1c level before breeding may reflect the foraging ability of individuals when food is scarce, and thus be related to better provisioning for chicks when they are in need of high energy. HbA1c during the breeding period on the other hand, was not significantly correlated to any variable of breeding. One of the possible explanations could be that the energy intake of a parent does not necessarily predict the amount of food provided to chicks.

The BCI was measured using tarsus length and weight of breeding Varied Tits. Females with poor BCI figures ended up with larger chicks in the end, although their chicks did not show any difference in condition at earlier stages nor in growth rate when compared to the chicks of females in better body condition. Since the condition of chicks at fledging determines their survival and future breeding success, this seems like a strategy of females for successful breeding in exchange for their fitness, by investing excessive amounts of energy. The laying date showed a similar pattern where early nests had larger chicks on day 3 but late chicks showed higher growth rates, probably due to the excessive investment of late mothers. Males showed opposite results, where a higher BCI led to rapid growth in early to mid-stages of chicks.

Finally, no relationship between HbA1c and the BCI of the breeding period was found. With considerable influence of energy consumed in breeding, the mechanism of glycation still needs to be studied further so as to devise it as a better indicator.

This study suggests that HbA1c may represent an individual's body condition before the breeding period and predict its breeding performance. However, the applicability of HbA1c as an index of body condition throughout the breeding period is uncertain because the HbA1c level while rearing is influenced by the provision for chicks and consequently likely to change over time. During the breeding period, therefore, conventional indicators such as the BCI are thought to better reflect an individual's condition.

References

Andersson, M. S., and L. Gustafsson. 1995. Glycosylated haemoglobin: A new measure of condition in birds. Proceedings of the Royal Society B: Biological Sciences **260**:299–303.

Ardia, D. R. 2006. Glycated hemoglobin and albumin reflect nestling growth and condition in American kestrels. Comparative Biochemistry and Physiology – A Molecular and Integrative Physiology **143**:62–66.

Bailly, J., R. Scheifler, M. Belvalette, S. Garnier, E. Boissier, V. A. Clément–Demange, M. Gète, M. Leblond, B. Pasteur, Q. Piget, M. Sage, and B. Faivre. 2016. Negative impact of urban habitat on immunity in the Great Tit *Parus major*. Oecologia **182**:1053–1062.

Bates, D. M., M. Maechler, and B. Bolker. 2012. lme4: Linear mixed–effects models using S4 classes. R package version 0.999999–0.

De Laet, J. F., and A. A. Dhondt. 1989. Weight loss of the female during the first brood as a factor influencing second brood initiation in Great Tits Parus major and blue tits *P. caeruleus*. Ibis **131**:281–289.

Dillon, K. G., and C. J. Conway. 2015. Elevational gradient in clutch size of Red–faced Warblers. Journal of Field Ornithology **86**:163–172.

Dubiec, A. 2011. Condition–dependent clutch desertion in Great Tit (*Parus major*) females subjected to human disturbance. Journal of Ornithology **152**:743–749.

Dubiec, A., and M. Cichoñ. 2001. Seasonal decline in health status of Great Tit (*Parus major*) nestlings. Canadian Journal of Zoology **79**:1829–1833.

Dufva, R. 1996. Blood parasites, health, reproductive success, and egg volume in female Great Tits *Parus major*. Journal of Avian Biology **27**:83–87.

Fargallo, J. A. 2004. Latitudinal trends of reproductive traits in the Blue

Tit *Parus caeruleus*. Ardeola **51**:177–190.

Garnett, M. C. 1981. Body size, its heritability and influence on juvenile survival among Great Tits, *Parus major* (Wytham Woods, Oxford). Ibis **123**:31–41.

Gill, F., and D. Donsker. 2016. IOC World Bird List (v 6.4). Doi 10.14344/IOC.ML.6.4. http://www.worldbirdnames.org/.

Guindre–Parker, S., H. G. Gilchrist, S. Baldo, and O. P. Love. 2013. Alula size signals male condition and predicts reproductive performance in an Arctic–breeding passerine. Journal of Avian Biology **44**:209–215.

Hõrak, P., S. Jenni–Eiermann, and I. Ots. 1999. Do Great Tits (*Parus major*) starve to reproduce? Oecologia **119**:293–299.

