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## Possible link between prey quality, condition and growth of juvenile hake (*Merluccius merluccius*) in the Gulf of Lions (NW Mediterranean)

## Relation possible entre la qualité des proies, la condition et la croissance des jeunes merlus (*Merluccius merluccius*) dans le golfe du Lion (NO Méditerranée).

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### Abstract:

The energy value of the main prey and the growth of juvenile hake in the Gulf of Lions were analysed to determine whether fish condition could be linked to difference in prey quality and resulted in growth difference. Large differences in energy content (15.6 to 23.6 kJ g<sup>-1</sup> DW) among prey types were recorded with low values found in amphipods and natantid shrimps, intermediate values in euphausiids and mysids, and high values in fish. High condition in juvenile hake was linked to the consumption of prey with high energy content. However, no difference in fish growth was observed by the analysis of otolith micro-increments.

### Résumé

La valeur énergétique des proies principales ainsi que la croissance des jeunes merlus dans le golfe du Lion ont été analysées afin de déterminer si la condition pouvait être liée à une différence d'énergie apportée par les proies et se traduisait par une différence de croissance en longueur. Les contenus énergétiques diffèrent entre groupes de proies (15,6 à 23,6 kJ g<sup>-1</sup> DW), avec des valeurs faibles pour les amphipodes et les crevettes Natantia, intermédiaires pour les euphausiacés et les mysidacés, et fortes pour les poissons. Une condition élevée des juvéniles de merlus a été reliée à la consommation des proies les plus énergétiques. Cependant, aucune différence de croissance en longueur n'a été observée par l'analyse des microstries de leurs otolithes.

**Keywords:** Merlucciidae ; *Merluccius merluccius* ; Hake ; Mediterranean Sea ; Energy ; Condition ; Otolith microstructure

## 1. Introduction

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The European hake (*Merluccius merluccius* Linnaeus, 1758) is an economically important fish in the NW Mediterranean (Oliver and Massuti, 1995) and the major demersal species in the fishery landings of the Gulf of Lions (GoL) with a mean annual catch of ~2400 tons (Aldebert *et al.*, 1993; Jadaud *et al.*, 2006). Hake recruitment occurs all year round in the NW Mediterranean (Maynou *et al.*, 2003) with a peak in spring in the GoL (Recasens *et al.*, 1998), and juveniles are mainly concentrated on the continental shelf (Orsi-Relini *et al.*, 2002). The recorded large interannual and spatial fluctuations in hake recruitment are explained by differences in environmental conditions (Lloret *et al.*, 2001; Olivar *et al.*, 2003; Bartolino *et al.*, 2008) or food supply (Maynou *et al.*, 2003). Other factors such as habitat quality, food source composition and intra- and inter-specific competition may influence hake condition, growth and abundance after benthic settlement (Lloret *et al.*, 2001; Maynou *et al.*, 2003; Hidalgo *et al.*, 2008). In ecological studies, fish condition (relative weight-length factor) is used as an indicator of habitat quality, food source availability and energetic reserves (Lloret *et al.*, 2002, 2005; Shulman *et al.*, 2005). The abundance and availability, but also the quality of the food, are critical determinants of reproductive success and population dynamics of marine top predators, like sea lions and seabirds (Österblom *et al.*, 2008). The question of prey quality is thus becoming an important parameter in ecosystem modelling and management (Spitz *et al.*, 2010). In the GoL, Ferraton *et al.* (2007) observe a decline in the relative condition factor (Le Cren, 1951) and abundance of juvenile hake (<20 cm TL) on the continental shelf in 2003 compared to 2002 and relate this pattern to differences in diet composition. They hypothesize that the main prey consumed in 2002 and 2003 may differ in energetic content, possibly resulting in a difference in fish condition, growth and survival.

To validate or rebut the hypothesis that prey quality can influence juvenile hake condition, this study was performed to determine whether the main prey types consumed by juvenile hake differed in energy content, and whether difference in hake condition was associated with a difference in somatic growth (= growth in length) between 2002 and 2003. The main questions addressed in the present paper were (1) did the quality of the different prey types consumed by juvenile hake differ, and (2) did the somatic growth of juvenile hake vary between 2002 and 2003? To answer the first question, the energetic value of the main prey types was determined, and related to juvenile hake diet and condition. The second question was addressed through the analysis of otolith microstructure for determining juvenile hake age and growth.

