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MEDITERRANEAN SEABIRDS AND THEIR CONSERVATION. *E. MÍNGUEZ, D. ORO, E. DE JUANA and A. MARTÍNEZ-ABRAÍN (eds.)*

Using stable isotopes of nitrogen and carbon to study seabird ecology: applications in the Mediterranean seabird community*

MANUELA G. FORERO[1] and KEITH A. HOBSON[2]

[1] Instituto Mediterráneo de Estudios Avanzados (CSIC-UIB), C/Miquel Marqués, 21, 07190 Esporles, Mallorca, Spain. E-mail: gforero@uib.es
[2] Canadian Wildlife Service, 115 Perimeter Road, Saskatoon, SK, Canada, S7N0X4

SUMMARY: The application of the stable isotope technique to ecological studies is becoming increasingly widespread. In the case of seabirds, stable isotopes of nitrogen and carbon have been mainly used as dietary tracers. This approach relies on the fact that foodweb isotopic signatures are reflected in the tissues of the consumer. In addition to the study of trophic ecology, stable isotopes have been used to track the movement of seabirds across isotopic gradients, as individuals moving between isotopically distinct foodwebs can carry with them information on the location of previous feeding areas. Studies applying the stable isotope methodology to the study of seabird ecology show a clear evolution from broad and descriptive approaches to detailed and individual-based analyses. The purpose of this article is to show the different fields of application of stable isotopes to the study of the seabird ecology. Finally, we illustrate the utility of this technique by considering the particularities of the Mediterranean seabird community, suggesting different ecological questions and conservation problems that could be addressed by using the stable isotope approach in this community.

Key words: stable isotopes, Mediterranean Sea, seabirds, trophic ecology, conservation, nitrogen, carbon.

RESUMEN: EL EMPLEO DE ISÓTOPOS ESTABLES DE NITRÓGENO Y CARBONO PARA ESTUDIAR LA ECOLOGÍA DE LAS AVES MARINAS: APLICACIONES A LA COMUNIDAD DE AVES MARINAS DEL MEDITERRÁNEO. – La aplicación de la medición de isótopos estables a los estudios ecológicos es cada vez más frecuente. En el caso de las aves marinas, los isótopos estables de nitrógeno y carbono han sido empleados principalmente como indicadores del régimen alimenticio. Este enfoque se basa en el hecho de que las concentraciones isotópicas de las cadenas tróficas se reflejan en los tejidos del consumidor. Además de para el estudio de la ecología trófica, los isótopos estables se han utilizado para estudiar el movimiento de las aves marinas a través de gradientes isotópicos, ya que los individuos que se mueven entre cadenas tróficas isotópicamente distintas pueden llevar con ellos información sobre la ubicación de áreas de alimentación anteriores. Los estudios que aplican la metodología de los isótopos estables al conocimiento de la ecología de las aves marinas muestran una clara evolución, desde los enfoques amplios y descriptivos, a los análisis detallados y basados en los individuos. El objetivo de este trabajo es mostrar los diferentes campos de aplicación de los isótopos estables en el estudio de la ecología de las aves marinas. Finalmente, ilustramos la utilidad de esta técnica considerando las particularidades de la comunidad de aves marinas del mar mediterráneo, sugiriendo diferentes cuestiones ecológicas y problemas de conservación que se podrían abordar empleando la técnica de los isótopos estables en esta comunidad.

Palabras clave: isótopos estables, Mediterráneo, aves marinas, ecología trófica, conservación, nitrógeno, carbono.

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BACKGROUND

For some time the measurement and use of naturally occurring stable isotopes has been restricted largely to investigations in the earth sciences. However, during the last two decades the application of this methodology to ecological studies has become widespread (Lajtha and Michener, 1994; Hobson, 1999; Kelly, 1999). In addition to a growing interest by ecologists and physiologists, the increase has also resulted from substantial technical advances that allow minimal sample preparation and the automated processing of a large number of samples. Stable isotope ratios are measured using an isotope ratio mass spectrometer (IRMS) with values reported as the ratio between the heavy and light isotopes compared to standards which vary depending on the stable isotope used. In the case of nitrogen and carbon, the most commonly used in marine systems, the standards are AIR (N_2) and Pee Dee Belemnite (PDB) respectively. The deviation (δ) of the ratio (R) is expressed per thousand (‰):

$$\delta X = [\,(R_{sample}/R_{standard}) - 1] \times 1{,}000$$

where X is the heavier isotope and R is the corresponding ratio between the heavier and the lighter isotope.

