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## Background

STATE & CONSERVATION 4-2015 discussed HELCOM monitoring guidelines for NIS in the Baltic Sea (paragraphs 2MA.6 - 2MA.13 of the outcome). STATE & CONSERVATION 4-2015 considered the proposal by Germany to apply the Extended Rapid Assessment Survey of Alien Species as minimum monitoring in the Baltic Sea (para 2MA.6 of the outcome) and recalled that, except for in Estonia, there is no regular monitoring of NIS in the Baltic Sea. It was noted that Extended Rapid Assessment is restricted to benthic organisms while the HELCOM/OSPAR Port survey protocol also includes planktonic organisms, mobile fauna and pathogens and that Extended Rapid Assessment cannot be used for the requirements under the IMO Ballast Water Management Convention.

STATE & CONSERVATION 4-2015 did not conclude on the issue of HELCOM monitoring guidelines for NIS and recommended further discussion on suitable guidelines. It was proposed that combining the Rapid Assessment protocol and the Port Survey protocol could be considered and that monitoring data stemming from this approach could be complement with NIS observations stemming from biological monitoring programmes (Outcome, para 2MA-11).

This document contains a proposal for an integrated NIS HELCOM monitoring programme, which combines all types of surveys and approaches, which may provide information on NIS findings, establishment and spread, including routine HELCOM biological monitoring (HELCOM COMBINE), Port Biological Baseline Surveys (HELCOM/OSPAR), Extended Rapid Assessment Survey (RAS), HELCOM coastal fish monitoring and other already ongoing and developing monitoring approaches. The proposal recommends using AquaNIS, the Information system on Aquatic Non-Indigenous and Cryptogenic Species, as a central database for data storage, analysis and reporting. Monitoring guidelines for the Extended Rapid Assessment Survey, one of the methods that could be considered as part of the integrated HELCOM NIS monitoring programme, will be provided as a separate meeting document (document 5J-2).

## Action requested

The Meeting is invited to consider the proposal on an integrated HELCOM NIS monitoring programme and amend it as appropriate.

# Suggested outline for non-indigenous species monitoring for HELCOM monitoring guidelines

## 1. Background

### 1.1. Introduction

Non-indigenous species (NIS) are species introduced outside of their natural range (past or present) and outside of their natural dispersal potential by intentional or unintentional human activities. Natural shifts in distribution ranges, e.g. due to climate change or dispersal by ocean currents, do not qualify a species to be a NIS. However, secondary spread of NIS from the area(s) of their first arrival may occur without human involvement due to dispersal by natural means. The most common pathways for NIS introductions into the Baltic Sea will have been vessels (via ballast waters and as ships' and leisure craft biofouling), then deliberate stocking into the wild (especially during the 1960s and 1970s) and natural spread of NIS from neighboring regions, mainly from the North Sea, but also from inland waters (Ojaveer et al., 2017).

Some NIS may reach a high abundance, become widespread and can pose harm to the environment, economy and/or human health. However, it is very difficult to predict which of the NIS may become invasive and result in detrimental effects. This is why biological invasions remain high on the environmental conservation and management agenda. At the European level, the EU Regulation on the Prevention and Management of the Introduction and Spread of Invasive Alien Species (EC, 2014) was adopted, indicating, inter alia, "*...Prevention is generally more environmentally desirable and cost-effective than reaction after the fact, and should be prioritized...*". Here, the clear distinction is made between the primary introduction of an alien species, which should be prevented, and its secondary spread within a region, which in the marine realm is practically unmanageable. Several legal and administrative instruments aimed to reduce the spread of NIS were developed at international and national levels, such as the International Convention for the Control and Management of Ship's Ballast Water and Sediments (BWMC; IMO, 2004), the International Council for the Exploration of the Sea (ICES) Code of Practice on the Introductions and Transfers of Marine Organisms (ICES, 2005), the EC Regulation on Invasive Alien Species (IAS, EC 2014) and the Marine Strategy Framework Directive (MSFD; EC, 2008). Finally, at the regional scale, the HELCOM Baltic Sea Action Plan (BSAP) sets the environmental management objective "*...No introductions of alien species from ships...*" (HELCOM, 2007).