## 2. Materials and methods

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### 2.1. Study site and sampling

Sampling was conducted in the GoL in two zones located at 70-150 m depth on the continental shelf at similar sites in 2002 and 2003 (Fig. 1). Zone 1 was located to the east, off the Rhone River delta, and Zone 2 to the west, in the middle of the GoL. Juvenile hake (<20 cm total length = TL, Mellon-Duval *et al.*, 2009) were sampled in June 2002 and 2003 by diurnal standardized trawling performed during the annual international MEDITS surveys (Bertrand *et al.*, 1998). Diet and condition of juvenile hake were studied by Ferraton *et al.* (2007) in both years. These authors observed that diet and condition differences between years were the highest in the 10-14 cm TL size class (Table I). The main fish prey of juvenile hake, small (< 10 cm = TL) anchovy (*Engraulis encrasicolus*), sardine (*Sardina pilchardus*), poor cod (*Trisopterus minutus*) and gobiids (*Gobius niger* and other species), were collected by trawling, along with juvenile hake. Small crustaceans living in the benthic boundary water layer, which are the dominant prey of juvenile hake (Table I), were collected in the same

zones and depth range with a suprabenthic sledge equipped with zooplankton nets (0.5 mm mesh size) (Sorbe, 1999). Crustaceans were sorted into major groups (mysids, euphausiids, amphipods, natantid shrimps) and kept frozen independently at -20°C before analysis. For determining age and growth of juvenile hake, a sub-sample of 74 individuals 10-20 cm TL was selected, 35 in 2002 and 39 in 2003 equitably distributed between the two zones. A two-way ANOVA revealed no difference in mean size of the selected fish between year ( $F = 0.600$ ,  $p = 0.442$ ) and site ( $F = 0.084$ ,  $p = 0.773$ ) and no interaction between the two factors, allowing interannual comparison.

## 2.2. Prey energetic content

Prey energetic content was estimated from the heat provided by the complete combustion of 0.1 g of homogenized freeze dried sample in an adiabatic calorimeter (IKA calorimeter C 4000). Benzoic acid was used as standard and the caloric content of prey was reported in kilo Joules per gram of dry weight ( $\text{kJ g}^{-1}$  DW). The caloric content of juvenile hake preys was quantified on three different pools of individuals (5-10 individuals per pool depending on species size) for each of the four crustacean types (mysids, euphausiids, amphipods and shrimps), whereas analyses were performed on individuals for fish prey (7 to 10 fish per species). The main suprabenthic crustaceans analysed in each group, *Westwoodilla* spp. for amphipods, *Leptomysis* spp. and *Siriella* spp. for mysids, *Meganyctiphanes norvegica* and *Nyctiphanes couchi* for euphausiids and *Philocheras* spp. for natantia, were also the main prey found in juvenile hake stomach contents (Ferraton *et al.*, 2007). The size ranges of fish analysed for caloric content corresponded to those of prey eaten by juvenile hake, i.e. 3-6 cm TL for *Gobius niger*, 5-8 cm TL for *Trisoperus minutus*, 5-10 cm TL for *Engraulis encrasicolus* and *Sardina pilchardus*.

## 2.3. Otolith preparation

Growth and age determination in European hake is well studied (e.g. Morales-Nin *et al.*, 1998; de Pontual *et al.*, 2006; Mellon-Duval *et al.*, 2009) and otolith microstructure provides a useful tool for age determination of juvenile hake (Morales-Nin and Aldebert, 1997; Arneri and Morales-Nin, 2000; Belcari *et al.*, 2006). Sagittae of juvenile hake were used for age determination. The daily formation of increments on hake otolith was validated indirectly (Arneri and Morales-Nin, 2000; Belcari *et al.*, 2006) as well as directly (Morales-Nin *et al.*, 2005). The right sagittal otoliths ( $N = 74$ ) were mounted on slides with an epoxy resin and ground on sagittal plane on both sides until the central zone (CZ) was readable. They were examined using a light microscope connected to a video camera and an image-analysis system (TNPC software). Otolith radius (OR) was measured ( $\mu\text{m}$ ) from the centre of the nucleus to the edge of the otolith and increments (DGI) counted. Increments were counted twice and the mean of the two readings was considered as the individual's age when differing by less than 10%. A total of 57 sagittal otolith sections could be read, 28 in 2002 and 29 in 2003.

## 2.4. Data analysis

Linear regressions were used to determine the relationships between OR and age estimated by DGI count, OR and fish length, and fish length and DGI count in 2002 and 2003. Linear regressions were used as they best fitted the data and allowed comparison of slopes and elevations for testing inter-annual fish growth. Differences in slope and elevation between years were tested by ANCOVA with size as covariate, followed by appropriate *t*-test (Zar, 1999). One-way ANOVA was used to test differences in mean caloric value of prey.

