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Protocols of the EU bottom trawl survey of Flemish Cap

by

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Abstract

Methods and procedures used in the EU bottom trawl survey of Flemish Cap (NAFO Division 3M) are described in detail. The objectives of publicising these protocols are to achieve a better understanding of its results, and to contribute to the routines being unaltered.

Introduction

The following protocols have been routinely followed since the beginning of the survey series in 1988. They describe the working routines used in the past as well the procedures proposed to be followed in the future. However, there were some changes along the years, which include:

- Hauls schedule was of 24 hours during the first survey in 1988, and only then; it was from 6:00 to 22:00 local time in the remaining surveys.
- The former RV Cornide de Saavedra was substituted by RV Vizconde de Eza in 2003.
- Coverage area also changed in 2003, increasing its limits from 730 m isobaths to 1100 m in 2003 and 1460 m since 2004.
- Non-commercial invertebrates, sponges and corals among them, are recorded since 2007.
- Division 3M is divided into 39 sampling strata up to 1460 m depth, but only 32 of them are considered. Strata 35-39 in the Beothuk bank and strata 26 and 27 located in the SE Flemish Cap were not visited since 2007 as it was almost impossible to carry out standard hauls. Results from 2004 onwards refer to the area of those 32 strata.
- Sampling of target species, including cod, is done independently by sex, even it isn't required by the NAFO criterion.

Contents¹

Introduction	Objective
	Flemish Cap
	Former research
	Fisheries
	Work already done
	Importance of the survey
Survey Design	Trawl station methodology
	Criteria for rejecting a haul
	Vessels
	Survey gear
	Personnel
	Standardization
	Dates
Data Collection	Catch record
	Length sampling
	Biological sampling
	Redfish (<i>Sebastes</i> spp.) sampling
	Shrimp (<i>Pandalus borealis</i>) sampling
	Sampling benthonic invertebrates
	Conservation of specimens
	Taxonomy
	Physical oceanography
	Feeding sampling
	Recording data
Data Analysis	Results
	Calculations
	Missing data
Validation of Survey Results	
References	
ANNEXES:	
	Data forms
	Species working codes
	Shrimp (<i>Pandalus borealis</i>) sampling
	Publications 2002-2012

¹ The covered items follow the scheme proposed by NAFO (1975a) and used in the Manual of Groundfish Surveys in the Northwest Atlantic (Doubleday 1981).

Objective

The objective of the survey is to know the stock status of target species: their abundance, biomass and demographic structure, and the oceanographic conditions on the bank. This objective implies the following actions:

- A random stratified survey of the Flemish Cap area until 1460 m (800 fathoms) depth, making 181² bottom trawl hauls with a Lofoten fishing gear, at daytime: between 6:00 and 22:00, and 30 minutes effective fishing time.
- Recording catches of fish species and invertebrates.
- Detailed biological sampling in each haul, including length, sex, weight, otolith and gonad's sampling for each one of the target species. Only length and length-weight sampling will be done for all the other species.
- Feeding analysis of most abundant species, to be done every two years.
- Sampling of invertebrates, with special attention to corals and sponges, to allow identification of potentially vulnerable marine ecosystems.
- Collecting environmental data through a reticule of CTD stations separated 15 nautical miles both in latitude and longitude.

Target species:

- cod (*Gadus morhua*)
- redfish (*Sebastes marinus*, *S. mentella* and *S. fasciatus*)
- American plaice (*Hippoglossoides platessoides*)
- Greenland halibut (*Reinhardtius hippoglossoides*)
- roughhead grenadier (*Macrourus berglax*)
- shrimp (*Pandalus borealis*)

Flemish Cap

Flemish Cap is an isolated bank on the American continental shelf, with an approximated surface of 17 000 squared nautical miles within the 1460 m (800 fathoms) isobath and 10 555 within the 730 m (400 fathoms) one (Figure 1). Flemish Pass, an area deeper than 1000 m, separates it from the Newfoundland Grand Bank and gives it its isolated character by limiting the migration of many species, particularly those occurring in the shallowest zones. The position of the bank was first determined by the Marquis de Chabert in 1750, who indicated that the bank already appeared in Dutch charts, but in a wrong position (Chabert 1753).

The general circulation in the vicinity of the Flemish Cap consists of the offshore branch of the Labrador Current which flows through the Flemish Pass on the Grand Bank side and a jet that flows eastward north of the Cap and then southward east of the Cap. To the South, the Gulf Stream flows to the northeast to form the North Atlantic Current and influences waters around the southern areas of the Cap. In the absence of strong wind forcing, the circulation over the central Flemish Cap is dominated by a topographically induced anti-cyclonic (clockwise) gyre (Akenhead 1986, Stein 1996).

² The cited 181 hauls is not a proper quantitative objective, but it corresponds to the most appropriate number to reach the main objective of the survey, which is an adequate sampling of the whole bank. The survey is planned based on this figure and, because of that, it is seldom achieved and never exceeded.

Templeman (1976) made the first full description of the Flemish Cap Bank and he made reference to the smaller cod caught in that bank by American fishermen in the late nineteenth century compared with the sizes of cod fished in the Grand Banks.

Former research

Regular fluctuations on the magnitude of the cod and redfish year-classes have been recorded in Flemish Cap. This was the reason to develop an international research project on the factors affecting the production of good or bad year-classes (Lilly 1986). On these studies was concluded that the predominance of the anticyclonic flow around Flemish Cap was the main factor for the larval survival.

Lilly (1986) made a review of the objectives and achievements of this project developed between 1978 and 1982. The Flemish Cap Bank was chosen to study the real processes and mechanisms included in fish production for the following reasons:

- Fluctuations in year-class strength of both cod and redfish were regularly observed in this area.
- The stocks of cod are discrete and confirmed as separate from those of the Grad Bank.
- The circulation patterns are likely quite amenable to study.
- The area is reasonably restricted in size.
- The area is one which, because of its major oceanographic features, has been of interest to physical oceanographers for many years and there exist a useful historical data base of fish production and physical environmental data.

Thus, there were three important issues to consider for the prediction of the survival in the different year-classes:

- The effect of water circulation patterns and the abundance and size composition of the planktonic food supply on the retention and the survival of fish larvae on Flemish Cap.
- The effect of intraspecific and interspecific predation on the survival of juvenile fish.
- Improved assessment of the size of the spawning stocks³.

The survey season was chosen in accordance to the initial objectives, and cod was the most important. Since cod spawning occurs mainly in March, sampling for larval survival studies had been proposed to be held between February and June, for survival of juveniles in March and September and for estimating the abundance of spawning stock population in March. For studying the way in which the eggs and larvae are swept by the currents, the period from March to May had been also proposed.

Cod annual recruitments were very weak along the years when the project was developed, hindering to complete the planned studies. One of the most important conclusions was that the 1981 strong year-class was originated from a very small spawning stock. This conclusion reinforced the general knowledge that the oceanographic conditions and circulation patterns on the bank at the time of spawning were the determining factors in the concentration or dispersion of the larvae to regions outside the bank causing the fluctuations in the size of cod year-classes.

One of the most important concerns about the fishing resources in Flemish Cap, not yet fully resolved, was the isolation of the cod in the Flemish Cap bank. According to de Cardenas (1994), results of two tagging surveys conducted by the European Union in 1991 and 1992 do not allow to reject the hypothesis that there is some interconnection with neighbouring populations. According to this author, these tagging results are similar to those obtained in the Canadian surveys of 1962 and 1964, and they suggest that there is some rate of emigration of individuals who have reached maturity (probably aged 5 or 6). The occurrence of such migration, when it is not

³ The same issues, as well as the interest of the Spanish and Portuguese fishing fleet in those fisheries, were the reasons to initiate the EU survey series.

taken into account in the assessment, could produce significant errors in the estimation of the parameters and consequent mistakes in the advice of management measures (mesh sizes, reference fishing mortalities, etc...).

Fisheries

Flemish Cap is entirely outside any 200-mile EEZ, and the exploitation of its resources is regulated by the NAFO.

Inaccuracy in catch statistics was a constant problem in the history of fishing in Flemish Cap. It occurred as a result of, first, the overfishing of national quotas by NAFO member countries and the subsequent unreported catches and, second, the presence of uncontrolled fleet belonging to non-member countries, whose declaration of catches, when they were made, did not offer guarantees. In this scenario, the results of the research surveys in Flemish Cap were the most reliable information source on the state of stocks. Russia also conducted annual surveys during the period 1977-1993, but there was a great disparity between the results of these surveys when both coincided in the same year. A Canadian survey covered the period 1977-1985.

Work already done

This survey series was initiated by the EU in 1988, and the following surveys were done until 2012:

Year	Vessel	Valid hauls	Dates of hauls
1988	Cornide de Saavedra	115	8/7 – 22/7
1989	Cryos	116	12/7 – 1/8
1990	Ignat Pavlyuchenkov	113	18/7 – 6/8
1991	Cornide de Saavedra	117	24/6 – 11/7
1992	Cornide de Saavedra	117	29/6 – 18/7
1993	Cornide de Saavedra	101	23/6 – 8/7
1994	Cornide de Saavedra	116	6/7 – 23/7
1995	Cornide de Saavedra	121	2/7 – 19/7
1996	Cornide de Saavedra	117	28/6 – 14/7
1997	Cornide de Saavedra	117	16/7 – 1/8
1998	Cornide de Saavedra	119	17/7 – 2/8
1999	Cornide de Saavedra	117*	2/7 – 20/7
2000	Cornide de Saavedra	120*	10/7 – 28/7
2001	Cornide de Saavedra	120*	3/7 – 20/7
2002	Cornide de Saavedra	120	30/6 – 17/7
2003	Vizconde de Eza	177 (114)**	2/6 – 2/7
	Cornide de Saavedra	62***	7/6 – 17/6
2004	Vizconde de Eza	177 (124)**	25/6 – 2/8
	Cornide de Saavedra	61***	23/7 – 2/8
2005	Vizconde de Eza	176 (117)**	2/7 – 21/8
2006	Vizconde de Eza	179 (115)**	1/7 – 26/7
2007	Vizconde de Eza	174 (117)**	23/6 – 19/7
2008	Vizconde de Eza	167 (111)**	21/6 – 19/7
2009	Vizconde de Eza	178 (119)**	23/6 – 20/7
2010	Vizconde de Eza	153 (97)**	21/6 – 21/7
2011	Vizconde de Eza	127 (77)**	27/6 – 9/8
2012	Vizconde de Eza	174(118)**	26/6 – 24/7

*) 20 additional hauls were done every year to test the Campelen gear.

**) hauls in the original zone (less than 730 m depth) are in brackets.

***) parallel hauls for calibration.

From 1988 to 2002, the survey was carried out on board RV Cornide de Saavedra, covering the 19 strata defined up to 730 m (400 fathoms) depth (Figure 2); its primary objective was to assess the populations of cod and American plaice. In 2003, taking advantage of new fishing capacities of RV Vizconde de Eza, the surveyed area was increased to prospect 31 strata up to 1100 m (600 fathoms) depth, to cover the wider area of the Greenland halibut distribution, which was the commercial species of greatest interest to the EU fleet at that time. In 2004, the range of depths was extended up to 1460 m (800 fathoms) with 34 strata and it was reduced to 32 from 2008 onwards.

The results of the Surveys are presented systematically in the NAFO Scientific Council and they are published as research papers in the SCR Doc. series (Scientific Council Research Documents) (Vázquez 1989, Casas y González 2011, Vázquez 2012).

Calibration of RV Cornide de Saavedra versus RV Vizconde de Eza catch rates was made from 111 parallel hauls of the two vessels in the 2003 and 2004 surveys (González-Troncoso and Casas 2005); it allowed transforming the Cornide de Saavedra catch data in their equivalence in Vizconde de Eza scale, to produce homogeneous abundance indices series.

