

MANUAL

**FOR QUANTITATIVE SAMPLING AND SAMPLE
TREATMENT OF MARINE SOFT-BOTTOM
MACROZOOBENTHOS**



Compiled by Valentina Todorova¹ and Tsenka Konsulova²

¹Institute of Oceanology, Bulgarian Academy of Sciences, PO Box 152, 9000 Varna, Bulgaria
vtodorova@io-bas.bg

²Institute of Oceanology, Bulgarian Academy of Sciences, PO Box 152, 9000 Varna, Bulgaria
konsulova@io-bas.bg

October 2005

Table of contents

Table of content	1
1. Introduction	2
2. Purpose	3
3. Sampling strategy	3
4. Logistics	5
4.1 Site location	5
4.2 Sampling equipment	5
4.3 Vessel.....	5
4.4 Positioning equipment.....	6
4.5 Personnel	6
5. Ship-board routines	6
5.1 Grab deployment	6
5.2 Sieving	6
5.3 Fixation	7
5.4 Staining.....	8
5.5 Labelling	8
5.6 Sample registering	8
6. Laboratory routines	9
6.1 Sorting and taxonomic identification.....	9
6.2 Abundance determination	9
6.3 Biomass determination.....	9
7. Data reporting	10
8. Recommendations for Quality Assurance	11
8.1 Equipment calibration	11
8.2 Training.....	11
8.3 Repeatability of site positioning.....	11
8.4 Quality and quantity of the sample	11
8.5 Accuracy and traceability of sample numbering and registration	11
8.6 Accuracy of sample sorting and taxon identification	12
8.7 Accuracy of data compilation	12
8.8 In-house Quality Assurance	12
9. Quality Control routines	12
9.1 Extraction efficiency – total taxa target.....	12
9.2 Extraction efficiency – total individuals target.....	12
9.3 Total wet weight biomass target.....	13
9.4 Bray-Curtis comparison.....	13
10. Data analysis and metrics	13
11. Acknowledgements	15
12. References	17
Annex 1. SAMPLE RECORD - MACROZOOBENTHOS	19
Annex 2. DATA REPORT SHEET - MACROZOOBENTHOS	20
Annex 3. List of taxonomic literature to be employed for the identification of macrozoobenthic species in the Black Sea	22
Annex 4. Provisional check list of macrozoobenthic Polychaeta, Crustacea and Mollusca encountered in the Black Sea and Azov Sea	25

1. Introduction

This guideline has been adapted from established benthic grab sampling methods described in a range of documents the most important of which are Davies *et al.* (2001), Gray *et al.* (1992), Holme and McIntyre (1984), ISO 16665: 2005 (E), the Manual for Marine Monitoring in the COMBINE Programme of HELCOM (2003), Rees *et al.* (1991), Rumohr (1990), Rumohr (1999) and UK NMMP Green Book (2003). Further consideration has been given to quality assurance from other texts including Rees (2004), ICES (2004) and ICES (2005).

The aim of these recommendations is to standardise the methods used by different scientists in the riparian Black Sea countries for sampling and treatment of macrozoobenthos in order to increase the comparability of results for different areas and to enable detection of large-scale changes in the system that would not otherwise be detected by scientists or groups working independently of each other.

The manual provides detailed procedures for quantitative sampling and analysing soft sediment macrozoobenthos. Soft bottoms are defined as those with sediments ranging from mud to, and including, sand. The zoobenthos comprise animals living in the sediment (infauna), on the sediments (epifauna) or in close association with the seabed. Conventionally, these organisms are sub-divided on the basis of size (Table 1, from McIntyre, 1978). Size categories, though somewhat arbitrary, identify the major functional groups of organisms, each of which requires different approaches to sampling and analysis. Macrofauna is defined as animals retained on a 0.5 - 1 mm sieve.

Table 1. Size categorization of zoobenthos (from McIntyre, 1978).

Category	Size	Biological features	Sampling techniques	Taxonomic position
Microbenthos	Pass finest sieves	High rates of respiration and reproduction	Plating and culturing. Cores of < 2 cm diam.	Most Protozoa
Meiobenthos	Pass 0.5-1 mm sieves	Medium respiration rates Two or more generations per year	Cores of 2 – 10 cm diam.	Large Protozoa Small Metazoa
Macrobenthos	Retained on 0.5-1 mm sieves	Low respiration rates. Two or less generations per year. Mostly infauna	Grab sampling at least about 0.1 m ²	Medium-sized Metazoa
Megabenthos	Handpicked from samples	As above, mostly epifauna	Towed gear, trawls, dredge	Large Metazoa

Analysis of macrofaunal communities in soft-bottom sediments is an integral part of marine environmental assessment. The use of macrozoobenthic communities for estimation of the extent of environmental impact has been an enduring approach due to the following advantages of the macrofauna as an indicator of the environmental conditions:

- long (comparatively to plankton) life span of species, which therefore integrate environmental change over time;
- sedentary or sessile mode of life, therefore organisms are not able to escape stress and integrate the environmental quality in a given area;
- relatively easy to sample quantitatively;

- relatively easy taxonomic identification and available taxonomic keys for most groups;
- well-documented and predictive response to a number of environmental stressors (thus, community changes can be interpreted with a degree of confidence).

2. Purpose

The specific objectives to be accomplished by sampling and analysing benthic macrofauna depend on the general goal of the study. According to the Manual for Marine Monitoring in the COMBINE Programme of HELCOM (2003) in eutrophication assessment studies benthic communities are examined for the following purposes:

- “to monitor the spatial variability in species composition, abundance and biomass within the maritime area resulting from anthropogenic nutrient inputs;
- to monitor temporal trends in species composition, abundance and biomass within the maritime area (at a timescale of years) in order to assess whether changes can be related to temporal trends in nutrient inputs;
- to support the development and implementation of a common procedure for the identification of the status of the benthic communities;
- to understand the relationship between nutrient concentrations and temporal trends in species/community characteristics.”

The following basic attributes can be met by using benthic grab sampling in studies aimed at assessing species and habitat diversity:

- establish the benthic community composition and measure the species richness within and between habitats;
- establish the species which are present at a site/habitat, including their abundance and biomass within statistical limits;
- measure the abundance of key species (rare, sensitive, declining, representative) in habitats;
- determine the distribution of different habitats and the associated communities (biotopes);
- identify rare, fragile, representative or rich biotopes at a site;
- ground-truth mapped areas (established by video or acoustic ground discrimination techniques, e.g. side-scan sonar) occupied by biotopes.

3. Sampling strategy

A strategy is defined as an elaborate and systematic plan of action designed to achieve a particular goal. The monitoring strategy will define what is to be determined (measured), where, in which media, at which time and frequency, as well as its required quality. The strategy should also include information on the final use of data, including data analysis, compilations, statistical calculations, and evaluations.

Some general considerations regarding macrozoobenthos monitoring strategy include:

- **Establishment of the baseline community structure and the natural variability.** This involves review of literature on the benthic biota and supporting habitat. If the existing information is insufficient, then an initial spatially extensive “baseline” survey is necessary to describe the distribution of the benthos, identify spatial patterns and to relate this to habitat type.
- **Sampling habitats and sites.** Characteristic habitats and substrates typical of the whole monitoring area must be sampled. In general soft-sediment sea beds are more at risk from the consequences of nutrient enrichment and pollution, being depositional environments than are hard bottoms (including gravel), being usually high-energy environments, therefore soft bottom macrozoobenthic communities are usually targeted

by monitoring studies for the assessment of the ecological status of coastal and transitional water.

- **Number of sampling stations.** The number of stations is governed by spatial heterogeneity at the sea bed, predicted dispersal pathways of pollutants, and resource limitations (time, money, laboratory facilities, staff). Sample points must be spread out over the extent of the habitat studied to ensure an adequate consideration of spatial variation. It cannot be assumed that one point is representative of the habitat as a whole.
- **Number of replicate samples.** Replicate sampling is recommended to allow statistical comparisons between stations in space and/or time. The sufficient number of replicate samples depends on the natural variability of benthic community qualitative and quantitative attributes.
- **Timing and temporal scales.** Sampling time should avoid recruitment periods, since the temporary presence of many newly settled juveniles may lead to misinterpretation of the quantitative trends in adult populations. The meteorological conditions are also of importance for selecting the optimum time for field work at sea. Annual sampling at the same time each year is considered as adequate for monitoring of point-source discharges (Rees *et al.*, 1991). However, severe effects of eutrophication with seasonal manifestation such as hypoxia/anoxia may dictate a higher sampling frequency. If the sampling frequency is twice per year, then sampling should take place in late winter/early spring (March/April) to establish the stable community conditions and in late summer/autumn (August/September) with a view to detecting the possible effects of nutrient enrichment (such as hypoxia) on the macrozoobenthos.
- **Establishment of reference conditions.** The ecological status of benthic macrofauna is assessed by measuring the deviation of key benthic community attributes from reference values expected under non-degraded conditions in similar habitat types. Therefore a control reference site is required for each test site when measuring anthropogenically induced change. Reference sites are generally defined as those sites having minimal exposure to human activities and are representative of the waterbody type and region of interest (Hughes *et al.* 1986). Reference conditions, as used here, are numerical descriptions of the variability of biological measurements taken from a composite of multiple reference sites (Gibson *et al.* 1996).
- **Selection of relevant environmental descriptors.** To obtain the most reliable and complete picture of the state of the environment, an integrated ecosystem-level monitoring approach should be adopted, involving coordinated chemical, physical, and biological sampling. The following physical and chemical environmental parameters are considered essential for the interpretation of macrozoobenthic data:
 - Organic carbon content in sediments. Gives an indication of food availability to benthic fauna. Can also indicate degree of organic enrichment.
 - Sediment granulometry (fractions expressed as % dry mass, median diameter, sorting coefficient). Indicates sediment homogeneity and sorting. Important in determining community composition and diversity and relationship to organic or contaminant content.
 - Dissolved oxygen in bottom water. Despite that momentary values are not indicative of the long-term oxygen conditions measurements during summer could provide detection and evidence of hypoxia/anoxia events. Measurements should be done at the seabed-water interface where a strong declining gradient of dissolved oxygen is usually observed.

- Secchi disk transparency. Despite that momentary values are not indicative of the long-term conditions it is considered as the simplest and easiest to measure indicator of the trophic conditions.
 - Long-term data of Chl *a*. Best indicator of the long-term trophic conditions.
 - Toxicants (selected heavy metals, petroleum hydrocarbons, chlorinated pesticides, etc.). These measurements and analyses are expensive, therefore should be done initially to establish the baseline concentrations and repeated at lower frequency compared to macrozoobenthos monitoring.
 - Depth. Determines the vertical gradients of dissolved oxygen and other gasses (i.e. H₂S), trophic resources, light conditions, temperature, etc. and therefore governs the diversity and distribution of macrozoobenthic communities.
 - Temperature. Major physical variable important in determining the distribution of macrofauna.
 - Salinity. Major physical variable important in determining the diversity and distribution of macrofauna.
- **Adherence to standard protocols for sampling and analysis.** Adoption of standard methodology for sampling and analysing macrozoobenthos on regional basis is of primary importance for data comparability and large-scale assessments.

