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Technical Guideline for Sturgeon Habitat Monitoring

Support Document for the Implementation of the Pan-European Action Plan for Sturgeons

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Once the Guideline has been approved by the Commission, the results of this Guideline will belong to the European Commission who may decide to publish them in due course.

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1 Executive Summary

Sturgeon populations in European rivers and coastal waters have undergone a dramatic decline over the last 150 years. In addition to overharvest, the intensive developments of hydropower and river channelization have led to massive habitat loss and fragmentation affecting all stages of their life-cycle. As a consequence, all eight sturgeon species found in European waters are threatened with extinction (International Union for Conservation of Nature - IUCN) and are reported as being in "unfavourable" conservation status within the frame of the reporting under Article 17 of the Habitats Directive.

To improve this situation, the Pan-European Action Plan for sturgeons (PANEUAP) was adopted by the Standing Committee to the Convention on the Conservation of European Wildlife and Natural Habitats (Bern Convention) in the form of Recommendation No. 199(2018) and endorsed for implementation under the Habitats Directive and provides a guiding framework of actions to be implemented in sturgeon range countries by regional stakeholders including regional sea and river commissions.

The Action Plan requests all signatory countries to *"restore all existing sturgeon populations to "least concern" (IUCN) or "favourable" status and re-establish self-sustaining sturgeon populations as well as their life-cycle habitat in their historic range to an extent that ensures species survival and representation of the subpopulations where possible."*

The Action Plan underlines the importance of functional habitats and migration corridors as prerequisite for the long term survival of self-sustaining sturgeon populations. Therefore, the purpose of this technical guideline is to specifically support the implementation of the Action Plan's Objective 3 "Sturgeon habitats are protected and restored in key rivers" as well as Objective 4 "Sturgeon migration (up- and downstream) is secured or facilitated".

The sturgeon life-cycle requires specific habitats and their accessibility to support specific habitat use such as spawning, feeding, and wintering to ensure viable populations. Therefore, sturgeons are key indicators for the ecological integrity of rivers (Schiemer, 2000), as their habitats may be distributed over whole catchments and adjacent marine areas.

As such, the rapid identification and monitoring of sturgeon habitats and habitat use is crucial for habitat protection and restoration, as well as for understanding the dynamic interactions between populations and their environments and to further inform sturgeon conservation measures.

Habitat monitoring is defined for this document as recurring measures to document habitat functionality, which also includes assessments for habitat identification, verification, and confirmation of habitat use as basic prerequisites. All such measures should be part of a habitat monitoring program; synchronized and coordinated with all other aspects of sturgeon conservation and restoration in a given system.

A four-step approach for a habitat assessment and monitoring program for sturgeon is recommended:

1. **Identification of areas of past presence and general relevance through data and information research** – The collection and analysis of relevant information on former habitat and waters, as well as areas and sections bearing the characteristics of sturgeon habitats. This should include data mining on specific sturgeon biology, ecology, and former habitats with regard to species presence, accessibility (migration barriers), population dynamics, distribution ranges, habitat use, life-cycle, hydromorphology, and hydrology, as well as data from open water monitoring and field assessments documenting general water quality, but also on specific habitat traits such as substrates and current velocities.

2. **Verification of habitats** – The measuring and documentation of actual relevant environmental variables in the field targeting specific locations, timings, and conditions, including their quantification in the form of temporal and spatial extensions, as well as their frequency with a given system, supported by statistical habitat modelling.

3. **Confirmation of actual habitat and habitat types** – The observation of sturgeon presence and movement in rivers and sections as a first step, and the documentation of actual habitat use and its concrete results in a second step, mainly by population monitoring.

4. **Recurring and real-time monitoring measures** – The measurement and documentation of the ongoing functionality of habitat and habitat types over time by both observing and measuring relevant environmental variables and by population monitoring.

Relevant criteria for habitat identification and verification, as well as methods for their documentation and confirmation, are provided in this document. Criteria for the determination of recurrent and real-time monitoring measures, a monitoring road map, an exemplary work plan including examples from sturgeon research, messages for decision makers, and a list of relevant literature complete this technical guideline on sturgeon habitat monitoring.

2 Introduction

Eight sturgeon species are native to Europe's rivers and seas, and all are featured in the IUCN Red List of Threatened Species. Seven of the eight species have been assigned "critically endangered" status. Sturgeon, although threatened with extinction, are key indicators for the ecological integrity of rivers, as habitat for completing the life-cycle may cover entire catchments (Schiemer, 2000). Sturgeon are thus considered flagship species for many conservation actors for healthy and free flowing river systems.

Reflecting the high risk of extinction for this species group, a Pan-European Action Plan for Sturgeons (PANEUAP)1 was adopted by the Standing Committee to the Bern Convention in the form of Recommendation No. 199(2018) to which all important European sturgeon range countries, as well as both the EU and its Member States, are Parties. In May 2019, the EU Nature Directive Expert Group (NADEG) also recommended the implementation of the Action Plan to EU Member States. The PANEUAP was designed to serve as a framework of almost 70 actions which aim to *"restore all existing sturgeon populations to "least concern" (IUCN) or "favourable" conservation status (Habitats Directive) and re-establish self-sustaining sturgeon populations as well as their life-cycle habitat in their historic range to an extent that ensures species survival and representation of the subpopulations where possible."*

The recommendation mandated the Secretariat of the Bern Convention to closely monitor the implementation of the Action Plan and to coordinate the implementation of regular reporting on the implementation of the Action Plan at national levels.

Since its adoption, the European Commission has closely followed the implementation of the PANEUAP and in 2022 issued a service contract (09.0201/2022/885601/SER/D.3) to support its implementation. The scope of the contract covers the assessment of the implementation of the PANEUAP in 18 key sturgeon range countries, including 15 EU Member States (Romania, Bulgaria, Croatia, Slovenia, Hungary, Slovakia, Austria, Germany, Italy, Poland, Lithuania, Latvia, Estonia, France, and The Netherlands) as well as Serbia, Ukraine, and Georgia. Existing knowledge about sturgeon habitats and migration obstacles in 11 key river basins including Danube, Rioni, Po, Vistula, Oder, Nemunas, Gauja, Narva, Elbe, Rhine, and Gironde have been collected and displayed in maps (Popp, S., 2024. Characteristics and locations of Sturgeon Habitat in European Rivers). Further, the contract encompasses (1) a study about sturgeon bycatch and possible measures to avoid or mitigate it, (2) technical guideline for sturgeon population monitoring, (3) technical guideline for habitat monitoring as well as a (4) technical guideline for best practice *ex situ* breeding and release programs.

This document presents the technical guideline for habitat monitoring supporting explicitly the implementation of the PANEUAP objective 3, "Sturgeon habitats are

[1] https://rm.coe.int/pan-european-action-plan-for-sturgeons/16808e84f3

protected and restored in key rivers", focusing specifically on identification and monitoring of habitats. It also supports objective 4 ""**Sturgeon migration (up- and downstream) is secured or facilitated". This Guideline** complements the Technical Guideline for Population Monitoring developed under the same service contract. The population and its habitats are ecological twins and cannot be sustained without the other. For a successful and sustainable conservation approach, a rapid implementation of measures targeting viable sturgeon populations and a functional habitat are needed. Restoring populations, habitats, or migration routes requires substantial resources, political will, and a sound knowledge base, all of which are required to make informed decisions concerning the conservation priorities.

In general, as all sturgeon species are protected under the Habitats Directive (Council Directive 92/43/EEC) EU Member States are obliged to ensure that the species covered by the Directive are maintained, or restored, to a favourable conservation status throughout their natural range within the EU. The monitoring of conservation status includes assessment on the availability of the species habitat and is an obligation arising from Article 11 of the Directive for all species (as listed in Annex II, IV and V) of community interest. The specific reporting obligation derives from Article 17, with the reporting for the conservation status assessment to be repeated every six years (last available report 2013-2018). For 3 species, Acipenser naccarii, *A. oxyrinchus* and *A. sturio* listed on Annex II and IV Member States are required to "designate core areas of their habitat as sites of Community importance and are to be included in the Natura 2000 network to be managed in accordance with the ecological needs of the species" and to "apply a strict protection regime across their entire natural range, within and outside Natura 2000 sites" (art 12 & 16).

At the same time the Convention on the Conservation of European Wildlife and Natural Habitats (Bern Convention) also aims to protect sturgeons and their habitats. The Conventions *Emerald Network is an ecological network made up of areas of special conservation interest (ASCIs), subject to management, monitoring and reporting measures, with the objective to ensure the long-term survival of the species and habitats of this treaty that require specific site protection measures.*

Keeping mind the flagship status of sturgeons, improved knowledge about their habitat and migration corridors, thus also supports the better implementation of the Habitats Directive and the Bern Convention in a wider sense and contributes to the ecosystem approach, necessary to maintain Europe's biological diversity.

2.1 Monitoring – a basis for decision making

The status of long-distance migratory fish populations is an excellent indicator for the functions of ecological corridors, the existence of sufficient breeding, feeding, and wintering habitats, habitat accessibility and connectivity, as well as ecosystem health, resilience and water quality in general.

The Pan-European Action Plan for Sturgeons provides the following Actions as **rationale and basis for habitat monitoring** within a general sturgeon restoration and conservation approach under **Objective 3 - Sturgeon habitats are protected and restored in key rivers:**

- 3.1.1 Identify existing critical habitats (time and location/conditions and resources) leading to a common database
- 3.1.2 Ensure legal protection of identified priority habitats and their functions
- 3.1.3 Identify conflicts and common interests between economic development plans, identified habitats and their functionality
- 3.1.4 Mitigate conflicts between economic development and the ecological requirements and functions of sturgeon habitat
- 3.2.1 Identify habitat restoration possibilities
- 3.2.2 Develop an integrated concept for the restoration of key habitats to reach near natural ecosystem functions, providing sufficient carrying capacity for self-sustaining sturgeon population in a given river basin
- 3.2.3 Implement pilot restoration actions
- 3.2.4 Monitor the habitat quality in pilot restoration actions, with special emphasis on criteria relevant for sturgeons

Also, under **Objective 4: Sturgeon migration (up- and downstream) is secured or facilitated:**

- 4.1.1 Prohibit any further construction of migration obstacles based on existing legislation, laws, treaties, and conventions
- 4.1.2 Establish legal prerequisites for future in-river construction development, including a minimum bypass with suitable conditions for fish migration of 30% of the discharge at all times
- 4.2.1 Identify relevant obstacles for sturgeon migration
- 4.2.2 Prioritize mitigation of migration obstacles according to criteria such as: existing stocks, former habitat, existing or former spawning sites, river length, existing habitat, and recolonization potential
- 4.2.3 Conduct feasibility studies (comprising hydrological and hydrodynamic monitoring and modelling and fish monitoring (telemetry, Didson sonar, etc.)) for facilitating up and downstream migration at highest priority barriers (based on results of 4.2.2)
- 4.2.4 Allocate funds for feasibility studies as well as mitigation measures
- 4.2.6 Implement functional passage solutions (proven by monitoring results)

- 4.2.7 Establish monitoring guidelines, identify suitable devices, and implement programs to assess fish pass efficiency
- 4.4.2 Monitoring of distribution, migration patterns, and behaviour of sturgeon populations on a catchment basis in marine and freshwaters

The knowledge gained deriving from population and habitat monitoring can serve multiple uses and would provide the best available knowledge to make data-driven decisions and to implement evidence-based management.

Any measures to protect and improve ecological corridors will highly benefit from habitat monitoring data, ranging from e.g. the prevention or mitigation of detrimental interventions, the active restoration of critical spawning, feeding, or wintering habitats or provide an important information base for large investments, such as the necessary and priority setting for the implementation of fish passages.

Monitoring sturgeon habitat is crucial for understanding the dynamic interactions between sturgeon populations and their environments, and the identification of potential key habitats through documentation of the use of habitats by sturgeons and the adverse impacts upon them. A general understanding of the functionality of these habitat types is essential to:

- Make informed decisions to plan and implement targeted habitat protection measures.
- Understand key factors affecting sturgeon distribution, reproductive success, and population viability.
- Assess impacts of existing infrastructure upon habitat quality, functionality, and suitability.
- Forecast effects of planned infrastructure developments in the frame of Environmental Impact Assessments, general pre-planning processes or plans for appropriate mitigation measures.
- Decide on necessary protection measures for habitats including the creation of dedicated protected areas, or spatial limitation of specific activities.
- Plan and implement measures to restore key habitats or ecological corridors for migration in cases these are not functional.

Ultimately, population and habitat monitoring are essential to evaluate and confirm the potential success or failure of the measures undertaken in the frame of the PANEUAP.

The guideline at hand shall provide guidance to responsible national authorities and institutions on best practice approaches to design monitoring programs or to decide on funding priorities. Descriptions of methodologies and technologies, their purpose, and advantages and disadvantages provide orientation and guidance for practitioners to develop their own individual solutions to implement targeted methodological approaches to address specific research questions and to close existing knowledge gaps.

Technical chapters are complemented with a compilation of the main pros and cons, required resources, practical examples from the field of applied science, and a compilation of key references on the respective topic for further reading and research.

2.2 Scope

The information given in this guideline is based on the general similarities of species within the sturgeon family of *Acipenseridae*. However, variations exist between species and also catchments, and as such there are still gaps in knowledge with regard to habitat conditions and requirements. Whenever available, specific information is provided, but it is the responsibility of the document user to do one's own research and adapt the given information as needed.

There is a clear focus on riverine freshwater habitats for sturgeon in the document at hand for two reasons:

1. All sturgeons require freshwater for reproduction and spawning sites as well as sites for early development play a pivotal role for the viability of populations.
2. Therefore, much of the available information and literature deals with the freshwater phase of the sturgeon life-cycle.

Major knowledge gaps that need to be filled with regard to habitat criteria still exist for certain species, catchments and specific life-cycle stages.

Apart from acquiring additional information on the system to be worked on, the basic principles of the "four Cs" should be applied with regard to any activities for sturgeon habitat monitoring, within a system of interconnected habitats and populations.

These principles are:

- **Consistency** – build on knowledge and results that have already been achieved/ established.
- **Compatibility** – strive for a standardization and harmonization of methodologies and technologies within and among managing jurisdictions.
- **Communication** – network within the "habitat community" and beyond, share your own expertise, experience, and data for the sake of the common cause.
- **Collaboration** - build on already existing resources and team up with other actors and stakeholders. Sturgeon conservation including sturgeon habitat monitoring requires the involvement of different fields of expertise, disciplines, and jurisdictions in a coordinated approach.

2.3 Supportive tools - Mapping and data base solutions

The use of Geographic Information Systems (GIS) in combination with data base storage solutions in habitat monitoring is recommended for both the preliminary mapping of relevant areas and sections in the system, based on data mining and preliminary surveys, as well as for the documentation of the results of habitat verification, confirmation, and recurrent monitoring.

GIS offers a powerful toolset to gather, store, process, analyze, and present diverse information and data for aquatic habitat monitoring. It integrates various forms of spatial information, such as maps, satellite imagery, topography, and georeferenced data into one digital platform and allows for the visualization, analysis, and interpretation of spatial relationships by layering them. This enables researchers to gain additional insights and make informed decisions regarding habitat use, distribution, and conservation efforts. It also facilitates interdisciplinary approaches to habitat monitoring by the integration of information and data from different fields such as ecology, hydrology, geology, and human land and water use (Gordon 1994). The potential benefits of GIS in the context of habitat monitoring are:

- **Layered Information**: GIS allows overlaying various data layers, such as potential and actual habitat types, results from population monitoring, historic catch locations, water depths, substrate types, temperature, migration routes and barriers, in association with human activities, for example. This layering enhances the identification of habitat areas within the system, potential threats, and conservation opportunities (a selection of basic layers can be found at https://www.copernicus.eu/en/copernicus-services).
- **Spatial analysis**: GIS enables an advanced spatial analysis, such as habitat suitability modeling and connectivity assessments, facilitating the identification of habitat type arrays and potential migration corridors for sturgeon.
- **Temporal trends**: By storing current but also historical data, GIS allows for tracking changes in habitat quality and quantity over time, providing insights into possible long-term population dynamics and habitat trends.
- **Data visualization**: GIS generates maps, graphs, and charts that are easy to understand even by politicians and decision-makers, enhancing the communication and negotiation among different stakeholders, such as decision-makers and conservation practitioners.
- **Collaboration**: GIS platforms support the collaboration among researchers, practitioners, and policymakers by providing a centralized repository for data, ensuring consistency and transparency in decision-making processes.

Required resources are hardware and software, such as computers capable of running GIS software (e.g., ArcGIS, QGIS, packages developed in R or Python), including peripherals such as GPS devices and high-resolution monitors. Accurate and up-to-date spatial data must be available to be fed into GIS, and trained GIS professionals need to oversee and implement operations. Adequate storage capacity and backup systems to manage and secure large datasets in database form are also required. Prior to the generation of initial assessment and monitoring data, it is imperative that mapping and database solutions are fully operational.

2.4 Work and operational safety

Ensuring safe working conditions during field research is paramount for the well-being of researchers and the success of monitoring activities. When conducting field research, researchers operate in potentially hazardous environments; it is essential to prioritize safety by adhering to legal obligations, regulations, and employ common sense practices, as provided below:

- Remain vigilant and avoid complacency, even after prolonged periods of successful fieldwork.
- Provide and maintain safe working environments for all team members.
- Take responsibility for personal safety and the safety of colleagues.
- Check weather forecasts and river discharge conditions before each sampling trip.
- Assign at least two individuals to each monitoring team.
- Maintain communication with base or emergency contacts throughout fieldwork.
- Equip team members with appropriate nautical gear, life vests, rain gear, and protective clothing.
- Ensure hydration, sun protection, and carry a first aid kit.
- Remain alert and vigilant during all phases of fieldwork.

Before commencing fieldwork, ensure all necessary licenses for scientific monitoring are obtained and readily available. Compliance with legal regulations and permit requirements is essential for conducting research in protected areas or involving endangered species.

2.5 Animal welfare and handling

Maintaining the welfare of sturgeon during handling and data collection is essential for proper ethical work involving live animals. Adhere to the following guidance:

- Ensure all necessary permits for working with live sturgeon are obtained.
- Prioritize safe and respectful handling of sturgeons to minimize stress and potential harm (example Figure 1).
- Implement best practices and adhere to highest standards for sturgeon handling and care.
- Minimize handling time and impact on sturgeons during all stages of research activities.
- Utilize experienced individuals and predefined crew roles to minimize risks during handling.

- Employ non-invasive sampling methods where possible to reduce stress on sturgeons.
- Ensuring proper training and competency for researchers involved in handling sturgeons.
- Release sturgeons safely and immediately following data collection, ensuring gentle release practices.
- Assess the cumulative effects of handling and minimize stress during reproductive seasons to safeguard population sustainability.

For more information on animal welfare and the proper handling of live sturgeon, see "Gessner et al., 2024. Technical Guideline for *ex situ* Conservation Measures in Sturgeon".

Figure 1: *The tagging of large sturgeon utilizing a tube and electronarcosis directly in the river to reduce stress and to ensure safe handling (© Danube Delta National Institute for Research and Development, Romania).*

3 Background information

3.1 Habitat terminology

The "**habitat"** in general is any area offering the conditions and resources that promote the utilization by a life-cycle phase of a given organism (Maddock 1999).

"Habitat use" is how an individual or a population uses a distinct habitat type.

"Habitats" or **"habitat types"** are areas (location and timing), which offer certain conditions and resources to support specific habitat use during distinct developmental stages of sturgeon, such as **spawning**, **hatching**, **feeding**, or **overwintering**. Habitats may belong to different spatial scales of physical habitat (e.g. macro- and mesohabitat) and cover mere patches, stretches, single banks, or entire reaches and sections of a river or catchment (Muhar 1996).

Potential habitats have been identified as bearing the characteristics of habitat types and/ or possessing the conditions and resources to potentially support certain types of habitat use (e.g. potential spawning sites), but their utilization has not yet been verified. **Current** or **actual habitats** have been **confirmed** by sturgeon habitat use.

"Habitat criteria" are descriptive parameters and features which serve to identify habitat types and monitor habitat functionality.

The "**migration route"** connects different habitat types during the sturgeon life-cycle both physically and ecologically. The technical term **"system"**, as used within this document, refers to the functioning and interaction of populations and habitat types within a respective general habitat in its entirety. It is synonymous with the "**ecological corridor for migratory fishes"** (Haidvogl et al., 2021) and comprises the different habitat types, inherent habitat use, "habitat-using"-fish populations, and all processes and exchanges such as information (e.g. behavioral, genetic) and turnovers (e.g. energy, biomass, bedload) necessary for the ecological functioning of the system to support viable populations of native fish and migratory species in particular. **"Fragmentation"** by migration obstacles, on the other hand, can disrupt the connectivity between different habitats, impacting populations by hindering their ability to migrate, access habitat, or find suitable resources.