Importance of the survey

The main interest of the fisheries research in Flemish Cap is to know adequately the evolution of fishing grounds where cod, redfish and American plaice have traditionally been fished and, more recently, Greenland halibut, grenadiers and shrimp. Spain and Portugal are the EU countries most directly concerned in those fisheries.

Survey results provide independent information about the stock status of commercial fisheries, which is in some cases the only available information. The results are provided regularly to the NAFO Scientific Council, and they are also the base for many later studies.

These results are used by the NAFO Scientific Council to make an assessment on the state of the resources, which is the key tool for the NAFO Fisheries Commission to take the appropriate management measures. Results are used in the following stocks:

- cod (Div. 3M) – results are the only available fishery independent reference
- American plaice (Div. 3M) – “ “
- redfish (Div. 3M) – “ “
- Northern shrimp (Div. 3M) – “ “
- Greenland halibut (SA2 and Div, 3KLMNO) – together with two Canadian surveys
- roughhead grenadier (SA2+3) – together with two Canadian surveys

Furthermore, results have contributed to the preliminary identification of vulnerable marine ecosystems.

The Annex contains a list of publications supported by the survey series since 2002, when it was co-funded by the EU Data Collection Framework. The annex also lists several PhD theses where the information obtained from the survey series since its inception in 1988 was essential.

Survey design

The survey has a stratified random design, covering the area with 181 bottom trawl fishing stations, following the methodological specifications of NAFO (Doubleday 1981).

The adopted stratification of Flemish Cap is that described by Doubleday (1981), which considers 19 strata up to 730 m (400 fathoms) depth. Stratification was later extended by the Department of Fisheries and Oceans (DFO) of Canada (Bishop 1994) to cover up to 1460 m (800 fathoms) depth, considering 39 strata (Figure 2). Two strata of this bank (numbers 26 and 27) have fishing grounds unsuitable for trawling due to the huge abundance of sponges, and the same goes for the five strata belonging to the Beothuk Knoll (numbers 35-39) due, presumably, to the massive presence of corals. All these strata have been removed from the survey, resulting in the current 32 strata surveyed (Table 1). Each stratum is divided in rectangles of equal area. i.e. the number of rectangles is proportional to the stratum area (Figure 2b, inlet). A total of 478 rectangles are therefore considered in the current survey design (Table 1). Each rectangle is in turn divided in 10 fishing units of equal area, leading to 4780 possible bottom trawl fishing hauls (Figure 2b).

Trawl station methodology

The selection of the hauls is set with the following conditions:

- The number of hauls in each stratum (Table 2) is fixed, distributed proportionately to the number of units, and ensuring at least two hauls by stratum.
- Hauls (fishing units) are randomly chosen within each stratum with the following constraints: only one haul can be selected within a given rectangle, and two hauls cannot coincide in adjacent fishing units.
- Information from previous surveys and commercial fishing is used to eliminate hauls in unsuitable fishing grounds.
- The allocation of the hauls into each fishing unit could be made more accurate using the bathymetry of the area obtained by the NEREIDA project, reducing the risks of snagging in the bottom.

In accordance with Table 2, 181 hauls will be selected at random, 120 of them in less than 730 m depth.

The criterion used to change the position of a previously selected random haul has always been the information from the commercial fishing and from previous surveys about the suitability of the bottom trawling. This information is contrasted with the more detailed bathymetric charts of the bottom that have been developed in the project NEREIDA.

Criteria for rejecting a haul:

- Snag of the trawling gear in the bottom.
- Damages in the cod-end or severe damages in large sections of the wings or belly.
- Less than 20 minutes of effective trawling time.
- Gear malfunction, i.e., when it is considered that gear contact with bottom was not correct, or the geometry of the gear was not maintained properly through the whole trawl.

Rejected fishing hauls means that, because standard conditions were not achieved, such station cannot be used to quantify the biomass and abundance neither to determine the structure of the population. However, the specimens caught in any non-valid hauls can be used to make all kind of biological sampling.

Vessels

The survey is carried out on board RV Vizconde de Eza, 1400 GRT, 1800 kW.

Former RV Cornide de Saavedra was 1200 GRT and 1680 kW.

Fishing gears

The trawling gear used is the Lofoten (NAFO 1990) (Figure 3), built and rigged as specified in Table 3. This gear is similar to that used by the commercial fleet engaged in American plaice fishing on Flemish Cap in the years when the survey started. It is characterized by being well adapted to the frequent hard bottoms of the bank, and it showed good performances throughout the years.

The cod-end mesh size is 35 mm, which is adequate for fishing juveniles of most important commercial species, particularly for cod at age one.

The cod-end mesh size (35 mm) is inefficient to retain juvenile shrimps (ages 1 and 2), and delays in one or two years the estimation of each new year-class entering the fishery. After several attempts in different surveys, an auxiliary net bag of 10 mm mesh size is used since 2000 to retain the youngest individuals of shrimp escaping through a small square of the cod-end. The base of the bag is a diamond of 36 cm in each side, and it is attached to the cod-end in a central-dorsal position, 26 cm from the seam end, just in a position where it is believed that the escape is maximum (Aschan and Sunnanå 1997).

Personnel

Catch sampling (2 teams of 5 persons each)	10
Taxonomy	1
Identification of redfish species	1
CTD and data processing	1
Survey leader	1
TOTAL	14 scientists

Every two years two persons will carry out feeding studies with exclusive dedication. In those years, the identification of redfish species and taxonomy tasks will be carried out by the teams in charge of catch sampling.

Standardization

Daily fishing period: 6.00 to 22.00

The target trawling speed is 3.5 knots. It is not possible to maintain the speed when trawling at deeper grounds due to insufficient weight of the trawl doors used. While this problem is not solved, deeper sets are made at the highest speed possible, which is always around 3.00 knots.

The 30 minutes trawling time is counted from the moment the gear, after its first contact with the bottom, acquires its characteristic mouth opening, until the beginning of the haul in. Its control is done, whenever possible, by using net sounders (ITI or SCANMAR), which enables accurate measures of those times. The start of the haul in is kept as the haul's end to be consistent with previous criterion used (with the exception of 2005).

In the surveys previous to 2003, when net sounder was not available, the 30 minutes were counted according to the expression:

$$t \text{ (min)} = 32 + \text{depth (m)} / 100$$

where t was the time between the end of the cable veering and the start of haul in. This criterion was established in the 1992 survey, in which it was made a systematic control of the gear behaviour with the SCANMAR sounder. The interpretation that was then made for counting the trawling time per haul was "as long as the art is in contact with the bottom before the start of the haul in" (Vázquez 1993).

Whenever possible during the haul in, the time when the gear loses its characteristic shape and it moves off the bottom is also recorded, which reveals the effective trawling time and, in his day, apply corrections to previous surveys.

As will be indicated (VI-Results, Estimates), the catch data are transformed to catch per trawled mile for processing, provided that fishing has lasted at least 20 and no more than 40 minutes.

The order of execution of selected stations is determined during the survey, setting each day the hauls to be held the next day, trying to minimize the routes between stations. A detailed plan of the order of the stations is impractical because it is necessary to make changes due to unforeseen malfunction of the gear (e.g. obstruction, breakages...).

The distance travelled in each haul is the geographical distance between the GPS positions of the start of the haul (when the gear comes into contact with the bottom and it acquires its characteristic shape) and the start of the haul in (when cable starts to be recovered).

The length of the wire released is determined by the following relationship (meters) and the results are in Table 4.

$$\text{Cable length} = 2 * \text{depth} + 200$$

The dimensions of each gear in use are verified using the data forms in Figure 3, by reporting in each cell the observed value versus the specified one. Groundrope and bobbins dimensions are detailed in Figure 4.

Dates

The survey starts in the second half of June, and needs 30 fishing days.

Data collection

A haul's data form (see annex) is filled in each set. It will contain information gathered in the bridge during and immediately after finishing the haul, as well as catch information by species. This form is available in the sampling area before sorting the catch starts.

The personnel in charge of analysing the catch is divided into two shifts of five people each, with the following schedule:

	Hour																	
	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23
Team A																		
Team B																		

The teams exchange shifts in the middle of the survey (90 hauls).

The designated *Team Leader* is responsible for distributing the work for the rest of the team and for verifying that all data forms are covered adequately by reviewing them at the end of the haul analysis. It is also responsible for report to the other *Team Leader* of the work that remains when the haul analysis is not completed during the shift.

Catch record

One member of each team, and always the same, is responsible for logging the catches in the haul's data form (see annex). All fish species, as well the commercial cephalopods and crustacean are recorded. Non-commercial invertebrates, sponges and corals among them, are recorded since 2007 in a specific data form (see below). All species should be identified. Species already found are listed in the Annex, as well their working codes. Their occurrence in past surveys is listed by Vázquez et al. (2013).

Species that are not possible classify immediately are labelled with the number of the station and with a letter by alphabetical order (e.g. #xx species A, #xx species B, ...), weighed and recorded in the summary report with the sequence described. Unidentified species are then set aside and sent to the dry laboratory for proper identification. The identification is made by a scientist appointed at the beginning of the survey (and always the same scientist) and regardless whether it is able to identify the samples, the specimens are frozen and stored in the freezer.

Sometimes redfish catch is excessive to sort all individuals (the Survey Leader or in his place the Team Leader determines when a catch is excessive). In these hauls, all species other than the redfish are separated and processed as usual. A number of boxes (number defined by the Team Leader) of redfish are separated for sampling, these boxes are chosen randomly without separating the three species and juveniles (see redfish sampling section below). The rest of the redfish catch is not weighed, but put in boxes; these boxes are counted and thrown overboard. The person in charge of recording catches places near the discard strip, counts and records the number of discarded boxes. He is also responsible for controlling that only redfish is discarded and the rest of the species are separated and transported to the sorting zone.

Later, total catches of redfish species are calculated by extrapolation. In this case it is necessary to state, on the reverse of the haul summary report, the number of discarded boxes and the number of boxes with redfish that were sorted.

Length sampling

The length sampling follows the sampling NAFO recommendations on length ranges and sex discrimination (NAFO 1999)⁴ (Table 5). Therefore, the length measurements of fish are made on the total length and to the centimetre below, except the redfish (*Sebastes spp*) which is measured to the fork length and also to the centimetre below. The standard for the Grenadiers (pre-anal fin length⁵ to the half centimetre below) extends to: *Macrourus spp.*, *Coryphaenoides spp.*, *Nezumia spp.* and *Trachyrhynchus spp.* Mantle length to the half centimetre below is measured for cephalopods, and carapace length (distance from the posterior edge of the optic pit to the postero-dorsal edge of the carapace) to the half millimetre below for shrimp.

Following the same recommendations, length sampling is performed independently by sex in all flatfish, redfish, cod and grenadier (*Macrourus berglax*). In these species, the selected sample is classified by sex before measuring lengths in order to reduce the causes of error.

Table 5 shows how the sample data is submitted for consideration by the NAFO Scientific Council. This does not exclude the possibility of the data being gathered in greater detail; therefore, for example, fish whose frequencies are grouped into 2 or 3 centimetres are always measured to the centimetre below for convenience. Likewise, although the NAFO Scientific Council considers that the separation of the sexes in cod sampling is not required, this species is sampled independently by sex since 2010.

⁴ This recommendation was approved by the NAFO Scientific Council in 1974 (NAFO 1974, NAFO 1975b). The standard for the grenadiers was afterwards amended (NAFO 1980, p. 94, NAFO 1984, p. 75).

⁵ Pre-anal fin length is considered the distance between the end of the snout and the first ray of the anal fin.

