

| |
|--|
| Sampling protocol: Survey_plankton_Atlantic |
| Sampling objective(s): data collection for fishery-independent timeseries for by sampling ichthyoplankton |
| Start of sampling: 1983 (mackerel and horse mackerel egg sampling in the Atlantic) |
| Sampling ongoing: yes |
| <p>Data use</p> <p>Primary data use: data from eggs of commercial fish species is used as tuning series in single stock assessments. The data users are stock coordinators in ICES Working group on widely distributed species (WGWIDE). The main target species can be found in Table 1 in http://data.europa.eu/eli/dec_impl/2021/1168/oj. The most up to date data use can be found in the advice sheets per species.</p> <p>Other users are individual scientists or institutions interested in the ichthyoplankton composition for the sampled species in the North Sea.</p> |
| <p>Sampling design and method</p> <p>The survey takes place triennially, and is coordinated by the ICES working group on mackerel and horse mackerel egg surveys (WGMEGS).</p> <p>Sampling design is in detail described in the (inter)national survey protocols. Gear is standardised, so the data per survey can be used as a timeseries.</p> <p>In short, the trawl surveys follow the following stratifications:</p> <ul style="list-style-type: none"> • MEGS plankton: systematic, one stations per 0.5 ICES statistical rectangle. The ICES rectangles are split horizontally or vertically, depending on the area. In the year before the survey, WGMEGS decides on the allocation of survey area and period per country. • MEGS fecundity and atresia: hauls have to be spread out over the sampling area. As many hauls as possible have to be conducted, to get the most accurate indication of realised fecundity in the area. <p>Based on the target stage the survey gear and mesh size is chosen.</p> <ul style="list-style-type: none"> • The MEGS plankton sampling is conducted with a Gulf7 plankton sampler with a 20 mm nose cone and 280 µm mesh. Oblique hauls of at least 10 minutes are made, sampling depth ranging from the water surface to 5 m from the bottom (max. depth 200 meter). If a thermocline of more than 2.5°C/10 meter depth occurs, fishing takes place till 20 meters below the thermocline. Fishing speed is 5 knots, lowering and hauling the gear is done at continuous warp speed: 10 meter/1 minute. • MEGS fecundity and atresia: a pelagic trawl is used to catch sufficient adult mackerel and horse mackerel. As only the ratio female:male and fecundity estimates have to be collected, the gear does not have to be standardised. |
| <p>Sampling protocol and data capture</p> <p><i>In the field</i></p> <p>Plankton sampling</p> <p>The majority of the trawl information (date, time, position, haul duration, flow meter revolutions) is registered automatically, using the vessel's system information. This information is transformed by in-house developed software (TRIHIP Gulf) to an interoperable file format. On each station a measurement of temperature and conductivity (CTD) is done, using a CTD (Seabird or Valeport) which</p> |

is attached to the sample gear, registering temperature and conductivity during the haul. The surface and 20 m depth temperature and salinity is automatically added to the same file as the trawl information.

Plankton samples are as soon as possible fixed in buffered 4% formaldehyde. Fixed samples are treated similarly on board and ashore (see: 'In the lab'). As much samples as possible are processed on board.

Fecundity sampling

If available, 100 adult mackerel and 100 adult horse mackerel are collected from each trawl haul. Individual length measurements are done using an analogue measuring board, of which the set-off is checked before the start of the survey. Individual wet weights are taken using electronic scales, to the gram. Scales are maintained annually and calibrated at least daily. Data is noted down on paper and entered in the computer directly after processing the fish. After the fish selected for biological sampling has been treated following national animal welfare conditions, the otoliths are collected, and sex and - if relevant- maturity is registered by opening the body cavity. Individual fish information is written down on paper and soon after entered to the Billie file with length measurements.

From a pre-defined (In the year before the survey, WGMEGS decides on the allocation of samples per period and area) number of females (per maturity stage) ovary samples are taken: pipette samples of a known volume (25 µl) to estimate potential fecundity, a piece of the ovary for screening purposes -to decide if the ovary should be used for potential fecundity and/or atresia analyses-, and one lobe of the ovary for atresia analyses. All ovary samples are fixed in 3.6% buffered formaldehyde, following the international [manual](#).

In the lab

Plankton sampling

The samples that have not been sorted on board, are processed in the lab ashore. After the eggs have become opaque (approx. after being fixed for 24 hours) the sample can be sorted on board in a black tray after washing out the formaldehyde and using a sorting solution to prevent evaporation of the remaining formaldehyde. All fish eggs are sorted from the sample, by using the 'spray method' (Eltink 2007). After removing the majority of eggs using this method, the sample is checked manually for remaining eggs. Eggs are photographed using an SLR camera, and measured using ObjectJ/ImageJ. Species and egg stage are defined based on the picture, and directly entered to the files with the length measurements. All fish eggs are sorted and identified. Of mackerel and horse mackerel, at least 100 eggs have to be measured and staged if the sample contains more than 100 eggs of the species. In case of large numbers of eggs the sample may be subsampled, using a Folsom splitter (Griffiths *et al.* 1984). Initials of the person(s) sorting the sample, identifying and measuring the larvae are noted down.

Fecundity and atresia sampling

The ovary samples are processed in accordance with the international [manual](#). All ovaries are prepared for screening as soon as the samples have arrived in the laboratory. From the ovary piece a histological slide is prepared and photographed using a slide scanner. These photographs are analysed for presence and stage of vitellogenic oocytes, eggs, POFs (Post Ovulatory Follicles) and atretic oocytes. Based on these screening results the WGMEGS mackerel and horse mackerel fecundity coordinators decide which samples should be analysed for potential fecundity and which for atresia. For potential fecundity the pipette samples are photographed under a dissecting microscope. Images are analysed in ObjectJ/ImageJ. For atresia analyses a histological slide is prepared from the whole preserved lobe of the ovary. The slide is photographed using a slide scanner. A cross-section of the ovary lobe is analysed for presence and area of α -atretic oocytes using ObjectJ/ImageJ.