The main components of the ecological corridor and their ecological connections also allows for the distinction between **habitat- and population monitoring** (Figure 2).

Habitat monitoring (left) characterizes the status of habitat and the physical connectivity within, whereas population monitoring (right) observes the status and viability of the respective population. Both deal with movements, habitat use and its results, and thus the ecological connectivity to a certain extent. For a complete guideline on population monitoring see the Technical Guideline for Sturgeon Population Monitoring (Neuburg, et al. 2024).

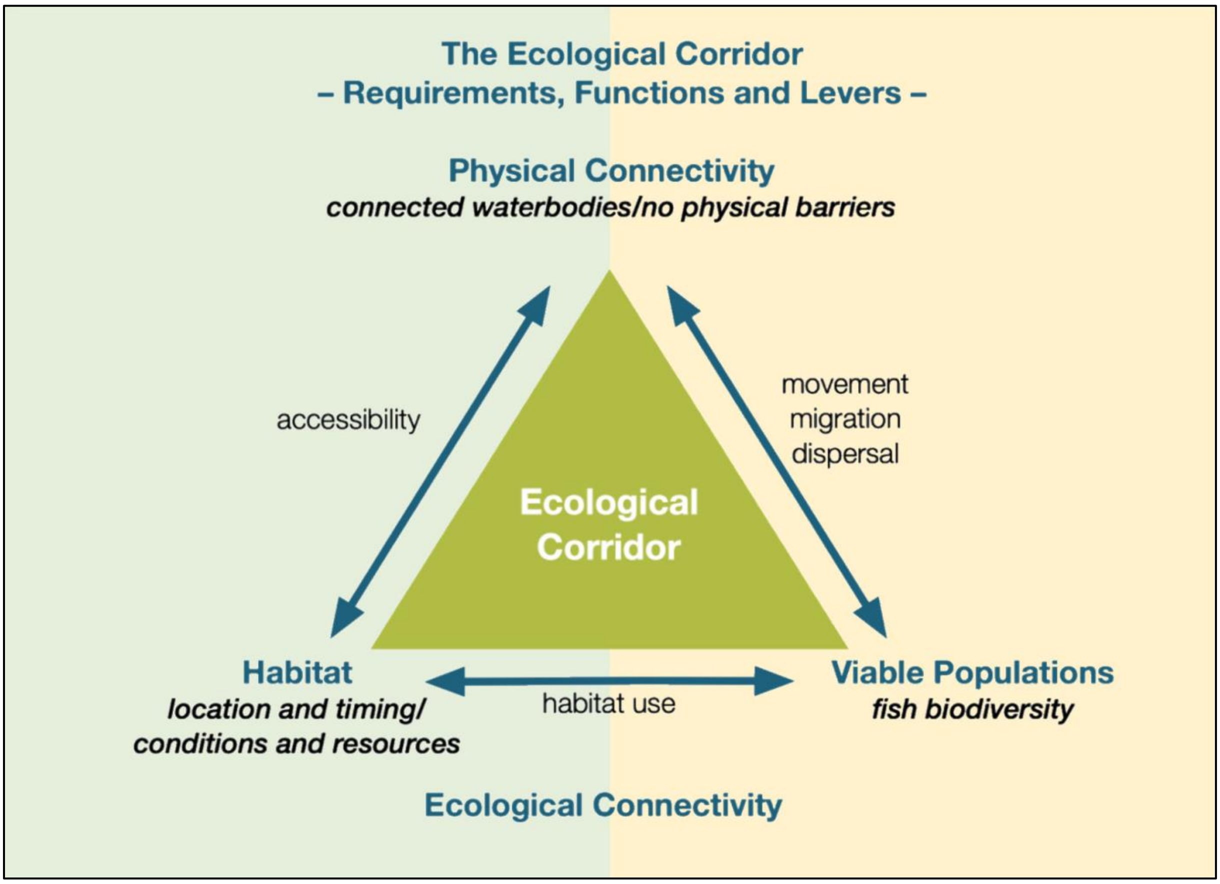

Figure 2: *The Ecological Corridor for migratory fish (from Haidvogl et al., 2021, modified). The left side is the main focus of habitat monitoring, the right population monitoring.*

3.2 Sturgeon traits, habitat requirements and life-cycle

Distinct traits and habitat requirements differ with species, but also between catchments, due to specific habitat conditions in different water bodies. However, there are certain ecological and habitat related traits that European sturgeon species have in common, summarized in Table 1. For more information on the habitat characteristics of European sturgeons and habitats in European rivers, see Popp, S., 2024, "Characteristics and locations of Sturgeon Habitat in European Rivers".

Table 1: *Main habitat-related traits of European sturgeons (after Jungwirth et al., 2003, amended).*

Scientific name	**common name**	**temp. preference**	**habitat**	**migration type**	**ecological guild**	**Reproduction guild**	**spawning site selection**

***Acipenser gueldenstaedtii* (complex)**	Russian sturgeon	meso-eurytherm	freshwater-marine	anadromous	rheophilic A	lithophilic	rheopar
Acipenser naccarii	Adriatic sturgeon		freshwater-marine	anadromous/ potamodromous			
Acipenser nudiventris	Ship sturgeon		freshwater-marine/ freshwater	anadromous/ potamodromous			
Acipenser oxyrinchus	Atlantic sturgeon		freshwater-marine	anadromous			
Acipenser ruthenus	Sterlet		freshwater	potamodromous			
Acipenser stellatus*	Stellate sturgeon		freshwater-marine	anadromous			
Acipenser sturio	European sturgeon		freshwater-marine	anadromous			
Huso huso	Beluga sturgeon		freshwater-marine	anadromous			

**Acipenser stellatus* is adapted to higher temperatures than *A. gueldenstaedtii* or *Huso huso,* especially during early development (Dettlaff et al., 1993, Igumnova 1985).

Explanatory notes on table 1

meso-eurytherm	temperature requirements vary with stage and respective environment / minimum temperatures required for reproduction in spring and summer / higher temperatures are tolerated during summer
rheophilic A	adapted to current and exclusively found in river (not in stagnant water situations of the floodplain)
lithophilic	spawns over hard substrates
rheopar	spawning in current

anadromous	migrates from salt- to freshwater for spawning
potamodromous	migrates in freshwater exclusively

Table 1 shows the similarity of many traits in different sturgeon species. All sturgeons reproduce in freshwater under lotic conditions that may include freshets prior to reproduction (McAdam et al., 2018). Sturgeon are broadcast spawners, requiring sufficiently high current velocities for the distribution of fertilized eggs/embryos over the reproduction site (Table 2; Bruch & Binkowski 2002).

All species of sturgeon can migrate over long distances (Auer 1996) while potamodromous, anadromous, and amphidromous migration patterns have been described (Bemis & Kynard 1997). In the anadromous species, at least four forms with different migration timing and distances were known to exist in the Ponto-Caspian sturgeons (Berg, 1934). An early and a late vernal form migrating and spawning in spring, a summer or early hiemal form spawning in the lower river in late summer or fall, and the late hiemal form migrating during fall, overwintering in the river and migrating further upstream to spawn in the following spring. Smaller runs enter the river throughout the year (Khodorevskaya et al., 2009).

Fertilized eggs/embryos need clean hard substrates with no or only scant periphyton growth (Manny & Kennedy 2002) and low levels of sedimentation of fine substrate for their development. Moreover, sedimentation may smother embryos and yolk sac larvae (Du et al., 2011, McAdam et al., 2005). These hard substrates may consist of pebbles (minimum size Ø of 25 – 30 mm), cobble, boulders, rocks, bedrock (*A. oxyrinchus*) or even hard clay bars (*A. stellatus*) (Gessner & Schütz 2011; Kynard et al., 2013; McAdam et al., 2018; Honţ et al., 2022, NOAA). Interstitial spaces utilized by eggs/ embryos and yolk sac larvae prevent displacement of eggs, protect from predation while their utilization requires sufficient water exchange through the gravel bed to provide sufficient oxygenation (Gessner & Schütz 2011; Du et al., 2011; McAdam 2011) because sturgeon are especially sensitive to low oxygen conditions, respectively hypoxia (Secor & Niklitschek 2002; HELCOM 2019; Delage et al., 2020).

After the yolk sac is resorbed, larvae switch to exogenous feeding and require “productive zones” with high abundances of feeding organisms closely associated with the main river (Gessner & Schütz 2011). At this stage, knowledge of the feed preferences of the species helps in locating suitable feeding sites (Margaritova et al., 2021).

Sturgeon juveniles feed in freshwater and remain above the salt front since tolerance towards salinity is species dependent and develops over time in anadromous species; potamodromous species cannot tolerate salinity. Some species enter brackish water immediately, such as *A. stellatus* in the North Caspian Sea (Khorodevskaya et al., 2009), or after their first year like *A. oxyrinchus* (Allen et al., 2014; Niklitschek & Secor 2009 a and b). Oxygen consumption rates suggest that increasing ionic and osmotic regulation impacts the metabolism of younger fish (Allen et al., 2014).

The table below provides examples of spawning conditions of Eurasian sturgeon species.

Table 2: *Examples of observed spawning conditions for European sturgeon species (after Billard & Lecointre 2000 modified, data mostly from Holcik 1989; Rochard et al., 1991; Chapman et al., 1996; Birstein et al., 1997; Honţ et al., 2022).*

Species	Spawning season	Temperature [°C]	Location of spawning beds	Spawning substrate	Depth [m]	Current velocity [m/s]
Acipenser gueldenstaedtii	May – June	8 – 15	main stem	gravel	4 – 25	1 – 1.5
A. naccarii	February – March	12 – 15	middle and lower reaches of the Po River and its tributaries	along the riverbanks	n.a.	low current
A. nudiventris	April – June	15 – 25	middle reaches	gravel, pebbles	4 – 15	1 – 2
A. oxyrinchus	May – June June – August September	13 – 24	lower reaches	hard bottom, rocks	11 – 13	0.5 – 0.8
A. ruthenus	April – June	12 – 17	riverbed and inundated areas during spring floods	gravel, pebbles	2 – 15	1.5
A. stellatus	May – September	12 – 24 (Volga) 15.0 – 29 (Kura)	lower reaches, along the banks	gravel/clay bars	2 – 14	0.7 – 1.8 bottom | 1.1 – 1.9 surface
A. sturio	March – August	7.7 – 22	middle reaches	gravel, stones	> 5	1.5 – 2

Huso huso	March – April	6 – 21 | 9 – 14 optimum	upper reaches / higher water	stones, pebbles, gravel	4 – 15	1.5 – 2

All species have a narrow window of optimal temperature for the successful development of early life-cycle stages. Temperatures exceeding this thermal range lead to increased deformations and mortalities, whereas reduced temperatures prolong development and enhance the risk for increased mortalities by fungal infection and predation (Table 3).

Table 3: *Optimal species-specific temperature range during embryogenesis (after Chebanov & Galich 2013, Dean 1895, Delage et al., 2020, Gessner & Schütz 2011, Jatteau 1998, Mohler 2004, fishbase.org).*

Species	**Optimal temperature range during embryogenesis [° C]**
Acipenser gueldenstaedtii	16 – 20
A. nudiventris	14 – 18
A. oxyrinchus	18 – 20
A. naccarii	14 – 18
A. ruthenus	13 – 16
A. stellatus	17 – 24
A. sturio	17 – 20
Huso huso	9 – 14

Yolk sac larvae hatch after few days (depending on temperature) and move to hiding places until the yolk is resorbed (Table 4).

Table 4: *Duration (h) of incubation until hatching at different temperatures (modified from Chebanov & Galich 2013; HELCOM 2019).*

	Duration of egg incubation [h]				
Temperature [° C]	***Huso huso***	***A. nudiventris***	***A. gueldenstaedtii***	***A. stellatus***	***A. oxyrinchus***
10 – 11	240 – 235				

11 – 12	230 – 220	190 – 180			
12 – 13	210 – 200	170 – 168			
13 – 14	190 – 180	155 – 145			
14 – 15	170 – 160	135 – 125			
15 – 16		115 – 105			
16 – 17		105 – 100			
17 – 18		95 – 90	150 – 145		
18 – 19			140 – 130		
19 – 20			120 – 115		105 – 95
20 – 21			110 – 95	100 – 90	95 – 85
21 – 22			90 – 85	80 – 70	
22 – 23			80 – 75	70 – 60	85 – 75
23 – 24				60 – 50	

Larvae emerge from their hiding places immediately prior to the resorption of the yolk, disperse to feeding areas, and start first feeding (planktonic: e.g. rotatoria, daphnia / benthic: e.g. chironomidae, tubificidae (Chiasson et al., 1997; Jatteau 1998; Muir et al., 2000; Gessner et al., 2007; Zarri & Palkovacs 2019; Holley et al., 2022) (Table 5).

Table 5: *Duration of sturgeon prelarval development in days before transition to first exogenous feeding in relation to water temperature (modified from Chebanov & Galich 2013; HELCOM 2019).*

Water temperature [° C]	**Duration, d**			
	A. gueldenstaedtii	***A. stellatus***	***A. oxyrinchus***	***Huso huso***
12	20	-		18
13	18	-		16
15	12	-		12
17	9.5	12		10
19 (20)	8	9	11	8

21	7.5	8		7
23	-	6.5	8	-

3.3 Sturgeon life-cycle stages and habitat types

A generalization of the (anadromous) sturgeon life-cycle, its main stages, and threats are shown in the figure below.

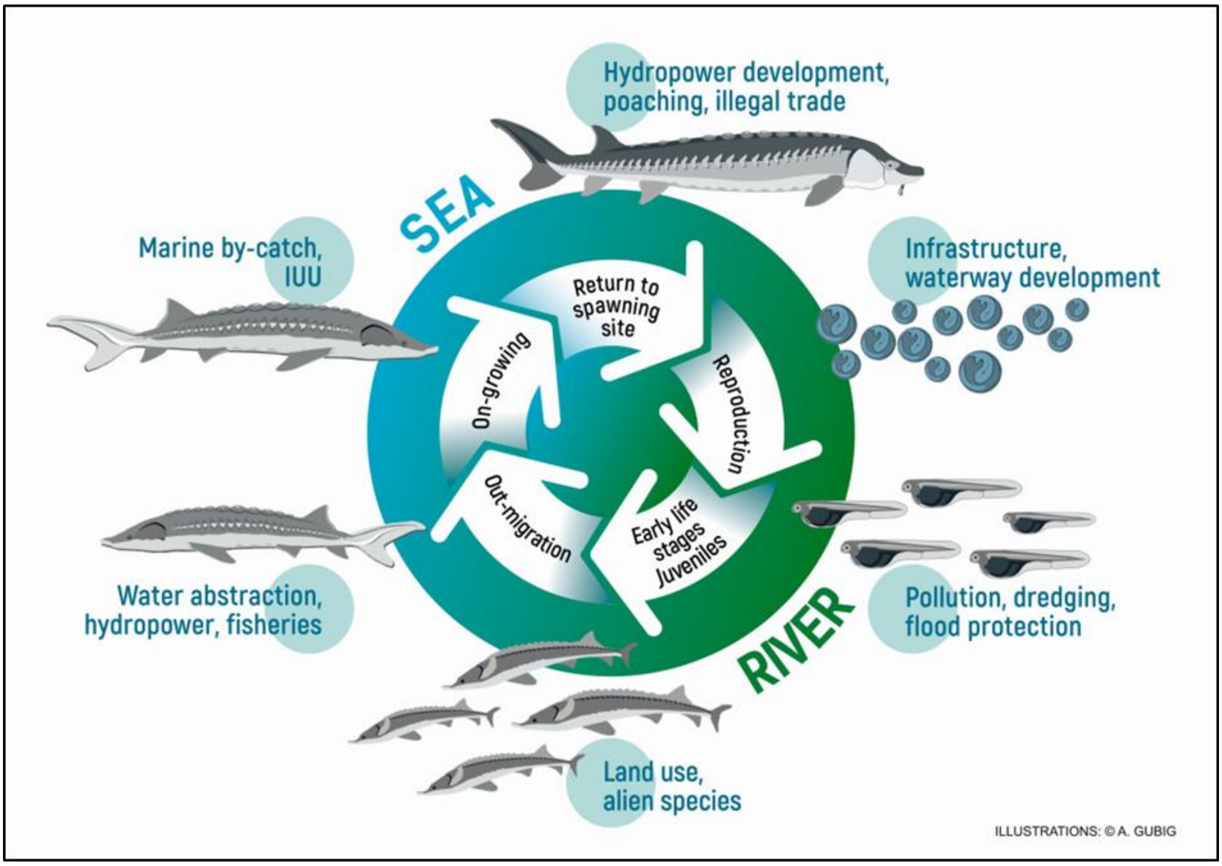

Figure 3: *The sturgeon life-cycle and main threats.*

There are still deficits in knowledge on distinct life-cycle stages and requirements for most species, catchments, and no official standards for their categorization across all sturgeon species and associated habitat types that exist. Different terms and wordings are used in scientific literature.

Table 6 provides an example of the sturgeon life-cycle and associated habitat conditions using the Atlantic sturgeon (*A. oxyrinchus*) as a reference. A common categorization for the description of life-cycle stages is applicable also to other sturgeon species.

Table 6: *Life-cycle stages of the Atlantic sturgeon (A. oxyrinchus) in the Greater Atlantic Region (modified from NOAA).*

Phase	Size (mm)	Duration	Description	Habitat utilization
Eggs	~2 - 3 mm	Hatching occurs ~4 - 6 days after egg deposition and fertilization	Fertilized or unfertilized	Eggs are deposited in fresh water (0.0 - 0.5 ppt) over hard bottom substrate (e.g., cobble) and become adhesive shortly after fertilization.
Free embryos (Yolk Sac Larvae (YSL))	~6 - 14 mm	8 - 12 days post hatch	Tigmotactic behavior (Gessner et al. 2009, Bates et al., 2014), nourished by yolk sac.	The YSL phase lasts approximately 8 - 12 days, during which larvae are absorbing the yolk sac and are completing embryonic development. Upon hatch the free embryos emerge from the gravel (e.g., exhibit a "swim-up and drift-down" behavior), seek refuge in interstitial spaces of hard bottom substrate (e.g., rock, cobble, gravel, limestone, boulder, etc.).
Feeding Larvae (Post Yolk Sac PYL)	~14 - 37 mm	12 - 40 days post hatch	Free swimming: feeding: Silt/sand bottom, deep channel; freshwater	During (days 13 - 40 post hatch) the larvae disperse downstream of the spawning/rearing area. Sturgeon larvae are intolerant of saline waters; thus, their habitat must be upstream of the salt front, in waters that have a salinity of 0.0 - 0.5 ppt. PYL occur in the water column but feed at the bottom as they move downstream and forage for aquatic insects, insect larvae, and other invertebrates.
Young of Year (YOY)	0.3 grams < 410 mm total length	From 40 days to 1 year	Fish that are >3 months and <1 year old; capable of capturing and consuming live food	YOY are fish between age 0 and the summer of the following year. Prey items may include aquatic insects, insect larvae, and other invertebrates.
Juveniles	>410 mm and <760 mm total length	1 year to time at which first coastal migration is made From first coastal migration to sexual maturity	Fish that are at least 1-year-old, are not sexually mature, and do not make coastal migrations	After their first year (YOY), juvenile Atlantic sturgeon become increasingly tolerant to saline water and may use the full extent of the river to opportunistically forage, particularly in areas with soft substrate (e.g., sand, mud). Migrating and foraging late juvenile Atlantic sturgeon may enter the lower estuary as early as mid-March and remain as late as mid-November. Juveniles close to maturation and adult Atlantic sturgeon exhibit seasonal coastal movements

	>760 mm and <1,500 mm total length		Fish that are not sexually mature, but make coastal migrations	in the spring and fall. They typically remain within the 50-meter depth contour but are not limited to that depth. Prey items may include benthic prey such as molluscs, gastropods, amphipods, annelids, decapods, isopods, and fish such as sand laces.
Adults-Migrating & Foraging	>1,500 mm total length	Post-maturation	Fish that are sexually mature	Juveniles close to maturation and adult Atlantic sturgeon may aggregate in estuarine (seaward of river mouth), bay, sound, and ocean areas over the winter months. They typically remain within a 50-meter depth contour but are not limited to that depth. Prey items may include molluscs, gastropods, amphipods, annelids, decapods, isopods, and fish such as sand laces.
Adult-Spawning	>1,500 ♂ - 1,800♀ mm total length		Fish with fully developed gonads entering rivers for reproduction	Adult Atlantic sturgeon migrate into rivers in the spring or early summer or in early fall for spawning and return to coastal waters after reproduction. For spawning, adults use habitat in fresh water with hard bottom substrate (e.g., rock, cobble, gravel, limestone bedrock, etc.), and water temperatures between 13 and 26° C. Sturgeon require well-oxygenated, flowing water, absence of physical barriers for passage (e.g., locks, dams, reservoirs, gear, thermal plumes, turbidity, sound, etc.) between the river mouth and spawning sites. Adults may stage around the spawning time downstream of the spawning area.