As a rule, all individuals in the catch are measured. Exception is made when the capture of some species is very abundant and then a subsample containing, as reference at least 200 individuals, is measured. This is a situation that occurs frequently with redfish; in these cases the number of boxes to be measured is determined by the person in charge of this species and the details of the capture and the selection of the sample are clearly reflected in the back of the haul's data form.

When the capture of a species is very large and there is a large number of small fish and few large fish, it is not sometimes reasonable to measure all of them; in these cases the sampling is done independently for each size groups and details are noted on the reverse of the haul summary report. This procedure must be exceptional

The sample weight is always recorded in the haul summary report, whether or not equal to the capture.

The lengths shall be recorded in the appropriate data form for this purpose (see Annex). After the measurement, it is clearly indicated the beginning and end of the size range and measures are counted and registered the total number of individuals measured by size. For species with sex discrimination, frequencies are recorded in separate columns with indication of the sex on headings. Measurements of species which are measured in half-centimetre below are recorded in a specific data form.

The length frequency of each species is recorded in separated data forms. Only in the case of the species in which the numbers of individuals is very low, the length frequency of several species are recorded in the same form, always clearly stating to what species refers each measurement. Length measurements are made by at least two persons, so that always a person measure and other records.

Biological sampling

In each haul a full biological sampling is done for all or at least two species of the following ones: cod, American plaice, *Sebastes marinus*, *S. mentella*, *S. fasciatus*, juvenile *Sebastes*, Greenland halibut and roughhead grenadier. This biological sampling includes:

- length
- round weight
- sex
- gonad collection (if applicable)
- gutted weight (gonad free)
- otolith collection

The target sample size within each haul and for each species is 50 fish, except for juvenile *Sebastes* which is only 20. In the species where the size sampling is done by sex, the sample is separated by sex before measurement; the sample goal is measuring 25 fish of each sex. The purpose of this sampling is to get at least 10 fish of each cm or ½ cm length (Table 5) and sex (male and female, or indeterminate) at the end of the survey.

Only female gonads (ovaries) are collected. The number of ovaries collected by species and size range is as follow:

Species	Minimum length (cm)	Gonads per cm
Cod	25	6
American plaice	25	10
Greenland halibut	40	10
Roughhead grenadier	20	6 (1/2 cm)
Redfish - <i>marinus</i>	20	10
Redfish - <i>mentella</i>	18	10
Redfish - <i>fasciatus</i>	15	10

The total number of gonads collected is always smaller than otoliths, therefore in the biological sampling form it is requested to annotate in the column “G” when a gonad is collected. This allows for precise control of the fish sampled by length and the number of ovaries collected. Control sheets for each species are available at the beginning of each day based on data collected from previous hauls. It is requested that all fish which ovaries are collected also the otolith are taken.

Gonads are stored in perforated (to allow formalin to penetrate) plastic bags with the corresponding label where it is written the name of the species, number of the haul, the number of the observation and the fish size. Redfish is labelled with its scientific name (*S. marinus*, *S. mentella*, *S. fasciatus*). When sampling work of a haul is finished, and not later, bags are closed and stored in their corresponding container. The fixative is phosphate based neutral buffered 4% formaldehyde (10% formalin), normally produced in 20 l batches: 18 l of water, 2 l of 35-40% high grade formaldehyde, 163.8 g of sodium phosphate dibasic dihydrate and 81.4 g of sodium phosphate monobasic monohydrate.

Biological sampling is stratified by fish length and sex, and continues till having completed the required number in each length class.

A simpler sampling is done to other species which include measuring only length and weight, to calculate their relationship, which is routinely used to estimate the sampling weight based on length composition and confirm the correctness of the catch records.

The requirement of sampling shrimp (measuring lengths of several stages of maturity) makes its sampling sometimes very time consuming to be done on each haul. If this happens, a random sample of approximately 1-2 kg is frozen to be worked out later. Two people from each team are responsible for working the samples (fresh or frozen). It is intended that the samples of shrimp caught are measured and recorded in the ARGO program before the end of the survey. Once on land, it is necessary to make a length and weight sample, so in addition to samples for length measurements, specific samples are frozen for the study of the length-weight relationship in the laboratory.

Likewise, the shrimp caught in the small net bag with 10 mm mesh, placed in the dorsal-central part of the cod-end, is collected, the catch recorded independently (*Pandalus* bag, *Sebastes* bag) and individuals measured by maturity stage. If the whole catch is not measured, the sample weight is recorded as in the rest of the species.

Redfish (*Sebastes spp.*) sampling

As pointed earlier, three species of redfish occur in Flemish Cap: *Sebastes mentella*, *S. marinus* and *S. fasciatus*. The morphological resemblance between *S. mentella* and *S. fasciatus* is high, making difficult their identification in a quick and routinely way, and both are commonly designated as beaked redfish. In 1988 and 1989 these species were not classified and hence two redfish entities were considered: *S. marinus* and beaked redfish, the latest included the unclassified fish (roughly those smaller than 15 cm) of the three species. Since 1990 the unclassified small fish, i.e. those where *S. marinus* was not possible to be distinguished from beaked redfish, was considered as a different category, named juvenile redfish. It is important to stress that many of the specimens classified into species are sexually immature, i.e. juveniles too. Therefore the category “juvenile redfish” does not refer to all sexually immature redfish but only those not able to be classified into species.

In 1990 and 1991, a subsample of beaked redfish catch (50 specimens) from each haul was classified into species (*S. mentella* and *S. fasciatus*) using the gasbladder musculature (Ni, 1981; Power and Ni, 1982) which is a more precise attribute than external characteristics. The fish dissection and musculature inspection was done but a designated expert in charge of this task solely. In 1990 these specimens were used exclusively for individual biological sampling, i.e. length, weight and age, as they were not taken randomly. In 1991, the subsample (maximum of 150 Kg) was taken randomly and then used also to estimate the haul catch species composition.

In 1991, during the process of redfish identification, it was developed sufficient skills to distinguish *S. mentella* and *S. fasciatus* by their external appearance. As a consequence, since 1992 the three redfish species are routinely separated in the whole catch, except the smallest ones still considered as juvenile redfish. A number of specimens were still dissected for inspecting the gasbladder musculature; these included those fish still difficult to classify by their external morphology, but also a selection of fish at different sizes for checking the correctness of the classification. The number of dissected fish reduced in each survey as experience was gained. The same person classified redfish species from 1992 to 2002, and since then different trained persons are in charge of this task. In the current procedure still a number of specimens are still dissected for inspecting the gasbladder musculature.

To optimize the identification process, difficult individuals are often retained for a later and more detailed identification; those individuals are generally the ones with smallest size. A violation of the random sampling criterion could occur if those selected fish, once they are classified, are not reintegrated to their corresponding sampling group/box, i.e. if all of them are put in the same box, and length sampling does not requires all boxes, a skew sampling will occur. Special care is taken to avoid this situation when a subsample for length measurements is taken.

The target of biological sampling is to get 20 otoliths by cm and sex from the whole bank. A data form is used to facilitate 2 individuals for each sex, cm and stratum are collected. It may allow getting larger sizes, which are scarce or only occurring in few strata. The person responsible for redfish classification does the biological sampling control.

Shrimp (*Pandalus borealis*) sampling

Shrimp is a protandrous hermaphrodite species. Every individual is born and mature as male, and after a transition period, becomes female. In some cases the external male characters are hardly visible. Sex can be identified attending to the external structure of the endopod of the first pair of pleopods.

Shrimp sampling requires some specific consideration: the absence of hard parts for ageing makes it necessary to obtain good length distributions to identify year-classes through modal analysis. Each length frequency sample usually contains around 300 individuals. Sex and maturity are recorded according to the following categories: "male", "transition", "primiparous female", "multiparous female" and "ovigerous female". Oblique carapace length (CL) is measured with a gauge from the optic pit's back edge on the postero-dorsal edge of the lower half millimetre carapace.

Additional samples are taken for study in laboratory to calculate the length-weight relationship. These samples are frozen on board. Each individual is measured in the laboratory to the hundredth of millimetre and hundredth of gram accuracy. Samples are taken from all strata.

Benthonic invertebrates

Non-commercial invertebrates are sorted after fish and commercial invertebrates, and after taken a picture of the whole invertebrate catch. Additional photos of rare or first recorded species are taken. Catch records are written down in a specific data form (see Annex), where weight and number of each best identified group is noted, as well as any observation, e.g. on photos taken. A sample should be taken when catch is too large.

Available guides in NAFO are:

- Coral Identification Guide NAFO Area, (Kenchington *et al.* 2009)
 - Sponge Identification Guide NAFO Area (Best *et al.* 2010)
- <http://www.nafo.int/publications/frames/science.html>

All specimens of less frequent species are retained, particularly those from species not included in the invertebrates' identification cards or those with uncertain or incomplete classification. Samples are stored in plastic bags, labelled with survey, haul and species, and they are preserved in the appropriate conservation media.

Corals and sponges are of primary interest in studying vulnerable marine habitats, so special attention is paid in recording catches and retaining samples.

- Corals: all corals not included in the identification cards are preserved. A piece of each colony of Gorgonians and Antipatarians marked with an * in the invertebrates form is also retained, and the remaining frozen after a photo is taken.
- Sponges' large catches: a photo is taken and samples of main species retained. In large specimens only a piece, like a cheese slice, containing most important elements is taken and preserved.

Sampling fixatives are:

- 70% alcohol for cnidarians (actinides excluded), crustaceans, molluscs and Equinodermata.
- 4% formalin for Polychaeta, sponges, actinides and all others.

Taxonomy

All species should be identified. List of species already found are listed by Vázquez et al. (2013). Their working codes are presented in the Annex. When species are not easily recognized, the item is reserved for late identification, putting a note on the haul's data form. Those items are identified on board to the most precise taxonomic level using the appropriate references.

If species are not identified, individuals are labelled, frozen and stored for their study in the lab. When the species is a prime cite for Flemish Cap, the item is photographed and adequately stored.

It is necessary to complete the photographic record of species in the area.

Physical oceanography

Temperature and salinity profiles are taken with a CTD according to a predefined square grid (Figure 5). It contains 80 stations with 15 nautical miles mean distance among them. Stations are done when time is available, trying to disturb the hauls program as little as possible, and anyway before the first haul of the day. A person is in charge of the whole issue.

Data are incorporated to tie IEO data base, and copy are sent to the *Integrated Science Data Management* (ISDM, former MEDS), the Canadian data base for that zone.

Feeding sampling

Feeding sampling is done every two years as a minimum, and it is under the exclusive task of two persons. To impede that the same specimens be required for different samplings, feeding sampling is normally done after catch sampling is finished and closely coordinated with the standard biological sampling work in such a way that the same fish measured in biological sampling are also analysed for their stomach contents. If there are sufficient individuals in the catch feeding analysis can be done on fish where biological sampling was not performed.

Studied species are:

<i>Sebastes fasciatus</i>	<i>Macrourus berglax</i>
juvenile redbfish	<i>Nezumia bairdi</i>
<i>Sebastes marinus</i>	<i>Phycis chesteri</i>
<i>Sebastes mentella</i>	<i>Urophycis tenuis</i>
<i>Gadus morhua</i>	<i>Amblyraja radiata</i>
<i>Hippoglossoides platessoides</i>	<i>Amblyraja hyperborea</i>
<i>Reinhardtius hippoglossoides</i>	<i>Bathyraja spinicauda</i>
<i>Glyptocephalus cynoglossus</i>	<i>Malacoraja senta</i>
<i>Anarhichas denticulatus</i>	<i>Rajella fyllae</i>
<i>Anarhichas lupus</i>	<i>Lycodes reticulatus</i>
<i>Anarhichas minor</i>	<i>Centroscyllium fabricii</i>

The target sample size is considered by fish length of 10 cm (0-9, 10-19, 20-29 cm, etc.) total length (LT) to the lower cm for most of the species, except for grenadiers, where 5 cm range (0-4.5, 5-9.5, 10-14.5 cm, etc.) pre-anal fin length (LPA) to the lower half cm is used. Length to the notch of the dorsal fin (LEAD) to the lower half cm is used for wolffish.

