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Contrasting years in the Gironde estuary (Bay of Biscay, NE Atlantic) springtime outflow and consequences for zooplankton pyruvate kinase activity and the nutritional condition of anchovy larvae: an early view

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Abstract:

A major spawning ground of the European anchovy (*Engraulis encrasicolus*) population in the Bay of Biscay is located near the mouth of the Gironde estuary. Variations in the river discharge affect the supply of nutrients to the coastal marine area encompassing anchovy spawning grounds. Two "Pegase" cruises (May–June 1997 and 1998) observed two contrasting situations: with and without a superficial layer of less saline water. Two new nutritional indices were used to assess potential effects (1) on carbohydrate assimilation rates by zooplankton estimated via pyruvate kinase (PK) activity measurements and (2) on nutritional condition of the anchovy larvae measured with the DNA/C index. Both indices indicated a much poorer situation in 1997 compared to 1998. Results of this study implicate river outflow rate as a potential factor influencing larval anchovy condition and perhaps to some extent recruitment variability.

Keywords: anchovy larvae; Bay of Biscay; DNA/C index; pyruvate kinase; river outflow

Introduction

Unlike other species of anchovy living in different parts of the world's oceans, the population dynamics of the European anchovy (*Engraulis encrasicolus*) do not appear to be controlled predominantly by a major physical process, such as the strength and extent of upwelling along western coasts of continents (*E. mordax* in California, *E. ringens* in Peru, *E. encrasicolus*, previously known as *E. capensis*, in Namibia and South Africa). The spawning region of the stock exploited in the Bay of Biscay (NE Atlantic) by the French and Spanish fleets is subject to many extrinsic drivers. It is a patchwork of different systems including: 1) coastal systems influenced either by river plumes or local upwellings caused by special wind regimes and 2) "oceanic" systems along the shelf break where deep water may be upwelled or large eddies may form. Run-off from large rivers such as the Gironde (Fig. 1) may particularly influence the supply of nutrients to these coastal and oceanic systems. Moreover, large interannual fluctuations may occur in the discharge rates.

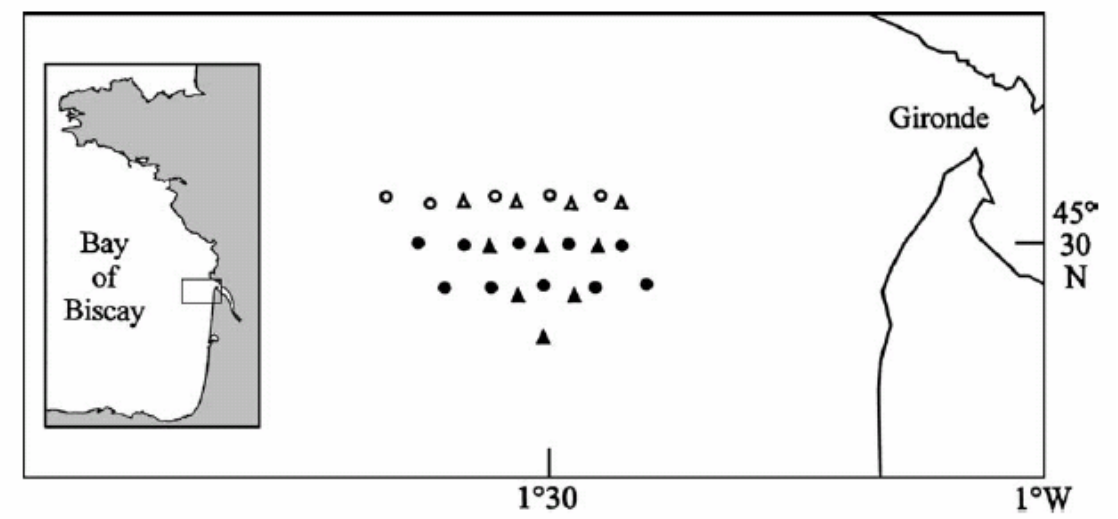

Fig.1 Location of mesozooplankton sampling stations (circles for 1997, triangles for 1998) near the mouth of Gironde in the Bay of Biscay (insert). Open symbols indicate stations selected for salinity sections shown in Fig. 3.

In 1997 and 1998, two Pegase ("Pélagiques Gascogne") cruises were carried out between May-June on the RV Thalassa. These cruises coincided with a period of low (1997) and high (1998) river discharge. During these cruises, estimates of carbohydrate (essentially synthesized by primary producers) assimilation rates by mesozooplankton were obtained via pyruvate kinase (PK) activity analysis (Bergeron and Herbland, 2001) and estimates of individual larval anchovy nutritional condition were made using the DNA/C index (Bergeron *et al*., 1991; Bergeron *et al*., 1997). Results of an early field application were recently presented by Bergeron (2000). The potential effects of different rates of river discharge could be assessed using these two new nutritional indices.