### 3. Results

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#### 3.1. Prey caloric value

Significant differences in mean energetic value were observed among the different prey types consumed by juvenile hake (Tab. II). Fish were the most highly caloric prey (20.8-23.6 kJ g<sup>-1</sup> DW) with *E. encrasicolus* and *S. pilchardus* presenting a significantly higher caloric value than *G. niger* and *T. minutus*. Among crustaceans, mysids were the most highly energetic prey (20.6 kJ g<sup>-1</sup> DW) followed by euphausiids (18.4 kJ g<sup>-1</sup> DW), whereas shrimps and amphipods displayed significantly lower and similar energetic values (15.6 – 15.9 kJ g<sup>-1</sup> DW). Juvenile hake ingested thus preys with higher energy content in 2002 than 2003 at both sites.

#### 3.2. Juvenile hake age and growth

DGI counts in juvenile hake sagittal otoliths ranged from 149 to 407 in 2002 and from 124 to 325 in 2003. The radius of the CZ did not differ between years ( $F = 3.651$ ,  $p > 0.05$ ) and zones ( $F = 0.033$ ,  $p > 0.05$ ) and averaged  $238 \pm 41 \mu\text{m}$  for a mean increment width of  $5.0 \pm 0.9 \mu\text{m}$ . OR was significantly related to fish age in 2002 and 2003 ( $\text{OR}_{2002} = 0.004 \text{ DGI} + 0.466$ ,  $R^2 = 0.79$ ,  $p < 0.001$ ;  $\text{OR}_{2003} = 0.004 \text{ DGI} + 0.560$ ,  $R^2 = 0.82$ ,  $p < 0.001$ ) and the relationships did not differ in slope and elevation (appropriate  $t$ -test  $p > 0.05$ ) between years. OR was also significantly correlated to fish length in 2002 ( $\text{OR} = 0.097 \text{ TL} - 0.041$ ,  $R^2 = 0.94$ ,  $p < 0.001$ ) and 2003 ( $\text{OR} = 0.082 \text{ TL} + 0.206$ ,  $R^2 = 0.86$ ,  $p < 0.001$ ). Fish age was significantly correlated to fish length (Fig. 2) and did not differ between 2002 and 2003 years (appropriate  $t$ -test  $p > 0.05$  for slope and elevation). It could be thus inferred that the somatic growth of juvenile hake did not differ between the two years.

### 4. Discussion

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#### 4.1. Prey energetic content and hake condition

In ecology, fish condition is often used to estimate the effect of environmental factors at the population level. Difference in condition reflects variations in physical and biological environmental conditions, food availability, growth and reproduction (Shulman and Love, 1999; Lloret *et al.*, 2002; Hidalgo *et al.*, 2008). In the Gulf of Lions, the relative condition factor  $K_n$  (Le Cren, 1951) of juvenile hake located at 70-150 m depth on the continental shelf was significantly higher in 2002 ( $K_n = 1.04 \pm 0.01$ ) than in 2003 ( $K_n = 0.96 \pm 0.01$ ) ( $p < 0.001$ ) (Table I, Ferraton *et al.*, 2007). In 2002, juvenile hake fed mainly on mysids and euphausiids (prey of high energetic content) in Zone 1 and 2 respectively, while they preyed mainly on shrimps (prey of low caloric content) in the two zones in 2003 (Fig. 1). Thus, the decrease in condition factor observed in juvenile hake in 2003 compared to 2002 could be related to the consumption of less energetic prey. Moreover, Ferraton *et al.* (2007) observe that juvenile hake located in shallow waters (30-50 m) which prey mainly on fish in 2002 and 2003, present similar high condition factors ( $K_n = 1.03 \pm 0.01$ ) both years. Fish appeared to be the most energetic prey consumed by juvenile hake in the Gulf of Lions. Spitz *et al.* (2010) place also clupeids (sardine) and engraulids (anchovy) in the high quality of forage species in the Gulf of Biscay. Thus, the consumption of high energetic prey such as fish in shallow waters and small energetic crustaceans like euphausiids and mysids in deeper waters could provides more energy to juvenile hake than shrimps and amphipods. Better fed juveniles can then reach a higher condition factor, which means a heavier weight for the same length.