### 1.2. Purpose and aims

A regionally harmonized, scientifically based NIS monitoring program is the only possible way to measure the effectiveness of the above-mentioned legal and administrative instruments (Ojaveer et al., 2014). However, it is not practical to establish separate monitoring programs to serve the needs of each international or regional instrument. The principle of the integrated NIS monitoring program should be "*...Collect once, use many times...*", this is a frequently cited principle in both national and international efforts to promote the collection, archiving and sharing of marine monitoring data (Whomersley et al., 2015).

As most of the new NIS are transported into the Baltic Sea via shipping, routine marine monitoring should be complemented with the port biological monitoring. Ports with the most intensive international ship traffic should be taken under regular monitoring. This monitoring should be conducted following the Joint HELCOM/OSPAR Harmonized Procedure tailored for the Baltic Sea conditions to fulfill the data requirements of the BWM Convention (for exemption procedures). As the protocol includes sampling of phytoplankton, zooplankton, soft bottom benthos and mobile and sessile epifauna on hard surfaces, it covers all taxonomic groups of NIS and thus gives valuable information needed not only for the BWMC for granting exemptions but also for the MSFD and IAS regulation purposes.

The NIS monitoring program should integrate all types of surveys and approaches, which may provide information on NIS findings, establishment and spread, including routine HELCOM biological monitoring (HELCOM COMBINE), Port Biological Baseline Surveys (HELCOM/OSPAR), HELCOM coastal fish monitoring, Baltic International Trawl Surveys (BITS), public observations, rapid assessments, etc. The results of NIS

monitoring will provide baseline information for serving many purposes should the data be collected within one database where it is available for indicator updates, reporting purposes and research. For example, such monitoring system will determine the number of new arrivals in periodic assessments for MSFD purposes (HELCOM, 2012; Olenin et al., 2016a). Also, it will provide substantially more information needed for making decisions on granting exemptions under BWMC (Olenin et al., 2016b).

The main aims of NIS monitoring program are:

- early detection of NIS in high-risk invasion areas and dispersal hubs, along with determination of the pathways and vectors involved,
- assessment of new arrivals at different geographical scales, from a country coast within the Baltic Sea to the entire sea basin, and even to a larger biogeographical scale (Baltic and the North seas together),
- determination of spread and rate of expansion of established NIS, along with evaluation of the long-term trend of NIS population abundance/biomass;
- evaluation of environmental effects of NIS on the structure and function in the invaded ecosystems.

## 2. Methods for the integrated NIS monitoring program

### 2.1. Surveys and monitoring approaches already ongoing

#### 2.1.1. HELCOM/OSPAR Port survey protocol

The NIS monitoring program should integrate both targeted and non-targeted methods. Such targeted method as the HELCOM/OSPAR Port survey protocol provides information on NIS found in ports to support decisions on granting exemptions (HELCOM, 2015). Information obtained during port surveys should also be used to complete NIS assessments for HOLAS and the MSFD (D2) purposes.

#### 2.1.2. HELCOM biological monitoring program

The HELCOM biological monitoring program (HELCOM COMBINE), which dates back to the early 1980s and has comprehensive quality control system, initially was not targeted on NIS. However, several plankton and soft bottom macrofauna species were first found at the routing monitoring stations (AquaNIS, 2016 and references therein). This well-established monitoring system should be used for records of presence-absence of NIS, thus providing the number of NIS found in a given area, and also temporal occurrence and spatial distribution of NIS (Lehtiniemi et al., 2015). The present coordinated COMBINE should be complemented with a regular shallow hard bottom fauna and flora monitoring.

#### 2.1.3. Surveys of fish, birds and underwater habitats

Another non-targeted approach, which may provide information on NIS presence-absence and spread, is HELCOM coastal fish gill-net monitoring and BITS surveys. During such surveys, non-indigenous fishes and mobile epifauna (e.g. crabs) can be caught and such records should be made available for the national authority responsible for managing NIS records (for example, see Ojaveer et al., 2011). Ornithological surveys performed according to MSFD requirements or for other purposes may detect bird species, which clearly are non-native and could be introduced by humans, like Canada goose (Nummi, 2002). Also, underwater habitat surveys, which are being conducted in many Baltic Sea countries, may provide information on the spread of conspicuous NIS such as mobile epifauna (e.g. crabs) and habitat engineers, e.g. the zebra mussel. The use of unmanned and preprogrammed underwater vehicles may pictographically survey extensive areas of seabed at comparatively low overall cost.

