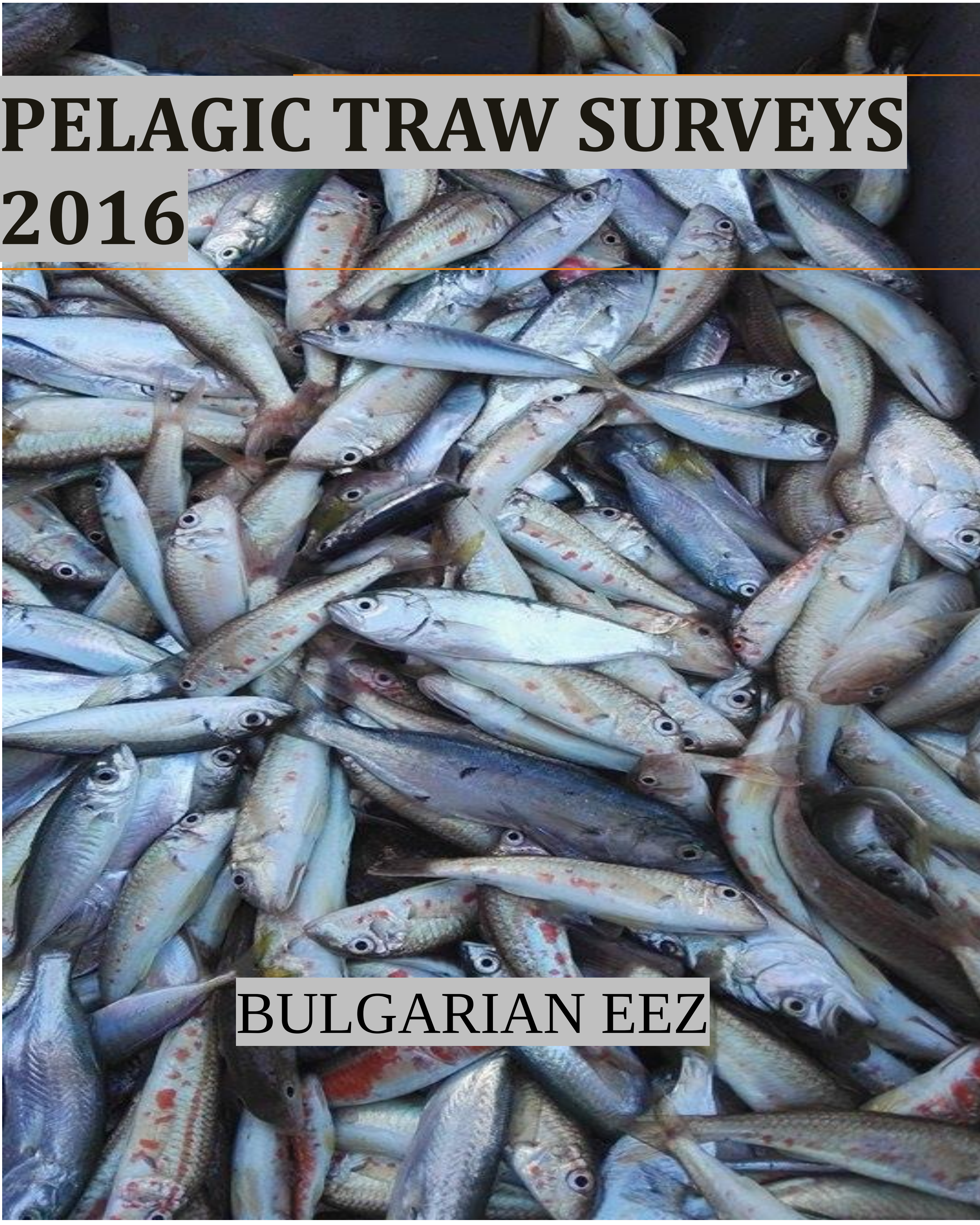
PELAGIC TRAW SURVEYS 2016



BULGARIAN EEZ

The present study was conducted owing to the financial support from the European Commission under Council Regulation (EC) No.199/2008 establishing a community framework for the collection, management and use of data in the fisheries sector and support for scientific advice regarding the common fisheries policy, National Agency for fisheries and Aquaculture – Bulgaria, and:

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The survey was carried out during the period August – September and December 2016 in Bulgarian Black Sea area on board of R/V HAITHABU" in execution of National Programs of Bulgaria for data collection in 2016.

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1 Biological results from International Pelagic Trawl Survey in August – September and December 2016

1.1. Summary

Pelagic Trawl Survey was accomplished in **August – September and December 2016** in the Bulgarian Black Sea area. Scientific team has produced a biological analysis of the results obtained in the marine area.

The analysis is based on the reference species biomass (*Sprattus sprattus*) at the scale of the whole survey. Furthermore, an analysis of the distribution and abundance of the rest species caught as by-catch, is presented. The Black Sea Sprat (*Sprattus sprattus*) is a key species for the Black Sea ecosystem. Together with the anchovy, sprat is one of the most abundant, planktivorous, pelagic species. The level of its stocks depends on the conditions of the environment mainly and on the fishing effort.

The changes in the environment due to anthropogenic influence, affect the dry land as well as the world ocean. The level of the sea pollution and its “self-purifying” ability are completely different. There is a clear indication of changes in the nature equilibrium in the corresponding ecological niches.

The greatest impact in the world ocean has the commercial fishery, which directly devastates a significant part of the given species populations. As a result of this some of the species stocks are declined or depleted.

As a result of the excessive exploitation, altered habitats and climatic variations numerous of the commercial species are critically endangered or vulnerable.

The abundance of the given fish species generations is dependent on different abiotic and biotic factors. With great importance are: the level of fishing mortality, changes in trophic levels due to mass occurrence of the ctenophore *Mnemiopsis leidyi*, algal blooms which lead to hypoxia in the shallower waters with mass mortality of the bottom dwelling organisms and etc.

Recent state of the sprat stock biomass (aggregations) off Bulgarian Black sea coast show relative stability i.e. taking into consideration almost constant level of exploitation (in western and north-western part of the Black Sea) in the last years the stock possibly is underexploited yet.

All analyses are based on the biomass and density estimates and by geographical strata. All the teams calculated their standard statistical estimates using the same software.

This report presents successively the results obtained at these two levels. The regional reports are presented in an order following the coast, from the northern to southern part of the Black Sea. The document is completed by a series of tables and figures related to the biomass/abundance indices and length frequency distributions of the species included in the reference list.

2 Fishing vessel and fishing gear

The Pelagic Trawl survey (PT) was accomplished on board of research vessel “HaitHabu”,. The main characteristics of the ship are given bellow:

Picture 2.1. R/V HaitHabu

R/V HaitHabu

- IMO: 8862686
- MMSI: 207139000
- Позивна: LZHC
- Flag: Bulgaria [BG]
- AIS Vessel Type: Other
- Gross Tonnage: 142
- Length Overall x Breadth Extreme:24.53m × 8m
- Crew: 6

-

The dimensions of the pelagic trawl (Pict. 2.2) employed are as follows:

- type of pelagic trawl 50/35 – 74 m
- Length of the head rope – 40 m.
- Horizontal spread of trawl – 16 m
- Vertical spread of trawl – 7 m
- Mesh size of the net – 7x7 mm.
- Effective part of wing spread – 27 m.
- Pelagic doors – 3.5 m^2.

The hauls were carried out during the day with single haul duration between 30 - 40 min., depending on hydro-meteorological conditions at average trawl speed 2.7 knots (2.7-2.9).

Picture 2.2. Catches in trawl codend

3. Material and Methods

Pelagic Trawl survey was accomplished with accordance with National Programs for Data Collection in Fisheries sector of Bulgaria for 2015. The study held during the period of December 2015, in the area enclosed between Durankulak and Ahtopol (Bulgaria) with total length of coastline of 370 km. Study area encloses waters between 42^{o}05’ and 43^{o}45’ N and 27^{o}55 and 29^{o}55 E.

During the survey, total 36 mid-water hauls were carried out in Bulgarian area (August-September and December 2016). The survey undergoes during the day and the following types of data were collected:

- Coordinates and duration of each trawl
- Sprat total catch weight
- Separation of the by-catch by species
- Composition of by-catch
- Conservation of the samples

3.1 Sampling design

To establish the abundance of the reference species (*Sprattus sprattus*) in front of the Bulgarian coast a standard methodology for stratified sampling was employed (Gulland, 1966;). To address the research objectives the region was divided in four strata according to depth – Stratum 1 (15 - 35 m) Stratum 2 (35 – 50 m), Stratum 3 (50 – 75 m) and Stratum 4 (75 – 100 m).

The study area in Bulgarian waters was partitioned into 128 equal in size not overlying fields, situated at depth between 10 - 100 m. At 36 of the fields chosen at random, sampling by means of mid-water trawling was carried out .

Each field is a rectangle with sides 5' Lat × 5' Long and area around 62.58 km^2 (measured by application of GIS), large enough for a standard lug extent in meridian direction to fit within the field boundaries. The fields are grouped in larger sectors – so called strata, which geographic and depth boundaries are selected according to the density distribution of the species under study. At each of the fields only one haul with duration between 30 - 40 min. at speed 2.7-2.9 knots was carried out.

As a result of the trawling survey a biomass index was calculated.

3.2 Onboard sample processing

The data recorded and samples collected at each haul include:

- Depth, measured by the vessel's echo sounder;
- GPS coordinates of start/end haul points;
- Haul duration;
- Abundance of sprat caught;
- Weight of total sprat catch;
- Abundance and weight of other large species;
- Species composition of by-catch;
- 4% Formaldehyde solution with marine water was used for conservation of sprat for stomach content examination.

3.3 Laboratory analyses

The samples collected onboard were processed in the laboratory for determination of age and food composition.

The age was established in otoliths under binocular microscope.

The food spectrum was determined by separation of the stomach contents into taxonomic groups identified to the lowest possible level.

3.4 Statistical analyses

Swept area method

This method is based on bottom trawling across the seafloor (area swept), weighted with chains, rock-hopper and roller gear, or steel beams. Widely used direct method for demercal species stock assessment.

The main point of the method: the trawl doors are designed to drag along the seafloor for defined distance. Trawling area was calculated as follows:

(1) $a = D * hr * X2$
$D = V * t$

(Where: a – trawling area, V – trawling velocity, hr* X2 – trawl door distance, t – trawling duration (h), D – dragged distance on the seafloor;

(2) $D = 60 * \sqrt{(Lat_1 - Lat_2)^2 + (Lon_2 - Lon_1) * \cos(0.5 * (Lat_1 + Lat_2))}$

(3) $D = \sqrt{VS^2 + CS^2 + 2 * VS * CS * \cos(dirV - dirC)}$,

Where, VS is vessel velocity, CS - present velocity (knots), dir*V* vessel course (degrees) and *dirC*- present course (degrees).

Stock biomass is calculated using catch per unit area, as fraction of catch per unit effort from dragged area:

(4) $(\frac{C_{w/t}}{a/t}) = C_{w/a} kg / sq.km$

Where: *Cw/t* – catch per unit effort, a/t – trawling area (km2) per unit time;

Stock biomass of the given species per each stratum could be calculated as follows:

(5) $B = (\overline{C_{w/a}}) * A$

Where: $\overline{C_{w/a}}$ - mean CPUA for total trawling number in each stratum, A- area of the stratum.

The variance of biomass estimate for each stratum is (equation 4):

(6) $VAR(B) = A^2 * \frac{1}{n} * \frac{1}{n-1} * \sum_{i=1}^{n} \left[Ca(i) - \overline{Ca}\right]^2$

Total area of the investigated region is equal to the sum of areas of each stratum:

A = A1+A2+A3

Average weightened catch per whole aquatic territory is calculated as follows:

(7) $\overline{Ca}(A) = Ca1 * A1 + Ca2 * A2 + Ca3 * A3 / A$

Where: Ca1- catch per unit area in stratum 1, A1 – area of stratum 1, etc., A- size of total area.

Accordingly, total stock biomass for the whole marine area to:

(8) $B = \overline{Ca}(A) * A$

Where: $\overline{Ca}(A)$ - average weightened catch per whole investigated marine area, A – total investigated marine area.

Estimation of Maximum Sustainable Yield (MSY)

The Gulland's formula for virgin stocks is used – equation 7:

(9) MSY = 0.5*M*Bv

where: M – coefficient of natural mortality; Bv – virgin stock biomass.

Relative yield-per-recruit model with uncertainties

(10) $$\mathrm{Y'/R = E * U^{M/k} \left\{ 1 - \frac{3U}{(1+m)} + \frac{3U^2}{(1+2m)} - \frac{U^3}{(1+3m)} \right\}}$$

where: $U = 1-(Lc/L\infty)$

$m = (1-E)/(M/k) = k/Z$

$E = F/Z$ – exploitation coefficient.

Age and growth
Lenght-converted catch curve

A number of methods are available with the help of which total mortality (Z) can be estimated from length-frequency data. Thus it is possible to obtain reasonable estimates of Z from the mean length in a representative sample, or from the slope of Jones' cumulative plot. In this article, a variety of approaches for analysing length-frequency data are presented which represent the functional equivalent of [age structured] catch curves; these "length-converted catch curves" are built around assumptions similar to those involved in age-structured catch curves.

3.5. Age estimation

As it is well known, the Calcified Structures (CS) are usually used to assign age useful to obtain their growth model and so, to reconstruct age composition of exploited fish populations. Fish ageing implies the presences in the CS of a structural pattern, in terms of succession of opaque and translucent zones and the knowledge of the periodicity of this deposition pattern. Calcified structures available for fish ageing are different: otoliths (sagittae, lapilli, asterischi), scales, vertebrae, spines and opercular bones (Panfili et al., 2002). For the selected stocks the CS utilized is the sagittae. The most important aspects (difficulties, extraction, storage, preparation method, ageing criteria) regarding the age analysis are addressed by species.
Otoliths are important for fish and fisheries scientists. Otoliths are playing role in balance, motion and sound. These structures are effective from growth to death in entire life cycle. They are most commonly used for age in order to determine growth and mortality research. Research on otoliths began in 1970s and continues to 21st century. Periodic growth increments which in scales, vertebrae, fin rays, cleithra, opercula and otolith are used to determine annual age in many fish species. Researchers have used otolith reference collections and photographs in publications

to aid in identifications. Otoliths have a distinctive shape which is highly specific, but varies widely among species16. Biologists, taxonomists and archaeologists, based on the shape and size of otoliths determined fish predators feeding habits (Kasapoglu and Duzgunes, 2014). In teleost fishes, otoliths are the main CS for the age determination and it is widely used in fisheries biology. On the other hand analysing O2 isotopes in their structure is useful to determine fish migrations between fresh water and sea as well as species and stock identification.

Otoliths are the balance and hearing organs for the fish. They are in three types located on the left and right side of the head in the semi rings; “sagitta” in the saccular, “lapillus” in the lagenar and “asteriskus” in the utricular channels. Place, size and shape of these three types are different by species, the biggest one is sagitta and the smallest one is asteriscus. So, sagitta is the one mostly used in age determination in bony fishes (Aydın, 2006). Other reasons for the preference to otoliths are;

-Their formation in the embryonic phase which shows all the changes in the life cycle of the fish,

- Existence in the fish which have no scales,
- Giving better results than the scales and more successful age readings in older fish than their scales,
- No resorption or regeneration,
- Having same structure in all the individuals in the same species (Jearld, 1983).

On the other hand, their disadvantages are the obligation of dissecting the fish and some failures in age determination due to crystal like formations by irregular $CaCO_3$ accumulations on the otoliths.

3.5.1 Otolith Preperation for sprat

Sampling of the fish for otolith extraction from the overall samples is very important to have representative samples for the catch. Number of otoliths needed is lower for the species having smaller size range than the species having larger size range. According to the availability 5 fish for each length group may be better for age readings to be representative for the population. Each of the individuals should be recorded individually with place of catch, date and ID number. These steps are useful for the process:

- For each fish total length (±0,1 cm), total weight (±0,01g), sex, maturation stage (I-V), gonad weight (±0,01g) are recorded.
- Sagittal otoliths of each fish are removed by cutting the head over eyes after all individual measurements. Then, rinsed and immersed in 96% ethyl alcohol to get rid of organic wastes/residuals and finally kept in small chambers in plastic roomed boxes with the sample number and other operational information.

3.5.2 Preperation of the otoliths for the age determination

Otoliths are put into small black convex glasses containing 96% ethyl alcohol for age readings under binocular stereo microscope which is illuminated from top and sides (Fig 3) (Polat ve Beamish, 1992). Magnifying level depends the size of the otolith; X4 is good for sprat and X1 for turbot.

Fig 3.5.2. Binoculer stereo microscobe with top and side illumination (Yoraz, 2015)

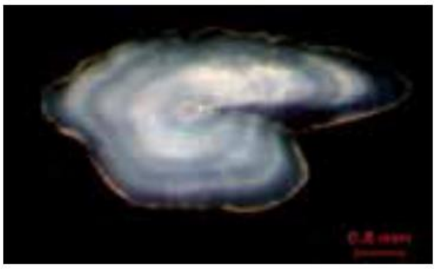
3.5.3. Age readings and commenting on annuluses

In order to prevent bias, during age reading reader should not refer length and weight of that fish. But information on the date of the catch and gonadal state is very important.

First step is to clarify the place of the center and the first age ring. After that, observation of the successive rings, whether they are continuous or not is important. Finally, determination of the fish in growth or just at the end of the growth period by checking characteristics of the ring at the edge of the otolith to decide it is opaque or hyaline. After these procedures otoliths can be read under these protocols which are very important to provide data on age to determine realistic population parameters and reduce uncommon procedures and biases by standardized age reading criteria.

3.5.4. Sprat (*Sprattus sprattus*)

In sprat left and right otoliths shows isometric growth. These are small and transparent (Fig 2). Age readings can be done over otolith surface by clear ring views. Due to summer and winter growths there are two different nucleus formation in the center; spring recruits has opaque, late fall recruits has hyaline rings which is taken into consideration during age readings (Pısıl, 2006).

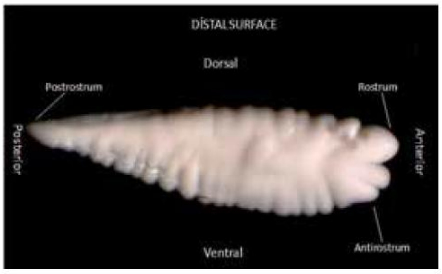


a

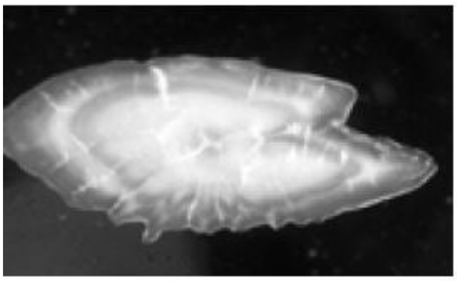
b

TL: *a* – 6.2 cm; *b* – 6.7 cm

S.sprattus



Merlangius merlangus *Trachurus mediterraneus*



E.encrasicolus

M.barbatus

P.salstarix

Figure 3.5.4. Sprat, anchovy, scad, red mullet, bluefish otoliths

3.5.5. Age reading protocol

1. Dissected otoliths rinsed and threated with 96 % ethyl alcohol and stored dry.

2. Readings are carried out by inspecting the whole otolith in 96% ethyl alcohol in black colored convex glass bowl under reflected light against a dark background.

3. Magnification set considering the biggest otolith size which is totally fit the visual capacity of the lens. It is aimed not to change magnification rate which may enable false rings visible in bigger otoliths and permits to see true rings (hiyalins) better by unchanging the color contrasts. Thats why magnifation rate X4 is selected for the sprat otoliths.

4. Otolith samples observed from distal surface as a whole, broken ones are not used.

5. Birthday of the sprat accepted as 1st of January as the common principle for the fish living in the Northern semiphere in line with the sub-tropic fish growth models.

6. Central point surrounded by the hyalin rings which is one in some cases or two for the others, is formed after the end of consumption of yolc sac and starting of the free feeding, and known as "stock rings". Next opaque accumulation is known as "first year growth ring". This ring keeps its circular form in the postrostrum region. Together with this ring and the next hyalin ring forming "V" shape in the rostrum, is accepted as first age rings.

7. Tiny and continious consantric rings prolonge close to real hyalin ringed are counted togetherwith the real one as one age. This ring may be either a very tiny and opaque inside the hyaline band or tiny hyaline ring near the outer edge of the opaque ring.

8. Sprat and some other short lived species has very fast growth rate especially in the first two years. Width of the growth bands after 2nd year ring has relatively getting narrower. This issue should be kept in mind in the older age ring readings.

9. Number of tiny and weak hyaline rings, known as false rings, in the opaque region, is not so high and, their seperation from age rings is rather easy. When they are so much and unseperable, these otoliths should not be used.

3.6 SEX AND MATURITY ESTIMATION

3.6.1 SPRAT

The European sprat (*Sprattus sprattus* L.) is a small short-lived pelagic species from the family Clupeidae. Sprat has a wide distribution including shelf areas of the Northeast Atlantic, the Mediterranean Sea and the Baltic Sea. Sprat is most abundant in relatively shallow waters and tolerates a wide range of salinities. Spawning is pelagic in coastal or offshore waters and occurs over a prolonged period of time that may range from early spring to the late autumn. Sprat is an important forage fish in the North Sea and Baltic Sea ecosystems. Commercial catches from pelagic fisheries are mainly used for fish meal and fish oil production. Three subspecies of sprat have been defined i.e. *Sprattus sprattus sprattus* L., distributed along the coasts of Norway, the North Sea, Irish Sea, Bay of Biscay, the western coast of the Iberian peninsula down to Morocco, *Sprattus sprattus phalericus*, R) in the northern parts of the Mediterranean and the Black Sea, and *Sprattus sprattus balticus* S. in the Baltic Sea. Knowledge about stock structure, migration of sprat and mixing of populations among areas is limited. Questions have been raised about the geographic distribution and separation of stocks and their interaction with neighboring stocks (ICES 2011). The apparent overlap e.g. between North Sea sprat and English Channel sprat seems very strong, whereas the overlap between North Sea sprat and Kattegat sprat is not as strong and varies between years. A distribution wide phylo-geographic study showed that sprat in the Western Mediterranean is a subgroup of the Atlantic group and that these two populations are closer to each other than to sprat in the Eastern Mediterranean and Black Sea (Debes et al., 2008).

3.6.2. Maturity Stages of Sprat

It is very important to use standardized maturity scales for sprat (and all species) to evaluate sampling strategies and timing for accurate classification of maturity in order to provide reliable maturity determination for both sexes. For sprat, small gonad size and the batch spawnings by several cohorts of eggs over a long period of time are the main challenges for standardizing a maturity scale.

According to the ICES (2011), present standardized maturity scales of sprat include 6-stages for both sexes (Fig3.6.2.Table 3.6.2.)

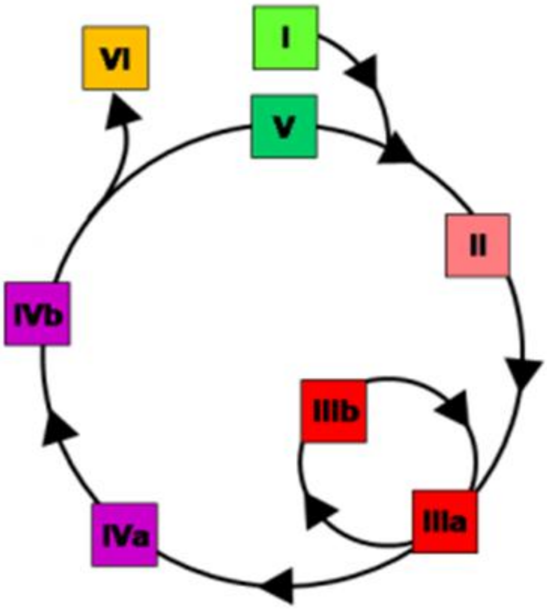


Figure3.6.2. Scale with six maturity stages in sprat (Name of the stages are given in Table 3.6.3)

In particular, specimens without visible development have been combined into Immature and Preparation, whereas the spawning stage has been sub-divided into a non-active spawning stage (maturing and re-maturing characterized by visible development of gametes) and an active spawning stage indicated by hydrated eggs/running milt. The integration of maturing and re-maturing into the spawning stage allows an accurate determination of maturing and spawning specimens and reliable assessment of the spawning fraction of the population.

Table 3.6.3. Macroscopic and histological characteristics of gonadal development stages
C

Stages	*Macroscopic characteristics*	*Histological characteristics*
FEMALES (OG: Oogonia, PG1: Early previtellogenic oocytes, PG2: Late previtellogenic oocytes, CA: Cortil alveoli oocytes, VT1: Early vitellogenic oocytes, VT2: Mid vitellogenic oocytes, VT3: Late vitellogenic oocytes, HYD: Hydrated oocytes, POF: Postovulatory		

follicles, SSB: Spawning stock biomass).		
I-Immature	*Juvenile: ovaries threadlike and small; transparent to wine red and translucent in color; sex difficult to determine; distinguishable from testes by a more tubular shape; oocytes not visible to the naked eye*	*OG+/-PGI*
II-Preparation	*Transition from immature to early maturing; oocytes not visible to the naked eye; ovaries yellow-orange to bright red in color; ovaries occupy up to half of the abdominal cavity. This stage is not included in SSB.*	*PG1, PG2, CA*
III. Spawning *a. Spawning(inactive)* *b. Spawning (active)*	 *Maturing and re-maturing: yolked opaque oocytes visible to the naked eye; ovaries change from semi-transparent to opaque yellow-orange or reddish in color as more oocytes enter the yolk stage; ovaries occupy at least half of the body cavity; re-maturing ovaries may be red to grey-red or purple in color and less firm than an ovary maturing first batch, few hydrated oocytes may be left* *Spawning active. Hydrated eggs are visible among yolked opaque oocytes; hydrates oocytes may be running; ovaries fill the body cavity; overall color varies from yellowish to reddish.*	 *PG1, PG2, CA, VT1, VT2, VT3, +/- POF* *PG1, PG2, CA, VT1,VT2, VT3, HYD, POF*
IV.a Cessation IV.b. Recovery	*Baggy appearance; bloodshot; grey-red translucent in color; atretic oocytes appear as opaque irregular grains; few residual eggs may remain*	PG1, PG2, POF, atretic oocytes, residual HYD PG1, PG2, atretic

	Ovaries appear firmer and membranes thicker than in sub-stage IV.a; these characteristics together with the slightly larger size distinguish this stage from the virgin stage; ovaries appear empty and there are no residual eggs; transparent to wine red translucent in color	VT oocytes
V. Resting	*Ovaries appear more tubular and firmer; oocytes not visible to the naked eye; transparent or grey-white to wine red in color with well-developed blood supply; this stage leads to stage II.*	PG1, PG2 +/- atretic oocytes
VI. Abnormal	*a) infection; b) intersex - both female and male tissues can be recognized; c) one lobe degenerated; d) stone roe (filled with connective tissue); e) other*	Abnormal tissue
MALES (SG: Spermatogonia; PS: Primary spermatocytes; SS: Secondary spermatocytes; ST: Spermatids; SZ: Spermatozoa; SSB: Spawning stock biomass)		
I. Immature	*Juvenile: Testes threadlike and small; white-grey to grey brown in color; difficult to determine sex, but distinguishable from ovaries by a more lanceolate shape (knife shaped edge of distal part of the lobe).*	SG, PS
II-Preparation	*Transition from immature to maturing: Testes easily distinguishable from ovaries by lanceolate shape; sperm development not clearly visible; reddish grey to creamy translucent in color; testes occupy up to ½ of the abdominal cavity; this stage is not included in SSB.*	SG, PS, SS, potentially few ST
III. Spawning a. Spawning(inactive)	*Maturing and re-maturing: Testes occupy at least half of the body cavity and grow to almost the length of the body cavity; the empty sperm duct may be visible; color varies from reddish light*	*SG, PS, SS, ST, SZ*

c. Spawning (active)	*grey, creamy to white; edges may still be translucent at the beginning of the stage, otherwise opaque; re-maturing testes may be irregularly colored with reddish or brownish blotches and grey at the lower edge with partly whitish remains of sperm* *Spawning active: testes fill the body cavity; Sperm duct filled and distended throughout the entire length; sperm runs freely or will run from the sperm duct, if transected; color varies from light grey to white..*	*SG, PS, SS, ST, SZ*
IV.a Cessation IV.b. Recovery	*Baggy appearance (like an empty bag when cut open); bloodshot; grey to reddish brown translucent in color; residual sperm may be visible in sperm duct.* *Testes appear firmer and the testes membrane appear thicker than in stage IVa due to contraction of the testes membrane; these characteristics together with the slightly larger size distinguish this stage from the virgin stage; testes appear empty and no residual sperm is visible in the sperm duct; reddish grey to greyish translucent in color.*	*SG, PS, atretic SS, ST and SZ* *SG, PS, potentially SS, atretic SZ*
V. Resting	*Testes appear firmer, development of a new line of germ cells; grey in color; this stage leads to stage II.*	*SG, PS, SS*
VI. Abnormal	*a) infection; b) intersex - both female and male tissues can be recognized; c) one lobe degenerated; d) other.*	*e.g. oocytes visible among spermatogenic tissues*

Batch fecundity

All fish were measured to the nearest 1 mm in the Total Length (TL) and weighted to the nearest 1 g. Gonads of the fish were examined under a dissecting microscope for its external features such as turgidity and colour in order to determine a maturity stage. The sex ratio also calculated in this study (i.e., No. of males/No. of females (Simon et al., 2012). The female was determined by the macroscopic observation of matured ovary (Laevastu, 1965a).

