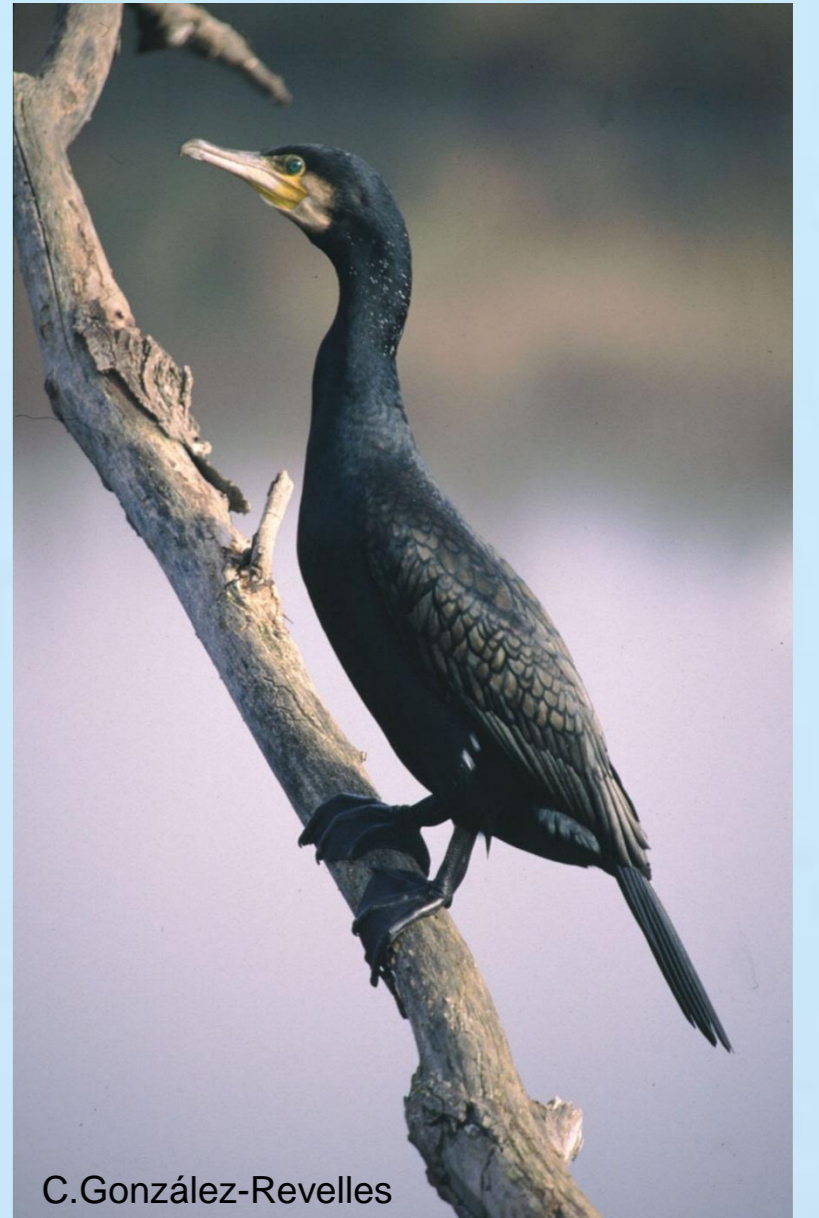


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Introduction

The Great Cormorant *Phalacrocorax carbo sinensis* is a piscivorous species showing an increasing trend over recent decades in Europe. Therefore, the requirement of new feeding areas or the more intensive exploitation of existing ones by wintering birds has been confirmed in many countries like Spain. In view of this, there is a need to manage the potential conflict between birds and local fisheries. Many studies have examined it, but the results are not generalizable, a different type and degree of impact being inferred depending on the specific situation. This has not impeded the removal of the Great Cormorant from the Spanish list of specially protected bird species.