#### a. Other useful surveys and monitoring approaches

A variety of approaches and methods have been and are being developed, which may be used for NIS monitoring purposes to complement ongoing monitoring programs.

### 2.2.1. Rapid assessment surveys

A rapid assessment survey (RAS) is a method to detect species that can be recognized in the wild from conspicuous morphological characteristics and whose abundance and distribution can be determined for a particular area. RAS may be part of a regular monitoring program and may also be activated following a particular NIS event, e.g. a report of a NIS finding, requiring confirmation. Rapid assessment monitoring for targeted species enables direct reporting to management when a notable species is encountered and the field work can be undertaken by a small group of experts (Lehtiniemi et al., 2015).

Target lists of NIS reduce sampling effort, over full inventories of biota present, and are more relevant for a swift management response (Lehtiniemi et al., 2015). One approach to select NIS for a RAS is to follow IMO (2007) definition of target species: “...*Species identified by a Party that meet specific criteria indicating that they may impair or damage the environment, human health, property or resources and are defined for a specific port, State or biogeographic region...*”. However, not all NIS may be easily recognized in the field and further systematic examination in laboratory may be required. This, in turn, may essentially increase time needed to obtain RAS results.

The selection of the sampling sites should be based on the analysis of most likely “entry” points/hubs where introductions are likely to arrive and “hot spots” containing elevated numbers of NIS, such as ballast water discharge areas, docks, marinas and aquaculture sites with stock movements by undertaking rapid surveys for targeted species (Ashton et al., 2006; Minchin, 2007). In addition, areas of special interest or concern, such as nature conservation sites, should be included in lists of the sampling sites. It is obvious that RAS cannot be conducted equally in the countries with an extensive coastal marine environment; therefore, careful selection of the representative sampling sites and adequate research methods should be performed prior to the start of any large scale field campaign (Olenin et al., 2011).

The time periods to perform RAS may vary depending on the purpose. An *ad hoc* decision may be taken and a survey may be activated following a particular NIS event, e.g. a report of NIS finding, requiring confirmation, especially if potentially harmful species are targeted. RAS within a regular monitoring program may be performed in different places during a certain period, especially in countries with extended coastline. The periodicity of such survey should be harmonized with the six years reporting period for MSFD.

Decision on RAS sampling areas and periodicity will be taken by national authorities, but it is feasible to coordinate such surveys regionally. RAS focused on selected target species may be arranged simultaneously by several countries within the Baltic Sea, in the same way as it is done for fishery surveys (e.g. ICES, 2014).

RAS can provide sound semi-quantitative information on the status of the NIS within the environment even before they become established. Such surveys are undertaken with a small investment of time, effort and cost and may lead to the evaluation of the mode of arrival and potential impacts of NIS in marine ecosystems (Minchin, 2007). Protocols for rapid assessment of marine and coastal biological diversity are available (Pedersen et al. 2005; Patrick et al., 2014) and the method proved to be useful in many aquatic regions (Pedersen et al. 2005; Minchin, 2007; Marić et al., 2016; Minchin et al., 2016).

The RAS method is cost-efficient and may provide timely information for managers and policy-advisers focusing on particular NIS at particular localities. However, there are substantial limitations using this approach, for example, the RAS method alone do not cover some important groups of organisms (see Table 2), and therefore cannot provide full set of data needed even for the HELCOM CORE indicator. Thus, the RAS approach alone should be considered as unsuitable for the long-term targeted NIS monitoring (Lehtiniemi et al., 2015).

### 2.2.2. Monitoring of Marine Protected Areas

A Marine Protected Areas observation program was used to identify occurrences of NIS around the UK coast. A standard list of NIS was compiled against which, infauna and epifaunal data records from the MPAs were compared and reported to the appropriate national authorities (Whomersley et al., 2015). Such approach may be used in the Baltic Sea NIS monitoring program. Within MPAs monitoring programs a

series of conventional and novel methods of surveillance are likely to become part of a protocol, and their potential use for detection of NIS should be considered.