Hargitai, R., G. Hegyi, M. Herényi, M. Laczi, G. Nagy, B. Rosivall, E. Szöllosi, and J. Török. 2014. Winter body condition in the Collared Flycatcher: Determinants and carryover effects on future breeding parameters. Auk **131**:257–264.

Hinks, A. E., E. F. Cole, K. J. Daniels, T. A. Wilkin, S. Nakagawa, and B. C. Sheldon. 2015. Scale–dependent phenological synchrony between songbirds and their caterpillar food source. American Naturalist **186**:84–97.

Johnson, D. H., G. L. Krapu, K. H. Reinecke, and D. G. Jorde. 1985. An evaluation of condition indices for birds. Journal of Wildlife Management **49**:569–575.

Ju, J., J. Yin, P. Racey, L. Zhang, D. Li, and D. Wan. 2014. Extra–pair paternity in Varied Tits *Poecile varius*. Acta Ornithologica **49**:131–136.

Kaliński, A., M. Bańbura, M. Gladalski, M. Markowski, J. Skwarska, J. Wawrzyniak, P. Zieliński, I. Cyzewska, and J. Bańbura. 2015. Long–term variation in hemoglobin concentration in nestling Great Tits *Parus major*. Comparative Biochemistry and Physiology – Part A : Molecular and Integrative Physiology **185**:9–15.

Kilgas, P., R. Mänd, M. Mägi, and V. Tilgar. 2006. Hematological parameters in brood-rearing Great Tits in relation to habitat, multiple breeding and sex. Comparative Biochemistry and Physiology - A Molecular and Integrative Physiology **144**:224-231.

Kubota, H., and M. Nakamura. 2000. Effects of supplemental food on intra- and inter-specific behaviour of the Varied Tit *Parus varius*. Ibis **142**:312-319.

Labocha, M. K., and J. P. Hayes. 2012. Morphometric indices of body condition in birds: A review. Journal of Ornithology **153**:1-22.

Lack, D. 1947. The significance of clutch size. Ibis **89**:302-352.

Lambrechts, M., and A. A. Dhondt. 1988. Male quality and territory quality in the Great Tit *Parus major*. Animal Behaviour **36**:596-601.

Lee, J. K., O. S. Chung, and W. S. Lee. 2011. Altitudinal variation in parental provisioning of nestling Varied Tits (*Poecile varius*). Wilson Journal of Ornithology **123**:283-288.

Lee, J. K., W. Jang, O. S. Chung, and W. S. Lee. 2016. The relationships between prey size, nestling age, provisioning rate, and elevation in the Varied Tit *Parus varius*. Ornithological Science **15**:29-36.

Lee, W. S., T. H. Koo, and J. Y. Park. 2000. A field guide to the birds of Korea. LG Evergreen Foundation. Seoul, Korea.

Linden, M. 1988. Reproductive trade-off between first and second clutches in the Great Tit *Parus major*: an experimental study. Oikos **51**:285-290.

Ma, R. Q., P. Chang, D. M. Wan, J. Ju, L. Zhang, and D. L. Li. 2015. Extra-pair paternity is unrelated to the spouse’s genetic compatibility in Varied Tit (*Parus varius*). Acta Ecologica Sinica **35**:5019-5025.

Mikšik, I., and Z. Hodný. 1992. Glycated hemoglobin in mute swan (*Cygnus olor*) and rook (*Corvus frugilegus*). Comparative Biochemistry and Physiology - Part B: Biochemistry and **103**:553-555.

Naef-Daenzer, B., and L. F. Keller. 1999. The foraging performance of great and blue tits (*Parus major* and *P. caeruleus*) in relation to caterpillar development, and its consequences for nestling growth and fledging weight. Journal of Animal Ecology **68**:708–718.

Naef–Daenzer, B., F. Widmer, and M. Nuber. 2001. Differential post–fledging survival of great and coal tits in relation to their condition and fledging date. Journal of Animal Ecology **70**:730–738.

Nakamura, M. K. H. 1998. Food supply in early spring and stability of pair bonds in the Varied Tit *Parus varius*. Journal of Avian Biology **29**:201.

Nilsson, J. A., and E. Svensson. 1993. Energy constraints and ultimate decisions during egg–laying in the blue tit. Ecology **74**:244–251.

Norte, A. C., J. A. Ramos, H. L. Sampaio, J. P. Sousa, and B. C. Sheldon. 2010. Physiological condition and breeding performance of the Great Tit. Condor **112**:79–86.