Diet studies are a fundamental tool to ensure the compatibility of potentially conflictive species with fisheries. Stable isotopes provide a non-destructive technique that can address many aspects of the trophic ecology of birds. C informs about the source of carbon (marine vs freshwater) entering the food web, and N about the trophic position of organisms. On the basis of classical ¹³C and ¹⁵N stable isotopes analysis in feathers we combine different specific techniques to determine the potential trophic niches occupied by *P. carbo sinensis* along a marine-continental gradient (including the transitional waters of the Mar Menor Lagoon). **Specific objectives are:**

- [1] to evaluate if the type of feather collected influences the isotopic signal,
- [2] to analyze the food source and inferred movement patterns of wintering groups along the marine-continental gradient
- [3] to test the suitability of methods commonly used at the scale of species assemblage for assessing, at the population scale, differences in trophic niche and directional changes in the isotopic signature of N and C along the gradient studied; and
- [4] to discuss the implications of the feeding distribution of *P. carbo* for traditional fishing on an internationally important wetland like the Mar Menor Lagoon.

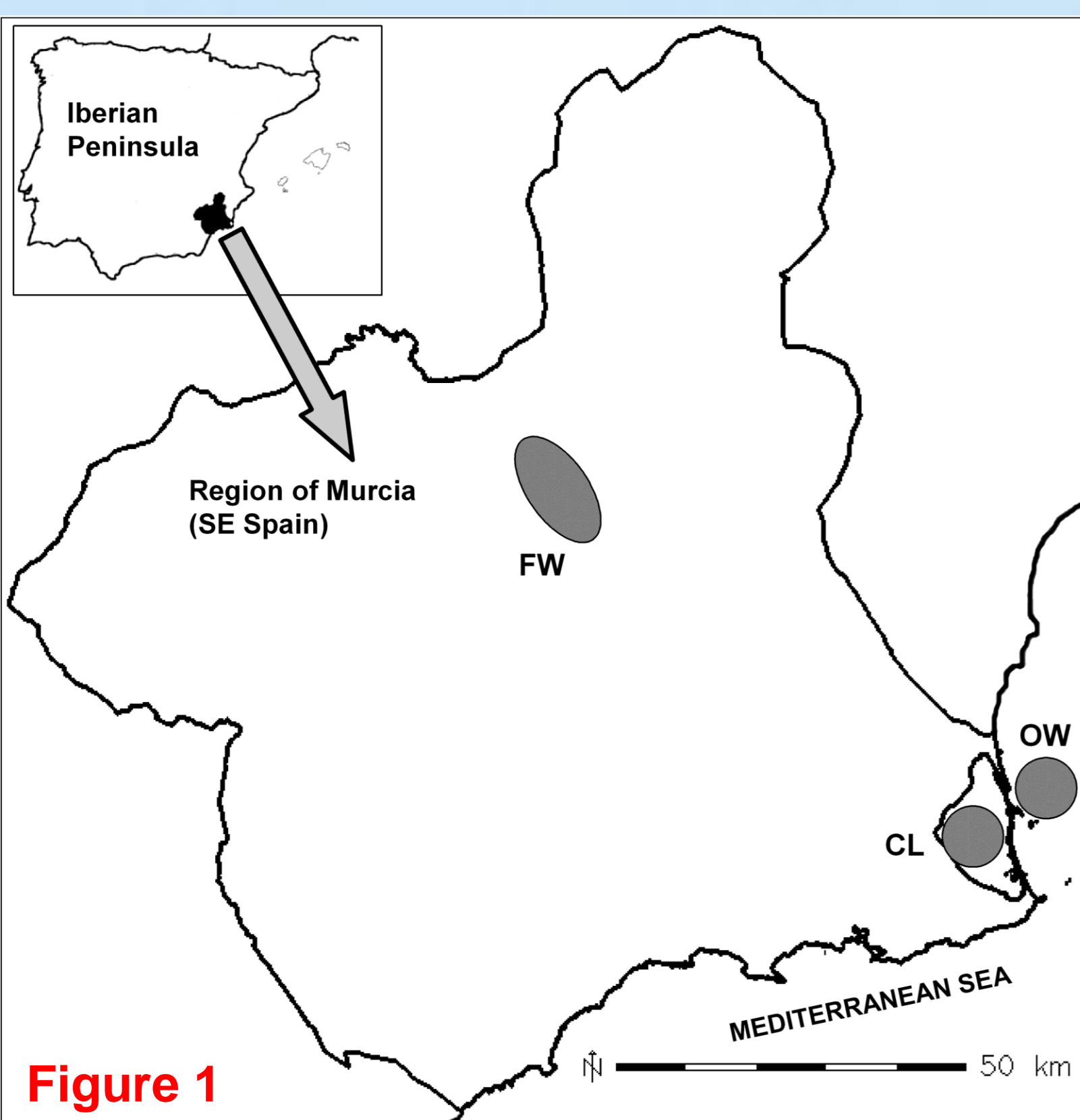


Figure 1



Methods

Study area

We selected three locations in Murcia Region, southeastern Spain (Figure 1) representing a marine-continental gradient (FW=freshwater, CL=transitional water and OW=open sea water)

Sample collection and preparation

From January 2008 to April 2009, five types of feathers (growing or freshly grown ones) were obtained from different parts of adult birds (primaries, secondaries and tail). Feathers were collected from recently dead birds (drowned in fish nets) or under roosting places.

To obtain the isotopic signature of potential prey bogue (*Boops boops*) and mackerel (*Trachurus sp*) were selected in OW, grey mullet (*Mugil sp*) in CL, and barbel (*Barbus sclateri*) in FW.

Feathers were washed with distilled water and dried at 60°C during 24 h. 1 mg sample was taken from each one, sonicated in distilled water for 30 minutes and then in diethyl ether (stabilized with BHT) for 30 minutes to remove any possible contaminants.

From each fish, we took 1 mg of abdominal white muscle tissue and froze it at -20°C until analysis of stable isotopes. Later, samples were freeze-dried, ground to a fine powder and homogenized.