Data taken to each fish are: length, sex, sexual maturity, round weight and total stomach repletion in volume measured with a *trophometre* (Olaso 1990). Stomach content analysis is done on board, identifying all components to the lowest possible taxonomic level, and noting for each prey: number, digestion level (fresh, half-digested and full-digested), percentage of total volume, length in mm, and size and number of hard pieces.

Fish with everted stomach, which is common in roughhead grenadier (*M. berglax*) and redbfish (*Sebastes spp.*), or containing preys taken in the cod-end, which is common if dogfish (*Anarhichas spp.*), are discarded. Fish with total or partially regurgitated food, which is common in rays, cod and roundnose grenadier (*C. rupestris*) are only considered for calculating feeding intensity indices. Size and colour of the bladder is observed and recorded to distinguish an empty stomach or containing scarce food from a total or partially regurgitated.

Storing data in electronic media

All data recorded during the survey are entered in a computer as soon as possible, as data is validated and potential errors corrected in an easy way. The data collected each day is always inputted before the next day work starts, to allow updating control of samples already taken.

After stored, data from each fishing haul will be printed to verify that the stored information is equal to that in the forms. Printing formats should be similar to that of the forms in use.

Data are stored and initially managed in an *ad hoc* program, ARGO. The system provides a reliable way of data storage and elaboration of results, as well as the possibility of transferring data to any other programs. Once they are corrected, they are transferred to the shared database SIRENO, which is managed by the IEO.

Data analysis

Results

Initial data analysis, done on-board after the end of the survey, provides the following preliminary results, without prejudice to a later and detailed analysis:

- Biomass and abundance estimates for all species and comparison with some previous surveys figures.
- Shrimp stock structure.

Later work in laboratory includes:

- Concluding otoliths ageing for fish target species, and estimating their abundance at age.
- Concluding shrimp length distribution analysis, using program MIX to identify their normal distributed components.
- Performing the histological analysis of gonads to determine maturity ogives of fish target species.
- Processing and analysing CTD data.
- Reviewing benthonic invertebrates' records based on photos taken in each haul to validate identification done on board.
- Updating benthonic invertebrates notes: list of species, and their distribution and abundance.
- Updating the photographic collection of species.

Calculations

Biomass and abundance estimates are calculated by the swept area method, which produces quite useful approximations of stock sizes, even if they admittedly sub-estimate real values, so they should be interpreted as indices and not as absolute figures. Total biomass is calculated with the expression:

$$B = \sum_e (Cpm_e \times area_e / 0.0075)$$

- B** – total biomass
e – stratum
Cpm_e – mean catch per mile of hauls in stratum **e**.
area_e – area of stratum **e** in square miles.
 0.0075 – accepted width of the fishing gear in miles.

And its standard error:

$$s_B = \sqrt{[\sum_e V(Cpm_e) \times (area_e / 0.0075)^2]}$$

$$V(Cpm_e) = \sum_p [(C_p / m_p - Cpm_e)^2 / (n_e - 1) / n_e]$$

- p** – any haul in stratum **e**
C_p – catch in haul **p**
m_p – miles towed in haul **p**
n_e – number of hauls in stratum **e**

Length frequencies that correspond to the previous total biomass estimates are also calculated based on data per mile in each haul:

$$F(t) = \sum_e (fpm(t)_e \times area_e / 0.0075)$$

$F(t)$ – frequency at length t

e – stratum

$fpm(t)_e$ – mean frequency per mile for length t of hauls in stratum e .

$area_e$ – stratum e area in square miles.

0.0075 – accepted width of the fishing gear in miles.

When only female biomass is the issue, like in the case of shrimp, calculations are the same but they are based on the female catch at each haul. Total catch is the only recorded at each haul, so this amount is divided between male and female in the same proportions as their respective SOP⁶.

Missing data

Some strata were not visited in 1993, and only one haul was done in some stratum in 2011. Missing biomass and length frequencies estimates follow different procedures in each case.

If no haul is done in a stratum, biomass is estimated by comparison among data of that survey and those of previous 5 years surveys, adjusting values to a multiplicative model with two factors: year and stratum (Vázquez and Larrañeta 1980):

$$B_{e,a} = E_e * A_a$$

being:

$B_{e,a}$ – biomass of stratum e in year a

E_e – stratum e factor

A_a – year a factor

Factors are calculated by iteration until convergence of the equations;

$$E_e = \sum_a (B_{e,a} / A_a * w_{e,a}) / \sum_a w_{e,a}$$

$$A_a = \sum_e (B_{e,a} / E_e * w'_{e,a}) / \sum_e w'_{e,a}$$

being:

$$w_{e,a} = A_a^2 / V(B_{e,a})$$

$$w'_{e,a} = E_e^2 / V(B_{e,a})$$

$$V(B_{e,a}) = B_{e,a} \text{ variance} = B_{e,a}^{1,8}$$

This assumed variance is preferable to the one calculated in each stratum and year to prevent very low results due to a reduced number of hauls.

Length frequencies of non-visited strata were calculated by extrapolation of results in the whole visited strata, using the same proportion as biomass estimates.

When only one haul was done in a stratum, biomass is calculated as the mean between the one estimated with only that haul (B_1) and the result obtained by the former described method for non-visited strata (B_0). If length sampling

⁶ Sum Of Products: sum of length frequencies times mean weight at length.

was also available for that single haul, length frequencies of the stratum will be, like in the case of biomass, the mean between haul frequencies extrapolated to biomass B_1 and frequencies of total sampled biomass extrapolated to B_0 . If no such sampling was available, length frequencies will be calculated by extrapolating frequencies of total sampled biomass to $(B_1 + B_0) / 2$.

For age-length keys, when a length lacked assigned ages, frequencies at age for that length are assumed to be equal to an age-length key done with data of the previous five surveys.

Validation of Survey Results

All collected data are recorded in paper, in the forms available for each class of data, and later stored in a PC. Verification of the stored information follows these steps:

- 1 – Once all data of a haul have been typed, sample catch must be compared with SOP (based on length frequencies of the sample): if an important difference between them exists both original and recording are revised. Consistency between length distribution and biological sampling is also checked.
- 2 – All the data of each haul is printed to check that all typed information is equal to the one in the forms. Printing uses similar formats to those of the original forms for an easy reading and comparison.
- 3 – Once the survey is finished, length-weight relationships for each species are updated and discrepancies again checked. When discrepancies are important (greater than 15%) and the source of them detectable, corrections are done. All corrections are made also in the original forms but never erasing the original annotation but using a red ink pen to write down the change. This allows in the future an easy traceability of the modifications made during or after the survey.

Survey results are regularly presented to the NAFO Scientific Council (Casas y González-Troncoso 2011, Vázquez 2012).

Acknowledgements

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Table 1 – Specification and characteristics of the survey area, and number of selected hauls.

	Area sq. miles	Strata	Rectangles	Fishing units	Selected hauls
depth < 730 m	10 555	19	309	3 090	120
depth: 730-1460 m	5 515	13	169	1 690	61
Total	16 070	32	478	4 780	181

Table 2 – Stratification of Flemish Cap and hauls plan.

Stratum	Depth interval (fathoms)	Area (sq. miles)	Fishing units	Selected hauls
1	70- 80	342	100	4
2	81-100	838	250	10
3	101-140	628	180	7
4	"	348	100	4
5	"	703	200	8
6	"	496	150	6
7	141-200	822	240	9
8	"	646	190	7
9	"	314	90	3
10	"	951	280	11
11	"	806	240	9
12	201-300	670	200	8
13	"	249	70	3
14	"	602	170	7
15	"	666	200	8
16	301-400	634	190	7
17	"	216	60	2
18	"	210	60	2
19	"	414	120	5
20	401-500	525	160	6
24	"	253	80	3
28	"	530	160	6
33	"	98	30	2
21	501-600	517	160	6
25	"	226	70	3
29	"	488	150	6
32	"	238	70	2
34	"	486	150	5
22	601-700	533	160	6
30	"	1134	350	11
23	701-800	284	90	3
31	"	203	60	2
Total (strata 1-34)		16 070	4 780	181

Table 3 – Technical data of the survey. Characteristics and deployment of the fishing tackle.

procedure	specification
Survey type Haul selection method Criterion to change position of a selected haul Criterion to reject a haul Daily fishing period Species to be sampled Species for aging	Stratified sampling Random Bottom unsuitable for trawling according to commercial fishing or former surveys. - snag in the bottom - severe damages in the net or in the cod-end - trawling time inferior to 20 minutes - gear malfunction 6:00 to 22:00 local time All fishes, cephalopods, shrimp and non-commercial invertebrates. cod, American plaice, redfish, Greenland halibut and roughhead grenadier.
Vessel TRB Power Maximum trawling depth Area to be surveyed Time to survey	RV Vizconde de Eza 1400 GT 1800 kW 1460 m Div. 3M (depth < 1460 m) 30 days
Fishing gear Groundrope / headrope Groundrope Floats Bridles Vertical opening Horizontal opening Rigging warps Trawl doors Wire Wire length Cod-end mesh size Towing speed Trawling time	Lofoten 17.70 m /31.20 m 27 steel bobbins Ø 35 cm Ø 20 cm (2 × 16) + Ø 24 cm × 20 8 m Ø 16 mm 3.5 m 14 m = 0.0075 miles 100 m, 45 mm, 200 kg/100m Oval polyvalent, 850 kg Ø 20 mm 2 × Depth + 200 m 35 mm 3.5 knots 30 minutes of effective fishing time determined by net sounder or “32 + depth (m)/100” minutes from the time the winches are locked.

Table 4 – Trawling wire length as a function of depth, both in metres.

depth	length
100	400
150	500
200	600
250	700
300	800
350	900
400	1 000
450	1 100
500	1 200
550	1 300

depth	length
600	1 400
650	1 500
700	1 600
750	1 700
800	1 800
850	1 900
900	2 000
950	2 100
1 000	2 200
1 050	2 300

depth	length
1 100	2 400
1 150	2 500
1 200	2 600
1 250	2 700
1 300	2 800
1 350	2 900
1 400	3 000
1 450	3 100

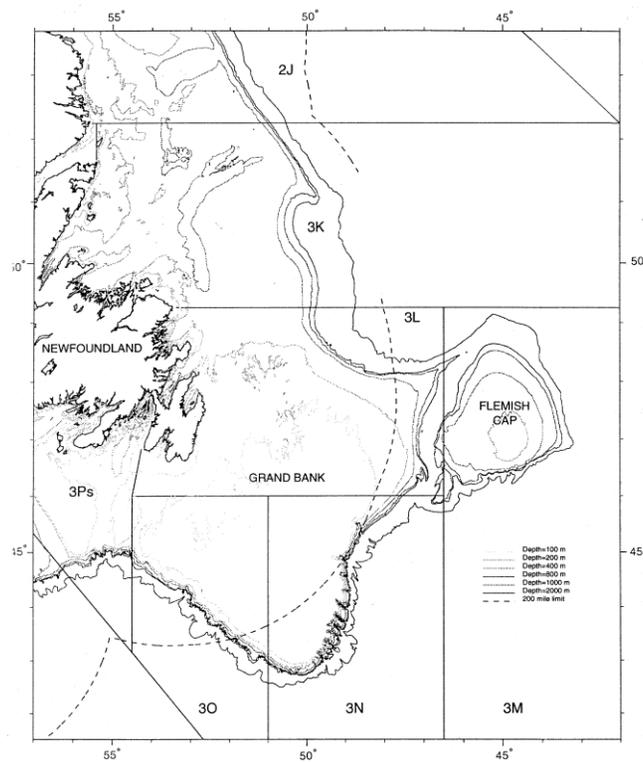
Table 5 – Length intervals and sexing criteria for sampling data transmission to NAFO (NAFO 1999, page 10)