It is important to note that not all observed conditions at sites of sturgeon presence inherently define habitat; therefore, careful consideration is essential when selecting the criteria for habitat identification and monitoring purposes.

Habitat suitability criteria for the European sturgeon (*A. sturio*) as an example, are detailed in Table 7.

Table 7: *Habitat suitability criteria for sturgeon with A. sturio as reference (modified and translated from Gessner & Schütz 2011).*

Life-cycle stage / Criteria	**Fertilized eggs/embryos**	**Yolk sac larvae**	**Feeding larvae**	**YOY**	**Juveniles > 30 cm**	**Adults (spawning)**
Current velocity (min, max, spatial variability)	0.6 - 2.2 m/s, water exchange in interstitial maintains >70% O2 saturation	0.6 - 2.2 m/s, water exchange in interstitial maintains >70% O2 saturation	low-flow, high productivity areas	low-flow areas 0 - 0.8 m/s	low-flow areas 0 - 1.4 m/s	High variability of current velocities with resting zones (meandering structures, pools), spawning: see fertilized eggs
Bottom substrate (particle size, quality, extension)	cobble/gravel > 25 mm, clean, scant periphyton growth suboptimal: cobble 3 – 30 cm	cobble/gravel > 25 mm, clean, scant periphyton growth	sand to gravel	sand and soft substrates (silt, clay) with high abundance of benthic food organisms (Tubificidae, Chironomidae)	sand and soft substrates (silt, clay, bank structures) with high abundance of benthic food organisms (Tubificidae, Chironomidae)	cobble/gravel > 25 mm, clean, scant biofouling, no sedimentation
Navigation	not above spawning sites	not above spawning sites	moderate suction and wave action	sufficient distance between ship hull and riverbed, moderate suction (< V crit.) and wave action	sufficient distance between ship hull and riverbed	not above spawning sites, sufficient distance between ship hull and riverbed
Water temperature (min, max)	17 - 20 °C	15 - 22 °C	15 - 22 °C	1 - 26 °C	1 - 26 °C	17 - 22 "C (June - July)
Suspended particles	low	low	up to 10 g/l	up to 10 g/l	up to 10 g/l	low (spawning)
Oxygen	> 6 ppm in gravel bed	> 6 ppm in gravel bed	> 6 ppm	> 5.5 ppm	> 5.5 ppm	> 6 ppm in gravel bed
pH	6.5 - 8	6.5 - 8	6.5 - 8	6.5 - 8	6.5 - 8	6.5 - 8
Conductivity (salinity, ions)	< 800 µS/cm-	< 800 µS/cm	< 1000 µS/cm	< 1200 µS/cm		< 800 µS/cm (spawning)
Organic load (BOD)	low	medium	high	high	high	low (spawning)

3.4 Habitat associated threats

The combination of threats to sturgeon habitats may be specific in a given river system. Yet, there are common threats that adversely affect the different habitats for sturgeons (Table 8).

Table 8: *Common threats for sturgeon habitat, habitat types and sturgeons in their habitat (modified and amended from Friedrich et al., 2019).*

Impact/Threat	Physical Effect	Ecological Effect
Dams	**Migration barriers**	Upstream migration barrier for spawning migration
		Downstream migration barrier for spent adults
		Downstream migration barrier for juveniles
	Alteration of habitat downstream (scouring, erosion, lack of sediment transport, alteration of discharge dynamics, changed temperatures due to deepwater release or increased irradiation)	Loss of spawning grounds in vicinity of dams, alteration of substrate availability, changes in hydrology, alteration of temperature regime, all leading to reduced reproductive efficiency, altered timing of maturation and reduced productivity
	Change of habitat upstream - impoundment	Loss of orientation, Interruption of upstream migration of adults
		Delay of downstream migration of juveniles & increased risk of predation & risk of suffocation (O_2 deficit in stratified reservoir)
		Loss of spawning & nursery habitat
		changed fluvial dynamics
		sedimentation processes
		changed thermal regime and potential stratification

		Reduced productivity of the river
Hydropower operation	**Migration barriers & change of habitat**	See "Dams" above
	Sediment flushing	Loss of spawning and nursing habitat
		Increased mortality of juveniles
		Reduced productivity
	Hydropeaking	Loss of nursing habitat
		Increased mortality of juveniles
		Reduced productivity
		Dewatering of spawning areas
		reduced flows during non-peaking hours, affecting oxygenation processes for developing embryos and larvae in interstitial spaces
	Turbine passage	Increased mortality of spent adults and juveniles migrating downstream
Changes in Hydromorphology	**Straightening of river, loss of sidearms and backwaters**	Loss of habitat heterogeneity
		increased flow in main channel, increased sediment transport, loss of habitat
		Reduced productivity
	Deepening of riverbed	Loss of habitat, altered sediment composition, increased flow
Navigation	**ship traffic**	Vessel strikes, displacement through wave action
	Deepening of riverbed	See ""Changes in Hydromorphology" above Direct mortalities due to encounter of fish with dredges

	Migration barriers	See "Dams" above
Pollution	**heavy metals and hydrocarbons**	Exposition causes direct mortality
		Accumulation causes reduced fertility
	Increase in nutrients and BOD	Increased mortality of eggs and larvae due to increased bacterial and fungal pressure
		Reduced reproductive success due to oxygen depletion
Gravel extraction	**Other adverse physico-chemical impacts**	Loss of spawning and nursery habitat
		Risk of increased fine sedimentation

4 Habitat Monitoring Program

4.1 Definition, recommendation, and rationale

Gruijter et al., (2006) defines **monitoring** as "collecting information on an object through repeated or continuous observation in order to determine possible changes in the object". A comprehensive overview of the monitoring of species and habitats is provided by Schmidt & Van der Sluis (2021).

Habitat monitoring in the context of this document is the recurring assessment of the functionality of sturgeon habitats. However, basic prerequisites for doing so are also assessments of information, data, conditions, timings, and habitat utilization for the identification, verification, and confirmation of habitats within a respective system. Therefore, a monitoring approach in form of an internally and externally coordinated and synchronized program (with other fields of sturgeon conservation and restoration), including all necessary assessments and recurring activities, is strongly recommended.

Such an approach comprises steps for the **identification** and **verification** of habitats for different sturgeon life-cycle stages, the **confirmation** of such assessments by the documentation of habitat use, while **recurring and real-time monitoring** measures are carried out to document the functionality of these habitats over time.

This four-step approach for habitat assessment and monitoring includes:

1. **Identification of areas of general relevance through data and information research**
    - Collecting relevant information and data on historic and current presence of species, populations, habitats, anthropogenic impacts, and their interaction.
    - Identification of rivers and areas of general relevance and preliminary mapping by identifying current and former sturgeon catchments and rivers as well as sections and stretches potentially being sturgeon habitat by e.g. historic documents, literature, documented presence, catch data.
    - Documenting areas and stretches potentially bearing the physico-chemical and hydromorphological characteristics of sturgeon habitats by e.g. documentation of water quality by data, remote sensing, analysis of aerial photography and maps, stream surveys.
    - The inclusion of data and information in GIS and databases to create layered maps for further analysis.

2. **Verification of identified habitats**
    - Verification of prior mapping efforts by measuring and documenting key habitat criteria in the field like e.g. temperatures, water depths, flow velocities, dissolved oxygen, and substrate compositions **under the actual conditions of habitat use** (e.g. season, hydrology, hydropower operation), statistical modeling to support findings in the field.
    - Including the results of field assessments in databases and for instance as a separate layer in GIS for the visualization of the outcome of the habitat assessment.
    - Development, verification, and adaptation of specific working protocols to assess the functionality of the respective habitats over time.

3. **Confirmation of sturgeon habitat utilization**
    - The documentation of actual habitat use in form of species presence as a first step by telemetry/tracking approaches of sturgeon individuals and groups and genetics (eDNA).
    - In combination with the assessment of habitat functionality by population monitoring, documenting the actual results of habitat use.
    - The inclusion of data on actual habitat use in databases and GIS.

4. **Recurring and real-time monitoring measures**
    - Monitoring of the functionality of sturgeon habitat over time.
    - Assessment of impacts and threats for sturgeon habitat.
    - Established under a dedicated, mid- to long-term **program**.

It is important to note, that the subsequent assignment of methods to the steps provided above are merely recommendations, as some of the methods may serve multiple purposes/ steps and the sequence of their application depends on the specific availability of data, the respective objectives and conditions of the monitoring program, and characteristics of the habitat being monitored.

4.2 Identification of areas of general relevance

In order to identify sturgeon habitat in catchments formerly or presently utilized by sturgeon the knowledge of the extent of sturgeon range within these catchments (Kinzelbach 1994) and the connectivity of the habitat are paramount. Also, identifying areas and sections containing potential life-cycle habitats are a prerequisite for their verification (Limnoplan 2017). Location, timing, and frequency of occurrence of potential habitats, as well as their spatial extension must be assessed and apply the criteria provided. The results are documented and mapped preliminary using GIS.

4.2.1 Habitat criteria for the identification of relevant rivers and sections

- Historic presence of native/indigenous sturgeon (e.g. by anecdotal catches, sightings documented in old documents, fisheries journals, photos etc.).
- Current presence of native sturgeon (e.g. by catches, sightings, and documentation/tracking of individuals, groups and aggregations of distinct life-cycle stages, eDNA gained e.g. through population monitoring, see also "Confirmation of sturgeon habitat utilization").
- Water quality meeting the criteria for relevant sturgeon life-cycle stages in particular (e.g. high levels of dissolved oxygen, low levels of BOD, appropriate temperature regime, see Table 12).
- Hydromorphological features indicating potential habitat types (e.g. meandering river structure comprising riverine dynamics in form of changing current patterns, deposition and translocation of varying substrates and materials resulting in sequences of cutbanks and point bars, alternating river sections with fast flowing deeper stretches and potentially hard substrates, gravel bars, soft-bottomed low-flow zones in or adjacent to the main channel, zones without sedimentation, deep pools for overwintering).
- Flow regime supporting sturgeon spawning and early development, with freshets prior to spawning season and early development.
- Temperature regime supporting all life-cycle stages of sturgeon (narrow requirements and only minor changes during early life-cycle stages).
- Zones with high abundance of feed organisms for different life-cycle stages.
- Accessibility of potential habitats (absence/presence and distribution of migration barriers and migration aids).

- Presence and reproduction of species with similar habitat requirements like sturgeon.

4.2.2 Methods

4.2.2.1 Data mining

The initiation of a sturgeon habitat monitoring program necessitates a systematic approach to retrieve and compile data on **sturgeon biology, ecology, and habitat**. Areas of focus comprise both current and historical data, covering species **presence, population dynamics, distribution ranges, and life-cycle**, with a special emphasis on **seasonal habitat use**. Additionally, **official data** from open water monitoring sources such as **water quality assessments** and **gauging stations** should be included.

Relevant information resides in various outlets, including universities, libraries, research organizations, museums, national/international administrations, NGOs, companies, and fishery associations. This information may exist in printed or digital formats, ranging from databases and maps to GIS applications or as anecdotal evidence only.

Stakeholders, such as current and former commercial fishermen, recreational fishermen, and fish traders, can provide valuable information since anecdotal information from local knowledge can complement scientific data, offering a more comprehensive view of sturgeon habitats. Stakeholder and expert interviews may help to address gaps in written and online information. They offer valuable insight, particularly with fishermen, with the caveat of potential intentional misinformation (Blaž et al., 2021).

Historical information sources such as historical digitized maps (e.g. from the 18th and 19th century in the MAPIRE (MAPs of the empIRE) project) may provide insight into past ecosystem conditions of natural or nature-like water bodies, which serve as reference points for sturgeon habitats. Historical reference conditions provide a temporal dimension, allowing for insights into long-term changes in sturgeon habitats and supporting adaptive management strategies. Caution is advised when utilizing scientific descriptions from past centuries, given potential discrepancies with contemporary standards. Historical analyses should therefore methodically identify and date impacts on the targeted system.

Habitat information can be supplemented by exploring historical **commercial fishery data**, though one must be aware of their limitations in accurately reflecting ecological abundance and species composition (Haidvogl et al., 2003). Catch data from angling and other recreational types of sturgeon fishing should also be considered in countries, where these activities are still allowed.

Utilizing various approaches, including citizen science, historical reference conditions, and commercial and recreational fishery data, can significantly **broaden the scope of data**, offering a more holistic view of sturgeon habitats. Incorporating local knowledge through stakeholder interviews and citizen science **enriches the dataset with insights** that may not be captured through scientific assessments alone.

Approaches like citizen science and stakeholder interviews **foster community involvement, raising awareness and promoting a sense of shared responsibility for sturgeon conservation**.

Required resources and effort are difficult to assess and largely depend upon the availability of documentation and the dimension of the catchment in question as well as the methods selected for the assessment. Citizen science and stakeholder engagement require resources for training, coordination, and support, potentially imposing constraints; while bridging the gap between scientific terminology and local knowledge or historical records demands effective communication, potentially hindered by language or interpretive differences. Citizen science and historical data may offer cost-effective options, while advanced technologies or extensive fieldwork may incur higher costs. Certain methods, like data mining historical records, may save time, while others, such as citizen science, may require careful planning and coordination as well as substantial input of personnel. Overall, the experience/expertise required for this initial step is not limiting for the overall approach.

4.2.2.2 Analysis of bathymetric maps, aerial photography and orthophotography

The analysis of bathymetric maps, aerial photography, and orthophotography also represents an approach for identifying areas of potential relevance for sturgeon habitat monitoring within catchments. These methods include the examination of underwater topography, aerial imagery, and corrected aerial photographs to discern and assign key features related to sturgeon habitats (Blaž et al., 2021).

Bathymetric maps provide information on underwater topography, offering depth contours, and thus potential sturgeon habitat features (e.g. deep pools as resting, staging, or wintering areas).

Aerial photography captures high-resolution images of the Earth's surface, while **orthophotography** corrects distortions, allowing for accurate measurements and thus potential quantification of habitats. The analysis of these images involves identifying surface features, such as riverbank structures, water flow patterns, and potential sturgeon habitat indicators such as submerged structures, gravel bars, rapids, and migration barriers.

Pros and Cons

- Allows for a detailed visual assessment of both underwater (partially) and surface features.
- Can cover large areas remotely, providing a broad overview of potential habitats.
- Enables historical comparisons to identify changes in habitat features over time.
- Does not provide detailed information on underwater structures or substrate composition compared to direct underwater surveys.

- Weather affects data quality, particularly in areas prone to cloud cover or adverse weather conditions.
- Resolution limitations may hinder the identification of smaller-scale habitat features.
- Requires expert knowledge for the interpretation of visible features with regard to sturgeon habitats.

Required resources and effort

- **Cost**: Medium (Moderate costs associated with acquiring high-resolution aerial imagery and orthophotography, with potential variations based on area coverage and image resolution.)
- **Time**: Medium (Efficient coverage of large areas, but processing time depends on image resolution and the extent of analysis required.)
- **Equipment**: Low (Basic equipment for accessing and analysing aerial imagery and orthophotography, such as GIS software and computing resources.)
- **Experience/Expertise**: Medium (While basic skills may be sufficient for a general image analysis and interpretation, advanced expertise is needed for precise identification of sturgeon habitat indicators.)

4.2.2.3 Remote sensing

Remote sensing is a collective term that stands for the assessment of habitat features from afar, using a variety of platforms, technologies, and sensors. It is used to identify terrestrial and aquatic habitats, but also for the documentation of habitat dynamics and changes. More information on remote sensing in the monitoring of species and habitats can be found in Schmidt & Van der Sluis (2021).

In freshwater habitats it is used mainly for hydromorphological and some physico-chemical features. Tables 9 and 10 provide an overview of the different platforms, technologies, sensors, and data sets on potential habitat conditions. Remote sensing has been used to identify freshwater fish habitats (Kuiper et al., 2023). Current literature research for the document at hand did not reveal any specific use for the identification of sturgeon habitats. This may be due to the fact of limitations for application in large and deep-water bodies. It is included in this document since the potential beneficial use for the identification of certain sturgeon habitat features cannot be ruled out. Required resources and effort are given in the tables below.

Table 9: *Overview of remote sensing technologies, strengths and weaknesses and applications for freshwater habitat characterization (from Kuiper et al., 2023, modified and amended).*

Technology	**Strength**	**Weakness**	**Example freshwater habitat application**
Platform			
Ground based	Very high spatial resolution (cm), easy to pair with field data	Very small geographic coverage	Single reach
UAV	High spatial resolution (cm - m), decreased cost in recent years	Small geographic coverage	Multiple reach, single stream
Aerial	High spatial resolution, moderate geographic coverage	Costly, generally a one-off collection	Single watershed / multiple watersheds
Satellite	Moderate to high spatial resolution, large geographic coverage, repeated data acquisition on a regular cycle	Spatial resolution may not be sufficient depending on application	Multiple watersheds / continental
Sensor			
Optical imagery	Most common sensor type, broad range of available information, many open access data, archives, and long-term calibration enable time series applications,	Difficult to get vegetation structural information, passive sensor relies on suitable illumination conditions, obscured by clouds and haze, turbidity can limit performance, most effective in shallow water only	Habitat type and complexity, landcover, spawning (Salmonids), can differentiate between different substrate types based on color and reflectance properties (unique spectral signatures)
Thermal	Best for temperature information	Only useful for temperature information	Stream and lake surface temperature

Lidar	High spatial resolution 3D information on terrain and vegetation structure, active sensor, can penetrate water to some extent, UAV lidar provides high-resolution bathymetry.	Lack of spectral information, costly, Limited penetration in turbid water, satellite data may lack the required resolution,	Habitat type and complexity, hydrological features, spawning (Salmonids) water depth: Lidar, bathymetric lidar, can provide detailed bathymetric data, allowing for the mapping of river bottom morphology, can distinguish between rougher surfaces (e.g., boulders) and smoother surfaces (e.g. sandy bottoms)
Radar	Active sensor, able to penetrate cloud, can provide some information on vegetation structure	Limited data availability for longer wavelength radar, difficult to process	Ice. cover, hydrological features water depth: depth-sounding radar
Digital Photogrammetry	High spatial resolution information on forest structure in floodplain that is similar to that provided by Lidar (but not the same), can have some limited spectral information	Limited geographic coverage, most often UAV based, lack of penetration through vegetation and cloud	Habitat type and complexity, hydrological features, spawning (Salmonids), delivers digital surface models, and three-dimensional models of the terrain

Table 10: *Required effort for remote sensing technologies (from Kuiper et al., 2023, modified and amended).*

Optical imagery	**Cost:** Medium **Time:** Low to Medium (depends on satellite revisit frequency and data availability) **Equipment:** Low (access to satellite or aerial imagery providers) **Experience and Expertise:** Low to Medium (basic image interpretation skills, familiarity with remote sensing principles)
Thermal	**Cost:** High (acquisition and processing costs are typically higher than optical imagery) **Time:** Medium (availability of thermal data may vary, and processing can take time) **Equipment:** Medium to High (specialized thermal sensors and processing infrastructure) **Experience and Expertise:** Medium to High (knowledge of thermal remote sensing principles, data processing, and interpretation)
Lidar	**Cost:** high (satellite) to medium (aerial, UAV) **Time:** low to medium (depending on scale) **Equipment:** medium to high (depending on platform) **Experience and expertise:** medium to high (depending on technology)
Radar	**Cost:** High (acquisition and processing costs can be relatively high) **Time:** Medium to High (availability of radar data may be less frequent than optical data) **Equipment:** Medium to High (specialized radar sensors and processing infrastructure) **Experience and Expertise:** Medium to High (advanced knowledge of radar technology, data processing, and interpretation)
Digital Photogrammetry	**Cost:** Medium to High (depends on the scale and complexity of the project) **Time:** Medium to High (processing time can be significant, especially for large datasets) **Equipment:** Medium to High (requires specialized photogrammetric software and computing resources) **Experience and Expertise:** Medium to High (advanced knowledge of photogrammetric principles, software usage, and data processing)

4.2.2.4 Back-calculation of spawning time and location

Back-calculation has been used in sturgeon habitat monitoring to either determine the timing of spawning, by exactly identifying the developmental phase of documented embryos and larvae and taking water temperature and required duration into account, or to estimate the rough location of sites for sturgeon spawning and early development, by considering the timing of known spawning events and the average downstream movement speeds of sturgeon juveniles within the system (Duncan et al., 2004; Wei et al., 2009; Chiotti et al., 2008; Chapman & Jones 2010; Seesholtz et al., 2015). However, the documentation/catch of sturgeon early life-cycle stages in large rivers without knowledge on the location of reproduction areas is difficult. Rough locations of sites for spawning and early development have been assessed for juveniles by simply calculating distances from known timing of spawning and average downstream moving speed of juveniles from other such spawning habitats (e.g. Margaritova 2022; Mihov et al., 2022).