Similar differences in energetic values among crustacean groups are found in the literature, with low energy content in amphipods and shrimps, and high energy content in mysids and euphausiids (Wolowicz and Szaniawska, 1986; Davies, 1993; Torres *et al.*, 1994). Davies (1993) records mean values of 5.2 kJ g<sup>-1</sup> DW in shrimps, compared to 22.0 kJ g<sup>-1</sup> DW in euphausiids and 23.6 kJ g<sup>-1</sup> DW in mysids. These values are close to those recorded for the same groups in the GoL (Tab. II). Such a difference in energetic content between crustacean groups is likely linked to their lipid content, as mesopelagic mysids present a higher energy value and contain higher proportions of lipids (35-48%) than *Natantia* (5-12 %) (Donnelly *et al.*, 1993). The energy content of prey consumed is thus important to better understand the spatial and temporal differences in fish condition. Fish eating an energetic diet have the advantage for getting fatter than fish feeding on a similar biomass ration but with a low caloric content (Wootton, 1990). Paul *et al.* (1990) demonstrate that pollock converted the energy content of crustacean and fish tissue with similar efficiency, and that growth in weight was related to the energy content of food. The poorest growth in weight is shown by pollock fed amphipods, the prey with the lowest energy content, while the best growth is obtained by pollock fed herring, which have the highest energy content. These results are in accordance with those found for the European hake in the present study, i.e. a better condition associated with more energetic prey. We are aware that a delay exists between food intake and food assimilation, but difference in caloric content among prey types, both in this study and in the literature, is consistent with our results. To accurately relate juvenile hake condition to prey quality would necessitate to perform experimental studies with diet differing in caloric content, as those conducted by Paul *et al.* (1990)

#### 4.2. Growth

Despite the rather low number of fish analysed for growth, the high significance ( $p < 0.001$ ) of the relationships obtained gives us grounds for confidence with regard to our results. Juvenile hake did not display any difference of growth in length (= somatic growth) in the GoL between 2002 and 2003 despite a difference of growth in weight reflected in a higher condition factor. These results indicated that the somatic growth was not affected by the consumption of prey of different energetic quality, but that fish were slender in 2003 compared to 2002. It is generally admitted that hake may withstand food resource variations due to its vertical migration (Bozzano *et al.*, 2005) and opportunistic predator activity (Bozzano *et al.*, 1997; Hidalgo *et al.*, 2008). In the Balearic Islands, Cartes *et al.* (2009) observe that juvenile hake located at the shelf-slope break feed on mesopelagic species (particularly euphausiids) located deeper than hake during the day. This was not the case on the continental shelf of the GoL at 70-150 m where juvenile hake and their suprabenthic crustacean prey occurred within the same depth range (J.C. Sorbe, unpublished data). Hidalgo *et al.* (2008) record a higher condition in hake in the north than in the south of the Balearic Islands and related that to higher food resources and consumption in the north. The present study indicated that more energetic prey could result in a higher condition (= weight) of juvenile hake without any difference in growth in length. Our results suggested then that the composition and quality of food resources were important in explaining juvenile hake condition and not only the total abundance of prey, as it was indicated for marine mammals in the Gulf of Biscay (Spitz *et al.*, 2010). The importance of the quality of food on population dynamics of marine top predators, evidenced for sea lions and seabirds (see Österblom *et al.*, 2008 for review), has also to be taken into consideration for explaining population dynamics of fishes.

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## Tables

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Table 1. Diet of juvenile hake (10-14 cm) in deep water (70-150 m) expressed in percentage of relative importance (%IRI) and relative condition factor  $K_n$  (Le Cren, 1951) in the two zones investigated in the Gulf of Lions in 2002 and 2003.  $IRI = \%FO \times (\%N + \%W)$  where FO = frequency of occurrence, N = number of prey, W = dry weight of prey. No Fish = number of individuals analyzed with food in stomach, Others = other crustaceans and polychaetes (modified from Ferraton et al. 2007).

Year	Zone1		Zone 2	
	2002	2003	2002	2003
No Fish	150	26	74	24
Mysids	83.8	4.6	1.5	4.5
Euphausiids	0.0	0.0	74.0	0.0
Amphipods	0.2	4.4	0.1	1.5
Shrimps	5.7	82.3	6.1	76.0
Fishes	9.9	6.0	7.3	17.5
Others	0.4	2.7	11.0	0.5
Condition factor $K_n$	1.04	0.96	1.04	0.96

Table 2. - Mean ( $\pm$  SD) energy density in wet weight ( $\text{kJ g}^{-1}$  WW) and dry weight ( $\text{kJ g}^{-1}$  DW) of the main prey types of juvenile *Merluccius merluccius* in the Gulf of Lions. N = pools of individuals for crustacean types; individuals for fish. Similar letters indicate means not statistically different on DW values ( $p > 0.05$ )

