

Seasonality in the feeding ecology of Black-necked swans (*Cygnus melancoryphus*) in a temperate wetland of southern Chile

La estacionalidad en la ecología nutricional de Cisnes de Cuello Negro (*Cygnus melancoryphus*) de un humedal templado del sur de Chile

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ABSTRACT

The concomitant effects of environmental physical constraints associated with the availability and quality of food will finally affect the energy budget of wild animals. This study aimed to determine the effect of seasonality on water level, behavioural and nutritional status of Black-necked swans inhabit the Budi Lake, Chile. In winter and spring, the water level was recorded using a graduated rod and a time budget of 60 swans/hours was recorded from 8 am to 6 pm using binoculars and the focal method. In order to determine nutritional status a subset of 20 birds were captured and weight, length, wingspan, and tarsal length were measured. Also, blood sample was collected to determine cholesterol, triglycerides, and total proteins plasma concentrations. Budi Lake's water level increase in winter (111 ± 3 cm vs 54 ± 0.6 cm, $P = 0.04$). In addition, winter swan population increased significantly the time allocated to foraging at morning, and the proportion of effortful foraging behaviour ($37/222$ vs $14/185$, $P = 0.01$), assigning less time to resting activities (7 ± 4 % vs 11 ± 2 %, $P = 0.05$) compared with spring population. Nutritional status was not compromised in swan populations in both seasons. In conclusion, behavioral adjustments allow Black-necked swan populations to maintain an adequate nutritional status despite the increase in water level during winter season.

Keywords: black-necked swans, *Cygnus melancoryphus*, nutritional status, seasonality.

RESUMEN

El efecto de las restricciones físico-ambientales y de la disponibilidad y calidad del alimento determinan finalmente el presupuesto de energía de los animales silvestres. Este estudio tuvo como objetivo determinar el efecto de la estacionalidad sobre el nivel del agua, el comportamiento y el estado nutricional de los Cisnes de Cuello Negro que habitan el Lago Budi, Chile. En invierno y en primavera se registró el nivel del agua con una varilla graduada y se obtuvo un presupuesto de tiempo de 60 cisnes/hora desde las 8 am hasta las 6 pm utilizando binoculares y el método focal. Con el fin de determinar el estado nutricional, se capturó un subconjunto de 20 aves y se midió el peso, la longitud total, la envergadura y la longitud del tarso. Además, se obtuvo una muestra de sangre para determinar las concentraciones plasmáticas de colesterol, triglicéridos y proteínas totales. El nivel del agua del Lago Budi aumentó significativamente en invierno (111 ± 3 cm vs $54 \pm 0,6$ cm, $P = 0,04$). Además, la población de cisnes de invierno incrementó el tiempo asignado a la búsqueda de alimento en la mañana y la proporción de alimentación con esfuerzo

(37/222 vs 14/185, $P = 0,01$), asignando menos tiempo a la actividad de descanso ($7 \pm 4\%$ vs $11 \pm 2\%$, $P = 0,05$) en comparación a la población de primavera. El estado nutricional no se vio comprometido en las poblaciones de cisnes de ambas estaciones. En conclusión, los ajustes conductuales permitieron a los cisnes de cuello negro mantener un estado nutricional adecuado a pesar del incremento de los niveles de agua en la estación de invierno.

Palabras claves: cisnes de cuello negro, *Cygnus melancoryphus*, estacionalidad, estado nutricional.

INTRODUCTION

The processes of energy acquisition and expenditure play a central role in shaping the physiology, behaviour, and ecology of organisms (Bozinovic & Martínez del Río 2000; López-Calleja *et al.* 2003). The concomitant effects of environmental physical constraints associated with the availability and quality of food will finally affect the structure of the animal's energy budget. This is particularly true for large nomadic birds inhabiting contrasting habitats where they must cope with different environmental temperatures and food unpredictability (McKechnie & Wolf 2004; Nolet *et al.* 2002; La Montagne *et al.* 2004).