4. Logistics

4.1 Site location

Maps and charts to the appropriate scale should be obtained. The positions of sampling stations should be defined using geographic co-ordinates, e.g. latitude/longitude to at least two decimal points with reference to the appropriate system for graticules (such as European Datum: ED-50; World Geodetic System: WGS-84). Latitude and longitude for sample sites should be determined prior to beginning field work (or should be the same as for sites surveyed in the pilot survey or previous monitoring studies). When revisiting sampling stations poorly-defined in terms of geographical coordinates, the water depth, known landmarks (unless in open sea) as well as sediment features should be used as the main criteria for relocating the sampling stations. A minimum accuracy of ± 50 m and ± 20 m in open waters and estuarine areas, respectively, should be attained.

4.2 Sampling equipment

Remote sampling is most commonly carried out using a grab or corer. The nature of the sea bed will determine the most effective type of sampling gear. For soft bottom substrates, a grab of standard design is an appropriate sampler. Van Veen grab with a sampling area of 0.1 m² should be employed as a standard macrozoobenthos sampler for the Black Sea Integrated Monitoring and Assessment Programme (BSIMAP) since it is (i) an efficient sampler for the range of soft sediments encountered in the Black Sea, (ii) reliable and simple to operate and (iii) widely applied, which allows data comparison with other marine areas. Grabs should be equipped with hinged inspection ports. The biting depth of grabs can vary with sediment conditions. Weights can be added to adjust according to the sediment conditions.

4.3 Vessel

The survey vessel should be appropriately and adequately equipped for bottom sampling, with sufficient deck space. The size of vessel required should be chosen as appropriate to the conditions in the sampling area and the type of sampling gear to be employed. In all cases where heavy sampling gear is deployed the vessel must be fitted with a suitable power winch, wire of the appropriate dimensions rigged to a meter

wheel and an 'A' frame or gantry. The vessel should also be equipped with echosounder and satellite global positioning device.

4.4 Positioning equipment

A Differential Geographical Positioning System (DGPS) with monitor should be used in all sea areas if possible. If the differential is not available, accuracy should be assessed and a minimum of Global Positioning System (GPS) should be used.

4.5 Personnel

van Veen Grab can be operated by two survey staff in addition to a winch operator. At least one of the survey team should be experienced with handling grabs and have experience of sampling and sieving marine invertebrates.

5. Ship-board routines

Sampling on shallow stations (70 m or less) is recommended to be conducted during daytime, since some benthic species have semi-pelagic activity during the night.

5.1 Grab deployment

At each site the grab should be set down gently at a speed that avoids triggering the mechanism. The winch wire should remain vertical (wire angle must be kept as small as possible) to ensure an even bite of the grab. In the case of deep or fast-moving water this may require additional weights on the grab and maintaining position by motoring into the current or anchoring. Between approximately 5 m and 10 m above the sea floor, the lowering speed should be decreased (< 0.5 m/s) in order to further reduce the bow-wave and water turbulence in front of the grab. Contact with the sea floor is observed by the slack on the wire, after which the grab is gently raised for approximately the first 5 m. Then the recovery can proceed with maximum safe speed.

Appropriate equipment for receiving and processing the samples should be ready on deck. On retrieval the grab should be placed on stable landing table. The sample should be examined for adequacy via the top inspection ports immediately upon retrieval on deck. If the sediment depth in the grab is less than 7 cm in mud or 5 cm in sand, the sample is rejected. Other samples rejection criteria are given in the Recommendations for Quality Assurance, (see 8.4). If sediment characteristics make it impossible to collect approved samples, the best available samples should be retained, and the circumstances noted in the field record.

The faunal samples should be gently decanted into a receiving container (barrel).

The grab is to be rinsed thoroughly before redeployment.

Each laboratory shall regularly check the exact sampling area of its grab in order to make possible a correct calculation of the number of individuals per square metre. (The area of the grab has a tendency to increase, especially when sampling in stiff clayey sediments).

Sediment characteristics and background information should be recorded before sieving.

5.2 Sieving

Approved samples should be sieved in the field using seawater to remove the fine sedimentary material. Each sample must be sieved, stored and documented separately.

The standard sieve for the Black Sea Integrated Monitoring and Assessment Programme (BSIMAP) shall be of metal gauze (stainless steel, brass or bronze) and have a mesh size of 1.0 x 1.0 mm. In order to collect quantitatively developmental stages of the macrofauna and abundant smaller species (longer but thinner than 1 mm) it is, however, recommended using an additional sieve with mesh size of 0.5 x 0.5 mm. The additional sieve also ensures against loss of specimens when sieving because of using too high water pressure. The mesh size of the sieves has to be checked from time to time for damage and wear. The use of large sieves is encouraged because:

- the risk of clogging is kept low;

- the risk of spilling is reduced when transferring samples from the receiving container to the sieves.

Water should be added gently to the receiving container to produce a water sediment suspension. The use of sprinklers and hand-operated douches to suspend the sample is recommended. Very stiff clay can be gently fragmented by hand. The sample is transferred in small quantities to the sieve as a sediment-water suspension.

The sieving of the sample has to be done carefully in order to avoid damage of fragile animals. Sieving is done by washing the material in the sieve with gentle jets of seawater and shaking by hand. Deck hoses must be provided with shower nozzles. Visible fragile animals, e.g. some polychaetes, echinoderms, etc. or large, heavy molluscs shall be hand-picked during the sieving, placed in separate plastic bags/jars and fixed before being placed in the container along with the rest of the sample. Stones and big shells should be picked out and kept in separate containers or discarded if devoid of encrusting fauna to avoid the grinding effect.

In order to reduce damage of delicate organisms sieving may be done by placing the sieve in a water bath deep enough to cover the mesh screen and “paddled” until the sediments are washed out. However, this process is time consuming and therefore is not recommended in case of limited resources. Furthermore, long duration of sieving time should be avoided because small animals may actively pass through the sieve.

All residues retained on the sieve should be carefully flushed off the sieve with water from below into appropriate sample containers (e.g. plastic jars, plastic buckets with watertight lids).

Between the sample portions the sieves must be checked and cleared of trapped fauna and any sediment to avoid clogging and thus to ensure an equal mesh size during the whole sieving procedure.

According to the UKNMMP Green Book (2003) samples may be sieved to 0.5 mm and 1 mm fractions either in the field or in the laboratory and analysed separately. Whether separated in the field or laboratory, the sieving method employed should remain consistent from year to year.

Separation of 0.5 mm and 1 mm fractions in the laboratory is recommended because:

- The time for sieving a sample onboard is kept shorter, which is important in time-limited cruises and in bad meteorological conditions (rain, cold, rough sea).
- Since the fractions must be kept separately, onboard separation will double the number of specimen containers, which is inconvenient for storage and transportation.
- The risk of mixing up fractions from different samples increases if a sample is split in two specimen containers (for two fractions) onboard.

5.3 Fixation

Samples (hand-picked animals and the sieving residue) should be fixed as soon as possible after sieving using buffered 37-41 % formaldehyde (formalin). For small sample volumes, where no particularly large animals 4% formaldehyde:seawater solution should be appropriate. Where the sample contains debris, tube-dwelling polychaetes, large animals or a lot of residual sediment, especially in compact clay sediments, formalin concentration should be increased to 10 % even 20 %.

There should be at least the same amount of solution in the sample container as solid material. Large shells may be opened to allow the fixative to penetrate to animal tissues. Fixative and sample material should be gently mixed by stirring or inverting the sample containers.

For buffering, 100 g of hexamethylenetetramine (Hexamine = Urotropin) shall be used per 1 dm³ of 40% formaldehyde or sodiumtetraborate (= Borax) at a ratio of 1.5g/dm³ formaldehyde. Buffering is necessary to prevent the leaching of calcium from shell material within the sample.

All necessary measures should be taken to avoid health damage by formalin.

5.4 Staining

Staining facilitates the sorting process and increases the sorting efficiency. However, over-staining may hinder identification of species. Staining is optional according to staff preference.

Rose Bengal (1 g/dm^3 of 40% formaldehyde), which turns animal protein red is added to the fixation fluid. Alternatively the stain can be applied in the laboratory, where the sample should be washed free from formalin and then immersed in stain ($1 \text{ g Rose Bengal/dm}^3$ of tap water + 5 g of phenol for adjustments to pH 4-5) for 20 minutes.

5.5 Labelling

The sample containers should be indelibly pre-marked with the unique sample information (station designation, sample number, replicate number and date) externally.

In addition samples should be properly labelled internally. The information filled in labels should be sufficient to identify the sample with certainty. The mandatory fields are date, station designation, depth, sample number, replicate number (additionally the cruise and vessel designations, type of grab, time, sediment type, etc. may be indicated). Labels made of heavy weight and chemically resistant paper should be filled in with a soft carbon pencil, which will not fade in Formalin. Filled in labels are placed inside the sample containers.

5.6 Sample registering

Samples should be properly registered in sample recording sheets (the standard proforma for on-site records is given in Annex 1).

The following information should be recorded in the field:

- project or contract identifier (code);
- geographical coordinates for each sampling station;
- type of positioning system and its accuracy;
- whether or not a buoy was used;
- whether or not the ship was anchored;
- date and time of each sampling station/sample;
- the water depth from which the sample was taken (if more than one sample is taken at a station, the depth range of samples should be recorded);
- the name, type and sampling area of the sampler;
- sieve mesh aperture sizes;
- number of replicate samples;
- depth of sediment in grab as a measure of sample volume;
- comments such as rejected/unapproved samples together with the causes;
- a visual sediment description, including:
 - a description of sediment type (e.g. sand, silt, clay, etc. and their relative proportions), including important notes, e.g., main groups of large, easily visible species present, occurrence of concretions, stones, dead shells, etc.;
 - surface colour and colour change down the sediment profile, if visible;
 - smell, e.g. presence and severity of H_2S ;
 - anthropogenic debris, rubbish, plastics.
- near-bottom temperature and salinity;
- person responsible for sampling.

6. Laboratory routines

6.1 Sorting and taxonomic identification

The basic premise of all macrobenthic sample analyses in the laboratory is that all specimens extracted from the samples are to be identified to the lowest possible taxonomic level and counted.

Small portions of the unsorted material shall be put on a set of tightly connected sieves with mesh size 0.5 mm and 1 mm and washed with tap water under a flume extractor, so that sorters are not exposed to formalin vapour. In case of large megafauna (bivalves, gastropods, etc.) present in the sample the use of third sieve with mesh size 5 mm may be considered. Sample fractions should be analysed separately.

The sample material should always be sorted using suitable magnification (magnification lamp, stereo-microscope).

Initially sorting is done by a technician into four major taxonomic groups: segmented worms (Annelida), animals with shells (Mollusca), animals with jointed limbs (Arthropoda), other marine invertebrates, which are placed in separate sample vials with identification labels. Once the major sorting has taken place, it is best that each group is identified to the lowest possible level by a specialised researcher. When identifying species there inevitably will be cases when specimens cannot be identified to species due to damage or unsolved taxonomic problems. In case of doubtful identification the lowest reliable taxonomic level should be given. If there is only one species within a genus, then this is indicated by “sp.” following the genus (e.g. *Capitella* sp.), and if it is certain that more than one species is found then this is indicated by “spp.” (e.g. *Capitella* spp.).

