

2. PROTOCOL TO ESTIMATE GLASS EEL RECRUITMENT

The aim of this protocol is to develop a standard method to calculate a glass eel recruitment index using a fishing gear (a net or a trap), preferably at the river mouth, i.e., not influenced by any anthropogenic pressure. The method also aims to provide a quantitative estimate of the glass eel recruitment for each catchment so that the levels of recruitment can be compared among basins. It is hoped that the use of this protocol could be extended to other catchments throughout the SUDOE area, to provide a nationwide recruitment index.

The conditions in the 10 pilot basins covered by the SUDOANG project are different. The basins contacting the Atlantic Ocean are strongly influenced by the tide, unlike the Mediterranean Sea where there is little tide and glass eels enter the river migrating against the current. The methods to sample glass eels and estimate recruitment have therefore, to be adjusted to the local conditions. However, regardless of the method chosen to sample glass eels, the objective is to record/ obtain a number of glass eels per volume of water (in the case of river basins influenced by the tide) or cross-section (in the case of river basins draining into the Mediterranean).

Glass eels use selective tidal transport to save energy. Hence, in the Atlantic coast, where there is the influence of the tide, glass eels should be caught during the flood tide. In the Mediterranean, where the tide influences the activity of glass eels to a lesser extent, the wind plays an important role in the recruitment process.

2.1. Timing of surveys

The fishery should be conducted **monthly** during the more **intense migration period** (ideally 6 months). Sampling should be performed in **New Moon Day**. If the weather conditions are not favourable to fishing, the fishery can be done up to a maximum of 2 days following the New Moon Day. In pilot basins that flow into the Atlantic, sampling should always be conducted during the **night flood tide**. In the catchments flowing into the Mediterranean, sampling should be conducted at similar hours of the night or around the highest water level if those conditions are considered to influence catches. In any case, the sampling protocol should be set after the initial experiments carried out during the first year and maintained in the long term to ensure the standardized collection of data for a recruitment series.

2.2. Site selection

The **fishing location** should be as close to the sea as possible, to avoid the influence of other fishermen, in case there is a fishery.

2.3. Sampling procedures

- The fishing gear should be preferably the one used by fishermen or other that has already been used;
- A sieve should be used to separate glass eels from other organisms;

- When 50 or fewer eels are caught they should be taken to the laboratory. When more than 50 glass eels are caught, a subsample of 50 glass eels should be kept for analysis;
- Water parameters should be measured at the beginning and end of each fishing period. If fishing is continuous, two records will be enough (beginning and end of fishing). If sampling is conducted repeatedly, measures should be recorded at regular intervals, i.e., at each sampling occasion.
- In the **Atlantic area**:
 - Sampling should start at the beginning of the flood tide and last until the end;
 - It is necessary to record the value of the flowmeter at the beginning and at the end of sampling;
 - The number of glass eels per volume of water should be recorded;
 - If fishing is continuous during the entire flood tide, it is necessary to record the value of the flowmeter at the beginning and at the end of sampling, and also the number of glass eels per volume of water;
 - If sampling can be conducted repeatedly at regular intervals during the entire flood tide, then the value of the flowmeter at the beginning and at the end of each occasion should be recorded separately; Glass eels should also be collected and stored separately on each occasion;
 - At the end of fishing, the 50 glass eels to retain for laboratory analysis should be equally distributed throughout the time intervals of sampling;
 - The cross-sectional area of the river and average water velocity should be provided to estimate the overall volume of water flowing in the river.
- In the **Mediterranean area**:
 - The sampling should last long enough to cover the night peak migration.
 - If a fyke-net (or similar sampling gear) is used, the cross-sectional surface sampled by this trap should be recorded, and the overall cross-sectional river at the location of the trap should be measured. If possible, tag-recapture experiments can be carried out to estimate the efficiency of the trap;
 - If a ladder trap is used, the efficiency of the ladder should be assessed, either by carrying mark-recapture experiments or by carrying out bongo tows to provide a measure of glass eel densities in water (protocol similar to the Atlantic area protocol);

2.4. Environmental data to collect in the field

The following environmental variables should also be recorded:

- Date and time of fishing;
- Tide hour and height;
- Depth;
- Water temperature and salinity or conductivity (brackish water or freshwater) – via data loggers or Van Dorn Bottles/Niskin Bottles;
- Date of New Moon day;
- Flow rates across the stream width and at trap sites;
- Length of fishing period.

2.5. Laboratory procedures

Samples taken to the laboratory should be kept in water in the refrigerator, to be analysed as quickly as possible.