Batch fecundity can vary considerably during the short spawning season, low at the beginning, peaking during high spawning season and declining again towards the end.

Annual egg production is the product of the number of batches spawned per year and the average number of eggs spawned per batch.
Batch fee undi t Y. 0 f s prat was '. determined as'Hydrated Oocyte Method'. (HUNTER et ale 1985). Oilly hydrated females.were used. After sampling their body cavity was opened and they were 'preserved in a buffered formalin solution
(HUNTER 1985). The ovary free female weight ind the ovary"weight were determined: Three tissue samples of - 50 mg were removed from different parts of the ovary and their exact were determined. Under binocuklar number of hydrated oocytes, in each of the three subsamples was determined.

Hydrated oocytes can easily be separated from all other types of oocytes because of their large size t their translucent appearance and their wrinkled surface which is due to formalin preservation. Batch fecundity 'was estimated 'based, on· the average number of hydrated oocytes per unit weight of the three subsamples.

Gonadosomatic Index (GSI) was determined monthly. GSI was calculated as:

$$GSI = \frac{GW}{SW} X\, 100$$

where, GW is gonads weight and SW is somatic weight (represents the BW without GW)

For the estimation of sprat growth rate, the von Bertalanffy growth function (1938) is used, (according to Sparre, Venema, 1998):

(11) $L_t = L_\infty \left\{1 - \exp\left[-k\left(t - t_0\right)\right]\right\}$

(12) $W_t = W_\infty \left\{1 - \exp\left[-k\left(t - t_0\right)\right]\right\}^n$

where: Lt, Wt are the length or weight of the fish at age t years; L∞, W∞ - asymptotic length or weight, k – curvature parameter, to - the initial condition parameter.

The length – weight relationship is obtained by the following equation:

(13) $W_t = qL_t^{\,n}$

where: q – condition factor, constant in length-weight relationship; n – constant in length-weight relationship.

Coefficient of natural mortality (M)

Pauly's empirical formula (1979, 1980) is applied:

(14) $\log M = -0.0066 - 0.279 * \log L_\infty + 0.6543 * \log k + 0.4634 * \log T^\circ C$

(15) $\log M = -0{\cdot}2107 - 0{\cdot}0824 \log W_\infty + 0{\cdot}6757 \log K + 0{\cdot}4627 \log T°C$

where: L∞, W∞ and к – parameters in von Bertalanffy growth function; T^oC - average annual temperature of water, ambient of the investigated species.

Food composition

Zooplankton collection, preservation and laboratory analysis

Samples were collected by vertical plankton Juday net, 0.1 m^2 mouth opening area, 200 μm mesh size, from 2 meters above the bottom to the surface at integrated sampling layer. Total of 26 zooplankton samples were collected from 26 stations during the cruise. Before sample preservation, the gelatinous species (*Aurelia aurita, Pleirobrachia pileus, Mnemiopsis leidyi*) were removed, rinsed, measured and counted on board. The samples were preserved in 4% buffered to pH 8-8.2 with disodiumtetraborate (borax) ($Na_2B_40_3{\cdot}10H_20$) formalin solution (Korshenko&Aleksandrov, 2006).

Species were identified according to Morduhay-Boltovskoy (1968, 1969, and 1972). Species quantity was determined by the method of Dimov (1959). Biomass was estimated using species individual weight by Petipa, 1959.

To estimate the importance of each food item among the forage, an Index of Relative Importance IRI (Pinkas et al., 1971) was calculated as follows– equation 14:

(16) *IRI = (CN + CW)*F*

where: CN – is the percentage of food item i in total number; CW - is percentage of food item i in total weight; F – is percentage of occurrence frequency in the food item i.

To estimate the importance of each food item among the stomach contents, IRI expressed on a percent basis (Cortes, 1997) was also calculated:

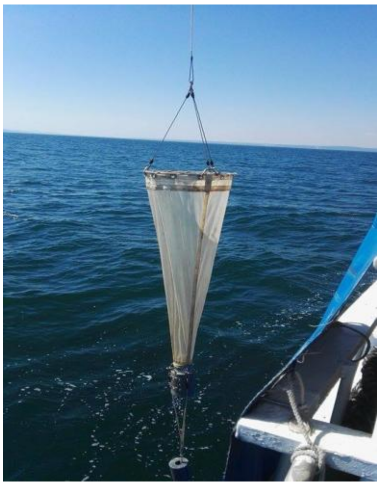
$$\%IRI_i = \frac{100 * IRI_i}{\sum_i^n IRI_i}$$

(17) where: n – is the total number of food categories considered at a given taxonomic level.

Zooplankton collection, preservation and laboratory analysis

Total of 11 zooplankton samples were collected from 11 monitoring stations. Samples were collected by vertical plankton Juday net, 0.1 m2 mouth opening area, 150 μm mesh size, from 2 meters above the bottom to the surface at integral sampling layers. Before sample preservation, the gelatinous species (*Aurelia aurita, Pleirobrachia pileus, Mnemiopsis leidyi, Beroe ovata*) were removed, rinsed, measured and counted on board. The samples were preserved in 4% buffered to pH 8-8.2 with disodiumtetraborate (borax) (Na2B4O3 •10 H2O) formalin solution (Korshenko&Aleksandrov, 2012).

Species were identified according to Morduhay-Boltovskoy (1968, 1969, and 1972) manuals. Species quantity was determined by the method of Dimov (1959). Biomass was estimated using species individual weights by Petipa, 1959.



Sprat feeding

The study of sprat diet was based on stomach content analysis of 132 fish individuals collected in front of Bulgarian Black Sea coast in the period 03.12 – 19.12.2015. The coordinates of the zooplankton sampling sites and sites of sprat diet were presented at Table 1. Two of stations were located at coastal area (st. 6 and 7) and shelf (30-200m).

After the each trawl catch about 20-25 sprat individuals were separated and immediately preserved in 10 % formaldehyde seawater solution. The total length (TL, to the nearest 0.1 cm) and weight (to the nearest 0.01 g) of 12 randomly selected sprat specimens from each sample were measured in laboratory conditions. Each stomach of selected animals was weighted with analytical balance (with 0.0001 g precision) after its removal. The food mass of each individual has been calculated as a difference between the weights of full and empty sprat stomach.

Zooplankton species composition of the sprat food and prey number of the stomach content was investigated under binocular microscope “Olympus”. The biomass of prey was estimated by multiplication of the registered number of consumed mesozooplankton species by their individual weights (Petipa, 1959).

The following indices were calculated: stomach fullness index (SFI) as a percent of body mass: (stomach content mass/fish mass)*100; and index of relative importance - IRI, Pinkas et al. (1971): IRI = (N+M) x FO; where N - the proportion of prey taxa (species) in the diet by numbers (abundance); M - the proportion of prey taxa (species) in the diet by mass; FO- frequency of occurrence among fish.

4. Results

4.1 Abundance index and biomass

Introduction

Total 36 hauls in the Bulgarian marine area were carried out on board of R/V "HaitHabu" Total number of identified species was 16, from which 12 fish, 1 crustacean, 2 mollusc, 1 macrozooplankton species. The most frequent species in the total of hauls (in terms of presence/absence) were (in decreasing order): in August –September 2016: *Tr.mediterraneus (52,3%), M.barbatus (25,05%), E.encarsicolus (10,43%) P.saltatrix (12, 22%).The rest of the species such as S.sprattus*, *M.merlangius , A. immaculata, N.melanostomus, G.niger, etc.* have negligible presence in the catches in August-September 2016. In December the bulk in catches was of sprat and whiting (68%) as the others presented with lower percentages.

In the Bulgarian Black Sea area were realized 36 hauling in the period 22.08-05.09.2016 and 36 hauling in the period of 30/11 – 29/12/2016. The towing time was of 30 and 40 minutes, on depths between 18.9 m and 85.6 m, being surveyed area between Ahtopol and Durankulak.

The surveyed area in Bulgarian waters was about 8135.40 km^{-2}. The sprat was observed on depths after 18 m. The densest sprat aggregations were detected in the deepest stratum 75-100 m with average 3832.02 $kg.km-^{2}$ and with average from all strata of 2192.44$kg.km^{-2}$. In December 4637.265 $kg.km^{-2}$ (50-75m) and 4483.566 $kg.km^{-2}$ (75-100m)

Global comments for Fish and other species

Fish

Sprattus sprattus L.

Small pelagic species, inhabiting the continental shelf area up to 100 – 120 m.

In August 2016 survey the highest biomass indices were established in stratum localized far from the shore 75-100 m in Bulgarian marine area – 9 592 t. In

December the bulk of biomass was in 50-75m – 12 769 t, followed by 75-100m depths – 11 223 t.

The size composition ranged from 6 to 11.75 cm, the age ranged from 0+ to 4-4+, as oldest age groups and young-of-the-year was presented with low percentage.

Whiting (*Merlangius merlangus*)

Whiting inhabits layer near the sea bottom and feed on predominantly with sprat. The species is predator on sprat and is important food component for top predators as turbot and dolphins. Whiting in August was not usual in the catches. This could be related with relatively high temperatures of SST and bottom temperatures. The catch amounts of this species was occasional, only few specimen. In December the catch was a bit higher than in August-September but not enough to make assessments on biomass.

Other species

Anchovy (*E.encrasicolus* L)

The species is migratory and pelagic. In– December, species not suppose to migrate in front of the Bulgarian for spawning but all observed individuals were with high level of maturation. In August the highest biomass index was detected in stratum 50-75 m - 260.95t, In December the highest biomass index was in startum 75-100m and equals to 776 t. The rest of the statums biomass average index varied between 430 – 560 t.

Scad (*Tr.mediterraneus*)

Migratory, small pelagic species, occurring in North-western part of the Black Sea in the time of spawning. In August- September the biomass indeces in 50-75m and 75-100m depths varied between 506-548 t. In 15-30m and 35-50m strata the indices were between 255 and 373 t. In December the highest biomass index was detected in 50-75 m depth polygon, 1 744 t, namely. The indices in 75-100m and in 15-30 m were also higer tha those in August –September 2016: 681(75-100m) and 886t (15-30m).

Red mullet (*Mullus barbatus*)

Benthic, commercially important species, inhabiting coastal zone. In August at depths 15-30m the average biomass index was 898 t. In the rest of strata the index was lower. In December at depths 15-30m the biomass was 952 t., followed by 820 t at 35-50m; 769t at 50-75m and 493t at 75-100m depths.

Bluefish (*Pomatomus satatrix*)

Pelagic, high migratory species, top-predator in Black Sea. The August September survey show that the biomass indices in 30-50m and 50-75m varied between 505 and 548t. The rest indices were lower in the study area. In December 2016, two strata 15-30m and 75-100 encountered high biomass indices: 878t and 844t. The highest average biomass index was detected in 50-75m – 2 141 t.

Round goby (*N.melanostomus*)

The species is benthic dwelling, with relatively good share in mid-water trawls. Only few specimen were detected in 5 hauls in August-September and in 3 hauls in December.

Shad (*Alosa immaculata*)

The species is rare in the catches from present surveys. Only separate specimen have been caught in the codend.

Comments about *Sparttus sprattus* **biomass by stratum in the Bulgarian area.**

Table 4.1.1. Swept area method from Bulgarian marine area, August-September 2016

CPUA sredno		B (kg)	Ax	№ Fields
1471.931	15-30	3039.744	2065.14	33
1519.404	30-50	2757.445	1814.82	29
2070.384	50-75	5700.844	2753.52	44
3832.021	75-100	9592.315	2503.2	40
		21.09035	9136.68	146

A.

CPUA sredno		B (kg)	Ax	№ Fields
1851.279	15-30	3823.15	2065.14	33
2460.126	30-50	4464.686	1814.82	29
4637.265	50-75	12768.8	2753.52	44
4483.566	75-100	11223.26	2503.2	40
		32279.9	9136.68	146

B.

The total surveyed area in Bulgarian part was 9136.7 km^{-2} and total estimated biomass was 21 090.35 t in August-September 2016 (*Table* 4.1.1.A). The total surveyed area in Bulgarian part in December was 9136.7 km^{-2} and total estimated biomass was 32 279.9 t (*Table* 4.1.1.B.)

Table. 4.1.2. A, B Descriptive statistics of Sprat Biomass indices, August-September 2016

Descriptive st. biomass indices sprat,August	
Mean	137203.085
Standard Error	13304.37699
Median	124464.0528
Mode	124464.0528
Standard Deviation	79826.26195
Sample Variance	6372232097
Kurtosis	- 0.274665732
Skewness	0.701269528
Range	305892.8444
Minimum	21849.48889
Maximum	327742.3333
Sum	4939311.065
Count	36
Confidence Level(95.0%)	27009.32104

A.

Descr.st.Sprat ,Dec 2016	
Mean	3456.629571
Standard Error	373.8797664
Median	2982.39449

Mode	3072.165562
Standard Deviation	2243.278598
Sample Variance	5032298.87
Kurtosis	2.773134304
Skewness	1.583472433
Range	10110.21758
Minimum	1061.293558
Maximum	11171.51113
Sum	124438.6646
Count	36
Confidence Level(95.0%)	759.0162732

B.

Comments about *E.encarsicolus* biomass by stratum in the Bulgarian area.

The total biomass in August-September of Anchovy was 722.5 t, and in December 2217 t in Bulgarian Black Sea area.

Table. 4.1.3.A,B Anchovy. Swept area in August-September and in December 2016

CPUA sredno		B (kg)	Ax	№ Fields
64.274	15-30	132.7348084	2065.14	33
75.19	30-50	136.4563158	1814.82	29
94.77	50-75	260.9510904	2753.52	44
76.84	75-100	192.345888	2503.2	40
		0.722488103	9136.68	146

A.

CPUA sredno		B (kg)	Ax	№ Fields
215.45	15-30	444.9344	2065.14	33
311.74	30-50	565.752	1814.82	29
156.12	50-75	429.8795	2753.52	44
310	75-100	775.992	2503.2	40
		2.216558	9136.68	146

B.

Table. 4.1.2. Descriptive statistics of Biomass (t) of sprat

Table. 4.1.4.A,B Anchovy. Descriptive statistics of biomass indices in August-September and in December 2016

Descr.st.Anchovy,bimass ind, August,16	
Mean	4757.899614
Standard Error	984.5074934
Median	2184.948889
Mode	0
Standard Deviation	5907.04496
Sample Variance	34893180.16
Kurtosis	0.864455451
Skewness	1.156802097
Range	22629.82778
Minimum	0
Maximum	22629.82778
Sum	171284.3861
Count	36
Confidence Level(95.0%)	1998.656455

A.

Desc.st, Anchovy,Biomass ind,December,16	
Mean	4757.899614
Standard Error	984.5074934
Median	2184.948889
Mode	0
Standard Deviation	5907.04496
Sample Variance	34893180.16
Kurtosis	0.864455451
Skewness	1.156802097
Range	22629.82778
Minimum	0
Maximum	22629.82778
Sum	171284.3861
Count	36
Confidence Level(95.0%)	1998.656455

B.

Comments about *Trachurus mediterraneus* biomass by stratum in the Bulgarian area.

The total biomass in August-September of Scad was 1682 t, and in December 4159 t in Bulgarian Black Sea area.

Table. 4.1.4. A, B Scad. Swept area in August-September and in December 2016

CPUA sredno		B (kg)	Ax	№ Fields
123.4477	15-30	254.9368	2065.14	33
278.3184	30-50	505.0978	1814.82	29
199.0579	50-75	548.1099	2753.52	44
149.1659	75-100	373.3921	2503.2	40
		1.681537	9136.68	146

A.

CPUA sredno		B (kg)	Ax	№ Fields
410.25	15-30	847.2237	2065.14	33
488.7	30-50	886.9025	1814.82	29
633.2	50-75	1743.529	2753.52	44
272	75-100	680.8704	2503.2	40
		4158.525	9136.68	146

B.

Table. 4.1.4.A,B Scad. Descriptive statistics of biomass indices August-September and in December 2016

Desc.st. Scad,August, 2016	
Mean	12220.97404
Standard Error	2726.56436
Median	2184.948889
Mode	0
Standard Deviation	16359.38616
Sample Variance	267629515.5
Kurtosis	0.057005129
Skewness	1.148388677
Range	49785.62111
Minimum	0
Maximum	49785.62111
Sum	439955.0656
Count	36
Confidence Level(95.0%)	5535.219889

A.

Desc.st, Scad, Biomass Dec,2016

Mean	28978.75168
Standard Error	5566.724875
Median	22629.82778
Mode	0
Standard Deviation	33400.34925
Sample Variance	1115583330
Kurtosis	6.904400676
Skewness	2.32631612
Range	158408.7944
Minimum	0
Maximum	158408.7944
Sum	1043235.061
Count	36
Confidence Level(95.0%)	11301.05223

B.

Comments about *Mullus barbatus* biomass by stratum in the Bulgarian area.

The total biomass in August-September of Scad was 1967.7 t, and in December 3035.2 t in Bulgarian Black Sea area.

Table. 4.1.5. A, B Red mullet. Swept area in August-September and in December 2016

CPUA sredno		B (kg)	Ax	№ Fields
435.03	15-30	898.3979	2065.14	33
200.42	30-50	363.7262	1814.82	29
170.7	50-75	470.0259	2753.52	44
93.84	75-100	234.9003	2503.2	40
		1.96705	9136.68	146

A.

CPUA sredno		B (kg)	Ax	№ Fields
461.12	15-30	952.2774	2065.14	33
452.2	30-50	820.6616	1814.82	29
279.32	50-75	769.1132	2753.52	44
197.02	75-100	493.1805	2503.2	40
		3.035233	9136.68	146

B.

Table. 4.1.4.A,B Red mullet. Descriptive statistics of biomass indices August-September and in December 2016

Descriptive st, red mullet,August biomass, 2016	
Mean	13857.5181
Standard Error	2452.956019
Median	11314.91389
Mode	0
Standard Deviation	14717.73611
Sample Variance	216611756.2
Kurtosis	0.914650406
Skewness	1.238460765
Range	49785.62111
Minimum	0
Maximum	49785.62111
Sum	498870.6517
Count	36
Confidence Level(95.0%)	4979.765429

A.

.

Desc.st, red mullet,biomass ind,Dec 2016	
Mean	22801.06881
Standard Error	4372.428302
Median	19664.54
Mode	0
Standard Deviation	26234.56981
Sample Variance	688252653.2
Kurtosis	-0.167098535
Skewness	0.917369086
Range	87397.95555
Minimum	0
Maximum	87397.95555
Sum	820838.4772
Count	36
Confidence Level(95.0%)	8876.501305

B.

Comments about *P.saltatrix* biomass by stratum in the Bulgarian area.

The total biomass in August-September of Scad was 1682 t, and in December 4159 t in Bulgarian Black Sea area.

Table. 4.1.6. A, B Bluefish. Swept area in August-September and in December 2016

CPUA sredno		B (kg)	Ax	№ Fields
123.4477	15-30	254.9368	2065.14	33
278.3184	30-50	505.0978	1814.82	29
199.0579	50-75	548.1099	2753.52	44
149.1659	75-100	373.3921	2503.2	40
		1.681537	9136.68	146

A.

CPUA sredno		B (kg)	Ax	№ Fields
425.37	15-30	878.4486	2065.14	33
367.6	30-50	667.1278	1814.82	29
777.63	50-75	2141.22	2753.52	44
337	75-100	843.5784	2503.2	40
		4.530375	9136.68	146

B.

Table. 4.1.7.A,B Bluefish. Descriptive statistics of biomass indices August-September and in December 2016

Des.st, Bluefish, biomass August, 2016	
Mean	17527.27849
Standard Error	3368.552937
Median	10924.74444
Mode	0
Standard Deviation	20211.31762
Sample Variance	408497360
Kurtosis	0.910512943
Skewness	1.234151786
Range	76473.21111
Minimum	0
Maximum	76473.21111

Sum	630982.0256
Count	36
Confidence Level(95.0%)	6838.525981

A.

Desc.st, Bluefish Biomass, Dec,2016	
Mean	28978.75168
Standard Error	5566.724875
Median	22629.82778
Mode	0
Standard Deviation	33400.34925
Sample Variance	1115583330
Kurtosis	6.904400676
Skewness	2.32631612
Range	158408.7944
Minimum	0
Maximum	158408.7944
Sum	1043235.061
Count	36
Confidence Level(95.0%)	11301.05223

B.

Figure 4.1.1. Biomass index (t.km^{-2}) in Bulgarian marine area

4.2. Catch per unit area

The calculated catches per unit area (CPUA) for the Bulgarian Black Sea area by strata are represented on Fig. 4.2.1.

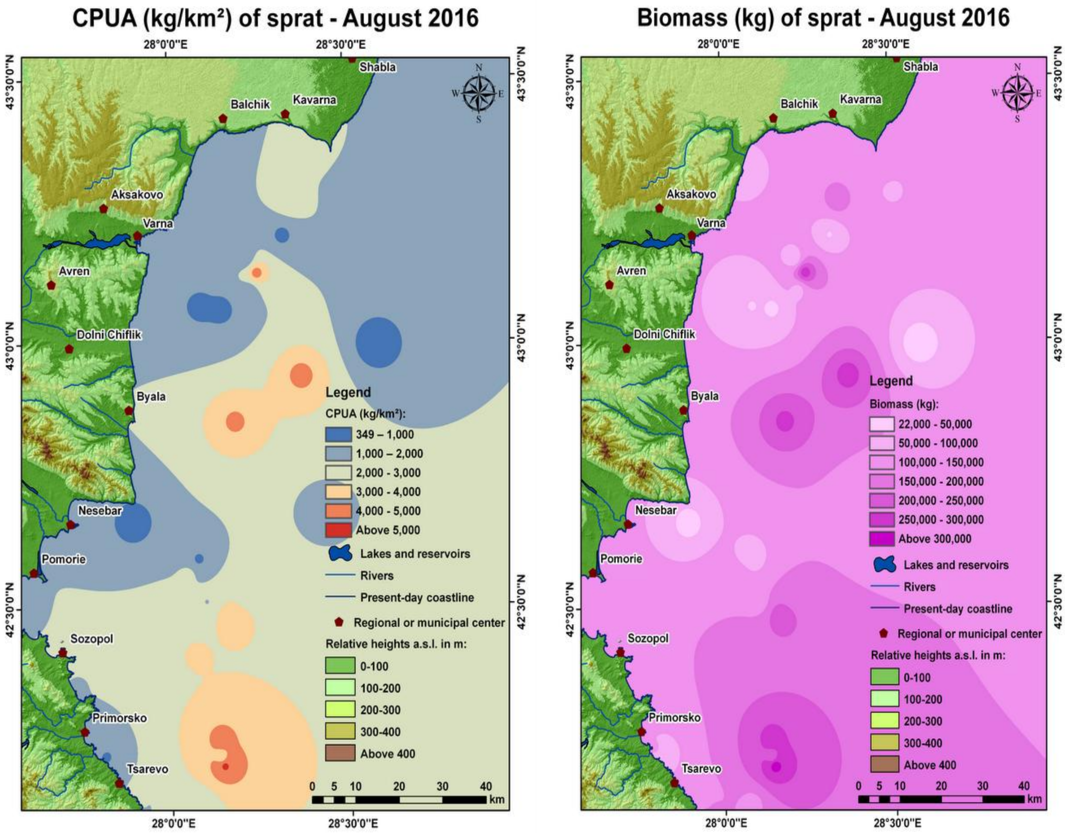


*Figure 4.2.1.*CPUA kg.km^{-2} and Biomass of sprat August-September 2016 from surveyed area.

Sprat, August-September, CPUA kg.km^{-2} -

In all strata CPUA kg.km^{-2}: the lowest value was 349 and the highest one was 5237 kg.km^{-2} detected in southern part of the coast. The densest agglomerations was detected in front of Biala at depths 50-75 and 75-100m and in front of Tzarevo (above 5000 kg.km^{-2}).

Sprat, December, CPUA kg.km^{-2}

The highest concentrations of sprat schools were detected close to the shore in front of Kranevo, River Batova mouth over 11 000 kg.km^{-2}). At depths of 50 m in front of Kaliakra, at depths of 82m in front of Varna and at similar depths in front of Burgas Bay were the rest of polygons with high agglomerations of sprat schools.

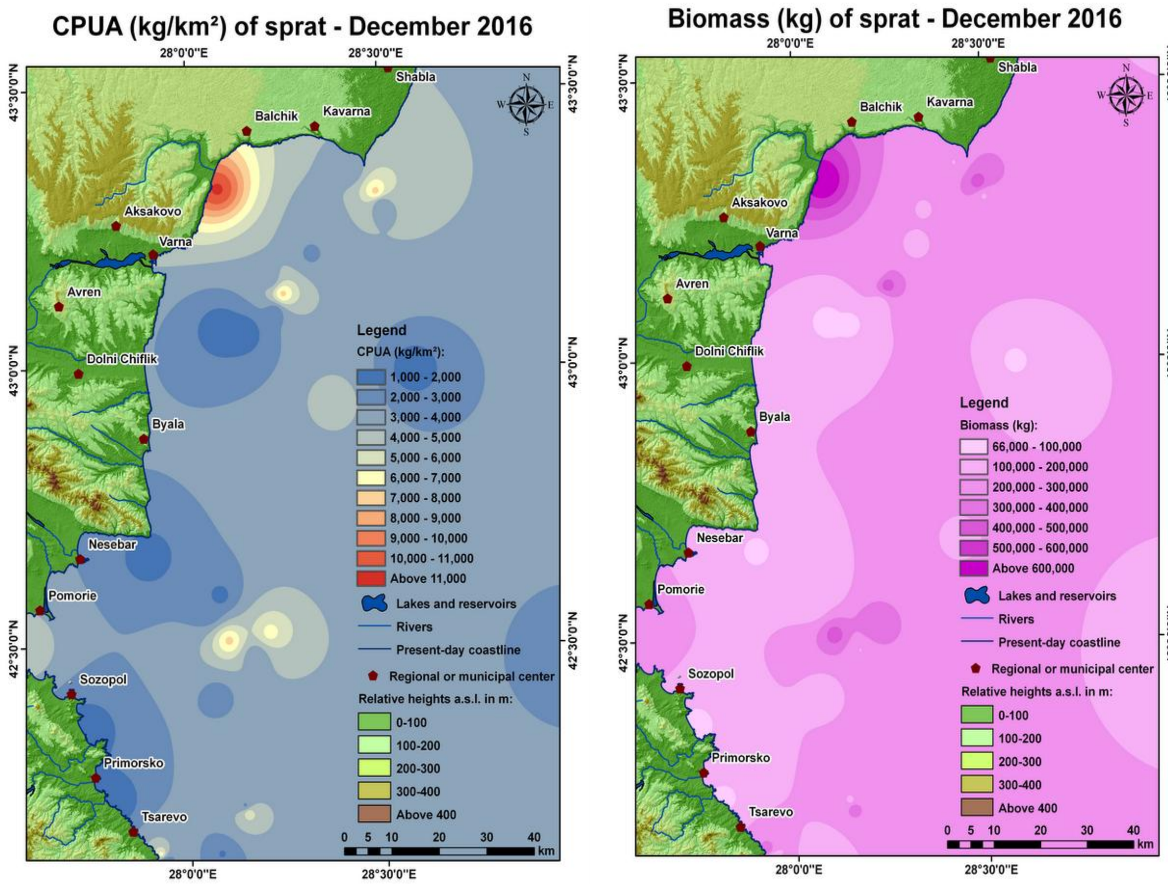


*Figure 4.2.2.*CPUA kg.km^{-2} and Biomass of sprat December 2016 from surveyed area.

Anchovy, August CPUA kg.km^{-2} and Biomass (t)

North-Western direction from Sozopol, at depths over 70m, were detected the highest concentrations of anchovy schools 9above 350 kg.km^{-1}. In December, In front of Kaliakra, Varna, North-west from Biala at depths around 90 m were detected the highest agglomerations of anchovy (over 600kg.km^{-2}). In shallower water (30m) - in Burgas Bay, at 30-50m depths in front of Burgas Bay, Pomorie and Primorsko also were observed dense concentrations f anchovy in December 2016.