Stable isotope measurements of carbon and nitrogen have been used mainly as dietary tracers in seabirds, but recent investigations also show that these have the potential for locating moulting areas of migratory seabird species (Hobson, 1993; Cherel *et al.*, 2000). The utility of $\delta^{15}N$ measurements in studies of seabirds and feeding ecology relies on trophic enrichment of the heavier isotope (^{15}N) in foodwebs. During the production of urea and uric acid, molecules of compounds containing the lighter atom ^{14}N are preferentially excreted over those containing the heavier isotope (Peterson & Fry, 1987). Tissues of consumers tend to be enriched in ^{15}N compared to their diet (Minagawa and Wada, 1984; Michener and Schell, 1994). Studies using $\delta^{13}C$ values have been valuable because the rate at which ^{13}C and ^{12}C isotopes are fixed by plants differ among photosynthetic pathways. In marine systems, phytoplankton have lighter $\delta^{13}C$ values than many inshore plants, suggesting that information about the sources of feeding, including inshore vs. offshore sources in marine habitats are available through stable isotope measurements (Fry and Sherr, 1989; Boutton, 1991; Hobson *et al.,* 1994; France, 1995; Hobson *et al.,* 1995). Studies that have determined the diet-tissue fractionation factor of nitrogen and carbon isotopes for marine consumers show that most tissues are enriched over the diet by about 3-5‰ and 1-3‰ respectively, although important variations may exist among tissues (DeNiro and Epstein, 1981; Mizutani *et al.,* 1992; Hobson and Clark, 1992a, 1992b; Michener and Schell, 1994; Bearhop *et al.,* 1999).

The advantages of the isotope methodology for studying seabird trophic ecology are numerous. Previous conventional studies of seabird diet relied upon examination of stomach contents of birds. However, this methodology has several sources of bias, including the rapid digestion of soft-bodied prey (see review in Duffy and Jackson, 1986 and González-Solís *et al.,* 1997a). In addition, there are some ecological problems in which the stable isotope technique provides solutions and conventional studies of diet perform poorly (e.g. quantifying trophic level). Combined approaches in which conventional studies and the stable isotope technique are complementary could avoid these biases and limitations. Feeding experiments have shown that the turnover rate of elements in a particular tissue varies with that tissue's metabolic activity (Tieszen *et al.,* 1983; Hobson and Clark, 1992*a*). Therefore, different tissues sampled from the same individual can provide dietary information reflecting different periods of time, allowing the evaluation of diet over various temporal scales and reducing the need for expensive sampling efforts. Recent investigations have also indicated that a great deal of dietary information can be obtained from the isotopic analysis of blood and feathers, thereby reducing the need to sacrifice birds just for analysis of diet (Hobson and Clark, 1992a, 1993; Thompson and Furness, 1995; Hobson *et al.,* 1997; Hodum and Hobson, 2000; Cherel *et al.,* 2000). In the case of feathers, since keratin is a metabolically inert tissue it conserves dietary signals integrated over the period in which they have grown. This quality, together with differences in isotope signatures between geographically separated foodwebs, offers an additional application of stable isotope analysis to the study of spatial and long-term changes in diet.

Evaluation of animal movement is an important component in understanding population dynamics and ecology. Conventional approaches used to track animal movements, such as external markers or tags, necessarily involve the subsequent recovery or sighting of individuals. In the case of seabirds, they are accessible for investigation only during a short period of the year because they spend most of their time offshore. Stable isotope analyses of feathers sampled on the breeding grounds offers an alternative means of studying diet and foraging areas during the non-breeding period. Because some ocean areas differ in the isotopic signature of foodwebs (e.g. Wada *et al.,* 1987), isotopic analyses may provide inference on location of moulting areas of seabirds (Schell *et al.,* 1999; Cherel *et al.,* 2000).