Pros and Cons

- Provides a method to predict potential spawning areas by analysing juvenile catches.
- Offers a temporally precise method by considering the timing of spawning events.
- May be cost-effective compared to extensive field surveys, especially when historical juvenile catch data is available.
- Relies on accurate and comprehensive data on sturgeon juvenile catches, spawning timings, and downstream movement speeds.
- Relies on specific knowledge of sturgeon ecology and behavior.
- Ignores environmental factors influencing juvenile movement, which might result in imprecise predictions.

Required resources

- Timing of Spawning: Accurate information on the timing of sturgeon spawning events within the system.
- Juvenile Movement Speeds: Average downstream movement speeds of sturgeon juveniles in the given watercourse.
- Juvenile Catch Locations: Data on catches of sturgeon juveniles at various points along the watercourse in relation to their respective locations of spawning.

4.2.2.5 Assessment of migration barriers

Barriers block the migration of sturgeon between their life-cycle habitats. Thus, barriers and migration aids/fish passes need to be assessed and function-controlled, respectively (Kemp et al., 2008; Noonan et al., 2012; Silva et al., 2017; Katopodis et al., 2019; Matica 2020). This information needs to be viewed in relation to the results of the identification of potential and current habitats to inform sturgeon conservation management on how to develop and implement further measures for restoring sturgeon migration routes and to identify priorities for habitat assessment and monitoring (Schmutz & Mielach 2013; PANEUAP 2018; Johnston et al., 2019; Bruch & Haxton 2023; Popp, S., 2024).

Figure 4: *Sindi dam on the Pärnu River in Estonia. Migration barrier for sturgeon and other fish. Note the fish pass on right side of photo (© Jarko Jaadla).*

Figure 5: *Pärnu River in Estonia after removal of the Sindi dam. River continuity for migratory fish has been restored (© Keskkonnaamet).*

Assessment of barriers is achieved by both data mining in databases and other information sources (ICPDR 2021; Amber 2020; Amber Barrier Atlas https://amber.international/), as well as by the assessment of migration barriers in the field. Numerous passability assessment protocols have been formulated for *in situ* barrier assessments. For example, within Europe, three key protocols exist that are well developed and widely accessible (SNIFFER (UK), ICE (France) and ICF (Spain)). These protocols were tested and reviewed by Kerr et al. (2016). The French ICE protocol (Baudoin et al., 2014) was found to be the least subjective and produces passability scores for the most species, whilst requiring fewer physical measurements to be recorded than other fine-scale protocols.

Thus, the ICE protocol has been chosen as the protocol of choice for use by the AMBER consortium (a project seeking to apply adaptive management to the operation of barriers in European rivers to achieve more effective and efficient restoration of stream connectivity) and for wider promotion as a European standard for barrier assessment.

One limitation of the ICE protocol is that passability scores are produced through a time consuming decision tree process. The Barrier Passability and Hydropower Potential Assessment Software Tool (AMBER Barrier track) automates this process, calculating the passability scores based on a few input parameters. In addition to generating ICE passability scores, the tool estimates the hydropower potential at the site through the assessment of discharge and head drop. As such, the tool produces data that can be used to populate barrier mitigation prioritisation tools and help with catchment level management decisions.

Table 11: *Attributes of the tier one "Record a New Obstacle" page(s) of the AMBER Barrier Tracker app (amended).*

Feature	Record entry	Automatic or manual	Answer types
Barrier Photo	Obligatory	Automatic opening, manual shutter control and option to retake image: camera opens upon opening “record obstacle” page	User defined (photo)
Date of record	Obligatory	Automatically, upon taking photo	Date in format Hours/minutes/seconds & Day/month/year
Barrier Type	Optional	Manual	Weir, dam, culvert, ford, sluice, ramp (with

			images to aid in making the choice)
Barrier Height	Optional	Manual	Height categories: <0.5 meter, 0.5 – 1.0 meter, 1.0 – 2.0 meter, 2.0 – 5.0 meter, 5.0 – 10.0 meter, >10.0 meter*
Does the barrier extend across the entire watercourse?	Optional	Manual	Yes/no
Is the barrier in working condition?	Optional	Manual	yes/no/don't know
Please add any additional notes	Optional	Manual	
Barrier Location (Geo-location of obstacle) Obligatory	Obligatory	Automatic, upon taking photo. Prompt for GPS (locate) to be used if not switched on. App records whether location was taken based on GPS, phone signal or both.	Lat./long. coordinates via GPS chipset on phone and where there is a suitable signal, the phone signal.
Please add any additional notes	Optional	Manual	Text

4.2.2.6 Presence of reproductive populations of species with similar ecological requirements

The presence of viable populations of other fish species than sturgeon, yet with similar habitat requirements with regard to reproduction, nursery and/or migration can hint at the presence of favourable conditions also for sturgeon (Marenkov & Fedonenko 2016). No protocols are available and such indicator fish species would be specific for a certain area and catchment.

Pros and cons

- May help in identifying potential sturgeon habitat types without sturgeon presence and identify systems of greatest recovery potential.
- List of indicator species may not be transferable between areas and catchments.

Required resources

- Information and data on the ecology, autecology, status, and habitat types of fish species in the respective catchment.

4.2.2.7 Stream surveys

A preliminary approach to identify river sections with habitat characteristics involves conducting a (boat) survey of the watercourse. This aims at the documentation of indicators of relevant habitat criteria, such as hydromorphological features or other macroscopically visible substrates and structures, rapid flow sections alternating with low-flow zones in direct connection to the main river, and the presence of migration barriers. Additionally, supplementary on-site interviews with stakeholders and other local experts (e.g. professional and recreational fishermen) can provide further insights into substrates, water depths and anecdotal information on ecological conditions. Such a survey can already provide sampling sites for habitat verification.

Pros and cons

- Provides direct, firsthand observation of the river and its features, allowing for real-time assessment.
- Offers a relatively quick method for collecting initial data on habitat features.
- Can cover a wide range of river sections, including remote or challenging areas that may be inaccessible by other means.
- Allows for visual confirmation of habitat characteristics, such as substrate types and flow patterns.
- Offers an opportunity to engage with local stakeholders on-site, gaining valuable insights from their knowledge and experience.
- May not provide detailed information on underwater features, such as deep channels or submerged structures.
- Observations may be subjective and dependent on the skills and experience of the surveyor.
- Survey effectiveness can be influenced by weather conditions, limiting the availability of suitable surveying days.
- Entails the risk of unintentional spread of invasive species if several catchments are surveyed and equipment is not properly cleaned between different water bodies.
- Involves inherent risks associated with on-site data collection, particularly in challenging environments.
- Could already be combined with a side-scan survey of bottom topography and substrates, but would require additional effort, time, and careful planning in advance.

Required resources and effort

- **Cost**: Low (Generally, costs are relatively low, especially compared to some high-tech methods like e.g. remote sensing).

- **Time**: Medium (While quicker than some methods, it still requires time for on-site surveying and data documentation).
- **Equipment**: Low (Basic equipment like boats and navigation tools are required, but it is less equipment-intensive compared to some advanced techniques.).
- **Experience/Expertise**: Low to Medium (Basic navigation skills are necessary, but deep technical expertise is not as critical. However, experience in interpreting observed structures and conditions is necessary to ensure good quality of observations).

4.3 Verification of identified habitats

This chapter deals with the pinpointing and verification of potential habitat areas in a second step from the previous identification of larger areas and sections within a catchment by field surveys, assessments, and habitat modelling. This step also includes quantification to a certain extent, as the frequency of habitat types, as well as their spatial extension must be assessed.

4.3.1 Criteria for habitat verification

Common criteria for the assessment of physico-chemical and structural parameters for sturgeon are available for a small range of habitat types only, due to gaps in knowledge on specific sturgeon requirements, ontogeny, autecology, and general ecology. Common habitat criteria were derived from literary sources (Holčík 1989; Bemis & Kynard 1997; Billard & Lecointre 2000; Gessner & Schütz 2011; McAdam et al., 2018). More information on habitat characteristics and criteria for different species, catchments, and habitat types from applied field research is available in the report on sturgeon habitats (Popp, S., 2024. Characteristics and locations of Sturgeon Habitats in European Rivers).

4.3.1.1 General water quality

The exact role of general water quality for sturgeon conservation still remains unclear. However, conditions hostile to aquatic life are a definite exclusion factor for a catchment or a section for the assessment of potential habitat. Sturgeon are sensitive to low levels of dissolved oxygen due to haemoglobin with a low oxygen affinity and lack of a compensatory strategy. Therefore, critical levels of O_2 concentrations for adults may already be reached when below 5 mg/l (Sullivan et al., 2003, Chebanov et al., 2011).

Hypoxic conditions are even less tolerable during early development and juvenile growth, as they are highly dependent on temperature, salinity, and dissolved oxygen, with optimal growth at oxygen concentrations >70% saturation (Gunderson 1998; Cech & Crocker 2002; Campbell & Goodman 2004; Niklitschek & Secor 2009a, b; Cech & Doroshov 2010; Kieffer et al., 2011). Tolerance for increasing salinities develops in anadromous sturgeon over time and with increasing body mass (Jenkins et al., 1993). Potamodromous species do not tolerate salinity. Oxygen consumption rates suggest increasing ionic and osmotic regulation impacts in younger fish at higher salinities (Allen et al., 2014).

An overview of values and ranges of water quality parameters to support sturgeon life under facility conditions, proven by experience and deemed beneficial, also as reference for open waters, are detailed in Table 12.

Table 12: *Recommended values and ranges of water quality variables for sturgeon (Chebanov et al., 2011, modified from Conte et al., 1988).*

Parameter	Value
Alkalinity, mg/litre as $CaCO_3$	50 – 400
Ammonia (unionized), mg/litre	< 0.01
BOD_5, O_2, mg/litre	< 2.5
Cadmium (soft water 100 ppm alkalinity), mg/litre (hard water 100 ppm alkalinity), mg/litre	0.004 0.003
Carbon dioxide, mg/litre	0 – 10
Copper, mg/litre in soft water	0.006
Dissolved oxygen, mg/litre	> 5.0 to saturation
Gas saturation	< 105 %
Hydrogen sulphide, mg/litre	0.002
Iron, mg/litre	< 0.01
Lead, mg/litre	< 0.03
Nitrite, mg/litre as N in soft water in hard water	< 0.1 < 0.2
Oxidability permanganate, O_2 mg/litre	≤ 10
Ozone, mg/litre	0.005
pH	6.5 – 8.5
Salinity, ppt for fry for juveniles for adults	0 – 0.5 0 – 3 3
Total hardness, mg/litre as $CaCO_3$	10 – 400
Total suspended and settleable solids, mg/litre	≤ 80
Zinc, mg/litre	0.03

4.3.1.2 Spawning and yolk sac larvae

Description:

- Absence of physical barriers for passage or presence of functional migration aids at barriers (e.g. locks, dams, reservoirs, thermal plumes, turbidity, sound, etc.) in the ecological corridor/habitat continuum, between the river mouth and spawning sites for anadromous populations, and between wintering sites, feeding sites, and spawning habitat for potamodromous populations.
- Deep river sections with elevated flow velocities compared to average current speeds, (McAdam et al., 2018; Kerr et al., 2010).
- Dominant hard clean bottom substrates with only scant periphyton growth; e.g. coarse gravel (Manny & Kennedy 2002), no sedimentation during spawning season and early development (McAdam et al., 2005).
- Interstitial spaces with good flow-through, well oxygenated, and only minor embeddedness of rough surface substrate in finer substrate fractions (Bain & Stevenson 1999; Du et al., 2011).
- BOD is low (empirical value from controlled propagation, not published) and water temperature supports timely early development until the yolk sac of larvae is resorbed and first (exogenously) feeding larvae transition to another habitat type (see "Externally feeding larvae" below).

Water depth: > 2 m

Water temperature: 13 – 20 °C, variation during the days of early development ≤ 2 °C (range and variation represent orientational values, not necessarily exclusion factors, have to be adapted to species, see also table 3: "Optimal species-specific temperature range during embryogenesis").

Current velocities (above spawning ground): > 0.6 m/s (maximum upper range not defined, observed values > 2.0 m/s).

Dissolved oxygen (DO) (interstitial spaces): > 6.0 mg/l.

Biochemical Oxygen Demand (BOD): Low, < 2 mg/l.

Salinity: 0.0 – 0.5 ppt.

Bottom substrates: gravel, cobble, boulders, rocks, pebble sizes > 25 mm,

also, hard clay bars (*A. stellatus*), bedrock (*A. oxyrinchus*) (Honţ et al., 2022; NOAA).

Sedimentation: Absent, scant embeddedness of substrate in fine sediment.

Flow: Steady after freshet, only minor variations, during the slow decline of the freshet.

Carrying capacity: The assumed natural incubation density of the female spawner. According to Derzhavin (1947) and Vlasenko (1970), applied by Gessner & Bartel (2000) and Arndt et al., 2006), the average fertility of a sturgeon spawner is 1 million eggs per female and an optimum density in the range of 1,000 to 3,500 eggs/m^2, resulting in an average spawning site area for one female of about 350 m^2.

Confirmation: Presence of spawning adult fish and documentation of embryos and/or yolk sac larvae.

4.3.1.3 Externally feeding larvae

Description:

- Calm-flow areas downstream (!) of spawning sites and sites of yolk sac larva development.
- Presence of food organisms for first feeding.
- Mixed substrates providing cover from predation.

Water depth: > 2.0 m/s.

Water temperature: 15 – 22 °C.

Current velocities (in habitat): N/A.

Dissolved oxygen (DO): > 6.0 mg/l.

Biochemical Oxygen Demand (BOD): Medium, 2 – 5 mg/l.

Salinity: 0.0 – 0.5 ppt.

Bottom substrates: Mixed substrates (clay, sand, and gravel).

Sedimentation: Low, < 10 mg/l (suspended particles).

Flow: N/A.

Carrying capacity: N/A. Potentially depending on the availability of appropriately sized feeding organisms (planktonic, e.g. copepods, daphnia; benthic, e.g. chironomidae, tubificidae). Species-specific and potentially depending on size of larva, gape of mouth and feeding behavior (Gessner et al., 2007).

Confirmation: Documentation of externally feeding larvae (yolk sac resorbed, digestive tract filled).

4.3.1.4 Young of the Year

Description:

- Low flow zones downstream of spawning sites and sites for yolk sac larva development in close connection to the main channel.
- Presence of food organisms.

Water depth: N/A.

Water temperature: 1 – 26 °C.

Current velocities (in habitat): 0 – 0.8 m/s.

Dissolved oxygen (DO): > 5.5 mg/l.

Biochemical Oxygen Demand (BOD): High, > 5 mg/l.

Salinity: 0.0 – 0.5 ppt.

Bottom substrates: Soft bottom substrates (e.g. silt, sand, clay), relatively stable to allow for the colonization and emergence of food organisms.

Sedimentation: Low, < 10 mg/l (suspended particles).

Flow: N/A.

Carrying capacity: N/A. Potentially depending on the availability of appropriately sized feeding organisms. Species-specific (e.g. piscivory in *Huso huso*) and potentially depending also on opportunistic feeding.

Confirmation: Documentation of feeding juveniles and alignment of digestive tract content with available spectrum of food organisms (gastric lavage).

4.3.1.5 Juveniles, subadults and adults > 1 year (riverine, marine for anadromous populations)

Description:

- Riverine and marine areas with presence of feeding organisms.

Water depth: N/A.

Water temperature: 1 – 26 °C.

Current velocities (in riverine habitat): 0 – 1.4 m/s.

Dissolved oxygen (DO): > 5.0 mg/l to saturation.

Biochemical Oxygen Demand (BOD): High, > 5 mg/l.

Salinities: If marine (anadromous), depending on respective environment (e.g. North Sea 34 – 35 ppt, Baltic Sea 2 – 30 ppt, Black Sea 17 – 20 ppt, Mediterranean Sea 38 – 39 ppt).

Bottom substrates: N/A.

Sedimentation: N/A.

Flow: N/A.

Carrying capacity: N/A. Potentially depending on the availability of appropriately sized feeding organisms. Species-specific (e.g. piscivory in *Huso huso*) and potentially depending also on opportunistic feeding.

Confirmation: Documentation of feeding individuals and alignment of digestive tract content with available spectrum of food organisms (gastric lavage).

4.3.1.6 Wintering (riverine)

Description: Deep pools or depressions in the riverbed, often in the vicinity of spawning sites.

Water depth: N/A.

Water temperature: N/A.

Current velocities: N/A (reduced near-bottom current).

Dissolved oxygen (DO): > 5.0 to saturation.

Biochemical Oxygen Demand (BOD): N/A.

Salinity: 0.0 – 0.5 ppt.

Bottom substrates: N/A.

Sedimentation: N/A.

Flow: N/A.

Carrying capacity: N/A.

Confirmation: Documentation of wintering sturgeon presence.

4.3.2 Methods

4.3.2.1 Water depth

Depth sounding, acoustic Doppler devices, total station surveying, or simpler yet effective (and also improvised) devices like tape measures or weighted lines may be used to measure water depth, if conditions like current velocities allow. No specific protocols are available for depth measuring at sturgeon habitat types.

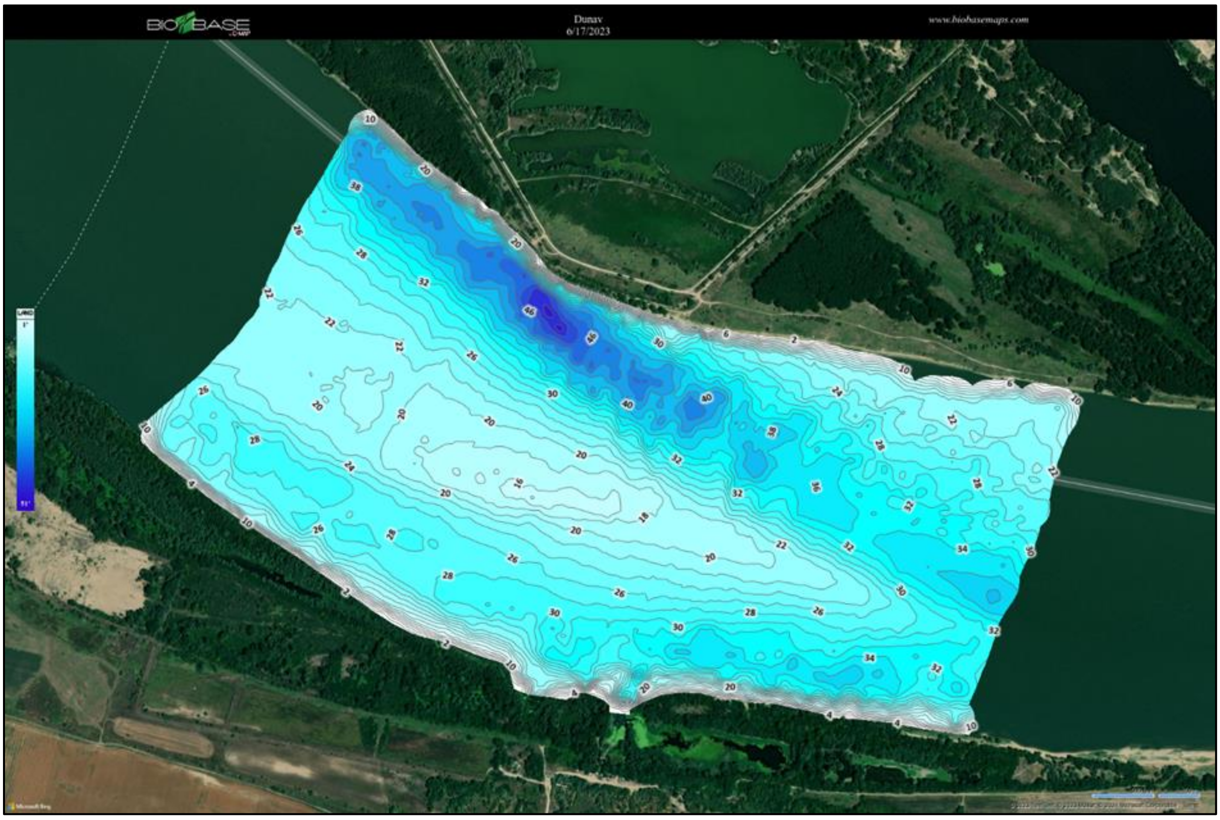

Figure 6: *Bathymetry map of a section of the Danube River near Belene / Bulgaria from sonar depth survey (© WWF-Bulgaria).*

4.3.2.2 Water temperature

Handheld temperature probes, temperature data loggers, thermocouples, and thermal imaging cameras may be used to measure and record water temperature at different locations and depths. There are no specific protocols available for measuring water temperature at sturgeon habitat types. However, it is an important variable for monitoring the timing of spawning, the length of incubation for predicting the timing of larval drift, and to monitor long term trends in terms of climate change.