The population groups of Great cormorants studied were linked to typical habitats present in three areas representing a marine-continental gradient. (A) Freshwater ecosystem (Segura river), (B) coastal lagoon (Mar Menor and its islands) and (C) open sea (birds moving to Farallón islet in the nearby Mediterranean)

Data analysis

The carbon and nitrogen isotope ratios of the samples were measured by Servicios de Apoyo a Investigación (Universidade da Coruña, Spain) following equation:

$$\delta^{13}\text{C} \text{ or } \delta^{15}\text{N} (\text{‰}) = [(R_{\text{sample}}/R_{\text{standard}}) - 1] \times 1000$$

Differences in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ signal between sites (for both cormorants and fishes), and between types of feathers, were checked. Also a Permanova analysis was performed, looking for differences in trophic niche breadth among the three groups of cormorants.

Layman *et al.* (2007) metrics were calculated to describe the shape of the trophic niche of each group of cormorants. To analyze the directional change of the isotopic signature of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ along the marine-continental gradient, we applied the circular statistics proposed by Schmidt *et al.* 2007.

Results

No significant differences in isotopic signals were detected between the types of feathers. As expected, significant differences were found for both isotopes between sampling areas. Isotopic signatures for both cormorants and fishes are presented in Table 1, and their representation in a bi-plot graph in Figure 2.

Permanova analysis showed significant differences between CL and FW cormorants, and between CL and OW cormorants, while not between OW and FW cormorants.

Layman's metrics indicate that OW and FW cormorants display larger $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ ranges than CL ones (Table 2). Total Area (TA) of the polygons represented in Figure 3, shows how OW and FW cormorants have a greater trophic niche breadth than CL ones, the latter constituting the most compact group in terms of trophic breadth and similarity between individuals.

The circular statistics show how the displacement of cormorants from FW or OW to CL (Figure 4) causes a decrease in the $\delta^{15}\text{N}$ signature and an increase in $\delta^{13}\text{C}$ signature (more oceanic).

