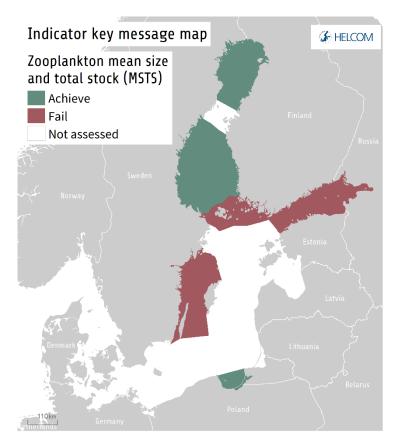


HELCOM core indicator report July 2018

Zooplankton mean size and total stock (MSTS)

Key Message

This core indicator evaluates zooplankton community structure to determine whether it reflects good environmental status. As a rule, good status is achieved when large-bodied zooplankters are abundant in the plankton community. Due to strong environmental gradients and community variations, size distribution and total stock of the zooplankton corresponding to good status vary between the Baltic Sea sub-basins.



Key message figure 1. Evaluation of the status assessment results for zooplankton indicator 'Mean size and total stock' (MSTS). The assessment is carried out using Scale 2 HELCOM assessment units (for more information see the <u>HELCOM Monitoring and</u> <u>Assessment Strategy Annex 4</u>. **Click here to access interactive maps at the HELCOM Map and Data Service: <u>Zooplankton mean size</u> <u>and total stock</u>.**

The indicator-based status evaluation has been completed for the northern Baltic Sea, namely the Gulf of Bothnia, Gulf of Finland, Åland Sea, and Western Gotland Basin, and for the Gdansk Basin in the southern Baltic Sea. For the other basins, work to establish the threshold values needed to carry out the evaluation is still in progress.

Good status during the assessment period 2011-2016 was found in the Bothnian Bay, Bothnian Sea and Gdansk Basin. By contrast, in the Åland Sea, Gulf of Finland and Western Gotland Basin, zooplankton mean size and/or total biomass have declined during the last decades, and MSTS does not reflect a good status

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during the assessment period 2011-2016. This negative development results from both an increased contribution of small zooplankton species, a probable consequence of eutrophication, and a decreased share of copepods, a probable consequence of increased predation by zooplanktivorous fish. It is also possible, albeit not verified, that altered environmental conditions (e.g. decreased salinity, increased temperature and deep water hypoxia) have contributed to these trends. The detected trends in the mean size and total stocks of zooplankton communities indicate that today's pelagic food web structure is not optimal for energy transfer from primary consumers (phytoplankton) to fish.

The confidence of the indicator evaluation is **moderate** since the data used cover fairly long time periods for the sub-basins where the evaluation results are completed, but also for the sub-basins where these results are not yet available.

The indicator is applicable in the waters of all the countries bordering the Baltic Sea. However, currently the indicator is only operational in some assessment units, and further development work is needed to make it operational in the remaining assessment units in the future.

Relevance of the core indicator

Zooplankton includes an array of macro and microscopic invertebrates. They play a vital role in the marine food web. The herbivorous zooplankton feed on phytoplankton and in turn constitute prey to animals at higher trophic levels, including fish. Therefore, zooplankton are an essential link in aquatic food webs, influencing energy transfer in the pelagic food webs and recruitment to fish stocks as well as ecosystem productivity, nutrient and carbon cycling. Hence, the evaluation of zooplankton communities is a prerequisite for analysis of pelagic food web structure.

The mean size of a zooplankter in the community is indicative of both fish feeding conditions and grazing pressure from zooplankton on phytoplankton. Large stocks of zooplankton composed of large-bodied organisms have a higher capacity for transfer of primary producers (phytoplankton) to fish, i.e. higher energy transfer efficiency. By contrast, dominance of small-bodied zooplankton is usually associated with lower energy transfer efficiency, due to higher losses. Thus, a high community biomass of zooplankton with large individual body size represents both favourable fish feeding conditions and a high potential for efficient utilization of primary production. According to ecological theories, this would represent an efficient food web and correspond to a good environmental status. All other combinations of zooplankton stock and individual size would be suboptimal and imply food web limitations in terms of energy transfer through the food web and productivity.



Policy relevance of the core indicator

	BSAP segment and objectives	MSFD Descriptor and criteria			
Primary link	Biodiversity	D4 Food-web			
	• Thriving and balanced	D4C3 The size distribution of individuals across the			
	communities of plants and	trophic guiled is not adversely affected due to			
	animals	anthropogenic pressures			
Secondary link	Eutrophication	D1 Biodiversity			
	 Natural distribution and occurrence of plants and animals 	D1C6 The condition of the habitat type, including its biotic and abiotic structure and its functions (e.g. its typical species composition and their relative abundance, absence of particularly sensitive or fragile species or species providing a key function, size structure of species), is not adversely affected due to anthropogenic pressures.			