Material and methods

Portions of the Pegase 97 (24-26 May) and 98 (10-13 June) cruises were devoted to sampling sites near the mouth of the Gironde in the estuarine plume (Fig. 1).

At each fixed station,

- temperature and salinity vertical variations were recorded with a CTD (Sea Bird SBE 19);
- water samples were collected with 5 Niskin bottles at standard depths (0, 10, 20, 30 and 40 m) in order to estimate nutrients concentrations: immediately frozen at – 30 °C, these samples were thereafter entrusted to collaborators (see the "Acknowledgements" section) for later analysis in laboratory, according to the well-known method of Strickland and Parsons (1972);
- a mesozooplankton sample was collected by a 1 knot vertical tow of a WP2 net (200µm mesh size) from bottom to surface, following two similar (or, at least, centred on the same geographic point, i.e. 45°30 N and 1°30 W) grids of stations (15 in 1997, 10 in 1998; cf. Fig. 1).

Sieving the samples through 5 mm mesh eliminated macrozooplankton. The samples were ground in iced distilled water with a Polytron and immediately frozen in liquid nitrogen ; they were thereafter stored at –90°C until analysis in the laboratory. After thawing, the crude extract was homogenised with a Potter-Elvehjem and centrifuged (10 min at 4000 rev min^{-1}, 3°C). An aliquot of 200 µl of the supernatant fluid was used for enzyme assay or protein determination. Pyruvate kinase (PK) activity was estimated according to the method of Bucher and Pfleiderer (1955), in the conditions specified by Bergeron and Herbland (2001). Protein (for determination of specific activities) was estimated by the method of Lowry *et al.* (1951) with bovine serum albumin as standard.

Between fixed stations, anchovy larvae were collected using double oblique, 2 knots tows between bottom and surface with a "carré net" (1 m^2 mouth opening; 380 µm mesh size). Collected larvae had to be shared for different purposes, especially otolith-based age and growth rate determination. Larvae for DNA/C estimates were taken from 3 samples in 1997, from 4 samples in 1998 and, owing to their greater sensitivity to environmental trophic conditions (Bergeron, 2000), only individuals (4.9 to 9.7 mm long) at development stages earlier than the notochord flexion one were selected. They were quickly sorted, frozen at –40°C, and subsequently stored at –30°C. In the laboratory, larvae were thawed and individually ground in varying volumes (0.8 to 2.4 ml according to size) of cold distilled water (4°C) with a Potter Elvehjem homogenizer. An aliquot was rapidly placed in a tin capsule for carbon analysis and the remaining sample was immediately processed for DNA determination by a fluorimetric method (Le Pecq and Paoletti, 1966) modified according to Karsten and Wollenberger (1972, 1977). Type I DNA (Sigma) from calf thymus was used as the standard. The sample for carbon determination was dried (60°C) in an oven and then processed in a Perkin Elmer CHNS/O 2400 analyzer. It must be reminded that a low DNA/C index characterises a good nutritional state (Bergeron *et al.*, 1991; Bergeron *et al.*, 1997) and

vice versa: Bergeron (2000) defined 60 µg DNA.mg C^{-1} as the upper value of the threshold under which anchovy larvae are in good condition.

Results

The seasonal variations of Gironde flow rates show strong interannual differences (Fig. 2).