Although the application of stable isotopes to ecological research is increasing, in many cases the correct interpretation of the results obtained from wild studies depends on laboratory experiments that clarify many of the assumptions on which these studies rely (Gannes *et al.,* 1997). So far, most of the applications of stable isotopes to ecological studies have reported results for wild populations, while experimental data are scarce (see review in Kelly, 1999). More research is required about the fundamental principles governing the behaviour of stable isotopes and how parameters such as elemental turnover rates and diet-tissue isotopic fractionation factors are affected by different ecological and individual conditions such as age, nutritional status and fasting (Hobson and Clark, 1992a; Hobson *et al.,* 1993; Bearhop *et al.,* 2000; Thompson *et al.,* 2000).

This review will focus on the use of stable isotopes in the study of seabird ecology. Specifically, we review some of the primary studies dealing with the delineation of diet and trophic ecology as well as the tracking of movement of seabirds across isotopic gradients. We also consider the potential contribution of this technique to issues involving seabird conservation and management and end by examining specific applications to seabird studies in the Mediterranean region.

TROPHIC ECOLOGY

Ecological questions: from individuals to communities

Studies using the stable isotope approach in seabird ecology have emerged during the last decade (Table 1) with a clear evolution from broad approaches, mainly descriptions of the trophic relationships in seabird communities, to individual based analyses. These more recent analyses explore different sources of intraspecific variability in isotope values and reveal the importance of trophic relationships and diet characteristics on population parameters and dynamics.

Hobson and Welch (1992) studied $\delta^{15}N$ and $\delta^{13}C$ in an Arctic food-web from primary producers to higher-level carnivores including seabirds. They found that although Arctic cod (*Boreogadus saida*), previously thought to be critical in the transfer of energy from primary producers to higher level vertebrates, was an important component, lower trophic level invertebrates also play a critical role in this process of energy coupling. Hobson (1993) continued these isotopic studies and Hobson *et al.* (1994) also analysed $\delta^{15}N$ and $\delta^{13}C$ values in 22 species of seabirds in the Gulf of Alaska and coastal British Columbia (Northeast Pacific Ocean, NPO). In both studies, according to the isotope data, seabird species consumed more lower-trophic-level prey than suggested by previous conventional dietary studies. Based on isotopic signatures of tissues with different turnover rates, Hobson (1993) also determined that the consumption of lower-trophic level prey in the Arctic seabird community increased on breeding compared to wintering grounds.

Sydeman *et al.* (1997) showed that stable isotope analyses combined with conventional methods improved the evaluation of the trophic relationships and temporal changes in the diet of seabirds in the Gulf of Farallones (California), an ecosystem characterised by extensive changes in physical oceanographic processes, which could have substantial effects on seabird feeding strategies and their demography. Analysis of stable isotopes in egg albumen and observations of adults feeding their chicks revealed that only one of the 6 studied species, the common murre (*Uria aalgae*), shifted its diet between spring (egg formation) and summer from krill to fish, which could be an adaptation to cope with limited or uncertain food resources. Another study underlying the importance of coupling conventional approaches and stable isotope analysis is that by Thompson *et al.* (1999) on the seabird community of Iceland. These authors demonstrated that although stable isotope measurements refined small-spatial scale differences in trophic relationships, the contribution to the diet of rare prey species using isotopic mixing models was overestimated. More recently, advances in the use of isotopic mixing models will undoubtedly refine this approach (Phillips, 2001).

Recently, some studies have applied stable isotope analysis to study intra and interspecific trophic segregation (e.g. Hodum and Hobson, 2000; Bocher *et al.*, 2000; Thompson *et al.,* 2000; Forero *et al.,* 2002). In relation to interspecific analyses, results varied

TABLE 1. – Summary of major studies dealing with application of stable isotopes of nitrogen and carbon to the study of seabird ecology in wild populations. Different categories have been defined in relation to the main focus of the study and wide geographic areas where the study was carried out are indicated.