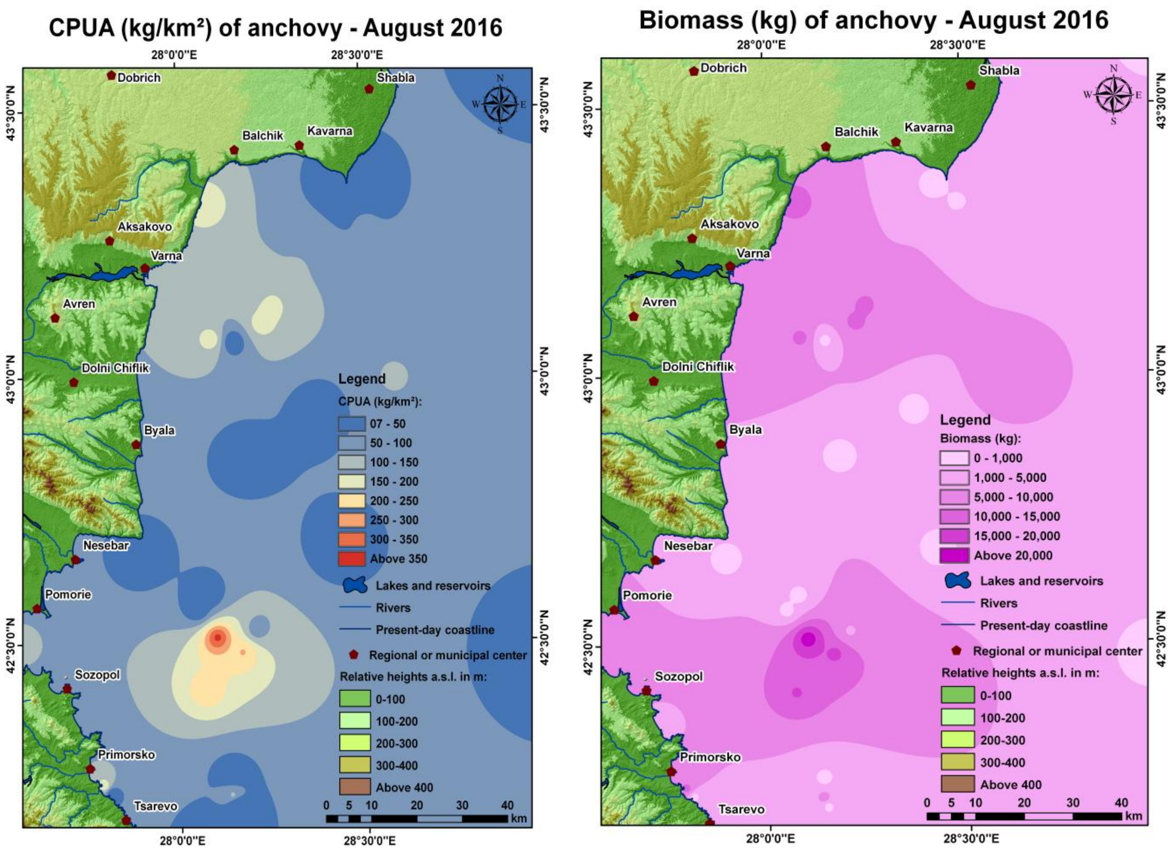


Figure 4.2.3.A, B CPUA kg.km^{-2} and Biomass of anchovy in August –September 2016 from surveyed area.

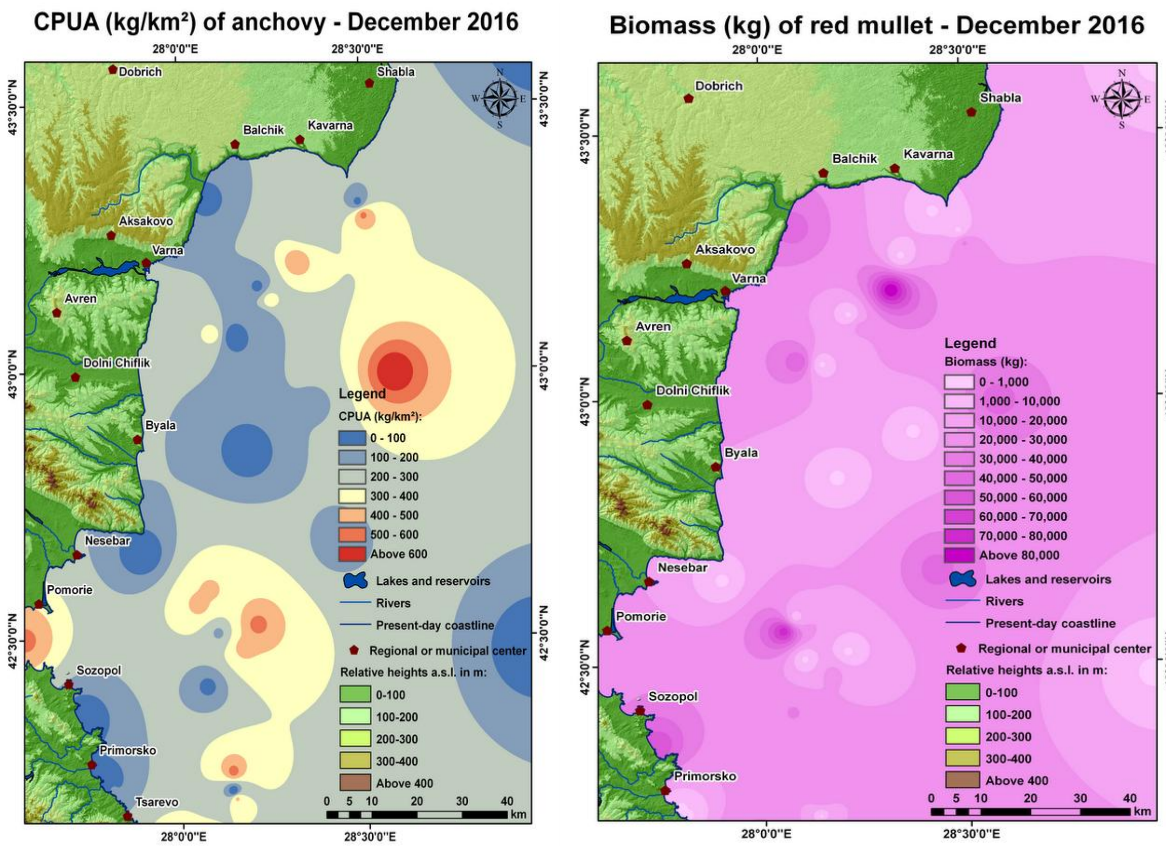


*Figure 4.2.3.*CPUA kg.km^{-2} and Biomass of anchovy in December 2016 from surveyed area.

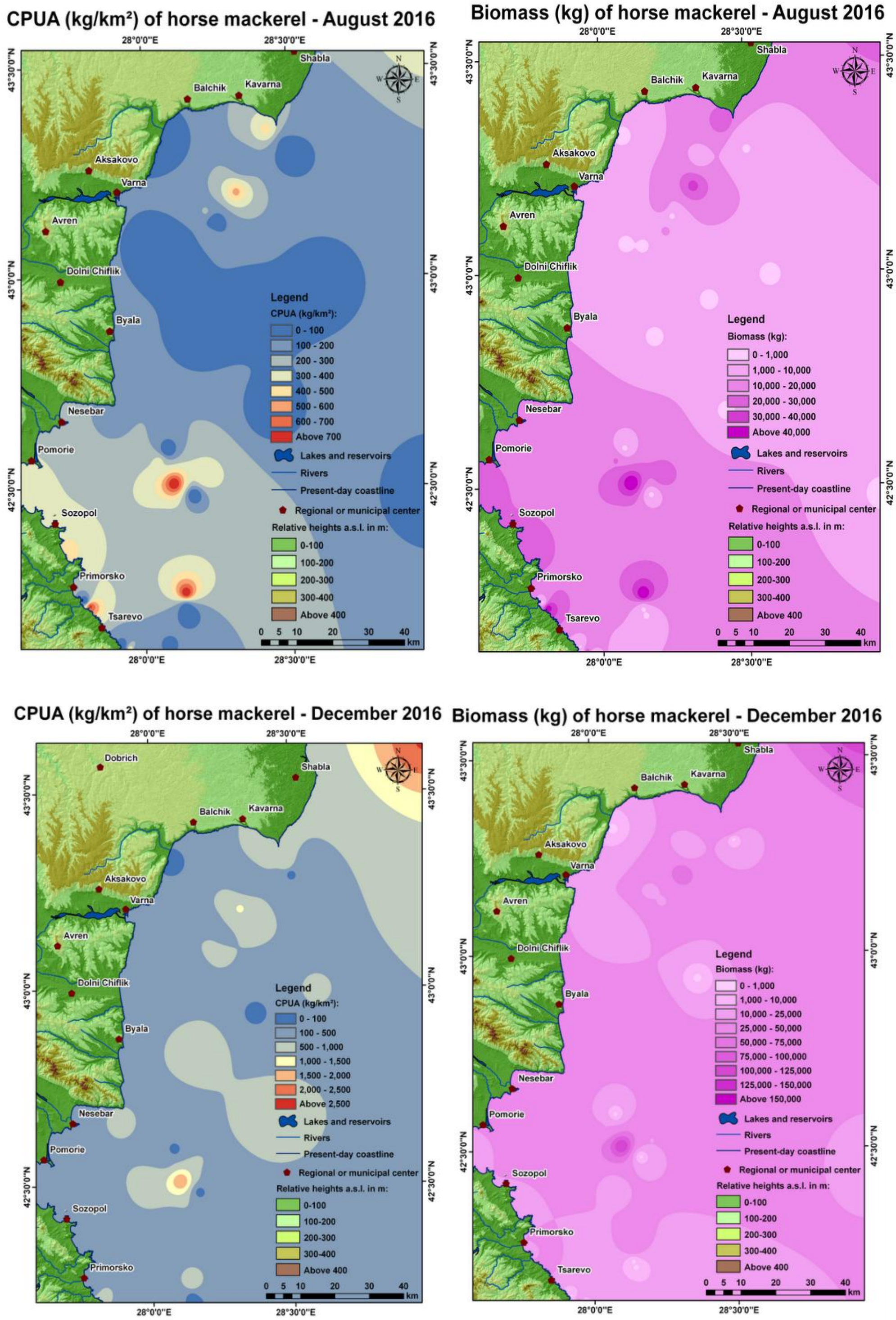


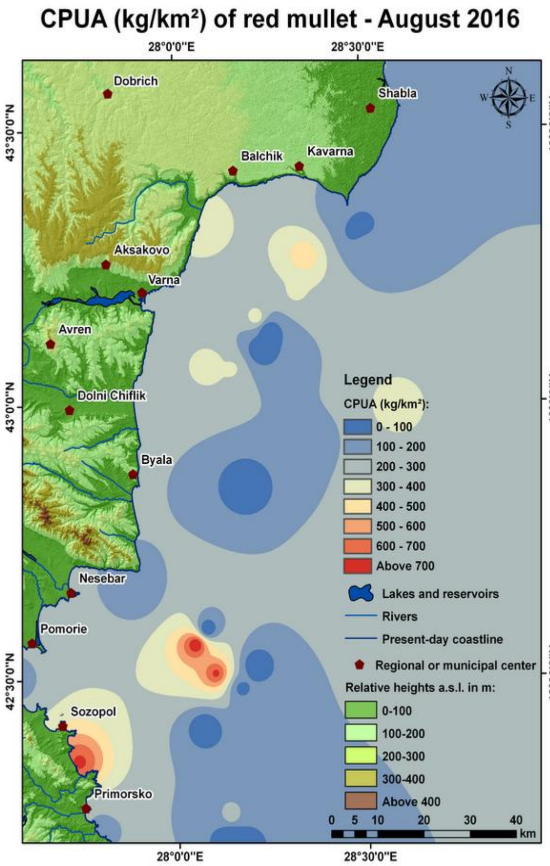
Figure 4.2.5.. CPUA kg.km^{-2} and Biomass of Scad August-September and December 2016 from surveyed area.

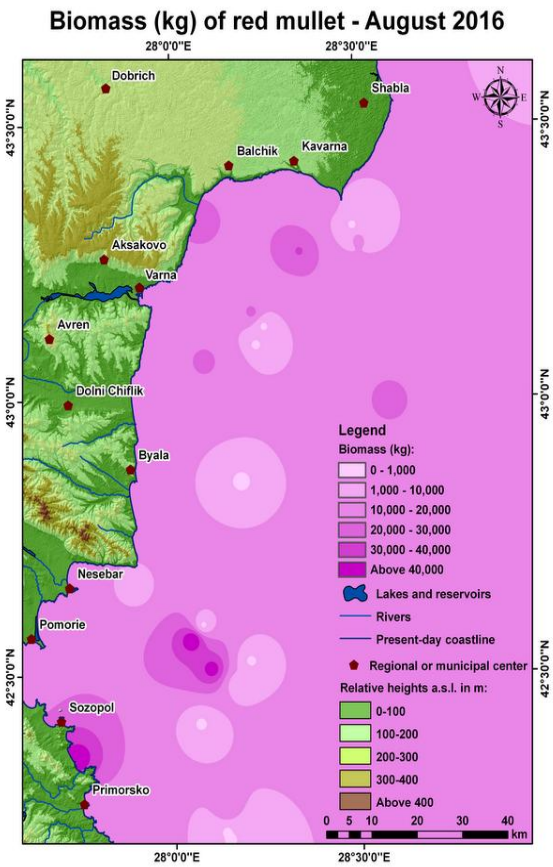
Scad, CPUA kg.km^{-2}, August-September 2016

In August, 2016 the highest concentrations were detected in southern part North from Tzarevo (22m depths, over 700 kg.km^{-2}), in front of Primorsko 30-50m depths, NW direction from Sozopol. Next, (over 500 kg.km^{-2}) agglomerations at depths 75m in front of Varna Bay were detected. Over 400 kg.km^{-2} were observed in front of Kavarna at depths 30-50m.

Scad, CPUA kg.km^{-2}, December 2016

Concentrations between 1500-2000 kg.km^{-2} were detected at depths of 65m in western direction ashore from Burgas Bay.





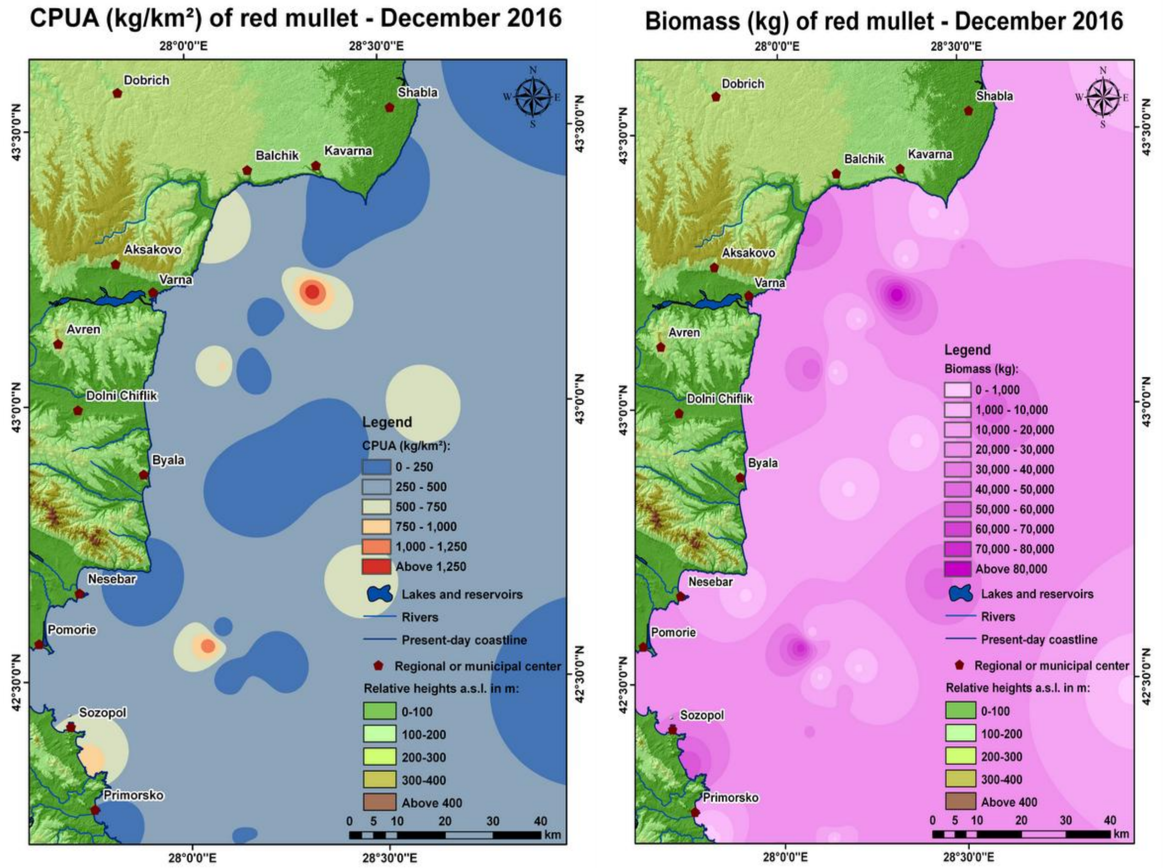


Figure 4.2.6.A, B. CPUA kg.km^{-2} and Biomass of Red mullet August-September 2016 from surveyed area.

Red mullet, CPUA kg.km^{-2}, August – September, 2016

4.3. Catch per unit effort

Values of sprat CPUE kg.h^{-1} from August-September and December 2016 - pelagic trawl survey is presented on Fig.4.3.1.

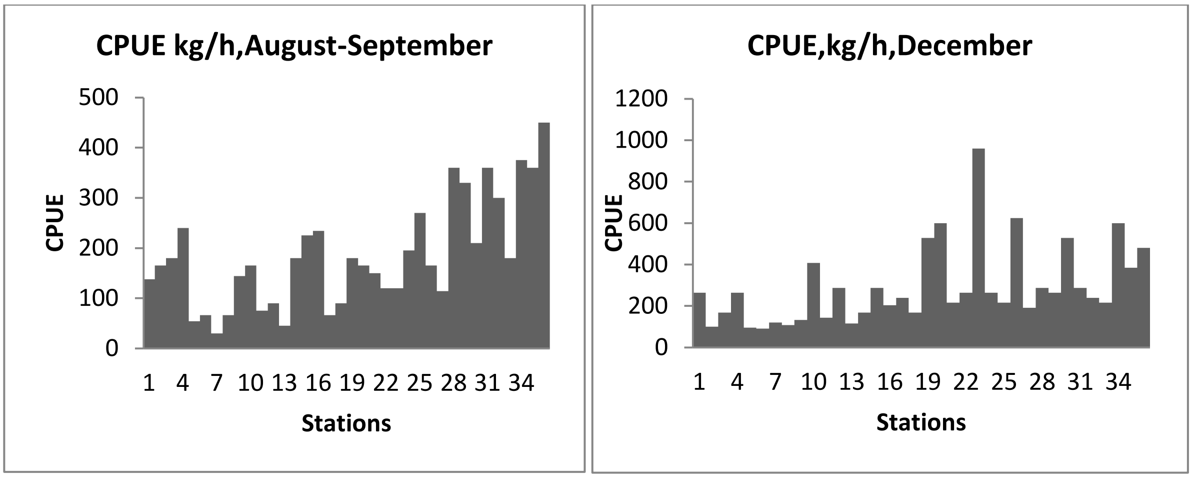


Figure 4.3.1. CPUE kg.h^{-1} from surveyed area.

4.4. Size structure of *S.sprattus*

The size composition comprised of length classes (TL, cm) from 6.5 cm up to 11.5 cm in the samples from Bulgarian marine area, (*Fig*.4.4.1. to 4.4.14).

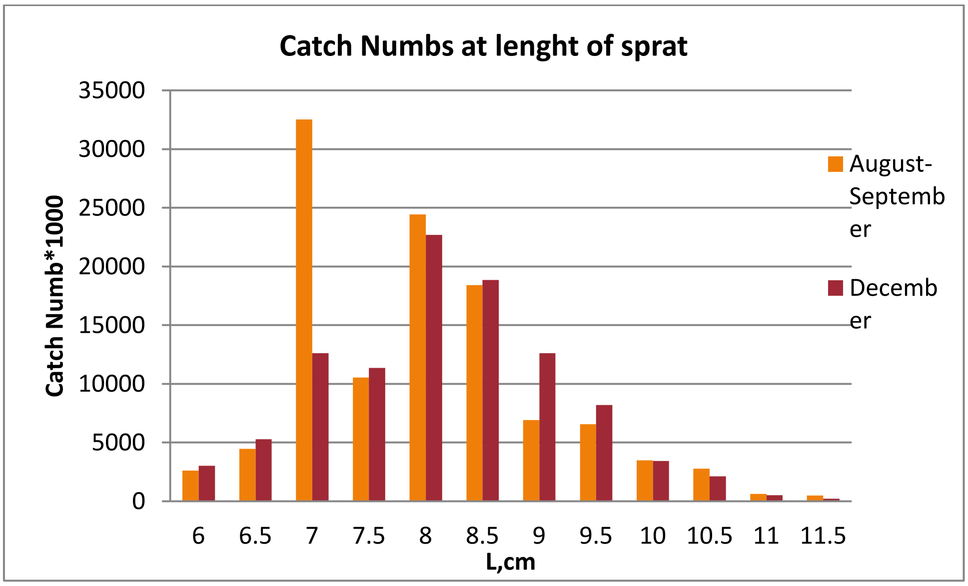


Figure 4.4.1. Share of size classes of sprat from Bulgarian marine area.

It is evident that the sizes classes from 7.0-8.5 cm are predominate, as the bigger classes were presented with low percentage. In August-September length calss 7.0 was with very high percentage, followed by L = 8.0 and 8.5 cm. The situation with the absence (or low share) of the larger (oldest) individuals was the same in the period of 2007-2015 (Raykov et al., 2007, 2008, 2009, 2010).

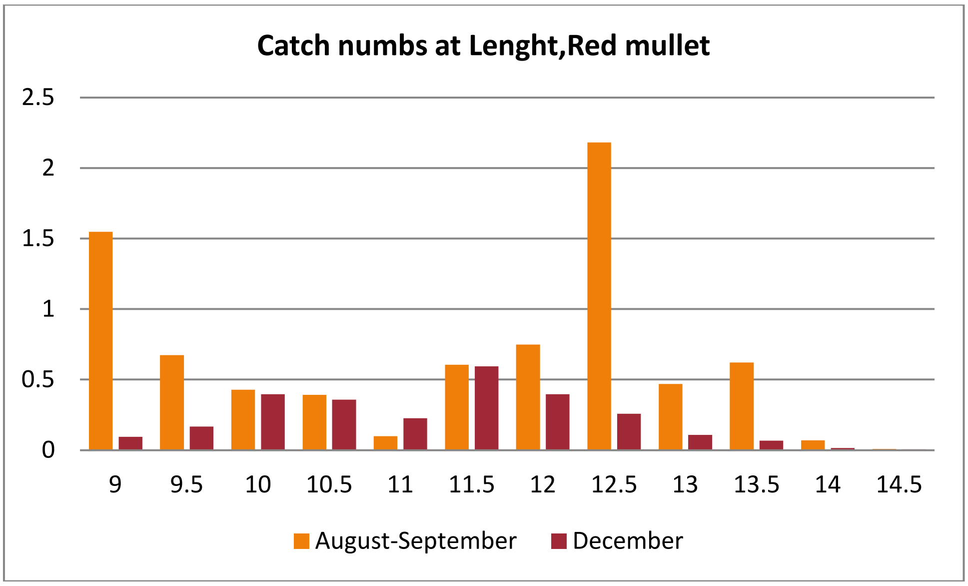


Figure 4.4.2. Share of size classes of red mullet from Bulgarian marine area.

Predominate L class in August September was 12.5cm, followed by Lclass= 9 cm.In December low share of all classes was detected.

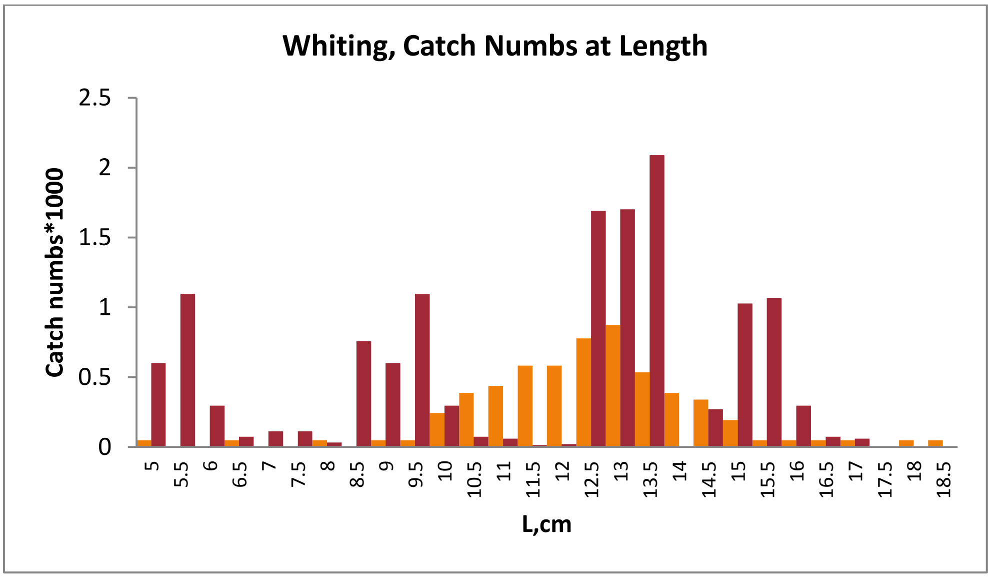


Figure 4.4.3. Share of size classes of whiting from Bulgarian marine area.

In August the share of 12.5 and 13 cm were the highest, although the presence of all length classes were very low in the catches.Later, in December the share of all length classes increased, as 12.5 ,13 and 13.5 classes increased twice and more.The largest classes of 15 and 15.5 cm increased significantly in December 2016.

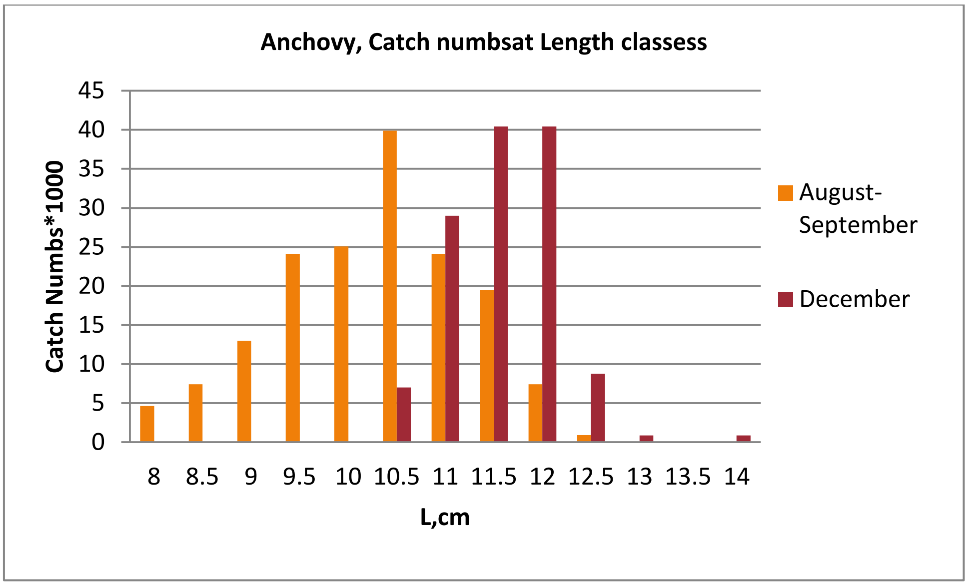


Figure 4.4.4. Share of size classes of anchovy from Bulgarian marine area.

In August-September the representation of length calssess begun with L=8 till 12.5 cm. In December the length classes from 10.5 to 14 were presented as the peak was at 11,11.5 and 12 length classes.