Post-processing data

Billie files are not post-processed, only quality checked (see Data quality). ObjectJ/ImageJ information is transposed to the exchange format and combined with the plankton sampling Billie files.

Fecundity and atresia data is copied from the ObjectJ file to the ICES format for fecundity and atresia datasheet.

Post-processing of CTD information is described in a separate factsheet [Surveys_CTD](#).

Data quality

Quality assurance procedure

Plankton sampling

Flow meters are calibrated at the start of the survey or during the survey. When a new flowmeter is installed, it is calibrated. During the survey, the relation between haul duration and flow meter revolutions is checked after each haul.

All persons sorting the plankton samples and identifying the species are trained via (inter)national exchange workshops. The national workshop is organised in the survey year, prior to the survey. Quality assurance of sorting is done by routinely re-sorting five samples per person sorting the samples. If more than 5% of the eggs has remained in the samples, all samples sorted by that person will be resorted. Species identification is checked by letting a second person identify the eggs of two samples per person. If there is more than 5% discrepancy in identification other (if needed: international) colleagues may have to identify eggs as well to reach agreement.

Data quality checks of the plankton data are conducted upon processing at the institute, and before entry into the national database FRISBE. Standardised SAS scripts are used for the data quality checks (available upon request). Essentially, the trawl haul data are checked for outliers on numerical values (either by plotting or by providing minimum, mean, and maximum values), consistency in text variables (e.g. station coding, crew members).

Fecundity and atresia sampling

All persons processing the fecundity/atresia samples are trained via (inter)national exchange workshops. The national workshop is organised in the survey year, after the survey.

Data quality checks of the fecundity and atresia sampling are conducted upon processing at the institute, and before sending to the WGMEGS mackerel and horse mackerel fecundity coordinators. The fish biological data (length, weight, sex, maturity and ovary weight) are checked using a standardised SAS script for the data quality checks (available upon request) before entry into the national database FRISBE. The ICES fecundity and atresia database is currently under development.

Quality checked parameters

Plankton sampling

- Haul information: survey code, vessel name, gear type, sampling date, time, shooting and hauling positions (map), crew members, station code, sample number, haul duration, flow meter revolutions, duration vs. flow meter revolutions, temperature, salinity, sample fixation coding;

- Species information: species list (expert judgement), maturity coding, subsampling type coding, minimum and maximum length, subsampling factor, number measured, measurement unit by species.

Fecundity and atresia sampling

- Haul information: survey code, vessel name, gear type, sampling date, time, shooting and hauling positions (map), crew members, station code, sample number, haul duration, sample fixation coding;
- Species information: species list (expert judgement), maturity coding, subsampling type coding, minimum and maximum length, subsampling factor, number measured, measurement unit by species.

Data storage

Plankton sampling

National database: Billie files are submitted to the national database FRISBE. The relevant aspects of this database are described in [Proc databases](#).

International database: MEGS data is stored in the ICES eggs and larvae database (<https://www.ices.dk/data/data-portals/Pages/Eggs-and-larvae.aspx>).

Fecundity and atresia sampling

National database: Billie files are submitted to the national database FRISBE. The relevant aspects of this database are described in Proc_databases.

Datasheets with screening, fecundity and atresia data are stored on the institute's shared drive and submitted to the WGMEGS fecundity and atresia coordinators via a ftp-server. The ICES fecundity and atresia database is currently under development.

Data availability

Institutional availability: accessibility of the national database FRISBE is described in [Proc databases](#), data is made available as soon as possible after the survey, mostly within 2 months after the sample processing has finished.

Public availability: data submission deadlines for submission to ICES eggs and larvae database and WGMEGS fecundity and atresia coordinators are driven by the end user's meeting. In general, the WGMEGS preliminary egg, fecundity and atresia data are publicly available before 1 August of the sampling year, the finalised data a year later.

Reference to full documentation:

National manual: CVO_h_003: Damme, C. van, U. Beier, I. de Boois, D. Burggraaf, B. Couperus, R. van Hal, T. Pasterkamp, J. Vrooman, 2023. Handboek bestandsopnamen en routinematige bemonsteringen op het water. Versie 17, maart 2023. CVO rapport 23.002

International manuals:

- MEGS at sea sampling: ICES. 2019. SISP 6 - Manual for mackerel and horse mackerel egg surveys, sampling at sea. Version 2.2. Series of ICES Survey Protocols. 82 pp. <http://doi.org/10.17895/ices.pub.5140>
- MEGS fecundity and atresia estimation: ICES. 2019. SISP 5 - WGMEGS Manual for the AEPM and DEPM estimation of fecundity in mackerel and horse mackerel. Version 12. Series of ICES Survey Protocols. 84 pp. <https://doi.org/10.17895/ices.pub/5139>

- Folsom splitter: Griffiths, F. B., Brown, G. H., Reid, D. D., and Parker, R. R. 1984. Estimation of sample zooplankton abundance from Folsom splitter sub-samples. *Journal of Plankton Research* 6(5): 721-731.
- Spray technique: Eltink, A.T.G.W. 2007. The spray technique: A new method for an efficient separation of fish eggs from plankton. *Journal of plankton research*. [10.1093/plankt/fbm065](https://doi.org/10.1093/plankt/fbm065)

Review frequency full documentation: national manual is annually reviewed; the reviewing frequency of the international manuals is unknown

Factsheet author(s): Boois, I.J. de; Damme, C.J.G. van

Factsheet latest update: 27/01/2023

Factsheet latest review: 27/01/2023