Study	Species	Type of sample	Category	Geographic place
Hobson, 1987	Glaucous-winged gull (*Larus glaucescens*)	Muscle	Terrestrial vs. marine feeding	Coastal British Columbia
Hobson and Montevecchi, 1991	Great auk (*Pinguinus impennis*)	Bone collagen	Reconstruction of diet in extinct animals	Northwest Atlantic and High Arctic
Mizutani *et al.*, 1991	Rockhopper penguin (*Eudyptes chrysocome*)	Soil	Identification of deserted colonies	New Zealand
Hobson and Welch, 1992	Whole marine community	Muscle	Trophic level and prey contribution	Northwest Territories
Hobson, 1993	Seabird community	Muscle, liver, bone collagen	Trophic relationships	Northwest Territories
Hobson *et al.,* 1994	Seabird community	Muscle, bone collagen	Trophic relationships	Northeast Pacific Ocean
Hobson *et al.,* 1995	Whole marine community	Whole specimens, muscle, blood	Trophic relationships	Northeastern Greenland
Minami *et al*., 1995	Sooty shearwater (*Puffinus griseus*)	Muscle	Migration	North Pacific
Thompson and Furness, 1995	Northern fulmar *(Fulmarus glacialis*)	Feathers	Seasonal variation	Northeast Atlantic
Thompson *et al.,* 1995	Northern fulmar (*Fulmarus glacialis*)	Feathers	Long-term changes in diet	Northeast Atlantic
Jarman *et al.,* 1996	Whole marine community	Egg albumin, homogenised composites, muscle	Trophic position and contaminant levels	Gulf of Farallones
Gould *et al.*, 1997	Laysan (*Diomedea immutabilis*) and black-footed albatrosses (*D. nigripes*)	Muscle	Interspecific, intraspecific differences and interaction with fisheries	North Pacific Ocean
Jarman *et al.,* 1997	Seabirds and marine mammals	Egg albumin, muscle	Trophic position and dioxins and dibenzofurans concentrations	Central California
Minami and Ogi, 1997	Sooty shearwater (*Puffinus griseus*), short-tailed shearwater (*P. tenuirostris*)	Muscle	Migration	North Pacific Ocean
Sydeman *et al.,* 1997	Seabird community	Egg albumen, muscle	Trophic relationships and temporal changes in diet	Central California
Atwell *et al.*, 1998	Whole marine community	Whole-body composites, muscle	Biomagnification and bioaccumulation of mercury	Northwest territories
Schmutz and Hobson, 1998	Glaucous gull (*Larus hyperboreus*)	Muscle, liver, blood	Geographic, temporal variation and age differences	Alaska
Bearhop *et al.,* 1999	Cormorants (*Phalacrocorax carbo*)	Feathers	Marine vs. prey from freshwater fisheries	England
Hobson *et al.,* 1999	Norway rats (*Rattus norvegicus*)	Muscle, liver	Predation on seabirds: marine vs. terrestrial diet	British Columbia
Thompson *et al.,* 1999	Seabird community	Liver	Trophic relationships and spatial changes	Iceland
Bearhop *et al.,* 2000a	Great skua (*Catharacta skua*)	Blood	Age-differences and influence of lipid and acid uric	Northeast Atlantic
Bearhop *et al.,* 2000b	Great skua	Feathers, blood	Trophic ecology and mercury accumulation	Northeast Atlantic
Bocher *et al.,* 2000	South Georgian and common diving petrels (*Pelecanoiddes georgicus* and *P. urinatrix)*	Feathers	Seasonally variation in interspecific segregation	Southern Indian Ocean
Cherel *et al.,* 2000	Black browed albatrosses (*Diomedea melanophrys*)	Feathers	Moulting foraging areas	Southern Indian Ocean
Hodum and Hobson, 2000	Antarctic fulmar (*Fulmarus glacialoides*), Antarctic petrel (*Thalassoica antarctica*), Cape petrel (*Daption capense*), Snow petrel (*Pagodroma nivea*)	Blood	Inter and intraspecific variability (age)	East Antarctica
Thompson *et al.,* 2000	Procellariiform	Liver	Dietary lipid- derived carbon	New Zealand
Forero *et al.,* 2002a	Magellanic penguin (*Spheniscus magellanicus*)	Blood	Age and sex segregation	Argentinean Patagonia
Forero *et al.* 2002b	Magellanic penguin	Blood	Intraspecific variability and population structuration	Argentinean Patagonia