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Results and Confidence

The evaluation of zooplankton mean size and total stock (MSTS) for the period 2011-2016 indicates that in the Bothnian Bay, the Bothnian Sea and the Gdansk Basin, the MSTS values are above the threshold values indicating good status. By contrast, in the Åland Sea, Gulf of Finland and Western Gotland Basin, the MSTS values are significantly below the threshold values, which implies that good status has not been achieved. The details for each of the evaluated sub-basins are presented below.

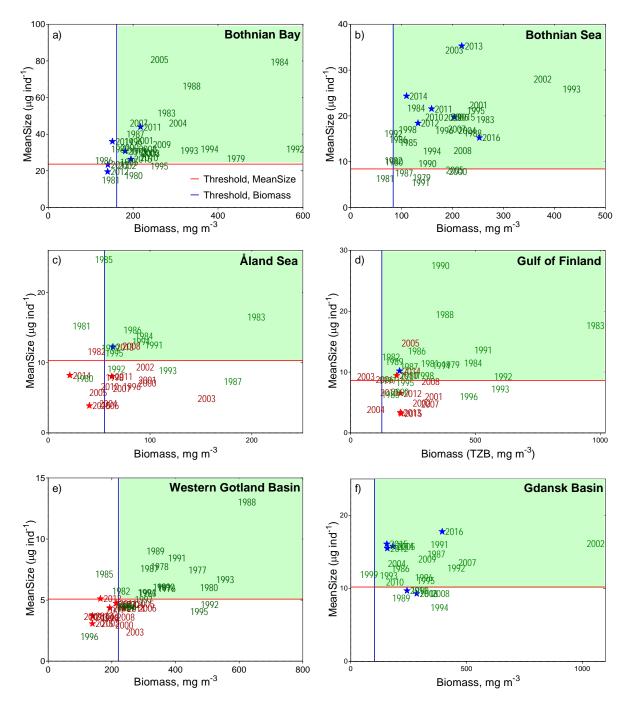
In the Bothnian Bay (Results figure 1a), MSTS has not changed considerably over the time period for which data are available (1979 – 2016). Although in some years values were below the threshold, these occasional deviations were not significantly different from the threshold value as indicated by CuSum analysis, implying that these fluctuations are stochastic and that MSTS reflects a good status.

In the Bothnian Sea (Results figure 1b) MSTS also suggests good food web structure, with no indication of the status decline over the assessed period. In the Åland Sea (Results figure 1c), starting from 1996, zooplankton mean size stayed significantly below the threshold and the total biomass values were often below the threshold values. Although the total biomass occurring during 2011-2016 are not significantly below the threshold, the mean size is, which implies that that the good status has not been achieved in this sub-basin.

In the Gulf of Finland (Results figure 1d), the values of the mean size indicate that the system was not in good status from 2001 onwards. Also, the biomass failed the threshold during the same years on multiple occasions, albeit not significantly. Thus, MSTS indicates that in 2011-2016, zooplankton community is not in good status.

In the Western Gotland Basin (Results figure 1e), the MSTS indicates that the system is not in good status since 1998, although some signs of recovery – at least in the coastal station Askö (monitoring station B1) – appear after 2007. Nevertheless, during the assessment period 2011-2016, zooplankton community is not in good status.

In the Gdansk Basin (Results figure 1f), the MSTS values indicate that the system is good status, with no deviations from its reference state for the last 30 years with regard to both mean size and biomass values.



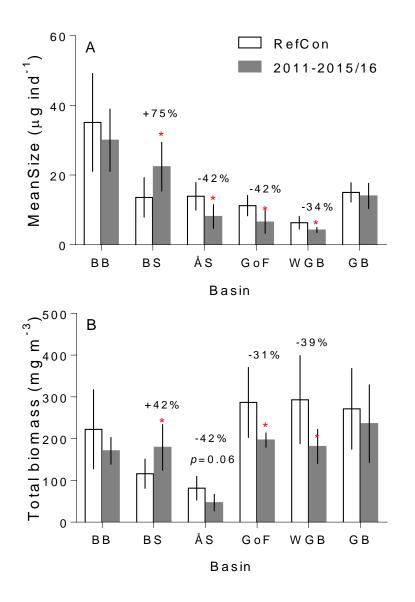
Result figure 1. Assessment results on the performance of MSTS indicator, which integrates mean size (Y axis) and total biomass of zooplankton (X axis). Blue and red lines show threshold values for the total biomass and mean size, respectively. Green shaded quartile indicates good status. Observations in good and not in good status are shown as green and red years, respectively. Stars indicate the assessment period years (2011 to 2016) with blue and red symbols (stars) for the observations that are in good and not in good status, respectively. Note that some years falling below the threshold values were assigned as being in good status, because these values were not judged as significantly different from the threshold value according to the CuSum analysis, which is based on the cumulative summing of the persistent deviations from the reference mean.

The difference in the MSTS components between the reference conditions and the assessment period varied from -34% to +75% for the mean zooplankter size (μ g ind⁻¹) and from -42% to +42% for the total biomass (mg

m⁻³) among the sub-basins (Results figure 2). Prominent decreases in both body size and total biomass of zooplankton were observed in the Åland Sea, Gulf of Finland and Western Gotland Basin, where size and total biomass decreased by 39% and 38%, respectively, from the reference period to the assessment period (2011-2016). Similar changes occurred in the Bornholm Basin (preliminary assessment) where mean size and biomass decreased by 20% and 39%, respectively.