Water sustains food for waterbirds (e.g., submerged plants, fish, and insects), and its quantity and quality primarily determine the state of the marsh vegetation (Euliss *et al.* 2004). Fluctuation in water level is a major factor that influences the relative abundance and distribution of waterbirds (Nolet *et al.* 2016, Rajpar & Zaparia 2011), affecting foraging behaviour through changes in patch use (Cursach *et al.* 2015), habitat selection (Clausen 2000, Wood *et al.* 2013), diet selection (Aharon-Rotman *et al.* 2017), and foraging strategies (Nolet *et al.* 2002, 2006).

The Black-necked swan (*Cygnus melancoryphus*) is a large herbivore waterbird (3-8 kg) endemic of South America. The population inhabits wetlands in the south cone ranging from subtropical southern Brazil to Tierra del Fuego, in Chile and Argentina (Schlatter 1998, 2005; Schlatter *et al.* 2002). In Chile, the Black-necked swan's diets include the aquatic plants *Egeria densa*, *Potamogeton pucillus*, *Myriophyllum aquaticum*, *Potamogeton lucens*, *Schoenoplectus californicus*, *Limnium laevigatum*, deposits of roots of death plants of *Ludwigia peploides* in rivers, and *Potamogeton striatus*, *Puccinellia glaucescens*, *Cotula coronopifolia* and *Sarcornia fruticosa* in Lakes (Corti & Schlatter 2002; Schlatter *et al.* 2002; Norambuena & Bonazinovic 2009a, Velasquez *et al.* 2019). Also, the algae *Ulva lactuca*, *Distichlis spicata*, *Gracillaria*

chilensis and *Gigartina skottsbergii* has been describes as the diet of swans in marine environments (Cursach *et al.* 2015). The diversity of their diets probably favors the energy homeostasis. Nevertheless, its high body weight, and the herbivore status suggest that energy balance could represent a great challenge for this species (Maurer 1996; Magnione & Bozinovic 2003). It's has been described that the basal metabolic rate (BMR) of Anatidae members escalates to a high degree with the body live weight (BLW) ($BMR = BLW^{0.74}$) (Miller & Eady 2006). Indeed, the average of BMR of Black-necked swan ($0.5 \text{ ml} \pm 0.07 \text{ ml O}_2 \text{ h}^{-1}\text{g}^{-1}$) was 110% from the allometrically predicted (Norambuena 2008). On the other hand, the high amount of cellulose and fiber content in the diet of swans negatively affect the digestibility and the nutrient assimilation (Corti & Schlatter 2002, Velasquez *et al.* 2019).

Black-necked swans undergo opportunistic displacements among different wetland areas occurring near the Pacific coast of Chile (Schlatter *et al.* 2002; Vilina *et al.* 2002). Although the factors that regulate their movements are not clear, some data suggest that the oceanic-atmospheric phenomenon called El Niño-Southern Oscillation (ENSO) could be implicated. It has been reported that the high rainfall of ENSO increases water level, primary productivity, and swan's population abundance of El Yali wetland, located in northern Chile (Vilina *et al.* 2002). During inter-ENSO years, swans leave this habitat due to droughts and move towards more stable southern wetlands. On the contrary, the southern population of swans reaches its maximum abundance during the dry years in the Carlos Anwandter Nature Sanctuary located in the Cruces River (Schlatter *et al.* 2002). Authors explained that the ENSO-rainfall increases the water level of this wetland, making the access to food difficult to Black-necked swans which lack the ability to submerge their body into the water (Blem 2000).

The understanding of the challenges faced by these birds in heterogeneous environments allows us to identify the plasticity of the behavioural and physiological responses to

promote their survival and reproduction. We hypothesize that seasonality will induce a behavioural response of Budi Lake swan population that guarantee the nutrients and energy acquisition. The objective of the study was to determine the effect of seasonality on water level, trophic behaviour, and nutritional status of Black-necked swan populations inhabiting the Budi Lake.

MATERIAL AND METHODS

SITE OF STUDY

The study was conducted in the Budi Lake (38°53'00"S 73°17'00"O), a shallow saline coastal lake located in the Araucanía Region, Chile (Fig. 1).