depending on the ecological characteristics of the seabird community. For example, Hodum and Hobson (2000) found a large overlap in $\delta^{15}N$ values of four related and sympatric petrels in the Antarctic, where available prey species are dominated by a few species. A similar study based on 1440 blood samples from individuals of different sex, age and colonies, and representing 14 species of seabirds breeding in the Argentinean Patagonia, showed a hihg degree of overlap in trophic level ($\delta^{15}N$) and $\delta^{13}C$ values (Forero *et al.,* submit.). Authors concluded that coexistence of species in the community could be interpreted as a consequence of superabundance of food or species diversification in morphology and foraging strategies. Results of this work also suggest a high vulnerability of the community to the reduction of fish stocks due either to comercial fishing or stockastic fluctuations. Bocher *et al.* (2000) studied seasonal segregation in the feeding ecology of two sympatric diving petrels, the South Georgia diving petrel (*Pelecanoides georgicus*) and the common diving petrel (*Pelecanoides urinatrix*) in the Southern Indian Ocean by measuring $\delta^{15}N$ and $\delta^{13}C$ in feathers of both species moulted during the breeding and inter-breeding period. These authors found a complete dietary segregation between the two species during the chick-rearing period but not during the inter-breeding period, when birds forage for themselves.

The identification of individual characteristics that determine differences in diet or feeding ecology within populations could be of crucial importance to understanding population dynamics. Age and sex are potential factors that could influence such differences. However, assessing trophic differences between adults and chicks using conventional diet-sampling methods is virtually impossible, and also the lack of molecular methods for sexing birds have prevented these kinds of studies until recently (Ellegren, 1996; Forero *et al.*, 2001, 2002a; Tella *et al*., 2001; Bertellotti *et al.,* 2002). Recent advances in stable isotope analyses and molecular sexing have permitted the exploration of these sources of variability in several seabird species. A common finding has been that chicks exhibit higher $\delta^{15}N$ values than adults (e.g. Hobson, 1993; Schmutz and Hobson, 1998; Hodum and Hobson, 2000; Forero *et al.,* 2002), suggesting that adults feed their chicks with prey of a higher trophic level and nutritive value than they feed themselves, although in some cases differences may be linked to unusual fractionation factors determined by differences in growth rates between age-classes (e.g. Bearhop *et al.,* 2000). In addition, Forero *et al.* (2002a), by isotopically studying diet of Magellanic penguins (*Spheniscus magellanicus*) in Argentinean Patagonia, revealed that first-year birds occupied the lowest trophic level in the population. This study was also one of the first showing sexual differences in diet and foraging ecology in any penguin species (see also Clarke *et al.,* 1998). Although relatively new, the stable isotope technique undoubtedly provides a great tool for studying how different life-history strategies could determine the magnitude of sex and age differences in seabird species (Forero *et al.,* in prep.).