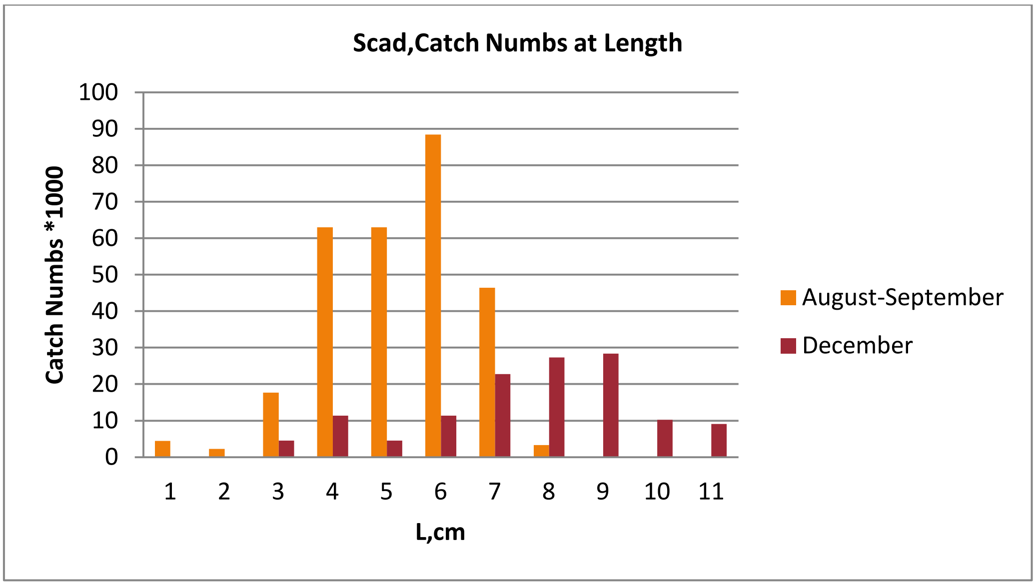
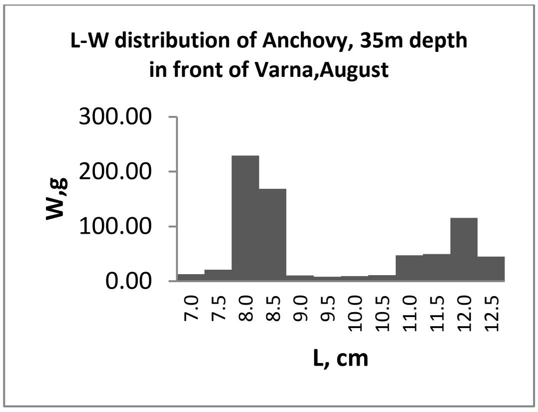
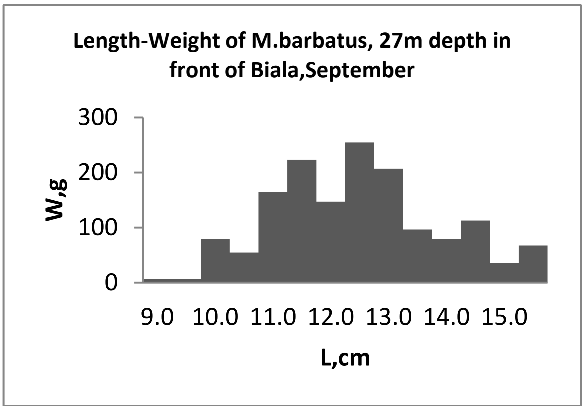
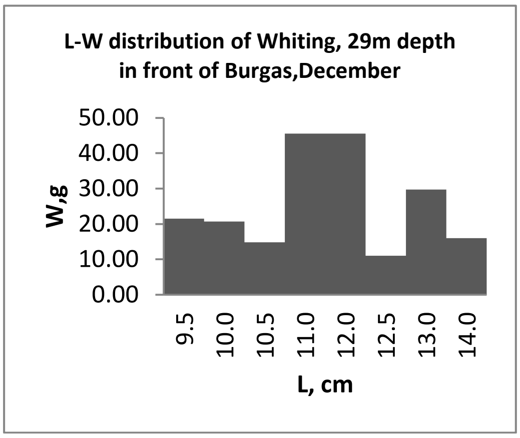
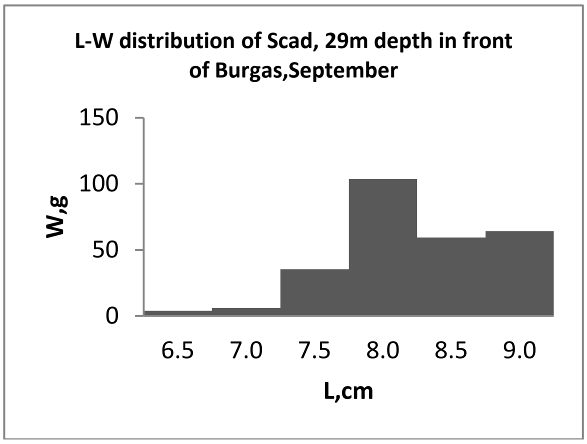


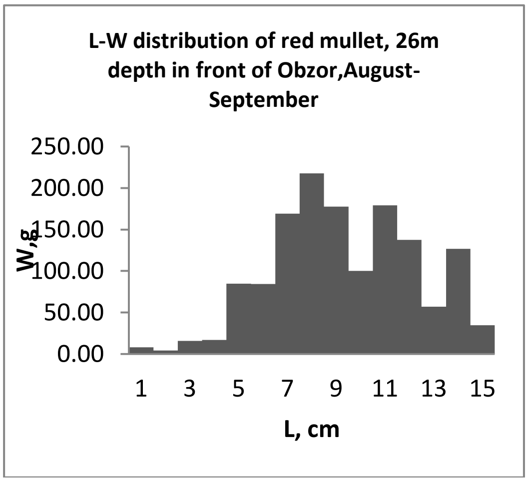
Figure 4.4.5. Share of size classes of scad from Bulgarian marine area.

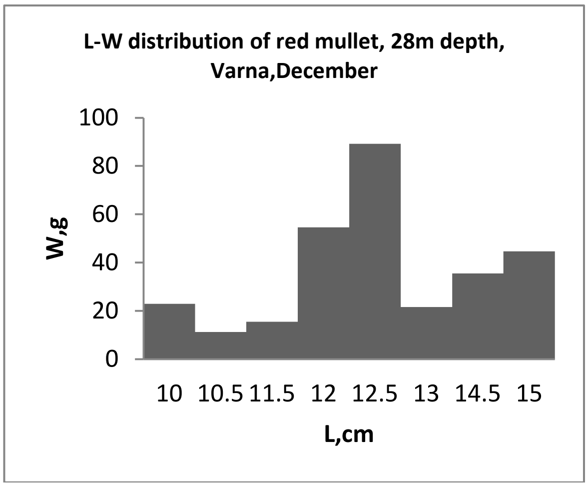

L-W distribution of Anchovy, 35m depth in front of Varna,August
300.00
200.00
100.00
0.00
W,g
7.0 7.5 8.0 8.5 9.0 9.5 10.0 10.5 11.0 11.5 12.0 12.5
L, cm

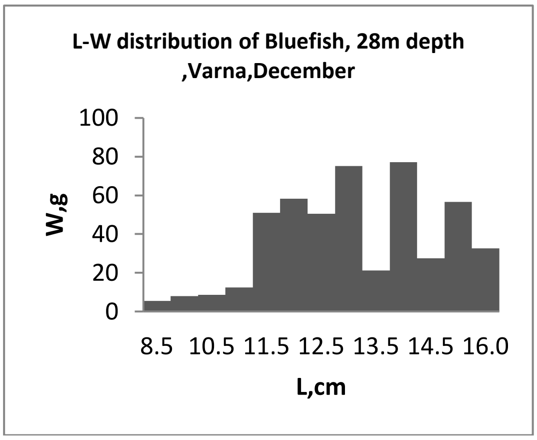

Length-Weight of M.barbatus, 27m depth in front of Biala,September
300
200
100
0
W,g
9.0 10.0 11.0 12.0 13.0 14.0 15.0
L,cm


L-W distribution of Whiting, 29m depth in front of Burgas,December
50.00
40.00
30.00
20.00
10.00
0.00
W,g
9.5 10.0 10.5 11.0 12.0 12.5 13.0 14.0
L, cm


L-W distribution of Scad, 29m depth in front of Burgas,September
150
100
50
0
W,g
6.5 7.0 7.5 8.0 8.5 9.0
L,cm


L-W distribution of red mullet, 26m depth in front of Obzor,August-September
250.00
200.00
150.00
100.00
50.00
0.00
W,g
1 3 5 7 9 11 13 15
L, cm


L-W distribution of red mullet, 28m depth, Varna,December
100
80
60
40
20
0
W,g
10 10.5 11.5 12 12.5 13 14.5 15
L,cm


L-W distribution of Bluefish, 28m depth ,Varna,December
100
80
60
40
20
0
W,g
8.5 10.5 11.5 12.5 13.5 14.5 16.0
L,cm

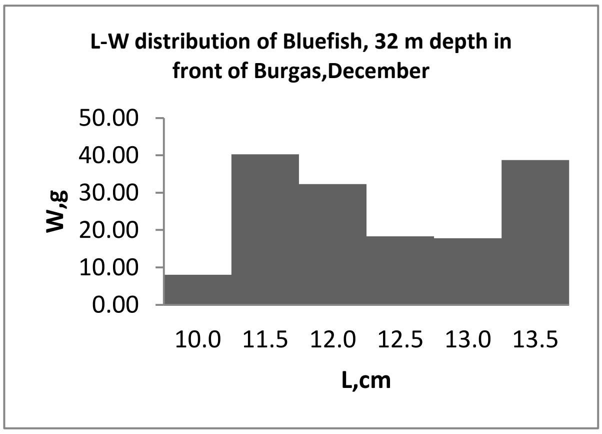

L-W distribution of Bluefish, 32 m depth in front of Burgas,December
50.00
40.00
30.00
20.00
10.00
0.00
W,g
10.0 11.5 12.0 12.5 13.0 13.5
L,cm

Figure 4.4.5. Length distributions of different species at different depths from pelagic surveys in 2016.

4.5. Age and growth of *S.sprattus* and bycatch

The age structure was determined on the basis of direct otolith reading with binocular on reflected light. Analysis shows that the percentage of one year old fish is greatest in both surveys. (*Fig*.4.5.1.A, B.)

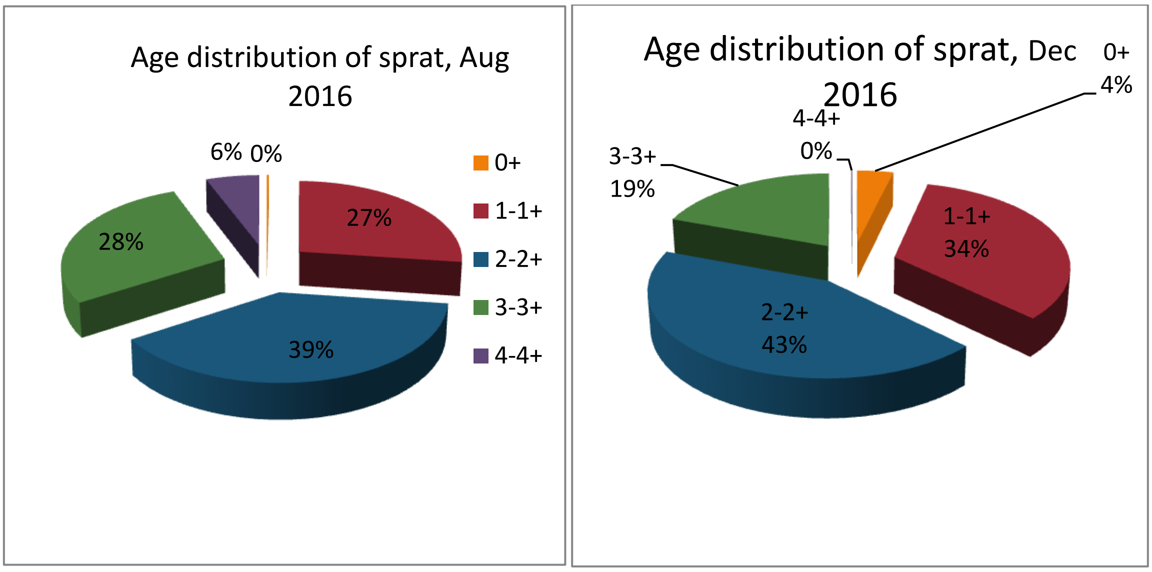
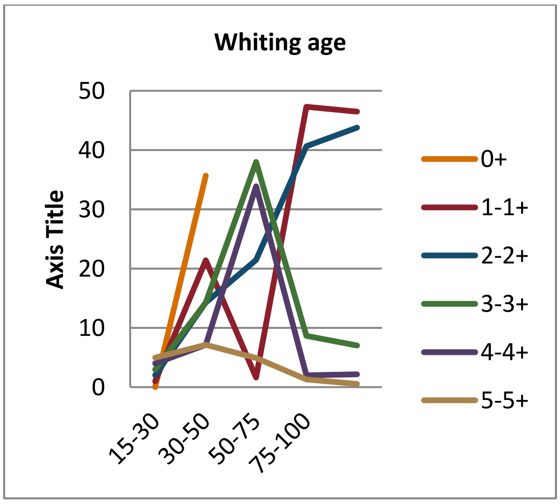
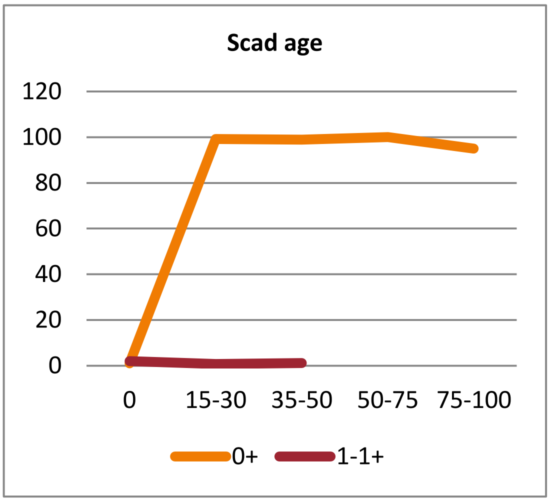


Fig.4.5.1.A, B. Age distribution of sprat in August-September and December 2016





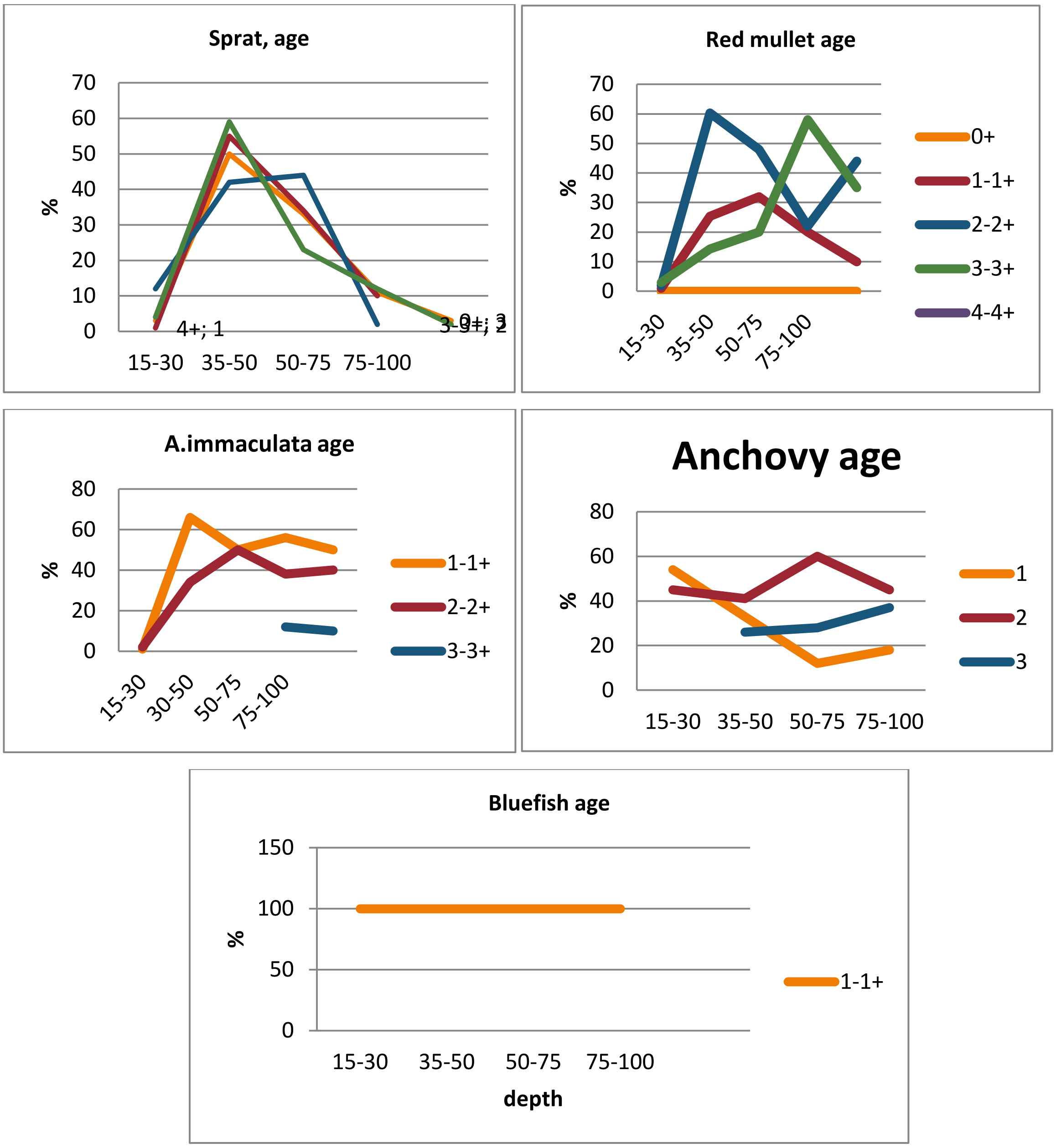


Fig.4.5.2. Age distribution of species in August-September 2016

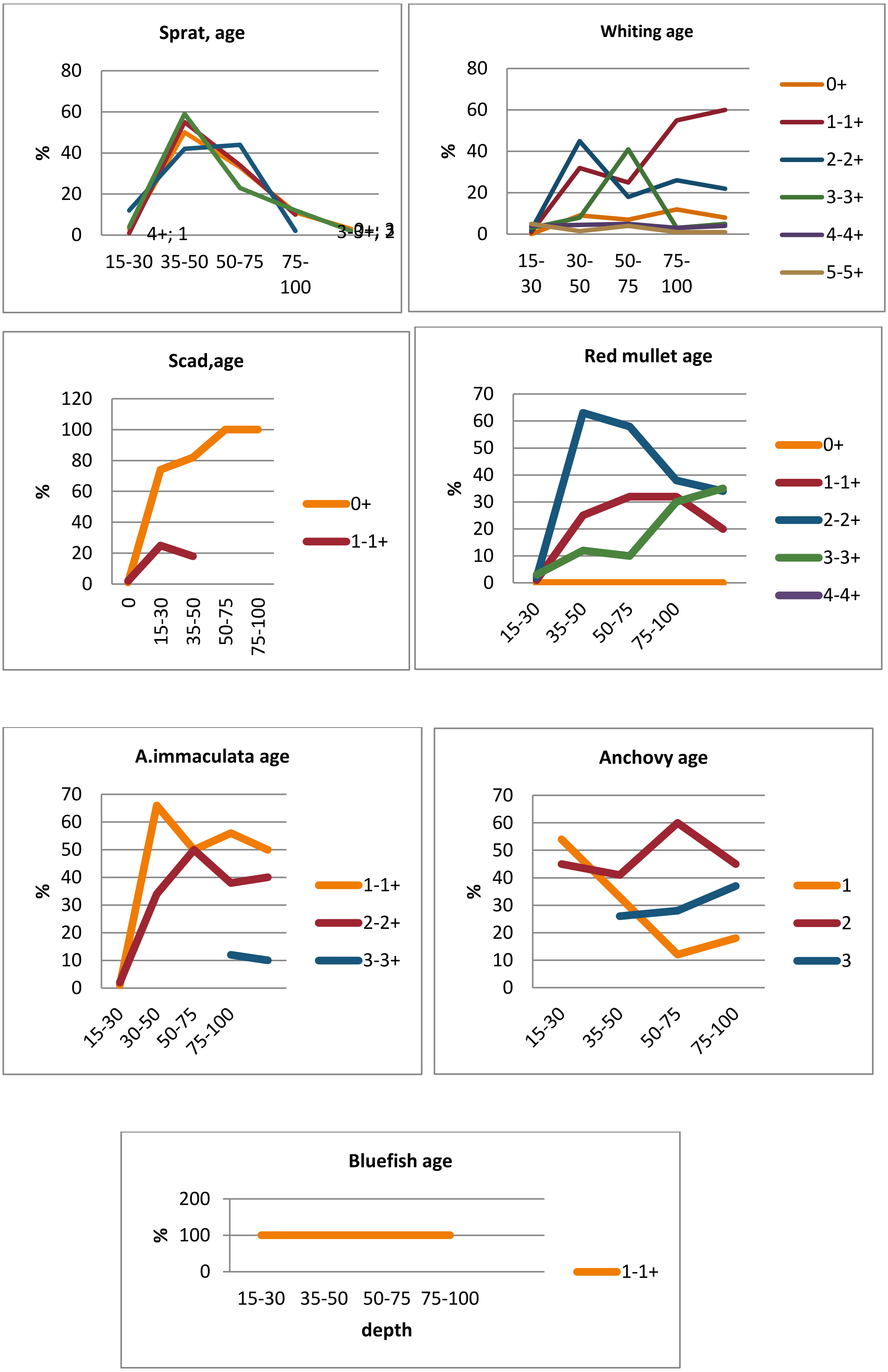


Fig.4.5.2. Age distribution of species in December 2016

4.6. Growth

To estimate the growth rate and population parameters of the sprat from Bulgarian and Romanian marine areas we used VBGF. The estimation of the asymptotic length, growth rate and related coefficients is presented on Table 4.6.1.

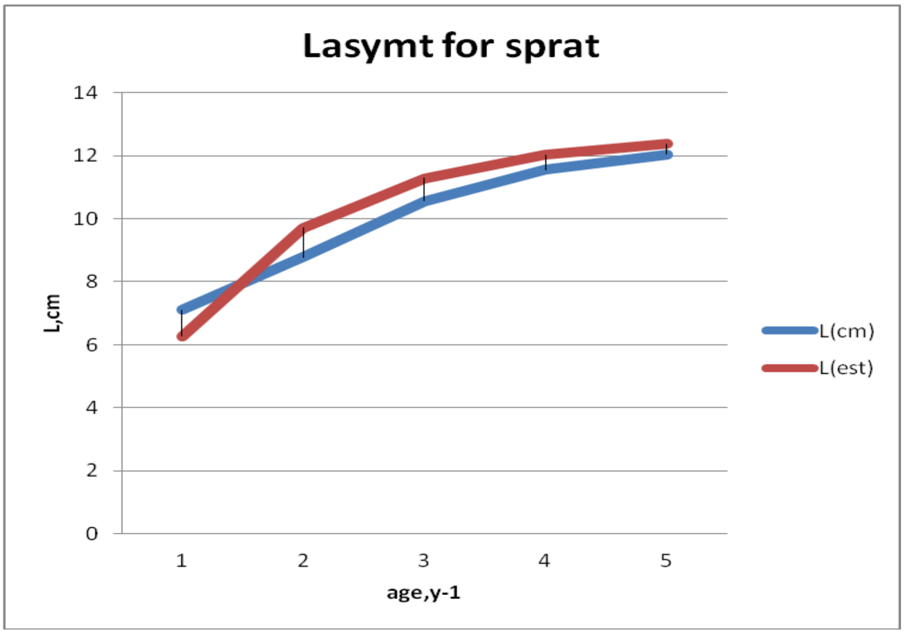


Fig.4.6.1. L asymptotic of sprat

Table 4.6.1. Values of parameters in VBGF.

Parameters	total
L_∞ (cm)	12.77
k	0.45
t_0	-0.22
q	0.0008
n	2.66
W∞(g)	11.27
k	0.24
t0	-7.33

Size growth

The asymptotic length reached 12.77cm and growth rate could be assessed as relatively high accounting 0.45 $^{y-1}$.The growth of sprat from present research is positive allometric (n=2.66) (Fig.4.6.1).

The important remark here is the fact that due to absence (or low share) of the oldest and individuals with large sizes, the VBGF accounts only the shorten size structure of sprat from present research. In this regard, the maximum or asymptotic length reached this value, which possibly not fully corresponds to the literature data about sprat size and marginal levels of length and growth rate. Hence, we could accept the growth analysis, as it is, reflecting only the present situation with large fish absence (low presence).

Somatic growth

The somatic growth of sprat from the present research shows that mean weight corresponding to the oldest age group is 8.05 g. The value corresponds to the marginal size of 11.75 cm observed in the samples from the trawl survey in Bulgarian waters (*Fig*.4.6.1.).

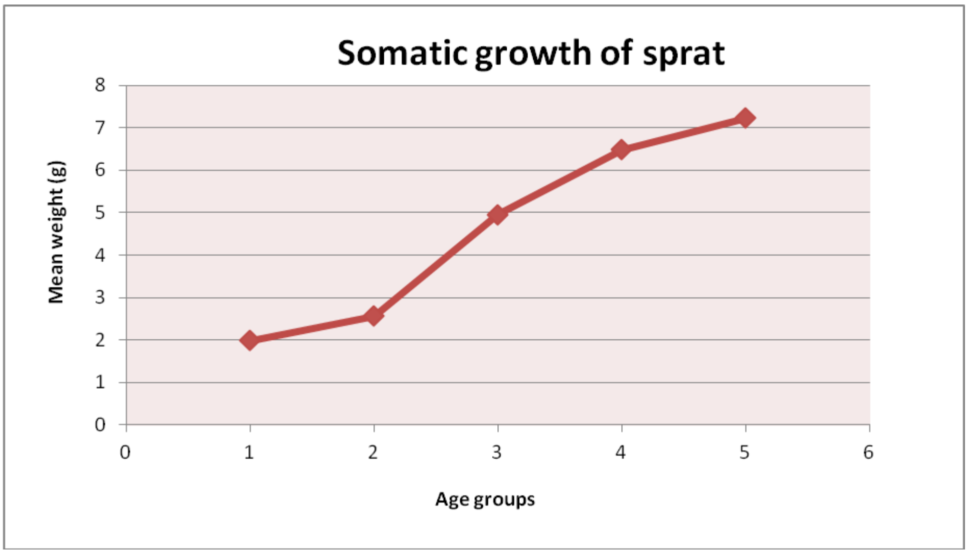


Figure 4.6.2. Somatic growth of sprat

Asymtotic somatic growth reached 11.41.The growth rate in weight is assessed as comparatively stable and high 0.46. This fact could possibly related to the fact that in December the species is in active gonad maturation and spawning with high batch fecundity.The batch fecundity of sprat was estimated in December since this is the active period of species spawning.

4.7. Natural mortality

We used the formula incorporated asymptotic length and weight derived by Pauly (1980).The average habitat temperature in the bottom layers was 6.9°C:

From asymptotic length:

M = 0.7632

From asymptotic weight:

M = 0.582

In the present research we used natural mortality coefficient for sprat as equal to **0.95** (Ivanov and Beverton, 1985; Prodanov et al., 1997; Daskalov, 1998).

4.7. Sex ratio

Females prevailed with 49%, followed by males (48%).The juveniles presented with low percent (3%)

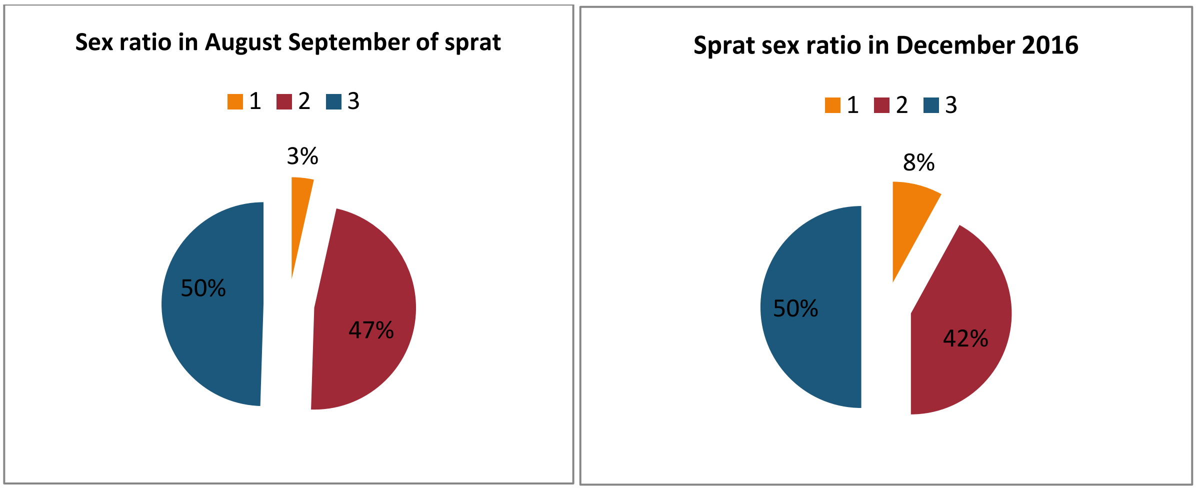


Figure 4.7.1. Sex ratio of sprat in August-September 2016.1-juv;2-male;3-female

4.8. Maturity

The sprat in active spawning during the present investigation in December. Most of the individuals are with III - IV stage of gonads. More detail analysis should be done in the active spawning period of the species (October-February).