Contrary to all other sub-basins, both mean size and biomass have increased in the Bothnian Sea from the reference period to the assessment period (Results figure 2). The increase observed in the Bothnian Sea is related to an increased population size of the large-bodied copepod *Limnocalanus macrurus*. This species, which is a glacial relict in the Baltic Sea, responded positively to the low salinity conditions during the last decade, which improved herring feeding conditions (Rajasilta et al. 2014) as well as MSTS values in this sub-basin. In the other sub-basins, species that contributed to the detected changes in the MSTS components varied. However, regardless of the variability among the species and species groups contributing to general declines in body size and biomass values among the sub-basins, an increase in proportion of small-sized taxa and groups was observed in all assessment units (except the Bothnian Sea). In the Gulf of Finland, the change is largely attributed to a decline in the biomass of large cladocerans. In the Western Gotland Basin and the Bornholm Basin, the decline in mean size and total biomass is mostly due to declining copepod populations and thus shifting size spectra and biomass of the zooplankton communities.





Results figure 2. Pair-wise comparisons between the MSTS values observed during the assessment period (2011-2015 for GoF and ÅS (n = 5), and 2011-2016 for BB, BS, WGB and GB (n = 6), where n is the number of years included in the analysis) and the reference period (RefCon; n varied from 8 to 17) for mean zooplankton size (MeanSize; A) and total zooplankton biomass (Total biomass; B) in the Bothnian Bay (BB), Bothnian Sea (BS), Åland Sea (ÅS), Gulf of Finland (GoF), Western Gotland Basin (WGB) and Gdansk Basin (GB). The basin-specific data were compared using unpaired t-test with Welch correction and statistically significant differences (p < 0.05) are indicated with percent change and red asterisk. For the nearly significant difference in ÅS, the *p* value is shown. Percentage values indicate change (positive or negative) in the value observed for the assessment period relative to the reference period. Data are shown as means and standard deviations for the untransformed data; the statistical comparisons were done using Box-Cox transformed values that were normally distributed.

Future work

At present, the MSTS indicator has not been evaluated for all open sea assessment units in the Baltic Sea where zooplankton monitoring is conducted. The applicability of the indicator and the determination of relevant threshold values are still needed in the northern Baltic Proper, much of the eastern, south-easternand southern Baltic Sea before evaluation for these areas can be conducted.

Temperature- and salinity-induced MSTS responses also need to be further evaluated and, if relevant and significant, they need to be accounted for in the indicator-based assessment of the pelagic food webs.

In order to assess the status of the food webs in the Baltic Sea, further development of the interpretation of the indicator results in relation to other assessment results is needed. A full assessment of pelagic food webs is still to be developed, and the outcome of the MSTS-based assessment needs to be considered in conjunction with other food web indicators. The interpretation of MSTS should also be integrated with the results of the eutrophication status assessment.

Confidence of the indicator status evaluation

The overall confidence of the evaluation varies from **low** to **high** between the assessment units.

Confidence of the evaluation accuracy depends on the time series length and between-year variability during the reference period. Also, different number of stations per assessment unit contributes to the between-year data variability. The data availability is the main reason for the variation in the confidence across the assessment units.

Zooplankton monitoring stations are generally found in every Baltic Sea sub-basin, and suitable monitoring data series are available for relatively long (>18 years) time periods from most of the sub-basins. A similar confidence in the evaluation (moderate to high) is expected for the most assessed basins with fairly similar length of the data sets and similar number of observations (number of data points per basin and per year). However, in case of low observation frequency (for example, Gdansk Basin, where only August data were used from a single station each year), the confidence is low.

The accuracy component of the confidence is considered to be high. This confidence classification is due to: (1) the CuSum technique that is used to determine whether the observed value reflects good status or not is considered to be a very sensitive method for detecting persistent small changes (Lucas 1982),

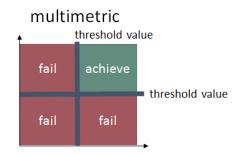
(2) the lower bound of 99% confidence interval around the baseline (reference condition) was used as threshold, thus minimizing the risk of false negatives (i.e., assigning not good status to an observation that is in fact reflecting good status), and

(3) using a pre-cautionary principle by selecting the higher value after comparing threshold values obtained for RefCon_{Fish} and RefCon_{Chl} for each part of the indicator (i.e. MeanSize and total biomass).



Thresholds and Status evaluation

This core indicator employs zooplankton mean size and total stock (MSTS) to evaluate pelagic food web structure, with particular focus on lower webs. MSTS evaluates whether good status is achieved using two threshold values, one for mean size and one for total standing stock (abundance or biomass) of zooplankton (Thresholds figure 1). An area is evaluated as having achieved good status using the MSTS indicator when **both** mean size and total stock are achieve their specific threshold values.