Intrapopulation variation in diet and foraging ecology could have important consequences for seabird population structure and dynamics, since such variations could affect different components of individual fitness (e.g. Golet *et al.,* 2000; Tella *et al.,* 2001). Although there is evidence that intraspecific food depletion could affect the size and distribution of colonies in seabirds (Furness and Birkhead, 1984), until recently it remained an unresolved question. However, Forero *et al.* (2002b), studying the Magellanic penguin in Argentinean Patagonia, extended the use of the stable isotope technique to answer this question. They demonstrated that $\delta^{15}N$ and $\delta^{13}C$ values are negatively and positively related, respectively, to local and broader-scale population densities, which suggest that when competition for food is high, individuals tend to feed closer to the colony on prey of lower quality. These authors also found a positive correlation between $\delta^{15}N$ values and two components of fitness, breeding success and offspring quality. Decreased breeding output was related to the colony-size structure of the population, since colony size was negatively correlated with the number of conspecifics within foraging ranges from colonies. This provided a connection between conspecific food depletion and the resulting population structure in colonial seabirds. The finding that stable isotopes, as indicators of diet, could be related to fitness components offers a great potential for studies of metapopulation dynamics in seabirds.

MOVEMENTS

So far, few studies have used stable isotopes to infer wintering grounds and foraging locations during non-breeding periods in seabirds (see review in Hobson, 1999). The first attempts to explore isotopic techniques to investigate wintering routes in seabirds relied on $\delta^{15}N$ and $\delta^{13}C$ analyses in sooty shearwater (*Puffinus griseus*) and short-tailed shearwater (*P. tenuirostris*) (Minami *et al.,* 1995; Minami and Ogi, 1997). These species breed in the Southern hemisphere and migrate to the North Pacific following two different routes, along the eastern and the western North Pacific. Although results of these works are subject to further sampling of the food webs along both migratory routes, these authors were able to segregate individuals following eastern or western routes during migration on the basis of differences in their $\delta^{15}N$ values. Cherel *et al.* (2000) analysed isotopically feathers of chick and adult Black-browed albatrosses (*Diomedea melanophrys*) moulted during the wintering and breeding period respectively. After addressing possible effects of diet shifts between both periods or of different diet-tissue fractionation factors for chicks and adults, these authors explained the differences they found in $\delta^{15}N$ and $\delta^{13}C$ values for feathers in terms of different foraging areas of this species during winter (i.e. north of the Subtropical Front, STF) and breeding period (south of the STF). Although previous studies show the potential of this technique to study migratory and wintering areas of seabirds, research in this field is limited by our relatively poor understanding of the biogeographic isotopic regions and the identification of small-scale biogeographic isotopic gradients in the food webs.

APPLICATIONS TO CONSERVATION AND MANAGEMENT

No review of applications of stable isotope analysis to study seabird ecology would be complete without a mention of the numerous and relevant uses of this technique for answering questions of conservation interest and delineating management strategies for seabird populations.

Fisheries

An obvious application of isotopic approaches to management is the interaction between seabirds and fisheries. Evaluation of long-term changes in the diet of seabirds is made difficult due to the scarcity of information from the past century. However, preservation of large numbers of skins in museum collections and the measurement of stable isotopes in feathers to infer past trophic habits offer an unprecedented opportunity for this kind of research. The northern fulmar (*Fulmarus glacialis*) has increased dramatically during the last century in the UK. By measuring stable isotopes of carbon and nitrogen in feathers from museum collections and from contemporary fulmars, Thompson *et al.* (1995) found that $\delta^{15}N$ values declined over the middle of the last century. They argued that the expansion of this species in the UK could be due to the utilisation by this species of offal from whaling during the last century. Another species that has increased rapidly during the last century throughout most of Europe is the common cormorant (*Phalacrocorax carbo*), which has led to increasing conflict with freshwater fisheries. Bearhop *et al.* (1999), measuring stable isotopes in feathers of this species in England, determined that they relied heavily on freshwater prey throughout autumn and winter.

Reconstruction of diet of extinct species

An extreme example of the use of stable isotope analysis to reconstruct the diet and trophic status of seabirds over long time scales is the work of Hobson and Montevecchi (1991), who investigated trophic characteristics of the extinct great auk (*Pinguinus impennis*) by measuring stable isotopes in extracted bone collagen. Results of this kind of study could be of special relevance to better understand the causes of seabird decline and extinction and for determining management decisions for seabird populations.