4.3.2.3 Current velocity

An overview on different methods of measuring current velocities, based on a review of methods for monitoring streamflow by Dobriyal et al., (2017), including relevant conditions for their application, is provided in Table 13.

Whereas surface velocities may be useful for identifying river sections with elevated current speeds for the identification of spawning sites, assessment of functionality for sturgeon spawning will include measuring velocities on-site in the water column and directly above the spawning substrate, leaving current velocimeters and Acoustic Doppler Current Profilers (ADCP) as applicable methods in the field.

Table 13: Overview of methods for measuring current velocities (from Dobriyal et al., 2017, modified and amended).

Method	Float	Current meter (flow meter)	Acoustic Doppler Current Profiler	Electromagnetic	Remote sensing	Particle image velocimetry
Operational ease	easy	difficult	difficult	difficult	difficult	difficult
Cost effectiveness	inexpensive	expensive	(very) expensive	expensive	expensive	expensive
Accuracy	low	high	high	high	low	high
Time effectiveness	efficient	efficient	efficient	efficient	efficient	efficient
Ecological significance	non-polluting	non-polluting	non-polluting	non-polluting	non-polluting	non-polluting
Remarks	for small streams only typically measures surface currents	short term study only can measure velocities at specific depths, including the surface current meters are often deployed at various depths using a vertical profiling system	capable of measuring current velocities throughout the water column requires a significant effort including fixed stations,	can be used to measure water velocity at the surface or at specific depths, depending on the design and deployment	covers large areas but needs ground truthing generally used for surface currents	covers large areas and yields accurate results but estimates need validation and cannot be used in hilly terrain often used in laboratory settings can measure velocities at different depths, but its application in rivers may be limited

Covering large and deep riverine areas at different depths and from a boat with current velocimeters can be a very time-consuming process. One pass with an ADCP on the other hand can produce velocity current patterns of whole transects (Yorke & Oberg 2002). Consequently, the use of ADCPs for conducting velocity and discharge measurements of rivers has increased in the last decades. A variety of proprietary solutions including various device sizes, beam configurations, and frequencies, for use in both shallow streams and deep rivers, are available. ADCPs can be mounted on small remote-controlled-, tethered-, or powerboats, depending on the characteristics of the water body. They provide a reliable means of making fast and accurate discharge measurements even on large rivers under varying flow conditions including flooding events and enable the documentation of spatial extensions of current velocity patterns. However, ADCPs cannot measure velocity near the surface, at the riverbed, or near riverbanks, which is compensated for by software provided with the devices that estimates velocity in the unmeasured zones or should be compensated *in situ* by taking additional orientational measurements with a current meter. ADCP equipment is expensive and requires significant expertise and training, thus a basic consideration for a monitoring program would be if equipment and training should need to be purchased, or if ADCP assessments should be purchased as a service. There are no specific protocols available for measuring current flow velocities among the sturgeon habitat types.

4.3.2.4 Oxygen and other water quality variables

Dissolved oxygen concentration (DO) and oxygen saturation (O_2-%) as well as other water quality variables considered relevant (e.g. pH, temperature) should be measured *in situ* by using the appropriate field measurement equipment.

Generally, on-site field measurement devices should be calibrated according to manufacturer instructions. Any on-site field measurement of variables should be performed prior to any biological sampling to avoid bias due to sediment turbidity caused by the sampling team. All on-site field data and observations must be entered in a field protocol (Wolfram et al., 2019).

To document the functionality at sites of spawning and early development, oxygenation should also be measured close to or directly in the interstitial water of the substrate using optical fluorescence sensor technology (Neill et al., 2014). Temporal trends of key habitat-relevant chemical and physical variables should be monitored over an extended period and throughout the daily cycle to capture both the minima and maxima of the values under consideration.

In case of actual or suspected detrimental impacts on water quality affecting sturgeon life and the life-cycle, regular monitoring events or even real-time monitoring in and at crucial locations and timings should be conducted. General guidelines for measuring DO in fresh- and seawater and for monitoring general water quality have been developed (Doudoroff & Shumway 1970; Culberson 1991; Best et al., 2007; Water Framework Directive (2000/60/EC); Behmel et al., 2016; HELCOM/Andreasson & Kronsell; U.S. Geological Survey 2020). No specific protocols are available for measuring oxygen and assessing water quality at sturgeon habitat types.

4.3.2.5 Biological Oxygen Demand (BOD) and water samples

Taking water samples and measuring BOD are standard procedures in the monitoring of surface waters and numerous protocols exist. It is mentioned separately here, as the BOD and its sampling as environmental variable has some significance for sturgeon. An example on the basis of Wolfram et al., (2019) is provided subsequently.

The material of the sample container, and any required treatment of water samples (e.g. filtration or stabilisation) depends on the variables to be analyzed and as specified by the laboratory. For many variables HD PE bottles are appropriate. For BOD_5 however, Karlsruhe bottles should be used. Sampling containers (as well as any filtration units and preservation/stabilization substances) should be labelled clearly attributable to the sampling site or river.

Taking water samples for chemical analyses must be done before taking biological samples to avoid bias effects from disturbed sediment. During sampling, care shall be taken concerning possible disturbing effects from upstream, which may cause enhanced turbidity.

Before filling the sampling bottle, it must be rinsed with sample water. The sample is taken directly by hand with a separate jug or by using a (telescope) rod with a jug at its end. If two or more bottles are sampled from one site, all bottles must be filled from one jug. Samples should be collected in a manner that does not permit contamination by debris, sediment, or (larger) particles of any kind. Samples should also not include surface film or floating layers. Sampling near or at the surface, bottom, or bank of a river should be avoided. Most representative samples are collected about 30 cm beneath the surface or in the middle of the water column.

Sampled water should be poured into the bottle without any turbulence along the inner wall of the bottle to avoid additional contact with air (oxygen). The HD PE bottle must be filled completely and slightly squeezed during the screwing on of the bottle cap so as to drive out air bubbles, as this allows lossless homogenizing in the laboratory.

For $BOD_{5,}$ two 250 mL Karlsruhe bottles (glass-stoppered conical flask) are filled. The sample must reach at least 1 cm into the glass funnel on top of the bottle. Then, a magnetic stirring rod is added, and the bottle closed with the glass stopper. The overlaying water in the funnel must not be poured away.

All bottles are put into cooling boxes immediately after sampling (Wolfram et al., 2019).

For own determination of BOD_5 and sampling specific sturgeon habitat types, the following instructions should be followed:

- Use clean, sterile containers for water sample collection and sampling procedure as described above.
- A reliable dissolved oxygen (DO) meter to measure initial (at time of sample collection) and final dissolved oxygen concentrations should be used.
- A controlled temperature incubator (20 ± 1°C) for BOD bottle incubation must be maintained.
- Clearly identify and document the exact location of sampling sites.
- Collect water samples at various depths and locations to capture potential variations in BOD_5.

- Plan sampling events to coincide with critical periods for sturgeon habitat use, considering seasonal and life-cycle phase factors.
- Preserve samples with appropriate preservation agents to minimize changes in BOD during transportation to the laboratory.
- Incubate BOD_5 bottles at 20 ± 1°C in the dark for 5 days, measuring dissolved oxygen again at the end.
- Include control bottles without a carbon source to assess baseline oxygen changes.
- Calculate BOD_5 as the difference between initial and final dissolved oxygen concentrations.
- Validate results by comparing with control bottles and consider repeating analyses if needed.
- Document sampling details, analysis procedures, and results accurately.
- Provide interpretations of BOD_5 levels in the context of sturgeon habitat requirements.
- Based on BOD assessments, offer recommendations for habitat management and potential remediation actions to maintain or enhance water quality for sturgeon.

4.3.2.6 Hydrological conditions

Stream gauges, flow meters, and automated water samplers may be used to observe and record relevant changes or patterns in discharge significant for habitat use (e.g. as trigger for spawning migration, impact on habitat conditions) (Burt 2003; Brierly et al., 2010; Chen & Wu 2011). No specific protocols are available for assessing and interpreting hydrological conditions at sturgeon habitat types. Examples for research into hydrological conditions with regard to sturgeon habitat and habitat use can be found in Hamel et al., 2014; Porter 2017; Porter & Schramm 2018; Klimley et al., 2020, and Chang et al., 2021.

Figure 7: *Historical sturgeon spawning site in the Narva River during sufficient discharge (© Meelis Tambets).*

Figure 8: *Same spawning site as in the photo above, yet under insufficient discharge conditions for sturgeon spawning (© Meelis Tambets).*

4.3.2.7 Hard bottom substrates

Substrates may be sampled directly, visually analyzed and classified using sediment corers, grab samplers, dredges or by underwater photography, videography, and visual sonar. Divers have also been deployed to survey substrates at hard-to-reach sturgeon spawning sites (Arndt et al., 2006).

General protocols for sampling of different substrates in water bodies are available (e.g. Mudroch & Azcue 1995, Clapcott et al., 2011, Skilbeck et al., 2017, DES 2018, Tuit & Wait 2020, BC Ministry of Environment, Lands and Parks), classification, processing of samples and analysis of substrate types and compositions vary between studies and are not standardized with regard to sturgeon habitat.

A range of methods have emerged over the past decade allowing for the measurement of clast dimensions. These clast dimensions are organized in the so-called "Wentworth" scale, classifying particles of loose sediment in a base-2 logarithmic fashion, ranging from small particles of clay to large boulders. Measurement of particle sizes can be performed with either manual- or image-based methods. Manual-based methods involve the physical measurement of sediment grains (e.g. by sieving of samples). At a basic level, grains are treated as triaxial ellipsoids that can be measured with three axis, a, b, and c, with the a-axis denoting the longest, b, the intermediate and c, the shortest axis.

Comprehensive descriptions and protocols of classic grain size measurement methods, as well as advantages and disadvantages of methods can be found in Bunte & Abt (2001).

However, direct sampling disturbs the riverbed, potentially interfering with habitat use, requires transportation of heavy sediments and subsequent laboratory analysis, is relatively expensive and time-consuming.

Hard bottom substrates, such as those which are encountered at sites of sturgeon spawning, embryonic, and yolk sac development pose specific challenges for monitoring. High current velocities and large substrate sizes at *Acipenser spp.* spawning locations often make quantitative direct sampling and the use of grabs and dredges ineffective (Chiotti et al., 2008). Therefore, qualitative techniques were often used when characterizing spawning substrate (Parsley et al., 1993; Sulak and Clugston 1998), leading to ambiguity when comparing spawning site substrate between studies. In contrast, photographs and underwater filming have been used successfully to quantify substrate particle sizes and substrate heterogeneity in aquatic systems resulting in a quantitative, more comparable measurement system (Boyero 2003; Whitman et al., 2003; Rubin 2004; Graham et al., 2005a; 2005b; Detert & Weitbrecht 2012), but requiring provisions to ensure a fixed distance, an optimal distance for identifying substrate particle sizes, for minimizing distortion effects, and for maximizing the covered image area. Thus, photographs and video do not require manual laboratory analysis, and allow for greater coverage and frequency of observations (Rubin et al., 2007), but are limited by a small field of view, possible positioning errors, and highly turbid conditions.

Emerged substrates can also be assessed by remote sensing using drones and piloted aircraft (Carbonneau et al., 2005; Buscombe et al., 2010; Dugdale et al.,

2010; Carbonneau et al., 2018; Woodget et al., 2018), but do not necessarily depict the situation of adjacent submerged substrate in deep water and high current velocities.
Obtaining sufficient underwater samples in this way, to characterize or map an entire reach or river may be too time-consuming and costly to be practical in large rivers and may not adequately capture the spatial distribution or pattern (e.g. patchiness) of sediments (Hamill et al., 2016). Alternatively, hydroacoustic devices (e.g. multibeam sonar) can be used to classify textures over large areas without a strict requirement of direct sampling of bed material (Kaeser and Litts 2010; Kaeser et al., 2012; Buscombe et al., 2014, 2015). Bed texture classification mapping is achieved from multibeam sonar either by analyzing high-resolution bathymetric maps or the acoustic backscatter of riverbed sediments.
Statistical algorithms have been developed to create bed texture maps from such high-resolution digital elevation models derived from multibeam sonar by developing a calibration between grain size and the standard deviation of local elevations (Brasington et al., 2012). However, editing multibeam sonar data requires expensive equipment, experienced operators, commercial software packages, and extensive amounts of post processing before an accurate digital elevation model can be produced (Kaplinski et al., 2009). These requirements limit the use of multibeam sonar echosounders to specialized cases where bathymetric information is the primary goal (Hamill et al., 2016).

4.3.2.8 Substrate assessment by side-scan sonar

Side-scan sonar (SSS) can be used for sturgeon habitats (Kaeser & Litts 2010; Hook 2011; Kaeser et al., 2012; Litts & Kaeser 2016; Walker & Alford 2016) and for population monitoring (Hughes et al., 2018; Kazyak et al., 2020; Flowers & Hightower 2013; Flowers & Hightower 2015; Fund et al., 2016; Andrews et al., 2020; Vine et al., 2019; Brown 2020). It allows for the identification of large sturgeon and individual fish under certain conditions, as well as for the relatively quick benthic mapping of river sections. It was found to be especially useful to create continuous high-resolution images of the floor of a waterbody, including bathymetry and substrate compositions, even under difficult conditions like in deep, turbid, or difficult to access rivers. A complete overview of this technology and associated methodologies can be found in Blondel (2010).
One must be aware however, that without calibration and subsequent processing, only the "roughness" of the riverbed is assessed. SSS is an active sonar system that consists of a projector, a hydrophone, and a recorder or display unit. The projector converts an electrical pulse into sound waves, the hydrophone performs the reverse. The projector and hydrophone are usually combined into one device, the transducer. A transducer can either be towed behind a vessel or fitted directly onto the boat, allowing surveys in all kinds of navigable water bodies. The transducer emits a fan shaped acoustical pulse outward in both directions perpendicular to the path of the tow vessel or boat. As the sound waves propagate outward and hit submerged surfaces, waves are reflected back to the transducer with an intensity determined by the shape, density, and position of the objects encountered. The variation in intensity is displayed by the recording/display unit

as variation in brightness of the displayed signal, with light and dark portions of the display representing strong and weak echoes, depending on the reflective properties of objects on the bottom. Each pulse is followed by another, and the resulting lines of display form a coherent picture of the bottom. Coupled with positional information from GPS, these images may be georeferenced for spatially accurate information about the river bottom and its mapping.

Commercial survey-grade side-scan sonar has been used for high-resolution seafloor imaging (Blondel 2009). This technology has been widely used, mainly for marine and lentic environments, as the equipment used to be large, bulky, and quite expensive. The sensors were contained within a torpedo shaped towfish, which had to be towed by a research vessel. The towfish itself could reach a length exceeding 2 meters. These conventional "towfish"- transducers, however, have limited applicability in rivers and streams because the transducer is towed at depth, which limits its utilization to large water bodies, may also impede accurate positioning of scans, and is quite expensive.

Comparably low-cost recreational-grade side-scan sonar platforms used for recreational fishing, for example, have become available recently (Hamill et al., 2016). These side-scan sonars can be deployed in shallow rivers, on a variety of vessels, operated remotely or by a single person with limited sonar experience, are not as limited by depth to the same degree as towed side-scan or by turbidity to the same extent as underwater video (Kaeser and Litts 2010). Side-scan sonar transducers can be mounted directly to a boat, facilitating the deployment in all kinds of navigable streams.

Processing of side-scan recordings has previously been achieved using commercially available software packages for maritime mapping and visual interpretation. Other software packages are used to convert the recordings from binary format to various spatial data formats where the data are easily visualized. Sediment patches can then be delineated to create bed texture maps (Kaeser et al., 2012). However, reproducing bed texture maps derived from visual interpretation is difficult as interpretations of the transitions from similar sized sediments are subjective. Buscombe et al., (2015) proposed an automated method to classify bed textures according to the Wentworth-style groupings, analyzing data recorded by a recreational-grade side-scan sonar with appropriate acoustic corrections applied.

Hamill et al., (2018) developed a method for side-scan sonar images for automated segmentation of bed textures into between two to five grain-size classes. Second-order texture statistics were used in conjunction with a Gaussian Mixture Model to classify the heterogeneous bed into small homogeneous patches of sand, gravel, and boulders with an average accuracy of 80%, 49%, and 61%, respectively. Reach-averaged proportions of these sediment types were within 3% compared to similar maps derived from multibeam sonar.

A combination of a specialized digital side-scan sonar system for bottom typing with GPS, GIS, and statistical software, may be used to provide georeferenced data and to acoustically map bottom substrate types, locations, and bathymetry in one process.

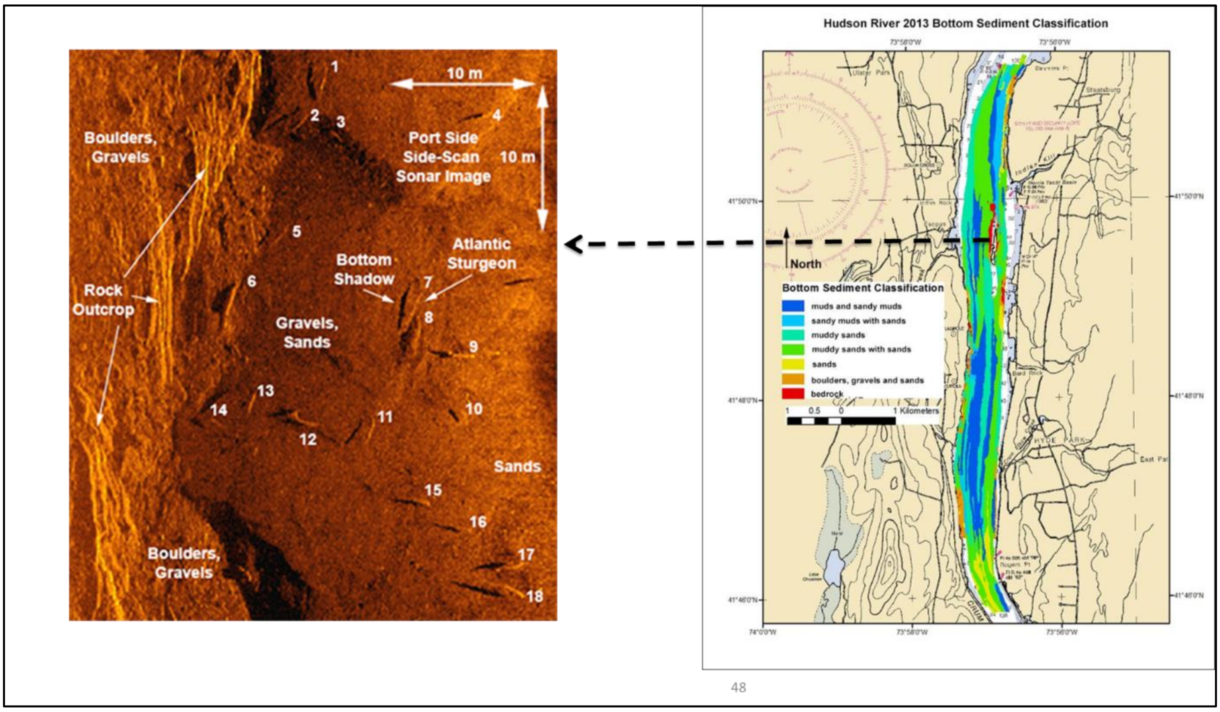

Figure 9: *The application of SSS in bottom sediment classification. Nos. 1 through 18 in the left sonar image serve as labels for individual sturgeon (© Dewayne Fox).*

No standard protocols for assessing sturgeon habitat with SSS exist to date and a variety of proprietary combinations of devices (adding capabilities like bathymetry and GPS positioning) and software are available, which also includes cheaper consumer-grade devices and open-source software that might be applicable and useful in a given scenario (Buscombe 2017; Hamill 2018). Table 14 summarizes main aspects for application of the above.