Gonado somatic index

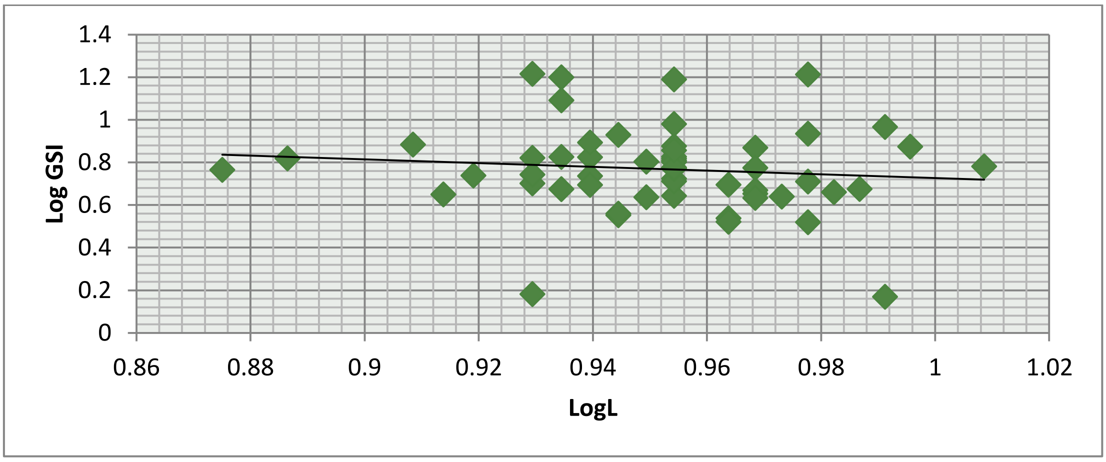
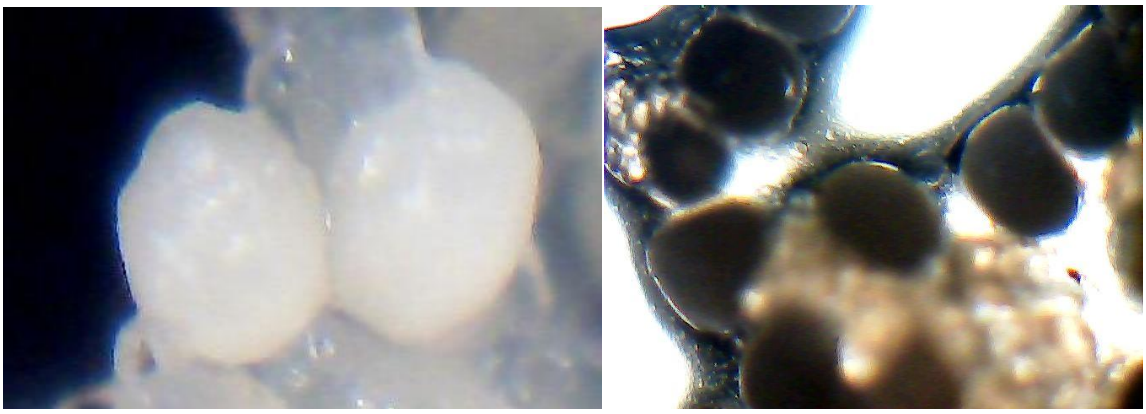


Figure 4.8.1. Gonado somatic index of sprat from present research (GSI, %)
The GSI (%) show that over 50% of females were active spawners. Very few specimens Were with early maturation stages, so we can conclude that in December 2016, the active spawning has been initiated, even the SST was relatively high for the season.



Picture 4. Eggs of sprat

Fecundity

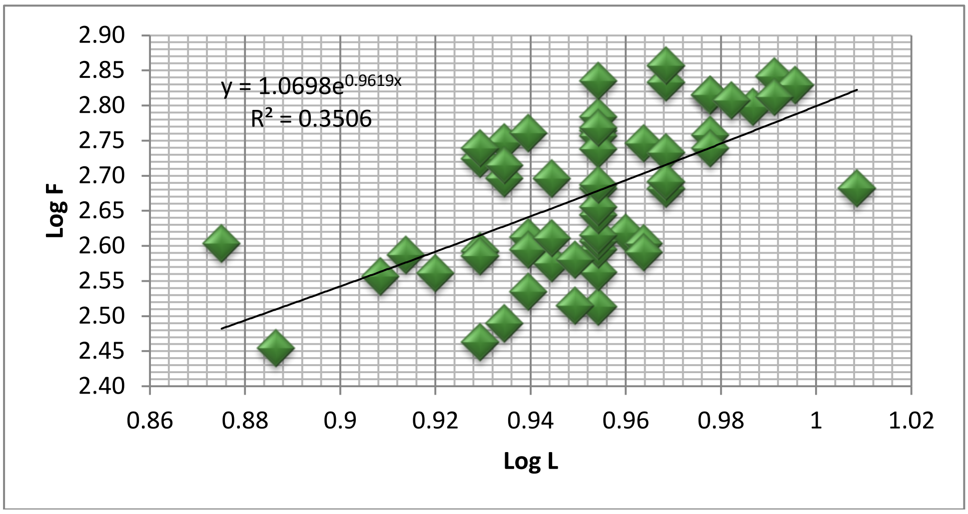


Figure 4.8.2. Batch fecundity (LogF) in relation to the length (LogL) of sprat in December, 2016

Fecundity of sprat correlate positive with sprat length (R^2 = 0.46), as the biggest length classes posses highest fecundity.

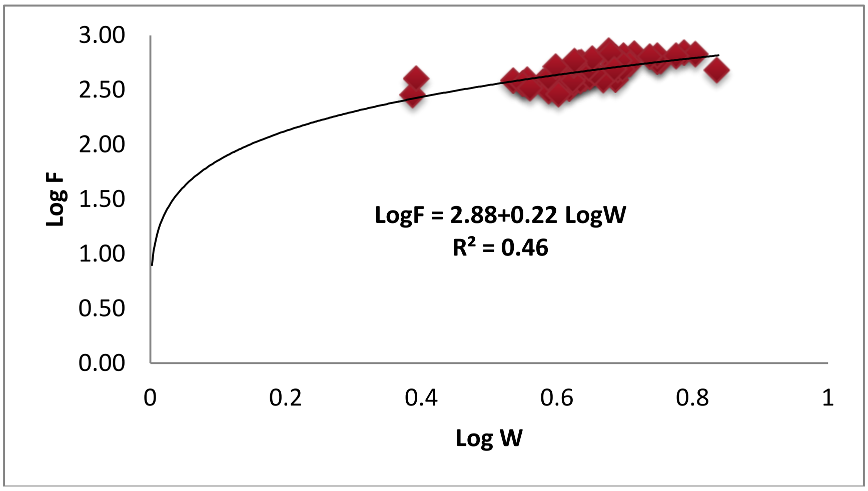


Figure 4.8.3. Batch fecundity (LogF) related to weight (LogW) of sprat in December, 2016

The correlation between fecundity and weight of sprat was much stronger (Fig.4.8.3.)

(R^2 = 4475)

4.9. Food composition

Results

Food composition and feeding patterns of sprat (*Sprattus sprattus*, L) and anchovy (*Engraulis encrasicolus*, L)

Material and method

The study of sprat and anchovy diet was based on analysis of stomach content composition of 38 sprats and 40 anchovy specimens, collected in front of Bulgarian Black Sea coast during 28.08 – 05.09. 2016. Additionally, this study encompassed analysis of the mesozooplankton species composition and biomass in the environment, as these pelagic organisms form the main food source for planktivorous fish species.

The coordinates and information about the sampling sites were presented at Table 4.9.1.

Table 4.9.1 Information on the investigated stations in 28.08 – 5.09.2016.

Date	№	Coordinates	Depth (m)	Zooplankton stations	Sprat food	Anchovy food
28.08.2016	1	42.22.10 N - 28.14.58 E	82	Zoo1		
29.08.2016	2	42.23.4 N - 27.52.5 E	40	Zoo2	FoodT1	FoodA1
29.08.2016	3	42.21.59 N - 27.51.59 E	37	Zoo3		FoodA2
29.08.2016	4	42.40.2 N - 27.41.1 E	27	Zoo4		FoodA3
30.08.2016	5	42.47.3 N - 28.17.0 E	84	Zoo5	FoodT2	
30.08.2016	6	42.56.72 N - 28.10.29 E	42	Zoo6	FoodT3	
4.09.2016	7	43.21.8 N - 28.33.5 E	44	Zoo7		
4.09.2016	8	43.32.30 N - 28.49.4 E	50	Zoo8	FoodT4	
4.09.2016	9	43.32.3 N - 28.46.2 E	76	Zoo9	FoodT5	
5.09.2016	10	43.05.7 N - 28.08.3 E	26	Zoo10		FoodA4
5.09.2016	11	42.02.7 N - 28.05.5 E	24	Zoo11		FoodA5

For the purposes of investigation about 10 - 12 fish specimens per trawl catch were separated and preserved in 10 % formaldehyde: seawater solution. The absolute length (TL, to the nearest 0.1 cm) and weight (to the nearest 0.01 g) of fish specimens were measured. Under laboratory conditions the stomachs of the selected animals were weighted with analytical balance (to the nearest 0.0001 g). The food mass of each individual has been calculated as a difference between the weights of full and empty sprat stomach.

The stomach content was investigated under microscope for estimation of species composition and prey number. The prey biomass was estimated by multiplication of the number of consumed mesozooplankton species by their individual weights.

The following indices were calculated: stomach fullness index (ISF) as a percent of body mass: (stomach content mass/fish mass) *100; and index of relative importance - IRI, Pinkas et al. (1971): IRI = (N+M) x FO; where N - the proportion of prey taxa (species) in the diet by numbers (abundance); M - the proportion of prey taxa (species) in the diet by mass; FO- frequency of occurrence among fish.

The zooplankton samples in open sea were gathered from the whole water layer (bottom- surface) with a plankton set (opening diameter d=36 cm; mesh size 150 µm). The samples were fixed onboard ships with 4% formaldehyde: seawater solution (Korshenko & Aleksandrov, 2013). The mesozooplankton species composition has been identified by Guides for the Black and Azov Seas (Morduhai-Boltovskii et al., 1968), and its quantity - by the method of Bogorov (Korshenko & Aleksandrov, 2013).

Sprattus sprattus: weight – length dependence, Index of stomach fullness (ISF)

The analysis of sprat feeding patterns was based on data collected from 38 fish samples processing. The mean absolute length of investigated specimens reached 88.81 ± 0.74 (SD) mm, varying between 72 - 105 mm, correspondingly the mean weight was 3.76 ± 0.90 (SD) g, varying from 2.31 g. to 5.86 g.

The weight-length dependence, established by this study, was described by the following equation: Log WW(g)=2.5283*Log L(cm) – 1.8334; (r2=0.67, p<0.001, Fig.4.9.1.).

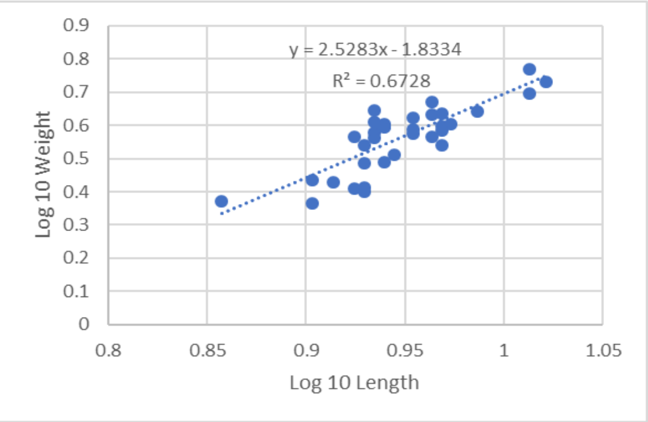


Figure 4.9.1.. Weight-length relationship for sprat, investigated in 2016.

In summer 2016, the mean value of the index of stomach fullness (ISF) reached 0.87 % ± 0.816 (SD) of sprat body weight (BW). Since the beginning of the sprat trawl surveys in 2007, we have observed a tendency of increasing values of ISF. Compared to the ISF mean value for the period 2007-2010, this index has increased with 1.6 times in 2016.

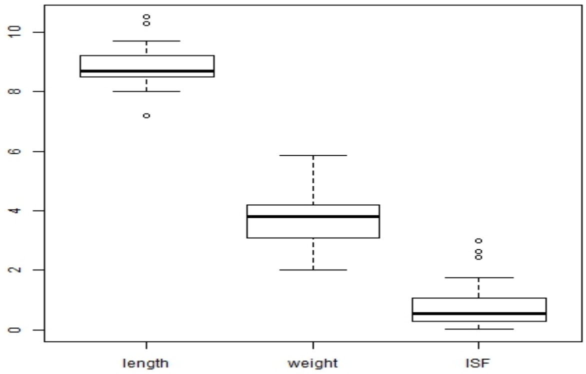


Figure 4.9.2. Boxplot: (median values, 25 – 75 % hinge, minimal and maximal values) for sprat weights, size and Index of stomach fullness (ISF) in 2016 (data extracted from the sprat feeding study).

The highest mean values of the stomach fullness index (ISF = 1 - 1.14 %) were found in the zone after 50 - meters isobaths (Fig. 4.9.3). In the shelf region, between 40 m - 50 m isobaths, the ISF has comprised 0.3-0.6% from the sprat body weight. Obviously, during the late summer in 2016, sprat find the more appropriate food conditions in the deep-water region, between 70 m and 80 m isobaths (Fig.4.9.3.).

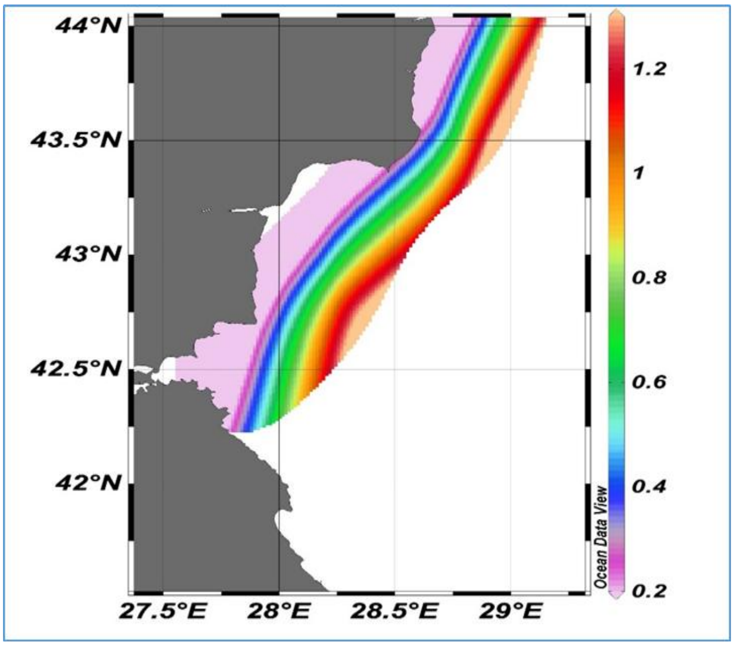


Fig. 4.9.3. Spatial distribution of ISF in 28.08-5.09.2016.

Among the ISF and sprat body weight (in the span - 2.31 - 5.86 g) a negative correlation (R2= -0.30) was established, which is not statistically significant. The maximal food amount, expressed as a percent of the sprat body mass (ISF) was observed by small sized specimens with weight 2.5 g, and by individuals with weight ~ 4 g (Fig.4.9.4). However, by the above-mentioned weight classes, individuals with low indexes of stomach fullness could be discovered (Fig. 4.9.4).

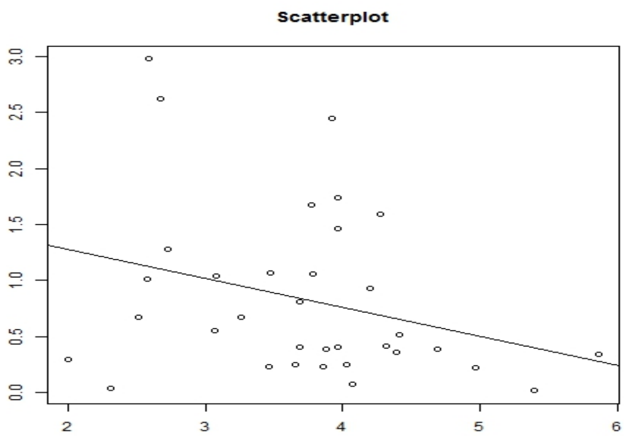


Figure 4.9.4. Scatterplot: Relationship between sprat weight (W, g) and ISF.

The collected data didn't permit us to obtain a statistically significant dependence between the observed variations of ISF and sprat body weight. Plausibly, other factors were crucial for the sprat feeding success, such as the amount of fodder mesozooplankton, and/or completed feeding migrations, that allow sprat to explore appropriate food sources.

Species composition and index of relative importance (IRI) of different mesozooplankton species in the sprat diet

In 91.67% of all investigated sprat specimens a presence of fodder mesozooplankton was found, which allow quantitatively and qualitatively identification through the microscope observations, i.e. it is possible to estimate the number and the species composition of the prey items. By 8.33 % of all sprat specimens only food remains were discovered, which didn't allow quantitative analysis.

The maximal number of prey items - 1506 ind/stomach was established in the sample from the station 5 (at 84-m depth in front of Obzor). At the same station, the mean prey number (PN) attained 680 ind/stomach, while in the whole studied area it accounted up to 120 ind/stomach. For comparison, during 2007-2010 the mean prey number in sprat stomach contents has reached ~ 64 ind/stomach. It is worth mentioning, that during the current survey the trend of elevated prey number was not ubiquitous. For instance, in the coastal and shelf region up to 50-m isobath, the amount of prey organisms varied among 10-79 ind/stomach, with minimum at station 8 (northern from Cape Kaliakra). Thus, in 2016, the increased mean value of PN was caused by a distinct raise of prey items at one deep-water station. Similar phenomenon was observed in 2010, when the mean PN reached akin levels of 649 ind/stomach at a sole open sea station, while consistently lower levels were detected through all other observations.

Analysis of the zooplankton samples, gathered from the marine environment, showed a total diversity of 21 mesozooplankton species/groups. From the latter, 11 species/groups were found in the sprat diet. From *Copepoda* representatives - *the species Pseudocalanus elongates, Calanus euxinus, Acartia clausi, Centropages ponticus, and Oithona spp*. were most often identified in the sprat diet; from the group of pelagic larvae of bottom species (meroplankton) - four taxonomic groups were found - Lamellibranchia veliger, Gastropoda veliger, Decapoda zoea, Cirripedia *nauplii* and cypris; planktonic crustaceans Cladocera were represented by genera Evadne; class Chaetognatha – from species *Parasagitta setosa.*

The indices of relative importance (IRI) of the main mesozooplankton representatives in sprat food spectrum (based on the subsequent percent shares from total abundance, biomass and frequency of occurrence in samples) were represented in Table 4.9.2.

.

Table 4.9.2. The sprat food composition of in 28.08-05.09.2016.

	N (%, from total abundance)	M (%, from total biomass)	FO – Frequency of occurrence	IRI – Index of relative importance
Pseudocalanus elongatus+copepodit stages	34.01	24.08	61.76	3587.64
Acartia clausi+copepodit stages	6.13	3.00	55.88	510.18
Copepoda copepodit (III -V)	16.73	23.42	50.00	2007.50
Calanus euxinus+copepodit+naupli stages	4.74	20.65	20.65	524.30
Copepoda adult	5.58	6.00	41.17	476.75
Lamellibranchia veliger	16.96	12.51	11.76	346.57
Centropages ponticus	0.31	0.33	26.47	16.94
Decapoda	0.49	0.85	8.82	11.82
Parasagitta setosa	9.21	8.99	32.35	588.77
other	5.84	0.17	23.22	139.55
total	100%	100%		

The sprat food was dominated by copepods - *Pseudocalanus elongatus*, *Acartia clausi, Calanus euxinus*, *Centropages ponticus* (Table 4.9.2, Fig. 4.9.5), as well as by copepodid stages (III-V) and Copepoda adults (that couldn’t be identified to species level due to partly decomposition). The cold-water copepod *Pseudocalanus elongatus* predominated the sprat food by abundance and biomass, and showed the highest frequency of occurrence (Table. 4.9.2, Fig.4.9.5).

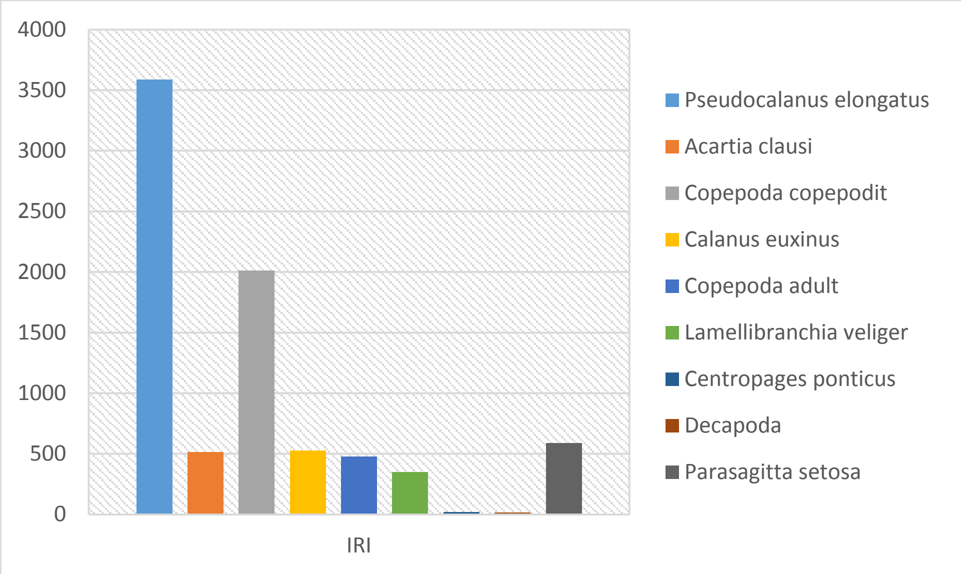


Figure 4.9.5.. IRI of different mesozooplankton species in the sprat food in 28.08.-05.09.2016.

Significant differences were observed in sprat feeding patterns per stations (Table 4.9.3). In front of the central sector - Biala/Obzor and between 42-84 m isobath (stations 5 and 6), the sprat food spectrum included mainly *Pseudocalanus elongatus* and copepodid stages of copepods. In the southern coastal area, the sprat diet was based on copepodid stages of crustaceans and meroplankton larvae of Lamellibranchia. In north direction - at depths 50-76 m, in front of Shabla, the sprat food consisted mainly from *Parasagitta setosa* and *Calanus euxinus.*

Table 4.9.3. Distribution of IRI (%) of main mesozooplankton representatives in sprat food per observed stations in August/September 2016.

Zooplankton

Zooplankton	Ст.2/40m	Ст.5/84m	Ст.6/42m	Ст.8/50m	Ст.9/76m
Pseudocalanus elongatus+copepodit stages	0.0	53.2	29.6	1.5	1.6
Parasagitta setosa	0.0	0.0	0.0	56.8	42.0
Acartia clausi+copepodit stages	0.0	17.8	9.8	0.0	8.7
Calanus euxinus+copepodit+naupli stages	0.0	5.1	6.5	38.5	29.4
Copepoda adult	0.0	5.9	13.1	0.0	17.2
Copepoda copepodit (III -V)	84.3	17.0	40.3	0.0	0.0
Lamellibranchia veliger	15.7	0.0	0.0	0.0	0.0
Centropages ponticus	0.0	0.6	0.3	2.9	0.2
Decapoda	0.0	0.3	0.1	0.3	0.7
others	0.0	0.3	0.2	0.0	0.3

Parasitic Nematodes were discovered by 16 % of a total of 38 sprat specimens.

Engraulis encrasicolus: weight – length relationship, Index of stomach fullness (ISF)

The mean absolute length of investigated anchovy specimens reached 11.57 ± 0.88 (SD) cm, varying between 8.7 - 13 cm, while the mean weight was 8.43 г. ± 1.90 (SD) g, varying from 3.02 г. to 13.78 g. (Fig.4.9.4).

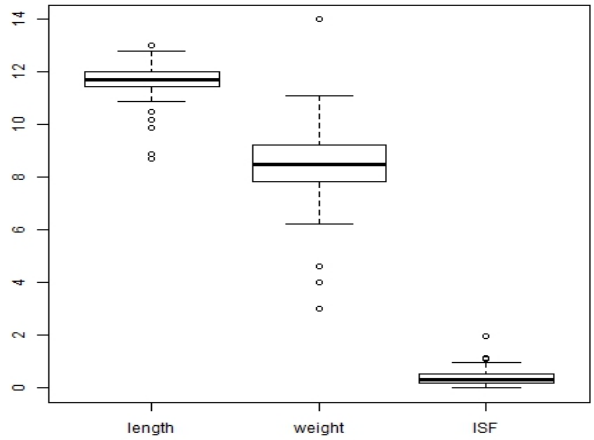


Figure 4.9.4 Boxplot: (median values, 25 – 75 % hinge, minimal and maximal values) for anchovy weight, size and Index of stomach fullness (ISF) in 2016 (data extracted from the anchovy feeding study).

The anchovy weight-length relationship could be described by the equation: Log WW(g)=3.0915*Log L(cm) – 2.3701; ($r2=0.89$, $p<0.001$, Fig.4.9.5).

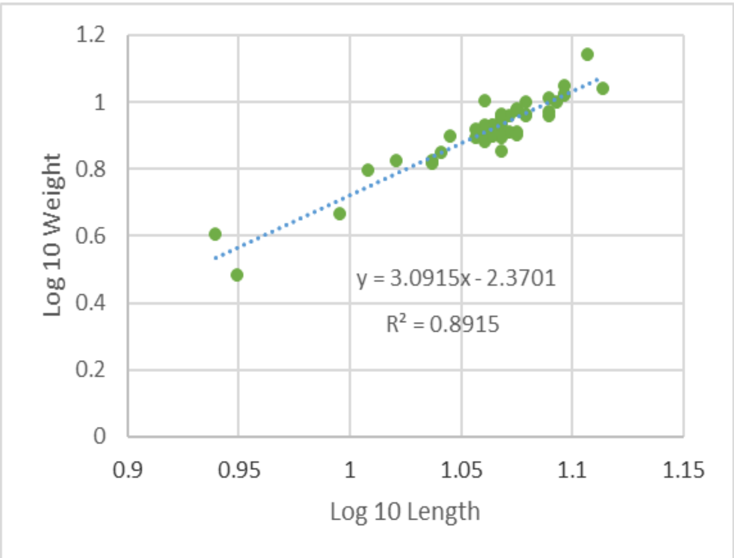


Figure 4.9.5. Weight-length relationship for anchovy, investigated in 28.08-5.09.2016

The mean value of stomach fullness index reached 0.43 % ± 0.37 (SD) of the anchovy body weight during the studied period. The highest mean values of ISF, up to 0.6 %, were found in the coastal area in front of Biala and Duni. The lowest vlevels of ISF - 0.25 - 0.38 % were registered in southern direction, near to Ahtopol (Fig. 4.9.6).

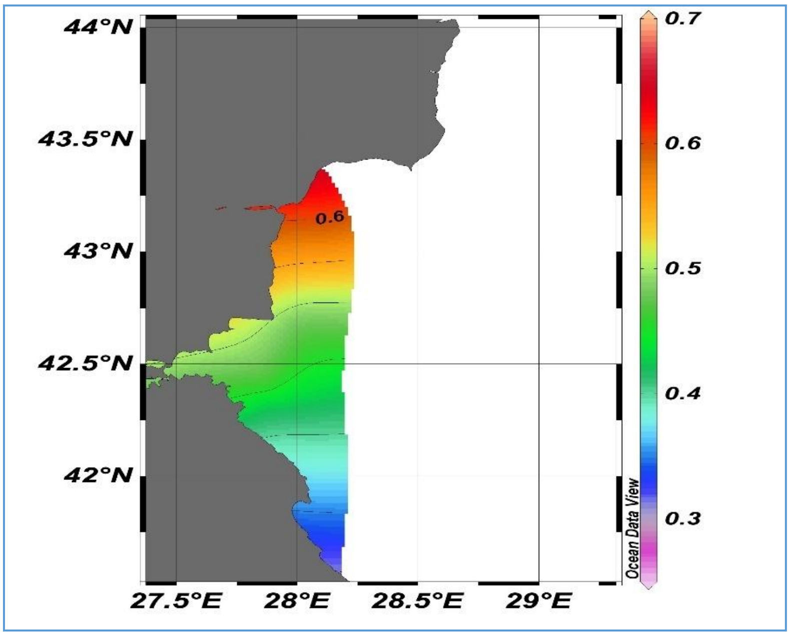


Fig.4.9.6 . Spatial distribution of anchovy ISF in 28.08-5.09.2016.

Among the ISF and anchovy body weight (in the span - 3.02 - 13.78 g), a statistically significant non-linear dependence was established, that explained 76 % of the observed variations by $p<0.001$, df =39 (Fig. 4.9.7). The food amount, expressed as a percent of the sprat body mass, has increased by small sized groups, with weight < 4 g. The second, less expressed pick of ISF was found by weight class - 6.5-7g (Fig.4.9.9), while by large-sized specimens with weight 9-13 g, the ISF showed comparable, but lower levels (Fig.4.9.7).

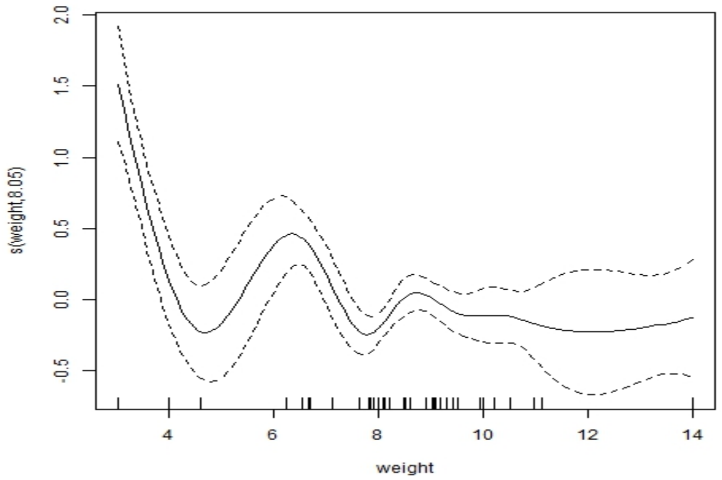


Figure 4.9.7. Contribution of the factor weight (W, g) to the fitted values of ISF. (With dotted line was estimated 95% confidence interval).