### 2.2.3. Molecular methods

Molecular methods are rapidly evolving which may become established within monitoring protocols. Such methods will be helpful in determining marine NIS identities, as well as their sources, routes of invasions, and the genetic make-up of founding populations (Lehtiniemi et al., 2015). Recent advances in molecular methods offer promising tools for assessment of early detection (Zaiko et al., 2015), fundamental connectivity within and among source and introduced populations, invasion dynamics, trophic ecology, interactions between NIS and native species – all of which are essential for quantifying their effects on recipient communities (Pochon et al., 2013). Molecular methods can also be used for the rapid identification of cholera bacteria (Mountfort et al., 2012) and NIS can be identified from genetic profiles within water (Zaiko et al., 2015). Molecular genetics methods are particularly useful for NIS detection at early life-history stages (due to difficulties in their identifications) and at initial stages of invasions or when occurring at low densities (Lehtiniemi et al., 2015).

### 2.2.4. Automated image analysis

Another rapidly developing approach is automated systems, which may pick up unfamiliar biological shapes (Culverhouse et al., 2006). Such in-situ continuous monitoring capacity initially images aliquots of sea water and rejects images of low-risk objects. This 'pre-filtering' identification method can reduce human sampling time by diminishing the false positive and false negative identification rates (e.g. Verikas et al., 2012). A managed web-based image database may be developed that acts a repository for images of identified NIS, together with metadata reflecting the scale of the object, its location, depth and date of image collection, and collector (Olenin et al., 2011). Although semi-automated classification of zooplankton allows increasing the number of processed samples cost-effectively, taxonomic accuracy of such methods still is limited, but it might be overcome in the future (Uusitalo et al., 2016). Currently, these methods may be used for abundance estimates of already known NIS in the sample which are identifiable by automatic image analysis.

### 2.2.4. Public involvement

Public involvement can aid in detection of NIS. Divers, anglers, leisure craft users, students and schoolchildren help to track the spread of NIS. Volunteers (citizen scientists) may look for a restricted number of species, and the data can be used to identify range expansions (Lehtiniemi et al., 2015). Partnerships with the aquaculture, fisheries and leisure craft industries may enable early detection of NIS arrivals. The advent of electronic communication facilitates the usage of online websites in reporting NIS observations. Websites also aid in providing up-to-date information on identification, distribution and means of preventing further spread. Public involvement increases general awareness on the NIS problem and may therefore help in preventing further intentional introductions (Lehtiniemi et al., 2015). In Finland, public observations are used already to gather information on NIS findings. For marine species, these are verified by experts and used for e.g. EU reporting. Also there are good examples of using citizen science for NIS monitoring purposes in the North Sea (Gittenberger et al., 2010), Mediterranean Sea (Bodilis et al., 2014), and for some species in the Baltic Sea (Katajisto et al., 2013).

### 2.2.5. Environmental impacts

Monitoring and assessment of environmental impacts of NIS is important for prioritizing management options, developing of target list of NIS, reporting for MSFD and IAS purposes (Lehtiniemi et al., 2015). EC Decision (2010/477/EU) recommends using "Impacts of non-indigenous invasive species at the level of species, habitats and ecosystem" for assessment of environmental status of marine waters. In MSFD assessments, several EU countries (Spain, Portugal, Estonia, Finland and Lithuania) used the Biopollution level (BPL) assessment method (ICES, 2016). The BPL method is based on a classification of the abundance and distribution range of NIS and numerically expresses the magnitude of their impacts on communities, habitats and ecosystem functioning aggregated in a BPL index which ranges from 'no impact' (BPL=0) to 'massive impact' (BPL=4) (Olenin et al., 2007). BPL is a semi-quantitative method, which has been used for impact assessments of macrozoobenthic NIS in the German Baltic estuaries and lagoons (Wittfoth, Zettler,

2013), and also various NIS at the pan-Baltic scale (Zaiko et al., 2011). It was also applied to assess the impacts of a single NIS, the dinoflagellate *Prorocentrum cordatum* (former *P. minimum*), based on the long-term phytoplankton monitoring data (Olenina et al., 2010). An online application BINPAS (Biological Invasion Impact / Biopollution Assessment System) is developed to aid the assessment of BPL and store relevant data (Narščius et al., 2012).

