ZERO BRINE

D9.1 Report on environmental impacts from brine discharge

October 2020

Draft

The ZERO BRINE project (www.zerobrine.eu) has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 730390.

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Deliverable lead	University of Aberdeen (ABDN)
Author(s)	Sergio Carlos Garcia Gomez, Taxonomist
Contact	fkuepper@abd.ac.uk eleni.avramidi@abdn.ac.uk
Reviewer	Frithjof Kuepper
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Name of researcher(s) with roles	Eleni Avramidi, Honorary Research Fellow, School of Biological Sciences (ABDN) Prof. Frithjof Kuepper, Chair in Marine Biodiversity, School of Biological Sciences (ABDN)

[1] **R**=Document, report; **DEM**=Demonstrator, pilot, prototype; **DEC**=website, patent fillings, videos, etc.; **OTHER**=other

[2] **PU**=Public, **CO**=Confidential, only for members of the consortium (including the Commission Services), **CI**=Classified

Revision no	Date	Description	Author(s)
0.1	30 May 2020	First draft	Prof. Frithjof Kuepper (ABDN) Eleni Avramidi (ABDN) Sergio Carlos Garcia Gomez, Taxonomist
0.2	14 Oct. 2020	Second draft The chapters “Executive summary” and “Conclusions” have been included. The comments of the internal review have been incorporated.	Prof. Frithjof Kuepper (ABDN) Eleni Avramidi (ABDN) Sergio Carlos Garcia Gomez, Taxonomist

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Executive summary

Seawater desalination plants may potentially affect the aquatic biological communities in the coastal environment due to the brine discharge. "Benthic macroinvertebrates" is the key biological indicator examined in this investigation for the assessment of the influence of brine discharge on benthic fauna, supported by data about macroalgal indicators and physicochemical and hydromorphological conditions. Benthic macroinvertebrates have an important role in sediment processes and predator–prey relationships and they usually have well-defined responses to environmental changes, especially those stressors that influence the sediment structure and its chemistry and quality. They are officially included among biological quality elements in the European umbrella regulations for water systems, namely the Water Framework Directive 2000/60/EC (WFD) and the Marine Strategy Framework Directive 2008/56/EC (MSFD).

The investigated area includes three sites, one in the vicinity of the EVIDES DWP1 in the Britanniehaven area, one in the vicinity of EVIDES DWP2 in the Hartelkanaal area, and one in Hartelkanaal in the Elbeweg area that is designated as reference site. In total, 4 seasonal sampling surveys were scheduled, namely in September 2019, January 2020, July 2020 and April 2021. The survey planned for April 2020 was cancelled due to the Covid-19 outbreak and postponed for April 2021. A total of 6 sampling stations have been established for benthic macroinvertebrates analysis. Biological quality descriptors (abundance A, species richness S, and Shannon's diversity H), biological quality indices (AMBI, BOPA), and statistics analysis tools will be applied after the completion of the surveys to objectively assess the effect of brine release on benthic fauna and the ecological status of the investigated areas. The approach will be in line with the guidelines of the WFD and the MSFD.

Contents

Abbreviations

AMBI	AZTI Marine Biotic Index
BOPA	Benthic opportunistic polychaeta amphipoda index
DWP	Demineralized Water Plant
EQR	Ecological Quality Ratio
ES	Ecological Status
ICPR	International Commission for the Protection of the Rhine
MSFD	Marine Strategy Framework Directive (MSFD)
POC	Particulate Organic Carbon
RAP	Rhine Action Programme
RBD	River Basin District
RBMP	River Basin Management Plan
SAC	Special Areas of Conservation of Natura 2000 Network
SPA	Special Protection Areas of Natura 2000 Network
SWB	Surface Water Body
TBT	Tri-butyl tin
WFD	Water Framework Directive 2000/60/EC

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1 Overview of the project

The ZERO BRINE project aims to facilitate the implementation of the Circular Economy package and the SPIRE roadmap in various process industries by developing necessary concepts, technological solutions and business models to redesign the value and supply chains of minerals and water while dealing with present organic compounds in a way that allows their subsequent recovery.

These resources will be recovered from saline impaired effluents (brines) generated by the process industry while eliminating wastewater discharges and minimizing the environmental impacts of brines from industrial operations (ZERO BRINE). ZERO BRINE brings together and integrates several existing and innovative technologies to recover products of high quality and sufficient purity to represent good market value.

A large-scale demonstration plant for the treatment of part of the brine effluent will be tested in the Energy Port and Petrochemical cluster of Rotterdam Port by using the waste heat from one of the factories in the port. The quality of the recovered products will be aimed to meet local market specifications. Additionally, three large-scale pilot plants will be developed in other process industries in Poland, Spain, and Turkey, providing the potential for immediate replication and uptake of the project results after its successful completion.

2 Objectives

The scope of the Deliverable 9.1 is to evaluate the results of the benthic macroinvertebrates analysis conducted in the context of the sub-task 9.2.2 "Assessment of environmental impacts associated with brine discharge" and assess the environmental quality status on the area of Britanniehaven due to the brine discharge and related activities.

Given the low operational capacity of the Pilot Plant, resulting in less than 1% of the total EVIDES brine discharge, it is not feasible to observe significant changes in the benthic communities and the ecological quality of the environment as a result of the implementation of the Zero Brine technology. In addition to that, the Britanniehaven area is a heavily industrialized district of the Rotterdam port and also a dead-end waterway which means that the environmental quality of the area is affected by several major sources and any effects related solely to the operation of EVIDES DWP cannot be detected.

Considering the aforementioned conditions, sampling surveys took place in the vicinity of the two EVIDES DWPs areas, in Britanniehaven and Hartelkanaal with a priority given to the Hartelkanaal area which is surrounded by less intense industrial activity in relation to Britanniehaven area. The aim of the surveys is to establish a baseline understanding of the environmental conditions in the area of EVIDES DWPs. This will provide an essential background/baseline information necessary for the assessment of potential environmental benefits of Zero Brine technology in the future.

Apart from the analysis of benthic macroinvertebrates, analysis of physical and chemical parameters of water, toxicity analysis of sediment, macroscopic identification of algae are performed as they are considered valuable in the assessment of the environmental quality in Britanniehaven and Hartelkanaal areas even if these were not foreseen in the original scope of work.