Species composition and index of relative importance (IRI) of different mesozooplankton species in the anchovy diet

In 85% of all investigated anchovy specimens a presence of fodder mesozooplankton was found, which could be quantitatively and qualitatively identified. By 12.5 % of all specimens only food remains were discovered, and by 2.5% of fish empty stomachs were observed.

In the investigated samples, the mean prey number reached 138 ind/stomach. The maximal prey number - 893 ind/stomach was identified at st.4 (located in front of the Cape Emine, at 27 m depth), while at the same station, the mean PN sustained 178 ind/stomach. In the coastal area and shelf zone up to 40 m isobath, the mean prey number varied from 17 ind/stomach to 178 ind/stomach, with a minimum at station 2, located near to Duni.

In the anchovy stomach content, a total of 17 zooplankton species were identified. From the latter, the meroplankton included five taxonomic groups - Lamellibranchia veliger, Decapoda zoea and mysis, *Gastropoda veliger, Polychaeta larvae, Cirripedia cypris;* from crustacean copepods, the following species were recorded - *Acartia clausi, Pseudocalanus elongatus, Calanus euxinus, Centropages ponticus, Paracalanus parvus and Oithona spp*; the planktonic crustaceans Cladocera were represented by the species *Pleopis polyphemoides* and *Penilia* *avirostris;* class Chaetognatha – from species *Parasagitta setosa*. In anchovy food spectrum were discovered also Ostracoda, Appendicularia and Hydromedusae.

The indices of relative importance (IRI) of the main mesozooplankton representatives in the anchovy diet were represented in Table 4.9.4.

Table 4.9.4 The anchovy food composition of in 28.08-05.09.2016.

Зоопланктон	*N (%, от общата численост)*	*M (%, от общата биомаса)*	*FO - Честота на срещане*	*IRI - Индекс на относителна значимост*
Lamellibranchia veliger	58.52	30.70	66.67	5948.30
Decapoda zoea, mysis	15.83	28.31	39.39	1738.67
Acartia clausi+copepodit stages	6.48	8.78	35.48	541.42
Calanus euxinus+copepodit+naupli stages	3.71	11.25	12.90	192.98
Pseudocalanus elongatus+copepodit stages	2.22	2.05	9.68	41.33
Gastropoda veliger	2.18	0.24	30.30	73.33
Copepoda copepodit (III -V)	2.17	2.47	21.21	98.41
Copepoda adult	1.74	2.99	21.21	100.32
Ostracoda	1.38	6.78	30.30	247.25
Parasagitta setosa	1.08	3.08	9.09	37.81
Centropages ponticus	0.20	0.36	9.68	5.42
other	6.67	3.23	48.48	479.95
total	100%	100%		

The representatives of meroplankton (larvae of benthic species) - Lamellibranchia veliger, Decapoda zoea and mysis, and copepods: *Acartia clausi*, *Calanus euxinus* and Centropages ponticus had leading position in the anchovy diet (Table 4, Fig. 10). The larvae of benthic Lamellibranchia predominated the anchovy food by abundance, biomass and by frequency of occurrence (Table. 4.9, Fig.4.9.8).

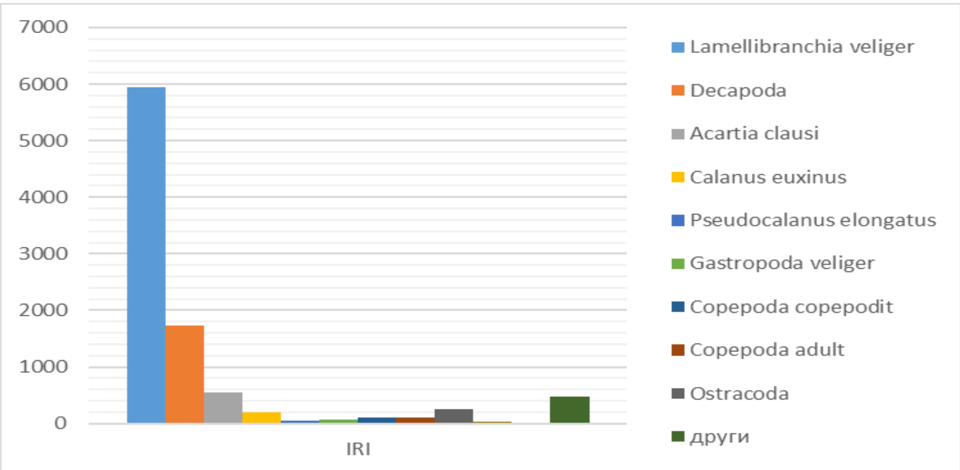


Figure 4.9.8. IRI of different mesozooplankton species in the anchovy diet during 28.08-05.09.2016.

At almost all investigated stations, located between 24-40 m isobaths, the anchovy diet was composed predominantly by meroplankton larvae of Lamellibranchia and Decopda. The other representatives of the fodder mesozooplankton created maximum 30 % by IRI (Table 4.9. 5).

Table 4.9.5. Distribution of IRI (%) of main mesozooplankton representatives in the anchovy food per stations in August/September 2016

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Zooplankton	Ст.2/40m	Ст.3/37m	Ст.4/27m	Ст.10/26m	Ст.11/24m
Lamellibranchia veliger	0.31	91.43	60.32	50.40	53.58
Decapoda zoea, mysis	99.39	1.14	9.89	39.37	38.75
Acartia clausi+copepodit stages	0.00	4.64	7.85	0.53	3.54
Calanus euxinus+copepodit+naupli stages	0.00	0.00	5.55	0.00	0.00
Pseudocalanus elongatus+copepodit stages	0.00	0.00	1.07	0.00	0.00
Gastropoda veliger	0.16	0.21	0.69	2.12	3.02
Copepoda copepodit (III -V)	0.00	0.00	3.98	0.00	0.00
Copepoda adult	0.14	0.28	3.73	0.00	0.00
Ostracoda	0.00	2.26	6.37	0.25	0.94
Parasagitta setosa	0.00	0.00	0.00	2.32	0.00
други	0.00	0.04	0.55	5.00	0.18

Parasitic Nematoda were discovered in 12 % of all investigated anchovy specimens.

Zooplankton in the marine environment: species composition and biomass

During the studied period the zooplankton biodiversity was formed by 21 species (Table 4.9.6).

Table 4.9.6. Species diversity of zooplankton.

	28.08 - 5. 09.2016
1.	*Noctiluca scintillans*
2.	*Pleurobrachia pileus*
3.	*Beroe ovata*
4.	*Mnemiopsis leidyi*
5.	*Hydromedusae*
6.	*Aurelia aurita*

7.	*Rotatoria spp.*
8.	*Penilia avirostris*
9.	*Evadne spinifera*
10.	*Evadne tergestiona*
11.	*Centropages ponticus*
12.	*Acartia clausi*
13.	*Paracalanus parvus*
14.	*Oithona davisae*
15.	*Polychaeta laravae*
16.	*Cirripedia nauplii, cypris*
17.	*Lamellibranchia veliger*
18.	*Gastropoda veliger*
19.	*Parasagitta setosa*
20.	*Decapoda zoea, mysis*
21.	*Oicopleura dioica*

In the total zooplankton biomass, as a sum of meso-and microzooplankton biomasses, the warm-water Cladocera showed dominating position with share 38.5 %, followed by the species *Beroe ovata* - 30%; copepods - 23.2 %, Appendicularia – 2.7% and *Parasagitta setosa* - 2.4 % (Table 4.9.6., Fig. 4.9.9.).

By abundance, the dominating group were copepods, and mainly the species *Oithona davisae*, that consisted 61% of the total zooplankton abundance. The species *Oithona davisae* is characterised by small sizes and low individual weight, thus, in spite of the high abundance, the copepods created a relatively low share of the total zooplankton biomass.

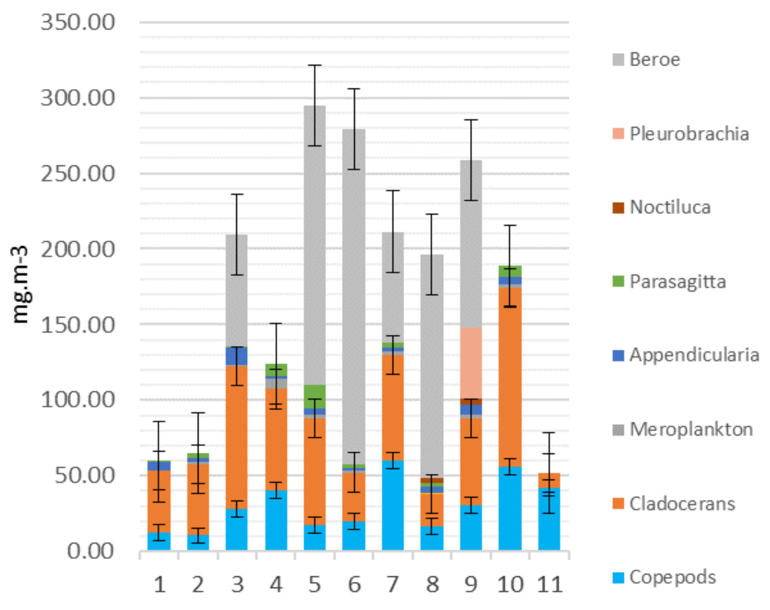
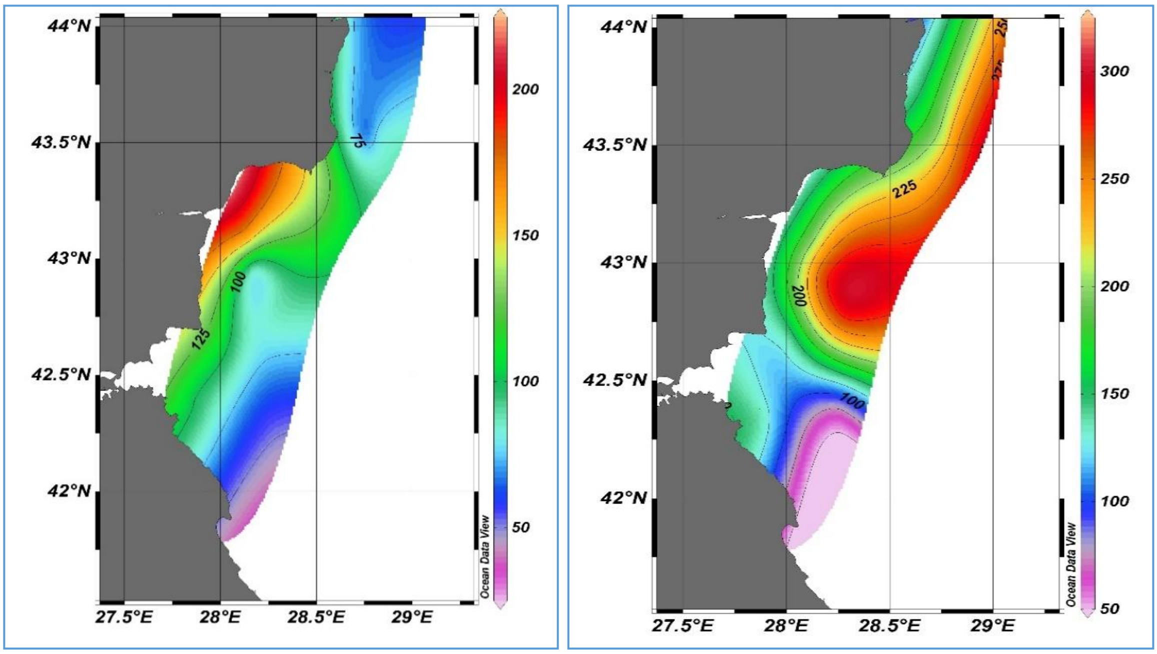


Fig. 4.9.9. Distribution of the biomass of the main mesozooplankton groups (mg.m-3) per stations during 28.08 - 5.09.2016. (The variability of biomasses of copepods, Cladocera and Beroe ovata was shown with error bars).

Table 4.9.7. Percentage distribution (%, from total biomass) of main mesozooplankton groups - Copepoda, Cladocera, Meroplankton, Rotatoria, Appendicularia, Noctiluca, Parasagitta setosa, Pleurobrachia, Beroe per stations during 28.08 - 5.09.2016.

Stations	*Copepods*	*Clad*	*Mero*	*Rot*	*Append*	*Parasag*	*Noct*	*Pleuro*	*Beroe*	*Zooplankton biomass ($mg.m^{-3}$)*
1	20.9	69.3	0.6	0.0	8.6	0.7	0	0	0.0	59.27
2	16.1	72.0	2.9	0.0	3.5	5.5	0	0	0.0	65.32
3	13.5	44.9	0.4	0.0	5.5	0.4	0	0	35.3	209.20
4	32.3	54.1	5.8	0.0	1.0	6.8	0	0	0.0	124.06
5	5.8	24.1	0.6	0.0	1.6	5.3	0	0	62.6	294.84
6	7.1	11.6	0.4	0.0	0.8	0.9	0	0	79.2	279.41
7	28.5	33.0	0.9	0.0	1.3	1.4	0	0	34.9	211.42
8	8.4	10.7	0.4	0.1	2.2	1.3	1.85	0.00	75.1	196.57
9	11.7	22.3	0.9	0.0	2.5	0.2	1.48	18.20	42.8	258.78
10	29.6	62.6	1.2	0.0	2.7	3.8	0	0	0.0	188.70
11	80.8	19.2	0.0	0.0	0.0	0.0	0	0	0.0	51.78
Total	**23.2**	**38.5**	**1.3**	**0.0**	**2.7**	**2.4**	**0.3**	**1.7**	**30.0**	**176.23**

The total zooplankton biomass reached 176.30 ± 88.67 (SD) mg.m-3, while the mean biomass of fodder zooplankton attained 93.49 ± 44.28 (SD) mg.m-3. The maximal biomass of the fodder zooplankton was registered in the coastal sector - between 20-50 m isobaths - southern from the Cape Kaliakra and up to Obzor and Cape Emine (Fig. 4.9.10). Meanwhile, the microzooplankton species were detected mainly in the area between 25 -100 m isobaths (Fig. 4.9.10).



1. 2.

Figure 4.9.10. Spatial distribution of the fodder mesozooplankton biomass (mg.m-3) (1) and the total zooplankton biomass (meso- + macrozooplankton (mg.m-3) (2) in 28.08 - 5.09.2016.

4.10. Food composition and feeding indexes of sprat (Sprattus sprattus, L) in autumn of 2016

Material and method

The current study of sprat diet was based on analysis of stomach content composition of 91 sprat specimens, collected in front of Bulgarian Black Sea coast during 23 – 26.12.2016. Additionally, the study included analysis of the mesozooplankton species composition, abundance and biomass in the environment, as these pelagic organisms present the main food source for planktivorous fish species.

The coordinates and information about the sampling sites were presented at Table 4.10.1.

Table 4.10.1. Information on the investigated stations in 23 – 27.12.2016.

Date	№	Coordinates	Depth (m)	Zooplankton stations	Sprat food
23.12.2016	1	42.67 N - 27.83 E	26	Zoo1	F1
23.12.2016	2	42.65 N - 27.79 E	24	Zoo2	F2
23.12.2016	3	42.69 N - 27.77 E	25	Zoo3	F3
24.12.2016	7	42.65 N - 27.75 E	25	Zoo4	F4
26.12.2016	16	43.27 N - 28.54 E	55		F5
26.12.2016	17	43.31 N - 28.57 E	68	Zoo5	F6
26.12.2016	19	43.39 N - 28.59 E	46	Zoo6	F7
26.12.2016	22	43.37 N - 28.26 E	16	Zoo7	F8
27.12.2016	33	43.36 N - 27.96 E	48	Zoo8	F9

For the purposes of investigation, about 10 - 11 fish specimens per trawl catch were separated and preserved in 10 % formaldehyde: seawater solution. The absolute length (TL, to the nearest 0.1 cm) and weight (to the nearest 0.01 g) of fish specimens were measured. Under laboratory conditions the stomachs of the selected animals were weighted with analytical balance (to the nearest 0.0001 g). The food mass of each specimen has been calculated as a difference between the weights of full and empty sprat stomach.

The stomach content was investigated under microscope for estimation of species composition and prey number. The prey biomass was estimated by multiplication of the number of consumed mesozooplankton species by their individual weights.

The following indices were calculated: stomach fullness index (ISF) as a percent of body mass: (stomach content mass/fish mass) *100; and index of relative importance - IRI, Pinkas et al. (1971): IRI = (N+M) x FO; where N - the proportion of prey taxa (species) in the diet by numbers (abundance); M - the proportion of prey taxa (species) in the diet by mass; FO- frequency of occurrence among fish.

The zooplankton samples were gathered from the whole water layer (bottom-surface) with a plankton set (opening diameter d=36 cm; mesh size 150 μm). The samples were fixed on board ships with 4% formaldehyde: seawater solution (Korshenko & Aleksandrov, 2013). The mesozooplankton species composition has been identified by Guides for the Black and Azov Seas (Morduhai-Boltovskii et al., 1968), and its quantity - by the method of Bogorov (Korshenko & Aleksandrov, 2013).

Length - weight relationship, index of stomach fullness (ISF)

The analysis of sprat feeding indexes was based on data, obtained after processing of 91 fish samples.

The mean absolute length of investigated specimens reached 85.22 ± 0.37 (SD) mm, varying between 78 - 96 mm, correspondingly the mean weight was 3.87 ± 0.55 (SD) g, varying from 2.92 g. to 5.26 g (Table 4.10.2, Fig 4.10.1).

Table 4.10.2. Summary statistics of length (L, cm) and weight (W, g) of sprat analysed for stomach content composition during XII.2016.

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	L, cm	W, g
Mean	8.52	3.87
Standard Error	0.04	0.06
Median	8.50	3.72
Mode	8.50	3.14
Standard Deviation	0.37	0.55
Sample Variance	0.14	0.30
Kurtosis	0.69	-0.74
Skewness	0.69	0.37
Range	1.80	2.34
Minimum	7.80	2.92
Maximum	9.60	5.26
Sum	775.50	351.88
Count	91	91
Confidence Level (95.0%)	0.08	0.11

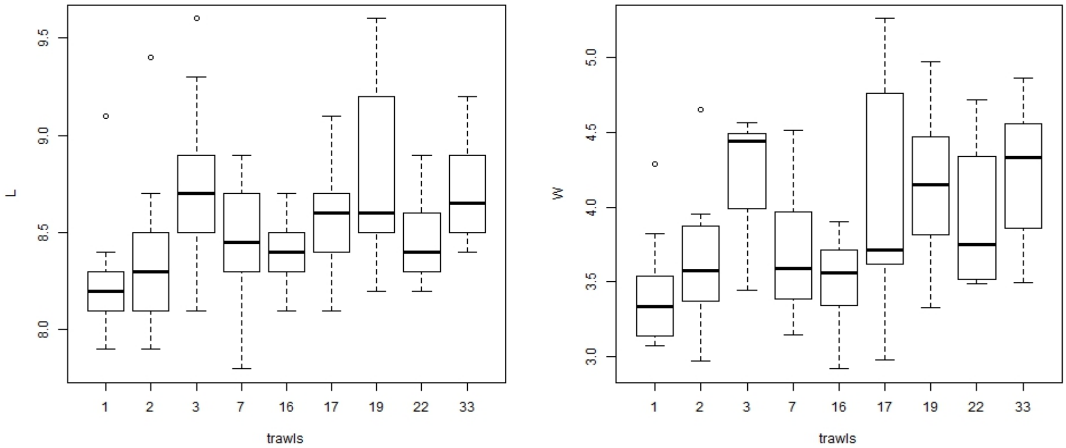


Figure 4.10.1.. Box plot: Distribution of length (cm) and weight (g) of sprat (included in stomach content composition analysis) per trawls (median values, 25 – 75 % hinge, minimal and maximal values) during XII.2016.

The length-weight dependence, established by this study, was described by the following equation: Log WW (g)=2.3476*log L(cm) – 1.6005; (R^2=0.52, p<0.001, Fig.4.10.2).

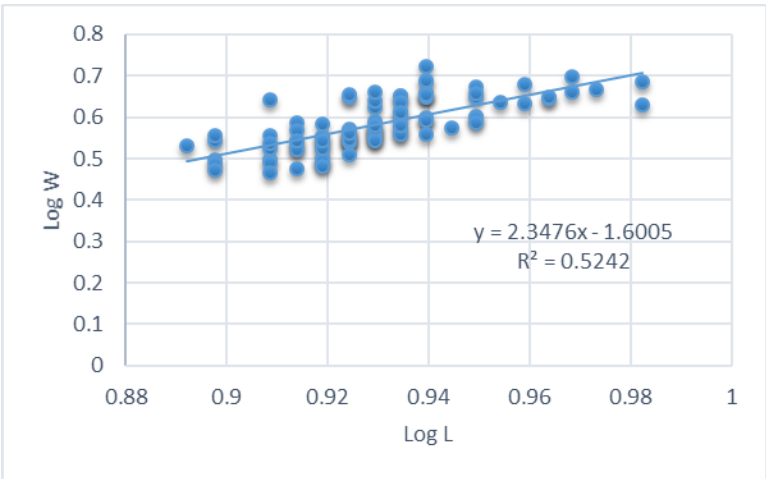


Figure 4.10.2. Length – weight relationship for sprat, investigated in XII.2016.

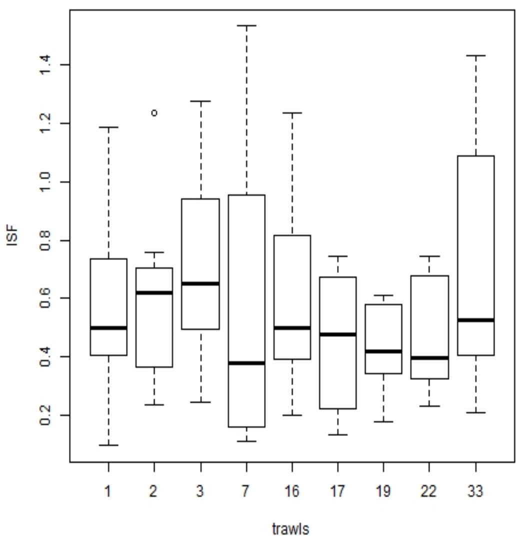
In autumn 2016, the mean index of stomach fullness (ISF) attained 0.56 % ± 0.32 (SD) of sprat body weight (BW) (Table 4.10.3.), nearing the multiannual average for 2007-2010 - 0.52 % (Mihneva et al., 2015. However, this value of ISF has decreased 1.5 times in comparison to the average ISF in August 2016.

Table 4.10. 3 Summary statistics of data about index of stomach fullness - ISF (% от BW) of sport during XII.2016.

Summary statistics of data about index of stomach fullness - ISF (% от BW) of sport during XII.2016.

	ISF, % of BW
Mean	0.56
Standard Error	0.03
Median	0.51
Standard Deviation	0.32
Sample Variance	0.10
Kurtosis	0.41
Skewness	0.88
Range	1.44
Minimum	0.10
Maximum	1.53

The highest average values of ISF = 0.7 % (Fig. 4.10.3 and 4.10.4) were determined in samples from trawls 3 and 33, located near to the cape Emine and Ahtopol. In general, along the southern costs, the ISF has reached 0.5 - 0.7% of sprat body weight, while in the northern sector, ISF formed ~0.43 % of BW. The mean values of ISF were homogenously distributed among the observed stations, indicating comparable levels of zooplankton consumption and/or similarity of food base quantity and composition.



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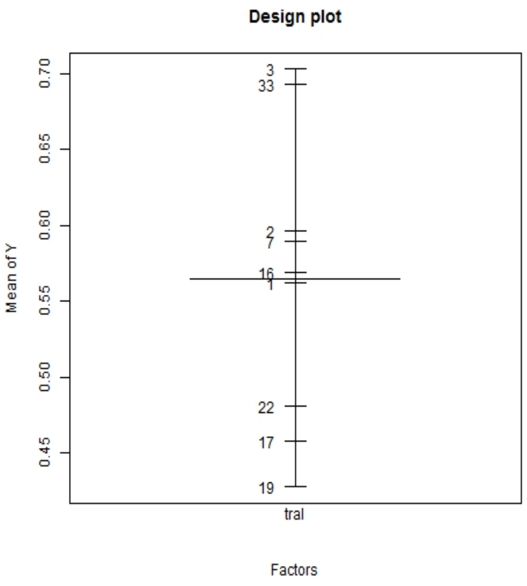

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Figure 4.10.3. Box plot: Spatial distribution of ISF (1) per trawls during 23 - 27.12.2016 г. Design plot (2): distribution of the mean ISF values per trawls.

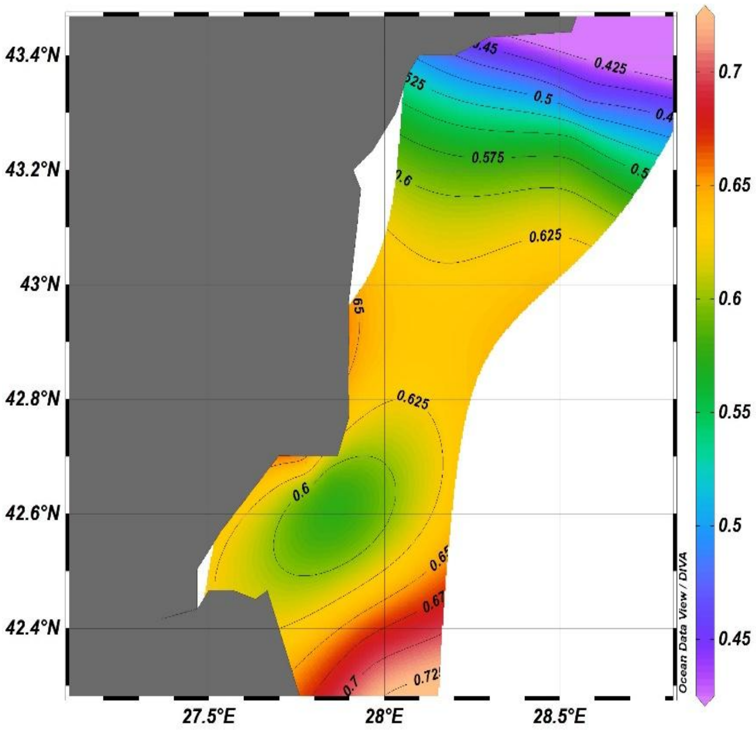

Fig. 4.10.4. Spatial distribution of mean ISF in 23-27.12.2016.

Among the ISF and sprat weight (within the limits of 2.92 - 5.26 g) was not established statistically significant correlation (Fig.4.10.5).

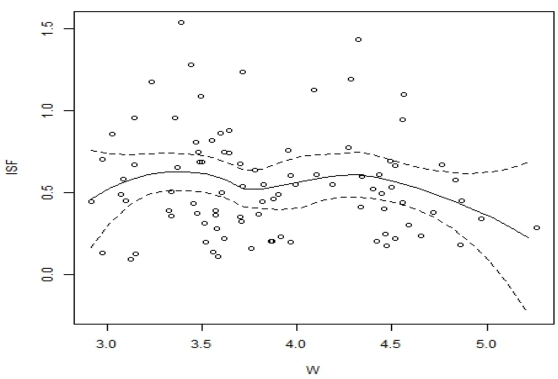

Figure 4.10.5. Scatterplot: relationship between sprat weight (W, g) and ISF in XII.2016 г.

Prey number, species composition of sprat food and index of relative importance (IRI)

In 98.72% of all investigated sprat specimens, a presence of fodder mesozooplankton was found, which allow quantitative and qualitative identification through the microscope observations, i.e. it is possible to estimate the number and the species composition of the prey items. By 1.28 % of all sprat specimens, only food remains were discovered, which didn't allow quantitative analysis.

The maximal individual number of prey items (PN) - 846 ind/stomach was found in specimen, collected from station F5 (located at 55-m depth in front of Balchik), where the mean prey number attained 391 ind/stomach. The maximal average value of PN – 458 ind/stomach was registered at st. F9 - located in front of Ahtopol with depth 48 m. Among the most of observed stations, the average PN values ranged between 300-350 ind/stomach, but highest values - 400-450 ind/stomach were established at depths 45 -55 m (Fig. 4.10.6)

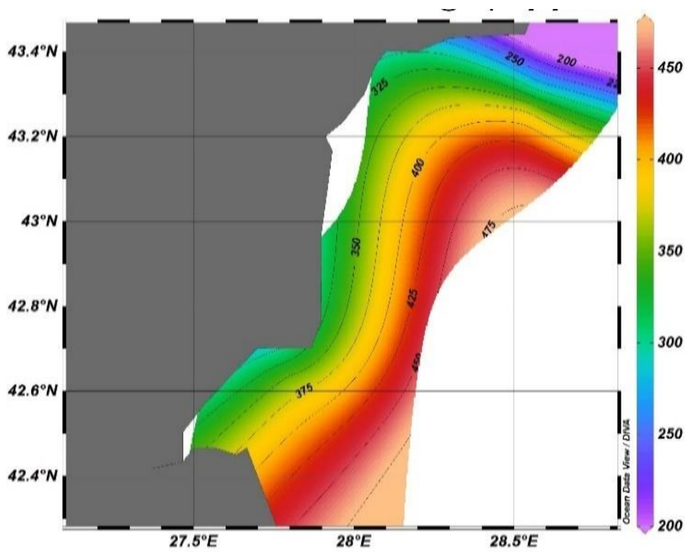

Fig. 4.10.6. Spatial distribution of average prey number per stations in XII.2016.