In the lab, the glass eels should be placed in paper to remove extra water, and the following data should be recorded:

- Individual length (mm);
- Individual weight (0.01g);
- Pigmentation stage (according to the classification from Briand, 2009).

2.6. Field Equipment

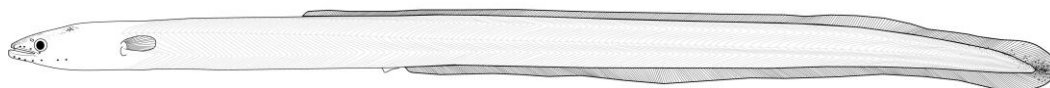
- Net/trap;
- GPS;
- Flowmeter;
- Sieve;
- Van Dorn or Niskin Bottles or Data logger.

2.7. Pigmentation stages

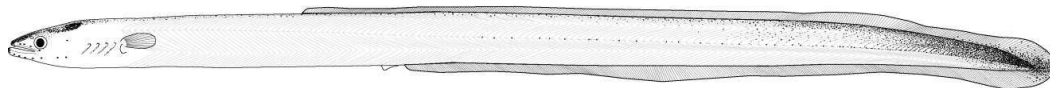
The identification of pigmentation stages should follow the classification by Briand (2009).

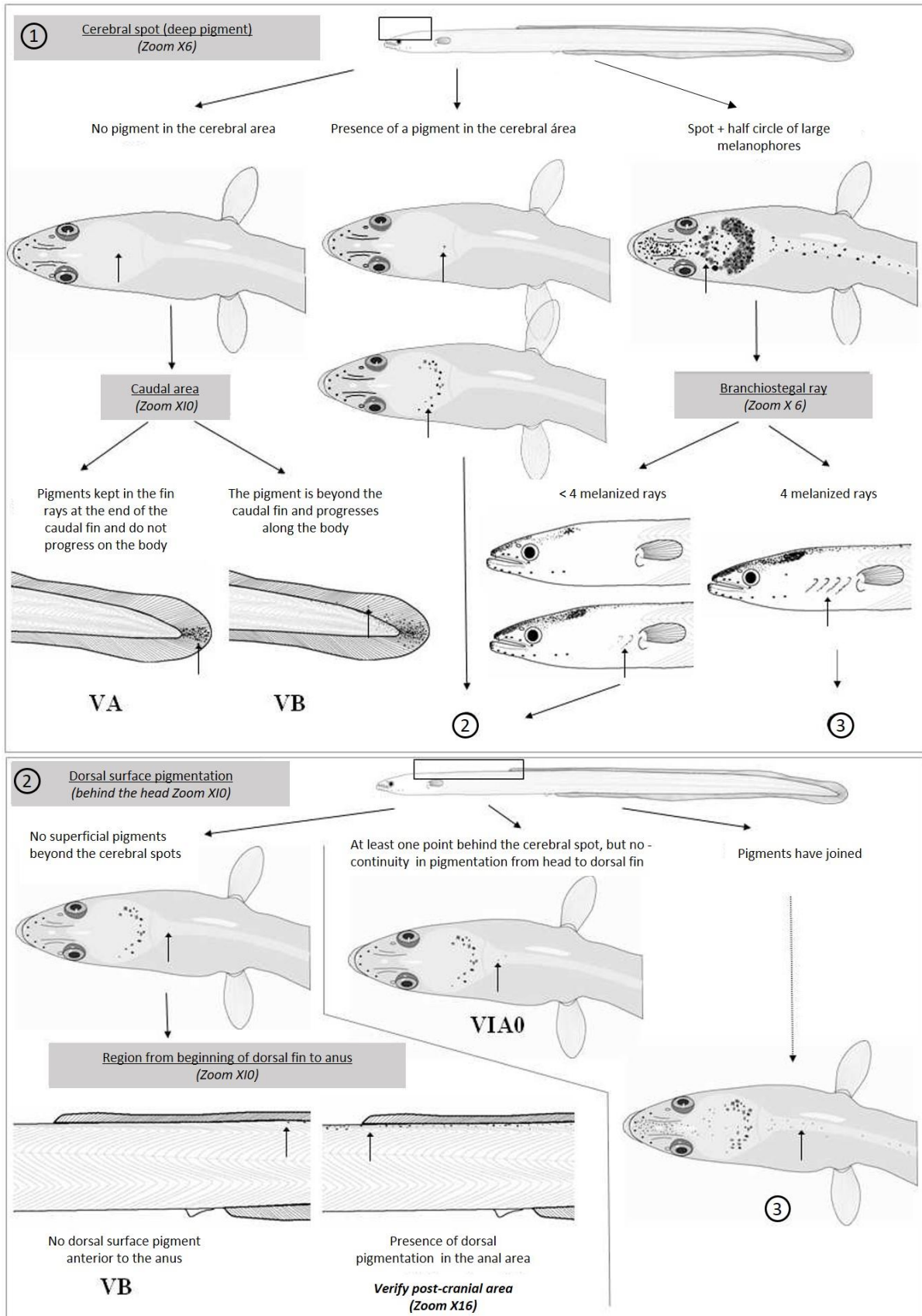
From (Strubberg, 1913; Elie et al., 1982; Lecomte-Finiger, 1983).

