



SOLICITUD DE BUQUE OCEANOGRÁFICO PLAN DE CAMPAÑA

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DATOS DEL PROYECTO:

Título del proyecto: Ecology of wild Common Octopus: towards **S**ustainable Management and **A**quaculture

PLAN DE CAMPAÑA.

The research plan for the Project ECOSUMA project can be summarized in Figure 1. In this document we will tackle only the research cruise applied for the R/V *Sarmiento de Gamboa*.

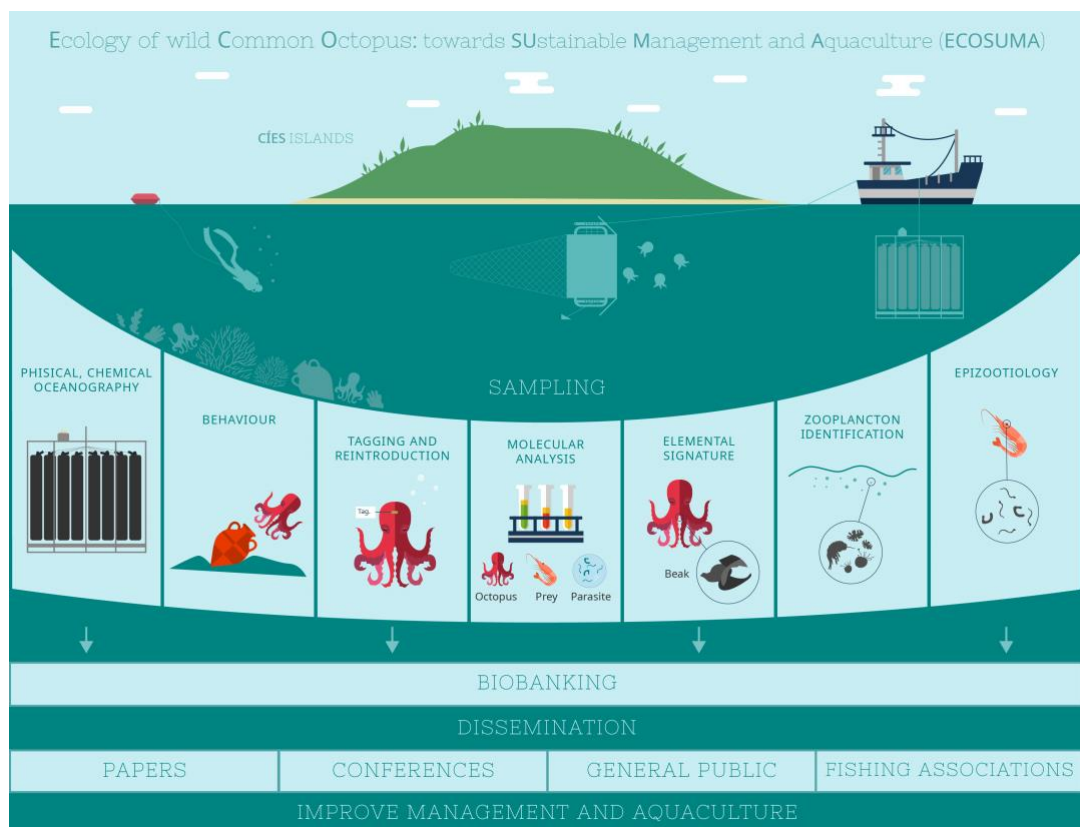


Figure 1. Scheme of the tasks to be undertaken during the project ECOSUMA.

Research cruise to collect *Octopus vulgaris* pelagic paralarvae offshore the southwestern Galician coast and oceanographic parameters

An area will be delimited to study the larval phase of *Octopus vulgaris* in Galicia, targeting animals with more than 3 suckers in each arm, which are washed offshore to complete its paralarval cycle in western Galician waters (Figure 2). The area will be studied at depths ranging from 100 to 2000 m during 5 biological and oceanographic transects including 50 CTD (25 at night and 25 at daytime, red stars and yellow circles) and 9 rosettes in total (3 the first day and last sampling days, T1 and T5, and one the three middle transects, T2-T4) during the month when oldest common octopus planktonic paralarvae are probably the most abundant, October 2021, onboard the R/V *Sarmiento de Gamboa*.