Thresholds figure 1. Schematic illustration of the core indicator applying two threshold values.

Due to strong environmental gradients in the pelagic communities in the sub-basins of the Baltic Sea, the exact threshold values are specific for each assessment unit (Thresholds table 1).

Assessment unit	Threshold value			
	mean size(μ g wet weight ind $^{-1}$) / total stock (mg m $^{-3}$)			
Gdansk Basin	10.2/103			
Western Gotland Basin	5.0 / 220			
Gulf of Finland	8.6 / 125			
Åland Sea	10.3 / 55			
Bothnian Sea	8.5 / 84			
Bothnian Bay	23.7 / 161			

Thresholds table 1. Assessment unit specific threshold values.

The threshold values are set using a reference period which defines a status when the food web structure was not measurably affected by eutrophication and represents good fish feeding conditions within the time series of existing data. Thus, the reference periods for MSTS reflects a time period when effects of eutrophication (defined as 'acceptable' chlorophyll *a* concentration) are low, whereas nutrition of zooplanktivorous fish is adequate for optimal growth. Hence, these are the periods when eutrophication and overfishing related food web changes are negligible. In some cases, reference periods can be adopted from neighbouring areas, for which longer datasets are available.

As the indicator evaluates the structural- and functional integrity of the food web, the threshold values are conceptually achieved when:

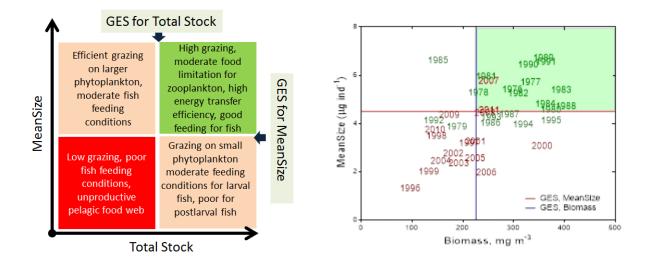
• there is a high proportion of large-sized individuals (usually copepods) in the zooplankton community that efficiently graze on phytoplankton and provide good-quality food for zooplanktivorous fish, and

• the abundance (biomass) of zooplankton is at an adequate level to support fish growth and exert control over phytoplankton production.

Two alternative strategies for setting reference conditions are possible.

- The first approach should be used when the data series are very short. Conceptually this approach is similar to using a trend as a threshold value. When using this approach, the longterm mean and corresponding variance (95% confidence interval, CI) for both the mean size and the total stock parameter are calculated based on the entire available dataset. The lower bound of 95%-CI is then used as threshold value to evaluate deviations in the current observations. This approach is possible; however, it was not used in the MSTS-based evaluation in 2011-2016.
- 2. The second approach is based on (i) specific reference conditions for chlorophyll *a* concentrations (RefCon_{Chl}) that have been defined for the different sub-basins of the Baltic Sea (either observed in the past or based on models), and (ii) reference data on clupeid fish (young herring and sprat) that are used to identify the reference time periods (RefCon_{Fish}) when both the fish growth (i.e. weight-at-age, WAA, or other body condition indices, such as fat content) and fish stocks were relatively high in the relevant ICES subdivisions. Once the reference time periods have been identified based on chlorophyll *a* and fish time series, the threshold values for both mean size and total stock were defined as the lower bound of the 99%-CI for the respective mean values calculated for zooplankton time series during the reference time period. This approach was used for the 2011-2016 assessment period (Thresholds figure 2).





Thresholds figure 2. The MSTS concept (left) and a data example (right) to illustrate the use of the indicator. The green area on the left panel represents good status conditions, orange areas represent not good status where only one of the two parameters is adequate and the red area represents not good status where both parameters fail to meet the threshold value. On the right panel, an example of long-term zooplankton data for mean size and total biomass (station B1, Askö, Western Gotland Basin) were analysed. The corresponding thresholds are shown as red and blue lines, respectively. The years in green were classified as in good status and those in red as not in good status. Generally, all years located in the right upper quadrant (green area in panel A) reflect good status. However, some years (e.g., 1979, 1985, 1994, etc.) are classified as reflecting good status, although they are placed outside of the green area. For these years, even though the absolute values for the indicator components (MeanSzie and biomass) are below the threshold value, the deviation is not significant as determined by CuSum. To achieve a significantly not good status value, the change must be persistent and cumulative negative change must exceed 5 σ difference from the threshold value. Similarly, some years (e.g., 2007) are classified as not good status, although they are placed in the green area; during these years the observed values were above the thresholds, however this has not resulted in a significant shift in any of the MSTS component that was sufficiently persistent to return the MSTS values in the reference state. See the <u>Assessment protocol for details.</u>



Assessment Protocol

The indicator uses mean zooplankton size and total stock (MSTS) for evaluating whether good environmental status is achieved or not. The indicator uses the parameter mean zooplankter size (mean size) which is presented as a ratio between the total zooplankton abundance (TZA) and total biomass (TZB). This metrics is complemented with an absolute measure of total zooplankton stock, TZA or TZB, to provide MSTS. Thus, MSTS is a two-dimensional, or a multimetric, indicator representing a synthetic descriptor of zooplankton community structure.