Table 14: *Main aspects of substrate sampling and mapping for different methods as described in the text.*

Method	Effort	Pros and cons
Manual	**Cost:** High (labor for comprehensive sampling) **Equipment:** Low (for sampling, but involves laboratory analysis) **Time:** High (for larger spatial scales) **Experience/ expertise:** Medium	• methodologies and protocols readily available • useful for ground truthing and calibration of image-/acoustic based methods • disturbs the habitat • deep water/ high current velocity habitat difficult or impossible to sample • might involve sampling by divers • not suitable for large-scale mapping and quantification of habitats, low spatial coverage

Photographic (videographic)	**<u>Cost</u>:** Low (ground-based and handheld, medium to high for drones and piloted aircraft) **<u>Equipment</u>:** Low to High (ground, air based, piloted) **<u>Time</u>:** Low to Medium (depending on mapping length) **<u>Experience/ expertise</u>:** Medium to High	• sampling possible in deep water/ high current velocity habitat • useful for ground truthing and calibration of acoustic- based methods • non-invasive • limited by a small field of view, possible positioning errors and highly turbid and low light underwater conditions
Side-scan sonar	**<u>Cost</u>:** Medium or High (consumer- or commercial survey grade) **<u>Equipment</u>:** Medium to High (consumer grade vs. commercial survey grade), boat, towfish or transducer, towing vehicle, hard- and software **<u>Time</u>:** Low **<u>Experience/ expertise</u>:** Medium to High	• suitable for large-scale substrate mapping by continuously imaging several kilometres of channel in few hours • provides high-resolution imaging capabilities, allowing for detailed visualization of river bottom and habitat features • non-invasive • requires calibration by other methods and post processing for quantification • combinations of devices, software, and functionalities (e.g. multibeam bathymetry, GPS referencing, GIS integration, continuous mapping) available for high-priced proprietary devices • lower priced consumer grade devices and application of open-source software solutions requires adaptation and the development and adjustment of individual solutions

4.3.2.9 Soft bottom substrates and food organisms (riverine)

Gastric lavage and analysis of the content of the digestive tract of sturgeon can provide information on preferred food organisms for different life stages of sturgeon and thus potentially certain habitat criteria (Holčík 1989; Damon-Randall et al., 2010; Crossman et al., 2016; Zarri & Palkovacs 2019; Margaritova et al., 2021). For the description of gastric lavage in sturgeon, see Technical Guideline for ex situ Conservation Measures in Sturgeon (Gessner et al., 2024). For a comprehensive overview of gastric content analysis in fish see Manko (2016).

Figure 10: *Gastric lavage for the identification of food organisms performed on a juvenile sturgeon (© WWF-Bulgaria).*

The available literature lists different types of food organisms for different species and life-stages, but no information with regard to mandatory key food species (opportunistic feeding and changes in the macroinvertebrate communities also plays a role (Strel'nikova 2012)), required minimal abundances or carrying capacities of nursery and feeding habitats based on biomass or abundance of food organisms as well as specific habitat criteria to be derived.

The two ecological groups of zooplankton and benthic invertebrates on and in soft bottomed substrates (low-flow zones in or in close connection with the main channel in a riverine environment) play a major role in feeding of sturgeon larvae, juveniles, subadults, and adults, including organisms from such groups as crustaceans, insect larvae, oligochaetes, and polychaetes (Muir et al., 1988; Holčík 1989; Chiasson et al., 1997; Muir et al., 2000; Nilo et al., 2006; Gessner et al., 2007; Brosse et al., 2011; HELCOM 2019; Sun et al., 2019; Holley et al., 2022). Numerous protocols are available for the sampling of zooplankton and benthic invertebrates for qualitative, semi-quantitative, and quantitative purposes. According to Blaž et al., (2021), the primary goal of sampling for sturgeon food organisms is the confirmation of presence and relative abundance, so that qualitative sampling is generally sufficient. This, however, depends on the specific objectives of the respective study.

Examples are given as reference below (adapted from BC Ministry of Environment, Lands and Parks). Specific protocols should be developed or adapted, if required by study design and purpose.

4.3.2.9.1 Zooplankton (e.g. Rotatoria, Cladocera) consists of free-floating animals suspended in open or pelagic waters. They are generally collected with a conical net that has a specific mesh size (ranging from as small as 64 μm to as large as 256 μm). Small mesh openings will clog more readily than larger ones, but small organisms will pass readily through larger openings. The mesh size required for a particular water body will depend on its productivity and the purpose of the study. The preferred net mesh, when appropriate, is 64 μm with a net mouth diameter of 20 cm.

The net is lowered to a particular depth and pulled up directly through the water column (vertical tow). Alternatives to the vertical tow are horizontal and oblique tows in which various strata of the lake are sampled individually (horizontal tow) or as a composite (oblique tow). These are elaborate techniques that require specialized equipment rigged to the boat and a tow net that has remote open and close capabilities. Unless there is specific need for data from horizontal and oblique tows, they are not used. Therefore, the vertical tow is the only protocol that will be described below.

PROTOCOL (zooplankton, vertical tow)

(a) Ensure rope is securely fastened at the plankton net opening and that the dead end is tied to the boat.

(b) At the designated site, lower the net to depth outlined in the project design.

Note: The actual distance that the net travels through the water must be recorded and the total volume of water that passes through the net must be calculated (see formula for quantification below).

(c) In smaller water bodies, haul the net hand over hand with a steady, unhurried motion at a rate of 0.5 m/s. In large water bodies, when long net hauls are conducted, use a davit, meter wheel, and winch. The maximum tow speed used should be 1 m/s.

(d) Once the net is at the surface, wash the net by raising and lowering the net body below the net mouth in the water. Then squirt de-ionized water against the outside of the netting and from the top downward. This washes any adhered plankton down into the cod-end (removable container at the end of the net).

(e) Disconnect the cod-end and carefully decant the water and plankton into a pre-labeled bottle. Rinse the cod-end several times, pouring each rinsate into the bottle (this ensures that all plankton are collected).

(f) Wash the net by rinsing (pulling it through the water without the cod-end). This is an absolute necessity before proceeding to the next sample site (particularly between different water bodies).

(g) Preserve the sample with 70% ethanol (70mL of 100% ethanol for each 30 mL of sample volume) and place in the cooler for shipping or transportation to the lab for analysis.

Formula: Volume (V) of water through a zooplankton tow
$V = \pi r^2 d$
V: Volume of water filtered through sampler
π: 3.1416
r: radius of net mouth
d: depth of net sampler at start of vertical haul (total length of course through water)

Pros and cons

- Provides a method for quantitative assessment by calculating the volume of water filtered.
- Requires large diameter net to avoid evasive movement of mobile zooplankton.
- Can be adapted to various water bodies, including both small and large water bodies.
- Allows for preservation of collected samples for further laboratory analysis.
- Requires specialized equipment for horizontal and oblique tows, if needed.

Required resources and effort

- **Cost**: Low
- **Equipment**: Low (Conical net with specified mesh size, boat, rope, bottles for sample preservation, de-ionized water, ethanol for preservation)
- **Time**: Low
- **Experience/Expertise**: Low to medium (Basic training for net handling and sample preservation)

4.3.2.9.2 Benthic invertebrates in lakes or large, slow-moving rivers are generally collected in the same fashion as sediment samples. The processing of the sample once it has been collected is where the techniques differ. The type of sampler to be used at a particular site will depend on the site conditions and the purpose of the study (e.g. Ekman grab, Petersen grab, Ponar grab, Van Veen grab, core sampler). The equipment to be used (grab or core sampler) will be dictated by the project design and must be outlined in the field logbook and pre-sampling checklist.
PROTOCOL (boat sampling with a grab sampler)

(a) Ensure that the rope is securely fastened to the sampler and that the loose end of the rope is tied to the boat.

(b) Set the grab sampling device with the jaws cocked open. **Great care should be taken while dealing with the device while it is set as accidental closure can cause serious injuries.**

(c) Lower the sampler until it is resting on the sediment (its own weight is adequate to penetrate soft sediments). At this point, the slackening of the line activates the mechanism to close the jaws of the Ponar and Petersen grabs.

(d) For the Ekman grab, send the messenger down to 'trip' the release mechanism.

(e) Retrieve the sampler slowly to minimize the effect of turbulence (which may result in loss/disturbance of surface sediments).

(f) Place a container (i.e., a shallow pan) beneath the sampler just as it breaks the surface of the water.

Note: If the jaws were not closed completely, the sample must be discarded. Discard the sample into a bucket if the second collection attempt is made from the same general area. Dump the unwanted sample only after the "real" sample has been successfully collected.

(g) Place a sieve between the sampler and the pan and gently open the jaws and allow the sediments to empty into the sieve. The size of the sieve mesh will depend on the purpose of the study, but a common mesh size is 0.20 mm (200 μm). This size represents the practical lower limit for general study of benthic organisms. It is not as crucial to have small mesh size when the only analysis to be conducted is biomass.

(h) Immediately record (in the field logbook) observations regarding the appearance of the sediment (i.e., texture, color, odor, presence of biota, presence of detritus).

(i) Rinse the sieve with de-ionized or on-site water to remove as much sediment as possible.

(j) Transfer the organisms to a pre-labeled sample bottle and preserve with 70% ethanol. Formalin may be used as a fixative for initial preservation but should be subsequently washed and transferred to 70% ethanol.

Figure 11: *Collecting substrate from a grab-sampling for further analysis of presence and composition of food organisms (© WWF-Bulgaria).*

PROTOCOL (boat sampling with a core sampler)

(a) Open the valve and set the trigger mechanism. Ensure the rope is securely fastened to the corer and attach the loose end of the rope to the boat.

(b) Most corers are designed to be simply lowered into the sediment and fill the core tube by their own weight and need not be dropped from any height. Consideration of the type of corer used and the nature of the sediment being sampled will need to be taken into account.

(c) Send the messenger down to release the trigger mechanism.

(d) Carefully retrieve the sampler and place a stopper into the bottom opening **before** removing from the water to prevent loss of the sample.

(e) Remove the core tube or liner from the corer and stopper the upper end.

Note: Once on shore, the sample can be treated as a bulk sample, or it can be sectioned and the organisms separated from the sediment in strata.

(f) For bulk samples all the sediment may be sieved as per the grab samples above. Otherwise, the sediment should be sectioned in regular intervals as it is extruded (record the thickness of each stratum the length of entire core). Each stratum may be sieved, and its contents placed in pre-labeled sample bottles or, the unsieved sediments can be placed directly into pre-labeled sample bottles.

(g) Preserve the samples with 70% ethanol.

Note: It is preferable to section the core as soon as possible after it is retrieved. As the sediment warms, it tends to expand in the core tube. With warming, decomposition gases are liberated at a much faster rate and if they bubble through the core they will disturb the stratigraphy.

A description of subsequent processing of samples is provided by Soucek et al., (2023).

After sampling in the field, preserved samples are typically returned to a laboratory for separation, identification, and the counting of collected invertebrates. Out of numerous methods available for counting, the primary choice is between the subsampling method and the full enumeration method. Qualitative (like in this case) and semiquantitative sampling methods typically involve subsampling, where a fixed count of organisms is separated from the litter material. Fixed counts can be limited to between 100 to 550 organisms (Carter and Resh, 2001). Although full enumeration of every organism collected in a sample is quite time consuming, there are multiple benefits to this approach. These benefits include:

- The identification of a greater number of taxa, and, in particular, a greater number of rare taxa (Pence et al., 2021).
- The ability to estimate total abundance and, if a fixed area was sampled, density of organisms per unit area.

Once invertebrates have been sorted and enumerated, organisms have to be identified to some level of taxonomic specificity. Identifications are usually made at least to family level, but preferably to "lowest practical taxonomic level", which for many taxa is genus. Identification to species level has its benefits, but typically requires a high degree of specific expertise, as well as often rearing organisms to adult stage for identification.

Pros and cons

- Penetrates into sediments, providing a comprehensive view of benthic organisms.
- Allows for sampling and analysis of sediment in strata, providing detailed information on sediment characteristics.
- Enables preservation of samples for later laboratory analysis.
- Includes protocols for recording observations about sediment appearance, texture, and biota.
- The sampling process can potentially disturb surface sediments, affecting the accuracy of results and habitat use.
- Primarily designed for soft sediments and may not be suitable for all types of substrates.
- The full enumeration method can be time-consuming, especially when dealing with a large number of samples.
- Depending on the research objectives, a combination of methods may provide a more comprehensive understanding of especially food sources for sturgeon larvae.

Required Resources and effort

Cost: Medium.

Equipment: Medium to high.

Grab sampler or core sampler (Boat, rope, sieve with specified mesh size, bottles for sample preservation, de-ionized water, ethanol for preservation).

Time: Medium to high.

Experience/Expertise: Medium to high (Handling specialized equipment, specific knowledge required for identification of species groups, species, and developmental stages).

4.3.2.10 Habitat modeling

Statistical modeling in sturgeon conservation enables the assessment of habitat preferences, the identification of key environmental factors influencing sturgeon presence, and ultimately informing targeted conservation strategies to safeguard sturgeons and their habitats (De Kerckhove et al., 2008; Jarić et al., 2014). Habitat modeling may serve both the identification and verification of habitats, depending on its implementation within a habitat monitoring program. Habitat modeling within the context of this document is assigned to the verification of areas and sections bearing the criteria of potential habitat types.

Foremost, habitat models can help to create predictions of sections and areas utilized which can be verified later in tracking/telemetry, observational or netting studies to improve the model predictions, and as such, the quality of fit between expected and real habitat utilization in a system under changing conditions. Modeling combines scientific data, geospatial information, and mathematical algorithms to describe habitat quality or the processes which enable habitat use in terms of modeling and the relation of distinct habitat features to the species' preferences for certain boundary conditions.

These models help identify the factors that influence sturgeon habitat selection and distribution. GIS can be utilized to incorporate geospatial data into habitat models. This can include pertinent data on river morphology, bathymetry, land use, and other spatial information.

A variety of models have been used in ecological sciences and for different research questions. **Species Distribution Models** (SDMs) and **Habitat Suitability Index** (HSI) Models are two main types used as examples in this document, because they are often used for aquatic environments and sturgeon habitat types as well.

SDMs predict the probability of sturgeon occurrence based on environmental variables, helping identify suitable habitat types, and potential range shifts (Yi et al., 2010; Melo-Merino et al., 2020; Charbonnel et al., 2023). They provide a tool not only for understanding sturgeon habitat preferences, but also for predicting how changes in environmental conditions might impact their distribution. In order to do so, SDMs need to have clear criteria for habitat choice of the species in question and the associated drivers, as well as sufficiently sound model data for the environment to be assessed. This requires statistical analysis of the 1. **observed distribution data** in combination with the potentially underlying 2. **impacting factors** (e.g. depth, current velocities, turbidity, temperature, substrate composition, abundance of food organisms). Since in most cases either one or the other is lacking, SDMs can be produced if fish are present (without any skewed collection methods) and this can help to describe habitat characteristics that are utilized. The data requirements and quality constraints limit the applicability and the robustness of the results either way.

SDMs can help assess the cumulative effects of multiple stressors on sturgeon habitat such as the potential impacts of climate change on sturgeon habitat by projecting how shifting temperature and precipitation patterns might alter habitat suitability.

HSI models are also valuable tools for assessing the potential impacts of various factors on sturgeon habitat and habitat types by providing a structured framework to quantify habitat quality and suitability for sturgeon, based on specific environmental variables (Haxton et al., 2008; Collier 2018; Collier et al., 2022). HSI models allow for the evaluation of the potential impacts of human activities, changes in water quality, or alterations to habitat features on sturgeon populations. By adjusting the assigned scores for habitat variables affected by these impacts, one can also predict how the overall habitat suitability might change and explore the potential consequences of different management actions or assist in identifying priority areas for habitat restoration efforts.

The main features of SDM and HSIs and differences between these two types of habitat models are outlined in Table 15.

Table 15: *Main features and differences of Species Distribution- and Habitat Suitability Index Models*

Feature	Species Distribution Models (SDMs)	Habitat Suitability Index (HSI) Models
Objective	Prediction of the potential distribution of a species in geographic space. Aiming at answering the question within a given large area or catchment: "Where is suitable habitat for sturgeon likely to be found?"	Evaluation of quality of specific habitat types for a species or a developmental phase. Aiming at answering the question: "How suitable is this particular area as sturgeon habitat type?"
Data Input	Relying on species occurrence data (presence-absence) and environmental variables (e.g., temperature, depth, water quality) to model the distribution.	Requiring more detailed data on habitat characteristics, often involving expert knowledge and field surveys to assess factors like substrate type, water flow, and availability of food organisms.
Output	Spatial distribution maps that highlight areas with a high likelihood of sturgeon presence.	Scores or indices that reflect the suitability of specific habitat patches for sturgeon
Applicability	A good tool for understanding the broader ecological requirements of sturgeon and identifying potential habitat locations, excelling at	Valuable for fine-scale assessments of habitat quality within a given area, excelling at identifying habitat types

	identifying suitable habitat over a large geographic scale.	within larger areas identified by SDMs.
Research questions	Useful for questions related to habitat suitability and large-scale conservation planning, such as identifying regions where conservation efforts should be prioritized or assessing how climate change might affect species distribution.	Useful for questions related to site-specific conservation and management decisions. Helpful in identifying areas where habitat restoration or protection efforts should be concentrated and assessing the potential impact of specific threats or interventions on sturgeon habitat types.

In summary, SDMs are more suitable for identifying large-scale habitat suitability and understanding broad habitat preferences of sturgeon (usually not transferrable between catchments, Haulsee et al., 2020). On the other hand, HSI models are valuable for assessing the quality of specific habitats within a region and making detailed conservation decisions. Combining both approaches can provide a comprehensive understanding of sturgeon habitat requirements and guide effective conservation strategies at various spatial scales.

Pros and cons

- Habitat models allow for the verification (or identification) of potential habitat types for sturgeon, as well as areas with a high potential for restoration. They may also provide predictive insights into species distribution and impacts like climate change or into the effects of human interventions in the system.
- However, a model can only be as good as the data it is built on; accurate and reliable data is fundamental for robust habitat modeling. Also, data collection methods have to be coherent and standardized.
- HSIs may vary among systems and are not necessarily transferrable, therefore widespread application is limited.
- Models should also always be validated to assess their accuracy and reliability and since habitat is dynamic, regular updates to models are mandatory. Habitat monitoring can be quite resource-intensive and requires funding, skilled personnel, equipment, and the acquisition or transformation of suitable data from existing sources or through own field assessments can be challenging.

Required resources and effort

Developing and implementing habitat models for sturgeon habitat types involves several specific resources. While the overall approach may share some common elements between SDMs and HSI models, there can be differences in the required resources due to the nature of the models. While both SDMs and HSI models require data, modeling software, and domain expertise, HSI models place a stronger emphasis on expert input to define habitat preferences and assign scores. However, there recently has been a movement away from expert opinion to using modelled suitability factors (e.g. Generalized Linear Mixed Models (GLMM) or occupancy based) towards more data-driven and statistically rigorous methods (Haxton 2023, pers. comm.). Additionally, SDMs may require more sophisticated statistical or machine learning skills for model development. Below is a breakdown of the resources needed for both types of models.

Species Distribution Models (SDMs) require:

- High-resolution environmental data, including bathymetry, temperature, substrate type, and flow characteristics.
- Presence and absence data of sturgeon occurrences in the study area.
- Geographic Information System (GIS) software to manipulate and analyze spatial data.
- Access to relevant GIS datasets such as topography, hydrography, and land use/land cover.
- Statistical or machine learning software for building SDMs. Common tools include R, Python (with libraries like scikit-learn, TensorFlow, or Keras), or specialized modeling software such as MaxEnt.
- Expert knowledge to identify and select key environmental variables that influence sturgeon distribution, growth, and survival.
- Knowledge of the reference points for these environmental variables.
- Reliable and comprehensive data on sturgeon occurrences (presence) and non-occurrences (absence) across the study area.
- Metrics to assess model performance, such as AUC-ROC, AIC, or cross-validation techniques.
- Access to high-performance computing resources that can expedite model training and evaluation for computationally intensive modeling techniques (optional).

Habitat Suitability Index (HSI) Models require:

- Similar high-resolution environmental data as for SDMs.
- Expert knowledge to assign scores to different habitat variables and create the HSI formula.
- Software for calculating the HSI based on assigned scores and habitat variables.
- Historical data for calibrating and validating the HSI model.
- Collaboration with well-versed experts familiar with sturgeon ecology and habitat preferences to ensure accurate scoring of variables.
- GIS tools for mapping the resulting HSI values to visualize habitat suitability across the study area.
- Long-term environmental data to account for temporal variations in habitat suitability.

4.4 Confirmation of sturgeon habitat utilization

Confirmation of actual habitats is achieved by the documentation of actual habitat utilization, first by documentation of presence of individuals or groups of a respective species within a river or section and second by methods and approaches utilized by population monitoring (see the Technical Guideline for Sturgeon Population Monitoring, Neuburg, et al., 2024). Confirmed habitat use also must be assessed by location, timing, frequency, and spatial extension (pattern), included in GIS and put into relation to the results of the identification of potential habitat types and their verification for further adaptation and refinement of habitat- and population monitoring objectives and procedures.

4.4.1 Molecular genetics and environmental DNA (eDNA)

Applying genetic techniques allows a researcher to understand the presence and/or origin of a species in a given area (e.g. native, non-native, hybrid, or aquaculture origin), the movements of sturgeons on multiple spatial scales and through evolutionary time. Also, for some sturgeons, multiple spawning populations may exist within a single river system, leading to different (maternal) lineages utilizing different spawning sites. Population differentiation can result from differences in timing or geographic location of spawning and can occur in the absence of any physical barriers separating populations. Genetic methodologies and technologies may thus be used to assign individuals to certain natal rivers in marine catchments, where sturgeon from different spawning populations may aggregate. A comprehensive overview on genetic techniques is provided by the Technical Guideline for EX SITU Conservation Measures in Sturgeon (Gessner et al., 2024).