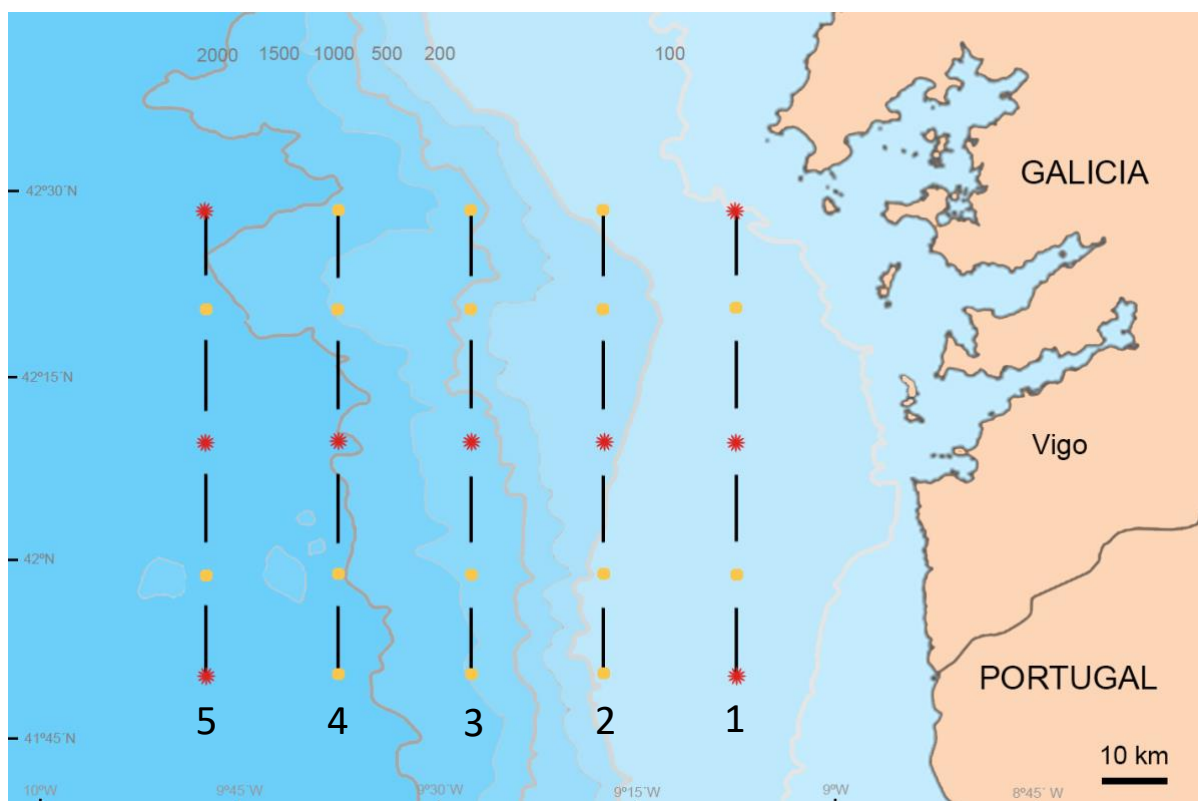


Figure 2. Map of the sampling area including the approximate biological transects and the oceanographic stations in Galician offshore waters (NE Spain). Red stars: Rosette stations; yellow circles, CTD stations.

The research cruise would need 8 days to complete the samplings. Northwards pelagic hauls during the day and southwards at night will be undertaken in each of the four transects parallel to the coast described in Figure 2 in six or eight strata depending in the depth we will filter seawater to collect mesozooplankton using a multi-trawl (MultiNet®) sampler (0.71x0.71 m opening frame). Thus, we will undertake 40 biological tows at bottoms ranging from 100 to 2000 m during daylight and at night (20 at daylight and 20 at night). However, due to the potential situation of the paralarvae, we will only collect biological and oceanographic data up to 500 m depth.