Prey type	N	Energy ( $\text{kJ g}^{-1}$ WW)	Energy ( $\text{kJ g}^{-1}$ DW)	Post-hoc test
<b>Crustaceans</b>				
Amphipods	3	$8.72 \pm 0.65$	$15.61 \pm 1.15$	a
Natantia	3	$8.78 \pm 0.15$	$15.99 \pm 0.27$	a
Euphausiids	3	$10.64 \pm 0.18$	$18.41 \pm 0.31$	b
Mysids	3	$11.52 \pm 0.26$	$20.61 \pm 0.33$	c
<b>Fishes</b>				
<i>Trisoperus minutus</i>	8	$11.92 \pm 0.75$	$20.85 \pm 1.31$	c
<i>Gobius niger</i>	10	$12.02 \pm 0.38$	$21.20 \pm 0.67$	c
<i>Engraulis encrasicolus</i>	7	$12.81 \pm 0.66$	$22.20 \pm 1.13$	c d
<i>Sardina pilchardus</i>	10	$14.14 \pm 1.49$	$23.61 \pm 2.48$	d

## Figures

Figure 1. - Location of the two zones investigated in the Gulf of Lions.

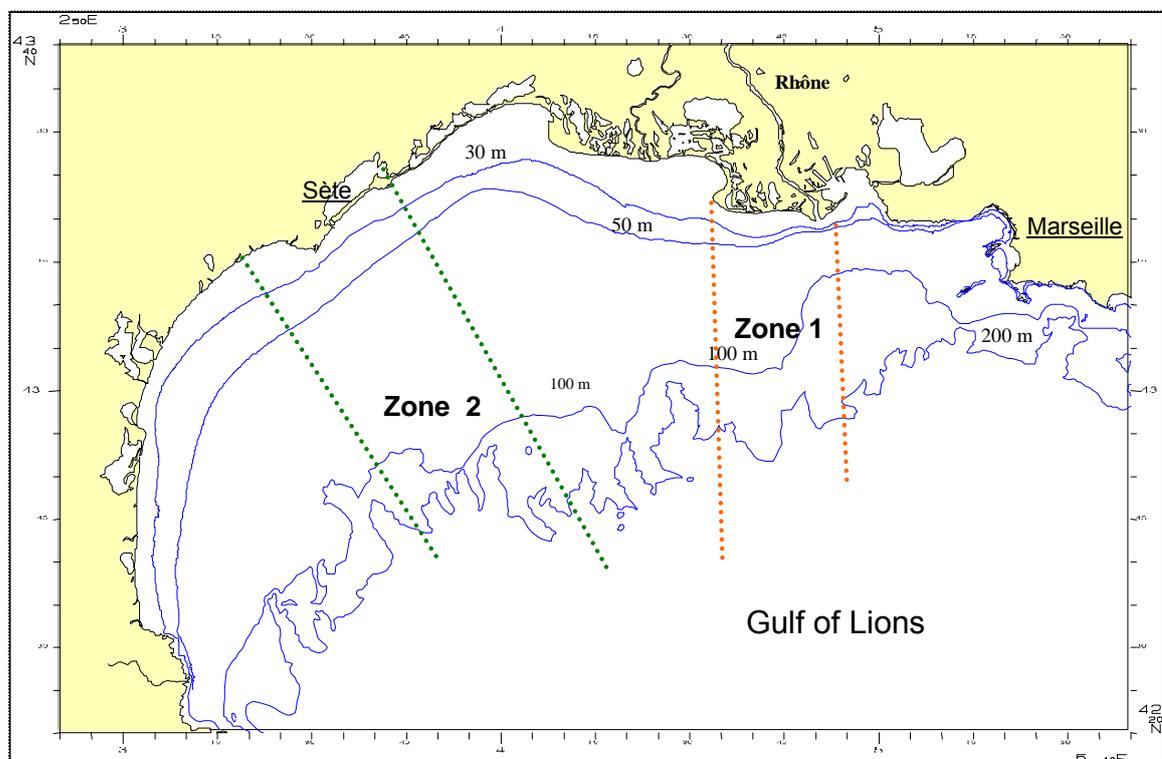


Figure 2. - Linear relationship between fish length (TL cm) and age (days) estimated from the daily growth increments (DGI) of juvenile hake in 2002 (black diamonds and solid line) and 2003 (white squares and dotted line).