3 Introduction

The Port of Rotterdam is situated in the estuary of the main branch of the river Rhine at a connection of fresh and marine ecosystems. Before the Delta Project which was carried out after the storm surge of 1953 (Smits et al. 2006), the intertidal zone of the estuary consisted mostly of beaches, salt and brackish marshes, sand and mud flats, tidal creeks, immense fresh and brackish rush and reed beds and intertidal forests. In the northern part of the Rhine–Meuse estuary, many of these soft substrate ecotopes disappeared gradually with the development of the Port of Rotterdam between 1870 and 1970 (Paalvast 2002, 2012). Nowadays, the port of Rotterdam is a highly engineered estuarine environment and the only completely open access into the river Rhine is through the Rotterdam Waterway (Nieuwe Waterweg), the main navigation channel of the Rotterdam Port (Fig. 1).

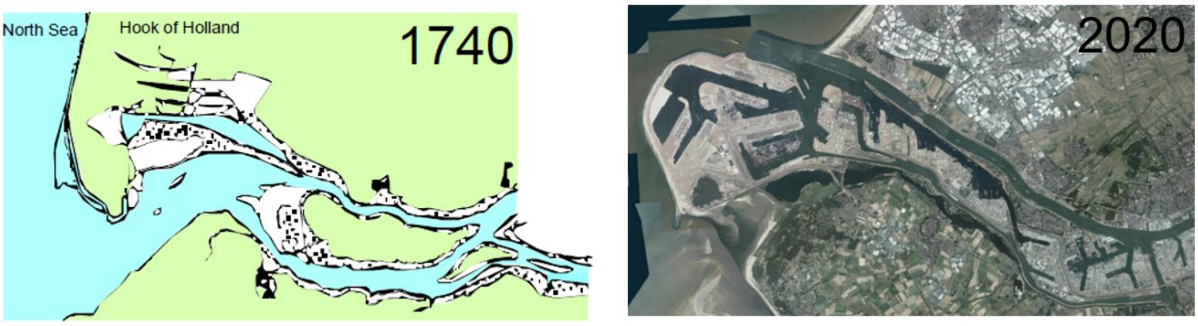


Figure 1 Topography of the Rotterdam waterway in 1740 and 2020

The total area of the Port of Rotterdam is 12,713 ha of which the land area is 7,903 ha and the water area is 4,810 ha. The total length is 42 km and the maximum water depth relevant to New Amsterdam Level is 24 m. In 2019, the Port of Rotterdam was the Europe's largest seaport. Shipping in the Port of Rotterdam is intensive. 29,491 seagoing vessels and 85,969 inland vessels visited the port of Rotterdam in 2019 (Port of Rotterdam Authority 2019).

The Port of Rotterdam is highly industrialized (Fig. 2). The main commercial activities are aggregates (sand, gravel etc.), ship repair, marine engineering, petroleum refining and product processing, roll-on/roll-of cargo transfer, chemical industry, general manufacturing, storage and packaging, refrigerated cargo and energy production. The main types of cargo handled are dry bulk, liquid bulk (non-oil), trade vehicles, perishable goods, petroleum/oil products, roll-on/roll-off and general cargo. The port itself is not designated as Natura 2000 site, however in the immediate vicinity there are areas of the Natura 2000 network that host numerous protected species. These sites are the habitats directive sites (Directive 92/43/EEC, SAC sites) Voordelta NL4000017, Voornes Duin NL9803077, Solleveld & Kapitteldulnen NL1000016 and the birds directive sites (Directive 2009/147/EC, SPA sites) Voornes Duin NL2002017 and Voordelta NL4000017.

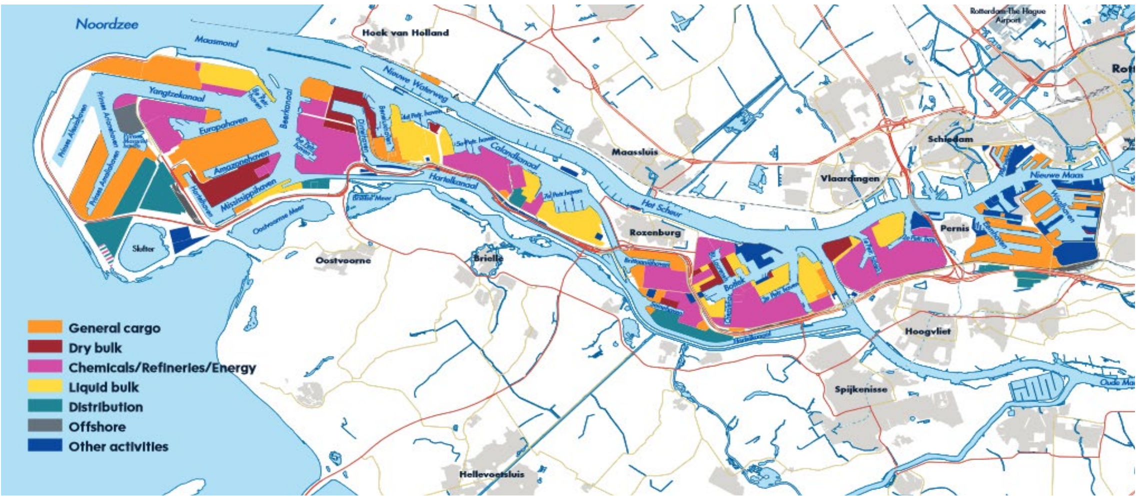


Figure 2 Distribution of activities in the Port of Rotterdam (Source: PoR)

Between 1960 and 1970 the pollution of the Port of Rotterdam was severely degrading the ecosystem, reducing biodiversity to a low number of pollution tolerant species (Wolff 1978). During more recent years, the pollution of the Rhine and of many of its tributaries was distinctly improved due to the implementation of the Rhine Action Programme (RAP) introduced by the International Commission for the Protection of the Rhine (ICPR) in 1987, a year after the Sandoz chemical accident. This is mainly due to the reduction of point source pollutant inputs of industrial and municipal origin. However, pollution originating from the surface, (diffuse) inputs of nitrogen and pollutants from agriculture, pollution due to historically polluted river sediments and substances measured in very low concentrations in water bodies (micro-pollutions) continue to be problematic. Furthermore, navigation continues to accidentally or deliberately discharge substances into the water. The ban of the anti-fouling agent tri-butyl tin (TBT), dredging and removal of the heavily polluted sediment, prevention of oil spills and the change from a bulk harbour to a container harbour also contributed to a better water and sediment quality (Anonymous 1999, 2006). Also, the European Parliament and the European Council adopted the Water Framework Directive 2000/60/EC (WFD) with the purpose to establish a framework for the protection of European waters. For artificial water bodies, like the Port of Rotterdam, WFD sets that Member States shall protect and achieve good ecological potential (GEP) and good chemical status by 2015 extended to 2021 and 2027. The responsible authority for the implementation of the WFD in Netherlands is the Ministry of Infrastructure and Water Management.