Zooplankton collection: At a ship's speed of 3 knots, the multinet is first lowered down to the desired depth with net bags open and stabilized near the bottom for a period of 3 min, then hauled parallel to the bottom. It is planned to filter 300 m³ of seawater per tow and depth, and then the net bag will be closed and hauled up to the next level. The sample is filtered using a 200 µm mesh and fixed onboard with 100% ethanol for using in ecological, molecular and parasite studies. Onboard it has to be transferred to 70% ethanol and stored at -20°C. An integrated Pressure Sensor (measuring range according to customer's requirements) allows continuous supervision of the actual operating depth, which is indicated together with all relevant system data at the LCD-display of the Deck Command Unit. Two Electronic Flow Meters with automatic angle compensation are mounted to the Underwater Unit: one inside the opening of the Underwater Unit for the determination of the amount of water passing through the opened nets, one outside the opening for the determination of clogging effects. For horizontal collections the MultiNet is used with a V-Fin Depth Depressor. Furthermore, to undertake multidisciplinary studies, the gear is also equipped with two electronic flow meters with automatic angle compensation (for towing speeds from 0.2m/sec) and temperature, salinity, chl_a and depth sensors obtaining, among others, those way profiles of the thermohaline conditions of the water masses where we are making the trawls. These data will allow to relate the temporal, spatial, and bathymetric variations with the atmospheric, hydrographic (physical, biological and geochemical) and dynamic frameworks where the paralarvae inhabit. We have a Mutinet to be used during the survey, which will be used as contingency plan in case the same equipment demanded to the Unidad Técnica Marina have any functioning event during the cruise.

The sampling could be summarized as follows (the samplings will be made at daylight and at night):

- √ Total route ~ 600 km (350 km between tows and 250 of sampling tows). 350 km of crossing at 8 knots / h ~ 24h of crossing; 250 km at 2.5 knots / h ~ 55 h sampling
- √ 5 transects parallel to the coast of 70 km, separated from each other by 20 km, consisting of 4 trawls of 10 km each, separated by 5 km between drags.
- √ 6 networks (strata from 0 to 200 m) in each drag in the 2 transects on the platform and slope edge (8 trawls per day and night, 98 samples)
- √ 8 networks in each drag in the other 3 transects (12 trawls between 0 and 400 m, 192 samples).
- √ Approximate duration of the drag (~ 1:30 h 8 networks and 1:10 6 networks, filtering at least 300 m³ per stratum at daylight and at night)
- √ CTDs at the beginning of the first drag and in between trawls each drag (Total 50 CTDs).
- √ Rosette in the middle of each parallel transect and another one at the beginning and end of transects 1 and 5 (9 rosettes in total).

The following morphometric and meristic measurements, as well as weights, will be taken to all paralarvae: total length, mantle length, eye diameter, body weight, arm sucker count and mantle width. Morphological and meristic measurements will be made to all

paralarvae employing an optical microscope (Nikon H600L, Eclipse 80 i) and the image analysis system NIS Element 3.0®.

All paralarvae of *Octopus vulgaris* collected from the fishing ground will be separated and preserved in 95% ethanol. The additional cephalopod paralarvae obtained during the hauls (sepiolids, common octopus, ommastrephids and other octopods) will allow to prove the presence of species belonging to other biogeographic areas, mainly Lusitanic and Subtropical, as a response of the global change, which actually has been observed in adults. These early stages of development will be identified according to reference collections that are accessible to the ECOBIOMAR Research Group.

Sampling of physical, biological and geochemical conditions: Continuous salinity, temperature, density, stability, dissolved oxygen, Chl fluorescence and turbidity profiles will be obtained with the multiparameter probe available in the R/V Sarmiento de Gamboa (See project task T.1.1.1) in the 25 hydrographic stations indicated in Figure 2.