Metric	OW	CL	FW
$\delta^{13}\text{C}$ range	12.25	4.9	9.9
$\delta^{15}\text{N}$ range	4.05	2.1	5.7
TA	25.36	5.2	37.4812
CD	3.57	1.96	3.48
NND	1.439	0.4	1.59
SDNND	0.5809	0.37	1.24

Table 2

Table 1	n	$\delta^{13}\text{C}$ (‰)		$\delta^{15}\text{N}$ (‰)	
		Mean ± SD	Range	Mean ± SD	Range
Marine cormorants	10	-20.53 ± 4.07	-28 to -15.7	16.78 ± 1.38	14.6 to 18.7
Lagoon cormorants	10	-15.66 ± 2.1	-17.8 to -12.9	13.70 ± 0.56	13 to 15.1
Freshwater cormorants	10	-21.36 ± 3.25	-24.9 to -15	17.45 ± 2.27	14.9 to 20.6
Marine fishes	8	-18.34 ± 0.17	-18.65 to -8.1	9.68 ± 0.35	9.3 to 10.2
Lagoon fishes	8	-11.00 ± 0.53	-11.65 to -10.3	8.41 ± 1.88	6 to 12
Freshwater fishes	8	-23.55 ± 0.62	-24.3 to -22.6	13.19 ± 1.26	10.5 to 14.8