Benthic macroinvertebrates have an important role in sediment processes (e.g., enhancing the flow of nutrients and materials between the sediments and the water column, and vice versa, trough bioturbation and bioirrigation) and predator–prey relationships, and they usually have well-defined responses to environmental changes, especially those stressors that influence the sediment structure and its chemistry and quality (Quintino et al. 2006, Borja et al. 2015). For this reason, benthic macroinvertebrates are officially included among biological quality elements in the WFD for the classification of the ecological quality status / potential of surface water bodies.

4 Methods

4.1 Study area

Three sites are sampled within the framework of this study (Fig. 3): one in the vicinity of the EVIDES DWP1 in Britanniehaven area, one in the vicinity of EVIDES DWP2 in Hartelkanaal area, and one in Hartelkanaal in Elbeweg area that was designated as reference site. DWP1 operates since December 2009 and DWP2 operates since January 2018.

In Britanniehaven, the main port’s activities are general cargo and chemicals, refineries and energy industries. This site is a dead-end waterway, i.e. it has no river input and it is entirely marine. This site receives effluents from the DWP1 and treated industrial wastewater from nearby chemical plants. In the vicinity of DWP2 in Hartelkanaal area, the main port’s activities are freight distribution, chemicals/refineries/energy and liquid bulk. This site has inherent variable salinities and current directions and receives effluent from the DWP2. The main port’s activities in the reference site are chemical/refineries/energy industries and liquid bulk cargo transportation. The reference site has naturally changing salinities and current directions due to tidal influence.

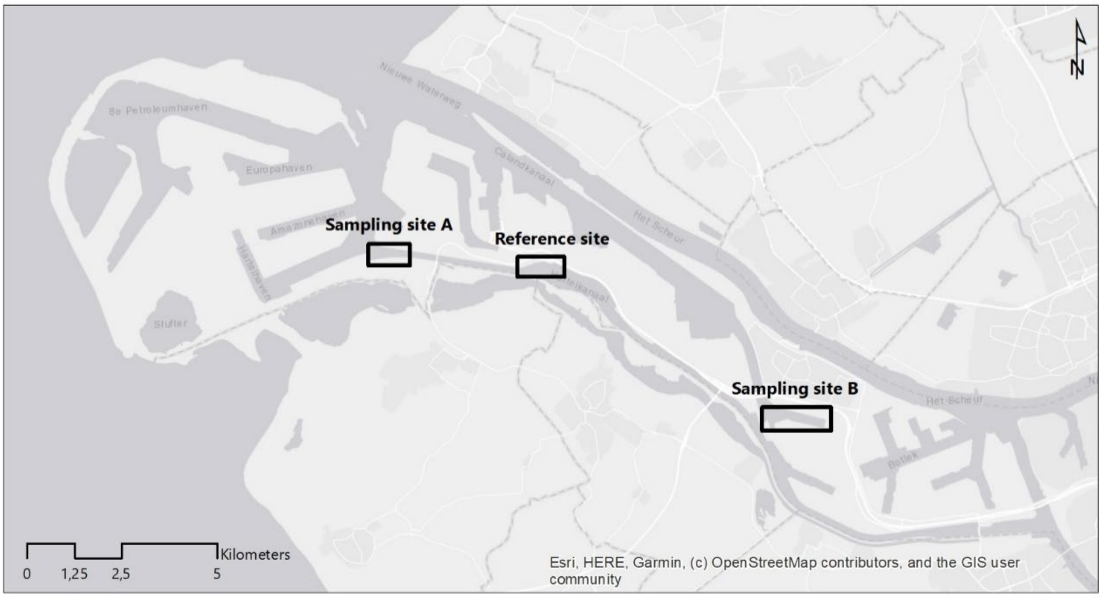


Figure 3 Sampling sites

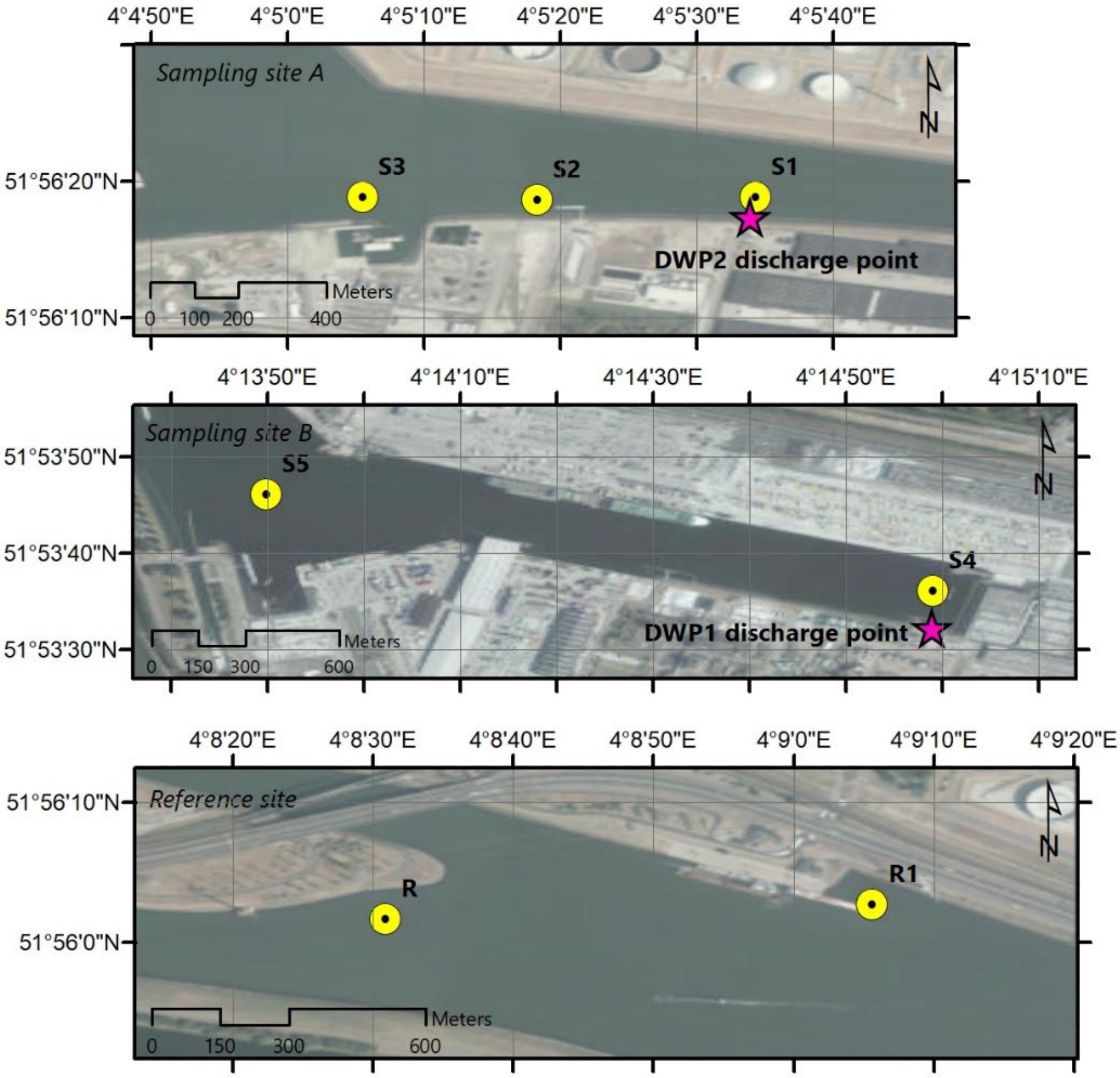
Based on the information provided in the River Basin Management Plan (RBMP) 2015 of the Rhine river basin district that was conducted in the framework of the WFD, the study area is located within the River Basin District (RBD) coded NLRN and specifically in the Surface Water Body (SWB) coded NL94_9. The type of the SWB NL94_9 is classified transitional since it is a SWB in the vicinity of river mouths which are partly saline in character as a result of their proximity to coastal waters, but which are substantially influenced by freshwater flows. The category of the SWB NL94_9 is classified artificial since it is created by human activity.