Environmental DNA (eDNA) is a method to detect the presence of species within a waterbody and is useful for narrowing down its presence to specific sections and during specific seasons. It is less invasive and often less expensive than traditional sampling methods that require individuals to be captured, which always includes handling stress (Pfleger et al., 2016). The technique is based on species continuously shedding DNA into their environment and has also proven to be effective for sturgeon. A comprehensive description, including instructions for taking tissue samples and water samples for eDNA analysis, can be found in the Technical Guideline for Sturgeon Population Monitoring (Neuburg, et al., 2024). Required resources and effort depend decisively on the scope of the study and the specific genetic methods applied.

Pros and cons

- Molecular genetic techniques enable precise identification of sturgeon species and their presence in a given area, also distinguishing between native, non-native, hybrid, or aquaculture origins.
- Provides insights into sturgeon movements on multiple spatial scales and throughout evolutionary time.

- Allows for the identification of multiple spawning populations within a single river system.
- Enables the assignment of individuals to specific natal rivers in marine catchments.
- Implementation of genetic techniques can be cost-intensive, requiring specialized equipment and expertise.
- Obtaining representative samples may be challenging, especially in lower river reaches and estuaries.
- Sampling eDNA will not yield information on population parameters or spawning and does not identify life stages or quantity of the species. It only confirms the presence or absence of the species.

4.4.2 Tracking, telemetry, and observation

Observation, tracking, the documentation of habitat use and comprehending the presence and movements of sturgeon within their habitat, can be key to identifying or confirming habitat types within the system, understanding how they function and they are ecologically interconnected (Lucas & Baras 2000; Fox et al., 2000; Cooke et al., 2013; Nelson et al., 2013; Acolas et al., 2017; Honţ et al., 2018).

Several relevant methods and technologies are used for tracking and observing fish in their underwater habitat and are described below. Comprehensive overviews can be found in Lucas & Baras (2000), Cooke et al., (2013) and Nelson et al., (2013). These technologies vary in their capabilities, ranging from simple visual observation to sophisticated remote sensing and imaging methods. In the end, the respective habitat monitoring team will decide on the most appropriate methodologies and equipment, depending on the available skill set, previous habitat research in the system, the available resources, and the respective research questions. The application of methods and technologies will also decisively depend on the determined objectives of the respective study, like the use of static (receivers and receiver arrays) or mobile detection of tagged individuals for the documentation of large- or small-scale movements (triangulation), also with regard to specific habitat use (e.g. spawning migration, negotiation of fish passes/function controls).

Tracking of sturgeon in their natural environment is achieved by using electronic tags of several different categories. Tags that transmit real-time data on the fish (telemetry tags), ones that store the data for later download, or bulk upload (archival tags). Not all electronic tags require recapture of the fish to recover generated data.

In this context, the "European Tracking Network" (ETN) aims at acquiring knowledge on the movements, habitat use, and survival of aquatic animals using telemetry, in an effort to inform the stewardship and sustainable management of aquatic life. The ETN strives especially for the compatibility of equipment, protocols, and software for and between different manufacturers (https://europeantrackingnetwork.org/en).

Tracking and observing sturgeon presence and movement only makes sense with regard to habitat monitoring if one can interpret such presence, aggregation, and movement by well-founded hypothesis on, or better yet, actual documentation of the associated habitat use. This can be achieved by means of preliminary maps of the system showing potential habitat types identified by information and data research (data mining), habitat modelling, field surveys (habitat verification), and by sufficient accuracy of locational telemetry data as well as by the documentation of actual habitat use in population monitoring (confirmation of habitat types by e.g. spawning, presence of early life-stages, feeding sturgeon).

4.4.2.1 Telemetry

Telemetry systems comprising transmitters and receivers are available as radio or acoustic data transmission mode. Acoustic tags can transmit identification codes and sensor data to receiving hydrophones over limited ranges.

Figure 12: *Acoustic tag (© WWF-Bulgaria).*

Radio tags and their waves transmit well through air and freshwater, yet poorly in hard- or seawater and at greater depths. Combined acoustic and radio tags (CART) allow the use of both acoustic and radio receivers for either environment.

Telemetry transmitters can be equipped with different sensors for temperature, depth, salinity, movement, or predation for example. The data are either emitted constantly or are stored until contact with a receiver is established. A special form of archival tags is the Data Storage Tag (DST) that also archives the environmental data but requires the recapture of the fish and the dismounting of the tag.

The use of archival tags, which may store data from various sensors, is advised, when fish move outside the range of the detection areas of receiver arrays (Erickson et al., 2011). The information recorded by archival tags may for example include pressure (depth), temperature, and light level, which allows for the estimation of location based on sea surface temperature and the time of sunrise and sunset.

Figure 13: *Testing of acoustic tag with a mobile receiver before the release of a tagged sturgeon (© WWF-Bulgaria).*

Other sensors may be electromyograms (EMG) recording muscle contractions, accelerometers, and tilt sensors that provide data on movement and orientation.

Data from archival tags can be recovered through subsequent recapture, data download, or by upload of archived data to satellites after the pre-programmed release from the tagged animal and ascension of the tag to the surface.

In all cases, telemetry receivers require strategic placement to ensure that the range of the tags allows for the clear identification of the signal and the download of large data sets for archival telemetry tags. The successful placement of receiver and receiver arrays depends on the potential movements and range of the tagged individual(s) or groups as well as on the capabilities of the electronic equipment and the predominant environmental conditions (e.g. natural noises, sediment transport, biological production (algae), interfering radio/acoustic sources in the vicinity).

Figure 14: *Hydrophones to be deployed as a receiver array ready for setup on the shore below the Freudenau hydropower station (© IHG/BOKU).*

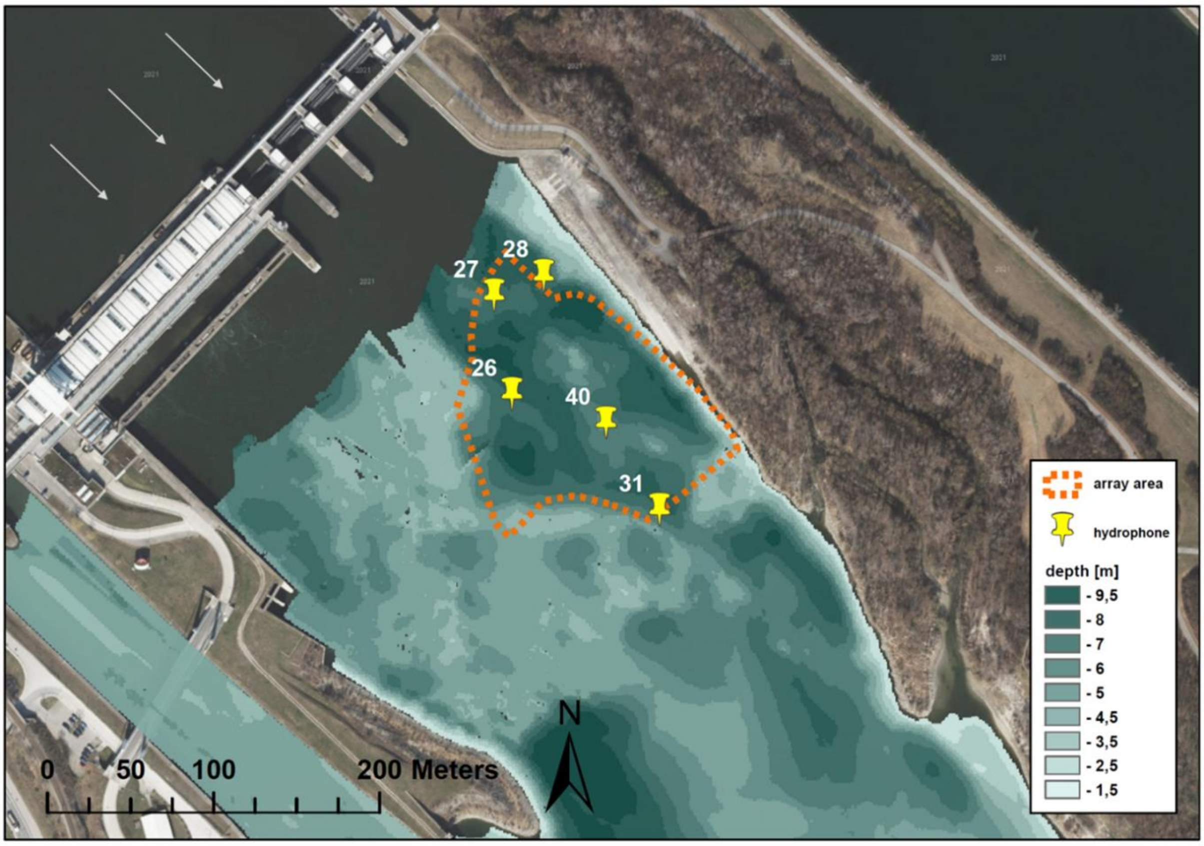

Figure 15: *Area covered by receiver array for fine tracking of sterlet (A. ruthenus) below the Freudenau hydropower station on the Danube River (© IHG/BOKU).*

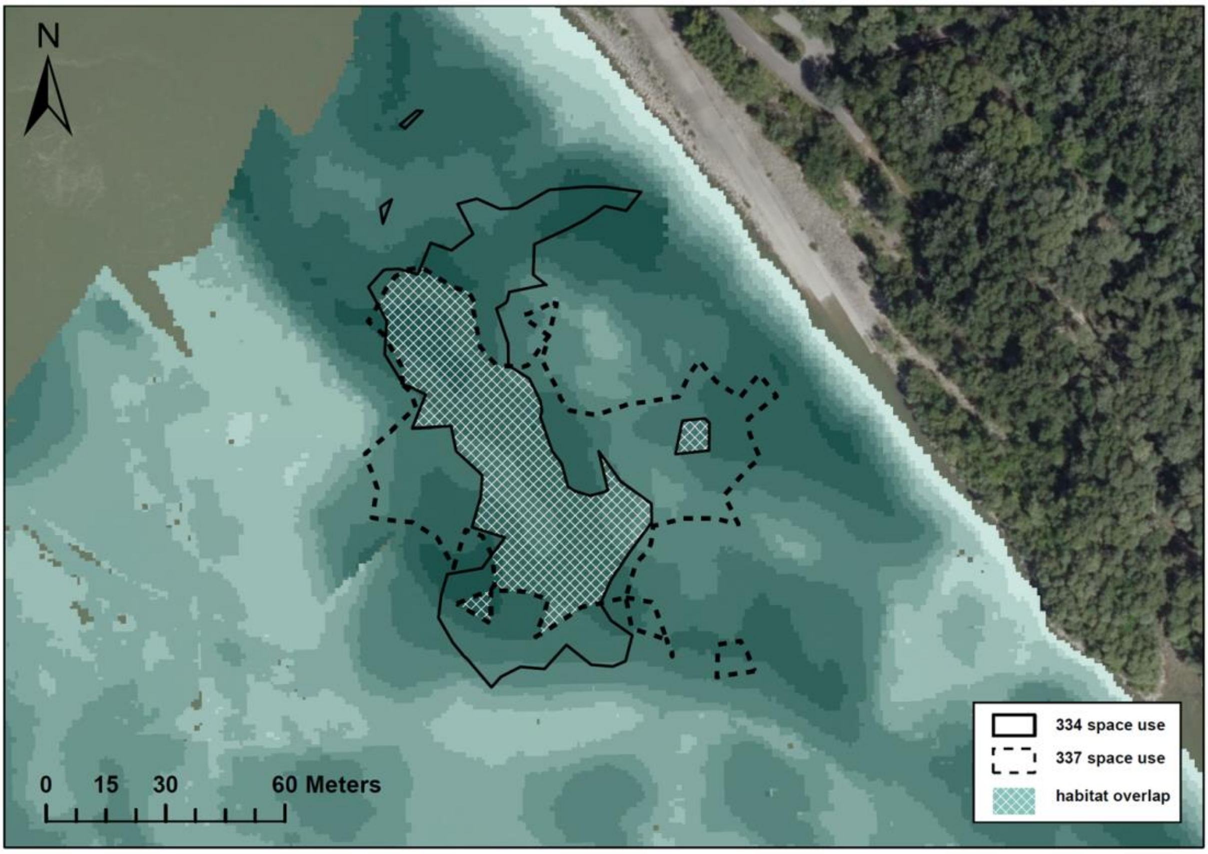

Figure 16: *Area utilized by two sterlet (A. ruthenus) individuals (Nos. 334 and 337) and habitat overlap during spawning season below the Freudenau hydropower station on the Danube River (© IHG/BOKU, Popp 2022).*

The alternative to receiver dependent transmitters are satellite tags that either pop up after a predetermined time after release or connect to satellites whenever the tag is in contact with the air (pop-up archival tag). The main disadvantage of these tags is their size and the cost, which can reach several thousand Euros per unit, and cases of premature detachment have been reported as well (Musyl et al., 2011).

With regards to habitat monitoring, the presence of the fish must be put into relation to the habitat, the habitat type and habitat use (status of the individual). This can be achieved on the basis of listings or maps of known current and potential habitat types, in combination with knowledge of the animal status (e.g. developmental stage, sex, and reproductive status, see *ex situ* guideline for determination of gonadal status).

The direct **observation** of sturgeon in their natural environment is a challenging task. Technologies for direct observation may be grouped into the two main categories: light-based and sonic-based imaging procedures.

4.4.2.2 Marking and tagging

The marking and tagging of sturgeon is useful for the identification of habitat and the documentation of movement in the case of subsequent catches.

A comprehensive description is given in the Technical Guideline for EX SITU Conservation Measures in Sturgeon (Gessner et al., 2024).

Figure 17: *External tag on juvenile Russian sturgeon (A. gueldenstaedtii) (©WWF-Bulgaria).*

4.4.2.3 Light-based underwater photography and videography

This allows for the direct observation of fish. The possibilities for underwater photo- and videography were mainly improved by the development and use of small submersible Remote Operated Vehicles (ROVs) and technological advances in low-light digital and video cameras. Such technologies are also useful for the ground truthing of substrate conditions in the course of habitat verification.

4.4.2.4 Hydroacoustic techniques

Side-scan sonar (SSS) can be used for both sturgeon habitat (Kaeser & Litts 2010; Hook 2011; Kaeser et al., 2012; Litts & Kaeser 2016; Walker & Alford 2016) and population monitoring (Hughes et al., 2018; Kazyak et al., 2020; Flowers & Hightower 2013; Flowers & Hightower 2015; Fund et al., 2016; Andrews et al., 2020; Vine et al., 2019; Brown 2020). It allows for the identification of large sturgeon and individual fish under certain conditions, as well as relatively quick benthic mapping of river sections (see also 4.3.2.8. Substrate assessment by Side-Scan Sonar).

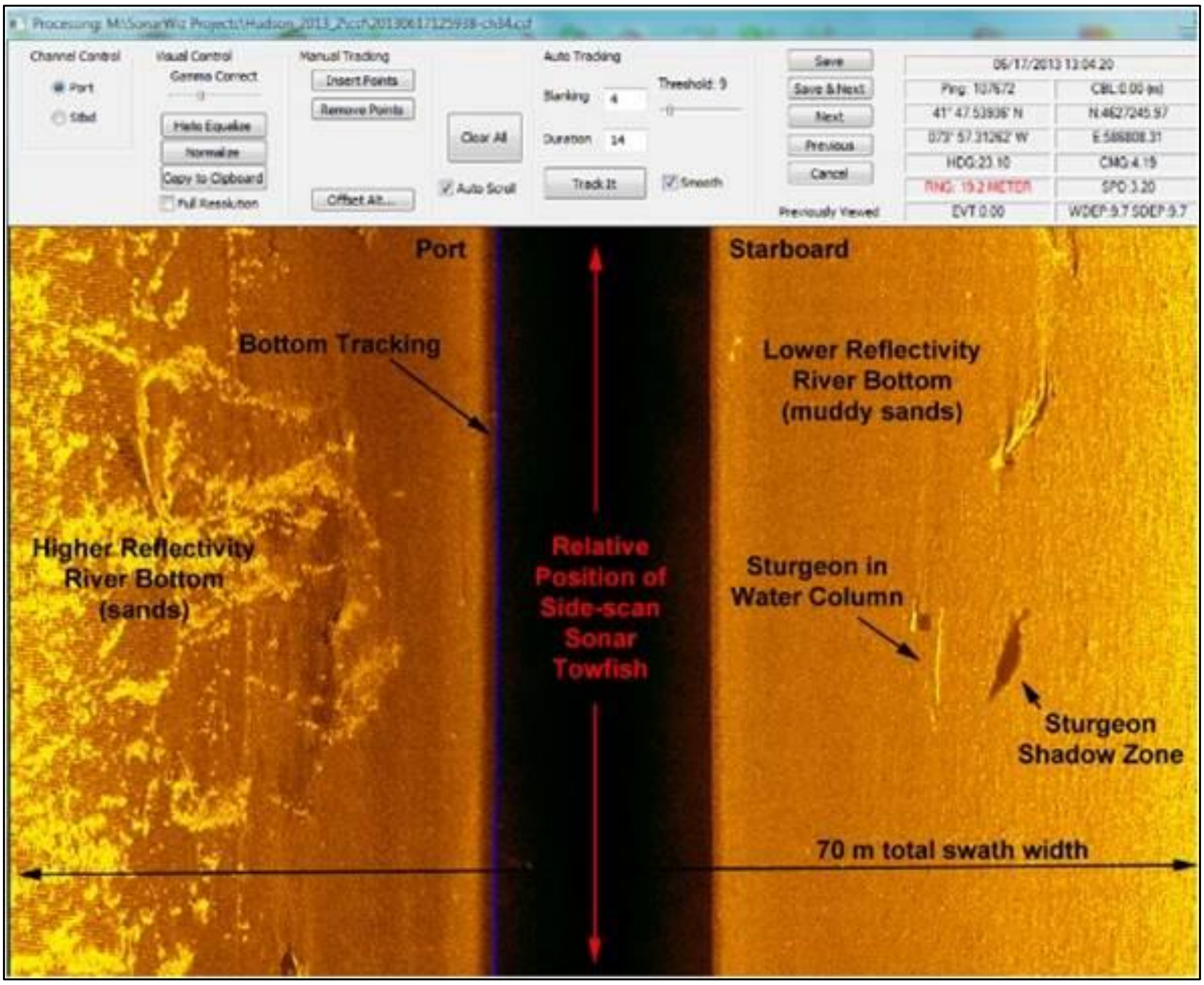

Figure 18: *Image from SSS demonstrating the potential for the identification of bottom substrates and fish (© Dewayne Fox).*

This method has been developed and proven effective for the mobile and static assessments of fish populations in both freshwater and marine environments. Mobile assessments are generally conducted from a boat while travelling along predetermined transects of the water body and sample both fish presence and bottom characteristics. Sampled fish produce characteristic acoustic signals which can be processed using specialized software to produce estimates of fish density, abundance, behavior, and size distribution.

Low frequency sonars have been used for continental shelf wide surveys while **high frequency sonars** are used for more detailed surveys of smaller areas. **Synthetic aperture sonars** (SAS) combine a number of acoustic pings to form an image with much higher resolution than conventional sonars. This technology has become commercially viable, and the technique is well suited for towed or remotely operated underwater vehicles. One must be aware of the fact, however, that all acoustic systems have sampling limitations with respect to their ability to resolve targets very close to boundaries like the bottom of a water body or with their spatial resolution which requires thorough planning of surveys to map an area of interest.

Fish finders, such as those used by recreational anglers, may also be useful in sturgeon research on habitat and habitat use, as it will facilitate the detection of sturgeon under certain conditions. For sturgeon research, a fish finder with multiple zoom settings, bottom lock, and split-screen option is recommended (Nelson et al 2013).

The main advantages of **split-beam systems** over other hydroacoustic techniques are improvements in location within the acoustic beam and in minimized susceptibility to ambient noise. Because of identical levels of bias in angular resolution, the split-beam system can locate fish within the beam with greater resolution than single-beam, dual-beam, or side-scan systems. It is therefore often used for counting migrating fish at fixed sites traveling upstream in large rivers. However, species identification using this technique is very limited.

An acoustic (dual-frequency identification sonar) camera, which is a high-definition imaging sonar that provides near-video-quality images, can also be used to count upstream migrants. In addition, this method generates video files which clearly show body shape and swimming behavior of individual fish, when used at a range of 5 – 10 m. Split-beam gear may provide more accurate information about fish location, but acoustic camera data are much easier to interpret, including the ability for on-screen measurement of fish lengths.