In December 2016, the average PN attained 328 ind/stomach. For comparison, through the interval 2007-2010, the mean PN reached 64 ind/stomach. Based on the increased value of PN in Dec. 2016, by ISF near to the multiannual average, we can deduce that sprat was feeding predominantly on small-sized zooplankton.

Analysis of the zooplankton samples, gathered from the marine environment, showed that the total diversity comprised of 14 mesozooplankton species/groups. From the latter, 12 species/groups were found out in the sprat diet. From copepods -

the species *Pseudocalanus elongates, Calanus euxinus, Acartia clausi, Oithona spp. and Paracalanus parvus* were most often identified in the sprat diet; from the group of pelagic larvae of bottom species (meroplankton) - three taxonomic groups were established - *Lamellibranchia veliger, Decapoda zoea, Cirripedia nauplii* and cypris; planktonic crustaceans Cladocera were represented by genera Pleopis; class Chaetognatha - from species *Parasagitta setosa*. Presence of Pisces ova was also registered in the sprat food content.

The indices of relative importance (IRI) of the main mesozooplankton representatives in sprat food spectrum (based on the percent shares from total abundance and biomass, and frequency of occurrence in samples) were represented in Table 4.10.4.

Table 4.10.4. The sprat food composition in 23-27.12.2016

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	N (%, from total abundance)	*M (%, from total biomass)*	*FO – Frequency of occurrence*	*IRI – Index of relative importance*
Pseudocalanus elongatus+copepodit stages	7.81	1.65	79.49	751.98
Acartia clausi+copepodit stages	20.45	27.73	89.74	4323.67
Copepoda copepodit (III -V)	20.29	33.94	91.03	4936.56
Calanus euxinus+copepodit+naupli stages	0.82	10.93	61.54	723.10
Copepoda adult	2.46	3.23	26.92	153.17
Lamellibranchia veliger	32.86	7.51	93.59	3778.23
Cirripedia nauplii + cypris	8.18	4.15	73.08	901.08
Pleopis polyphemoides	4.64	2.33	61.54	428.93
Oithona spp.	1.06	0.17	33.33	41.00
други	1.43	8.36	33.33	326.30
total	100%	100%		

The sprat food was dominated by copepods - Acartia clausi and copepodid stages (III-V) (that couldn't be identified to species level due to partly decomposition) and meroplankton larvae of Lamellibranchia (Table. 4.10.4, Fig.4.10.7). The eurytherm species predominated in frequency of occurrence, as well as in abundance and biomass of food.

The sprat food composition show slightly pronounced differences among the observed stations (Tabl. 4.10.5.). In front of the southern costs, the sprat ration consisted mainly of A. clasui, Copepoda copepodit and Pseudocalanus elongatus (stations F1, F2, F3, F4, F9), while L. veliger, A. clasui and Calanus euxinus were mainly detected in the sprat diet within the zone Albena -Kaliakra (stations F5, F6, F7, F8).

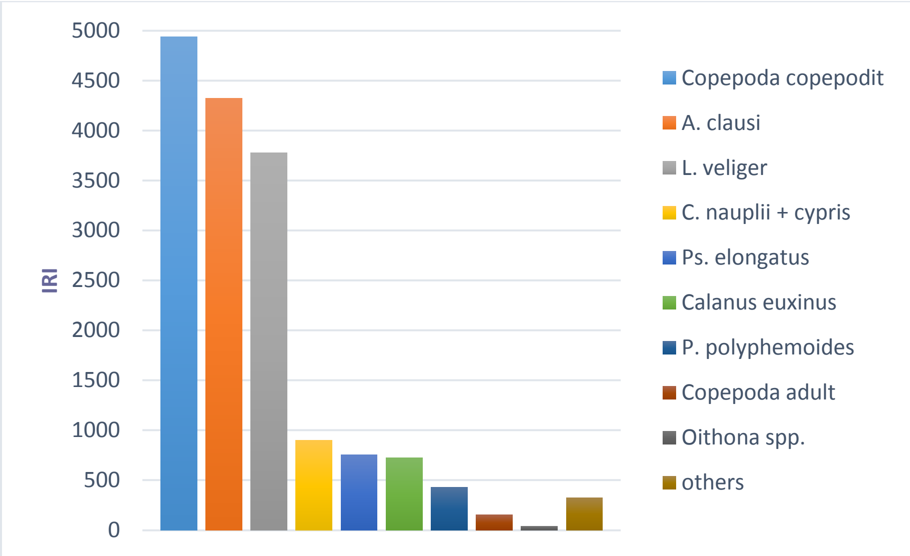


Figure 4.10.7. IRI of different mesozooplankton species in the sprat food in 23-27.12.2016.

Table 4.10.5. Distribution of IRI (%) of main mesozooplankton representatives in sprat food per observed stations in XII.2016.

Distribution of IRI (%) of main mesozooplankton representatives in sprat food per observed stations in XII.2016.

Zooplankton groups/species	F1 26m	F2 24m	F3 25m	F4 25m	F5 55m	F6 68m	F7 46m	F8 16m	F9 48m
Ps.elongatus+copepodit stages	10.59	6.88	7.91	3.78	3.44	3.20	5.50	3.31	7.99
A. clausi+copepodit stages	31.1	19.3	28.1	37.5	11.0	14.0	18.8	30.3	22.1
C. copepodit (III -V)	23.14	40.17	36.27	16.23	23.85	21.33	31.75	27.16	37.12
C.euxinus+copepodit+naupli stages	3.36	0.87	2.78	3.63	4.17	5.39	3.27	6.15	1.52

C. adult	0.56	2.07	1.37	0.00	2.22	2.62	0.00	0.14	0.08
L. veliger	26.03	25.06	15.56	26.03	25.55	44.66	31.77	23.42	20.15
C.nauplii + cypris	2.78	3.49	5.51	11.13	3.03	6.63	5.35	5.49	5.70
P.polyphemoides	1.66	1.95	0.59	1.57	0.00	1.88	2.82	3.09	4.43
Oithona spp.	0.25	0.09	0.00	0.13	0.00	0.13	0.63	0.61	0.68
други	0.53	0.12	1.91	0	26.74	0.16	0.11	0.33	0.23

Parasitic Nematodes were discovered by 3.84 % of a total number of investigated sprat specimens.

Zooplankton in the marine environment: species composition and biomass

During the studied period, the zooplankton was presented by 14 species (Table 4.10.6). Usually, in autumn, the zooplankton species diversity has been reduced in comparison to the summer months, due to lack of warm water species that form diapause eggs for overwintering.

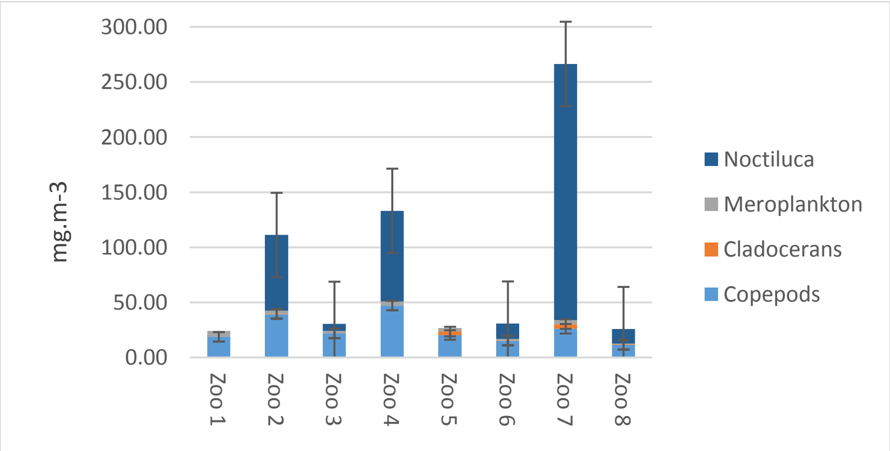
Table 4.10.6 Species diversity of zooplankton.

	23 - 27. 12.2016
1.	*Noctiluca scintillans*
2.	*Tintinopsis spp.*
3.	*Hydromedusae*
4.	*Aurelia aurita*
5.	*Acartia clausi*
6.	*Pseudocalanus elongatus*
7.	*Calanus euxinus*
8.	*Paracalanus parvus*
9.	*Oithona davisae*
10.	*Oithona similis*

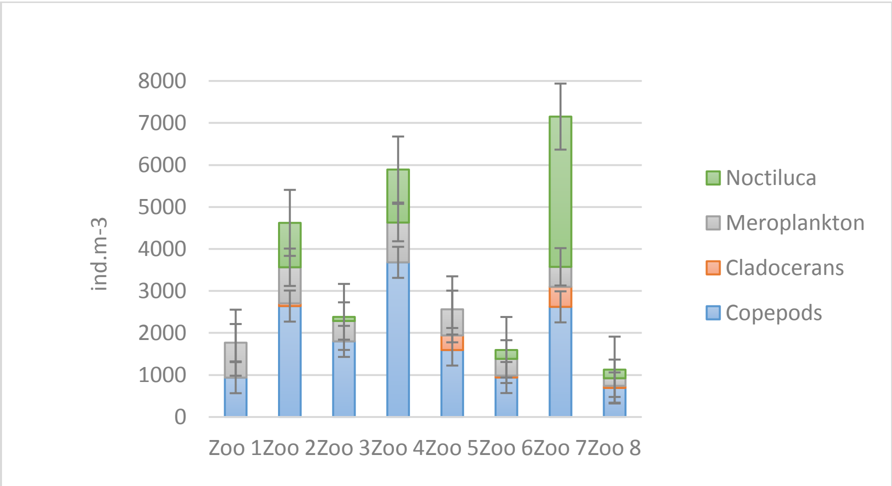
11.	*Pleopis polyphemoides*
12.	*Cirripedia nauplii, cypris*
13.	*Lamellibranchia veliger*
14.	*Pisces ova*

The total zooplankton biomass was mainly formed by two groups - Protozoa - creating 64.31 % and Copepods, with share - 30.78 %, while percent shares of Cladocera and Meroplankton were negligible - 1.37 % and 3.54 % (Fig. 4.10.8, Table 4.10.7)

Protozoa and copepods were dominating groups by abundance and the species Oithona davisae comprised 47 % of the total zooplankton abundance.



A



B

Figure 4.10.8. Distribution of biomass (A) and abundance (B) of main zooplankton groups per stations during 23 - 27.12.2016.

Table 4.10.7. Percent shares (%, of biomass) of main zooplankton groups - Copepoda, Cladocera, Meroplankton, Noctiluca (Protozoa), per stations during 23 - 27.12.2016.

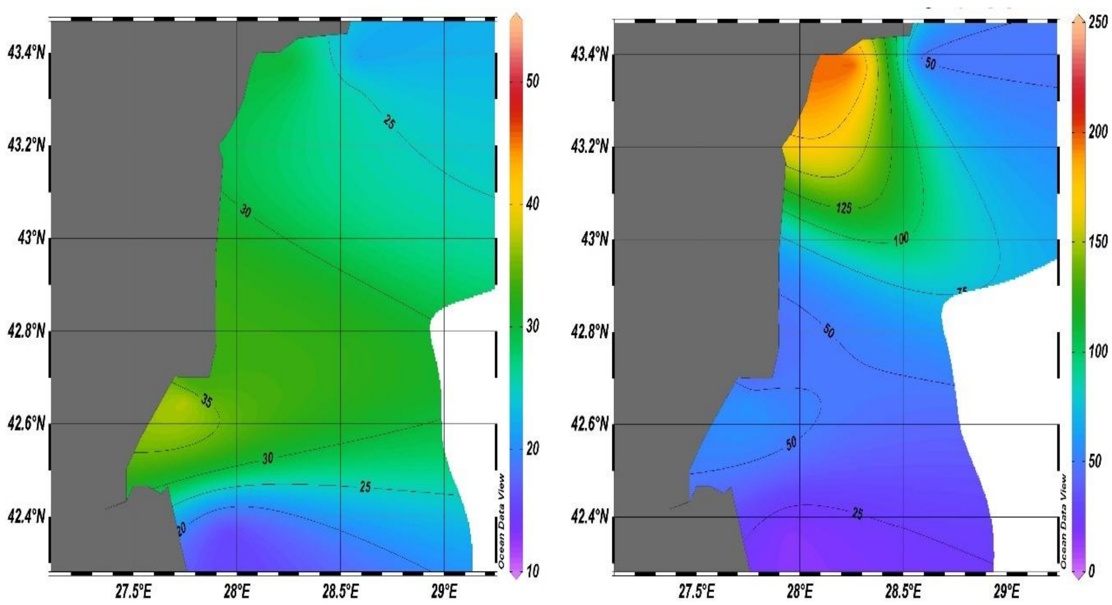
Станции	*Copepods*	*Cladocera*	*Meroplankton*	*Noctiluca*	*Total zooplankton biomass (mg.m^{-3})*
Zoo1	77.88	0.00	22.12	0.00	24.00
Zoo2	35.30	0.53	2.32	61.85	111.09
Zoo3	71.59	0.00	7.67	20.74	30.40
Zoo4	35.43	0.00	2.85	61.73	132.99
Zoo5	75.96	11.66	12.38	0.00	26.79
Zoo6	49.28	1.38	4.57	44.76	30.64
Zoo7	9.75	1.61	1.34	87.30	266.41
Zoo8	43.87	1.79	2.41	51.94	25.66

The total zooplankton biomass averaged at 81.00 ± 86.47 (SD) mg.m^{-3} and the biomass of Protozoa reached 52.09 ± 79.51 (SD) mg.m^{-3}. The biomass of fodder mesozooplankton could be characterized as relatively low for the season and reached 28.91 ± 12.87 (SD) mg.m^{-3}. Table 8 shows the summary statistics of total zooplankton biomass and the main groups, contributing to it - mesozooplankton - Copepoda, Cladocera, Meroplankton and Protozoa/Noctiluca.

Table 4.10.8. Summary statistics of zooplankton biomass - by groups and total, XII.2016.

	Copepods	*Cladocerans*	*Meroplankton*	*Noctiluca*	*TZB*	*MZB*
Mean	**24.93**	**1.11**	**2.86**	**52.09**	**81.00**	**28.91**
Standard Error	4.33	0.58	0.52	28.11	30.57	4.55
Median	21.06	0.44	2.95	13.52	30.52	25.44
Standard Deviation	12.25	1.65	1.47	79.51	86.47	12.87
Sample Variance	150.12	2.72	2.15	6321.11	7476.44	165.69
Kurtosis	0.12	0.84	0.04	4.41	2.65	-0.34
Skewness	1.03	1.51	0.07	2.05	1.70	0.60
Range	35.86	4.29	4.69	232.57	242.42	38.57
Minimum	11.25	0.00	0.62	0.00	24.00	12.33
Maximum	47.11	4.29	5.31	232.57	266.41	50.90
Sum	199.46	8.89	22.91	416.72	647.98	231.26

The analysis of spatial distribution of mesozooplankton biomass (without *N. scintillans*) show rather low levels - 25-30 mg.m-3, with slightly increasing values in southern direction (Fig. 4.10.9.). “Bloom” of *N. scintillans* (Protozoa) with biomass 150-250 mg.m-3 was established in the zone Kalikara - Kamchia (Fig. 4.10.9.).



1. 2.

Figure 4.10.9. Spatial distribution of the biomasses of mesozooplankton (1, without *N. scintillans*, $mg.m^{-3}$) and *N. scintillans* (2, $mg.m^{-3}$) during 23 - 27.12.2016.

Correlation relationships

Table 4.10.9. Correlations between measured parameters per stations: average biomass of fodder mesozooplankton (MZB), sprat length (L) and weight (W), index of stomach fullness (ISF) and prey number (PN).

	MZB	L	W	ISF	PN
L	-0.60 (p=-.02)	1			
W	-0.56 (p=-.05)	0.96 (p=-.0001)	1		
ISF	-0.04	0.14	0.09	1	
PN	0.01	-0.26	-0.25	0.64	1

Among MZB and average sprat length and weight were established statistically significant negative correlations (df = 8, p< .05, Table 4.10.9), indicating predatory effect on the mesozooplankton quantity.

5. Predictions and possibilities for exploitation

As it could be seen from Fig.5 the Catch numbers in December is 3 times higher than those in August. The highest numbers is accounted for 3-3+ old individuals in December.

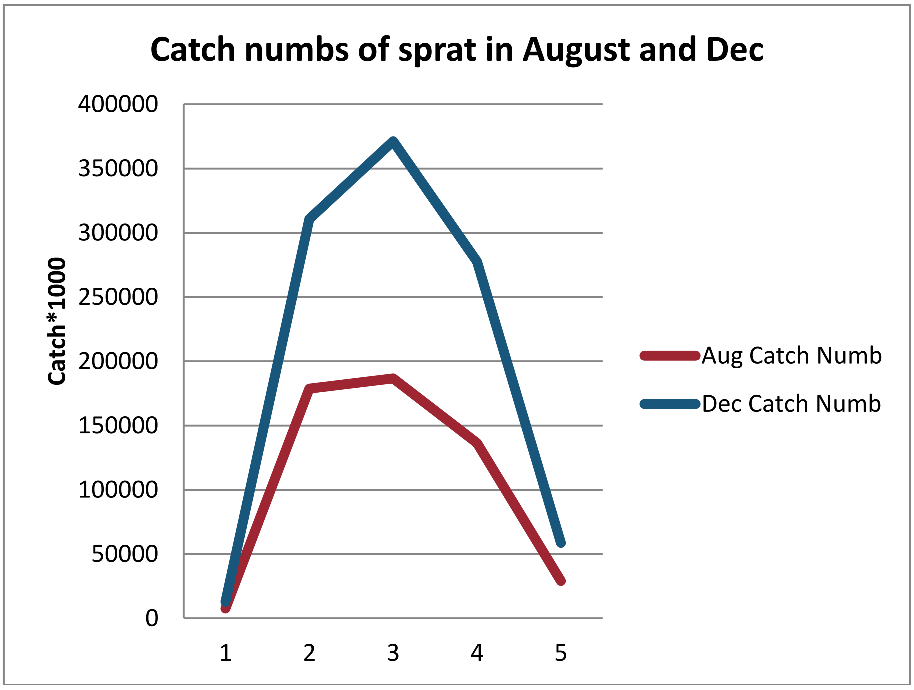


Fig.5 Catch numbers in August and December 2016 of sprat related to the age

Equilibrium state of the sprat stock

Equilibrium yield and related biomasses of sprat from Bulgarian and Romanian Black Sea waters have been presented graphically on *Fig*.5.1.1. On first graphic Equilibrium Yield with Confidence intervals (uncertainties) showed very low values at CI_{med} and $CI_{2.5\%}$. Y/R with $CI_{97.5\%}$ reached its maxima and corresponded to fishing mortality at around 1.16 then plateau of the curve follows and F_{max} determination became impossible.

It is evident that levels above F=0.8 would lead to stock collapse. Sustainable levels of fishing mortality are those around F=0.5, which would correspond to the catch level of 12.5 thous.tons of sprat in Bulgarian and Romanian marine waters.

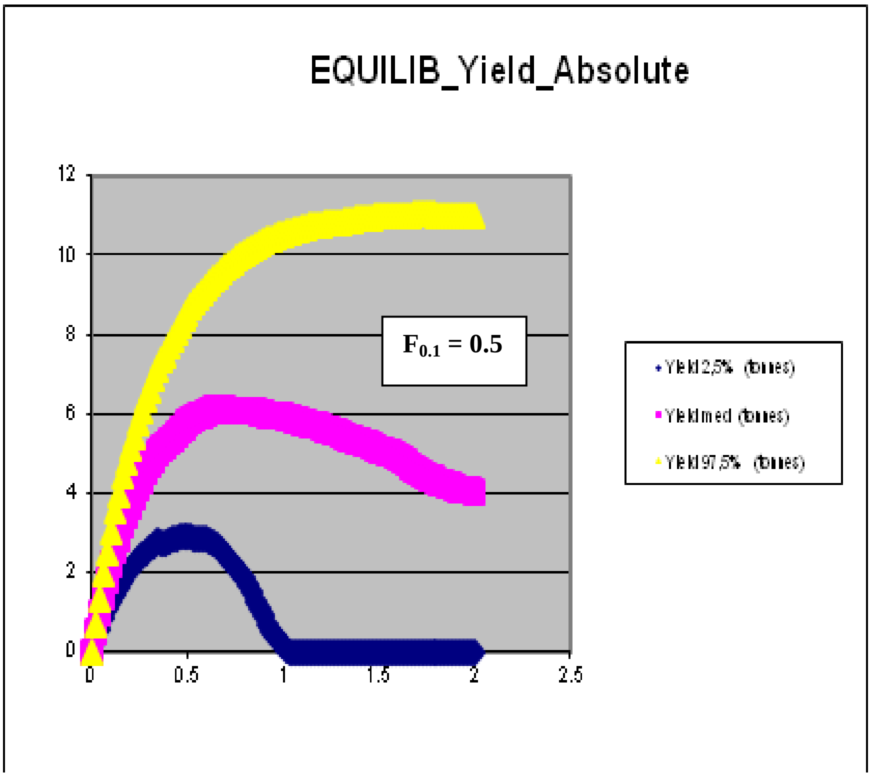


Figure 5.1.1.Equilibrium yield with CI. Optimal level of fishing mortality and corresponding catch of sprat from Bulgarian and Romanian waters.

Spawning stock biomass, fishable biomass and total biomass followed similar trend of steep decrease, since only values with $CI_{97.5\%}$ have relatively high values at corresponding lowest fishing mortality. Hence, with increasing fishing mortality all tested biomasses (*Fig*.5.1.2, *Fig*.5.1.3, and *Fig*.5.1.4.) followed diminishing trend, as after F=0.8 (at $CI_{2.5\%}$) and after 1.16 (at CI_{med}) the sprat stock would decrease below unsustainable levels – *Fig*.5.1.1.

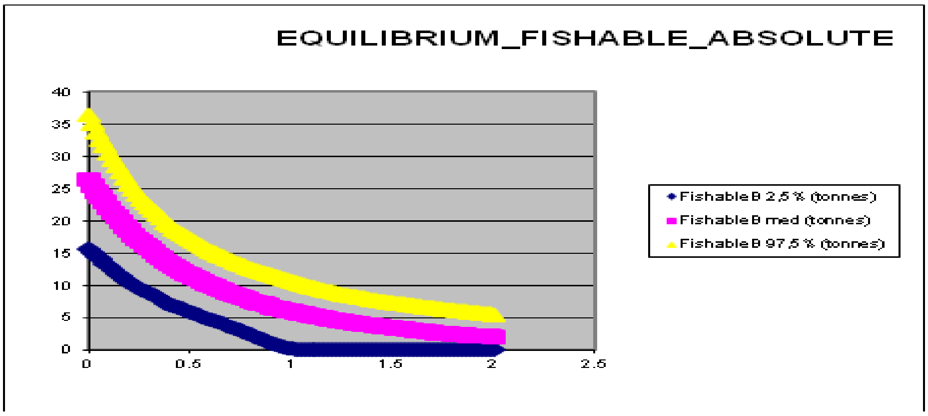


*Figure 5.1.2.*Equilibrium Fishable Biomass

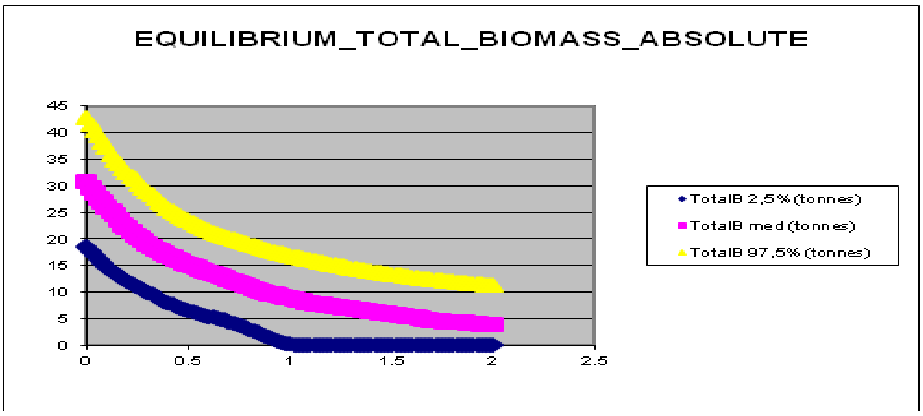


*Figure 5.1.3.*Equilibrium Total Biomass

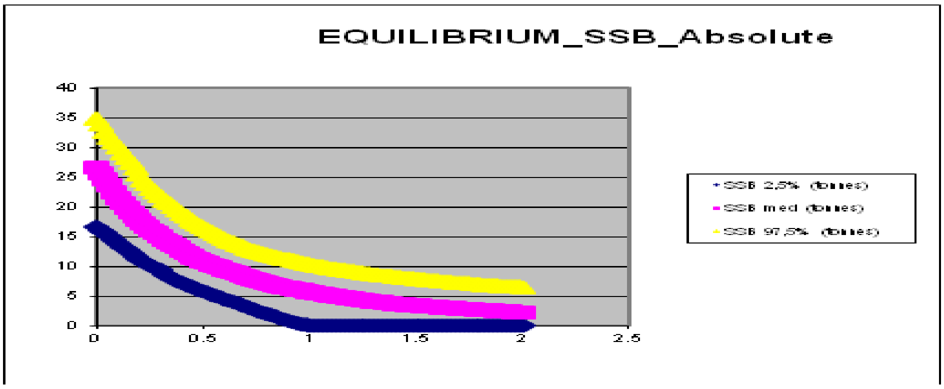


Figure 5.1.4 .Equilibrium SSB

Recruitment is highly affected by fishing mortality and after F= 0.5 drops very steeply – Fig.5.1.5.