The three major taxonomic groups – Polychaeta, Mollusca and Crustacea should be identified to the species level. These are the richest groups in the Black Sea and generally contribute mostly to the abundance and biomass of macrozoobenthos. Anthozoa, Echinodermata, Cephalochordata, Phoronidea and Pantopoda shall also be identified to the species level, since sufficient taxonomic expertise and keys are available in the Black Sea region. Nemertini, Turbellaria, Oligochaeta, Chironomidae and insects in general may be identified to higher taxonomic level (Phylum or Class) for routine monitoring purposes.

Taxonomic guides and keys used for the identification of organisms should be reported with the data. The reference list of taxonomic literature to be employed for the identification of the Black Sea macrofauna is attached as Annex 3.

In order to overcome taxonomic discrepancies due to usage of synonymous names common nomenclature shall be used according to the European Register of Marine Species (ERMS) available on <http://www.marbef.org/data/ermsearch.php>. This will facilitate comparison of data not only within the Black Sea region but also with other European seas.

A taxonomic reference collection should also be available for training and verification purposes.

A checklist of species encountered in the studied area should be established and regularly updated. Provisional list of species from the three major taxonomic groups – Polychaeta, Crustacea and Mollusca is attached as Annex 4.

6.2 Abundance determination

Broken animals shall only be counted as individuals by their heads (e.g. polychaetes) or hinges of bivalves with adhering pieces of tissue.

Taxa that are not sampled quantitatively or that are not truly indicative of sediment conditions shall not be quantified but their presence should be noted. These taxa include Foraminifera, Nematoda, planktonic organisms, benthic fish, and colonial epifauna (Poryfera, Bryozoa, etc.).

6.3 Biomass determination

Biomass can be expressed in a variety of ways (e.g. wet weight, dry weight and ash-free dry weight). As the evaluation of ash-free dry weight (AFDW) ignores the contributions of

inorganic material, water content and all non-living parts to the mass of an organism, it is considered as the most appropriate measure of living biological matter. However, as the determination of AFDW requires combusting specimens, thus removing any possibility for further taxonomic analysis, it is recommended that a non-destructive method be employed. This can be done by measuring wet weight, from which AFDW can be estimated by applying conversion factors obtained from the literature, backed up by local calibration where necessary.

Recently collected material is kept in buffered fixative for a recommended period of three months before wet-weight analysis, to stabilise the mass. Practical issues relating to survey demands may dictate earlier analysis, in which case absolute values may be unreliable, but spatial information can still be informative. The wet weight is obtained by weighing after the external fluid has been removed on filter paper. The animals are placed on filter paper and moved around until no more wet traces are left behind, ensuring that undue pressure is not applied. Animals with shells are generally weighed with their shells; the water should be drained off bivalves before weighing. All tube dwelling species (polychaetes) should be removed from their tubes. Echinoids and ascidians should be punctured to drain off the water before blotting on filter paper. As soon as dry, the specimens are transferred to tared container on the balance. Balance should be accurate to 0.0001 g. After 30 seconds has elapsed the weight of animals is recorded to 0.0001 g. However, where a taxon weighs less than this, the weight is recorded as 0.0001 g. The container should be re-tared before weighing the next taxon. If some of the laboratories are deficient in balances with the required accuracy, then they shall determine wet weight by means of standard biomass conversion tables, which shall be common for all of the laboratories involved in the Black Sea Integrated Monitoring and Assessment Programme (BSIMAP).

The dry weight shall be estimated after drying the formalin material at 60°C to constant weight (for 12-24 hours, or an even longer time, depending on the thickness of the material).

Ash-free dry weight should be estimated after measuring dry weight. It is determined after incineration at 500 °C in an oven until weight constancy is reached (~6 hours, depending on sample and object size). The temperature of the oven should be checked with a calibrated thermometer because there may be considerable temperature gradients (up to 50 °C) in a muffle furnace. Caution is advised to avoid exceeding a certain temperature (> 550 °C), at which a sudden loss of weight may occur owing to the formation of CaO from the skeletal material of many invertebrates (CaCO₃). This can reduce the weight of the mineral fraction by 44 %. Before weighing, the samples must be kept in a desiccator while cooling down to room temperature after oven drying or removal from the muffle furnace.

7. Data reporting

Data obtained from laboratory analysis are entered into standard data report sheets (an example pro-forma is given in Annex 2). The following information should be included in scientific reporting:

- complete list of taxa recorded, including those that are not quantified;
- abundance (number of individuals) within each taxon;
- biomass within each taxon;
- appropriate metadata (e.g. location of sampling site, sampling depth, type and area of sampler, mesh size of sieves, etc.), which are necessary for the correct interpretation of data.

The final dataset obtained from a survey should be recorded as a taxon by abundance (or biomass) by station matrix in electronic spreadsheet format. Abundance and biomass data should be normalised per m².

All data entered into electronic spreadsheets must be proof-read.

8. Recommendations for Quality Assurance

Quality assurance measures should focus on the following areas:

8.1 Equipment calibration

The technical quality of the equipment should be verified on regular basis. The most important of these involve:

- accuracy of depth and positioning fixing equipment;
- sampling area of grab;
- sieve aperture size;
- recalibration of eyepiece gratitudes and microscope maintenance.

8.2 Training

- Experienced and well-trained personnel are a basic prerequisite for maintaining high level quality standards of sampling and analysing procedures. Proper training and education should be given to all staff involved in field and laboratory work and documentation.
- Ring tests and intercalibration exercises on a regional basis should be undertaken regularly and be obligatory for institutions delivering data to the Black Sea Integrated Monitoring and Assessment Programme (BSIMAP). They should be open to all institutions including private industry. Technicians and taxonomic identifiers who carry out the actual procedures rather than managing scientists should take part in the exercises.

8.3 Repeatability of site positioning.

Exact positioning and correct depths when sampling should be noted in the Sample record sheet to avoid comparisons between samples taken at different localities (although noted as the same station in the protocols). If exact positioning due to weather or technical problems is impossible, then fix station work to the correct depth.

8.4 Quality and quantity of the sample.

- The criteria for sample rejection are as follows:
 - less than 5 litres of sample volume is obtained by 0.1 m² grab in soft sediments or less than 2.5 litres in hard-packed sand. (5 l approximates to a depth of 7 cm, while 2.5 l approximates to a depth of 5 cm. Measures of sample depth are taken vertically at the centre of the closed grab buckets.);
 - incomplete closure is noted;
 - an obvious uneven bite is noted;
 - spillage during transferring of samples is observed;
 - samples clearly deviate from the other samples (e.g., there is an observed change from clean sand samples to mussel bank samples). The samples should be nevertheless kept, in order to record faunal patchiness, but another sample should be taken to replace it in calculating the mean for the station.
- Sieving of samples in water bath is recommended as the gentlest way of washing samples.
- The use of large sieves is encouraged because:
 - the risk of clogging is kept low;
 - they reduce the risk of spilling when transferring samples from containers/buckets to the sieves.

8.5 Accuracy and traceability of sample numbering and registration

- Each sample must be sieved, stored and documented separately.
- Samples should be properly marked externally and labelled internally to insure unambiguous identification.

- Samples should be properly registered in standard sample recording sheets (an example pro-forma for on-site records is given in Annex 1).

8.6 Accuracy of sample sorting and taxon identification

- It is advisable to stain the samples to facilitate sorting, if this does not hamper species identification.
- The sorting efficiency of the personnel sorting the samples should be checked by an experienced sorter. At least 5 % of the processed samples, randomly selected, should be subjected to quality control of the sorting efficiency. If the laboratory staff is inexperienced, then the percentage should be increased to 10 %.
- The species determination should be checked by an experienced identifier. At least 5 % of the processed samples, randomly selected, should be subjected to quality control of the identification efficiency. If the laboratory staff is inexperienced, then the percentage should be increased to 10 %.
- A list of literature used for taxonomic identification should be compiled and reported with the data. The list should be updated regularly and should reflect recent advances in the taxonomic literature.
- Regional taxonomical workshops and intercalibration exercises should be held on a regular basis and be attended by every laboratory.
- A checklist of species in the area should be developed, distributed to the participating laboratories and updated regularly.
- Specimens of each taxon should be placed in separate vials in reference collections. A separate reference collection may be required for individual surveys to make later taxonomic checks possible.

8.7 Accuracy of data compilation

- All datasets must be proof-read after input to the computer, before usage.
- One way to check the quality of numbers in the database is to compare individual mean weights. If they are abnormally high or low, the figures need verification.

8.8 In-house Quality Assurance

- It is recommended that organisations should prepare their own in-house procedures and training records, including, but not limited to, the following aspects of the work:
 - records of training and experience of survey personnel;
 - records of training and experience of laboratory staff;
 - procedures for handling survey equipment;
 - procedures for collection, processing and analysis of macrobenthic samples;
 - procedures for recording biological and environmental data.
- Signed protocols should be obligatory for all steps in the analyses.
- Taxonomic certification of the persons responsible at the laboratories is recommended.

9. Quality Control routines

Independent re-analysis of samples of the benthic macrofauna by second researcher should be done for 5-10 % of samples.

9.1 Extraction efficiency – total taxa target

To achieve a pass, the number of taxa extracted should be within $\pm 10\%$ or ± 2 taxa (whichever is greater) of this total.

9.2 Extraction efficiency – total individuals target

The total should be within $\pm 10\%$ or ± 2 individuals (whichever is greater) of the total resulting from re-analysis of samples.

9.3 Total wet weight biomass target

The total value should be ± 20 % of the value obtained from re-analysis of the sample.

9.4 Bray-Curtis comparison

Comparison of the two untransformed data sets, arising from the work of the participating laboratory and from independent re-analysis, should result in a Bray-Curtis Similarity Index of ≥ 90 %.

$$S_{jk} = 100 \left\{ 1 - \frac{\sum_{i=1}^p |y_{ij} - y_{ik}|}{\sum_{i=1}^p (y_{ij} + y_{ik})} \right\} \quad (\text{IV.2.6})$$

where S_{jk} – Similarity index of samples j and k ;
 y_{ij} – abundance of i th species in sample j ;
 y_{ik} – abundance of i th species in sample k ;

Data flags are applied using a graded system related to the untransformed Bray-Curtis scores as follows:

100 % BCSI:	Excellent
95-<100 % BCSI:	Good
90-95 % BCSI:	Acceptable
85-90 % BCSI:	Poor – remedial actions suggested
<85 % BCSI:	Fail - remedial actions required

If the results obtained from the re-analysis of samples do not meet the set targets, than all the samples in the batch should be re-quantified after remedial actions have been taken.

10. Data analysis and metrics

Metrics are called all primary variables or derivative indices that describe the community attributes (biological descriptors) in terms of species richness/composition, quantity (abundance/biomass), structure and function.

The approaches that have been developed in order to explain and reveal the impact of pressures (physical, chemical and biological) on benthic communities can be grouped into three classes:

- **univariate measures/indices** such as number of species, abundance, biomass; diversity indices based on richness/abundance counts; taxonomic diversity indices based on the taxonomic spread of species; biotic indices based on functional attributes such as trophic mode or ecological strategy; abundance/biomass ratios, etc.;
- **multi-metric indices** combining several measures of community response to stress into a single index;
- **multivariate methods** describing the assemblages pattern, including modelling.

