



# Relationships between fresh weight, dry weight, ash free dry weight, carbon and nitrogen content for selected vertebrates



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## ABSTRACT

Top predators are relevant indicators of the ecological status of a system and can have a high impact on food webs. But top predators are difficult to include in network analyses because their biomass in ash free dry weight or carbon content is missing. Regression equations were determined for the relationships between fresh weight and dry weight, ash free dry weight, carbon and nitrogen contents respectively for six of the most abundant bird species in the Wadden Sea (*Calidris canutus*, *Limosa lapponica*, *Haematopus ostralegus*, *Chroicocephalus ridibundus*, *Larus canus*, *Anas penelope*) and harbor seals (*Phoca vitulina*). The relationships for all species were interpreted as linear through the origin. Carbon content vs. fresh weight ratios for birds ranged from  $0.16 \pm 0.01$  to  $0.22 \pm 0.02$ . Carbon content vs. fresh weight ratio was  $0.17 \pm 0.02$  on average for harbor seals. This work highlights that the biomass of top predators was often over- or underestimated in previous studies. The determined conversion factors will be useful for future studies to generate more realistic food web models.

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## 1. Introduction

In the last decades, food web models and ecological networks have become useful tools to describe the functioning of large and complex ecosystems encompassing numerous compartments interacting with each other and responding differently to external stressors (Ings et al., 2009). In many studies, network analyses have been used to define ecosystem properties. These properties include the ecosystem structural complexity, the structure and magnitude of the cycling of energy and material, the efficiency of energy transfer within the system, the rates of energy assimilation and dissipation, the trophic structure, the system activity, growth and development (Baird et al., 2004). Results from these models provide significant insights into the fundamental functioning of the ecosystem (Baird et al., 2004) and are very relevant for the management of marine ecosystems (Samhuri et al., 2009).

Abundance and distribution of top predators, such as sea birds and marine mammals, can have a large influence on community structures and on the functioning of the ecosystem they live in (Baird et al., 1985; Bowen, 1997; Moreira, 1997). As a corollary, they are good indicators for ecosystem's health (Furness and Camphuysen, 1997; Reddy et al., 2001; Bossart, 2011). Therefore, there is an increasing need to include marine birds and mammals in ecosystem models, especially in studies about trophodynamic to have a better understanding of food web functioning, allowing improvement of management plans for conservation.

Studies about marine bird and mammal populations are classically based on abundance data (Reijnders et al., 1997; Brasseur et al., 2013; Markert et al., 2013; Galatius et al., 2014; Mandema et al., 2015), which cannot be directly used to study matter or energy flow within ecosystems (Dumont et al., 1975). These abundance data can be converted to fresh weight values using average individual weight corresponding to the studied species. But the use of fresh tissue might lead to large approximations in the organic matter weight, as body water content can vary between taxa. The fresh weight is therefore a bad proxy for biomass comparison. In ecological studies it is a common practice to use standardized biomass units (e.g. dry weight, ash free dry weight, carbon content) allowing comparison of different species biomass from different locations or periods of time (e.g. seasons, years). Most of the mass balanced food web models such as ECOPATH with ECOSIM (Bradford-Grieve et al., 2003; Leguerrier et al., 2007; Pinkerton et al., 2010) and especially ecological network analyses (Baird et al., 2004; Scharler and Baird, 2005; Fath et al., 2007; Baird et al., 2012; Saint-Béat et al., 2013) also rely on these consistent and standardized biomass units (e.g. dry weight, ash free dry weight, carbon content).

Although a large database of conversion factors from fresh weight to standardized biomass units is available for macrobenthic invertebrates (Rumohr et al., 1987; Ricciardi and Bourget, 1998), to our knowledge, no such database exists for marine birds and mammals. As a result, including top predators in ecosystem models is very difficult. It is associated with a high degree of uncertainty and relies on large approximations that might bias the model outputs.

The aim of this study was to determine relationships useful for modeling between fresh weight (FW) and dry weight (DW), FW and ash free dry weight (AFDW), FW and carbon content (CC) and FW and

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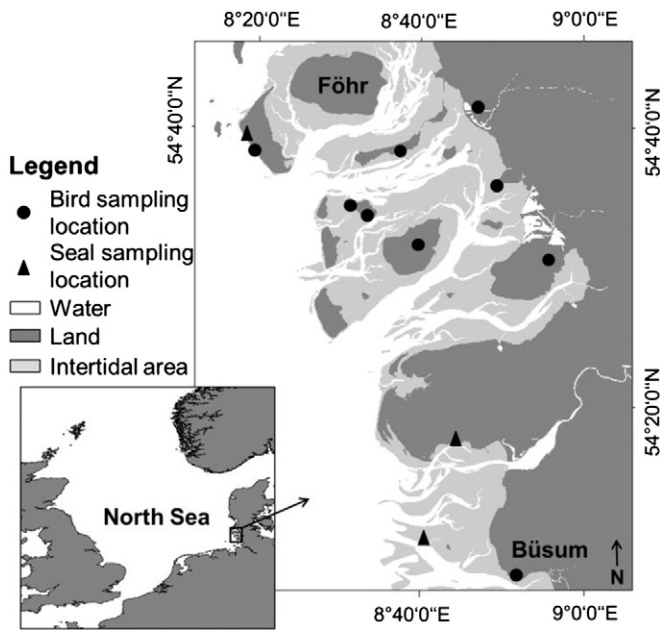


Fig. 1. Location and map of the study area. The circles and triangles refer to the locations where carcasses of birds and seals were respectively found.

nitrogen content (NC). These relationships were determined for six of the most abundant bird species in the Wadden Sea (Blew et al., 2013) (*Calidris canutus*, Linnaeus, 1758; *Limosa lapponica*, Linnaeus, 1758; *Haematopus ostralegus*, Linnaeus, 1758; *Chroicocephalus ridibundus*, Linnaeus, 1766; *Larus canus*, Linnaeus, 1758; *Anas penelope*, Linnaeus, 1758), and for harbor seal (*Phoca vitulina*, Linnaeus, 1758), one of the most abundant marine mammal species in this area (Reijnders et al., 2009).