Introduced predators

A second, but not less important, problem of conservation in seabird populations is the vulnerability of ground-nesting birds breeding on islands to introduced predators, especially rats (*Rattus* spp.) (Bailey and Kaiser, 1993; Bertram, 1995; Seto and Conat, 1996; Martin *et al.*, 2000). Stable isotope measurements offer a new tool for evaluating the effect of introduced predators by describing long-term diet preferences and identifying individuals preying on ground nesting seabirds before the application of expensive and often logistically difficult eradication programs. On Langara Island (British Columbia, Canada), the ancient murrelet (*Synthliboramphus antiquus*) has declined dramatically during the last 40 years (Beltran, 1995). Hobson *et al.* (1999) were able to quantify the dependence of Norway rats (*Rattus norvegicus*) on seabirds, intertidal invertebrates and upland foods on Langara Island, by measuring $\delta^{15}N$, $\delta^{13}C$ and $\delta^{34}S$ values in the liver and muscle of rats as a means of segregating marine and terrestrial dietary inputs. In addition, by comparing these results with those from stomach contents these authors determined that stomach analyses underestimated the true importance of seabirds and their eggs in the diet of rats.

Contaminants

Stable isotopes have also frequently been used to identify contaminant sources in the diet of seabirds. Several recent studies have used stable nitrogen isotope measurements to describe the biomagnification of lipophilic and metal contaminants in marine ecosystems. Jarman *et al.* (1996) showed that in a food web of the Californian Gulf of the Farallones, both organochlorine compounds and mercury were strongly associated with levels of stable isotopes of nitrogen and hence with trophic level, indicating that these contaminants biomagnified in that food web. Further, Atwell *et al.* (1998) found that in an Arctic food web, total mercury levels in bird muscle increased in variance, but not in mean value, with increasing δ^{15}N value. These authors argued that interspecific differences in mercury could have been associated with factors such as elimination of mercury during moult and/or age differences. Due to the importance of identifying intra-specific sources of variation in contaminants, recently, Bearhop *et al.* (2000b) used multivariate analyses to assess the relative contribution of different sources of variation in the feather mercury levels of Great skuas (*Catharacta skua*). As with previous studies, this study demonstrated that trophic status, as indicated by δ^{15}N values, was correlated with mercury concentrations, which also depended on other interacting factors that vary both spatially and temporally. Recent investigations of the seabird community associated with the Northwater Polynya (northern Baffin Bay, Canada/Greenland), has demonstrated the value of using δ^{15}N values as a proxy for trophic level for a variety of contaminants (Fisk *et al.,* 2001). Braune *et al.* (2001a,b) have also demonstrated the utility of retrospective analyses of contaminant changes in the diets of high Arctic seabirds by analysing archived egg material. An important component of this work was to use stable isotope analyses to determine whether or not contaminant level changes were related to dietary changes (see also Hebert *et al.,* 2000).

APPLICATIONS IN THE STUDY OF THE MEDITERRANEAN SEABIRD COMMUNITY

Most previous studies applying the stable isotope technique to the study of seabird ecology have been carried out in the Northern Hemisphere (but see Bocher *et al.,* 2000; Hodum and Hobson, 2000; Thompson *et al.,* 2000; Forero *et al.,* 2002; Forero *et al.,* 2002b) (Table 1). Although some information exists about the spatial variation in δ^{15}N and δ^{13}C values of foodwebs in the Mediterranean, this does not include seabirds (Jennings *et al.,* 1997; Godley *et al.,* 1998). However, the Mediterranean seabird community is especially amenable to applying this technique due to several characteristics.

A characteristic of special relevance to applying stable isotope analysis in the Mediterranean is the high regional heterogeneity of water temperature, salinity, and primary productivity (Zotier *et al.,* 1999), which could result in different isotopic compositions of food webs from different locations, a factor that should be taken into account in any study involving spatial comparisons (Schell *et al.,* 1999).