Data treatment and control charts

Data period: The MSTS evaluations are currently restricted to the analysis of zooplankton communities observed during June-September. This seasonal time period was chosen because it is covered most extensively by the monitoring sampling programmes supplying the data; moreover, this is also the period of the highest plankton productivity as well as predation pressure on zooplankton (Johansson et al. 1993; Adrian et al. 1999). The structure of the marine food web is naturally variable; therefore, the indicator is designed to detect changes in the community structure that significantly deviate from the natural variability during the growth season.

Control charts: The time series of the MSTS components (mean size and total stock) for each zooplankton community are analyzed with cumulative sum (CuSum) control charts. The CuSum methods are designed to detect persistent small changes when the long-term mean changes in observed processes or periods. A control chart uses information about the natural variation of the process that is evaluated to examine if the process, i.e. the structure of the zooplankton community, is moving beyond the expected stochastic variability which is defined as desirable tolerance. If the process is *in control*, i.e. the zooplankton community structure is not affected by pressures, then subsequent observations are expected to lie within the tolerance boundaries. The hypothesis that the process is *in control* is rejected if the observations fall outside the desired tolerance boundaries. As a test statistic, control charts employ the controlling mean (μ) and specify control limits of *n* × standard deviations (σ) above and below the mean or the confidence intervals (CI). The upper and lower control limits are defined using a conservative approach of ±5 σ for μ estimated for either RefCon_{Fish} (reference conditions for fish) or RefCon_{Chl} (reference conditions for chlorophyll *a* concentrations).

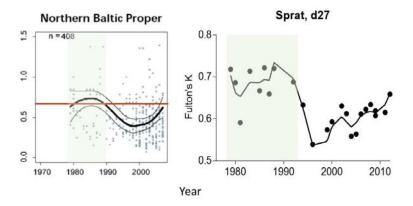
All datasets used for setting the thresholds values for evaluating status are >30 years of observations. The normality of each data series is first tested for normality (D'Agostino & Pearson omnibus normality test, Shapiro-Wilk and Kolmogorov-Smirnov normality tests). As both mean size and total zooplankton biomass often deviate significantly from the normal distribution, the values can be transformed using Box-Cox procedure and all calculations are then carried out on the transformed data. Once a controlling mean (μ_i) and standard deviation (σ_i) have been specified based on the chosen period used to determine the baseline against which status evaluation is made, indicator values ($x_{i,t}$) within the time series are standardized to *z*-scores ($z_{i,t}$) as:

$$Z_{i,t} = \frac{X_{i,t} - \mu_i}{\sigma_i}$$



The approach for setting the reference period used a window of the available data corresponding to the selected reference period, i.e. years representing sub-basin specific reference conditions for (*i*) food webs not measurably affected by eutrophication; these are based on environmental quality ratio (EQR) and historical data on chlorophyll *a* (HELCOM 2009) when defining RefCon_{Chl}, and (*ii*) high feeding conditions for zooplanktivorous fish when defining RefCon_{Fish} (Assessment protocol figure 1).

The μi and σi are defined based on the conditions during the reference period.



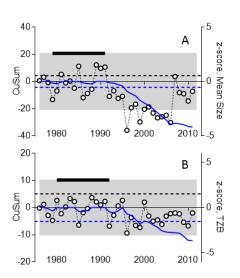
Assessment protocol figure 1. Examples for setting RefCon_{Chl} and RefCon_{Fish} using long-term variability in chlorophyll *a* expressed as ecological quality ratio (EQR) in the northern Baltic Proper (modified from HELCOM 2009) (left) and body condition index (Fulton's K) of sprat in the ICES subdivision 27 (right) used to identify time period (green area) when zooplankton community was sufficient to efficiently transfer primary production to secondary consumers.

To investigate trends in accumulated small changes for the zooplankton mean size and total stock over long time periods, the CuSum charts (Assessment protocol figure 2) are constructed by first determining a decision-interval CuSum (DI-CuSum) that is calculated by recursively accumulating negative deviations (one-sided lower CuSum) as:

$$S_i^- = \min[0, S_{i-1}^- + z_i + k]$$

with $S_{i=0} = 0$. The k value is the allowance value in the process, expressed in z units, reflecting natural variability of the mean shift one wishes to detect. Thus, deviations smaller than k are ignored in the recursions. The default choice of k = 0.5 is considered appropriate for detecting a 1- σ shift in the process mean (Lucas 1982).





Assessment protocol figure 2. CuSum analysis of mean size (A) and total zooplankton biomass, TZB (B) using data series for station B1 (Askö station, Western Gotland Basin). The data are normalized to z-scores (right Y axis, open symbols). The threshold values are shown as dashed blue lines (-5 σ from the mean for the reference period; σ is standard deviation) and the reference period (years) is indicated as a black bar on the top. The lower CuSum (solid blue line) indicates accumulated changes in the mean size and TZB; the CuSum lines are crossing the respective good status threshold values in 1995 (mean size) and 1999 (TZB). According to this chart, from 1995 onwards, MSTS indicates food web structure being in not good status.