4.2 Field and laboratory work

In total, 4 seasonal sampling surveys were scheduled, namely in September 2019, January 2020, July 2020 and April 2021. The survey planned for April 2020 was cancelled due to the Covid-19 outbreak and postponed for April 2021. The sampling survey in September 2019 played a key role in the design of the subsequent surveys, as it constituted the survey that highlighted a complex ecosystem in the Rotterdam Port. For this reason, some differences in the sampling schema are observed in this survey.

A total of 6 stations (Fig. 4) were selected and established for benthic analysis. The main characteristics of the sampling stations are given in the Table 1. Three replicates were collected at each sampling station using a Van Veen grab of 2L capacity. At each replicate, the Van Veen grab collected sediment twice, and the total volume collected were 4L. The sediment samples were sieved through a 0.5 mm mesh, stained with Rose Bengal and preserved in ethanol. In the laboratory, macrobenthic invertebrates were sorted, identified to the lowest taxonomic level possible, and counted. The type of bottom sediment in each station was visually estimated.

Samples from 3 stations were used for analysis of physical and chemical parameters of water. Water samples were collected from a depth of 1 m from the water surface and were stored in appropriate bottles and analyzed in the laboratories of SYNLAB Analytics & Services B.V. (Steenhouwerstraat 15, 3194 AG Rotterdam, Netherlands) and C-MARK B.V (Munsterstraat 9, 7418 EV Deventer, Netherlands). Sediment samples from 3 stations will be used for Particulate Organic Carbon (POC) analysis, granulometric analysis, heavy metal and hydrocarbon content. Sediment samples will be collected with a Van Veen grab and stored in appropriate bottles and analyzed in the laboratory Eurofins.



Service layer credits: Source: ESRI, DigitalGlobe, GeoEye, Earthstar Geographics, CNES/Airbus DS, USDA, USGS, AeroGRID, IGN, and the GIS User Community.

Figure 4 Location of sampling stations

Table 1: Main characteristics of the sampled stations

Station	Coordinate (latitude / longitude) WGS 84	Depth (m)	Port sector	Bottom sediment	Surveys in which the stations were investigated
S1	X: 51°56'18.84"N, Y: 4°5'34.32"E	6-8 m	Hartelkanaal	Hard substrate consisting of *Crassostrea gigas* (Pacific oyster) reef	1st, 2nd, 3rd, 4th
S2	X: 51°56'18.66"N, Y: 4°5'18.34"E	6-8 m	Hartelkanaal	Mainly hard substrate consisting of *Crassostrea gigas* (Pacific oyster) reef mixed with soft substrate of mud	1st, 2nd, 3rd, 4th
S3	X: 51°56'18.84"N, Y: 4°5'5.54"E	6-8 m	Hartelkanaal	Mainly hard substrate consisting of *Crassostrea gigas* (Pacific oyster) reef mixed with soft substrate of mud	1st, 2nd, 3rd, 4th
S4	X: 51°53'36.12"N Y: 4°14'59.04"E	6-8 m	Britanniehaven	Soft substrate, silt	2nd, 3rd, 4th
S5	X: 51°53'46.08"N Y: 4°13'49.86"E	9-11 m	Britanniehaven	Soft substrate, silt	3rd, 4th
R1	X: 51°56'2.76"N, Y: 4°9'5.64"E	13m	Hartelkanaal - Dolfijnweg	Soft substrate, silt	1st
R	X: 51°56'1.72"N, Y: 4°8'30.93"E	9-11m	Hartelkanaal - Dolfijnweg	Soft substrate, silt	1st, 2nd, 3rd, 4th

5 Results and Discussion

5.1 Historical data

Physical and chemical characteristics of the water in the Port of Rotterdam are monitored within the framework of the national long-term regular monitoring program (Monitoring Waterstaatkundige Toestand des Lands, MWTL). Responsible for the implementation of the program is the Dutch Rijkswaterstaat, which is part of the Ministry of Infrastructure and the Environment. MWTL is a national environmental programme for monitoring hydrochemistral, hydrobiologal and geomorphologal parameters. The results of this program fed the Dutch RBMPs in line with the guidelines of the WFD.

Based on the information provided in the RBMP of the Rhine river basin district (2015), there are 4 monitoring stations within the SWB NL94_9, which are NL94_BEERKNMDN, NL94_NIEUWEWATERWEG, NL94_NIEUWEWATERWEG_A, NL94_MAASSS (Fig. 5). None of them coincides with the study sites.