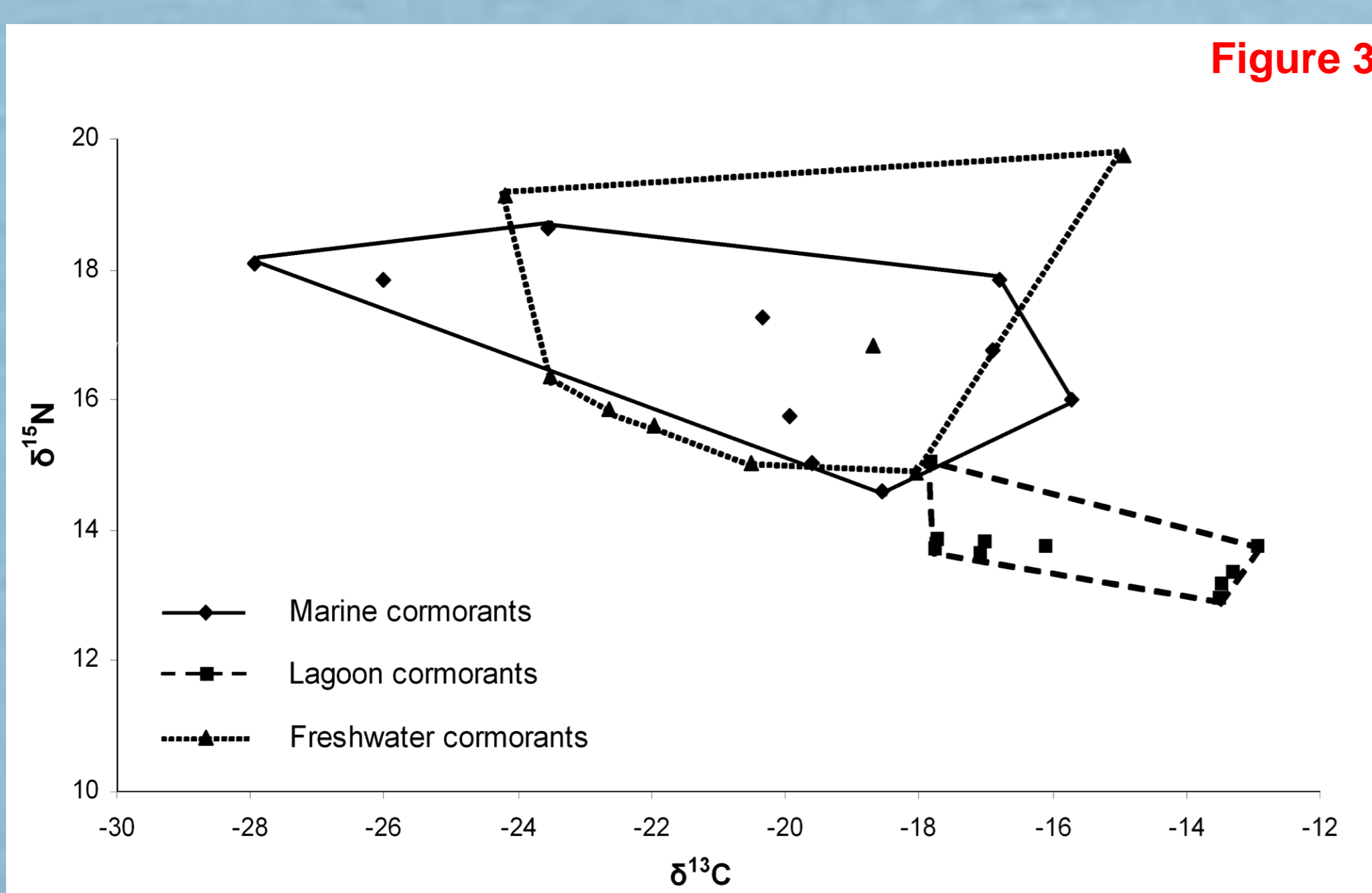


Figure 3

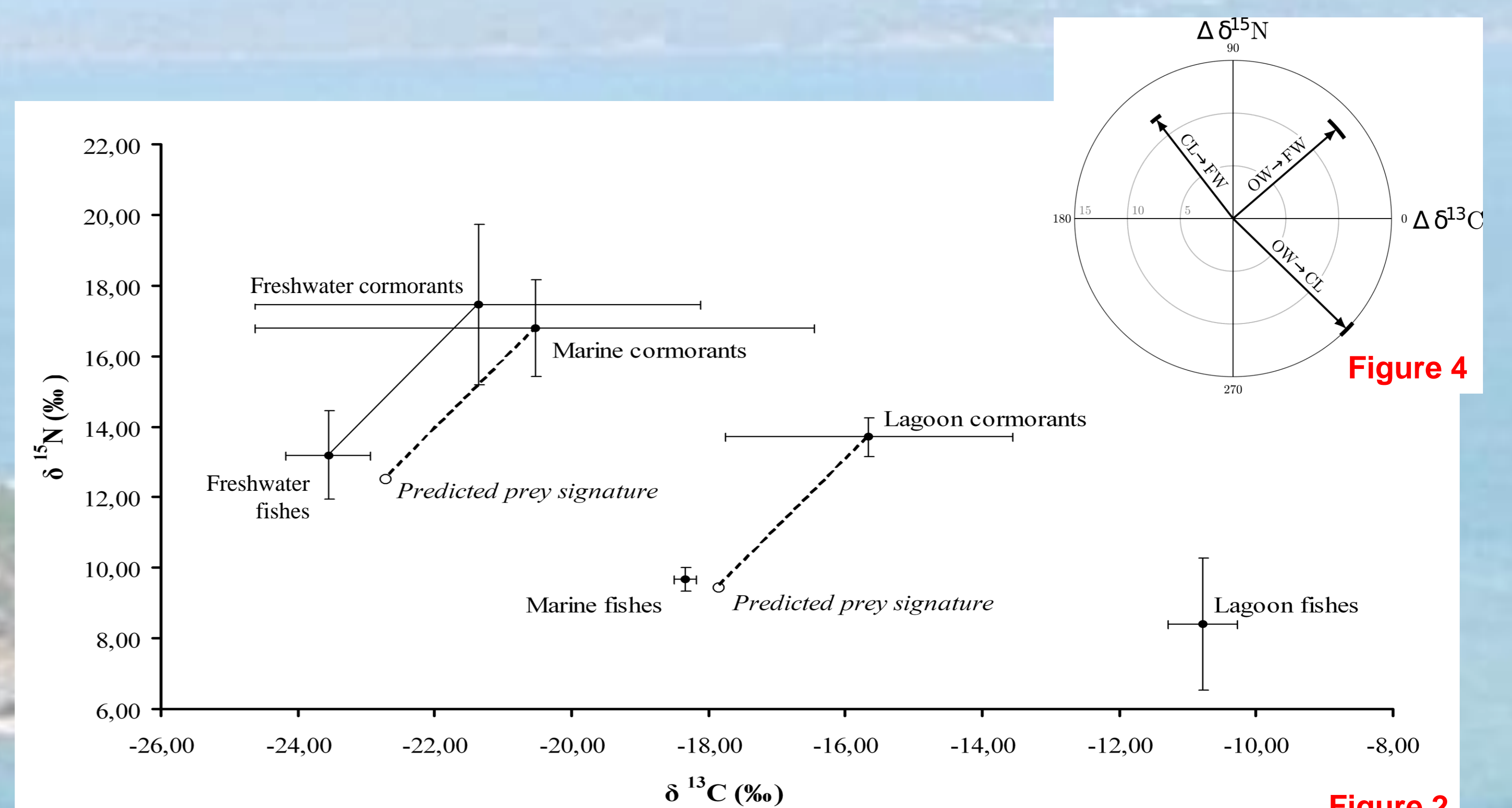


Figure 2

Discussion

The mean isotopic fractionation in FW bodies (from the barbel to the feathers) described in this study, suggests that the diet of FW cormorants has a terrestrial organic carbon origin and is based almost exclusively on barbel. OW cormorants (initially taken as a reference of marine diet) seem to have a mixed diet, mainly of continental origin but probably shared with some marine sources. Applying the same fractionation factor to the signal of CL cormorants, we get an isotopic signature for their predicted prey close to that obtained for *Trachurus sp.* and *Boops boops*, which are not present in the lagoon, so CL cormorants must leave to open sea regularly to hunt and return to roost in lagoon islands.

The $\delta^{13}\text{C}$ signature of grey mullets probably has an oceanic origin. If cormorants do not forage exclusively in transitional waters (as reflects their mixed diet of oceanic and lagoon fishes) the conflict with local lagoon fisheries (i.e. *Mugil sp.*) would be reduced.

Two separate population groups (FW and OW) apparently forage in freshwater bodies (following Layman's metrics interpretation, there is an "overlap" of their trophic niche) and the third group (CL), which moves around the coastal lagoon, shows a mixed diet. In addition, FW and OW cormorants have a higher internal variability in food sources than CL ones (CR and TA results). This is consistent with the well-known daily mobility of cormorants between roosting and feeding habitats (up to 60 km). It is probable that some individuals belonging to FW or OW also foraged in coastal areas. Circular statistics and directions of change in isotopic signatures between areas support this interpretation. The application of Layman (2007) and Schmidt (2007) techniques on a population scale seem to produce consistent and meaningful results.

The dependence of wintering *P. carbo* on transitional systems like the Mar Menor lagoon as feeding areas seems partial. At a regional scale, the growing population of the species looks equally dependent on the greater availability of inland, highly productive water bodies (river reservoirs, large irrigation ponds). The lagoon can be interpreted as an "information centre" (roost) for a metapopulation that would split into several feeding groups moving along the marine-continental gradient, and hence spreading their predation pressure.

Therefore, the impact of *P. carbo* on commercial fish species in the lagoon would be less important than that expected from the numbers recorded. The set of methods and metrics used in this work are an example of non-invasive tools that can be used to assess the real impact of the species on fisheries, before downgrading its protection or implementing control measures.

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Schmidt, S.N., Olden, J.D., Solomon, C.T., Vander-Zanden, M.J., 2007. Quantitative approaches to the analysis of stable isotope food web data. *Ecology*, 88, 2793-2802.