A strategy that was used for obtaining an overall status assessment when several datasets are available for an assessment unit is based on the integrated datasets. Since all zooplankton data are generated by national laboratories following HELCOM-Monitoring Manual guidelines and standardized gears and analysis methods, the data used for MSTS calculations are likely to be comparable. In order to arrive at a meaningful decision scheme, the main properties of the datasets should be considered. This includes issues such as length of the time series, their variability within defined reference periods, length of the time series overlapping with the reference periods, statistical properties of yearly mean values (i.e. number of samples contributing), quality control practices in the analyzing laboratories, etc. These issues were carefully considered and discussed before this two-stage assessment algorithm (first, comparing the datasets, and, second, generating integrated data for the assessment unit) was applied.

Assessment units

The indicator is evaluated using HELCOM assessment scale 2, which is consists of 17 Baltic Sea sub-basins. In the future it should be further discussed whether a higher spatial resolution (i.e. separating coastal and offshore areas) is needed.

The assessment units are defined in the HELCOM Monitoring and Assessment Strategy Annex 4.

Relevance of the Indicator

Biodiversity assessment

The status of biodiversity is assessed using several core indicators. Each indicator focuses on one important aspect of the complex issue. In addition to providing an indicator-based evaluation of the mean size and total stock of zooplankton, this indicator contributes to the overall biodiversity assessment in 2018 along with the other biodiversity core indicators.

Policy relevance

The indicator on zooplankton mean size and total stock addresses the Baltic Sea Action Plan (BSAP) Biodiversity and nature conservation segment's ecological objective 'Thriving and balanced communities of plants and animals', which has a direct connection to the food web structure. The background document to the Biodiversity segment of the BSAP describes a target for this ecological objective as 'By 2021 all elements of the marine food webs, to the extent that they are known, occur at natural and robust abundance and diversity'.

The core indicator also addresses the following qualitative descriptors of the MSFD for determining good environmental status (European Commission 2008):

Descriptor 1: 'Biological diversity is maintained. The quality and occurrence of habitats and the distribution and abundance of species are in line with prevailing physiographic, geographic and climatic conditions';

Descriptor 4: 'All elements of the marine food webs, to the extent that they are known, occur at normal abundance and diversity and levels capable of ensuring the long-term abundance of the species and the retention of their full reproductive capacity'.

and the following criteria of the Commission Decision (European Commission 2010):

- Criterion 1.6 (habitat condition)
- Criterion 4.3 (abundance/distribution of key trophic species)

This core indicator is among the few indicators able to evaluate the structure of the Baltic Sea food web with known links to lower and higher trophic levels.

The role of zooplankton in the ecosystem

Zooplankton play an important role transferring primary production to zooplanktivorous fish. However, different zooplankton taxa often have different preferences for trophic state of the ecosystem and are of different value as prey for zooplanktivores, because of the variations in size, escape response, and biochemical composition. In the Baltic Sea, alterations in fish stocks and regime shifts received particular attention as driving forces behind changes in zooplankton (Casini et al. 2009). With the position that zooplankton has in the food web – sandwiched between phytoplankton and fish (between eutrophication and overfishing) – data and understanding of zooplankton are a prerequisite for an ecosystem approach to management.

With respect to the eutrophication-driven alterations in food web structure, it has been suggested that with increasing nutrient enrichment of water bodies, total zooplankton abundance or biomass increases (Hanson & Peters 1984), mean size decreases (Pace 1986), and relative abundance of large-bodied zooplankters (e.g. calanoids) generally decrease, while small-bodied forms (e.g. small cladocerans, rotifers, copepod nauplii, and ciliates) increase (Pace & Orcutt 1981).

Total zooplankton abundance and biomass

In lakes and estuaries, herbivorous zooplankton stocks have been reported to correlate with chlorophyll *a* and phytoplankton biomass (Pace 1986; Nowaczyk et al. 2011; Hsieh et al. 2011), but also with total phosphorus (Pace 1986). In general, total zooplankton stocks increase with increasing eutrophication, which in most cases is a result of the increase in small herbivores (Gliwicz 1969; Pace 1986; Hsieh et al. 2011). Both parameters have been recommended as primary 'bottom-up' indicators (Jeppesen et al. 2011).

In most areas of the Baltic Sea, copepods contribute substantially to the diet of zooplanktivorous fish (e.g. sprat and young herring), and fish body condition and weight-at-age (WAA) have been reported to correlate positively to abundance/biomass of copepods (Cardinale et al. 2002; Rönkkönen et al. 2004). In coastal areas of the northern and central Baltic Sea, WAA has been suggested to be used as a proxy for zooplankton food availability and related fish feeding conditions to fish recruitment (Ljunggren et al. 2010).