Provisional list of macrozoobenthos metrics/indices to be tested in the environmental status assessment of the Black Sea is given below:

- Primary measures:
 - Number of species
 - Presence/absence of identified sensitive species
 - Abundance (total, of identified sensitive/tolerant/opportunistic taxa)
 - Biomass (total, of identified sensitive/tolerant/opportunistic taxa)
- Ratios and proportions:

- Total abundance/total biomass ratio
- Proportion of sensitive/tolerant/opportunistic taxa from the total number of taxa, abundance or biomass
- Diversity indices:
 - Margalef's Species richness index, d
 - Pielou's evenness index, j (Pielou, 1966)
 - Simpson's dominance index, c (Simpson, 1949)
 - Shannon-Wiener diversity index H' (Lloyd, Zar and Karr, 1968)
 - Expected number of species (ES) Hurlbert (1971)
 - Average taxonomic diversity Δ (Warwick & Clarke 1995, 1998),
 - Average taxonomic distinctness Δ^+ (Warwick & Clarke 1995, 1998, Clarke & Warwick 1998)
 - Variation in taxonomic distinctness Δ^+ (Warwick & Clarke 2001, Clarke & Warwick 2001)
- Biotic indices:
 - Infaunal Trophic Index (ITI) (Word, 1978)
 - Biological Quality Index (BQI) (Wilson *et al.*, 1985)
 - AZTI Marine Biotic Index (AMBI) (Borja *et al.*, 2000)
 - BENTIX (Simboura, Zenetos, 2002)
 - Benthic Quality Index (BQI) (Rosenberg *et al.*, 2004)
- Multi-metric indices:
 - Benthic Index of Biotic Integrity (B-IBI) (Weisberg *et al.*, 1997).
- Multivariate approaches. A range of data analyses procedures is available, extensively described in Clarke and Warwick (1994). Hierarchical clustering and ordination analyses have been recommended for assessments of benthic faunal data sets. Such techniques are most effective in the analysis of large data sets based on many samples and allow direct linkages to be made between biological variance and changes in specific environmental factors, including the ranking of such factors in order of importance.

Important remarks concerning the implementation of the above metrics/indices and multivariate statistical analysis methods include:

- Preliminary adaptation of the biotic and multimetric indices (ITI, AMBI, BENTIX and B-IBI) is necessary for their implementation in the Black Sea. This involves allocation of Black Sea species to respective trophic/ecological groups, selection and statistical testing of candidate metrics to be included in B-IBI.
- Reference values of community metrics (reference conditions) at different seabed habitats should be established.
- Threshold values of community metrics that delineate acceptable from unacceptable ecological status should be defined.
- Comprehensive list of Black sea species shall be compiled for the calculation of the taxonomic indices (Δ , Δ^+ , Δ^+).
- Relevant statistical packages are PRIMER, AMBI, Bio Diversity Programme.

11. Acknowledgements

The preparation of this guideline was supported by GEF-UNDP Project PIMS 3065: Control of eutrophication, hazardous substances and related measures for rehabilitating the Black Sea ecosystem: Phase 2 (BSERP).

We wish to acknowledge Nikita Kucheruk from Shirshov State Institute of Oceanology, Russian Federation for compiling the first draft of the Manual on the basis of HELCOM Manual.

We also wish to gratefully acknowledge the contribution of the following persons for the compilation of the Provisional check list of macrozoobenthic Polychaeta, Crustacea and Mollusca encountered in the Black Sea and Azov Sea (Annex 4): Camelia Dumitrache¹ (Mollusca), Christos Arvanitidis² (Polychaeta), Dragos Micu³ (Mollusca), Nikolai Revkov⁴ (Polychaeta), S. Ünsal Karhan⁵ (Crustacea, Mollusca), Valentina Todorova⁶ (Polychaeta), Victor Surugiu⁷ (Polychaeta).

¹ National Institute for Marine Research and Development "Grigore Antipa"
300 Mamaia Blv.
Ro-900581, Constanta, 3
Romania
cdumitrache@alpha.rmri.ro

² Institute of Marine Biology and Genetics,
Hellenic Center for Marine Research,
Former American Base of Gournes,
71003, Heraklion, Crete,
Greece.
arvanitidis@imbc.gr

³ National Institute for Marine Research and Development "Grigore Antipa"
300 Mamaia Blv.
Ro-900581, Constanta, 3
Romania

⁴ Institute of Biology of Southern Seas, National Academy of Science
2, Nakhimov Av., 99011 Sevastopol
Crimea, Ukraine
revkov@ibss.iuf.net

⁵ Istanbul University Institute of Marine Science and Management
Muskule Sok., №: 1
34116, Vefa-Istanbul
Turkey
unsalkarhan@yahoo.com

⁶ Institute of Oceanology-BAS
9000 Varna
P.O.BOX 152
Parvi maj Str., № 40
Bulgaria
vtodorova@io-bas.bg

⁷ "Al. I. Cuza" University of Iași, Faculty of Biology
Bd. Carol I, 20A,
6600, Iași

Romania
vsurugiu@uaic.ro

Cover photograph was generously provided by Lyubomir Klissurov:

Institute of Oceanology-BAS
9000 Varna
P.O.BOX 152
Parvi maj Str., № 40
Bulgaria
klisurov@ultranet.bg
www.klissurov.dir.bg

12. References

- Borja, A., Franco, J., Pérez, V., 2000. A marine biotic index to establish the ecological quality of soft-bottom benthos within european estuarine and coastal environments. *Mar. Pollut. Bull.* 40 (12), 1100–1114.
- Clark, K.R. and Ainsworth, M., 1993. A method of linking multivariate community structure to environmental variables. *Marine Ecology Progress Series*, 92, 205-219.
- Clarke, K. R. and Warwick, R. M., 1994. Changes in marine communities: an approach to statistical analysis and interpretation. Natural Environmental Research Council, Plymouth.
- Clarke, K.R. & Warwick, R.M., 1998. A taxonomic distinctness index and its statistical properties. *Journal of Applied Ecology*, 35, 523-531.
- Clarke, K.R. & Warwick, R.M., 2001. A further biodiversity index applicable to species lists: variation in taxonomic distinctness. *Marine Ecology - Progress Series*, 216, 265-278.
- Davies J., Baxter J., Bradley M., Connor D., Khan J., Murray E., Sanderson W., Turnbull C. and Vincent M., 2001. *Marine Monitoring Handbook*, ISBN 1 86107 5243, 405 pp.
- Eleftheriou, A. and Holme, N.A., 1984. Macrofauna techniques. p. 140-216. In: *Methods for the study of marine benthos* (N.A. Holm and A.D. McIntyre, eds.). Blackwell Scientific Publications, Oxford, 387 pp.
- Gibson, G. R., M. T. Barbour, J. B. Stribling, J. Gerritsen, and J. R. Karr. 1996. Biological criteria: technical guidance for streams and small rivers. EPA 822-B-96-001. U.S. Environmental Protection Agency, Office of Water, Washington, DC.
- Gray, J. S., McIntyre, A.D., and Štirn, J., 1992. Manual of methods in aquatic environment research. Part 11. Biological assessment of marine pollution with particular reference to benthos. *FAO Fisheries Technical Paper* 324: 49 pp.
- Holme, N.A. & McIntyre, A., 1984. *Methods for the Study of Marine Benthos*. Oxford, 387 pp.
- Hughes, R. M., D. P. Larsen and J. M. Omernik. 1986. Regional reference sites: a method for assessing stream pollution. *Environmental Management* 10: 629-635.
- Hurlbert, S.H., 1971. The nonconcept of species diversity: A critique and alternative parameters. *Ecology* 52: 577-586.
- ICES. 2004. Report of the ICES/OSPAR Steering Group on Quality Assurance of Biological Measurements in the Baltic Sea. 59 pp.
- ICES. 2005. Report of the ICES/OSPAR Steering Group on Quality Assurance of Biological Measurements in the Northeast Atlantic. 59pp.
- ISO 16665: 2005 (E). Water quality – Guidelines for quantitative sampling and sample processing of marine soft-bottom macrofauna.
- Jeffrey, D. W., Wilson, J. G., Harris, C. R. and D.L. Tomlinson, 1985. The application of two simple indices to Irish estuary pollution status. *Estuarine management and quality assessment*. Plenum Press, London. 147-165 pp.
- Lloyd, H., Zar, J.H., and Karr, J.R., 1968. On the calculation of information - theoretical measures of diversity. *Am. Mid Nat.* 79, 257-272.
- Manual for Marine Monitoring in the COMBINE Programme of HELCOM, 2003. Part C. Programme for monitoring of eutrophication and its effects. Annex C-8 Soft bottom macrozoobenthos. <http://sea.helcom.fi/Monas/CombineManual2/PartC/CFrame.htm>

- McIntyre, A. D., 1978. The Benthos of the western North Sea. Rapp. P.-v. Réun. Cons. Int. Explor. Mer, 172: 405-417.
- Pielou, E.C., 1966. The measurement of diversity in different types of biological collections. J. Theor. Biol., 13, 131-144.
- Rees, H.L., 2004. Biological monitoring: General guidelines for quality assurance. ICES Techniques in Marine Environmental Sciences, No. 32. 44 pp.
- Rees, H.L., C. Heip, M. Vincx and Parker M.M., 1991. Benthic communities: use in monitoring point-source discharges. ICES Techniques in Marine Environmental Sciences No. 16, 70 pp.
- Rosenberg, R., M. Blomqvist, H. Nilsson, H. Cederwall and A. Dimming, 2004. Marine quality assessment by use of benthic species-abundance distribution; a proposed new protocol within the EC Water Framework Directive. Marine Pollution Bulletin.
- Rumohr, H., 1990. Soft bottom macrofauna: collection and treatment of samples. ICES Techniques in Marine Environmental Sciences No. 8, 18pp.
- Rumohr, H., 1999. Soft bottom macrofauna: Collection, treatment, and quality assurance of samples. ICES Techniques in Marine Environmental Sciences No. 27, 26 pp.
- Simboura N., Zenetos A., 2002. Benthic indicators to use in Ecological Quality classification of Mediterranean soft bottom marine ecosystems, including a new Biotic Index. Mediterranean Marine Science, 3/2, 77-111.
- Simpson, E.H. (1949). Measurement of diversity. Nature, Lond. 163, 688.
- UK National Marine Monitoring Programme Green Book, 2003. <http://www.sepa.org.uk/marine/index.htm>
- Warwick, R.M. and Clarke, K.R., 1995. New "biodiversity" measures reveal a decrease in taxonomic distinctness with increasing stress. Marine Ecology Progress Series, 129: 301-305.
- Warwick, R.M. & Clarke, K.R., 1998. Taxonomic distinctness and environmental assessment. Journal of Applied Ecology, 35, 532-543.
- Warwick, R.M. & Clarke, K.R., 2001. Practical measures of marine biodiversity based on relatedness of species. Oceanography and Marine Biology: an Annual Review, 39, 207-231.
- Weisberg, S. B., Ranasinghe, J. A., Dauer, D. M., Schaffner, L. C., Diaz, R. J., and Frithsen., J. B., 1997. An estuarine benthic index of biotic integrity (B-IBI) for the Chesapeake Bay. Estuaries 20:149-158.
- Word, J. Q., 1979. The Infaunal Trophic Index. Sth Calif. Coast. Wat. Res. Proj. Annu. Rep., El Segundo, California. 19-39.