Ots, I., and P. Horak. 1996. Great Tits Parus major trade health for reproduction. Proceedings of the Royal Society B: Biological Sciences **263**:1443–1447.

Ots, I., A. Murumägi, and P. Hõrak. 1998. Haematological health state indices of reproducing Great Tits: Methodology and sources of natural variation. Functional Ecology **12**:700–707.

Owen, J. C. 2011. Collecting, processing, and storing avian blood: A review. Journal of Field Ornithology **82**:339–354.

Perrins, C. M. 1991. Tits and their caterpillar food supply. Ibis **133**:49–54.

Perrins, C. M., and R. H. McCleery. 1989. Laying dates and clutch–size in the Great Tit. Wilson Bull **101**:236–253.

Ralph, C. J., G. R. Geupel, P. Pyle, T. E. Martin, and D. F. DeSante. 1993. Handbook of field methods for monitoring landbirds. General Technical Report. U.S. Department of Agriculture, Forest Service, Pacific Southwest Research Station.

Rhim, S. J., S. H. Son, and K. J. Kim. 2011. Breeding ecology of tits *Parus* spp. using artificial nest boxes in a coniferous forest over a five–

year period. Forest Science and Technology **7**:141−144.

Robb, G. N., R. A. McDonald, D. E. Chamberlain, S. J. Reynolds, T. J. E. Harrison, and S. Bearhop. 2008. Winter feeding of birds increases productivity in the subsequent breeding season. Biology Letters **4**:220−223.

Rooyen, J. V., F. Lalubin, O. Glaizot, and P. Christe. 2013. Altitudinal variation in haemosporidian parasite distribution in Great Tit populations. Parasites and Vectors **6**:139.

Sanz, J. J., and J. M. Tinbergen. 1999. Energy expenditure, nestling age, and brood size: An experimental study of parental behavior in the Great Tit *Parus major*. Behavioral Ecology **10**:598−606.

Slagsvold, T., and J. T. Lifjeld. 1990. Influence of male and female quality on clutch size in tits (*Parus* spp.). Ecology **71**:1258−1266.

Smith, H. G., H. Kallander, and J. A. Nilsson. 1989. The trade−off between offspring number and quality in the Great Tit *Parus major*. Journal of Animal Ecology **58**:383−401.

Tinbergen, J. M., and M. C. Boerlijst. 1990. Nestling weight and survival in individual Great Tits (*Parus major*). Journal of Animal Ecology **59**:1113−1127.

Van Noordwijk, A. J., R. H. McCleery, and C. M. Perrins. 1995. Selection for the timing of Great Tit breeding in relation to caterpillar growth and temperature. Journal of Animal Ecology **64**:451−458.

Verhulst, S. 1998. Multiple breeding in the Great Tit, II. The costs of rearing a second clutch. Functional Ecology **12**:132−140.

Verhulst, S., and J. M. Tinbergen. 1991. Experimental evidence for a causal relationship between timing and success of reproduction in the Great Tit *Parus m. major*. Journal of Animal Ecology **60**:269−282.

Visser, M. E., and C. M. Lessells. 2001. The costs of egg production and incubation in Great Tits (*Parus major*). Proceedings of the Royal Society B: Biological Sciences **268**:1271−1277.

Yamaguchi, N. 2005. Cheek-patch coloration varies greatly within a subspecies of the Varied Tit *Parus varius*. Ibis **147**:836-840.

Yamaguchi, N., and H. Higuchi. 2005. Extremely low nesting success and characteristics of life history traits in an insular population of *Parus varius* namiyei. Wilson Bulletin **117**:189-193.

Yamaguchi, N., and K. K. Kawano. 2001. Effect of body size on the resource holding potential of male Varied Tits *Parus varius*. Japanese Journal of Ornithology **50**:65-70.

Yamaguchi, N., K. K. Kawano, K. Eguchi, and T. Yahara. 2004. Facultative sex ratio adjustment in response to male tarsus length in the Varied Tit *Parus varius*. Ibis **146**:108-113.

Ydenberg, R. C. 1984. The conflict between feeding and territorial defence in the Great Tit. Behavioral Ecology and Sociobiology **15**:103-108.