A total of nine seawater samples will be collected for the analysis of vertical profiles of salinity (S), O₂, nutrient salts, Chl, and particulate organic carbon (POC) and nitrogen (PON), at all stations (6 depths). These data will be processed by researchers from Organic Geochemical Laboratory (LGO) Research Group. S samples will be collected in 100 ml Pyrex flasks and will be analyzed at the laboratory with a PORTASAL salinometer. This instrument measures the conductivity of seawater samples at 20°C referred to an IAPSO certified standard of 35 and S is calculated with the algorithm of UNESCO (1983). Samples for the analysis of nutrient salts will be collected in 50ml polyethylene flasks. They will be measured by segmented flow analysis following Álvarez-Salgado (1993). Chl will be collected onto GF/F filters and frozen (-20°C) before fluorometric determination on board after 90% acetone extraction (Yentsch & Menzel, 1963). POC and PON will be collected on precombusted GF/F filters by gently vacuum (-0.15 atm) filtration, frozen to -20°C and will be analyzed in the laboratory following Álvarez-Salgado (1993).

The cruise duration is 8 days, and it should be centered, preferably in mid-October, when it is expected that the targeted paralarvae of *Octopus vulgaris* with more than 4 suckers are more abundant in the area studied. However, we have the flexibility of doing the survey in a window that could range from mid-September to early November. The expected completion of the samplings is six days. However, the potential weather condition and the intensive sampling that has to be performed suggest to have one and a half extra days as a sampling contingency plan.

The main objective of this project is to investigate in the wild some of the main bottlenecks that prevent the integral rearing of this species: the ecology of the pre-settled planktonic paralarvae (with more than 3 suckers per arm) washed to offshore waters offshore. The trophic relationships of the common octopus in phase of an area subjected of seasonal upwelling will be investigated to advance towards an ecosystem approach for the management of these socioeconomically important octopod resource as well as to transfer the data gathered to obtain an integral culture of this socioeconomic important cephalopod. This general objective include the following specific ones:

- Obtain, for the first time, information of wild paralarvae with a range of 4-22 suckers per

arm and the oceanographic influence during this planktonic phase.

- Extend historical series of common octopus paralarvae abundance of newly hatched paralarvae with 3 suckers per arm, which are the only collected during the 20 years monitoring program up to 120 m depth, near the fishing ground, as well as gather information on the influence of chemical, physical and biological parameters of water masses under different scenarios in Galicia. The time series that we already have will also allow us to estimate possible changes in the distribution associated with global warming.
- Estimate age by counting the rings deposited in the beaks of the specimens collected and try to identify marks of specific life cycle events.
- Study elemental signatures in beaks of subadults (8-200 g) and pre-settled planktonic animals to investigate the settlement strategy that is already unknown in the wild.
- Identify with molecular techniques the pre-settled planktonic paralarvae found offshore the Galician waters and the subadults of this species, collected by scuba diving.
- Comprehend the trophic ecology of the common octopus collected, their potential preys and pathogenic parasites involved.
- Study the population genetic structure of the zoonotic parasite *Anisakis* spp. in the mesozooplanktonic realm, and its potential transmission to planktonic predators, including the common octopus.
- Monitoring the mesozooplankton fraction (0.2-20 mm) to get a concise idea of the zooplankton communities that the different paralarvae inhabit, and test whether they are specialist or generalist predators.
- Quantify home range and identify habitat preferences of octopus juveniles in the wild using acoustic telemetry techniques.
- Create the first biobank on mesozooplankton samples and associated data under a 9001 certification.

Because of the tight schedule due to the sampling effort during the day will need to organize to organize the fieldwork in three groups, since we have to undertake the samplings during 24 hours. We would need the technical assistance to carry out the operations related to the depletion of the multitrawl gear, as well as the CTD and rosette samplings. We will carry the instruments necessary to complete the sampling of the zooplankton fraction, as well as to collect the water samples. We would necessitate -20 freezers to maintain the samples that have to be analyzed once back at the lab.

RESUMEN DEL PLAN DE CAMPAÑA (máximo 10 líneas)

During this research cruise we will collect biological and oceanographic data in waters offshore the western Galician coast. The main objective of this cruise is to investigate in the wild some of the main bottlenecks that prevent the integral rearing of this species: the ecology of the pre-settled planktonic paralarvae (with more than 3 suckers per arm) washed to offshore waters before reaching the fishing ground near the coast. The trophic relationships of the common octopus in an area subjected of seasonal upwelling will be



investigated to advance towards an ecosystem approach for the management of these socioeconomically important octopod resource as well as to transfer the data gathered to obtain an integral culture of this socioeconomic important cephalopod.