WATER LEVEL

Water level of the Budi Lake was measured in a pier located at the periphery of the wetland, using a graduated rod at

the same point during three non-consecutive days, between 12:00 h and 13:00 h, in July (winter) and December (spring), in 2018.

TIME BUDGET

The time budget included the observation of 60 swans (n=30 in July, and n=30 in December) without sex distinction. Every bird was monitored using binoculars to record its activity every 5 minutes for one hour. The observational period was performed from 8:00 am to 6:00 pm and it took 3 weeks in each season. Swans were randomly selected avoiding recording the same specimen through the movement of the observer. The activities recorded were classified into the following categories: 1) foraging activity classified as effortless or effortful foraging. The first one consisting of the submergence of the swan's neck into the water to foraging, while the second involving the submergence of the neck and the anterior part of the body exposing only the tail and hind limbs out of the water; 2) locomotion activity which grouped

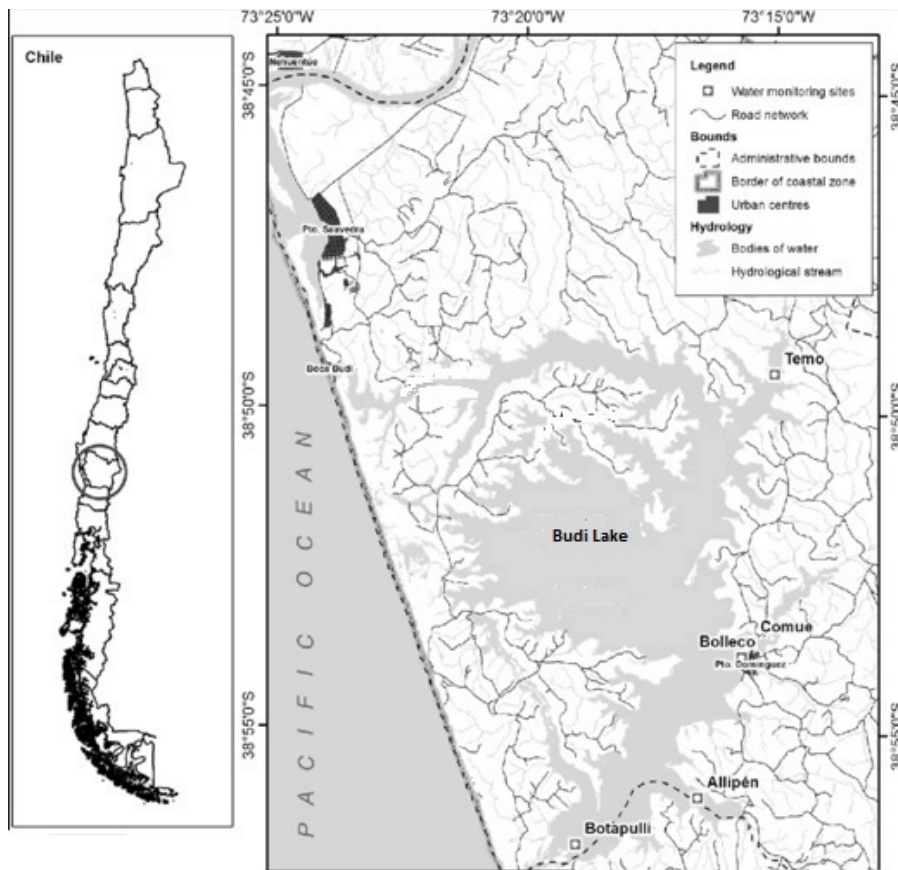


FIGURE 1. Study site. / Sitio de estudio.

swimming and walking; and 3) resting activity, including behaviours such as: standing on the water or land, grooming on the water or land, lying on their limbs on land or floating on the water without swimming, and the sleeping behaviour which was evident by the support of the swan's head on its back. Flying was not considered in this study. The proportion of effortful foraging behaviour was estimated over the total observations of foraging.