Appendix

Appendix 1

Birds observed at four study sites in June, 2016 (Dosolbong and Hanjae at 800 m: 27th, 1,000 m: 28th, 400 m: 30th)

Common name	Scientific name	1000 m	800 m		400 m
			Dosolbong	Hanjae	
Blue−and−white Flycatcher	*Cyanoptila cyanomelana*				1
Brown−eared Bulbul	*Hypsipetes amaurotis*			1	6
Coal Tit	*Periparus ater*	3		1	
Daurian Redstart	*Phoenicurus auroreus*				1
Eurasian Jay	*Garrulus glandarius*		1	1	
Eurasian Nuthatch	*Sitta europaea*	1	4		
Great Spotted Woodpecker	*Dendrocopos major*		1		
Great Tit	*Parus major*	4	1	1	3
Hazel Grouse	*Tetrastes bonasia*	2		2	
Large−billed Crow	*Corvus macrorhynchos*		2	4	
Lesser Cuckoo	*Cuculus poliocephalus*	2			
Long−tailed Tit	*Aegithalos caudatus*	1			
Marsh Tit	*Poecile palustris*	2			2
Oriental Cuckoo	*Cuculus optatus*			1	
Oriental Turtle Dove	*Streptopelia orientalis*		4	1	
Pale Thrush	*Turdus pallidus*	6	5	7	4
Siberian Blue Robin	*Lavivora cyane*		1		
Varied Tit	*Sittiparus varius*	8	11	3	5
White's Thrush	*Zoothera aurea*				1
Yellow−throated Bunting	*Emberiza elegans*	6	6	2	1
Number of species		10	10	11	9
Number of individuals		35	36	24	24

국문 초록

조류의 번식은 많은 에너지가 소모되는 과정이다. 다른 여러 종의 조류와 같이, 박새류의 건강 상태는 번식의 여러 단계에 걸쳐 영향을 미치는 것으로 알려져 있다. 이러한 개체의 건강 상태와 번식의 관계가 곤줄박이(*Sittiparus varius*)에서는 어떻게 나타나는지 알아보기 위해 광양 백운산에서 인공새집을 이용한 연구를 실시하였다. 부모의 개체 건강 상태는 2016년 번식기에 건강상태지표(Body Condition Index, BCI)와 당화혈색소(HbA1c)를 이용하여 번식 전과 번식 후기, 두 번에 걸쳐 측정하였다. 번식의 지표로는 산란일, 한배산란수, 번식성공률, 유조의 건강 상태와 성장률을 측정하였다.

번식 전 암컷의 당화혈색소 수치는 유조의 체중과 양의 상관관계를 보였는데, 이는 육추를 위한 더 많은 에너지 비축량과 더 나은 먹이 활동 능력의 결과인 것으로 생각된다. 번식 후기 부모의 BCI와 번식의 관계는 암컷과 수컷이 다른 양상을 보였으며, 이는 번식 전략의 성별 차이에 의한 것으로 생각된다. 암컷의 BCI는 번식 후기 유조의 건강 상태와 음의 상관관계를 보인 반면, 수컷의 BCI 값이 클수록 유조의 성장이 빠르게 나타났다. 암컷 BCI의 감소가 유조에 대한 직접적인 에너지 투자를 반영하는 반면, 수컷의 BCI는 유조의 성장률에 영향을 미치는 자원 보유 능력을 반영하는 것으로 생각된다. 번식을 일찍 시작한 개체들에게서 유조의 초기 크기가 크게 나타났지만 이후에는 암컷의 건강상태에 따른

둥지 간 차이가 나타나지 않았는데, 이는 번식을 늦게 시작한 개체들이 추가적으로 육추 에너지를 투자한 결과로 생각된다. 당화혈색소와 BCI 사이에는 유의한 상관관계를 보이지 않았고, 번식 후기에 측정한 부모의 당화혈색소 또한 번식 지표와 유의한 관계가 없는 것으로 나타났다.

본 연구에서 곤줄박이의 건강 상태는 번식의 지표들과 여러 상관관계를 보였다. 당화혈색소의 경우 번식 전이나 초기 조류의 건강 상태를 대표하는 지표로는 사용될 수 있지만, 번식 중에는 BCI와 같은 기존의 지표들이 부모의 건강 상태를 대표하고 번식의 과정을 반영하기에 더 적합한 지표인 것으로 생각된다.

주요어: 건강 상태, 건강상태지표, 곤줄박이, 당화혈색소, 번식 결과

학번: 2015-21502