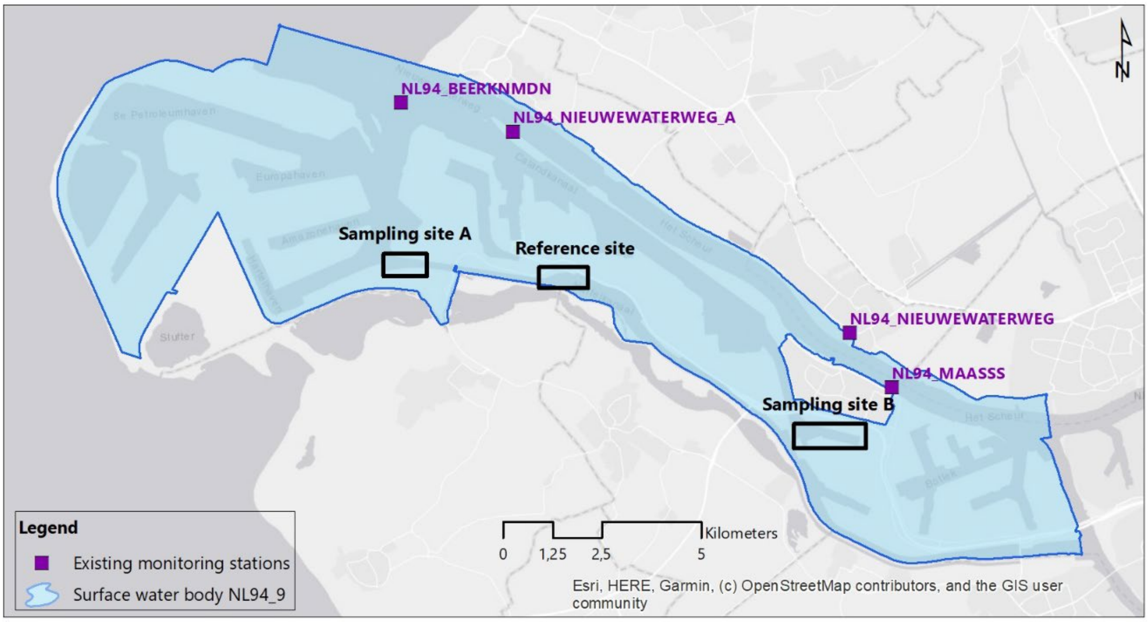


Figure 5 Existing monitoring stations in the broader area as reported in the RBMP of the Rhine river basin district (2015)

According to the monitoring results, the ecological potential of the SWB NL94_9 was characterized as "moderate" and the chemical status as "not good". Regarding the biological quality elements, phytoplankton, phytobenthos/macrophytes and macrozoobenthos were characterized as "good" ecological potential and the fish fauna with "moderate" ecological potential. Therefore, the goal of WFD for "good" ecological potential and "good" chemical status will not be achieved in 2021.

5.2 Water and sediment physical and chemical parameters

The water physicochemical parameters examined are pH, electrical conductivity, total suspended solids, ammonium ions, nitrates, nitrites, total N, phosphates, total P and sulphates. Water parameters and discharge locations are set by Rijkswaterstaat in compliance with the legal provisions.

Based on the macroinvertebrates results of the 1st and 2nd sampling survey, it was considered worthwhile to also investigate ecotoxicological parameters. For that reason, analysis of heavy metals and polycyclic hydrocarbons in the sediments of the sampling sites will be performed in the following two sampling surveys.

The values measured are presented in the Tables 2 and 3.

Table 2: Values of the water physical and chemical parameters measurements at sampling stations

	1st sampling survey		2nd sampling survey			3rd sampling survey			4th sampling survey		
Parameters	**S1**	**R**	**S1**	**S4**	**R**	**S1**	**S4**	**R**	**S1**	**S4**	**R**
pH	7.9	7.9	7.9	7.9	8.1						
EC (µS/cm)	18000	23000	8500	>13000	5600						
NH_4^+ (mg/l)	<0.2	<0.2	<0,06	0.10	0.064						
TSS	11900	15000	8153	25524	4806						
NO_2^- (mg/l)	<0.3	<0.3	0.066	0.099	0.066						
NO_3^- (mg/l)	<0.75	<0.75	12	8.8	12						
PO_4^{3-} (mg P/l)			<0.05	0.12	0.06						
N_T											
P_T (mg/l)	0.077	0.073									
SO_4^{2-}											

Table 3: Values of the sediment physical and chemical parameters measurements at sampling station

	3rd sampling survey			4th sampling survey		
Parameters	**S1**	**S4**	**R**	**S1**	**S4**	**R**
Granulometric analysis						
POC						
PAHs						
Cd						
Pb						
Cu						
Zn						
Fe						
Ar						
Cr						
Ni						
SO_4^{2-}						
PAHs						

5.3 Algae

Intertidal surveys of seaweeds (macroalgae) will be conducted at Hartelkanaal, Britanniehaven and other sites where the waterline is accessible from the shore. Immediately following each day of sampling, herbarium specimens will be prepared by mounting seaweed thalli on Bristol paper, or samples will be fixed as permanent mounts on microscope slides. They will be deposited in duplicate in the Herbarium of the University of Aberdeen and in a suitable natural history collection in the Netherlands. Fragments of all specimens will be kept in silica gel or CTAB buffer (Phillips et al. 2001), both of which conserve DNA for further molecular studies.

The presence of species will be evaluated in light of their potential significance as biological indicators of environmental quality.

5.4 Benthic macroinvertebrates

5.4.1 Community composition

In the 1st and the 2nd sampling surveys, a total of 15 benthic samples were analysed and 882 individuals, belonging to 44 species were identified. The species found and the total number of individuals per species are shown in the following table.

The biological descriptors that will be calculated when all the sampling surveys will be completed are abundance (A), species richness (S) and Shannon's diversity (H).