NUTRITIONAL STATUS OF BLACK-NECKED SWANS

Adult male swans were captured in July ($n=10$) and December ($n=10$) after being approached by boat, using a rod with a metal ring and mesh. Swans were aged before the capture by differentiating the size of the bird, and the caruncle size. When captured, adults and juvenile birds can be differentiated because the latter has black color at the end of the wings (Nascimento 2001). The couples were identified prior to the capture, differentiating males from females by their greater body size, and greater development of the neck and the caruncle. Once captured, the swan was sexed by eversion of the cloaca and identification of the erectile penis. Morphometric measurements such as length (from the end of the beak to the end of the tail feathers), the wingspan (length between the ends of the spread wings), and the length of the tarsus were obtained. Body weight was recorded using a digital scale (± 0.01 kg) and a blood sample (4 ml in tubes with heparin) was obtained from the brachial vein using a 21 G needle accoupled to a 5 ml syringe. Swans were captured on the same day and they were painted on the neck plumage previous to the release to avoid recapture. Blood samples were immediately centrifuged at 1500 rpm for 15 min and plasma was stored at -80°C until use. Cholesterol plasma concentration (CHODPAP method, HUMAN®), triglycerides (GPO-PAP method, HUMAN®) and total proteins (Biuret method) were analysed using Cobas-Mira-PlusH autoanalyzer (Roche, D-10587 Berlin, Germany) in the Austral University of Chile.

STATISTICAL ANALYSIS

The Shapiro-Wilk test and the Levene test were used to determine the normal distribution of the data and the homoscedasticity of variances, respectively. Water level, time budget and nutritional state data were compared between winter and spring using the Mann-Whitney U test. Proportions of effortful foraging were compared between months using Chi-square test. Data was analyzed in SPSS V 20 and a $P < 0.05$ was considered significant.

RESULTS

The water level recorded at the periphery of the wetland in winter (111 ± 3.4 cm) doubled that registered in spring (54 ± 0.6 cm, $P = 0.04$; Fig. 2). It was observed that in winter, swan population tended to allocate more time foraging ($58 \pm 5\%$ vs $51 \pm 2\%$, $P = 0.09$; Fig. 3), especially during the morning ($P < 0.05$; Table 3). Also, in winter, swan populations spent significant less time resting ($7 \pm 4\%$ vs $11 \pm 2\%$, $P = 0.05$) compared to in spring. The reduction of time allocated to resting were also mainly registered during morning suggesting that birds privilege the use of time in the foraging activity than resting ones. Locomotion activity was similar between populations ($35 \pm 4\%$ vs $38 \pm 4\%$, $P = 0.5$). The proportion of effortful foraging behaviour was higher in winter than spring ($37/222$ vs $14/185$ respectively, $P = 0.01$).

Both populations showed no significant differences in their morphology and biochemical parameters ($P > 0.05$) although the wingspan was greater in the Black-necked swans captured in winter (Table 3). During the study it was not observed reproductive activities in Budi Lake.

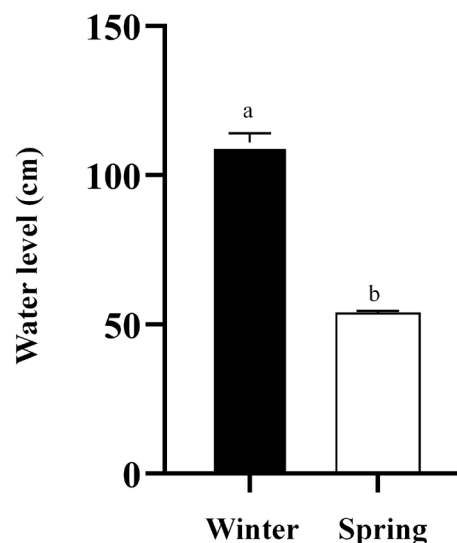


FIGURE 2. Water level (cm) registered at the periphery of Budi Lake in winter and spring. ^{a,b} Letters indicate significant differences ($P < 0.05$). / Nivel del agua (cm) registrados en la periferia del Lago Budi en invierno y primavera. ^{a,b} Letras indican diferencias significativas ($P < 0,05$).