Synchronized multi-method approaches are possible to gather a maximum of habitat- and habitat use related data within one survey. Intensive reach-scale hydroacoustic mapping using, for example, a suite of multi-beam bathymetry, may be combined with acoustic doppler current profiler (ADCP) for measuring current speeds, high-resolution side-scan sonar for documenting substrate types, acoustic camera imagery for detection of large fish, amongst others, and intensive tracking of individuals and groups. Such an approach would potentially provide measures of habitat availability and selection variables at sub-meter to bedform scales, corresponding with the observed scale at which fish occupy and use these habitat types. The required effort, cost, and thus funding, depend decisively on the scope and scale of the study, as well as on the respective brands and devices.

Pros and cons

- Direct observation and tracking provides highly accurate data on sturgeon movements and habitat use.
- Researchers may also gain a better understanding of sturgeon behavior and interactions with their habitat.
- Tagging and tracking may allow for continuous monitoring of individual sturgeon and groups over extended periods of time, providing even more information than one-time windows of observation opportunity (e.g. bycatch).

- Apart from data providing temporal and spatial information on habitat types and habitat use, critical information such as maximum ranges, migration bottlenecks, and preferred migratory routes may also be revealed.
- The handling and tagging of sturgeon may cause stress and potential injury to the fish, as well as potentially distort the results of research by artefacts.
- Location error must be determined for each telemetric technique, as this will accurately facilitate habitat use, especially when a habitat type is limited in the watercourse.
- This approach is also very resource intensive, requiring specialized equipment, skilled personnel, significant working time, and funding. Due to drops in cost for tags and transmitters in recent decades, increased numbers of individuals and larger groups of tagged sturgeon could be released. However, processing and analyzing large amounts of tracking data can be complex and time-consuming, calling for additional effort in form of expertise, working, and computing time.

Required resources and effort

- Necessary permits for working and moving in the field and on water (e.g. for protected areas) and for interacting with and performing surgery on live animals.
- Photography and videography gear (submersible), hydroacoustic equipment, sonars.
- Tags (e.g. radio transmitters, acoustic tags, satellite tags).
- Receivers and readers to detect and record data from the tags.
- Land based, vessel, and aerial platforms: Boats, aircraft, or drones for tracking sturgeon in aquatic environments, ROVs for underwater observation, protected spots to place stationary receivers, vehicles for transport and towing.
- Data bank applications: To store and grade tracking data.
- Data analysis software: To process and analyze tracking data.
- Skilled personnel: Biologists, researchers, and technicians with expertise in tagging and tracking of fish and sturgeon in particular, specialists for the storage and analysis of large amounts of tracking data.
- Institutional commitment especially for long-term studies in terms of funding and successional planning.
- Working time in office and field: Working schedules of skilled personnel need to be adjusted to allow for working on such an approach.
- Funding: Sufficient financial resources for equipment, personnel, and data analysis.
- Ethical considerations: Adherence to ethical guidelines for handling and tagging of sturgeon to minimize stress and potential harm.

4.5 Recurrent and real-time monitoring measures

Recurrent monitoring activities to document and assess the functioning of habitat and habitat types depend mainly on the specific conditions and drivers in the respective system. Their implementation should not consist of standalone regular single activities but be part of a habitat monitoring program within a catchment. The main objectives of such a program would be the detection and assessment of changes in habitat and habitat function, to inform sturgeon conservation and management efforts (Vos et al., 2000).

Examples of relevant changes are:

- Changes in reproduction and recruitment documented by population monitoring.
- Changes in hydrology, discharge and hydromorphology.
- Expected/suspected changes in the impact/threat scenario.
- The determination of monitoring activities and intervals and the need for real-time monitoring should be an adaptive process. The effectiveness of the monitoring program should be evaluated regularly, also with regard to new information and changing conditions, and adjusted accordingly.

Habitat monitoring should allow for early detection of potential threats and enable prompt emergency response planning. In general, monitoring activities should be aligned with the habitat for critical life-cycle stages, such as spawning, migration, and feeding. It should consider the relevant threats or impacts that are observed or suspected. Monitoring frequency and intervals should be based on the timing of these events, also considering the seasonal variations in habitat conditions. Resource constraints such as budget, manpower, and technology must be considered, and the resulting monitoring program should primarily focus on the assessment of the impacts of the anthropogenic pressures on the functionality of key habitats. As such, monitoring schedules should make the best use of available resources.

In cases where sudden changes of conditions occur, real-time monitoring should be conducted for critical events and parameters, such as sudden changes in water quality, extreme weather events, or unexpected human impacts. Automated systems and sensors should be used to provide instant alerts in case of occurrence. Technological advances in monitoring equipment should be exploited.

4.5.1 Monitoring roadmap

Reynolds et al., (2016) provides a road map for the development of a biological monitoring program in general. They recommend the design and implementation of a monitoring framework containing four general phases, including ten separate steps as follows.

1st phase: Frame the problem

1. Define problem or question
2. Clearly state the study's objectives

3. Sketch a conceptual model of the system (components, system drivers and stressors)
4. Specify management or policy actions or confirm none are planned

2nd phase: Design the monitoring

5. Decide on the approach
   5a. Monitor to understand the system. No action (status and trends monitoring)
   5b. Monitor to decide when to act. No initial action (threshold monitoring)
   5c. Monitor to assess the outcome of actions (effectiveness monitoring)
   5d. Monitor to assess outcomes of multiple actions in explicit framework for informing the next action (adaptive management)
6. Translate the conceptual model from step 3 into quantitative form (What attributes and covariates should be measured?)
7. Design the survey, the analytical approach, and the data management system
8. Collect and manage data

3rd phase: Implement and learn

9. Analyze data and report results

4th phase: Learn and revise

10. Update models, assess, or plan and implement actions, when relevant

Document all steps and repeat steps 8 – 10.

5 Exemplary workplan

A stepwise approach for the planning and implementation of a combined habitat assessment and monitoring program is recommended.

Step 1: Getting started and identification of areas of general relevance through data and information research

- Start and design of the habitat monitoring program in close coordination and synchronization with and as an integral part of all other actors and aspects of sturgeon conservation in a given system.
- Determination of the objectives, assessments and measures of the habitat monitoring program and securing funding for its implementation.
- Establishment of resources for data storage, analysis, and mapping (GIS). Databases must be up and running before the first habitat related data are generated.
- Identification of sources for river related data.
- Identification of areas of past presence.
- Collection and analysis of information on current conditions (biotic and abiotic) such as hydromorphological characteristics, sediment composition and water quality.

- Establishment of ecological profiles of the species and populations (life-cycle) in the system, including past species ranges, abundances, and main sites, as well as times of harvest and observation.
- Identification of data deficiencies for additional assessments to fill knowledge gaps.
- Establishment of list and map of potential habitats in your system and locations and timings for assessments in the field.

Case example: The range and distribution of sterlet (*Acipenser ruthenus*) in the Upper Danube

Kinzelbach (1994) describes and discusses the occurrences of the sterlet in the Upper Danube river on the basis of historical sources, with the uppermost sighting near Ulm dated to the year 1430. The author provides an overview on a number of historical records of this sturgeon species in the Danube catchment area upstream of Passau. A synopsis of all the available and presented information, in combination with certain species traits, such as the distance of migrations, suggests that there had to formerly be an autochthonous population of considerable size of this species in the Danube between the riverine cities of Regensburg and Passau. He concludes that with the regularity and distribution of documented occurrences, it is evident, that specimens caught in the Danube upstream of Passau during the 19th century were not single accidental migrants from far away downstream stretches but were the last remnants of a vanishing population. This defines the Upper Danube as also being a sturgeon habitat and the maximum uppermost range limit of this particular species within the Danube River as being near Ulm (rkm 2,600 from its mouth in the Black Sea). These findings support conservation and restoration measures for the sterlet in the Upper Danube of Austria and Germany.

Step 2: Verification of identified habitats

- Assessing the main hydromorphological characteristics and complementary information required, selection of the variables to be assessed in the field, and methods best suited for the purpose and the budget.
- Setting up a structured workplan identifying the times of field visits, their duration, number of collaborators needed, equipment to be employed, samples to be taken, measurements to be carried out, purchasing consumables and storage materials, ensuring necessary infrastructure is available, securing accommodation for the trips, and development of the respective distinct assessment protocols.
- Application of permits for accessing the river, running the sampling vehicles, sampling, and translocation of samples.
- Testing equipment and becoming acquainted with the methods to be applied, establishing the documentation to be carried out in the field, securing the function of the technical equipment, training of personnel on safety measures, and adaptation of the monitoring protocols.

- Implementation of field trips for sampling and measuring, verification and revision of the monitoring protocols.
- Transfer of samples, transcription of data, storage of protocols, and analysis of samples.
- Data processing and storage.
- Data aggregation and analysis.
- Quantification of verified habitats in the system and their documentation in GIS.

Case example: A first assessment of sturgeon spawning grounds in the Odra River tributaries

Gessner & Bartel (2000) conducted an assessment of spawning habitat within the Odra River catchment as part of a feasibility study for the re-establishment of sturgeon presence in its previous range in German and Polish waters. As spawning habitat for sturgeons is considered to be of major importance for the successful restoration and subsequent reproduction, it was considered a priority. Habitat requirements were identified based on published information on sturgeon reproduction, historical catch data, and early life history. Potential spawning habitats were determined in a stepwise approach (Figure 19). For the identified historic spawning sites, recent data on migration obstacles and water pollution were evaluated, thus excluding non-accessible or adversely affected sites. Data were gathered on the dynamic of the discharge, water quality, longitudinal profiles and cross-sections of the river, as well as substrate composition. Five river stretches in the Drawa River comprising approximately 15,000 m^2 were verified as being potentially suitable for sturgeon spawning. Assuming an average fertility of 1 million eggs per female and a maximum density of 3,500 eggs/m^2, the spawning-site surface required for an average female would be comprised of approximately 350 m^2. Thus, the Drawa River could provide a spawning habitat for approximately 50 females.

Step 3: Confirmation of habitat utilization

- Confirmation in a first step by documenting sturgeon presence in a respective system, river and/or section by tracking, observation or eDNA (population monitoring).
- Documentation of actual results of habitat utilization like embryos, larvae, juveniles, content of digestive tract, growth (population monitoring).
- Determination and documentation (GIS) of extent, behavioral aspects, and preferences for specific habitat utilization like spawning, nursery, and wintering.

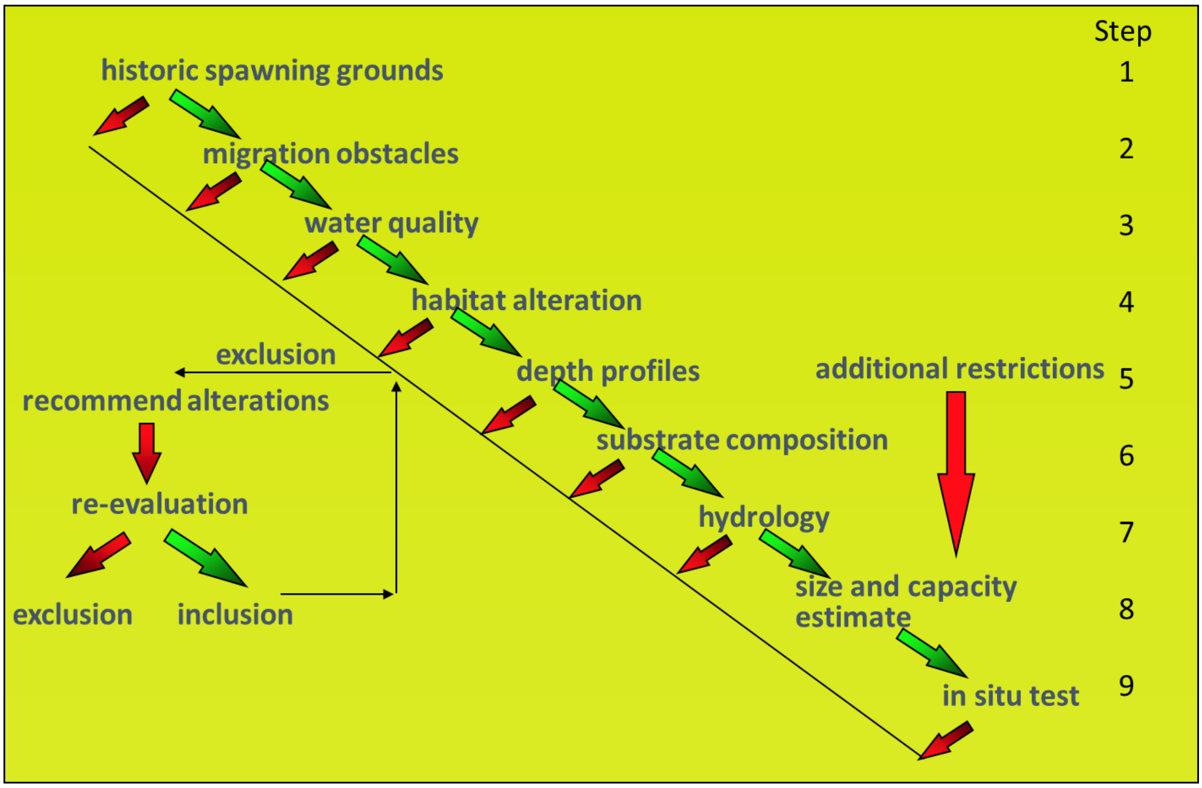

Figure 19: *Decision tree including subsequent steps for the evaluation of habitat suitability; red arrows relate to mismatch with criteria, green arrows indicate that criterion is met; in case of not meeting the criterion, the site is excluded from further assessment while countermeasures are proposed and tested for effectiveness (from Gessner & Bartel 2000, amended).*

Case example: Tracking the spawning migration and confirming spawning habitat and conditions for Gulf Sturgeon (*Acipenser oxyrinchus desotoi*) in the Southeastern U.S.

Fox et al., (2000) used a combination of ultrasonic- and radiotelemetry to monitor the movements of 35 adult Gulf sturgeons on their spawning migration in a marine-riverine system in the Southeastern U.S. during the spring of 1996 and 1997. The histological analysis of gonadal biopsies delivered the sex and reproductive status of each tagged and tracked individual. Tracking results and egg sampling provided locations and confirmation of Gulf sturgeon spawning sites as well as additional insight on the significance of sex and reproductive status for migratory behavior. Fertilized sturgeon eggs were able to be collected in six locations characterized by hard bottom substrate, steep banks, and relatively high flows (compared to average flow in the system).

Ripe sturgeon were found to occupy spawning areas from late March through early May. Ripe fish of both sexes entered the river significantly earlier and at a lower water temperature and migrated further upstream than nonripe fish did. Males entered the river at a lower water temperature than females. Results from histology and telemetry support the hypothesis that male Gulf sturgeon may spawn annually, whereas females require more than 1 year between spawning events. Upper river hard bottom areas were found to be important for the successful spawning of Gulf sturgeon, leading to the recommendation to protect such sites and actual known spawning habitat against habitat loss or degradation.

Step 4: Recurring and real-time monitoring measures

- Identification of system-specific habitat threats and impairments of habitat functionality and the parameters and variables to be monitored for the documentation of ongoing functionality.
- Determine necessary intervals for recurring monitoring assessments and samplings.
- Conduct regular habitat monitoring and regularly adjust and adapt the monitoring program to changing conditions in the system.

Case example: Fish habitat assessments along the Lower Danube River as basis for monitoring in the light of navigational interventions

Honţ et al., (2022) and their team from the Danube Delta National Institute for Research and Development (DDNI) in Tulcea, Romania conducted habitat assessments for sturgeon and other fish species in the Lower Danube between rkm 864 and rkm 375. This was done in the light of planned interventions to improve navigational conditions especially during low water level periods. Data and information from previous habitat assessments in other parts of the Bulgarian-Romanian Lower Danube River were used to identify new potential sturgeon habitat types for spawning, feeding (respectively nursery – feeding habitat for YoY sturgeon), and wintering in the study area.

To focus the efforts of the field surveys, bathymetry data were analyzed in advance during a desk study using specialized software to identify potential wintering sites and vertical clay banks (as potential spawning habitat for *A. stellatus*). Sediment data were also analyzed to identify gravel bottom as potential substrate for beluga (*Huso huso*) spawning sites. Potential sturgeon habitat types were identified by finding similarities (bottom substrate and fauna, water velocities, species captured) to habitat types previously studied in other parts of the river. Two fieldwork trips were conducted in 2017 and 2018 and 21 new potential habitat sites were identified, including 15 potential spawning sites for sturgeon (stony shores with gravel bottoms and boulders, steep riverbanks with clay sills), two sites for YOY feeding with specific sandy/muddy substrate with invertebrate fauna, and four potential wintering sites (deep areas with weak flow velocities) (Table 16).

Table 16: *Location of potential sturgeon habitat types in the Bulgarian-Romanian part of the Danube River between rkm 864 and rkm 375 as identified by field surveys (from Honţ et al., (2022), modified)*

No.	rkm	Habitat type	Perimeter [km]	Area [km2]	Observation/Rationale
1	843 – 841	spawning	4.57	0.39	Left bank, gravel substrate
2	831 – 824	spawning	14.6	1.5	Right bank, rocky banks, and gravel substrate
3	786 – 785	spawning	2.43	0.14	Left bank, vertical clay banks
4	778 – 776	spawning	5.70	0.45	Right bank, rocky banks, and gravel substrate
5	775 – 771	spawning	9.27	0.74	Right bank, rocky banks, and gravel substrate
6	770 – 769	spawning	2.30	0.14	Left bank, vertical clay banks
7	762 – 761	spawning	2.68	0.16	Left bank, vertical clay banks
8	678 – 677	wintering	0.145	0.00128	Left bank, 8 m deep, close to main channel
9	678 – 673	spawning	8.50	1	Right bank, bottom samples reveal rocky/gravel substrate and large soft stones
10	662 – 651	spawning	23.4	2.26	Right bank, bottom samples reveal rocky/gravel substrate and large soft stones
11	649 – 640	spawning	19.9	2.25	Right bank, rocky banks, and gravel substrate
12	626 – 624	YoY feeding	3.90	0.56	Left bank, bottom samples with worms
13	603 – 602	spawning	3.62	0.23	Right bank, rocky/gravel substrate
14	596 – 593	spawning	6.10	0.44	Right bank, rocky banks, and gravel substrate
15	586 – 585	wintering	0.363	0.0082	Left bank, deep water/pit
16	579 – 577	spawning	5.41	0.46	Right bank, gravel substrate

17	572 – 571	YoY feeding	1.12	0.0313	Left bank, at the island's tail, bottom sample revealed worms
18	570 – 569	spawning	2.00	0.1	Left bank, vertical clay banks
19	524 – 523	wintering	1.50	0.1	Left bank, deep water/pit
20	414 – 412	spawning	4.35	0.29	Left bank, gravel substrate
21	409	wintering	n.a.	n.a.	Left bank, deep pit at the tail of the island

6 Messages for decision makers

- Support and facilitate the design of a coherent monitoring approach in your country and catchment. Set monitoring priorities according to a national sturgeon action plan or transnational conservation strategy following e.g. the frame of the Pan-European Action Plan for Sturgeons (PANEUAP 2018).
- Include sturgeon habitat monitoring into other existing monitoring approaches (e.g. in the EU into the frame of the Water Framework Directive (2000/60/EC))
- Ensure that future projects subject to funding from public or national support follow best practice approaches as outlined in this document in the sense that the research question is clearly articulated, and the method appropriately chosen to answer this question.
- Ensure that results of monitoring actions are shared transparently with the public and other relevant stakeholders for sturgeon conservation, including different national research institutions, NGOs or other stakeholders from the navigation, fisheries, nature protection, or water management sector.
- Ensure regular data exchange transnationally with neighboring countries sharing the same sturgeon populations. For example, information on whether a key habitat such as a spawning place is available in an upstream country informs management decisions of downstream countries or vice versa.
- Include knowledge on sturgeon habitats into national and regional River Basin Management Plans, Navigation Plans or other strategically relevant documents.
- National governments and international organisations alike need to ensure necessary funding to support the implementation of monitoring actions. The combination of national sources with specific funding instruments (e.g. LIFE, Horizon, European Regional Development Fund (ERDF), the Cohesion Fund (CF), and the European Maritime, Fisheries and Aquaculture Fund (EMFAF) in the EU) may provide good opportunities for beginning implementation,

however, in the longer term, such costs should be integrated into national budgets.

- In the short and medium terms, relevant ministries (e.g. Environment, Agriculture, Fisheries, Development) of sturgeon range states should therefore ensure that monitoring of threatened migratory fish species is prioritized as critical habitats are under threat and immediate action is required. For example, the inclusion of such a monitoring in so-called Priority Action Frameworks (PAFs) is of paramount importance in the EU, as references to the PAFs is the enabling condition for accessing funding from EU instruments.

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