SAMPLE RECORD - MACROZOOBENTHOS

No

Project/contract identifier:

Cruise duration: from to

Vessel (name and type¹):

Positioning system²:

Sampling precision³:

Sediment sampler type/area:

Mesh size of sieves (mm):

Date:

Time:

Station/site designation:

Sample №:

Replicate №:

Coordinates: Lat.:

Long.:

Depth (m):.....

Depth of sediment in sampler (cm):

T_{H2O} bottom (°C)

S_{H2O} bottom (‰).....

Sediment type⁴ and observations⁵:

.....

.....

.....

.....

Person responsible for collecting:

.....

Annex 2

DATA REPORT SHEET - MACROZOOBENTHOS

Project/contract identifier:

Vessel (name and type¹):

Positioning system²:

Sampling precision³:

Date:

Time:

Station/site designation:

Coordinates:

Depth (m):

Sample No:

Replicate №:

Sediment sampler type/area:

Mesh size of sieves (mm):.....

Sediment type⁴ and observations⁵:

T_{H2O} bottom (°C):

S_{H2O} bottom (‰):

Person responsible for collecting:

[illegible]

Annex 3

List of taxonomic literature to be employed for the identification of macrozoobenthic species in the Black Sea

- Bacescu, M., 1951. Cumacea. In: Fauna Romaniei, 1(1), Ed. Academiei Romane, 1-94, Bucuresti.
- Bacescu, M., 1982. Contributions à la connaissance des Cumacés de la Mer de Marmara et d'Egée (Ile Eubea). Travaux Museum d'Histoire naturelle "Grigore Antipa", 24, 45-54.
- Ball, I.R. & Reynoldson, T.B., 1981. British planarians. Synopses Br. Fauna (N.S.) No. 19. Linnean Society of London. Estuarine and Brackish Water Sciences Association. Bath: Cambridge University Press.
- Bellan-Santini, D., Diviacco, G., Krapp-schickel, G., Myers, A.A. and Ruffo, S., 1989. Part 2. Gammaridea (Haustoriidae to Lysianassidae [Ruffo, S. (Ed.) The Amphipoda of the Mediterranean, Mémoires de l'Institut océanographique, 13, 365-576, Monaco]
- Bellan-Santini, D., Karaman, G., Krapp-Schickel, G., Ledoyer, M. and Ruffo, S., 1993. Part 3. Gammaridea (Melpodidae to Talitridae), Ingolfiellidea, Caprellidea. [Ruffo, S. (Ed.) The Amphipoda of the Mediterranean, Mémoires de l'Institut océanographique, 13, 577-813, Monaco]
- Bellan-Santini, D., Karaman, G., Krapp-Schickel, G., Ledoyer, M., Myers, A.A., Ruffo, S. and Schiecke, U., 1982. Part 1. Gammaridea (Acanthonotozomatidae to Gammaridae) [Ruffo, S. (Ed.) The Amphipoda of the Mediterranean, Mémoires de l'Institut océanographique, 13, 1-364, Monaco]
- Bellan-Santini, D., Karaman, G., Ledoyer, M., Myers, A.A., Ruffo, S. and Vader, W., 1998. Localities and Map, Addenda to Parts 1-3, Ecology, Faunistics and Zoogeography, Bibliography, Index. [Ruffo, S. (Ed.) The Amphipoda of the Mediterranean, Mémoires de l'Institut océanographique, 13, 814-959, Monaco]
- Bouvier, E.L., 1923. Pycnogonides. Faune de France, Vol. 7, Paris.
- Day, J.H., 1967. A monograph on the Polychaeta of southern Africa. Trustees of British Museum (Natural History), London.
- Ergen, Z., Çinar, M.E. and Ergen, G., 2000. On the Nereididae (Polychaeta: Annelida) of the Bay of Izmir. Zoology in the Middle East, 21, 139-158.
- Fage, L., 1951. Cumacés. Faune de France, Vol. 54, Paris.
- Fauchald, K., 1977. The polychaet worms. Definitions and keys to the orders, families and genera. Natural History Museum, Los Angeles Country, Sci. Ser. 28.
- Fauvel, P., 1923. Polychètes errantes. Faune de France, Vol. 5, Paris.
- Fauvel, P., 1927. Polychètes sédentaires. Faune de France, Vol. 16, Paris.
- Gaillard, J.M., 1987. Gasteropodes. [Fischer, W., Schneider, M. ve Bouchot, M.L. (Ed.) Fishes FAO d'identification des especes pour les besoins de la peche. Méditerranée et Mer Noire, Zone de peche 37, Vol. I.: Végétaux et invertébrés, 312-367, Rome]
- Ghisotti, F., Melone, G., 1972. Cotalogo Illustrate delle Conchiglie Marine del Mediterraneo 4. Trochidae, Conchiglie VIII., 11–12., 47–77.

- Ghisotti, F.–Melone, G., 1971. Catalogo Illustrato delle Conchiglie Marine del Mediterraneo 3. Trochidae, Conchiglie VII., 1–2., 47–77.
- Ghisotti, F.–Melone, G., 1975. Catalogo Illustrato delle Conchiglie Marine del Mediterraneo, 5 Trochidae, Conchiglie IX., 11–12., 147–208.
- Hayward, P.J. and Ryland, J.S. (Ed.), 1996. Handbook of the marine fauna of North-West Europe. Oxford University Press.
- Holthuis, L.B., 1987. Vrais Crabes. [Fischer, W., Schneider, M. and Bouchot, M.L. (Ed.) Fishes FAO d'identification des especes pour les besoins de la peche. Méditerranée et Mer Noire, Zone de peche 37, Vol. I.: Végétaux et invertébrés, 312-367, Rome]
21. Kirkim, F., 1998. Ege Denizi Isopoda (Crustacea) faunasının sistemiği ve ekolojisi üzerine araştırmalar. Doktora Tezi, Ege Üniversitesi., İzmir.
- Koehler, R., 1921. Echinodermes. Faune de France, Vol. 1, Paris.
- Manuel, R.L., 1987. British Anthozoa (Coelenterata: Octocorallia and Hexacorallia). Synopses of the British Fauna, new ser., no:18.
- Nordsieck, F., 1972. Die europäischen Meereschnecken (Opisthobranchia mit Pyramidellidae, Rissoacea) von Eismeer bis Kapverden, Mittelmeer und Schwarzes Meer. Gustav Fischer Verlag, Stuttgart.
- Parenzan, P., 1970. Carta d'identità delle conchiglie del Mediterraneo, Vol. I, Gasteropodi, Ed. Bios Taras, Taranto.
- Parenzan, P., 1974. Carta d'identità delle conchiglie del Mediterraneo, Vol. II, Bivalvi, Prima Parte, Ed. Bios Taras, Taranto
- Parenzan, P., 1976. Carta d'identità delle conchiglie del Mediterraneo, Vol. II, Bivalvi, Seconda Parte, Ed. Bios Taras, Taranto.
- Poppe, G.T. ve Goto, Y., 1993. European seashells. Vol. 2 (Scaphopoda, Bivalvia, Cephalopoda). Verlag Christa Hemmen, Wiesbaden.
- Poutiers, J.M., 1987. Bivalves. [Fischer, W., Schneider, M. ve Bouchot, M.L. (Ed.) Fishes FAO d'identification des especes pour les besoins de la peche. Méditerranée et Mer Noire, Zone de peche 37, Vol. I.: Végétaux et invertébrés, 312-367, Rome]
- Preda, S., 1995. Diversitatea Lumii VII. Determinatorul Ilustrat al florei Si faunei Romaniei. Vol. I – Medul Marin.
- Riedl, R., 1983. Fauna und Flora des Mittelmeeres. Verlag Paul Parey, Hamburg und Berlin.
- Tebble, N., 1966. British bivalve seashells. Trustees of British Museum (Natural History) London.
- Tortonese, E., 1965. Echinodermata. Fauna d'Italia. Vol. 6. Edizioni Calderini, Bologna.
- Walker-Smith, G.K. and Poore, G.C.B., 2001. A phylogeny of the Leptostraca (Crustacea) with keys to families and genera. Memoirs of Museum Victoria, 58 (2), 383-410.
- Zariquiey Alvarez, R., 1968. Crustáceos Decápodos Ibéricos. Investigación Pesquera, no: 32, Barcelona.
- Гурьянова Е.Ф., 1951. Бокоплавы морей СССР. Определители по фауне СССР, вып.41. Изд-во. АН СССР.
- Жадин В. И., 1952. Моллюски пресных и солоноватых вод СССР. Определители по фауне СССР, вып. 46. Изд-во АН СССР.

- Киселева М.И., 2004. Многощетинковые черви (Polychaeta) Черного и Азовского морей. Изд-во Кольского научного центра РАН, 409 с.
- Маринов Т., 1977. Фауна на България т. 6, Многощетинести червеи (Polychaeta), Издателство на БАН, София, 258 стр.
- Мордухай-Болтовской Ф. Д., 1968. Определитель фауны Черного и Азовского морей, Том первый, Наукова думка, Киев, 437 с.
- Мордухай-Болтовской Ф. Д., 1969. Определитель фауны Черного и Азовского морей, Том второй, Наукова думка, Киев, 536 с.
- Мордухай-Болтовской, Ф. Д., 1972. Определитель фауны Черного и Азовского морей, Том третий, Наукова думка, Киев, 340 с.

Annex 4

Provisional check list of macrozoobenthic Polychaeta, Crustacea and Mollusca encountered in the Black Sea and Azov Sea

POLYCHAETA

Acholoe astericola (Delle Chiaje, 1828)
Harmothoe boholensis (Grube, 1878)
Harmothoe extenuata (Grube, 1840)
Harmothoe imbricata (Linnaeus, 1767)
Harmothoe impar (Johnston, 1839)
Harmothoe minuta (Potts, 1910)
Harmothoe spinifera (Ehlers, 1864)
Lepidasthenia maculata Potts, 1910
Subadyte pellucida (Ehlers, 1864)
Polynoe scolopendrina Savigny, 1822
Lepidonotus carinulatus (Grube, 1869)
Lepidonotus squamatus (Linnaeus, 1758)
Eupanthalis kinbergi McIntosh, 1876
Pholoe synophthalmica Claparede, 1868
Sthenelais boa (Johnston, 1833)
Labioleanira yhleni (Malmgren, 1867)
Vigtorniella zaikai (Kisseleva, 1992)
Chrysopetalum debile (Grube, 1855)
Pisione remota (Southern, 1914)
Phyllodoce laminosa Savigny, 1818
Phyllodoce lineata (Claparede, 1870)
Phyllodoce maculata (Linnaeus, 1767)
Phyllodoce madeirensis Langerhans, 1880
Phyllodoce mucosa Oersted, 1843
Phyllodoce vittata Ehlers, 1864
Phyllotethys koswigi La Greca, 1949
Nereiphylla nana Saint-Joseph, 1906
Nereiphylla paretii Blainville, 1828
Nereiphylla rubiginosa (Saint-Joseph, 1888)
Eteone picta Quatrefages, 1865
Eteone siphonodonta (Delle Chiaje, 1822)
Eulalia viridis (Linnaeus, 1767)
Eumida sanguinea (Oersted, 1843)
Pterocirrus limbata Claparede, 1868
Pterocirrus macroceros (Grube, 1860)
Hesionura coineau (Laubier, 1962)
Pseudomystides limbata (Saint-Joseph, 1888)
Hesion *pantherina* Risso, 1826
Hesionides arenaria Friedrich, 1937
Microphthalmus fragilis Bobretzky, 1870
Microphthalmus szcelkowi Meczniow, 1865
Microphthalmus similis Bobretzky, 1870
Kefersteinia cirrata (Keferstein, 1862)
Gyptis propinqua Marion & Bobretzky, 1875
Ophiodromus flexuosus (Delle Chiaje, 1822)