Table 4: Benthic macrofauna results at sampling stations in the 1st sampling survey

					FIRST SAMPLING SURVEY (09/2019)												
TAXA				**NATIVE / NON NATIVE**	**S1**	**S1**	**S1**	**S2**	**S2**	**S2**	**S3**	**S3**	**S3**	**R1**	**R1**	**R**	**TOTAL**
Annelida	Class Polychaeta	Capitellidae	*Heteromastus filiformis*	Native					1		1					1	3
		Cirratulidae	*Chaetozone gibber*	Native							2						2
			Tharyx cf killariensis	Native					16			4	5				25
		Cossuridae	*Cossura longocirrata*	Native								1					1
		Nephtyidae	*Nephtys hombergii*	Native							2		1				3
		Nereididae	*Alitta (Neanthes) cf succinea*	Native						1							1
			Nereis zonata	Native												5	5
			Websterinereis glauca	Native											1		1
		Ophelidae	*Orbinia latreillii*	Native												1	1
		Orbiniidae	*Scoloplos sp.**	Native							2	1					3
		Phyllodocidae	*Phyllodoce lineata*	Native								1	1			1	3
		Serpulidae	*Ficopomatus enigmaticus*	Non native		5	1	4	1					1		1	13
		Spionidae	*Spiophanes bombyx*	Native								1					1
		Terebellidae	*Lanice conchilega*	Native								1					1
Arthropoda	Orden Amphipoda	Corophiidae	*Corophium volutator*	Native										9	7	1	17
			Monocorophium acherosicum♀	Native	1	4			1	5	1						12
		Ischyroceridae	*Ericthonius punctatus*	Native									1				1
		Melitidae	*Melita sp.*♀*	Native												4	4
	Order Decapoda	Varunidae	*Hemigrapsus cf. takanoi*	Non native	3	1		1								4	9
		Panopeidae	*Rhithropanopeus harrisii*	Non native										2	1		3
	Order Isopoda	Anthuridae	*Cyathura carinata*	Native												1	1
	Order Sessilia	Balanidae	*Amphibalanus improvisus*	Non native		16	18	1	1	7				42	3		88
			Balanus cf crenatus	Native			6				1		1				8
Mollusca	Class Bivalvia	Cardiidae	*Cardiidae juv.*	Native							1	4	1	2		1	9
		Dreissenidae	*Dreissena polymorpha*	Non native											1		1
		Mactridae	*Spisula subtruncata*	Native								1	4			1	6
		Myidae	*Mya arenaria*	Non native										2	2		4
		Mytilidae	*Mytilus edulis*	Native	3	11	1	2	2	2	4						25
		Ostreoidea	*Crassostrea (Magallana) gigas*	Non native			2		2								4
		Pharidae	*Ensis cf leei***	Non native							1						1
		Pholadidae	*Pholas dactylus*	Native											1		1
		Tellinidae	*Tellina (Fabulina) fabula*	Native								4					4
			Tellina tenuis	Native									1				1
	Class Gasteropoda	Hydrobiidae	*Peringia (Hydrobia) ulvae*	Native										1			1
		Nassariidae	*Tritia (Nassarius) reticulata (reticulatus)*	Native								1					1
Cnidaria	Class Anthozoa	Actiniidae	*Actinia equina*	Native							6	5	4				15
Number of individuals					7	37	28	8	24	15	21	24	19	59	16	21	279
Number of species					3	5	5	4	7	4	10	11	9	7	7	11	36

Table 5: Benthic macrofauna results at sampling station in the second sampling survey

TAXA				NATIVE / NON NATIVE	S1	S1	S1	S2	S2	S2	S3	S3	S3	S4	S4	S4	R	R	R	TOTAL
				SECOND SAMPLING SURVEY (01/2020)																
Annelida	Class Polychaeta	Capitellidae	*Heteromastus filiformis*	Native						3			1							4
			*Capitella capitata*****	Native									1	22	8	2				33
		Cirratulidae	*Chaetozone gibber*	Native				2			3	1			1					7
			Chaetozone setosa	Native								2								2
			Tharyx cf killariensis	Native				12	6		13	8	2		2					43
		Hesionidae	*Oxydromus flexuosus*	Native												1				1
		Magelonidae	*Magelona filiformis*	Native							1									1
		Nephtyidae	*Nephtys hombergii*	Native				1				1	1						1	4
		Nereididae	*Alitta (Neanthes) cf succinea*	Native										7	3	3				13
			Platynereis dumerilii	Native	1	1	4	2	1									1		10
		Orbiniidae	*Scoloplos sp.**	Native				1												1
		Phyllodocidae	*Phyllodoce lineata*	Native				1	4											5
		Sabellaridae	*Laonome kroyeri*	cf*							11		7					2		20
		Serpulidae	*Ficopomatus enigmaticus*	Non native	5	1	2													8
			*Hydroides sp.**	Native	2	1	2													5
		Spionidae	*Polydora ciliata*	Native							2		1	1		1				5
			Streblospio cf shrubsolii	Native			1	18	9		21	1	1						1	52
		Terebellidae	*Lanice conchilega*	Native							1									1
Arthropoda	Orden Amphipoda	Corophiidae	*Corophium volutator*	Native						1							18	41	7	67
			Monocorophium acherosicum♀	Native		1			1											2
		Melitidae	*Melita hergensis*	Native	1	5	4	7	2	1								4		24
	Order Decapoda	Palaemonidae	*Palaemon longirostris*	Native	1															1
		Panopeidae	*Rhithropanopeus harrisii*	Non native														1		1
		Varunidae	*Hemigrapsus cf takanoi*	Non native	1															1
	Order Isopoda	Anthuridae	*Cyathura carinata*	Native													1	3		4
	Order Mysida	Mysidae	*Gastrosaccus spinifer*	Native					1	1		6								8
	Order Sessilia	Austrobalanidae	*Austrominius modestus*	Non native			2													2
		Balanidae	*Amphibalanus improvisus*	Non native	35	21	12	2	3								3	108	2	186
			Balanus cf crenatus	Native		2														2
	Order Tanaidacea	Tanaididae	*Sinelobus stanfordi****	Non native		1														1
Mollusca	Class Bivalvia	Anomiidae	*Anomia ephippium*	Native					1											1

					SECOND SAMPLING SURVEY (01/2020)															
TAXA				**NATIVE / NON NATIVE**	**S1**	**S1**	**S1**	**S2**	**S2**	**S2**	**S3**	**S3**	**S3**	**S4**	**S4**	**S4**	**R**	**R**	**R**	**TOTAL**
		Corbulidae	*Corbula gibba*	Native						1							2			3
		Mactridae	*Spisula subtruncata*	Native							6	2			1					9
		Myidae	*Mya arenaria*	Non native									1							1
		Mytilidae	*Mytilus edulis*	Native	5	5	13											1		24
		Ostreoidea	*Crassostrea (Magallana) gigas*	Non native	8	6	4											1		19
		Pharidae	*Ensis cf leei***	Non native							1									1
			Phaxas pellucidus	Native							1									1
		Semelidae	*Abra nitida*	Native									1							1
		Tellinidae	*Tellina tenuis*	Native							1									1
		Veneridae	*Ruditapes philippinarum*	Non native				1	3	4	2	2							1	13
	Class Gasteropoda	Calyptraeidae	*Crepidula fornicata*	Non native		2	1	1	2											6
		Nassariidae	*Tritia (Nassarius) reticulata*	Native							3									3
Cnidaria	Class Anthozoa	Actiniidae	*Actinia equina*	Native	3	1		1					1							6
Number of individuals					62	47	45	49	33	11	66	23	17	30	15	7	24	162	12	603
Number of species					10	12	10	12	11	6	13	8	10	3	5	4	4	9	5	44