Atlantic cod (<i>Gadus morhua</i>)		3
Pollock (<i>Pollachius virens</i>)		3
Cusk (<i>Brosme brosme</i>)		3
White hake (<i>Urophycis tenuis</i>)		3
Wolffishes (<i>Anarhichas</i> sp.)		3
Striped wolffish (<i>Anarhichas lupus</i>)		3
Spotted wolffish (<i>Anarhichas minor</i>)		3
Haddock (<i>Melanogrammus aeglefinus</i>)		2
Red hake (<i>Urophycis chuss</i>)		2
American plaice (<i>Hippoglossoides platessoides</i>)	(by sex)	2
Witch flounder (<i>Glyptocephalus cynoglossus</i>)	(by sex)	2
Yellowtail flounder (SA 3-4) (<i>Limanda ferruginea</i>)	(by sex)	2
Greenland halibut (<i>Reinhardtius hippoglossoides</i>)	(by sex)	2
Atlantic halibut (<i>Hippoglossus hippoglossus</i>)	(by sex)	2
Summer flounder (<i>Paralichthys dentatus</i>)		2
Greenland cod (<i>Gadus ogac</i>)		2
Redfishes (<i>Sebastes</i> sp.)	(by sex)	1
Golden redfish (<i>Sebastes marinus</i>)	(by sex)	1
Beaked redfish (<i>Sebastes mentella</i>)	(by sex)	1
Silver hake (<i>Merluccius bilinearis</i>)	(by sex)	1
Yellowtail flounder (SA 5-6) (<i>Limanda ferruginea</i>)	(by sex)	1
Winter flounder (<i>Pseudopleuronectes americanus</i>)		1
Windowpane flounder (<i>Scophthalmus aquosus</i>)		1
Polar cod (<i>Boreogadus saida</i>)		1
Scup (<i>Stenotomus chrysops</i>)		1
Spotted hake (<i>Urophycis regia</i>)		1
Atlantic herring (<i>Clupea harengus</i>)		1
Atlantic mackerel (<i>Scomber scombrus</i>)		1
Atlantic butterfish (<i>Peprilus triacanthus</i>)		1
Atlantic menhaden (<i>Brevoortia tyrannus</i>)		1
Alewife (<i>Alosa pseudoharengus</i>)		1
Argentines (<i>Argentina</i> sp.) (by sex)		1
Black seabass (<i>Centropristis striata</i>)		1
Blueback herring (<i>Alosa aestivalis</i>)		1
Roundnose grenadier (<i>Coryphaenoides rupestris</i>)	(by sex)	1/2 cm or 5 mm
Roughhead grenadier (<i>Macrourus berglax</i>)	(by sex)	1/2 cm or 5 mm
Capelin (<i>Mallotus villosus</i>)	(by sex)	1/2 cm or 5 mm
Squid (<i>Illex</i> sp. and <i>Loligo</i> sp.)	(by sex)	1/2 cm or 5 mm
Sea scallop (<i>Placopecten magellanicus</i>)		1/2 cm or 5 mm
Northern prawn (<i>Pandalus borealis</i>)		1/2 cm or 5 mm

Note: Any other species not listed above should initially be reported in 1-cm groups.

Table 6 – Sampling objectives for otoliths and gonads

Species	Otoliths	Gonads	Length range for gonads
Cod	10 by cm and sex	4 by cm	≥ 25 cm
<i>A. plaice</i>	30 by cm and sex	5 by cm	≥ 25 cm
<i>S. marinus</i>	20 by cm and sex	10 by cm	≥ 10 cm
<i>S. mentella</i>	20 by cm and sex	10 by cm	≥ 10 cm
<i>S. fasciatus</i>	20 by cm and sex	10 by cm	≥ 10 cm
Juvenile <i>Sebastes</i>	20 by cm	---	---
<i>G. halibut</i>	10 by cm and sex	2 by cm	≥ 30 cm
<i>M. berglax</i>	10 by ½ cm and sex	4 for each ½ cm	≥ 20 cm

**Figure 1** – Situation of Flemish Cap: depth contours and dashed line indicating the 200 miles Canadian EEZ.

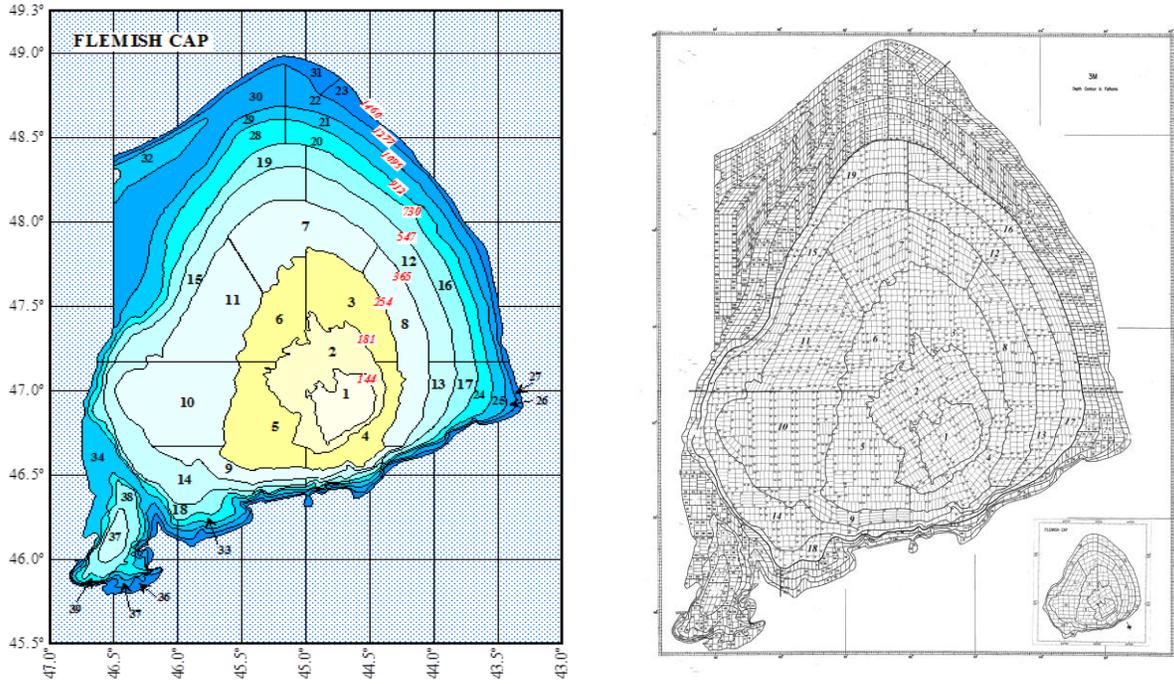


Figure 2 –Flemish Cap stratification: left) Distribution of the 39 strata, right) fishing units in the whole bank and rectangles of the first 19 strata (inlet).

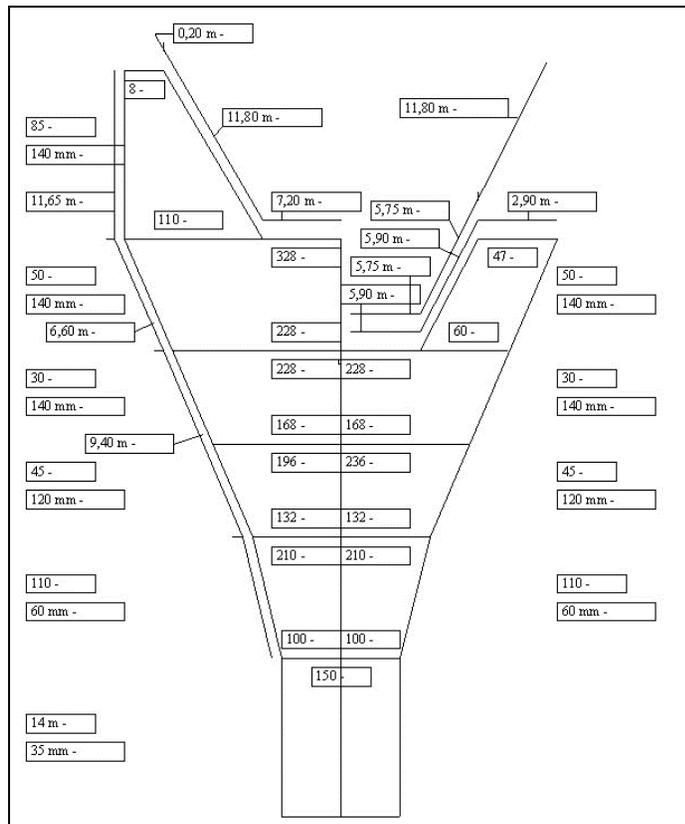


Figure 3 –Dimensions of the Lofoten trawl gear (31.20 m x 17.70 m)

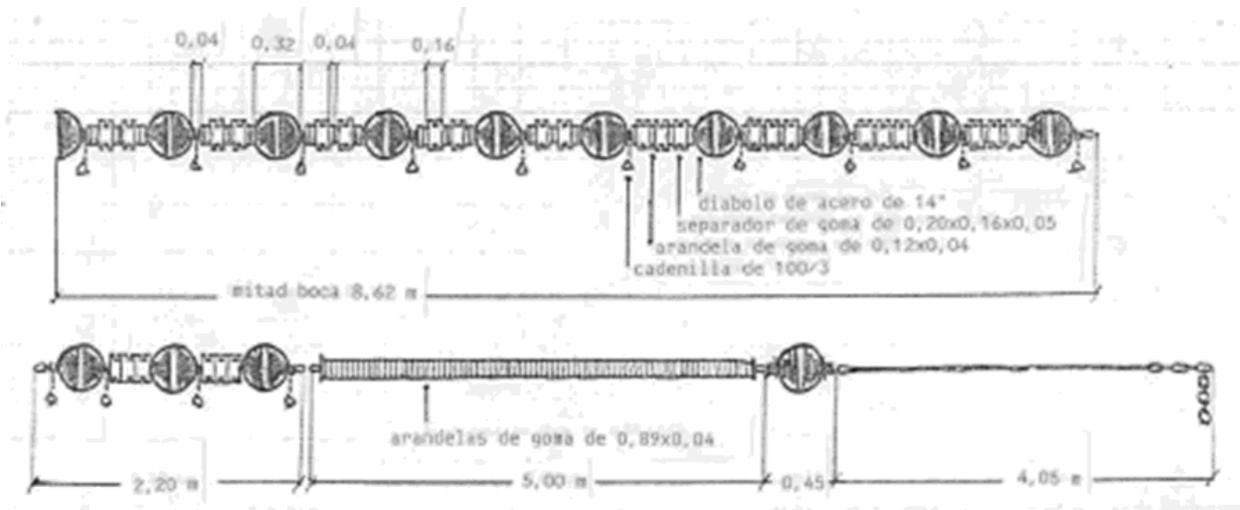


Figura 4 – Groundrope rigging.