Sigambra constricta (Southern, 1921)
Sigambra tentaculata (Treadwell, 1914)
Ancistrosyllis rigida Fauvel, 1919
Haplosyllis spongicola (Grube, 1855)
Syllis amica Quatrefages, 1865
Syllis armillaris (O.F. Muller, 1776)
Syllis gracilis Grube, 1840
Syllis hyalina Grube, 1863
Syllis monilaris Savigny, 1818
Syllis prolifera Krohn, 1852
Syllis variegata Grube, 1860
Syllis vittata (Grube, 1840)
Ehlersia cornuta (Rathke, 1843)
Amblyosyllis formosa (Claparede, 1863)
Pionosyllis pulligera (Krohn, 1852)
Pseudosyllis brevipennis Grube, 1863
Streptosyllis varians Webster & Benedict, 1887
Syllides longocirrata Oersted, 1845
Eusyllis assimilis Marenzeller, 1875
Xenosyllides violacea Perajaslawzewa, 1891
Trypanosyllis coeliaca (Claparede, 1868)
Trypanosyllis zebra (Grube, 1860)
Petitia amphophthalma Siewing, 1955
Pseudobrania limbata (Claparede, 1868)
Pseudobrania clavata (Claparede, 1863)
Brania tenuicirrata (Claparede, 1864)
Exogone naidina Oersted, 1845
Exogone hebes (Webster & Benedict, 1884)
Sphaerosyllis bulbosa Southern, 1914
Sphaerosyllis claparedii Ehlers, 1864
Sphaerosyllis hystrix Claparede, 1863
Sphaerosyllis ovigera Langerhans, 1879
Autolytus prolifer (O.F. Muller, 1776)
Autolytus rubrovittatus Claparede, 1864
Proceraea aurantiaca (Claparede, 1868)
Namanereis littoralis (Grube, 1872)
Namanereis pontica (Bobretzky, 1872)
Ceratonereis costae (Grube, 1840)
Nereis pelagica Linnaeus, 1758
Nereis rava Ehlers, 1868
Nereis zonata Malmgren, 1867
Eunereis longissima (Johnston, 1840)
Hediste diversicolor (O.F. Muller, 1776)
Neanthes fucata (Savigny, 1820)
Neanthes succinea (Frey & Leuckart, 1847)
Perinereis cultrifera (Grube, 1840)
Platynereis dumerilii (Audouin & M.-Edwards, 1833)
Platynereis coccinea (Delle Chiaje, 1841)
Websterinereis glauca (Claparede, 1870)
Nephtys caeca (Fabricius, 1870)
Nephtys ciliata (O.F. Muller, 1776)
Nephtys cirrosa Ehlers, 1868
Nephtys hombergii Savigny, 1818

Nephtys hystricis McIntosh, 1900
Nephtys incisa Malmgren, 1865
Nephtys longosetosa Oersted, 1842
Nephtys paradoxa Malmgren, 1874
Micronephthys stammeri (Ehlers, 1887)
Glycera alba (O.F. Muller, 1776)
Glycera capitata Oersted, 1843
Glycera gigantea Quatrefages, 1865
Glycera rouxii Audouin & Milne-Edwards, 1833
Glycera tessellata Grube, 1863
Glycera tridactyla Schmarda, 1861
Glycera unicornis Savigny, 1818
Goniadella bobretzkyi (Annenkova, 1929)
Ephesiella peripstus (Claparede, 1863)
Sphaerodorum claparedii (Greef, 1866)
Sphaerodorum gracile (Rathke, 1843)
Chloeia venusta Quatrefages, 1865
Aponuphis bilineata (Baird, 1870)
Hyalinoecia fauveli Rioja, 1918
Nothria conchylega (M. Sars, 1835)
Onuphis eremita Audouin & Milne-Edwards, 1833
Eunice harassii Audouin & Milne-Edwards, 1833
Eunice pennata (O.F. Muller, 1776)
Eunice torquata Quatrefages, 1865
Eunice vittata Delle Chiaje, 1829
Lysidice ninetta Audouin & Milne-Edwards, 1833
Marphysa bellii (Audouin & Milne-Edwards, 1833)
Nematonereis unicornis (Grube, 1840)
Lumbrineris gracilis (Ehlers, 1868)
Lumbrineris latreillii (Audouin & M.-Edwards, 1834)
Scoletoma debilis (Grube, 1878)
Scoletoma funchalensis (Kinberg, 1865)
Scoletoma tetraura (Schmarda, 1861)
Drilonereis filum (Claparede, 1868)
Dorvillea rubrovittata (Grube, 1855)
Protodorvillea kefersteini (McIntosh, 1869)
Schistomeringos neglectus (Fauvel, 1923)
Schistomeringos rudolphii (Delle Chiaje, 1828)
Dinophilus gyrociliatus O. Schmidt, 1857
Trilobodrilus heideri Remane, 1925
Nainereis laevigata (Grube, 1855)
Orbinia cuvierii (Audouin & Milne-Edwards, 1833)
Orbinia latreillii (Audouin & M.-Edwards, 1833)
Protoaricia oerstedii (Claparede, 1864)
Aonides oxycephala (M. Sars, 1872)
Aonides paucibranchiata Southern, 1914
Laonice cirrata (M. Sars, 1851)
Malacocerus fuliginosus (Claparede, 1868)
Malacocerus tetracerus (Schmarda, 1861)
Malacocerus vulgaris (Johnston, 1827)
Microspio mecznikowianus (Claparede, 1868)
Polydora caulleryi Mesnil, 1897
Polydora ciliata (Johnston, 1838)

Polydora cornuta Bosc, 1802
Polydora limicola Annenkova, 1934
Polydora websteri Hartman, 1943
Pseudopolydora antennata (Claparede, 1868)
Prionospio steenstrupi Claparede, 1867
Minuspio cirrifera (Wiren, 1883)
Pygospio elegans Claparede, 1863
Scolecopsis cirratulus (Delle Chiaje, 1828)
Pseudomalacoceros cantabra (Rioja, 1918)
Parascolecopsis tridentata (Southern, 1914)
Spio decoratus (O.F. Muller, 1776)
Spio filicornis (O.F. Muller, 1766)
Spio multioculata Rioja, 1919
Spiophanes bombyx (Claparede, 1870)
Spiophanes kroyeri reyssi Laubier, 1964
Streblospio benedicti Webster, 1879
Chaetopterus variopedatus (Renier, 1804)
Magelona mirabilis (Johnston, 1865)
Magelona rosea Moore, 1907
Protodrilus flavocapitatus (Uljanin, 1877)
Protodrilus purpureus (Schneider, 1868)
Saccocirrus papillocercus Bobretzky, 1872
Aricidea assimilis Tebble, 1959
Aricidea claudiae (Laubier, 1967)
Cirrophorus lyriformis (Annenkova, 1934)
Cirrophorus neapolitanus (Cerruti, 1909)
Paradoneis lyra (Southern, 1914)
Levinsenia gracilis (Tauber, 1879)
Paraonis fulgens (Levinsen, 1883)
Aphelochaeta marioni (Saint-Joseph, 1894)
Caulleriella bioculata (Keferstein, 1862)
Chaetozone caputesocis (Saint-Joseph, 1894)
Cirratulus cirratus (O.F. Muller, 1776)
Cirratulus filiformis Keferstein, 1862
Protocirrinieris chrysoderma (Claparede, 1868)
Cirriformia tentaculata (Montagu, 1808)
Timarete anchylochaeta (Schmarda, 1861)
Timarete dasylophius (Marenzeller, 1879)
Timarete filigera (Delle Chiaje, 1828)
Cossura soyeri Laubier, 1963
Ctenodrilus serratus (Schmidt, 1857)
Stygocapitella subterranea Knollner, 1934
Armandia cirrhosa Philippi, 1861
Ophelia bicornis Savigny, 1818
Ophelia limacina (Rathke, 1843)
Polyophthalmus pictus (Dujardin, 1839)
Scalibregma inflatum Rathke, 1843
Capitella capitata (Fabricius, 1780)
Capitella giardi (Mesnil, 1897)
Capitella minima Langerhans, 1880
Capitellethus dispar (Ehlers, 1907)
Heteromastus filiformis (Claparede, 1864)
Notomastus latericeus Sars, 1851

Notomastus lineatus Claparede, 1870
Notomastus profundus Eisig, 1887
Dasybranchus caducus (Grube, 1846)
Dasybranchus carneus Grube, 1870
Dasybranchus gajolae Eisig, 1887
Arenicola marina (Linnaeus, 1758)
Arenicolides branchialis (Audouin & M.-Edwards, 1833)
Euclymene collaris (Claparede, 1868)
Euclymene lombricoides (Quatrefages, 1865)
Euclymene oerstedii (Claparede, 1863)
Euclymene palermitana (Grube, 1840)
Clymene santanderensis Rioja, 1917
Maldane glebifex Grube, 1860
Praxillella lophoseta (Orlandi, 1898)
Praxillella praetermissa (Malmgren, 1866)
Petaloproctus terricolus Quatrefages, 1865
Leiochone clypeata Saint-Joseph, 1894
Micromaldane ornithochaeta Mesnil, 1897
Owenia fusiformis Delle Chiaje, 1842
Myriochele heeri Malmgren, 1867
Sternaspis scutata (Ranzani, 1817)
Pectinaria belgica (Pallas, 1766)
Pectinaria koreni (Malmgren, 1866)
Pectinaria neapolitana Claparede, 1870
Petta pusilla Malmgren, 1866
Ampharetre acutifrons (Grube, 1860)
Amage adspersa (Grube, 1863)
Amphicteis gunneri (M. Sars, 1835)
Hypania invalida (Grube, 1860)
Hypaniola kowalewskii (Grimm, 1877)
Melinna palmata Grube, 1870
Amphitritides gracilis (Grube, 1860)
Loimia medusa (Savigny, 1818)
Nicolea venustula (Montagu, 1818)
Pista cretacea (Grube, 1860)
Pista cristata (O.F. Muller, 1776)
Proclea graffii (Langerhans, 1884)
Polycirrus aurantiacus Grube, 1860
Polycirrus caliendrum Claparede, 1868
Polycirrus haematodes (Claparede, 1864)
Polycirrus pallidus (Claparede, 1864)
Streblosoma bairdi (Malmgren, 1865)
Thelepus cincinnatus (Fabricius, 1780)
Thelepus triserialis (Grube, 1855)
Terebellides stroemii M. Sars, 1835
Trichobanchus glacialis Malmgren, 1865
Sabellaria spinulosa Leuckart, 1849
Branchiomma vesiculosum (Montagu, 1815)
Chone collaris Langerhans, 1880
Chone filicaudata Southern, 1914
Potamilla torelli Malmgren, 1865
Fabricia stellaris stellaris (Muller, 1774)
Jasmineira caudata Langerhans, 1880