** Broken animals or females.*
*** Recently renamed, previously named as Ensis americanus.*
**** It can be the first record for the area. Van Haaren et al (2009) identified this specie in The Netherlands and Belgium.*
***** Common in harbour areas with hydrocarbon enrichment (Fauna Iberica, CSIC España). It is considered as instability indicator.*
cf recently studies are reviewing data because they suggest most of the registers for Laonome kroyeri are in fact Laonome xeprovala.*

5.4.2 Biotic indices

The AMBI index (Borja et al. 2000) will be applied when all the sampling surveys will be completed to classify the identified species into ecological categories, calculate the ecological quality ratio (EQR) and qualify the ecological status (ES) of the study area (Table 6). AMBI is a commonly used index and is for official use within the WFD as part of different multimetric indices in Portugal, the United Kingdom, Ireland, Denmark, Norway and the Netherlands. AMBI has been tested in different geographic regions and has been proved to have large geographical coverage. AMBI shows responsiveness to various pressures (Borja et al. 2015) and is considered suitable for the pressures met in the study area ▪ chemical pollution: industrial discharges or presence of metals and organic compounds in water and/or sediment, ▪ Dredging and sediment disposal: activity needed to maintain navigability in channels and harbours, creation of new harbours and disposal of sediments, ▪ Harbours: presence of ports and normal activity, excluding dredging.

Table 6: AMBI values and classification (Borja et al. 2000, 2003)

Index value	Dominating ecological group	Benthic community health	Site disturbance classification	ES
0.0 ≤ AMBI ≤ 0.2	I-II	Normal	Undisturbed	High
0.2 < AMBI ≤ 1.2		Impoverished		
1.2 < AMBI ≤ 3.3	III	Unbalanced	Slightly disturbed	Good
3.3 < AMBI ≤ 4.3	IV-V	Transitional to polluted	Moderately disturbed	Moderate
4.3 < AMBI ≤ 5.0		Polluted		Poor
5.0 < AMBI ≤ 5.5	V	Transitional to heavy pollution	Heavily disturbed	
5.5 < AMBI ≤ 6.0		Heavy polluted		Bad
6.0 < AMBI ≤ 7.0	Azoico	Azoic	Extremely disturbed	

Note: Group I: Species very sensitive to organic enrichment and present under unpolluted conditions. Group II: Species indifferent to enrichment, always present in low densities with non-significant variations with time. Group III: Species tolerant to excess organic matter enrichment. These species may occur under normal conditions; however, their populations are stimulated by organic enrichment. Group IV: Second-order opportunistic species, adapted to slight to pronounced unbalanced conditions. Group V: First-order opportunistic species, adapted to pronounced unbalanced situations. (Grall and Glemarec, 1997).

Apart from AMBI also BOPA index (Dauvin and Ruellet, 2007) will be implemented. BOPA index results from the refinement of the polychaeta/amphipoda ratio (Gomez-Gesteira and Dauvin, 2000). Accordingly, this index will be used to assign the estuarine communities into the five ES categories (Table 7).

Table 7: BOPA values and classification

Index value	Site disturbance classification	ES
0.00000 ≤ BOPA ≤ 0.06298	Unpolluted sites	High
0.04576 < BOPA ≤ 0.19723	Slightly polluted	Good
0.13966 < BOPA ≤ 0.28400	Moderately polluted	Moderate
0.19382 < BOPA ≤ 0.30103	Heavily polluted	Poor
0.26761 < BOPA ≤ 0.30103	Extremely polluted	Bad

6 Conclusions

Comprehensive conclusions will be made once field and laboratory work will be completed. So far, three seasonal surveys (September, January and July) have been conducted covering 2 EVIDES-operated desalination plants in the Port of Rotterdam. While a remarkable diversity of taxa has been observed, enabling a detailed characterization of biological communities (which constitutes significant asset considering how little published literature exists about the unique system of the Port of Rotterdam), it will be challenging to discern the impacts of desalination brine outfalls given that in particular the Britanniehaven area is heavily impacted by an adjacent oil refinery.

7 Bibliography

W. Admiraal, G. van der Velde, H. Smit & W.G. Cazemier, 1993, The Rivers Rhine and Meuse in The Netherlands: Present state and signs of ecological recovery, Hydrobiologia 265: 97-128.

Anonymous, 1999. De kwaliteit van de waterbodem in het Rotterdamse havengebied 1981–1998. Rijkswaterstaat, Directie Zuid-Holland, Gemeentelijk Havenbedrijf Rotterdam.

Anonymous, 2006. De kwaliteit van de waterbodem in het Rotterdamse havengebied 1998–2005. Syncera Water B.V. In opdracht van Havenbedrijf Rotterdam N.V. en Rijkswaterstaat, dienst Zuid-Holland.

A. Borja, J. Franco, and V. Pérez, A, 2000, Marine Biotic Index to establish the ecological quality of soft-bottom benthos within European estuarine and coastal environments, Mar. Pollut. Bull. 40 (2000), pp. 1100–1114.

A. Borja, I. Muxica, and F. Franco, 2003, The application of a Marine Biotic Index to different impact sources affecting soft-bottom benthic communities along European coasts, Mar. Pollut. Bull. 46 (2003), pp. 835–845.

Angel Borja, Sandra L. Marin, Inigo Muxika, Loreto Pino, Jose G. Rodriguez, 2015, Is there a possibility of ranking benthic quality assessment indices to select the most responsive to different human pressures?, Marine Pollution Bulletin 97 (2015) 85–94.

Natalia Belkin, Eyal Rahav, Hila Elifantz, Nurit Kress, Ilana Berman-Frank, 2017, The effect of coagulants and antiscalants discharged with seawater desalination brines on coastal microbial communities: A laboratoryandin situstudy from the southeastern Mediterranean, Water Research 110 (2017) 321-331

Dauvin, J.C., Ruellet, T., 2007, Polychaete/amphipod ratio revisited, Marine Pollution Bulletin 55, 215–224.

J.A. de-la-Ossa-Carretero, Y. Del-Pilar-Ruso, A. Loya-Fernández, L.M. Ferrero-Vicente, C. Marco-Méndez, E. Martinez-Garcia, F. Giménez-Casalduero, J.L. Sánchez-Lizaso, 2016, Bioindicators as metrics for environmental monitoring of desalination plant discharges, Marine Pollution Bulletin 103 (2016) 313–318.