## 2. Material and methods

Carcasses of birds and seals were collected along the shore of the eastern German Wadden Sea, between the coastal city Büsum in the South and the island of Föhr in the North (Fig. 1). Only fresh carcasses which did not show any noticeable signs of starvation or diseases were selected for this study.

Seventeen birds from six different species (*C. canutus*, *H. ostralegus*, *L. lapponica*, *C. ridibundus*, *L. canus*, and *A. penelope*) were collected by a network of volunteers. Three individuals were collected for each species, except for *A. penelope* for which only two birds were available. Most individuals died due to collision with lighthouses or cars (Table 1). Carcasses were stored frozen in plastic bags at  $-20\text{ }^{\circ}\text{C}$  until preparation for analyses. Each individual was unfrozen and grinded entirely using a kitchen cutter (RCKC-6000, Royal Catering, 750 W) in order to get a homogenized mixture composed of all the tissues. Four subsamples were collected from each grinded individual: three for determination of fresh weight (FW), dry weight (DW) and ash free dry weight (AFDW), and one for carbon content (CC) and nitrogen content (NC) analyses.

Three harbor seals were collected in 2015 (Table 2) as part of the stranding network established along the German coasts of Schleswig-Holstein (Benke et al., 1998; Siebert et al., 2006). Carcasses were stored frozen in plastic bags at  $-20\text{ }^{\circ}\text{C}$  until necropsies, which were carried out according to the protocol described by Siebert et al. (2007), at the Institute for Terrestrial and Aquatic Wildlife Research of the University of Veterinary Medicine, Hannover Foundation. The different tissues were dissected and weighed ( $\pm 0.1\text{ g}$ ). The contribution of each tissue to the total fresh weight was determined for each individual. Two subsamples were collected from each tissue and each individual: one for determination of FW, DW, AFDW and one for determination of CC and NC.

The FW of each subsample of birds and seals was measured to the nearest 0.1 mg. Subsamples were dried in an oven at  $50\text{ }^{\circ}\text{C}$  until constant weight and the DW was measured ( $\pm 0.1\text{ mg}$ ). Each subsample was then burned in a furnace at  $500\text{ }^{\circ}\text{C}$  for 5 h, cooled down in a desiccator and ash weight was measured ( $\pm 0.1\text{ mg}$ ). AFDW was determined by subtracting the ash weight from the DW. For CC and NC, subsamples were freeze-dried and grinded into a fine powder using a ball mill. An amount of each powder was precisely weighed ( $\pm 1\text{ }\mu\text{g}$ ) and sealed in a tin capsule. CC and NC were measured using an elemental analyzer (Flash EA 1112, Thermo Scientific, Milan, Italy) at the LIENSs stable isotope facility of the University of La Rochelle, France. Acetanilide (Thermo) and peptone (Sigma-Aldrich) were used as standards for CC and NC calibration.

Relationships between FW and DW, AFDW, CC and NC respectively were plotted for bird species and for each seal tissue. These plots were then made for entire seal individuals taking into account the mass proportions of each tissue in FW. Missing data for some tissues were estimated by assuming that the proportion of the weight of missing tissue is the same as in *P. vitulina* 1 (Table 6).

The regression equations for FW and DW, AFDW, CC and NC respectively were calculated for all individuals of bird species combined, for the seal tissues and for entire seals.

Table 1  
Species, date of collection, total fresh weight of individuals, season and cause of death of the birds.

Species #	Date of collection	Total fresh weight (g)	Season	Cause of death
<i>C. canutus</i> 1	4th Apr. 2014	114.8	Spring	Unknown
<i>C. canutus</i> 2	21st Sep. 2014	119.5	Autumn	Unknown
<i>C. canutus</i> 3	7th Jul. 2014	108.6	Summer	Unknown
<i>L. lapponica</i> 1	2nd Apr. 2004	246.2	Spring	Lighthouse collision
<i>L. lapponica</i> 2	20th Mar. 2007	270.5	Spring	Lighthouse collision
<i>L. lapponica</i> 3	25th Jan. 2007	299.2	Winter	Lighthouse collision
<i>H. ostralegus</i> 1	2nd Jun. 2014	464.7	Summer	Unknown
<i>H. ostralegus</i> 2	27th Mar. 2014	371.7	Spring	Unknown
<i>H. ostralegus</i> 3	27th Apr. 2009	501.3	Spring	Unknown
<i>C. ridibundus</i> 1	27th Sep. 2013	231.7	Autumn	Lighthouse collision
<i>C. ridibundus</i> 2	13th Sep. 2013	198.5	Autumn	Unknown
<i>C. ridibundus</i> 3	3rd Jun. 2012	150.1	Summer	Unknown
<i>L. canus</i> 1	6th May. 2013	521.1	Spring	Unknown
<i>L. canus</i> 2	4th Jul. 2014	332.4	Summer	Vehicle collision
<i>L. canus</i> 3	17th Nov. 2006	442.0	Autumn	Vehicle collision
<i>A. penelope</i> 1	15th Jan. 2002	777.5	Winter	Lighthouse collision
<i>A. penelope</i> 2	11th Nov. 2007	795.7	Autumn	Lighthouse collision

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