Manayunkia caspica Annenkova, 1929
Oriopsis armandi (Claparede, 1864)
Ditrupa arietina (O.F. Muller, 1776)
Hydroides norvegica Gunnerus, 1768
Salmacina incrustans Claparede, 1870
Serpula vermicularis Linnaeus, 1767
Pomatoceros triqueter (Linnaeus, 1767)
Ficopomatus enigmaticus (Fauvel, 1923)
Vermiliopsis infundibulum (Linnaeus, 1758)
Neodexiospira pseudocorrugata (Bush, 1904)
Janua pagenstecheri (Quatrefages, 1865)
Pileolaria militaris (Claparede, 1868)
Nerilla antennata O. Schmidt, 1848
Polygordius neapolitanus Fraipont, 1882

CRUSTACEA

OSTRACODA

Euphilomedes interpuncta (Baird, 1850)
Polycopse frequens G.W. Mueller, 1894
Paracypris polita G.O. Sars, 1866
Aglaioocypris complanata (Brady & Robertson, 1869)
Propontocypris intermedia (Brady, 1868)
Pontocythere bacescoi (Caraion, 1960)
Eucytherura bulgarica Klie, 1937
Cyprideis torosa (Jones, 1850)
Cytheridea acuminata (Bosquet, 1852)
Eucythere prava Brady & Robertson, 1869
Cytheromorpha fuscata (Brady, 1869)
Microcytherura nigrescens G.W. Mueller, 1894
Leptocythere fabaeformis (G.W. Mueller, 1894)
Leptocythere macallana (Brady & Robertson, 1869)
Leptocythere multipunctata (Seguenza, 1942)
Leptocythere ramosa (Rome, 1942)
Leptocythere rara (G.W. Mueller, 1894)
Callistocythere crispata (Brady, 1868)
Callistocythere diffusa (G.W. Mueller, 1894)
Callistocythere flavidofusca (Ruggieri, 1950)
Callistocythere mediterranea (G.W. Mueller, 1894)
Buntonia (Rectobuntonia) subulata Ruggieri, 1954
Carinocythereis carinata (Roemer, 1838)
Pterygocythereis jonesii (Baird, 1850)
Costa edwardsii (Roemer, 1838)
Costa runcinata (Baird, 1850)
Aurila convexa (Baird, 1850)
Cythereis amnicola (G.O. Sars, 1887)
Cythereis rubra pontica Dubowsky, 1939
Urocythereis margaritifera (G.W. Mueller, 1894)
Limnocythere inopinata (Baird, 1850)
Cytheroma karadagiensis Dubowsky, 1939
Cytheroma variabilis G.W. Mueller, 1894
Loxoconcha bairdi (G.W. Mueller, 1894)

Loxoconcha globosa Schornikov, 1965
Loxoconcha granulata G.O. Sars, 1866
Loxoconcha elliptica Brady, 1868
Loxoconcha pontica Klie, 1937
Loxoconcha rhomboidea (Fischer, 1855)
Hemicytherura videns (G.W. Mueller, 1894)
Semicytherura acuticostata (G.O. Sars, 1866)
Semicytherura alifera Ruggieri, 1959
Cytheropteron rotundatum G.W. Mueller, 1894
Xestoleberis decipiens (G.W. Mueller, 1894)
Xestoleberis corneli Caraion, 1963
Xestoleberis aurantia acutipenis Caraion, 1963
Xestoleberis aurantia aurantia (Baird, 1838)
Microcythere longiantennata Marinov, 1962
Microcythere varnensis Marinov, 1962
Parvocythere hartmanni Marinov, 1962
Bythocythere turgida G.O. Sars, 1866
Sclerochilus gewemuelleri dubowskyi Marinov, 1962
Cytherois vitrea (G.O. Sars, 1866)
Paradoxostoma abbreviatum G.O. Sars, 1866
Paradoxostoma guttatum Schornikov, 1965
Paradoxostoma intermedium G.W. Mueller, 1894
Paradoxostoma mediterraneum G.W. Mueller, 1894
Paradoxostoma ponticum Klie & Whittaker, 1942
Paradoxostoma simile G.W. Mueller, 1894
Paradoxostoma variabile (Baird, 1835)
Cyprinotus inaequalis Bronstein, 1928
Heterocypris incongruens (Ramdohr, 1808)
Heterocypris salina (Brady, 1886)
Potamocypris steueri Klie, 1935
Potamocypris villosa (Jurine, 1820)

CIRRIPEDIA

Balanus eburneus Gould, 1841
Balanus improvisus Darwin, 1854
Chthamalus stellatus (Poli, 1795)
Euraphia depressa (Poli, 1795)
Verruca spengleri Darwin, 1854

DECAPODA

Hippolyte longirostris (Czerniavsky, 1868)
Hippolyte sapphica d'Udekem d'Acoz, 1993
Lysmata seticaudata (Risso, 1816)
Alpheus dentipes Guérin-Méneville, 1832
Athanas nitescens (Leach, 1814)
Palaemon adspersus Rathke, 1837
Palaemon elegans Rathke, 1837
Palaemon serratus (Pennant, 1777)
Crangon crangon (Linnaeus, 1758)
Philocheras fasciatus (Risso, 1816)
Philocheras trispinosus (Hailstone, 1835)

Processa edulis (Risso, 1816)
Homarus gammarus (Linnaeus, 1758)
Astacus leptodactylus Eschscholtz, 1823
Astacus pachypus Rathke, 1837
Upogebia pusilla (Petagna, 1792)
Callianassa truncata (Giard & Bonnier, 1890)
Pestarella candida (Olivi, 1792)
Clibanarius erythropus (Latreille, 1818)
Diogenes pugilator (Roux, 1829)
Pisidia longimana (Risso, 1816)
Macropodia longirostris (Fabricius, 1775)
Macropodia rostrata (Linnaeus, 1761)
Callinectes sapidus Rathbun, 1896
Carcinus aestuarii Nardo, 1847
Pirimela denticulata (Montagu, 1808)
Polybius depurator (Linnaeus, 1758)
Polybius navigator (Herbst, 1794)
Portumnus latipes (Pennant, 1777)
Sirpus ponticus Vereshchaka, 1989
Potamon potamios (Olivier, 1804)
Eriphia verrucosa (Forskål, 1785)
Pilumnus hirtellus (Linnaeus, 1761)
Rhithropanopeus harrisii (Gould, 1841)
Xantho poressa (Olivi, 1792)
Brachynotus sexdentatus (Risso, 1827)
Pachygrapsus marmoratus (Fabricius, 1787)
Planes minutus (Linnaeus, 1758)

MYSIDA

Siriella jaltensis jaltensis Czerniavsky, 1868
Gastrosaccus sanctus (Van Beneden, 1861)
Leptomysis lingvura (G. O. Sars, 1866)
Leptomysis truncata (Heller, 1863)
Acanthomysis strauchi (Czerniavsky, 1882)
Hemimysis anomala G.O. Sars, 1907
Hemimysis lamornae (Couch, 1856)
Hemimysis serrata Bacescu, 1938
Diamysis bahirensis (G.O. Sars, 1877)
Diamysis pengoi (Czerniavsky, 1882)
Diamysis mecznikovi (Czerniavsky, 1882)
Limnomysis benedeni Czerniavsky, 1882
Mesopodopsis slabberi (van Beneden, 1861)
Katamysis warpachowsky G.O. Sars, 1877
Paramysis agigensis Bacescu, 1940
Paramysis arenosa (G.O. Sars, 1877)
Paramysis baeri Czerniavsky, 1882
Paramysis bakuensis G.O. Sars, 1895
Paramysis kessleri (Grimm, 1875)
Paramysis kosswigi Bacescu, 1948
Paramysis kroyeri (Czerniavsky, 1882)
Paramysis lacustris tanaitica Martinov, 1924
Paramysis pontica Bacescu, 1938

Paramysis sowinskii Daneliya, 2002
Paramysis ullskyi (Czerniavsky, 1882)

CUMACEA

Schizorhamphus eudorelloides (G.O. Sars, 1894)
Schizorhamphus scabriusculus (G.O. Sars, 1894)
Volgacuma telmatophora Derzhavin, 1912
Pterocuma pectinatum (Sowinsky, 1893)
Pterocuma rostratum (G.O. Sars, 1894)
Pterocuma sowinskyi (G.O. Sars, 1894)
Pseudocuma (*Pseudocuma*) *ciliatum* G.O. Sars, 1879
Pseudocuma (*Pseudocuma*) *longicorne* (Bate, 1858)
Pseudocuma (*Stenocuma*) *cercarioides* G.O. Sars 1894
Pseudocuma (*Stenocuma*) *laeve* G.O. Sars, 1914
Pseudocuma (*Stenocuma*) *graciloides* G.O. Sars, 1894
Bodotria arenosa mediterranea (Stener, 1938)
Iphinoe maeotica (Sowinsky, 1894)
Iphinoe tenella G.O. Sars, 1878
Iphinoe elisae Bacescu, 1950
Cumopsis goodsir (Van Beneden, 1861)
Nannastacus euxinicus Bacescu, 1951
Cumella (*Cumella*) *limicola* G.O. Sars, 1879
Cumella (*Cumella*) *pygmaea euxinica* Bacescu, 1950
Eudorella truncatula (Bate, 1856)
Leucon (*Epileucon*) *longirostris* G.O. Sars, 1871

TANAIDACEA

Apseudes acutifrons G.O. Sars, 1882
Heterotanaïs oerstedii (Kroyer, 1842)
Leptochelia savignyi (Kroyer, 1842)
Pseudoleptochelia merginellae (Smith, 1906)
Pseudotanaïs borceai Bacescu, 1960
Tanaïs dulongii (Audouin, 1826)

ISOPODA

Limnoria tuberculata Sowinsky, 1884
Eurydice dollfusi Monod, 1930
Eurydice racovitzai Bacescu, 1949
Eurydice pontica (Czerniavsky, 1868)
Eurydice spinigera Hansen, 1890
Eurydice valkanovi Bacescu, 1949
Anilocra physodes (Linnaeus, 1758)
Mothocya taurica (Czerniavsky, 1868)
Cymodoce erythraea euxinica Bacescu, 1958
Cymodoce aff. tattersalli Torelli, 1929
Dynamene bidentatus (Adams, 1800)
Dynamene bicolor (Rathke, 1837)
Exosphaeroma pulchellum Colosi, 1921
Sphaeroma serratum (Fabricius, 1787)

Idotea balthica (Pallas, 1772)
Idotea ostroumovi Sowinsky, 1895
Synisoma capito (Rathke, 1837)
Porcellio lamellatus Budde-Lund, 1885
Tylos europaeus Arcangeli, 1938
Tylos ponticus Grebnitsky, 1874
Jaera hopeana A. Costa, 1853
Jaera nordmanni (Rathke, 1837)
Jaera sarsi Valkanov, 1936
Bopyrina ocellata (Czerniavsky, 1868)
Bopyrissa diogeni (Popov, 1927)
Parathelges racovitzai R. Codreanu, 1940
Progebiophilus euxinicus (Popov, 1929)
Ligia italica Fabricius, 1798
Halophiloscia couchii (Kinahan, 1858)
Halophiloscia pontica Radu, 1985
Elaphognathia bacescoi (Kussakin, 1969)
Gnathia oxyuraea (Lilljeborg, 1855)