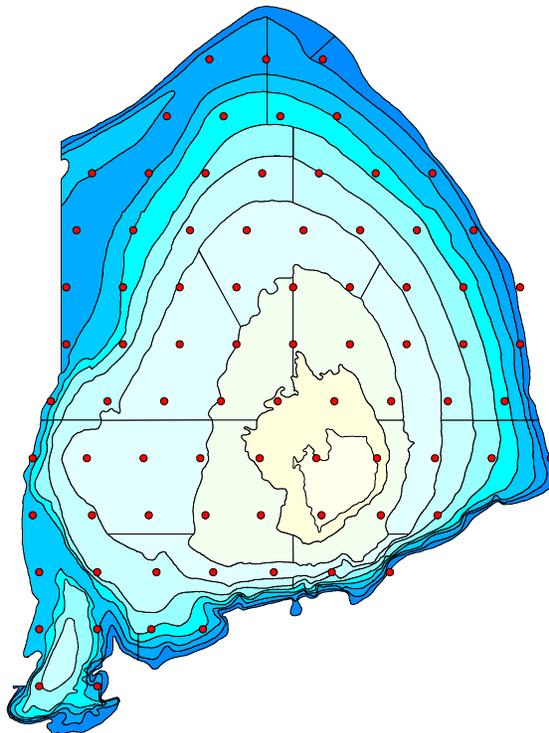


Figure 5 – Distribution grid of 80 CTD stations with a mean distance of 15 miles apart.

ANNEXES

DATA FORMS
SPECIES WORKING CODES
SHRIMP SAMPLING
PUBLICATIONS

Length frequencies forms: 1 and 1/2 cm

Campaña FLEMISH CAP 20

Pesca _____
 Día/mes _____ ESPECIE _____
 Peso muestra _____

talla (cm)		talla (cm)	
0		0	
1		1	
2		2	
3		3	
4		4	
5		5	
6		6	
7		7	
8		8	
9		9	
0		0	
1		1	
2		2	
3		3	
4		4	
5		5	
6		6	
7		7	
8		8	
9		9	
0		0	
1		1	
2		2	
3		3	
4		4	
5		5	
6		6	
7		7	
8		8	
9		9	
0		0	
1		1	
2		2	
3		3	
4		4	
5		5	
6		6	
7		7	
8		8	
9		9	

Campaña FLEMISH CAP 20

Pesca _____
 Día/mes _____ ESPECIE _____
 Peso muestra _____

talla (cm)		talla (cm)	
0		0	
0,5		0,5	
1		1	
1,5		1,5	
2		2	
2,5		2,5	
3		3	
3,5		3,5	
4		4	
4,5		4,5	
5		5	
5,5		5,5	
6		6	
6,5		6,5	
7		7	
7,5		7,5	
8		8	
8,5		8,5	
9		9	
9,5		9,5	
0		0	
0,5		0,5	
1		1	
1,5		1,5	
2		2	
2,5		2,5	
3		3	
3,5		3,5	
4		4	
4,5		4,5	
5		5	
5,5		5,5	
6		6	
6,5		6,5	
7		7	
7,5		7,5	
8		8	
8,5		8,5	
9		9	
9,5		9,5	

Biological sampling form: general and shrimp.

Campana FLEMISH CAP 20

Pesca _____
Dia/mes _____ ESPECIE _____

	talla (cm)	sexo	peso (g)	peso eviscer.	G
1					
2					
3					
4					
5					
6					
7					
8					
9					
10					
11					
12					
13					
14					
15					
16					
17					
18					
19					
20					
21					
22					
23					
24					
25					

	talla (cm)	sexo	peso (g)	peso eviscer.	G
26					
27					
28					
29					
30					
31					
32					
33					
34					
35					
36					
37					
38					
39					
40					
41					
42					
43					
44					
45					
46					
47					
48					
49					
50					

Pandalus borealis

Pesca _____

	talla (mm)	sexo	peso (g)
1			
2			
3			
4			
5			
6			
7			
8			
9			
10			
11			
12			
13			
14			
15			
16			
17			
18			
19			
20			
21			
22			
23			
24			
25			

Campana FLEMISH CAP 20

	talla (mm)	sexo	peso (g)
26			
27			
28			
29			
30			
31			
32			
33			
34			
35			
36			
37			
38			
39			
40			
41			
42			
43			
44			
45			
46			
47			
48			
49			
50			

161	<i>Arctozenus risso</i>		
262	<i>Paralepis speciosa</i>	222	<i>Brosme brosme</i>
17	<i>Paralepis coregonoides</i>	128	<i>Enchelyopus cimbrius</i>
181	<i>Sudis hyalina</i>	910	<i>Gaidropsarus argentatus</i>
160	<i>Magnisudis atlantica</i>	141	<i>Gaidropsarus ensis</i>
	Alepisauridae		Merlucciidae
32	<i>Alepisaurus ferox</i>	104	<i>Merluccius bilinearis</i>
371	<i>Alepisaurus brevirostris</i>	106	<i>Lyconus</i> sp.
	Anotopteridae		Melanonidae
154	<i>Anotopterus pharao</i>	232	<i>Melanonus zugmayeri</i>
	Notosudidae	215	Macrouridae
930	<i>Scopelosaurus lepidus</i>	426	<i>Coryphaenoides armatus</i>
	Bathysauridae	168	<i>Coryphaenoides rupestris</i>
715	<i>Bathysaurus ferox</i>	924	<i>Coryphaenoides guentheri</i>
	<u>Myctophiformes</u>	556	<i>Coryphaenoides carapinus</i>
173	Myctophidae	557	<i>Coryphaenoides brevibarbis</i>
367	<i>Ceratoscopelus maderensis</i>	985	<i>Coryphaenoides rudis</i>
913	<i>Lampanyctus</i> sp.	214	<i>Coryphaenoides mediterraneus</i>
370	<i>Notoscopelus kroeyeri</i>	134	<i>Coelorinchus caelorhincus</i>
366	<i>Benthoosema glaciale</i>	170	<i>Nezumia bairdii</i>
372	<i>Protomyctophum arcticum</i>	211	<i>Trachyrincus scabrus</i>
368	<i>Lampadena speculigera</i>	167	<i>Trachyrincus murrayi</i>
369	<i>Myctophum punctatum</i>	169	<i>Macrourus berglax</i>
981	<i>Taaningichthys</i> sp.		<u>Ophidiiformes</u>
	<u>Cetomimiformes</u>		Ophidiidae
	Rondeletiidae	177	<i>Brotulotaenia</i> sp.
705	<i>Rondeletia loricata</i>		<u>Perciformes</u>
	Cetomimidae		Zoarcidae
223	<i>Cetostoma regani</i>	166	<i>Lycenchelys paxillus</i>
249	<u>Lophiiformes</u>	162	<i>Lycodes</i> sp.
919	Linophrynidae	164	<i>Lycodes vahlii</i>
183	<i>Haplophryne mollis</i>	163	<i>Lycodes esmarkii</i>
209	<i>Linophryne coronata</i>	130	<i>Lycodes reticulatus</i>
26	Lophiidae	382	<i>Melanostigma atlanticum</i>
143	<i>Lophius americanus</i>	951	<i>Lycodonus flagellicauda</i>
	Ogcocephalidae		Polyprionidae
301	<i>Dibranchus atlanticus</i>	534	<i>Polyprion americanus</i>
	Melanocetidae		Howellidae
144	<i>Melanocetus johnsonii</i>	210	<i>Howella sherborni</i>
453	Oneirodidae		Chiasmodontidae
706	<i>Chaenophryne longiceps</i>	131	<i>Chiasmodon niger</i>
220	<i>Oneirodes eschrichtii</i>		Anarhichadidae
269	<i>Dolopichthys allector</i>	188	<i>Anarhichas</i> sp.
701	<i>Lophodolus acanthognathus</i>	189	<i>Anarhichas lupus</i>
	Ceratiidae	190	<i>Anarhichas minor</i>
146	<i>Ceratias holboelli</i>	121	<i>Anarhichas denticulatus</i>
7	<i>Cryptopsaras couesii</i>		Stichaeidae
	<u>Gadiformes</u>	281	<i>Lumpenus</i> sp.
	Moridae	280	<i>Lumpenus lampretaeformis</i>
139	<i>Antimora rostrata</i>	212	<i>Leptoclinus maculatus</i>
296	<i>Lepidion lepidion</i>		Ammodytidae
207	<i>Halargyreus johnsonii</i>	129	<i>Ammodytes</i> sp.
	Gadidae	191	<i>Ammodytes dubius</i>
100	<i>Boreogadus saida</i>		Trichiuridae
101	<i>Gadus morhua</i>	180	<i>Aphanopus carbo</i>
56	<i>Pollachius virens</i>		Centrolophidae
102	<i>Melanogrammus aeglefinus</i>	941	<i>Centrolophus niger</i>
140	<i>Micromesistius poutassou</i>		Caristiidae
	Phycidae	142	<i>Caristius fasciatus</i>
110	<i>Urophycis</i> sp.		<u>Beloniformes</u>
105	<i>Urophycis chuss</i>		Scomberesocidae
186	<i>Urophycis tenuis</i>	117	<i>Scomberesox saurus</i>
107	<i>Phycis chesteri</i>		<u>Stephanoberyciformes</u>

	Melamphaidae	845	<i>Neptunea despecta</i>
205	<i>Poromitra</i> sp.		Capuloidea (Superfamily)
290	<i>Poromitra megalops</i>	869	<i>Torellia delicata</i>
208	<i>Scopelogadus beanii</i>		Heterobranchia (Subclass)
	<u>Beryciformes</u>		<u>Cephalaspidea</u>
29	Dirietmidae	891	<i>Scaphander punctostriatus</i>
201	<i>Dirietmus argenteus</i>	846	<u>Nudibranchia</u>
	Trachichthyidae	764	Scaphopoda
202	<i>Hoplostethus atlanticus</i>	875	Polyplacophora
	Anoplogastridae	865	Bivalvia
248	<i>Anoplogaster cornuta</i>		Heterodonta (Subclass)
	<u>Scorpaeniformes</u>		Astartidae
	Sebastidae	840	<i>Astarte</i> sp.
52	<i>Sebastes</i> sp.		Cuspidariidae
51	<i>Sebastes norvegicus</i>	561	<i>Cuspidaria</i> sp.
53	<i>Sebastes mentella</i>		Pteriomorphia (Subclass)
54	<i>Sebastes fasciatus</i>		Pectinidae
50	<i>Sebastes</i> (juvenile)	992	<i>Chlamys islandica</i>
49	<i>Sebastes</i> (bag)	500	Cephalopoda
558	Cyclopteridae		<u>Sepiolida</u>
	Cottidae	964	Sepiolidae
258	<i>Triglops</i> sp.	503	<i>Semirossia</i> sp.
159	<i>Triglops murrayi</i>	929	<u>Teuthida</u>
	Psychrolutidae	912	<u>Oegopsida</u>
2	<i>Cottunculus</i> sp.		Gonatidae
115	<i>Cottunculus microps</i>	11	<i>Gonatus fabricii</i>
116	<i>Cottunculus thomsonii</i>		Onychoteuthidae
	Agonidae	509	<i>Onychoteuthis banksii</i>
135	<i>Aspidophoroides monopterygius</i>	30	Brachioteuthidae
136	<i>Leptagonus decagonus</i>	510	<i>Brachioteuthis</i> sp.
911	<i>Ulcina olrikii</i>		Histioteuthidae
917	Liparidae	12	<i>Histioteuthis</i> sp.
727	<i>Careproctus micropus</i>	506	<i>Histioteuthis reversa</i>
777	<i>Careproctus reinhardti</i>	511	<i>Histioteuthis bonnellii</i>
724	<i>Liparis</i> sp.	16	Ommastrephidae
726	<i>Liparis liparis</i>	516	<i>Todarodes sagittatus</i>
725	<i>Liparis fabricii</i>	810	<i>Illex</i> sp.
138	<i>Paraliparis copei</i>	504	<i>Illex illecebrosus</i>
	<u>Pleuronectiformes</u>		Chiroteuthidae
	Pleuronectidae	794	<i>Chiroteuthis</i> sp.
114	<i>Glyptocephalus cynoglossus</i>	707	<i>Chiroteuthis veranii</i>
112	<i>Hippoglossoides platessoides</i>	507	<i>Chiroteuthis picteti</i>
118	<i>Reinhardtius hippoglossoides</i>		Cranchiidae
120	<i>Hippoglossus hippoglossus</i>	768	<i>Taonius</i> sp.
	Tunicata (Subphylum)	996	<i>Taonius pavo</i>
932	Ascidiacea (Class)	512	<i>Teuthowenia megalops</i>
853	Didemnidae	942	<i>Liguriella</i> sp.
	Thaliacea (Class)		<u>Vampyromorpha</u>
962	Pyrosomatidae		Vampyroteuthidae
		798	<i>Vampyroteuthis</i> sp.
935	Mollusca	6	<u>Octopoda</u>
804	Gastropoda		Octopodidae
	Caenogastropoda (Subclass)	502	<i>Bathypolypus</i> sp.
	Stromboidea (Superfamily)	762	<i>Bathypolypus bairdii</i>
837	<i>Arrhoges occidentalis</i>	505	<i>Bathypolypus arcticus</i>
	Muricoidea (Superfamily)	513	<i>Graneledone</i> sp.
470	<i>Boreotrophon</i> sp.	508	Cirroteuthidae
	Buccinoidea (Superfamily)	515	<i>Cirroteuthis muelleri</i>
839	Buccinidae	931	Opisthoteuthidae
945	<i>Buccinum</i> sp.		
893	<i>Turrisipho</i> sp.		Arthropoda
944	<i>Beringius turtoni</i>	600	Crustacea (Subphylum)
900	<i>Colus</i> sp.		<u>Decapoda</u>