### 2.3. Sampling frequency

The suggested sampling frequency of various organism groups is different, depending on the life-history traits of organisms and trophic levels, which they belong to (Table 1).

Table 1. Suggested sampling frequency requirements for monitoring of presence–absence and population dynamics (abundance and/or biomass) of NIS of different taxonomic groups and varying life cycle lengths (Lehtiniemi et al., 2015 updated; Klais et al., 2016).

Organism group	Presence/absence	Population dynamics
Pathogens and other disease agents	Seasonal	Variable
Phytoplankton	Seasonal	Frequent, depending on biosecurity requirements
Zooplankton	Seasonal	Bi-weekly
Benthic vegetation	Seasonal/Annual	Seasonal/Annual
Zoobenthos	Annual	Annual
Fish	Annual	Annual at specific times ( <i>e.g.</i> , reproduction)

### 2.4. Summary of surveys and monitoring approaches

Thus, there are a broad suite of methods, which should be used for different types of NIS monitoring and are appropriate for detection of different groups of NIS (Table 2). Integration of existing monitoring schemes and developing surveillance approaches should be a grounding principle of the Baltic Sea NIS monitoring program.

Table 2. Existing (x) and potential (p) monitoring and surveillance approaches to be used in the Baltic Sea non-indigenous species monitoring program: Port surv. – HELCOM/OSPAR port survey protocol; Biol. mon. – HELCOM biological monitoring program (HELCOM COMBINE); Fish surv. – young fish and gill-net surveys, Bird surv. – ornithological surveys; UW hab. – underwater habitat mapping programs, RAS - Rapid assessment survey; MPA – monitoring of marine protection areas, eDNA –environmental DNA assessment (molecular methods); Public – involvement of citizen science (xp, already used in Finland and potentially can be expanded to other Member States); BPL – biopollution level index.

Groups of NIS	Port surv.	Biol. mon.	Fish surv.	Bird surv.	UW hab.	RAS	MPA	eDNA	Public	BPL
Human pathogens	x							p		
Phytoplankton	x	x						p		x
Zooplankton	x	x	p					p		x
Macrophytes	x	p			p	p	p	p	p	x
Soft bottom macrofauna	x	x	p			p	p	p		x
Hard bottom macrofauna, incl. fouling organisms	x	p			x	p	p	p	px	x
Mobile epifauna	x		x			p	p	p	x	x
Fish	x		x		x		p	p	px	x
Birds				x		p		p	px	p
Mammals						p		p	px	p
Parasites			p	p						

### 3. Data reporting, storage and analysis

A centralized database is the key element of the proposed integrated NIS monitoring system. There is little value in monitoring NIS unless the knowledge obtained is timely and can be directly used by research and management (Ojaveer et al., 2014). The organizational principles, structure and functionality of a NIS database should allow for the assembling, storing and disseminating of data compiled from various marine monitoring programs (Ojaveer et al., 2014; Lehtiniemi et al., 2015). Such information system should include data on:

- valid taxonomy of NIS, including notes on availability of molecular information;
- introduction event records at the level of particular countries, country regions and ports;
- introduction pathways and status of NIS populations in the invaded countries, country regions and ports;
- biological traits and environmental tolerance limits of NIS;
- documented evidence of NIS found in ballast water and/or on ship hulls;
- impacts on environment, incl. ecosystem structure and functioning.

It is recommended to use AquaNIS, the Information system on Aquatic Non-Indigenous and Cryptogenic Species (AquaNIS, 2016), which meets all above requirements and contains the most up-to-date and free-access information/data on NIS introduction events within the Baltic Sea, neighboring regions (e.g. North Sea) and other regions of the world. The system was developed in the course of the EU FP7 funded VECTORS project. It inherited and incorporated multiple NIS data collections from earlier projects and initiatives, including the Baltic Sea Alien Species Database, earlier supported by HELCOM. AquaNIS is regularly (at least annually, but sometimes more frequently) being updated by national reports to the ICES Working Group on Introductions and Transfers of Marine Organisms and ICES/IOC/IMO Working Group on

Ballast and Other Ship Vectors (ICES WGITMO, WGBOSV), and new records obtained within several ongoing regional and national research projects (such as BONUS BIO-C3).

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