AMPHIPODA

Orchomene humilis (A. Costa, 1853)
Nannonyx goesi (Boeck, 1871)
Nannonyx propinquus Chevreux, 1911
Ampelisca diadema (A. Costa, 1853)
Ampelisca pseudospinimana Bellan-Santini & Kaim-Malka, 1977
Bathyporeia guilliamsoniana (Bate, 1857)
Harpinia dellavallei Chevreux, 1910
Stenothoe monoculoides (Montagu, 1815)
Perioculodes longimanus longimanus (Bate & Westwood, 1868)
Synchelidium maculatum Stebbing, 1906
Monoculodes gibbosus Chevreux, 1888
Apherusa bispinosa (Bate, 1857)
Apherusa chiereghinii Giordani-Soika, 1950
Atylus guttatus (A. Costa, 1851)
Atylus massiliensis Bellan-Santini, 1975
Biancolina algicola Della Valle, 1893
Cymadusa crassicornis (A. Costa, 1857)
Amathillina cristata G.O. Sars, 1894
Cardiophilus baeri G.O. Sars, 1896
Dikerogammarus haemobaphes (Eichwald, 1841)
Dikerogammarus villosus (Sowinsky, 1894)
Echinogammarus foxi (Schellenberg, 1928)
Echinogammarus ischnus (Stebbing, 1899)
Echinogammarus olivii (Milne-Edwards, 1830)
Echinogammarus placidus (G.O. Sars, 1896)
Echinogammarus warpachowskyi (G.O. Sars, 1894)
Erichthonius punctatus (Bate, 1857)
Erichthonius difformis Milne-Edwards, 1830
Euxinia maeoticus (Sowinsky 1894)
Euxinia sarsi (Sowinsky, 1898)
Euxinia weidemanni (G.O. Sars, 1896)
Gammarellus angulosus (Rathke, 1843)

Gammarus aequicauda (Martynov, 1931)
Gammarus crinicornis Stock, 1966
Gammarus duebeni Liljeborg, 1852
Gammarus insensibilis Stock, 1966
Gammarus pulex (Linnaeus, 1758)
Gammarus subtypicus Stock, 1966
Gammarus zaddachi Sexton, 1912
Gammarus locusta (Linnaeus, 1758)
Gammarus marinus Leach, 1815
Iphigenella andrussowi (G.O. Sars, 1896)
Iphigenella shablensis (Carausu, 1943)
Megaluropus agilis Hoeck, 1889
Melita palmata (Montagu, 1804)
Obesogammarus crassus (G.O. Sars, 1894)
Obesogammarus obesus (G.O. Sars, 1894)
Pontogammarus robustoides (G.O. Sars, 1894)
Stenogammarus carausui Derzhavin & Pjatakova 1962
Stenogammarus compressus (G.O. Sars, 1894)
Stenogammarus macrurus (G.O. Sars, 1894)
Stenogammarus similis (G.O. Sars, 1894)
Uroniphargoides spinicaudatus (Carausu, 1943)
Yogmelina pusilla (G.O. Sars, 1896)
Dexamine spinosa (Montagu, 1813)
Tritaeta gibbosa (Bate, 1862)
Orchestia cavimana Heller, 1865
Orchestia mediterranea A. Costa, 1853
Orchestia stephenseni Cecchini, 1928
Orchestia gammarellus (Pallas, 1766)
Orchestia montagui Audouin, 1826
Parhyale aquilina (A. Costa, 1857)
Talitrus saltator (Montagu, 1808)
Talorchestia brito Stebbing, 1891
Talorchestia deshayesi (Audouin, 1826)
Platorchestia platensis Kroyer, 1845
Hyale crassipes (Heller, 1866)
Hyale dollfusi Chevreux, 1911
Hyale schmidtii (Heller, 1866)
Hyale perieri (Lucas, 1849)
Hyale pontica Rathke, 1837
Hyale prevosti (Milne-Edwards, 1830)
Microdeutopus algicola Della Valle, 1893
Microdeutopus anomalus (Rathke, 1843)
Microdeutopus damnoniensis (Bate, 1856)
Microdeutopus gryllotalpa A. Costa, 1853
Microdeutopus versiculatus (Bate, 1856)
Microtopus longimanus Chevreux, 1887
Megamphopus cornutus Norman, 1869
Leptocheirus pilosus Zaddach, 1844
Ampithoe gammaroides (Bate, 1856)
Ampithoe helleri Karaman, 1975
Ampithoe ramondi Audouin, 1826
Atylus guttatus (A. Costa, 1851)
Atylus massiliensis Bellan-Santini, 1975

Biancolina algicola Della Valle, 1893
Cymadusa crassicornis (A. Costa, 1857)
Jassa marmorata (Holmes, 1903)
Jassa oia (Bate, 1862)
Corophium acherusicum A. Costa, 1851
Corophium maeoticum Sowinsky, 1898
Corophium nobile G.O. Sars, 1895
Corophium orientale Schellenberg, 1928
Corophium robustum G.O. Sars, 1895
Corophium sowinskyi Martynov, 1924
Corophium bonnellii (Milne-Edwards, 1830)
Corophium crassicorne Bruzelius, 1859
Corophium curvispinum G.O. Sars, 1895
Corophium runcicorne Della Valle, 1893
Erichthonius difformis (Dana, 1855)
Erichthonius punctatus (Bate, 1857)
Siphonocetes dellavallei Stebbing, 1899
Chelura terebrans Philippi, 1839
Caprella acanthifera Leach, 1814
Caprella acanthifera discrepans Carausu, 1941
Caprella liparotensis Haller, 1879
Caprella rapax Mayer, 1890
Caprella danilevskii Czerniavski, 1868
Caprella mitis Mayer, 1890
Pseudoprotella phasma (Montagu, 1804)
Phtisica marina Slabber, 1749

MOLLUSCA

POLYPLACOPHORA

Lepidochitona caprearum Scacchi, 1836
Lepidochitona cinerea Linnaeus, 1767

GASTROPODA

Patella caerulea Linnaeus, 1758
Calliostoma granulatum (Von Born, 1778)
Gibbula adansonii adansonii (Payraudeau, 1826)
Gibbula albida (Gmelin, 1791)
Gibbula divaricata (Linnaeus, 1758)
Tricolia pullus pullus (Linnaeus, 1758)
Bittium reticulatum (da Costa, 1778)
Bittium submamillatum (de Rayneval & Ponzi, 1854)
Cerithiopsis minima (Brusina, 1865)
Cerithiopsis tubercularis (Montagu, 1803)
Marshallora adversa (Montagu, 1803)
Melarhaphe neritoides (Linnaeus, 1758)
Rissoa lilacina Récluz, 1843
Rissoa membranacea (Adams J., 1800)
Rissoa parva (da Costa, 1778)
Rissoa splendida Eichwald, 1830

Caecum trachea (Montagu, 1803)
Setia valvatoidea (Milaschewitsch, 1909)
Rudolphosetia turriculata (Monterosato, 1884)
Hydrobia acuta (Draparnaud, 1805)
Hydrobia ventrosa (Montagu, 1803)
Ventrosia maritima (Milaschewitsch, 1916)
Bella nebula (Montagu, 1803)
Truncatella subcylindrica (Linnaeus, 1767)
Calyptrea chinensis (Linnaeus, 1758)
Epitonium commune (Lamarck, 1822)
Trophonopsis breviatus (Jeffreys, 1882)
Nassarius incrassatus (Ström, 1768)
Nassarius nitidus (Jeffreys, 1867)
Nassarius reticulatus (Linnaeus, 1758)
Cyclope neritea (Linnaeus, 1758)
Rapana venosa (Valenciennes, 1846)
Omalogyra atomus (Philippi, 1841)
Chrysallida brusinae (Cossmann, 1921)
Chrysallida emaciata (Brusina, 1866)
Chrysallida indistincta (Montagu, 1808)
Chrysallida interstincta (Adams J., 1797)
Eulimella aciculata (Philippi, 1836)
Odostomia erjaveciana Brusina, 1869
Odostomia eulimoides Hanley, 1844
Odostomia scalaris MacGillivray, 1843
Odostomia plicata (Montagu, 1803)
Turbonilla delicata (Monterosato, 1874)
Ebala pointeli (de Folin, 1868)
Retusa mammillata (Philippi, 1836)
Retusa piriformis Monterosato, 1878
Retusa truncatula (Bruguière, 1792)
Cylichnina umbilicata (Montagu, 1803)
Corambe obscura (Verrill, 1870)
Tergipes tergipes (Forskål, 1775)
Tenellia adpersa Nordmann, 1845
Embletonia pulchra (Alder & Hancock 1844)
Myosotella myosotis (Draparnaud, 1801)

BIVALVIA

Anadara inaequalis (Bruguière, 1789)
Striarca lactea (Linnaeus, 1758)
Mytilus galloprovincialis Lamarck, 1819
Mytilaster lineatus (Gmelin, 1791)
Modiolus adriaticus (Lamarck, 1819)
Modiolus barbatus (Linnaeus, 1758)
Modiolula phaseolina (Philippi, 1844)
Pinna rudis Linnaeus, 1758
Chlamys flexuosa (Poli, 1795)
Chlamys glabra (Linnaeus, 1758)
Ostrea edulis Linnaeus, 1758
Loripes lacteus (Linnaeus, 1758)
Lucinella divaricata (Linnaeus, 1758)

Kellia suborbicularis (Montagu, 1803)
Mysella bidentata (Montagu, 1803)
Acanthocardia paucicostata (Sowerby G.B. II, 1841)
Acanthocardia tuberculata (Linnaeus, 1758)
Parvicardium exiguum (Gmelin, 1791)
Plagiocardium papillosum (Poli, 1795)
Cerastoderma glaucum (Poiret, 1789)
Macra stultorum (Linnaeus, 1758)
Spisula solida (Linnaeus, 1758)
Spisula subtruncata (da Costa, 1778)
Donacilla cornea (Poli, 1791)
Solen marginatus Pulteney, 1799
Tellina distorta Poli, 1791
Tellina donacina (Linnaeus, 1758)
Tellina fabula Gmelin, 1791
Tellina tenuis da Costa, 1778
Gastrana fragilis (Linnaeus, 1758)
Donax trunculus Linnaeus, 1758
Donax venustus Poli, 1795
Abra alba (Wood W., 1802)
Abra nitida (O.F. Müller, 1776)
Abra ovata (Philippi, 1836)
Abra prismatica (Montagu, 1808)
Venus casina Linnaeus, 1758
Chamelea gallina (Linnaeus, 1758)
Gouldia minima (Montagu, 1803)
Pitar rudis (Poli, 1795)
Paphia aurea (Gmelin, 1791)
Mya arenaria Linnaeus, 1758
Lentidium mediterraneum (O.G. Costa, 1829)
Pholas dactylus Linnaeus, 1758
Barnea candida (Linnaeus, 1758)