	Dendrobranchiata (Suborder)		Geryonidae
	Sergestoidea (Superfamily)	927	<i>Chaceon quinquedens</i>
635	<i>Eusergestes arcticus</i>		Peracarida (Superorder)
826	<i>Sergia robusta</i>	872	<u>Isopoda</u>
	Penaeoidea (Superfamily)		<u>Lophogastrida</u>
636	<i>Aristaeopsis edwardsiana</i>		Gnathophausiidae
918	<i>Benthesicymus bartletti</i>	816	<i>Gnathophausia</i> sp.
888	<i>Gennadas</i> sp.	903	<i>Gnathophausia zoea</i>
977	<i>Gennadas elegans</i>	568	<i>Gnathophausia gigas</i>
	Caridea (Infraorder)		Eucopiidae
	Oplophoroidea (Superfamily)	983	<i>Eucopia sculpticauda</i>
14	<i>Acanthephyra</i> sp.	928	<u>Mysida</u>
612	<i>Acanthephyra eximia</i>	895	<u>Amphipoda</u>
285	<i>Acanthephyra pelagica</i>	700	Hyperiidea
286	<i>Acanthephyra purpurea</i>		Eucarida (Superorder)
817	<i>Notostomus</i> sp.	956	<u>Euphausiacea</u>
980	<i>Notostomus elegans</i>	854	Cirripedia
813	<i>Ephyrina</i> sp.		
302	<i>Oplophorus spinosus</i>		Chelicerata (SubPhylum)
	Nematocarcinoidea (Superfamily)	933	Pycnogonida
761	<i>Nematocarcinus rotundus</i>	946	Colossendeidae
	Pasiphaeidae (Superfamily)		
825	<i>Pasiphaea tarda</i>	907	Porifera
909	<i>Pasiphaea multidentata</i>		Demospongiae
824	<i>Parapasiphae sulcatifrons</i>		<u>Poecilosclerida</u>
	Alpheoidea (Superfamily)		Cladorhizidae
921	<i>Spirontocaris spinus</i>	476	<i>Chondrocladia</i> sp.
999	<i>Spirontocaris liljeborgii</i>		Coelosphaeridae
620	<i>Lebbeus polaris</i>	499	<i>Forcepia</i> sp.
	Pandaloidea (Superfamily)		<u>Hadromerida</u>
802	<i>Pandalus montagui</i>		Stylocordylidae
632	<i>Pandalus borealis</i>	417	<i>Stylocordyla</i> sp.
631	<i>Atlantopandalus propinquus</i>	970	Polymastiidae
630	<i>Pandalus</i> (bag)	971	<i>Tentorium</i> sp.
	Crangonoidea (Superfamily)	785	<i>Radiella hemisphaerica</i>
613	<i>Argis dentata</i>		Suberitidae
827	<i>Sabinea</i> sp.	566	<i>Rhizaxinella</i> sp.
829	<i>Sabinea sarsii</i>		<u>Spirophorida</u>
828	<i>Sabinea hystrix</i>	618	Tetillidae
747	<i>Sabinea septemcarinata</i>	982	<u>Astrophorida</u>
640	<i>Pontophilus norvegicus</i>		Ancorinidae
	Palinura (Infraorder)	563	<i>Stryphnus</i> sp.
820	Polychelidae	799	Geodiidae
475	<i>Stereomastis</i> sp.		
821	<i>Stereomastis sculpta</i>	934	Cnidaria
601	<i>Stereomastis nana</i>	844	Hydrozoa
555	<i>Pentacheles laevis</i>	905	Anthozoa
	Anomura (Infraorder)	868	<u>Pennatulacea</u>
954	Paguridae		Kophobelemnidae
	Lithodidae	814	<i>Kophobelemnion stelliferum</i>
650	<i>Lithodes maja</i>		Halipteridae
651	<i>Neolithodes grimaldii</i>	880	<i>Halipteris</i> cf. <i>christii</i>
925	Galatheidae	851	<i>Halipteris finmarchica</i>
	Munididae		Anthoptilidae
699	<i>Munida</i> sp.	852	<i>Anthoptilum</i> sp.
	Munidopsidae		Umbellulidae
790	<i>Munidopsis curvirostra</i>	889	<i>Umbellula</i> sp.
	Brachyura (Infraorder)		Funiculinidae
	Oregoniidae	881	<i>Funiculina quadrangularis</i>
806	<i>Hyas</i> sp.		Protoptilidae
788	<i>Hyas araneus</i>	906	<i>Distichoptilum gracile</i>
901	<i>Hyas coarctatus</i>		Pennatulidae
789	<i>Chionoecetes opilio</i>	848	<i>Pennatula</i> sp.

- 767 *Pennatula grandis*
615 *Pennatula aculeata*
940 Antipatharia
Antipathidae
604 *Stichopathes* sp.
Schizopathidae
885 *Stauropathes arctica*
793 Alcyonacea
850 Nephtheidae
619 *Gersemia* sp.
832 *Duva florida*
Plexauridae
991 *Swiftia* sp.
860 *Paramuricea* sp.
Chrysogorgiidae
963 *Radicipes* sp.
Alcyoniidae
892 *Heteropolypus* sp.
419 *Anthomastus* sp.
Anthothelidae
879 *Anthothela* sp.
Paragorgiidae
763 *Paragorgia* sp.
Acanthogorgiidae
815 *Acanthogorgia* sp.
Primnoidae
959 *Primnoa resedaeformis*
898 Isididae
878 *Acanella arbuscula*
Zoanthidea
862 Epizoanthidae
936 Actiniaria
Liponematidae
774 *Liponema* sp.
Actinerniidae
775 *Actinernus* sp.
834 Hormathiidae
605 *Stephanauge nexilis*
471 *Stephanauge spongicola*
Actinoscyphiidae
974 *Actinoscyphia* sp.
987 Scleractinia
Caryophylliidae
886 *Desmophyllum dianthus*
Flabellidae
833 *Flabellum alabastrum*
960 Scyphozoa
863 Coronatae
616 Atollidae
617 Periphyllidae
800 **Ctenophora**
894 Nemertea
Annelida
955 Polychaeta
Sabellida
968 Sabellidae
Phyllodocida
976 Polynoidae
Aphroditidae
809 *Aphrodita* sp.
- 882 *Laetmonice* sp.
Clitellata
890 Hirudinea (Subclass)
836 **Sipuncula**
847 **Bryozoa**
864 **Brachiopoda**
975 *Terebratulina septentrionalis*
Echinodermata
807 Asterozoa
Asterozoa
Astropectinidae
787 *Plutonaster agassizi*
855 *Leptychaster arcticus*
871 *Psilaster andromeda*
796 *Bathybiaster vexillifer*
883 Brisingidae
Asterinidae
870 *Tremaster mirabilis*
Ctenodiscidae
849 *Ctenodiscus crispatus*
835 Benthoplectinidae
Goniasteridae
830 *Ceramaster granularis*
842 *Hippasteria phrygiana*
795 *Mediaster bairdi*
Pseudarchasteridae
961 *Pseudarchaster* sp.
904 *Pseudarchaster parelii*
760 *Pseudarchaster gracilis*
902 Solasteridae
949 *Lophaster furcifer*
841 Pterasteridae
428 Poraniidae
874 *Poraniomorpha hispida*
843 Echinasteridae
Zoroasteridae
818 *Zoroaster fulgens*
Asteriidae
896 *Stephanasterias albula*
805 Ophiurozoa
Ophiurozoa
Asteronychidae
943 *Asteronyx loveni*
897 Gorgonocephalidae
Ophiuridae
877 *Ophiura sarsii*
535 *Ophioplithus* sp.
Ophiolepididae
811 *Ophiomusium lymani*
Ophiacanthidae
876 *Ophiacantha* sp.
Ophiactidae
831 *Ophiopholis aculeata*
808 Echinozoa
822 Echinothuriidae
Phormosomatidae
791 *Phormosoma placenta*
Schizasteridae
838 *Brisaster fragilis*
866 Crinozoa
759 Holothurozoa

Shrimp sampling (*Pandalus borealis*)

The shrimp is a decapod crustacean Pandalidae. The distinctive characters of the species are the presence of prominent spines on the 3rd abdominal somite (in mid-dorsal position) and on the 4th abdominal somite (postero-dorsal margin); spines on the distal portion of the face (Figure 1) .

Berkeley (1930) first noted that *P. borealis* is a protandric hermaphrodite, i.e., each individual matures and functions first as a male, passes through a transitional or intersexual phase, and becomes a female. While this is the normal sequence of events, several authors have reported early maturing females in the southern portions of the distribution range.

The sex of *P. borealis* can be identified by changes in the external structure of the 1st pair of pleopods from the endopodite. However, a stricter classification should take into account also the observation of internal appendage and male appendage of the second pair of pleopods, allowing us to distinguish between male and female transition (Figure 2). This appendix is difficult to see without the aid of a binocular microscope, making impractical the sampling aboard commercial vessels.

Sampling

Due to the biological characteristics of shrimp, the study of this species presents some particularities to be considered in sampling.

The absence of hard parts that can be used to identify the age of the individuals, makes necessary to obtain size distributions in adequate numbers to enable us to identify different year classes through modal analysis or identification of trends in the size distribution of the different states considered.

Furthermore, the great plasticity of this species in terms of growth, change of sex and sexual maturity, which is affected, both spatially and temporally, determine an adequate stratification in the sampling and detailed analysis (by sex and maturity state).

The carapace length (CL) is measured with a calliper from the posterior margin of the eyestalk to the posterior mid dorsal edge of the carapace (Figure 3), to the lower half millimetre.