Herbivorous zooplankton biomass is indirectly impacted by eutrophication via changes in primary productivity and phytoplankton composition, whereas direct impacts are expected mostly from predation, and to a lesser extent, from introduction of synthetic compounds (at point sources) and invasive species (via predation). The latter can also be indirect if invasive species are changing trophic guilds, which may affect zooplankton species. Finally, zooplankton abundance and biomass are affected – both positively and negatively – by climatic changes and natural fluctuations in thermal regime and salinity.

Mean zooplankter size

Evidence is accumulating that a shift in zooplankton body size can dramatically affect water clarity, rates of nutrient regeneration and fish abundances (Moore & Folt 1993). Although these shifts can be caused by a variety of factors, such as increased temperatures (Moore & Folt 1993; Brucet et al. 2010), eutrophication (Yan et al. 2008; Jeppesen et al. 2000), fish predation (Mills et al. 1987; Yan et al. 2008, Brucet et al. 2010), and pollution (Moore & Folt 1993), the resulting change implies a community that is well adapted to eutrophic conditions and provides a poor food base for fish. It has been recommended to use zooplankton size as an index of predator-prey balance, with mean zooplankton size decreasing as the abundance of zooplanktivorous fish increase and increasing when the abundance of piscivores increase (Mills et al. 1987).



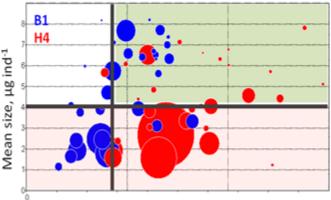
Human pressures linked to the indicator

	General	MSFD Annex III, Table 2a
Strong link	Fishery-induced mortality of larger zooplankters. Eutrophication leading to dominance of small-sized phytoplankton.	Biological - Extraction of, or mortality/injury to, wild species (by commercial and recreational fishing and other actitivies). Substances, litter and energy - Input of nutrients – diffuse sources, point sources, atmospheric deposition.
Weak link	 Higher salinity favouring larger zooplankters. Higher temperature favouring smaller zooplankters. Changes to oxygen concentration. Environmental contaminants. Invasive species. 	

The core indicator responds to fishing and eutrophication but also other pressures causing changes in the food web, such as salinity and temperature that are particularly relevant in the context of the Baltic Sea. Other pressures that might be involved are environmental contaminants and bottom hypoxia. The effects of fishery activities and eutrophication, although potentially co-occurring, would have different outcomes:

- Increased fishery that leads to increase in zooplanktivorous fish stocks affects both mean size and total zooplankton biomass negatively. Hence, declining trend in mean size and total stock (MSTS).
- Increased eutrophication and dominance of bacterio- and picoplankton leads to selective advantage for small-sized zooplankton. Hence, declining trend in mean size, but not total stock (Relevance figure 1) are likely to occur. In moderately eutrophied systems, an increase in abundance and/or biomass can be observed. The regression analysis conducted during the evaluation procedure, confirmed that all metrics in questions (MeanSize, total zooplankton abundance and total biomass) change significantly when both chlorophyll *a* and WAA values are outside of their reference conditions.





Total zooplankton biomass

Relevance figure 1. MSTS for two coastal stations (B1 and H4) in the Western Gotland Basin/northern Baltic Proper (years 1976-2010). Data are non-transformed mean values for summer (June-September) and circle size indicates average biovolume of filamentous cyanobacteria during the same period. In the Baltic Sea, the extensive cyanobacteria blooms are commonly considered a sign of eutrophication. Therefore, lower mean size observed during years with particularly strong blooms suggests negative effects of eutrophication primarily on mean size. By contrast, no clear effect on the total stock is apparent. Thick lines show threshold values and the green area corresponds to good status conditions.

In aquatic ecosystems, a hierarchical response across trophic levels is commonly observed; that is, higher trophic levels may show a more delayed response or a weaker response to eutrophication than lower ones (Hsieh et al. 2011). Therefore, alterations in planktonic primary producers and primary consumers have been considered among the most sensitive ecosystem responses to anthropogenic stress, including eutrophication (Schindler 1987; Stemberger & Lazorchak 1994).

Monitoring Requirements

Monitoring methodology

HELCOM common monitoring of relevance to the core indicator is described on a general level in the **HELCOM Monitoring Manual** in the <u>Sub-programme: Zooplankton species composition, abundance and biomass</u>.

Specific guidelines are under review with the aim to be included in the **HELCOM Monitoring Manual** at a later stage.

According to HELCOM guidelines for biological monitoring (HELCOM 1988), zooplankton were collected by vertical tows from either ~5 m above the bottom to the surface (shallow stations, \leq 30 m bottom depth) or by stratified tows (deep stations, \geq 30 m) as designed and specified by regional monitoring programmes. The standard sampling gear is a 100 μ m WP-2 net (diameter 57 cm) equipped with a flow meter.

Samples are preserved upon collection in formalin and analyzed by national laboratories within the respective monitoring programmes (see <u>Data</u> table 1). Copepods are classified according to species, developmental stage (copepodites CI-III and CIV-V classified as younger and older copepodites, respectively), and sex (adults); naupliar stages are not separated. Rotifers and cladocerans are identified to the lowest possible taxonomic level; moreover, the latter are classified according to sex, and females as ovigerous or non-ovigerous. Biomass is estimated using individual wet weights recommended by Hernroth (1985); for species not included in this list, either measured or calculated individual weights based on length measurements are used.