Both the population sizes and diversity of seabirds are relatively low in the Mediterranean and breeding colonies usually have low numbers of species and individuals (Zotier *et al.*, 1999). However, species composition of colonies, colony size and distribution, are highly variable geographically. The isotopic measurement of these species throughout their ranges in the Mediterranean would contribute a better understanding of their actual population structure.

In the Mediterranean, seabirds mainly from the western region migrate after the breeding season, while eastern populations are mainly sedentary but can disperse far from the breeding colonies. There are several lines of evidence that food conditions during the inter-breeding period could greatly affect breeding success and population dynamics of seabirds (Birkhead and Furness, 1985). To our knowledge, linkages between overwinter diet and breeding ecology have not been investigated for any seabird species in the Mediterranean. Sampling during the breeding season of feathers that were grown during the winter period represents a possible approach that might help to better understand this potential effect.

Applications of the stable isotope technique to address conservation problems are of crucial importance in the Mediterranean seabird community. This area holds several species of special conservation concern, with breeding distributions in some cases restricted to the Mediterranean (Zotier *et al*., 1999). Humans and their activities have been present in the Mediterranean and its islands for a long time and have impacted the distribution and number of seabirds. For example, Martin *et al.* (2000) revealed that the presence of introduced black rats (*Rattus rattus*) can partially explain the current distribution of colonial seabirds in the Mediterranean. Distributions of species such as European storm-petrel (*Hydrobates pelagicus*) or Cory's shearwater (*Calonectris diomedea*) clearly reflects the presence or absence of black rats. In cases where quantification of predation rates and measurements of success of eradication programmes are needed, measurement of stable isotopes in different rat tissues offer an alternative and effective methodology.

Another impact of human activity on seabirds in the Mediterranean area is the dramatic increase of some gull species as a consequence of the use of rubbish dumps. A clear example is the yellow-legged gull (*Larus cachinnans*), which has changed its feeding habits and tends to feed today mostly on terrestrial prey from human refuse (Bosch *et al.,* 2000). Although conventional dietary methods are useful for studies that require a detailed identification of prey choice, biases of this methodology (see references in *Background*) could be avoided by using stable carbon and sulfur ($\delta^{34}S$) isotope analysis where the identification of relative contribution of diet derived from terrestrial rather than marine reservoirs is required (e.g. Hobson, 1987; Hobson *et al.,* 1997).

Shallow seas of the Mediterranean have favoured the development of industrial fishing, which could have two different effects on seabird communities. First, commercial fishing can dramatically deplete fish stocks, particularly those of small pelagic schooling fish. In contrast, scavenging seabird species can benefit from the use of fishery waste, especially from discards of demersal fish that seabirds could not obtain naturally (Oro and Ruiz, 1997). Furthermore, changes in rates of discarding due mainly to fishing moratoriums and the consequent reduction of primary food resources for scavenging species could induce an increase in the rates of predation on other seabirds (e.g. Oro and Martínez-Vilalta, 1994; González-Solís *et al.,* 1997b) and also affect intra- and inter-species resource partitioning. In the Mediterranean, demersal fish constitute the majority of all reported fish discards. However, the degree of exploitation of discard by seabirds is usually difficult to assess. Stable isotope signatures of demersal fish (feeding mainly on benthic invertebrates) and natural food for most of the seabirds in the Mediterranean (pelagic fish, feeding mostly on zooplankton) are expected to be different. Thus, the measurement of stable isotopes in Mediterranean seabirds may offer a tool for evaluating the current dependence of seabirds on discarding and the effect of temporal changes of discarding rates on their population parameters. This approach could be of special interest in species of high conservation concern such as Audouin's gull (*Larus audouinii*), which have recently increased their population in the Western Mediterranean, partly influenced by discard availability (Oro *et al.,* 1997; Oro, 1998), or the Mediterranean shearwater (*Puffinus yelkouan*), which shows a higher dependence on discards than other *Puffinus* species (Oro and Ruiz, 1997). In addition, long-term studies could be carried out by measuring stable isotopes in tissues from museum collections. Such studies might help to explain how important the impact of fisheries has been in the population demographics of seabird populations in the Mediterranean.

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