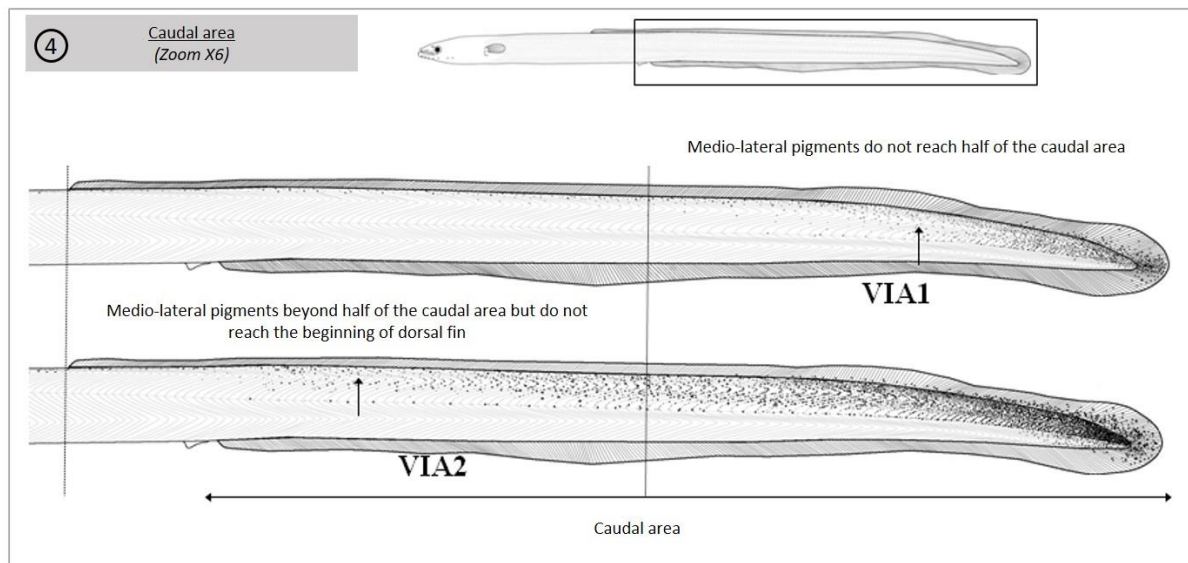
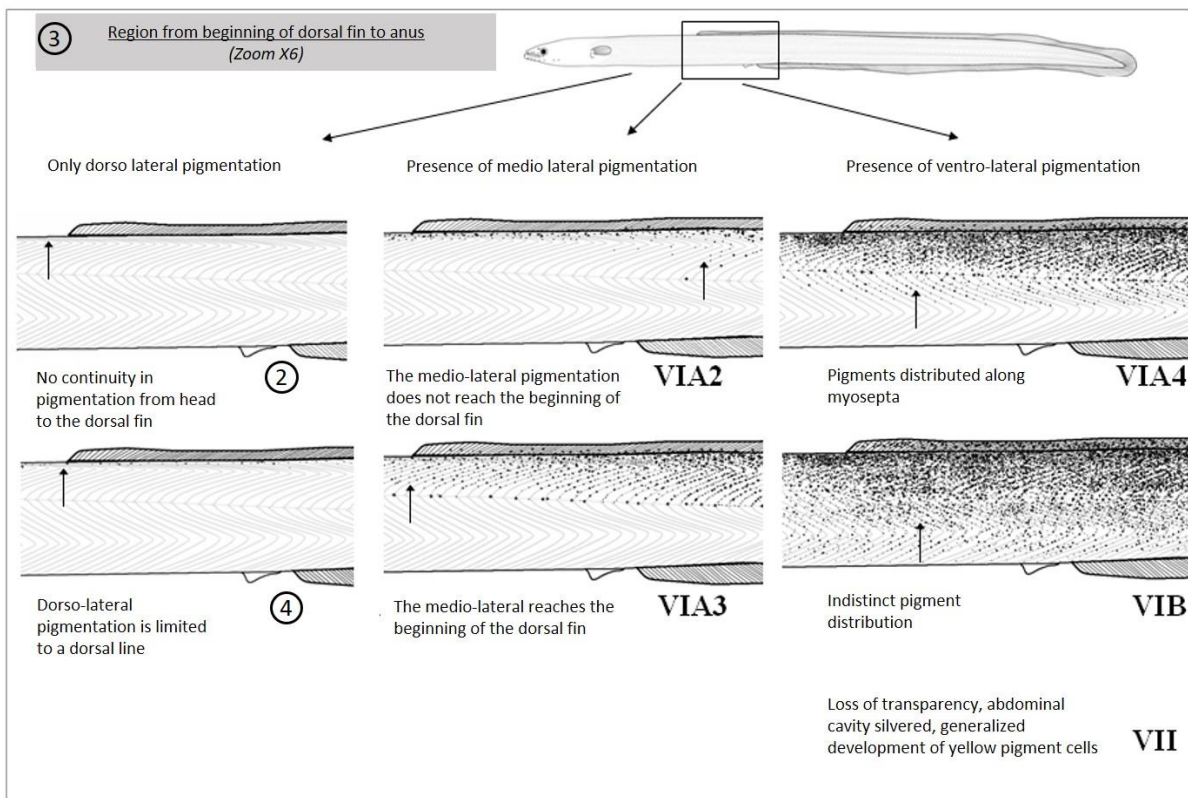
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References