Current monitoring

The monitoring activities relevant to the indicator that are currently carried out by HELCOM Contracting Parties are described in the **HELCOM Monitoring Manual** in the Monitoring Concepts table.

Sub-programme:Zooplanktonspeciescomposition,abundanceandbiomassMonitoring Concepts table

Zooplankton monitoring stations are located in every Baltic Sea sub-basin. Most of the stations are offshore but there are also some coastal stations.

Time series of zooplankton used for setting thresholds value for mean size and total stock (MSTS) assessment are > 30 years. Due to considerable variations in the sampling frequency between the monitoring programmes and datasets, the data that are currently recommended for use in the MSTS assessment are restricted to the summer period (June-September) as the most representative in the currently available datasets (due to sampling schedules in the national monitoring programmes).

Description of optimal monitoring

In general, current monitoring is considered sufficient, although effects of the sampling frequency on the indicator performance remain to be evaluated. Evaluating the effect of sampling frequency on the indicator performance would be relevant for evaluating the confidence of the indicator.



Different strategies are employed in the national monitoring programmes with regard to sampling frequency and spatial coverage. In future work, this should be addressed to provide recommendations for zooplankton monitoring in the Baltic Sea.

If more resources are available, they should be used for development and implementation of methods for automated analysis and growth rate assessment that may complement standard analysis at the existing monitoring sites and provide specific information on zooplankton productivity.



Data and updating

Access and use

The data and resulting data products (tables, figures and maps) available on the indicator web pages can be used freely given that the source is cited. The indicator should be cited as following:

HELCOM (2018) Zooplankton mean size and total stock. HELCOM core indicator report. Online. [Date Viewed], [Web link].

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Metadata

Result: Zooplankton mean size and total stock Data: Zooplankton mean size and total stock

The data are provided by national monitoring programmes with HELCOM COMBINE parameters and methods. The indicator is based on routine data obtained within current monitoring schemes in the Baltic Sea, and is applicable in all areas where the programme is implemented. All HELCOM Contracting Parties carry out relevant monitoring.



Data table 1. Overview of the datasets used for MSTS evaluation for the period 2011-2016; deviations in the sampling methods from the HELCOM COMBINE guidelines are indicated.

Data set code	Area	Monitoring station(s)	Geographic coordinates	Max. sampling depth (m)	Time period (gaps)	Sampling frequency ^a	Deviations in sampling methods from HELCOM guidelines
ASKÖ	Western Gotland Basin	В1	N 58° 48' 19, E 17° 37' 52	40 m	1976-2010 (1990, 1993)	8-10	Water bottle ^b (1983-1988), otherwiseWP2, 90-µm mesh size ^c
Landsort	Western Gotland Basin					2-10	WP2, 90-µm mesh size ^c
GoFFI	Gulf of Finland	LL7 LL3A	N 59.5101, E 24.4981 N 60.0403, E 26.8020,	95 m 60 m	1979-2010 (1999, 2009) 1979-2010 (1989, 1990, 1999, 2000, 2009)	1 ^d	none
ÅlandFl	Åland Sea	F64	N 59.5101, E 24.4981	280 m	1979-2010 (1988- 1990,1997, 1999, 2009)		
BoSFI	Bothnian Sea	SR5	N 61.0500, E 19.3478	125 m	1979-2010 (1989, 1997, 1999, 2009)		
		US5B	N 62.3517, E 19.5813	116 m	1980-2010 (1989, 1997, 1999, 2009)		
BoBFI	Bay of Bothnia	BO3 ^e	N 64.1812, E 22.2059	100 m	1979-2010 (1989, 1990,1997- 1999, 2009)		
		F2 ^f	N 65.2302, E 23.2776	90 m	1979-2010 (1983, 1989, 1990,1997- 2000, 2009)		
Gdansk Deep	Gdansk Basin	P1	N 54°50.042′ E 19°19.683′	112 m	1986-2016 (1988, 1997- 1998, 2000- 2001)	1 ^d	

^a if not specified otherwise, this frequency is a number of samples collected during June-September;

^b 23-L water bottle was used to sample water column every 5 m (bottom to surface) and pooled for counting using a 90-μm sieve; ^c WP2 nets with mesh size of 90 and 100 μm were compared in 2003 in the Western Gotland Basin/northern Baltic proper and found to provide statistically similar sampling efficiencies for all relevant zooplankton groups (Gorokhova, pers. observations); ^d August;

^e or stations BO3N and/or BO3S located in a close proximity;

^f or station F2A located in a close proximity;

^g total for all stations



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Archive

This version of the HELCOM core indicator report was published in July 2018:

Zooplankton mean size and total stock HELCOM core indicator 2018 (pdf)

Older versions of the core indicator report are available:

HOLAS II component - Core indicator report - web-based version July 2017 (pdf)

2013 Indicator report (pdf)

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