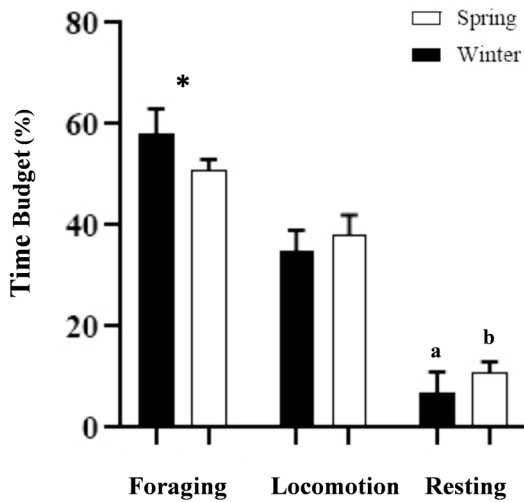


FIGURE 3. Time budget (%) of Black-necked swan populations during spring and winter at Budi Lake. * $P = 0.09$. ^{a,b} Letters indicate significant differences ($P < 0.05$). / Presupuesto de tiempo (%) de poblaciones de cisnes de cuello negro en invierno y primavera en el Lago Budi. * $P = 0,09$. ^{a,b} Letras indican diferencias significativas ($P < 0,05$).

TABLE 1. Time budget (%) comparisons between winter (n=30) and spring (n=30) Black necked swan’s populations of Budi Lake. / Comparación del presupuesto de tiempo (%) de poblaciones de cisnes de cuello negro del Lago Budi de invierno (n=30) y primavera (n=30).

Time (hours)	Activity	Winter	Springer	P
8 -9	Foraging	44 ± 10	33 ± 23	0.4
	Locomotion	56 ± 10	47 ± 13	0.5
	Resting	3 ± 5	0 ± 0	1
9 - 10	Foraging	56 ± 5	28 ± 19	0.02
	Locomotion	39 ± 13	47 ± 13	0.5
	Resting	8 ± 8	25 ± 8	0.02
10 - 11	Foraging	80 ± 10	39 ± 27	0.01
	Locomotion	28 ± 5	53 ± 19	0.03
	Resting	0 ± 0	8 ± 8	0.08
11 - 12	Foraging	64 ± 19	33 ± 14	0.01
	Locomotion	17 ± 8	36 ± 19	0.06
	Resting	19 ± 13	31 ± 17	0.09
12 - 13	Foraging	28 ± 13	64 ± 25	0.001
	Locomotion	39 ± 10	28 ± 17	0.3
	Resting	27 ± 21	6 ± 10	0.005
13 -14	Foraging	67 ± 17	54 ± 6	0.6
	Locomotion	31 ± 13	42 ± 8	0.3
	Resting	3 ± 5	0 ± 0	0.3
14 -15	Foraging	78 ± 17	53 ± 13	0.04
	Locomotion	17 ± 22	31 ± 5	0.2
	Resting	6 ± 5	14 ± 13	0.2
15 -16	Foraging	75 ± 14	58 ± 9	0.1
	Locomotion	25 ± 14	36 ± 10	0.3
	Resting	0 ± 0	8 ± 8	0.2
16 -17	Foraging	39 ± 5	58 ± 25	0.06
	Locomotion	61 ± 5	33 ± 17	0.009
	Resting	0 ± 0	11 ± 13	0.04
17 -18	Foraging	58 ± 8	69 ± 48	0.3
	Locomotion	42 ± 8	28 ± 5	0.009
	Resting	0 ± 0	3 ± 5	0.3

TABLE 2. Morphological and biochemical variables (Mean \pm SD) of Black-necked swans from Budi Lake during winter (n=10) and spring (n=10). / Variables morfológicas y bioquímicas (Prom \pm Desv Est) de cisnes de cuello negro del Lago Budi en invierno (n=10) y primavera (n=10).