Approximately 200 or 300 individuals from the catch at each station will be randomly measured by sex and maturity stage (Figures 4, 5, 6 and 7), according to the following categories:

- Males (M), who include individuals both young and adult (M). They are distinguished by the shape of the endopodite apex from the 1st pair of pleopods (Figure 4 and 5). Also, this sampling should be done by separating the males from the individuals in transition state (T), as these specimens in NAFO, at the survey time, are classified as immature females primiparous.
- Immature primiparous females (IF). They differ mainly in the lanceolate shape of endopodite from the 1st pair of pleopods (Figure 6), and the presence of sternal spines well defined on the abdomen (Figure 7). In addition, as noted above, individuals in transition (T) will be considered as immature females. The presence of eggs in the head is not a decisive character, as may be present in individuals in transition or in females that have matured in the past (in this case the sternal spines are absent or much less marked).
- Mature females (MF). They are females with sternal spines absent or barely marked. In this group are included the females in a new spawning process, without eggs in the abdomen but with eggs in the head and without sternal spines visible and the females at rest, without sternal spines and eggs in the abdomen or head.
- Mature ovigerous females (HMOV), which present eggs in the abdomen.

In addition to the samples analysed on board will be collect samples for further study in the laboratory. These samples will be frozen at sea and subsequently analysed to establish the length-weight relationship. The sampling precision in the laboratory will be one hundredth of a millimetre and one hundredth of a gram. On the other hand berried females will be analysed for further study of fecundity (egg counts). Therefore samples should be collected properly so that egg loss as small as possible.

The frozen samples will be taken randomly from all strata considered in the survey.

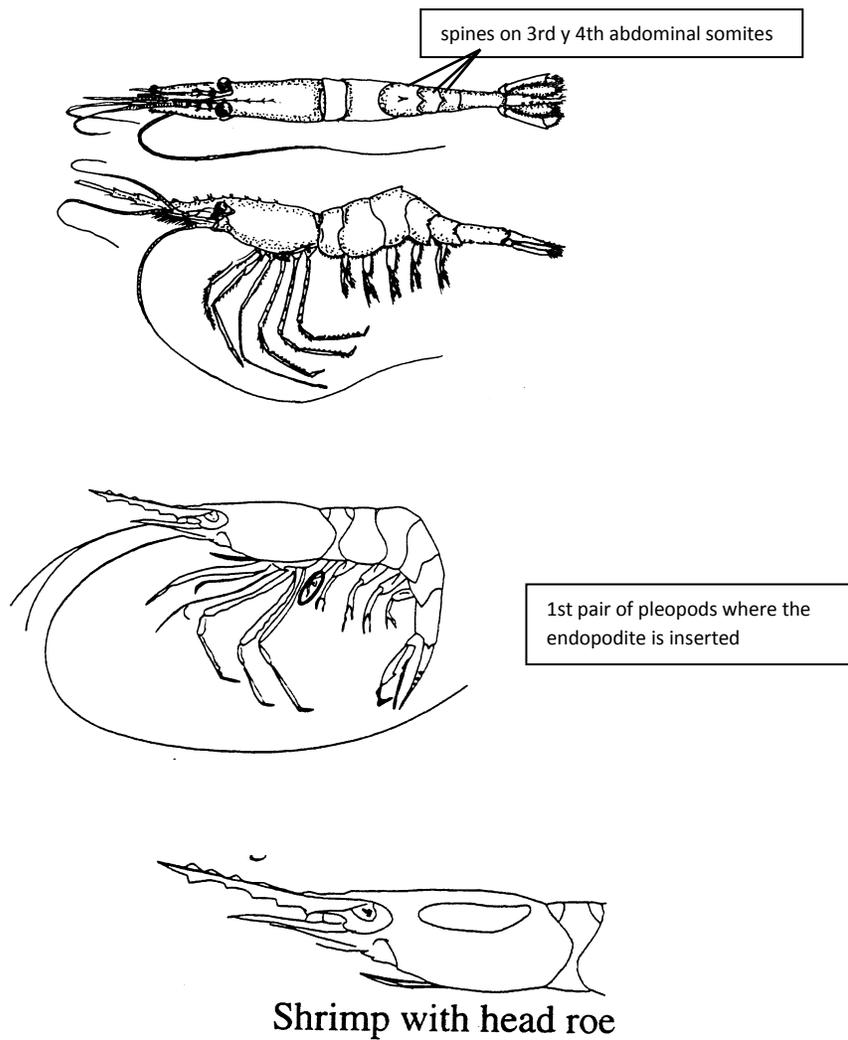


Figure1 – Dorsal and lateral view from *Pandalus borealis* (Butler, 1980); general aspects in its anatomy to determine the species and sex.

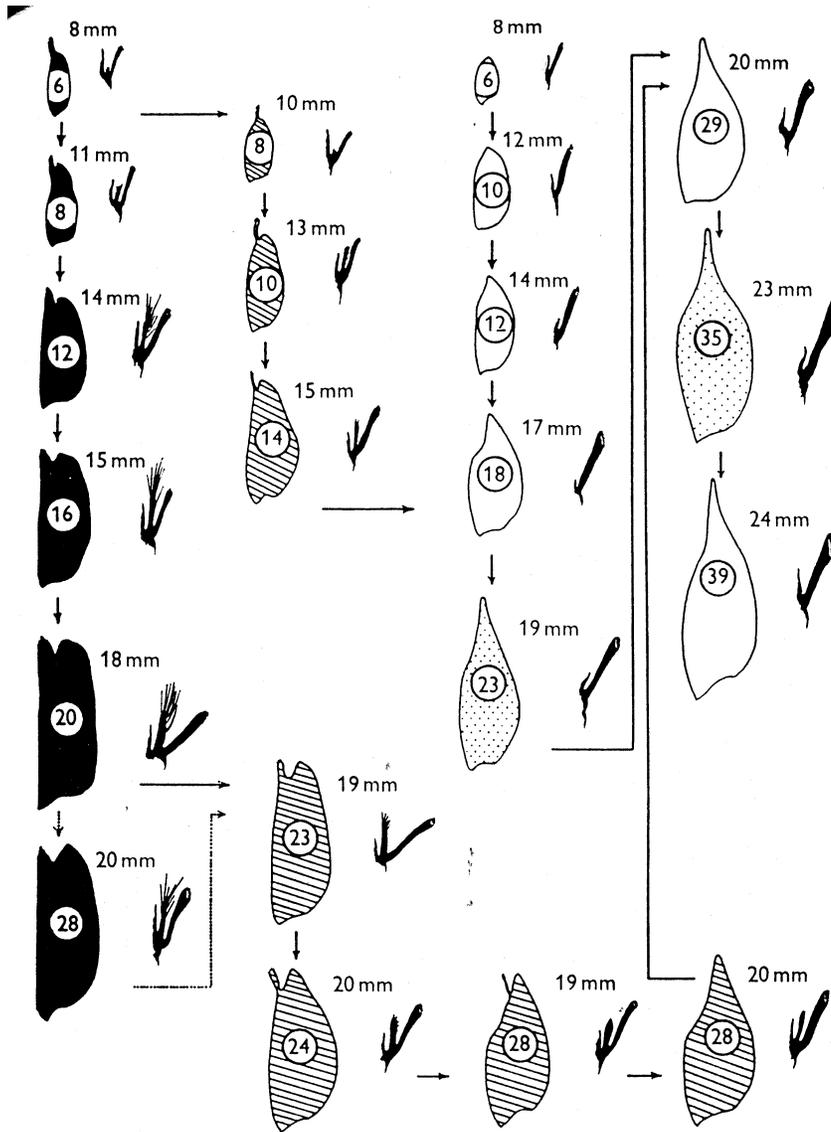
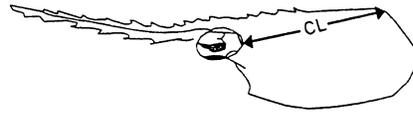


Figure 2 – Changes in form with increasing age of the endopodite of the first pleopod and the corresponding appendix internal and appendix masculine of the second pleopod of *Pandalus borealis*. Age in months is given in the ring in each endopodite and the carapace length (mm) above each figure. Male endopodite, black; transitional, cross-hatched; female, outline. Arrows indicate sequence (from Allen 1959)



Length measurement of shrimp

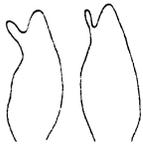
Figure 3 – Length measurement from the posterior margin of the eyestalk to the posterior mid dorsal edge of the carapace.



1 2
1= young male
2= older male

Figure 4 – Males (M).- Carapace lengths from 6 to 22 mm approximately. Endopodite transformed as reproductive organ with the internal appendix clearly visible:

- Narrow and long in young males, the internal appendix exceeds the height of the apex of the endopodite (1).
- In adult males this organ is considerably wider compared with its length and rarely exceeds the apex of endopodite.



3 4
3 and 4, changing sex

Figure 5 – Transitional o intersex (T).- Lengths from 18 to 23 mm. The reproductive organ gradually reduces in size at each moult and does not reach the apex of the endopodite.

In this phase two states can be distinguished:

- No eggs in the head.
- With blue-green eggs in the head. Furthermore, as in males have well defined abdominal sternal spines. They will be considered immature females.

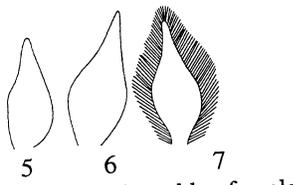
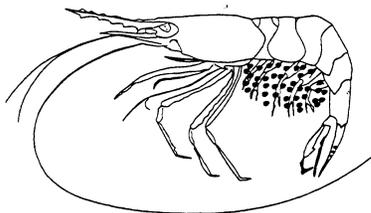


Figure 6 – Female.- Lengths from 18 to 30 mm. The male reproductive organ is disappeared. The endopodite shows a lanceolate shape (5 and 6). In females where there has been the release of eggs and larvae (hatching) the endopodite has long silks (7). Sternal spines have disappeared or are much less marked.

Females can present several maturity stages



Immature females (IF). They are hard to separate from the transition stage without observation of internal appendix in the 2nd pair of pleopods. In general they would be in this state those individuals with endopodite shape as females, and sternal spines well defined. Also, they will be considered as immature females those individuals at transition state with eggs in the carapace.

Mature females. (MF). They may have different stages that we will group into two: ovigerous females (HMOV) with eggs in the abdomen and non-ovigerous females (HM) no eggs in the abdomen and sternal spines barely marked or absent

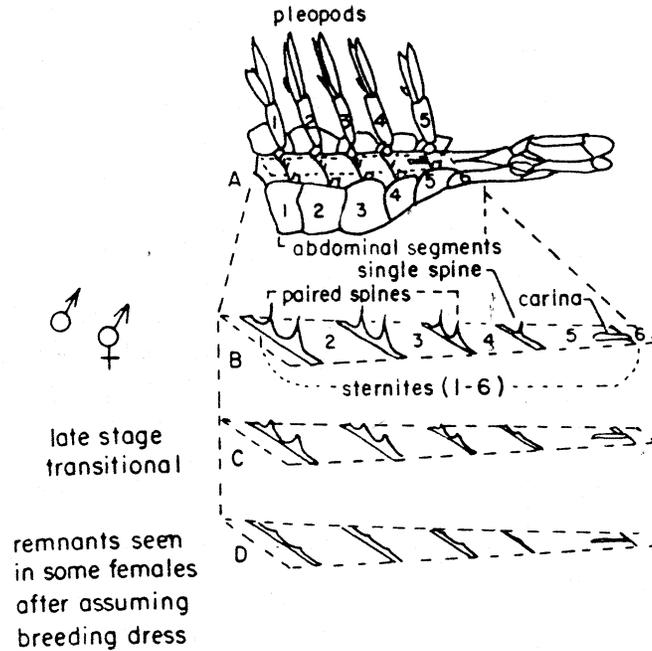


Figure 7 – Diagrammatic ventral view of the abdomen of a pandalid shrimp showing sternal spines (from McCrary 1971).

B and C.- Spines well defined (present in males, transitional stage and immature females).

D.- Spines barely marked or absent in mature females which have previously spawned

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