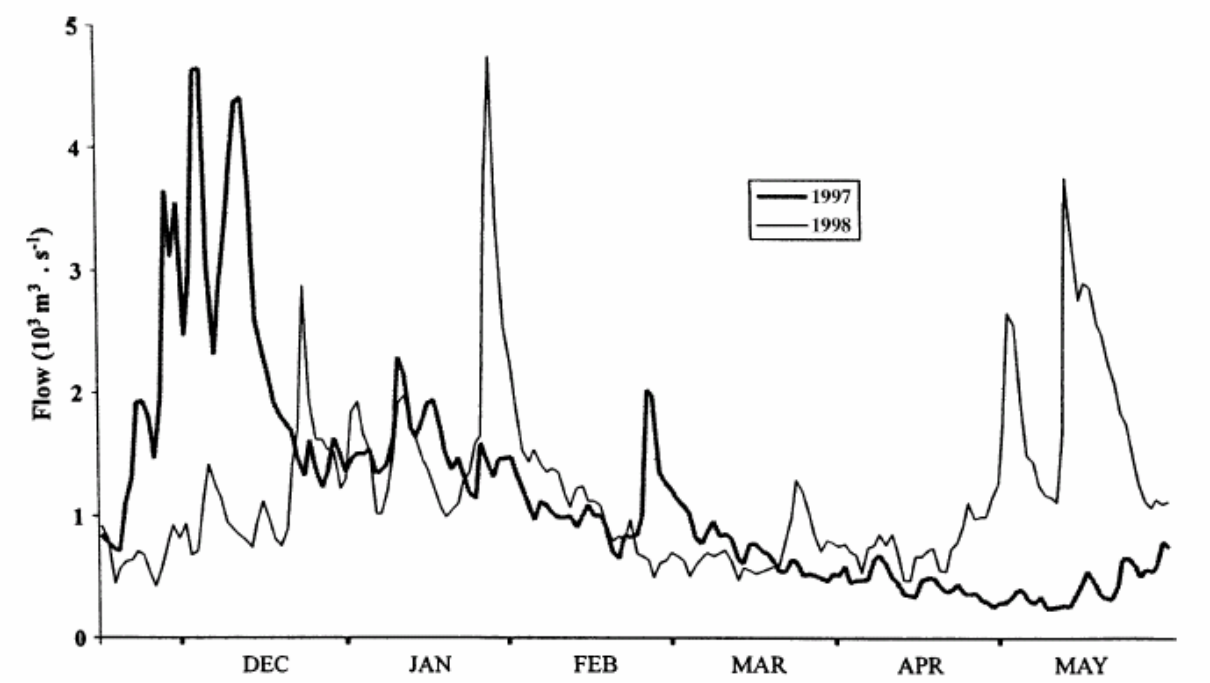

Fig.2. Seasonal variations of Gironde outflow during the early months of 1997 and 1998 (daily flow rates data provided by the "Port Autonome de Bordeaux").

The general pattern observed in 1997 was classical for temperate regions: after winter peaks, a long decreasing trend characterised the springtime period. However, in 1998, the pattern was different and high flow rates were measured in late April and May (Fig. 2). As a consequence, the vertical hydrological structure of the study area took very different aspects from one year to the other (Fig. 3). Quite unlike 1997, when the lowest surface salinities were > 34.9, the 1998 surface salinities implied an invasion of less saline water. This less saline water

contained elevated concentrations of NO_3 (from 1 to 4 µM) compared to more saline surface waters in 1997 (NO_3 = "undetected" to 0.1 µM). Nutritional indices for zooplankton (PK specific activity) and larval anchovy (DNA/C) were greatly enhanced in 1998 compared to 1997 (Table 1). In the latter year, these indices were quite poorer (differences are highly significant: *t*-test indicates $p < 0.001$ for PK specific activity mean values and even below for DNA/C index values).