Yoana Del-Pilar-Ruso, Elena Martinez-Garcia, Francisca Gimenez-Casalduero, Angel Loya-Fernandez, Luis Miguel Ferrero-Vicente, Candela Marco-Mendez, Jose Antonio de-la-Ossa-Carretero, Jose Luis

Sanchez-Lizaso, 2015, Benthic community recovery from brine impact after the implementation of mitigation measures, Water research 70 (2015) 325-336.

Panagiotis D. Dimitriou, Eva Chatzinikolaou, Christos Arvanitidis, 2020, Ecological status assessment based on benthic macrofauna of three Mediterranean ports: Comparisons across seasons, activities and regions, Marine Pollution Bulletin 153 (2020) 110997.

Elliott, M., Quintino, V., 2007, The estuarine quality paradox, environmental homeostasis and the difficulty of detecting anthropogenic stress in naturally stressed areas. Marine Pollution Bulletin 54, 640–645.

European Commission, 2000, Directive 2000/60/EC of the European Parliament and of the Council of 23 October 2000 establishing a framework for community action in the field of water policy. Off. J. Eur. Communities 2000.

Grall, J., Glemarec, M., 1997, Using biotic indices to estimate macrobenthic community perturbations in the Bay of Brest. Estuar. Coast. Shelf Sci. 44 (Suppl. A), 43–53.

Gomez-Gesteira, J.L., Dauvin, J.C., 2000, Amphipods are good bioindicators of the impact of oil spills on soft-bottom macrobenthic communities, Marine Pollution Bulletin 40 (11), 1017–1027.

Nadine Heck, Adina Paytan, Donald C. Potts, Brent Haddad, Karen Lykkebo Petersen, 2018, Management preferences and attitudes regarding environmental impacts from seawater desalination: Insights from a small coastal community, Ocean and Coastal Management 163 (2018) 22–29.

Internationale Kommission zum Schutz des Rheins (IKSR), Commission Internationale pour la Protection du Rhin (CIPR), Internationale Commissie ter Bescherming van de Rijn (ICBR), 2015, Internationaal gecoördineerd stroomgebiedbeheerplan 2015 van het internationaal stroomgebieddistrict Rijn.

I. Muxica, A. Borja, and W. Bonne, The suitability of the Marine Biotic Index (AMBI) to new impact sources along European coasts, Ecol. Indic. 5 (2005), pp. 19–31.

Alfonso Nebra, Nuno Caiola, Gloria Muñoz-Camarillo, Silvia Rodríguez-Climent, Carles Ibáñez, 2014, Towards a suitable ecological status assessment of highly stratified mediterranean estuaries: A comparison of benthic invertebrate fauna indices, Ecological Indicators 46 (2014) 177–187.

Paalvast, P., 2002. Historische ontwikkeling estuariene ecotopen in de Noordrand van het benedenrivierengebied. In opdracht van het Rijksinstituut voor Kust en Zee/RIKZ. Ecoconsult, rapportnr.

Peter Paalvast, Bregje K. van Wesenbeeck, Gerard van der Velde, Mindert B. de Vries, 2012, Pole and pontoon hulas: An effective way of ecological engineering to increase productivity and biodiversity in the hard-substrate environment of the port of Rotterdam, Ecological Engineering 44 (2012) 199– 209.

Phillips, N., C.M. Smith, and C.W. Morden. 2001. An effective DNA extraction protocol for brown algae. Phycological Research 49: 97-102.

Rute Pinto, Joana Patrıcio, Alexandra Baeta, Brian D. Fath, Joao M. Neto, Joao Carlos Marques, 2009, Review and evaluation of estuarine biotic indices to assess benthic condition, Ecological Indicators 9 (2009) I-25.

Port of Rotterdam Authority, 2019, Highlights of 2019 Annual Report.

Valentina Pitacco, Lovrenc Lipej, Borut Mavrič, Michele Mistri, Cristina Munari, 2018, Comparison of benthic indices for the evaluation of ecological status of three Slovenian transitional water bodies (northern Adriatic), Marine Pollution Bulletin 129 (2018) 813–821.

Quintino, V., Elliott, M., Rodrigues, A.M., 2006. The derivation, performance and role of univariate and multivariate indicators of benthic change: case studies at differing spatial scales. J. Exp. Mar. Biol. Ecol. 330, 368–382.

Rodrigo Riera, Fernando Tuya, Alicia Sacramento, Eva Ramos, Myriam Rodríguez, Óscar Monterroso, 2011, The effects of brine disposal on a subtidal meiofauna community, Estuarine, Coastal and Shelf Science 93 (2011) 359-365.

R. Riera, Ó. Monterroso, M. Rodríguez and E. Ramos, 2011, Biotic indexes reveal the impact of harbour enlargement on benthic fauna, Chemistry and Ecology Vol. 27, No. 4, August 2011, 311–326.

Rijkswaterstaat, Water info portal, https://waterinfo.rws.nl/#!/nav/index/, https://ihm-open-data-viewer-waterinfo.infoprojects.nl/index.htm

Smits, A.J.M., Nienhuis, P.H., Saeijs, H.L.F., 2006. Changing estuaries, changing views. Hydrobiologia 565, 339–355.

Almeida Vinagre, Antónia Juliana Pais-Costa, Stephen John Hawkins, Ángel Borja, João Carlos Marques, João Magalhães Neto, 2017, Addressing a gap in the Water Framework Directive implementation: Rocky shores assessment based on benthic macroinvertebrates, Ecological Indicators 78 (2017) 489–501.

Nikolaos Voulvoulis, Karl Dominic Arpon, &, Theodoros Giakoumis, 2017, The EU Water Framework Directive: From great expectations to problems with implementation, Science of the Total Environment 575 (2017) 358–366.

P.C.B. de Wit (Port of Rotterdam), G. Bolier (Delft University of Technology), P. Paalvast (Ecoconsult), G. van der Velde (Radboud University Nijmegen), M. de Vries (WL | Delft Hydraulics), 2007, Contribution to the Development of Ecological Goals and Indicators for the Aquatic Environment in Ports, Challenges in the Port of Rotterdam Area, Report Theme 2, NEW! Delta project.

Wolff, W.J., 1978. The degradation of ecosystems in the Rhine. In: Holdgate, M.W., Woodman, M.J. (Eds.), The Breakdown and Restoration of Ecosystems. Plenum Press, New York, pp. 167–187.