- Briand C. 2009. Dynamique de population et de migration des civelles en estuaire de Vilaine. PhD thesis, Agrocampus Ouest. Rennes, France. 207p.
- Elie P., R. Lecomte-Finiger, I. Cantrelle and N. Charlon. 1982. Définition des limites des différents stades pigmentaires durant la phase civelle d'*Anguilla anguilla* L. *Vie et Milieu* 32: 149–157.
- Lecomte-Finiger R. 1983. Contribution à la connaissance de l'écobiologie de l'anguille, *Anguilla anguilla*, L. 1758, des milieux lagunaires méditerranéens du golfe du Lion: Narbonnais et Roussillon. PhD Thesis, Université de Perpignan, France.
- Strubberg A.C. 1913. The metamorphosis of elvers as influenced by outward conditions. Meddelester fra Kommissionen for Havundersøgesler, serie Fiskeri Copenhagen 4: 1–11.

Site name:			Site code:
GPS Coordinates	Lat:	Long:	Coordinate system:
Date:	Date of New Moon:		Hours: (start - end) ___H___ - ___H___
Photos (ref):			Team:

Atmospheric conditions

Air temperature (°C):				
Nebulosity:	<input type="checkbox"/> <i>clear</i>	<input type="checkbox"/> <i>slightly cloudy</i>	<input type="checkbox"/> <i>averagely cloudy</i>	<input type="checkbox"/> <i>fully cloudy</i>
Wind:	<input type="checkbox"/> <i>null</i>	<input type="checkbox"/> <i>light</i>	<input type="checkbox"/> <i>moderate</i>	<input type="checkbox"/> <i>strong</i>
Rain:	<input type="checkbox"/> <i>yes</i>	<input type="checkbox"/> <i>no</i>	Before sampling:	

Sampling section

Total fishing time (minutes):	Tide hour: ___H___	Tide height (m):
Fishing net opening area (m ²):	River width (average - m):	

Environmental conditions *IF CONTINUOUS FISHING (if not, fill in next page)*

	<i>Start</i>	<i>End</i>		<i>Start</i>	<i>End</i>
Hour: ___H___	___H___	___H___	Conductivity (μS/cm):		
Water temperature (°C):			TDS (mg/L):		
Salinity:			Depth (m):		
Flowmeter:					

Remarks (please describe the fishing method, including the type of fishing net/ trap)

2. Glass Eel Recruitment

Site code:

Date:

Sampling data *IF NOT CONTINUOUS FISHING*
(Replicates - 20 minutes each; Water Temperature - °C; Salinity; Conductivity - µS/cm; TDS - mg/L; Depth - m; Total Weight fished – 0,01g; Sub-sample – 0,01g)

Repl. Nr.	Hours		Flowmeter	Environmental parameters					TW (g)	Sub-sample (g)	Remarks
				WT (°C)	Salinity	Cond (µS/cm)	TDS (mg/L)	Depth (m)			
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(Total Length - mm; Total Weight - 0.01g; Pigmentation stage - see protocol)

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