Variable	Winter	Spring	P value
Length (cm)	119 \pm 3	120 \pm 4	0.5
Wingspan (cm)	160 \pm 6	136 \pm 15	$P < 0.001$
Tarsus (cm)	8.9 \pm 0.5	9.2 \pm 0.2	0.09
Body weight (kg)	6.9 \pm 0.5	6.9 \pm 0.6	0.6
Cholesterol (mmol/L)	2.6 \pm 0.3	2.9 \pm 0.5	0.1
Triglycerides (mmol/L)	1.5 \pm 0.6	1.4 \pm 0.3	0.4
Total protein (g/l)	57 \pm 6	55 \pm 5	0.6

SD: Standard Deviation

DISCUSSION

Based on the results of the present study, the water level of the Budi Lake registered in winter doubled that register in spring, resulting in changes in the time budget and trophic behaviour of Black-necked swans. They increased time allocated to foraging and the effortful foraging behavior and reduced time allocated to resting, allowed them to maintain an adequate energetic and nutritional state.

The increase in the water level could be related to the seasonal changes in precipitation since the accumulated precipitation of winter (544 mm) was greater than that of spring (394 mm). Nevertheless, the water level is influenced by the entry of seawater, in both a superficial and an underground way (Rodríguez 2005). Therefore, the local authorities facilitate the drainage of the lake 2 to 3 times a year, removing the sand deposits from the place where the lake drain into the sea. This intervention was performed 1 month before the winter study was started.

The Black-necked swans of Budi Lake dedicated more than half of the time budget to foraging activities in winter and spring, which is very similar to the results of previous studies for *C. melancoryphus* in the same habitat (57%, Norambuena 2008). Also, both Cursach *et al.* (2015) and Corti (1996) pointed out that the Black-necked swans foraged about 57% of the day during the winter season in Caulín Bay and Chihuo wetland, in Southern Chile, respectively.

In the present study, the winter population increased the time allocated to foraging mainly at morning and increased the effortful foraging activity compared to spring. Both patterns are probably caused by the increase of water level and the consequent difficulty in accessing food. Indeed, the water level differences between months (57 cm) is similar to

the length of an adult swan neck (56-70 cm, Corti 1996). A plausible explanation for the increment of forage intensity during morning in winter, is that Budi Lake is located in a region whipped by strong cold wind from 2 pm in the afternoon onwards, therefore birds may find a calmer lake that facilitates their foraging behaviour during morning. This climatic phenomenon is increased in winter.

The increase of effortful foraging behaviour could also be explained by differences in diet selection. It has been described that Black-necked swans that consumed roots under the sediment had a higher frequency of effortful foraging than those that fed on aquatic plants (Norambuena 2008; Norambuena & Bozinovic 2009a). On the other hand, the birds might have tended to allocate more time to forage in winter considering the differences in light hours that exist between months (about 3 h), which favors the acquisition of nutrients and energy in spring. Nevertheless, it has been described that this species had some nocturnal forage activity (Corti 1996).

Winter and spring populations showed similarities ($P > 0.05$) in the morphological variables although the wingspan was lower in swans captured in spring due to the absence of some primary feathers which is explained by the process of molting that has been reported before for the species belonging to the same habitat, in the same season (Norambuena & Bozinovic 2009a, 2009b). The total length and tarsal length agreed with the values described for the adult males of this species (Boetcher *et al.* 2004; Norambuena & Bozinovic 2009a, 2009b). Regarding the nutritional status, the body weight of both groups agreed with reference values obtained from adult swans under captivity (Norambuena & Bozinovic 2009a, 2009b). Mean values of cholesterol, triglycerides, and total proteins plasma concentration were according to reference

values for the species (Boettcher *et al.* 2004), indicating an adequate energy and protein metabolism. Also, these variables were similar to those obtained in well-nourished Black-necked swans of Budi Lake (Norambuena & Bozinovic 2009a, 2009b).

Nevertheless, it is not possible to make a cause-effect relationship between the water level changes, behavioural adjustments and the nutritional status of the birds because the migratory status of swan populations is poorly known.

In conclusion, although there are differences in the water level of Budi Lake between winter and spring, Black-necked swan populations maintained an adequate nutritional status due to behavioral adjustments.

ACKNOWLEDGMENTS

The authors acknowledge to the Diaz Nahuel family, Azlilko Lewfu Budi indigenous association, the Agricultural and Livestock Service of Chile and the Bioethics Committee of the Catholic University of Temuco, who authorized the study and supported the field work.

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Received: 08.10.2021

Accepted: 04.01.2023