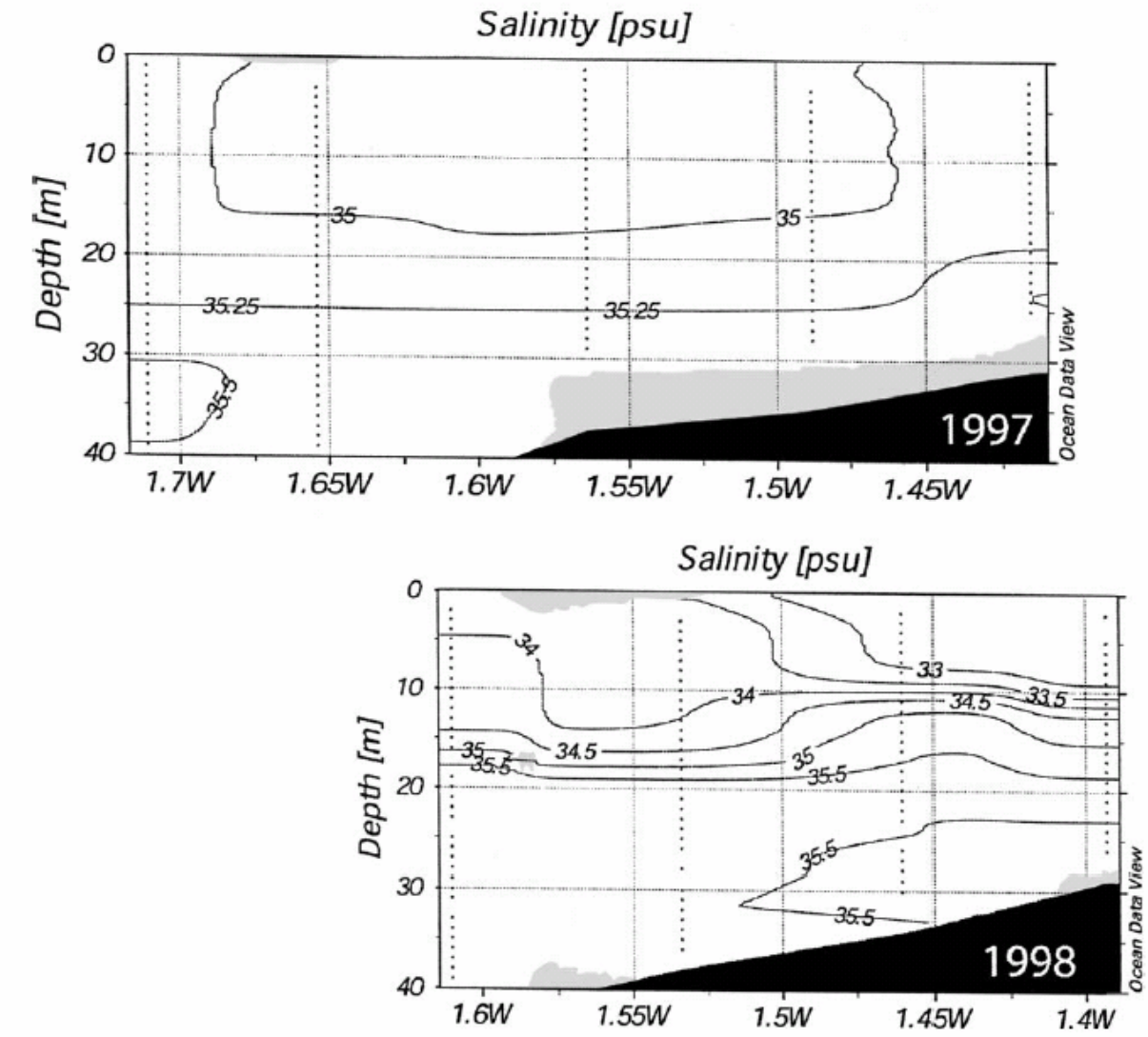

Fig.3. Salinity structure of hydrological sections constructed from data collected at stations indicated in Fig. 1. Images created using ODV software (Schlitzer, 2001).

Table I. Mean values and standard deviations of zooplankton pyruvate kinase specific activity (µmol min^{-1} mg $protein^{-1}$) and anchovy larvae DNA/C index (µg DNA.mg C^{-1}).

	PK			**DNA/C**		
	mean	s.d.	n	**mean**	s.d.	n
1997	**0.63**	0.33	15	**84**	16	15
1998	**1.22**	0.45	10	**59**	18	29

Discussion

Nutritional condition of larval fish is considered an important factor affecting recruitment fluctuations of the population, either directly by inducing mortality or indirectly by lengthening the duration of larval life and exposure to predators. In a previous study (Bergeron, 2000), larval anchovy DNA/C index in the Bay of Biscay indicated that good nutritional condition was only detected along the shelf break or in the river plume (ERAG 93 cruise, June 1993). In 1993, the Gironde run off was not characterised by peaks of the same magnitude as in 1998, but a brief peak occurred at the beginning of May (over 3.10^3 $m^3.s^{-1}$) proceeded by rates which were 2x greater than normal (oscillating around 10^3 $m^3.s^{-1}$). During this period in 1993, larval nutritional condition was also better (mean DNA/C = 54; s.d. = 6).

The anchovy population in the Bay of Biscay spawns in two main areas, one located near the shelf break and one in the Gironde plume (Motos *et al*., 1996). The enrichment of the shelf break environment is controlled by large scale oceanic physical processes (New and Pingree, 1990; Laborde *et al*., 1999): tidally-induced internal waves are very energetic in the

Bay of Biscay, it is particularly manifest near the Armorican shelf in the North (e.g. around 47-48° N according to Langlois *et al.*, 1990) and they are still strong enough in the more southern part of the region (around 45° N), where spawning of anchovies occurs, to induce notable enrichment in nutrients (Pichon and Correard, accepted paper to be published in Oceanologica Acta, pers. comm. from A. Pichon, Service Hydrographique et Océanographique de la Marine, Brest). Moreover, the superficial fertilisation of this area is reinforced by strong instabilities of the poleward slope current linked to bathymetric anomalies such as canyons, notably in the present case the "canyon du Cap-Ferret" (X. Carton, Laboratoire de Physique des Oceans, IFREMER, Brest, pers. comm.). It is obvious that these physical processes are potentially less interannually variable than rates of the Gironde run off. The latter is dependent on variable climate conditions, which may be from year to year notably different for a given season, over the south-western part of the French territory, while the former is driven by larger-scale, more stable and reproducible physical processes. Therefore it may be hypothesised that a considerable fraction of the anchovy population spawns in an environment which is sometimes unfavourable for larval feeding and survival (e.g. 1997). The early stages of larval life of fishes are recognised to be especially delicate (Hewit *et al.*, 1985; Heath, 1992) and their survival is still considered by many authors as a strong determinant of the annual recruitment level. Consequently, variation in the rate of Gironde outflow might be likely to play a role in the recruitment variability of anchovy. This aspect is worth being confirmed by future studies and should be taken into account for fluctuations forecasts of this fishery, possibly incorporated into predictive models such as those in course of development (Allain *et al.*, 2001).

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