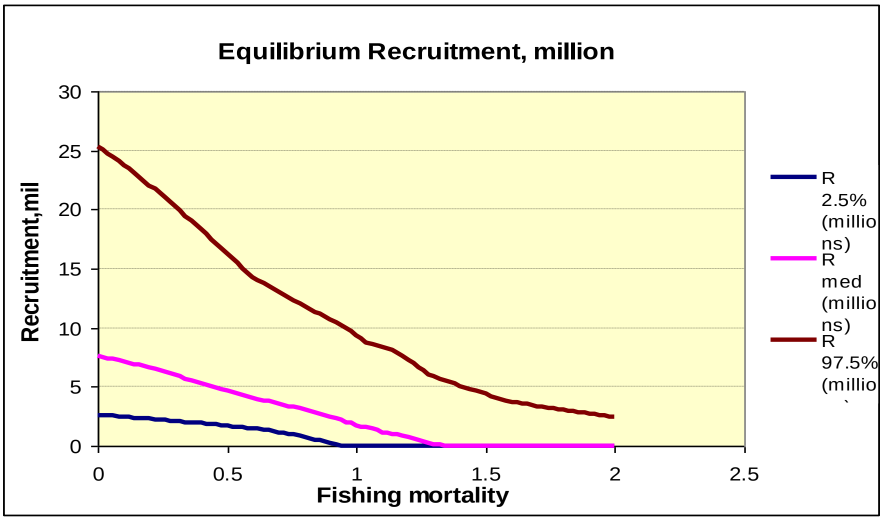
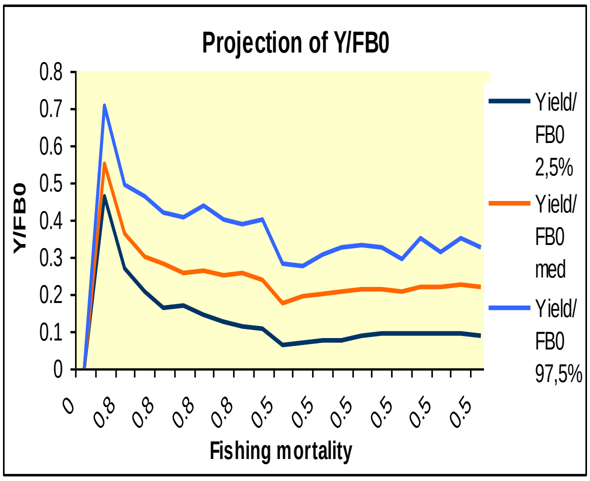
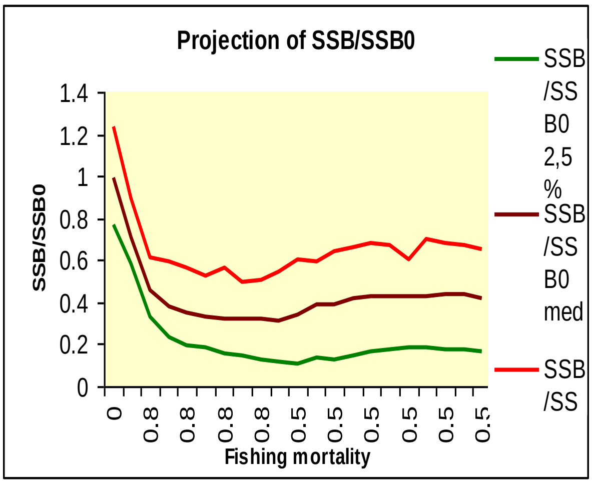


Fig.5.1.5. Equilibrium Recruitment

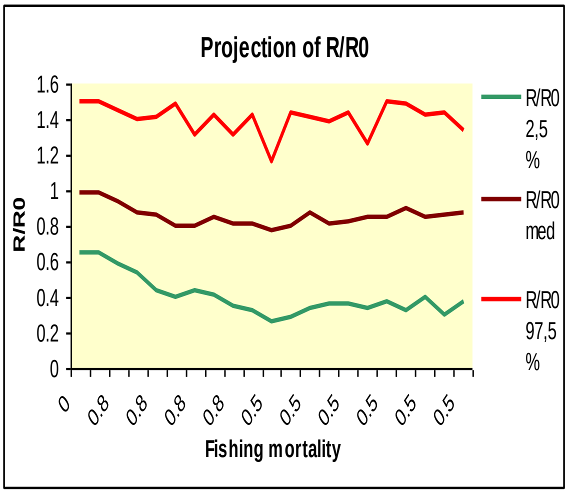
6. Prediction model of the stock parameters in relation with fishing mortality variation for 10 years

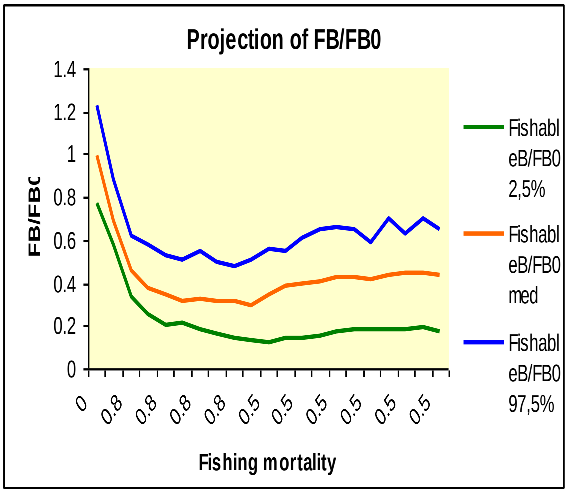


A)



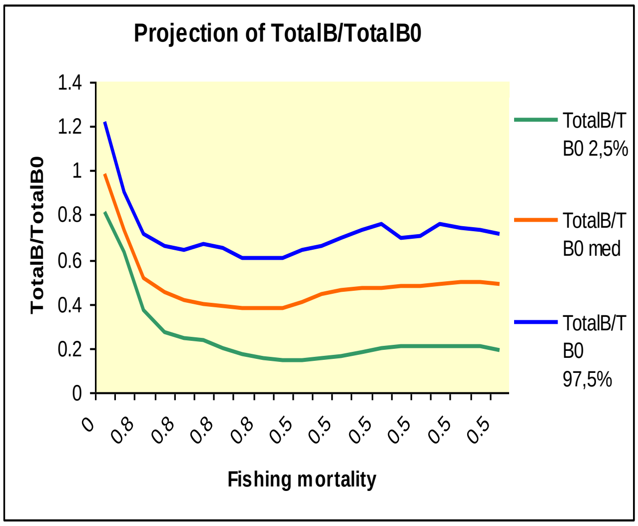
B)





C)

D)



E)

Figure. 6.1. Predictions of the stock parameters of sprat, related to fishing mortality Unexploited state

The relative yield (Y/F_0) at very low levels of the fishing mortality is high during the first projected year (Fig.6.1. A). At F = 0.8, in the second projected year fall of the relative yield was detected up to the levels of F = 0.5 (Fig. 6.1, A). After the fifth projected year, in all tested confidence intervals, plateau of the relation Y/F0 was observed. Similar is the case with the relative (SSB/SSB_0) (Fig. 6.1, B), and even slight increase at CI 97.5% and SSB/SSB_0 med, after change of fishing mortality (from F = 0.8 to F = 0.5). Recruitment (Fig. 6.1, C) is stable, none influenced by the changes in fishing mortality. Fishable and total biomass, represented as relation with the biomass at unexploited state, show similar trends with those of relative SSB (Fig. 6.1, D, E).

variations.

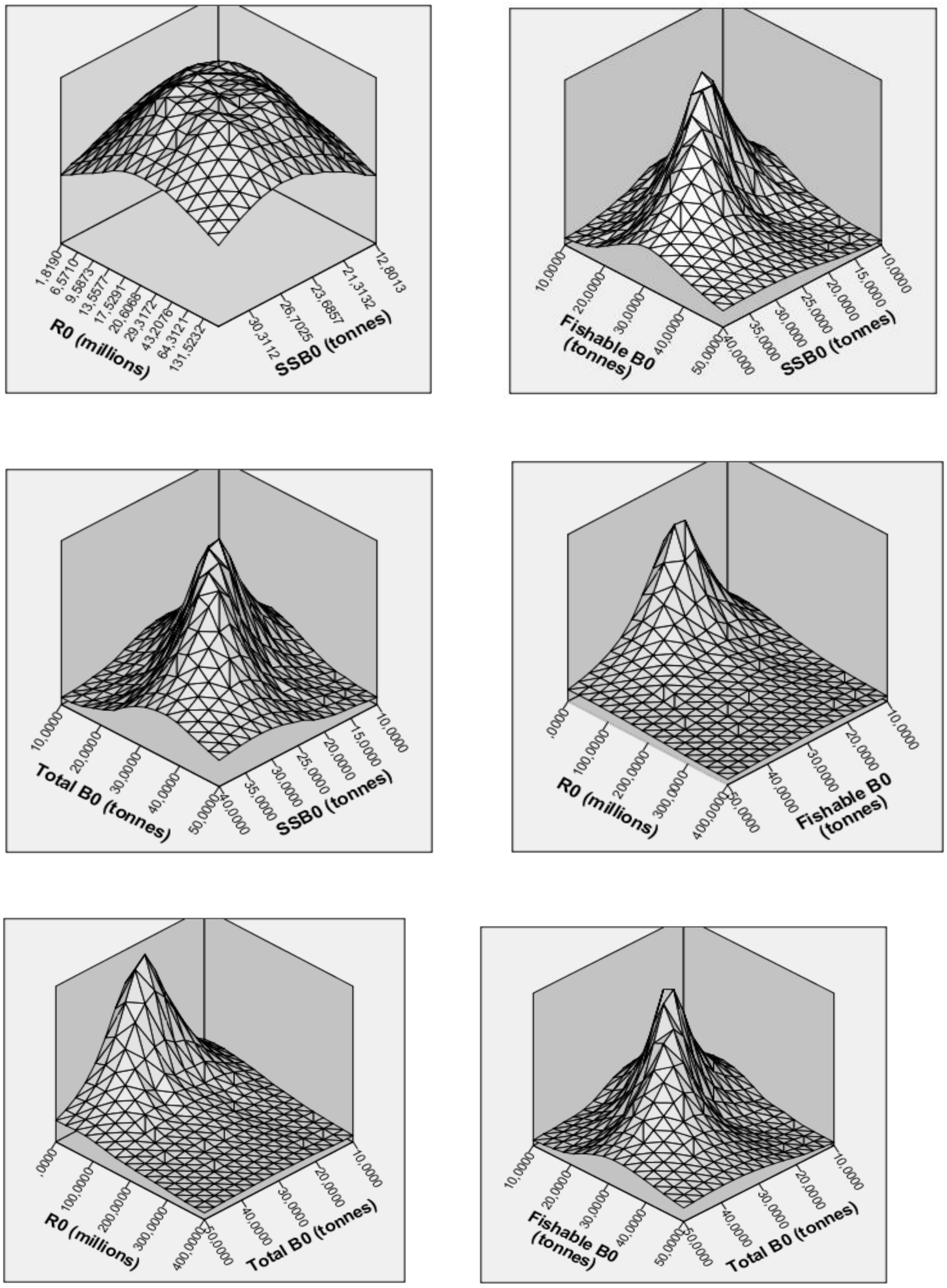


Figure. 6.2. Unexploited state

7. Maximum sustainable yield

Maximum sustainable yield (MSY) by countries, according to the method of Gulland (1970), was calculated for both exploitation biomasses – in Bulgarian and in Romanian waters. In the present research we used the natural mortality coefficient M=0.95 (Ivanov and Beverton, 1985; Prodanov et al., 1997; Daskalov, 1998).The results obtained are given on Tabl.7.1.

Table.7.1. Biomass (tons) by countries and MSY

	Biomass	MSY (t)	
Country BG	(t)	Gulland	BH steepness, $F_{0.1}$
August-September	21 090	10 545	
December	32 278	16 139	11 500

The estimated MSYs represent the maximum potential yield, including official landings, regulated by quota, as well as misreporting or illegal catches and by-catch in other fisheries. The calculated exploitation biomasses and equilibrium levels (MSYs) should not be regarded as absolute value for possibly future yields in view of the fact that the methods have some uncertainties and the share of misreported and illegal catches and the by-catch during fisheries on other target species has been still unknown. In such cases, special approaches has been applied, as use of 2/3 MSY (Caddy and Mahon, 1995).

8. Conclusions and recommendations

- ✓ Total number of identified species in Bulgarian waters was 16, from which 12 fish, 1 crustacean, 2 mollusk, 1 macro zooplankton;

- ✓ The total surveyed area in Bulgarian part was 9136.6 km^{-2} and total estimated sprat biomass was 21 090 t – in August-September and 32 280 t in December survey; Anchovy – 722.5 and 2217t; Scad – 1682 t and 4159 t;Red mullet-1971 t and 3035 t; Bluefish – 1682 t and 4530 t;

- ✓ The densest sprat aggregations were detected in the shallowest stratum 75-100 m (30.003 - 450.045 kg.km^{-2} CI (95%) = 36.22689. In contrast to the previous surveys the CPUA of *M.merlangius* was very low;

- ✓ In August –September the percentage of summer spawning species such as scad, red mullet and bluefish was relatively high;

- ✓ December is not the best month for biomass estimation due to the fact that sprat is batch spawning species (cold water species).The majority of population is in active maturation, not feeding actively. The fodder zooplankton in this period was dispersed, the sun activity was low and SST was relatively high, not favorite conditions for sprat aggregations;

- ✓ The highest sprat biomass aggregations were in 75-100 m isobaths.The densest aggregations were localized in front of Bourgas Bay predominantly, since in northern direction the indices were relatively low;

- ✓ The total anchovy biomass in August-September of Anchovy was 722.5 t, and in December 2217 t in Bulgarian Black Sea area;

- ✓ The total scad biomass in August-September of Scad was 1682 t, and in December 4159 t in Bulgarian Black Sea area;

- ✓ The total red mullet biomass in August-September of Scad was 1967.7 t, and in December 3035.2 t in Bulgarian Black Sea area;

- ✓ The total bluefish biomass in August-September of Scad was 1682 t, and in December 4159 t in Bulgarian Black Sea area;

- ✓ The size composition of sprat comprised of length classes (TL, cm) from 6.5 cm up to 11.75 cm in the samples from Bulgarian marine area;

- ✓ It is evident that the sizes classes of sprat from 7.0-8.5 cm are predominate, as the bigger classes were presented with low percentage. In August-

September length calss 7.0 was with very high percentage, followed by L = 8.0 and 8.5 cm;

- ✓ Predominate L class for red mullet in August September was 12.5cm, followed by Lclass= 9 cm.In December low share of all classes was detected;

- ✓ In August the share of whiting at 12.5 and 13 cm were the highest, although the presence of all length classes were very low in the catches.Later, in December the share of all length classes increased, as 12.5 ,13 and 13.5 classes increased twice and more.The largest classes of 15 and 15.5 cm increased significantly in December 2016;

- ✓ In August-September the representation of length calssess for scad begun with L=8 till 12.5 cm. In December the length classes from 10.5 to 14 were presented as the peak was at 11, 11.5 and 12 length classes;

- ✓ The sprat in active spawning during the present investigation in December. Most of the individuals are with III - IV stage of gonads;

- ✓ Analysis shows that the percentage of one and two years old fish was high. The three years old sprat specimen share increased in 2016;

- ✓ The asymptotic length reached 12.77cm and growth rate could be assessed as relatively high accounting 0.45 y^{-1}. The growth of sprat from present research is positive allometric (n=2.66);

- ✓ In the present research we used natural mortality coefficient for sprat as equal to 0.95;

- ✓ Levels above F =0.8 would lead to stock collapse. Sustainable levels of fishing mortality are those around F=0.5, which would correspond to the catch level of around 8.5 thous.tons of sprat in Bulgarian marine waters;

- ✓ The relative yield (Y/F_0) at very low *levels of the fishing mortality* is high during the first projected year. At F = 0.8, in the second projected year fall of the relative yield was detected up to the levels of F = 0.5;

- ✓ Sprat is a fast growing species with highly cycling nature of its recruitment and parental stock biomass dependant on the anthropogenic impacts different from fishing, as well as fishing press and dynamics in the environmental factors. Hence, when studied the continuity of the research on population parameter dynamics is of high importance. In studied months the observed length, weight and age structure were stable. The condition factor is high in the beginning of the spawning period and gonad maturation (IV-V; VI; III gonad stages were observed). To analyze and to make stronger recommendations regarding the sustainability of exploitation and measures for rational utilization of the marine living resources at least 4 samples per month should be collected from different depths and processed. New indicators as lipid content, otholith chemistry should be introduced when biological characteristics are studied;

- ✓ We strongly recommend the landings and catches sampling in all months, due to the fact that sprat fisheries is conducted whole year round with active and passive fishing gears;

- ✓ The calculated exploitation biomasses and equilibrium levels (MSYs) should not be regarded as absolute value for possibly future yields in view of the fact that the methods have some uncertainties and the share of misreported and illegal catches and the by-catch during fisheries on other target species has been still unknown. In such cases, special approaches has been applied, as use of 2/3 MSY (Caddy and Mahon, 1995).

9. References

Anonymous 2014. Balıkçılık istatistikleri. [Fisheries Statistics.] Türkiye İstatistik Kurumu, Ankara, Turkey. http://tuik.gov.tr [In Turkish.]

Alexandrov B. and Korshenko A., 2006. Manual for zooplankton sampling and analysis in the Black Sea Region.

Atılgan, E., Erbay, M., Aydın, 2010. Otolith characteristics of some economic species in the Eastern Black Sea. Yunus Research Bulletin.10(3)12-15

Aydın I., Sahin T. 2011. Reproductive performance of turbot (*Psetta maxima*) in the southeastern Black Sea. Turkish J. Zoology 35 (1): 109–113. DOI: 10.3906/zoo-0905-26.

Bertalanffy L. VON 1938. A quantitative theory of organic growth (Inquiries on growth laws. II). Human Biol. 10: 181-213.

Beverton, R.J. and S.J. Holt. (1957). On the dynamics of exploited fish populations. Fish. Invest. Ser. 2, Vol 19.

Debes, P. V., F. E. Zachos and R. Hanel (2008). Mitochondrial phylogeography of the European sprat (*Sprattus sprattus* L., Clupeidae) reveals isolated climatically vulnerable populations in the Mediterranean Sea and range expansion in the northeast Atlantic. Molecular Ecology 17: 3873-3888.

Dimov I., 1959. Improved quantitative method for zooplankton calculation. Rep. BAS, 12, 5, 427 429. (in Russian)

Foote, K.G. 1996. Quantitative fisheries research surveys, with special reference to computers. In: B.A. Megrey & E. Moksness. Computers in fisheries research. Chapman & Hall. 254 pp. 80-112.

Gulland J.A., 1966. Manual of sampling and statistical methods for fisheries biology. Part I: Sampling methods. FAO Manuals in Fisheries Science No. 3, Rome.

GullandJ.A., 1970. The fish resources of the ocean. FAO Fish. Techn. Pap. No. 97, 1-425, Rome.

FAO: Precautionary approach to fisheries. FAO Fish. Tech. Paper N. 350 (1), 1995.

ICES. 2011. Report of the Workshop on Sexual Maturity Staging of Herring and Sprat (WKMSHS), 20-23 June 2011, Charlottenlund, Denmark. ICES CM 2011/ACOM:46. 143pp.

ICES. 2012. Report of the Workshop on Sexual Maturity Staging of Turbot and Brill (WKMSTB 2012), 5–9 March 2012, Ijmuiden, Netherlands. ICES 2012/ACOM:56. 48 pp

Jerald, Jr., A. 1983. Age Determination. In: Fisheries Techniques (L.A., and D.L. Johnson, eds). Southern Printing Company, Inc., Blacksburg, VA, pp 301-324.

Kasapoglu, N., Duzgunes, E. 2014. Otolith Atlas for the Black Sea. Journal of Environmental Protection and Ecology 16, No 1, 133–144

Korshenko, A., Alexandrov B., 2012. Manual for mesozooplankton sampling and analysis in the Black Sea monitoring (Black Sea Zooplankton Manual) Online: http://bsc.ath.cx/documents/ExpertNetwork/default.asp?I=/Expert%20Network%20-%20 Zooplankton

Laevastu, T., 1965. Manual methods in fisheries biology. Observations on the chemical and physical environment. Chemical analysis of water FAO. Marine Physiological Sci., 6: 86-86.

Mordukhay-Boltovskoy, F.D. (Ed.). 1968. The identification book of the Black Sea and the Sea of Azov Fauna.- Kiev: Naukova Dumka Publ., T. 1 (Protozoa, Porifera, Coelenterata, Ctenophora, Nemertini, Nemathelminthes, Annelida, Tentaculata), 423 pp. (in Russian).

Mordukhay-Boltovskoy, F.D. (Ed.). 1969. The identification book of the Black Sea and the Sea of Azov Fauna.- Kiev: Naukova Dumka Publ., T. 2 (Artropoda: Cladocera, Calanoida, Cyclopoida, Monstrilloida, Harpacticoida, Ostracoda, Cirripedia, Malacostraca, Decapoda), 536 pp. (in Russian).

Mordukhay-Boltovskoy, F.D. (Ed.). 1972. The identification book of the Black Sea and the Sea of Azov Fauna.-Kiev: Naukova Dumka Publ., T. 3 (Arthropoda, Mollusca, Echinodermata, Chaetognatha, Chordata: Tunicata, Ascidiacea, Appendicularia), 340 pp. (in Russian).

Mihneva V., Raykov V., Grishin A., Stefanova K., 2015. Sprat feeding in front of the Bulgarian Black Sea Coast, MEDCOAST conference 2015, vol.1, 431- 443

Nelson J.S. 2006. Fishes of the world. 4th edn. Wiley, New York, NY, USA.

Nielsen J.G. 1986. Scophthalmidae. Pp. 1287–1293. In: Whitehead P.J.P., Bauchot M.-L., Hureau J.-C., Nielsen J., Tortonese E. (eds.) Fishes of the North-eastern Atlantic and the Mediterranean. Vol. 3. UNESCO, Paris.

Petipa, T.S. 1959. On the mean weight of the principle forms of zooplankton in the Black Sea. Tr. Sevast. Biol. St. 9, P. 39-57

Panayotova M., Marinova V., Raykov V., Stefanova K., Stanchev H., Shtereva G., Krastev A., Maximov V., Radu Gh., Apostolov A. 2011. Scientific report from pilot Black Sea study for application of hydroacoustic method in assessment of fish biomass and distribution to National Agencies of Fisheries and Aquaculture of Bulgaria and Romania in relation to National Data Collection programs for 2010. Report, 46 pp.

Panayotova M., Marinova V., Raykov V., Stefanova K., Stefanova E., Yankova M., 2014. Study and assessment of pelagic fish stocks along Bulgarian Black sea coast by hydroacoustic method. Biological monitoring European sprat (*Sprattus sprattus*) landings during August – November 2014. Institute of Oceanology – BAS, Varna, Bulgaria and National Agency for fisheries and Aquaculture – Bulgaria in accordance with the Council Regulation (EC) № 199/2008 establishing a community framework for the collection, management and use of data in the fisheries sector and support for scientific advice regarding the common fisheries policy. 82 pp.

Pinkas L.,. Oliver M.S, Iverson I.L.K., 1971. Food habits of albacore, bluefin tuna and bonito in Californian waters. California Fish Game 152:1-105.

Pısıl, Y., 2006. Karadeniz'de Yaşayan Çaça Balığı (Sprattus sprattus (L., 1758))'nda Kemiksi Yapıları ve Uzunluk-Frekans Metodu ile Yaş Tayini. 19 Mayıs Üniversitesi, Fen Bilimleri Enstitüsü, Y. Lisans Tezi, 32 s.

Prodanov K., Mikhailov K., Daskalov G., Maxim C., Chashchin A., Arkhipov A., Shlyakhov V., Ozdamar E. 1997. Environmental management of fish resources in the Black Sea and their rational exploitation. Studies and Reviews. GFCM. No. 68. FAO, Rome.

Raykov V., Panayotova M., Stefanova K., Stefanova E., Radu G., Maximov V., Anton E.. 2011. Statement to the Deputy Minister of Ministry of environment and waters in order to annual workshop of Black Sea commission National Report to GFCM 35 - Annual Assembly; Scientific report from international pelagic trawl survey in the Bulgarian and in the Romanian Black Sea area, June 2010 to National Agencies of Fisheries and Aquaculture of Bulgaria and Romania in relation to National Data Collection programs for 2010, 71 pp.

Raykov V., Schlyakhov Vl., Maximov V., Radu Gh., Staicu I., Panayotova M., Yankova M., Bikarska I., 2008 - Limit and target reference points for rational exploitation of the turbot (Psetta maxima L.) and whiting (Merlangius merlangus euxinus Nordm.) in the western part of the Black Sea.VI Anniversary Conferense of the Institute of zoology. Acta Zoologica Bulgarica, 2008, Suppl.2: 305-316, ISSN 0324- 0770.

Raykov V, Yankova M, Radu Gh., Lisichkov K., 2009 - Exploitation patterns call for limit the sprat catches off the north-western coast of the Black sea.Intern.Conf.:Sustainability in South-eastern Europe"Tekirdag, Istanbul 16-20 VI.(under review for JEPE).

Raykov V., I. Staicu, G. Radu, V. Maximov, S. Nicolaev, 2008 – Specificity of the fishery and common fishery policy implementation: a case study of the western part of the Black Sea. Cercetari marine. Recherches marines. INCDM. 38: 223-232, ISSN:0250-3069, Cod CNCSIS: 74.

Raykov.V 2006. From EU25 to EU27.El Anzuelo, European newsletter on fisheries and the environment Vol.17, 10-11p.

Raykov V 2008. Stock agglomerations assessment of sprat (*Sprattus spratus* L.) off the Bulgarian Black Sea coast. Cercetari marine. Recherches marines. INCDM. 37. ISSN: 0250-3069.

Raykov,V. M Yankova., V. Mihneva,.Dineva S., Petrova D. M. Panayotova, 2008 Stock Assessment of Sprat (Sprattus sprattus) by Swept Area Method during the spring season of 2008 along the Bulgarian Black Sea coast. Technical report, 2008.

Raykov V Primary management objectives for sustainable Sprat (Sprattus sprattus L.) stock exploitation at the Bulgarian Black Sea coast - preliminary results J.Environmental Protection and Ecology, 8 (2) (2007),302-318.

Raykov V.S, V.V.Mihneva, Daskalov, and G: Investigations on sprat (*Sprattus sprattus* L.) population dynamics related to its trophic base and climate change over the period 1996-2004 in Bulgarian waters of the Black Sea. J.Environmental Protection and Ecology, 8 (2) (2007), 319-332.

Raykov, V, Panayotova Stefanova K, E. Stefanova 2011 Stock assessment of sprat in Bulgarian marine area, Institute of Fish resources and National Agency for fisheries and Aquaculture – Bulgaria in accordance with the Council Regulation (EC) № 199/2008 establishing a community framework for the collection, management and use of data in the fisheries sector and support for scientific advice regarding the common fisheries policy. 100 pp.

Simon, K.D., Y. Bakar, A.G. Mazlan, C.C. Zaidi and A. Samat et al., 2012. Aspects of the reproductive biology of two archer fishes Toxotes chatareus, (Hamilton, 1822) and Toxotes jaculatrix (Pallas, 1767). Environ. Biol. Fish., 93: 491-503.

Yoraz, A., 2015. Orta Karadeniz Balıkçılığındaki Bazı Pelajik ve Demersal Balık Türlerinin Sagittal Otolitlerinin Morfometrik Tanımlanması ve Annulus Oluşumunun Görsel Analizi, Doktora Tezi, OMU Fen Bilimleri Enstitüsü, 407 s.

Zengin M., Düzgüneş E. 2003. Variations on the turbot (Scophthalmus maeoticus) stocks in the south-eastern Black Sea during the last decade and comments on fi sheries management. Pp. 9–26. In: Öztürk B., Karakulak S. (eds.) Workshop on Demersal Resources in the Black Sea & Azov Sea. 15–17 April Şile, Istanbul, Turkey.

ANNEX I

CPUE kg.h^{-1} and CPUAkg.km^{-2} Bulgarian marine area - Sprat

	August			**December**	
1	CPUEkg/h	CPUAkg/km2	1	CPUEkg/h	CPUAkg/km2
2	138.0138	1606.066617	2	263.9998	3072.165562
3	165.0165	1988.879079	3	100.7999	1214.901836
4	180.018	2169.686268	4	167.9999	2024.836393
5	240.024	2793.159334	5	263.9998	3072.165562
6	54.0054	628.4608501	6	95.99992	1117.151113
7	66.0066	768.1188168	7	91.19993	1061.293558
8	30.003	349.1449167	8	119.9999	1396.438892
9	66.0066	795.5516317	9	107.9999	1301.680538
10	144.0144	1735.749015	10	131.9999	1590.94288
11	165.0165	1920.297042	11	407.9997	4747.892232
12	75.0075	872.8622918	12	143.9999	1675.72667
13	90.009	1047.43475	13	287.9998	3351.45334
14	45.0045	523.7173751	14	115.1999	1340.581336
15	180.018	2094.8695	15	167.9999	1955.014449
16	225.0225	2712.107835	16	287.9998	3471.148102
17	234.0234	2723.33035	17	203.9998	2373.946116
18	66.0066	768.1188168	18	239.9998	2792.877784
19	90.009	1084.843134	19	167.9999	2024.836393
20	180.018	2169.686268	20	527.9996	6363.771521
21	165.0165	1988.879079	21	599.9995	7231.558547
22	150.015	1808.07189	22	215.9998	2603.361077
23	120.012	1396.579667	23	263.9998	3072.165562
24	120.012	1396.579667	24	959.9992	11171.51113
25	195.0195	2350.493457	25	263.9998	3181.885761
26	270.027	3142.304251	26	215.9998	2513.590005
27	165.0165	1920.297042	27	623.9995	7261.482237
28	114.0114	1326.750684	28	191.9998	2234.302227
29	360.036	4189.739001	29	287.9998	3351.45334
30	330.033	3977.758158	30	263.9998	3181.885761
31	210.021	2531.300646	31	527.9996	6363.771521
32	360.036	4339.372536	32	287.9998	3471.148102
33	300.03	3616.14378	33	239.9998	2892.623419
34	180.018	2094.8695	34	215.9998	2513.590005
35	375.0375	4520.179725	35	599.9995	7231.558547
36	360.036	4339.372536	36	383.9997	4628.19747
37	450.045	5237.173751	37	479.9996	5585.755567

ANNEX II

Species composition from Bulgarian area

Species	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	26	27	28	29	30	31	32	33	34	35	36	
Sq.acanthias																																					
R.clavata																																					
D.pastinaca																																					
A.immaculata																																					
S.sprattus																																					
M.merlangus																																					
E.encrasicolus																																					
Tr.mediterraneus																																					
M.barbatus																																					
P.maxima																																					
N.melanostomus																																					
G.niger																																					
Polybius vernalis																																					
M. galloprovincialis																																					
R.venosa																																					
A.aurita																																					

Species	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	26	27	28	29	30	31	32	33	34	35	36	
Sq.acanthias																																					
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M.barbatus																																					
P.maxima																																					
N.melanostomus																																					
G.niger																																					
Polybius vernalis																																					
M. galloprovincialis																																					

ANNEX III

Market sampling, Discards and Surveys indicator targets and results in 2016 (Bulgarian part)

Black Sea	Length @age	market, discards, surveys	2,50%	Market: **2026** Discard: - Survey: **12 427** 1250
Black Sea	Weight @length	market, discards, surveys	2,50%	Market: **2026** Discard: - Survey: **12 427** 5000
Black Sea	Weight @age	market, discards, surveys	2,50%	Market: **2026** Discard: - Survey: **12 427** 1250
Black Sea	Maturity @length	surveys	2,50%	5000 **140**
Black Sea	Maturity @age	surveys	2,50%	5000 **140**
Black Sea	Sex-ratio @length	market, surveys	2,50%	Market: **250** Survey: **250** 125
Black Sea	Sex-ratio @age	market, surveys	2,50%	Market: **250** Survey: **250** 500